

A green and simplified approach for the quantitative and sensitive analysis of heavy metal ions in sea and stream waters

Celal Duran ^{a,*}, Aslihan Yilmaz Camoglu^a, Duygu Ozdes^b and Olcay Bekircan^a

^a Faculty of Sciences, Department of Chemistry, Karadeniz Technical University, Trabzon, Turkey

^b Gumushane Vocational School, Gumushane University, Gumushane, Turkey

*Corresponding author. E-mail: cduran@ktu.edu.tr

 CD, 0000-0002-1306-9061

ABSTRACT

Elimination of the matrix effect is a major challenge in developing a method for the quantification of heavy metals (HMs) in water samples. In this regard, the current research describes the simultaneous analyses of Cu(II), Cd(II), and Ni(II) ions in water matrices through flame atomic absorption spectrophotometry (FAAS) after preconcentration with carrier element-free co-precipitation (CEFC) technique by the help of an organic co-precipitant, 3-[[5-(4-Chlorobenzyl)-3-(4-chlorophenyl)-1H-1,2,4-triazol-1-yl]-methyl]-4-[2,4-(dichlorobenzylidene)amino]-1H-1,2,4-triazole-5(4H)-thione (CCMBATT). Based on our literature research, CCMBATT was employed for the first time in this study as an organic co-precipitant for the preconcentration of HMs. Factors such as solution pH, concentration of co-precipitant, sample volume, standing time, centrifugation rate, and time were thoroughly examined and optimized to achieve the highest efficiency in terms of HM recovery. The limits of detection (LODs) (with 10 number of tests) of 0.54, 0.34, and 1.95 $\mu\text{g L}^{-1}$ and the relative standard deviations (RSD %) of 2.1, 3.3, and 3.0 were determined for Cu(II), Cd(II) and Ni(II) ions, respectively. Recovery results of HMs for the spiked samples were in the range of 92.8–101.0%, demonstrating the trueness of the method and its applicability to the water samples matrix.

Key words: cadmium, copper, co-precipitation, flame atomic absorption spectrometry, nickel, preconcentration

HIGHLIGHTS

- CCMBATT has been utilized for the first time as an organic co-precipitant in CEFC method for the precise determination of HMs at low levels in water samples.
- CCMBATT enables the direct quantification of hazardous HMs on itself, eliminating the need for the introduction of any additional carrier elements.
- The risk of analyte contamination and interferences caused by the carrier element has been eliminated.

1. INTRODUCTION

In recent years, contamination caused by HMs in the environment has increased gradually due to factors, such as population growth, industrialization, and human activities. Unlike some other pollutants that can break down naturally over time or be broken down by biological processes, heavy metals (HMs) persist in the environment for long periods causing toxic effects in living organisms (Khah *et al.* 2021; Vig *et al.* 2023). The extent of these effects depends on the concentration and chemical forms of the HMs. Common health effects include harm to the kidneys, liver, lungs, and nervous system. Human activities, such as industrial processes, mining, improper waste disposal, and the use of HMs in products, contribute to the release of these toxic substances into the environment (Melebari 2023). As a result, HMs contaminate soil, water bodies, and the atmosphere.

Cadmium (Cd) is a poisonous transition metal which is not essential for human and animal health. Cd is naturally present in the environment but is also released through agricultural and industrial activities, leading to pollution. Exposure to Cd primarily occurs through the consumption of contaminated food and water (Rehman *et al.* 2018). Scientific studies have indicated a potential relationship between Cd exposure and various types of cancer, such as breast, prostate, lung, pancreatic, and kidney cancers (Genchi *et al.* 2020). Copper (Cu) and nickel (Ni) are essential elements that serve as cofactors for

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numerous vital enzymes and play a crucial role in protein, carbohydrate, and lipid metabolism (Karadaş 2017; Shakerian *et al.* 2019). However, while Cu and Ni are necessary for normal metabolism, excessive intake or exposure to high amounts can lead to detrimental effects on the body. Hence, the ascertainment of Cu(II), Cd(II), and Ni(II) ions and other HMs in water is crucial for assessing water quality and identifying potential environmental and health risks.

The flame atomic absorption spectrophotometer (FAAS) is commonly performed for quantification of HMs in environmental matrices. FAAS is a commonly utilized technique due to its advantages of sensitivity, rapid analysis, reliability, simplicity, and cost-effectiveness (Ozdes *et al.* 2023). However, in most circumstances, the sensitivity of FAAS and also other instrumental methods may not be adequate for accurately analyzing metal ions at very low levels in complex sample matrices. To overcome this limitation, a variety of separation and preconcentration procedures including flotation (Wang *et al.* 2022), cloud point extraction (Li *et al.* 2022), membrane filtration (Xiang *et al.* 2022), liquid–liquid extraction (Nyamoto *et al.* 2022), solid phase extraction (Zhao *et al.* 2021), and co-precipitation (Moreira *et al.* 2020) are applied in conjunction with FAAS to enhance sensitivity and improve the precision of metal ion determination.

The co-precipitation technique has great advantages due to its simplicity, rapidity, and capability to obtain high preconcentration factors (PF). In addition, the minimized usage of organic solvents which may be ecologically harmful is considered as a notable advantage. The inclusion, occlusion, and surface adsorption mechanisms are effective in the accumulation of the HMs (Ozdes *et al.* 2023). In the case of inorganic co-precipitation, trace elements are deposited onto the precipitate formed by different metal hydroxides (Saracoglu *et al.* 2012; Mohammadi *et al.* 2019; Moreira *et al.* 2020). Conversely, organic co-precipitation involves the addition of a significant quantity of carrier element (Bi, Cu, Co, and Ni) to the medium, resulting in the formation of water-insoluble complexes with ligands like N-benzoyl-N-phenyl-hydroxylamine (Saçmacı & Kartal 2010), 4-(2-Pyridylazo)-resorcinol (Tokaloğlu & Daşdelen 2011), pyrrolidine dithiocarbamate (Baysal *et al.* 2008), and 8-Hydroxyquinoline (Feist & Mikula 2014). The trace elements are then deposited onto these organic precipitates. However, it should be noted that the excess carrier element added in both cases may introduce interference effects during the analysis stage.

In CEFC procedure, a soluble compound in organic solvents but insoluble in water is employed as the co-precipitant. A significant advantage of the CEFC method over conventional co-precipitation methods is that it does not necessitate the use of a carrier element for precipitate formation (Gouda *et al.* 2022). Thus, the risk of pollution caused by the carrier element can be eliminated. Moreover, the CEFC method is more environmentally friendly, as it requires fewer chemical reagents. These advantages, including reduced environmental impact and minimal chemical usage, make the CEFC method highly favorable for trace element determination and analysis (Uluozlu & Tuzen 2015; Roushani *et al.* 2016).

In this study, a CEFC process was proposed to simultaneously analyze Cu(II), Cd(II), and Ni(II) ions in water samples, including stream and sea water. A novel organic co-precipitant, CCMBATT, was utilized for the first time in the separation and preconcentration of HMs. Even, no previous research has been found in which CCMBATT was used in other analytical methods. To optimize the CEFC method, several analytical parameters were investigated to determine their influence on the simultaneous recovery of the target metal ions. Additionally, the impact of various matrix components including Na⁺, K⁺, Ca²⁺, Mg²⁺, SO₄²⁻, NO₃⁻, PO₄³⁻, NH₄⁺, Fe(III), Al(III), Mn(II), Zn(II), phenol, and 4-chlorophenol, commonly present in environmental samples was assessed to ensure the method's reliability and accuracy. Furthermore, spike tests were performed to validate the method's performance prior to its application in the analysis of water samples.

2. MATERIALS AND METHODS

2.1. Chemicals and instruments

Analysis of HMs was conducted using a PerkinElmer AAnalyst 400 flame atomic absorption spectrometer. The spectrometer was equipped with an air/acetylene burner and a deuterium background correction system. Hollow cathode lamps were employed as the radiation sources for the determination of HMs. The specific wavelengths used for each element were 324.75 nm for Cu(II), 228.80 nm for Cd(II), and 232.00 nm for Ni(II). The pH measurements of the solutions were realized by a Hanna pH-211 digital pH meter with a glass electrode. A centrifuge, model NUVE NF 200, was utilized for the separation of the metal-containing precipitate from the solution.

The experiments utilized high-purity analytical grade chemicals and reagents sourced from reputable suppliers such as Merck (Darmstadt, Germany) or Sigma-Aldrich (St. Louis, MO, USA). The standard and working solutions of HMs were prepared on a daily basis by diluting the certified stock solutions (1,000 mg L⁻¹) of them with deionized water. The solutions of

interfering cations and anions were prepared from the high-purity compounds (99.9%) of NaCl, KCl, CaCl₂, Mg(NO₃)₂, Na₂SO₄, NaNO₃, Na₃PO₄, and NH₄NO₃ salts. For cleaning the laboratory glassware and polypropylene falcon tubes were first stored in a 10% (v/v) HNO₃ solution for at least 24 h. Then they were washed well with tap water followed by distilled water.

The co-precipitating agent, 3-[[5-(4-Chlorobenzyl)-3-(4-chlorophenyl)-1H-1,2,4-triazol-1-yl]-methyl]-4-[2,4-(dichlorobenzylidene)amino]-1H-1,2,4-triazole-5(4H)-thione (CCMBATT), was synthesized in accordance with the methodology presented in the literature (Ozdemir *et al.* 2017). The chemical structure of CCMBATT was set out in Figure 1. The stock solution of CCMBATT was prepared at a concentration of 1.02 mM by dissolving 0.060 g of CCMBATT in a 100 mL mixture of ethanol and dimethyl sulphoxides (1:1 (v/v)).

2.2. Co-precipitation procedure

First, experiments were carried out for Pb(II), Co(II), Cr(III), Cu(II), Cd(II), and Ni(II) ions to assess which HMs the developed CEFC method can be used for simultaneous separation and preconcentration. It was noticed that the method can only be utilized for the simultaneous determination of Cu(II), Cd(II), and Ni(II) ions. Therefore, the method was applied for these HMs. The optimization process for the co-precipitation method involves the examination of the influences of experimental factors on the recovery of HMs using centrifuge tubes containing 15.0 mL of model solutions. These solutions contained 10 µg of Cd(II), 25 µg of Cu(II), and Ni(II) ions. To initiate the process, 1.0 mL of 1.02 mM CCMBATT solution was added to the tubes. Diluted NaOH or HNO₃ solutions were then used to adjust the pH of the solutions to 7.0. After a 30-min incubation period, the tubes were centrifuged at 3,000 rpm for 5 min. The resulting supernatant was carefully removed, and the precipitate at the bottom of the tube was dissolved with 1.0 mL of concentrated HNO₃. The solution was then brought to a final volume of 4.0 mL using distilled water. Finally, the contents of HMs in the solution were directly analyzed using FAAS.

To investigate the effect of sample volume on the analyte ions' recovery, a study was conducted within the sample volume range of 50–750 mL at pH 7.0. The samples contained 10 µg of Cd(II) and 25 µg of Cu(II) and Ni(II) ions, along with 0.07 mM of CCMBATT. Experimental procedures differed based on the sample volume used. For sample volumes up to 50 mL, polyethylene centrifuge tubes were employed. A centrifuge device was used to separate the precipitate from the solution after the necessary duration of precipitation. In contrast, sample volumes exceeding 50 mL underwent CEFC experiments using appropriately sized beakers. Precipitates formed in the beakers were collected using a cellulose nitrate membrane after allowing sufficient time for precipitation. The collected precipitates were subsequently filtered using cellulose nitrate membrane filter paper. Decomposition of the filter paper was carried out by heating it with 1.0 mL of concentrated HNO₃ on a heating plate at 80 °C. Following decomposition, the final volume was adjusted to 4.0 mL using distilled water. Analyte ions were then analyzed using FAAS.

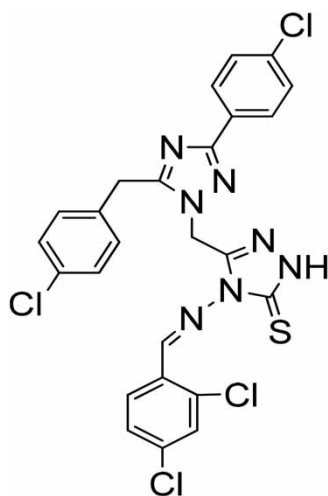


Figure 1 | Chemical structure of CCMBATT.

2.3. Preparation of water samples

The recommended methodology utilizing CCMBATT was carried out to ascertain the concentration of HMs in sea and stream water samples collected from five different regions in 2023. These samples were grouped as follows: sea water was taken from the coast of Yüzüncü Yıl Park, where traffic and human activities are intense in the Black Sea (Trabzon), and stream waters were taken from Değirmendere (Trabzon), which is close (approximately 1 km) to the organized industrial zone (coded as stream water 1), from Harşit (Gümüşhane), which is also close (approximately 2.0 km) to the organized industrial zone (coded as stream water 2), and from areas close (approximately 500 m) to two different mining enterprises operating in Gümüşhane province (coded as stream water 3 and 4). Water samples were collected from each region in duplicate into 500 mL precleaned polyethylene bottles. They were stored in a refrigerator at a temperature of 4 °C until they were ready for analysis. Concentrated HNO₃ was used to reduce the pH value of the samples below 2.0. Filtration of the water samples was performed using cellulose nitrate membranes with a pore size of 0.45 µm. To prepare the samples for analysis, the pH of the water samples was adjusted to 7.0. Subsequently, 6.9 mL of 1.02 mM stock CCMBATT solution was added to each water sample at the optimum sample volume (100 mL), so that the final CCMBATT concentration was 0.07 mM. The co-precipitation method detailed earlier was then implemented to determine the levels of HMs in the samples.

3. RESULTS AND DISCUSSION

3.1. Evaluation and optimization of the experimental conditions

pH value of the medium plays a significant role in determining the solubility and speciation of the metal ions, as well as their interaction with the co-precipitant. On the other hand, the surface structure of the precipitate occurred is directly affected by the pH of the solution. By carefully controlling the pH, it is possible to manipulate the electrostatic interactions and surface chemistry, leading to enhanced co-precipitation efficiency and the desired recovery of metal ions (Ozdes *et al.* 2023). Therefore, the optimization of pH value is crucial in co-precipitation processes. The influence of pH upon the co-precipitation of HMs was evaluated within the pH range of 2.0–8.0. The experimental conditions included the use of 0.07 mM of CCMBATT, with a standing time of 30 min, a centrifugation rate of 3,000 rpm, and a centrifugation time of 5 min. In highly acidic conditions, the analyte ions' recovery was found to be negligible. However, as the pH of the solution increased from 2.0 to 8.0, a significant improvement in the recovery of analyte ions was observed. Specifically, the recovery values increased from 39.3 to 98.9% for Cu(II), from 13.0 to 99.0% for Cd(II), and from 9.2 to 95.6% for Ni(II) ions as the pH of the aqueous solution was increased from 2.0 to 8.0 (Figure 2). At pH 7.0, the precipitate formed exhibits a negatively charged surface owing to the presence of OH⁻ ions in the medium. As a result, the electrostatic interaction between the metal cations and the negatively charged surface of the precipitate intensifies, promoting adsorption. This adsorption phenomenon, which is one of the mechanisms involved in co-precipitation, contributes to the increased recovery of the analyte ions. Based on the findings, the optimum pH for further experiments was decided to be 7.0. Similar to our results, Gouda *et al.* (2022) determined the

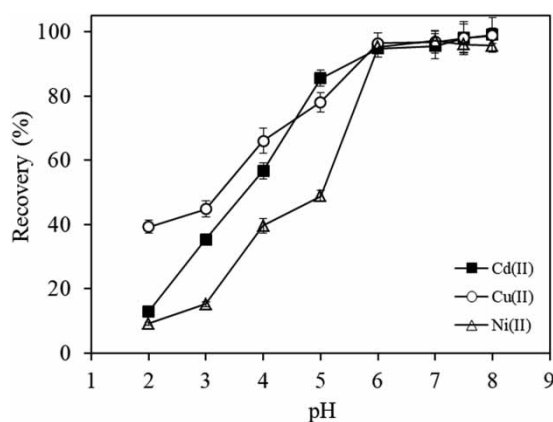


Figure 2 | Influences of pH on the recoveries of analyte ions (sample volume: 15 mL, concentration of CCMBATT: 0.07 mM, standing time: 30 min, centrifugation rate: 3,000 rpm, centrifugation time: 5 min, *N*:3).

optimum pH as 7.0 in the separation and preconcentration of Cd(II), Cu(II), Ni(II), and Pb(II) ions in water and food samples by the CEFC method using a triazole-derivative ligand as organic co-precipitant.

To assess the impact of CCMBATT quantity on the co-precipitation of HMs, a series of experiments were conducted using different concentrations of CCMBATT in the range of 0.0–0.20 mM at a solution pH of 7.0. The absence of CCMBATT led to considerably lower recovery values, with Cu(II), Cd(II), and Ni(II) ions exhibiting recovery values of 63.2, 69.1, and 62.4%, respectively. This outcome underscored the essential role and necessity of CCMBATT for the accurate and quantitative determination of HMs. Significant improvements in recovery values were observed as the concentration of CCMBATT increased from 0.02 to 0.07 mM. However, further increases in CCMBATT quantity resulted in relatively minor changes in the recovery values, reaching a plateau (Figure 3). Consequently, a CCMBATT concentration of 0.07 mM was determined as the optimal quantity for subsequent co-precipitation experiments. The amount of ligand used in the current study is less than many CEFC methods presented in the literature (Duran *et al.* 2011; Gouda *et al.* 2022; Ozdes *et al.* 2023). This is an advantage of the current study as it prevents the consumption of large amounts of chemicals.

Achieving high PF is crucial for the accurate and sensitive determination of analyte ions present at low levels in environmental samples using the proposed method. PF is determined by dividing the optimum sample volume, from which the analyte ions are quantitatively recovered, by the final volume. Thus, the sample volume is the primary factor influencing the PF. Consequently, it is imperative to designate the optimal sample volume for the developed CEFC procedure in order to achieve the desired analytical performance. The results of the experiments demonstrated that as the sample volume is increased from 50 to 750 mL, the recovery percentages decreased from 98.0 to 52.0% for Cu(II) ions, from 103.0 to 41.0% for Cd(II) ions, and from 97.9 to 37.9% for Ni(II) ions (Figure 4). It was observed that the maximum sample volume resulting

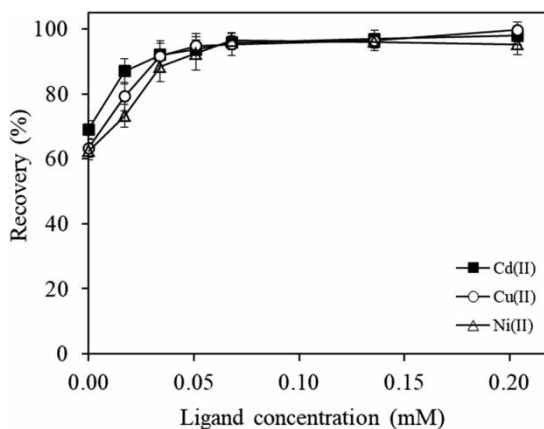


Figure 3 | Effect of CCMBATT concentration on the recoveries of analyte ions (Sample pH: 7.0, sample volume: 15 mL, $N:3$).

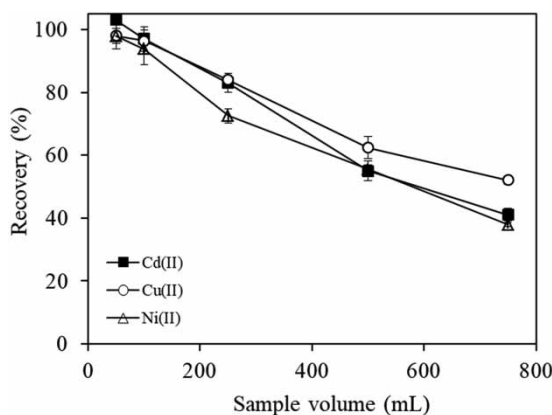


Figure 4 | Impact of sample volume on the recoveries of analyte ions (Sample pH: 7.0, $N:3$).

in quantitative recovery was 100 mL. At this volume, the recovery percentages for Cu(II), Cd(II), and Ni(II) ions were 96.4, 97.0, and 93.8%, respectively. Utilizing these recovery values, the PF was calculated as 25, with an eluent volume of 4.0 mL. Saracoglu & Soylak (2010) determined PF as 10 for Cd(II) and Cu(II) ions by using a cationic surfactant (N-cetyl N,N,N trimethyl ammonium bromide (CTAB)) as co-precipitant. Tokaloğlu & Yıldız (2009) obtained PF as 20 for Cu(II) ions utilizing Ni(II)-salicylaldoxime co-precipitant.

In order to facilitate the formation of precipitates and the co-precipitation of analyte ions in the solution, it is necessary to allow a specific duration of time after adding CCMBATT to the medium. This standing time enables the interaction of various mechanisms, such as surface adsorption, inclusion, and occlusion. To determine the optimum standing time, experiments were conducted over a range of 1–120 min at pH 7.0, using 0.07 mM of CCMBATT. It was found that the highest recoveries (99.5, 95.6, and 91.6% for Cd(II), Cu(II), and Ni(II), respectively) for all three analyte ions were achieved when a standing time of 30 min was applied. Saracoglu & Soylak (2010) also determined the standing time as 30 min for CEFC of Cd(II), Cr(III), Co(II), Cu(II), Fe(III), Pb(II), and Mn(II) ions in environmental samples by using a cationic surfactant as organic co-precipitant. Subsequently, the centrifugation rate and time were assessed to ensure proper separation of the precipitate from the solution without any loss. The experiments involved centrifugation rate and time in the range of 1,000–4,000 rpm and 1–10 min, respectively. The results demonstrated that 3,000 rpm of centrifugation rate and 5 min of centrifugation time were sufficient for achieving simultaneous quantitative recoveries. At 3,000 rpm centrifugation rate, the recoveries of Cd(II), Cu(II), and Ni(II) ions were obtained as 96.5, 94.0, and 96.1%, respectively and at 5 min of centrifugation time the recoveries were 93.5, 97.9, and 96.1%, respectively. Mendil *et al.* (2015) required higher centrifugation speed (4,000 rpm) and time (30 min) for quantitative recovery in the CEFC method developed using 2,9-dimethyl-4,7-diphenyl-1,10-phenanthroline for the determination of Cu(II), Pb(II), Zn(II), Fe(III), and Cr(III) ions from various water and food samples.

3.2. Influences of matrix ions

In the determination of HMs using different instrumental techniques, the presence of other components in the sample medium can lead to matrix interference, causing a significant challenge (Duran *et al.* 2011). Hence, it is necessary to evaluate the effects of various anions, cations, and organic species that may coexist with the analyte ions in water samples. Experiments focused on the impact of these species at certain levels on the recovery of analyte ions utilizing the developed co-precipitation method. This analysis was important to investigate the potential influences of these species on the accuracy and reliability of the method when applied to real water samples. When high levels of foreign ions are present, the recovery values for Cu(II), Cd(II), and Ni(II) ions exhibit variations ranging from 91.7 to 99.8%, from 91.9 to 104.0%, and from 91.8 to 104.0%, respectively. Even in a highly complex matrix where multiple ions coexist at specific concentrations, the recovery values for Cu(II), Cd(II), and Ni(II) ions were determined to be 99.6, 104.0, and 93.4%, respectively (Table 1). Based on the results obtained, it can be noticed that the presence of various ions at specific concentrations, including 5,000 mg L⁻¹ of Na⁺, 1,000 mg L⁻¹ of K⁺, Ca²⁺, Mg²⁺, SO₄²⁻, NO₃⁻, PO₄³⁻, and NH₄⁺, as well as 25 mg L⁻¹ of Fe(III), Al(III), Mn(II), and Zn(II), and their mixture, and also 10 mg L⁻¹ of phenol and 4-chlorophenol does not significantly interfere with the analysis of analyte ions using the developed method. These results demonstrate the outstanding performance of the developed method in effectively recovering the target analyte ions, even under challenging conditions with a diverse range of interfering species. Similar results were obtained in the separation and preconcentration of various metal ions using Co(OH)₂ (Mohammadi *et al.* 2019), copper(II)-N-benzoyl-N-phenyl-hydroxylamine complex (Saçmacı & Kartal 2010), and holmium hydroxide (Saracoglu *et al.* 2012) co-precipitation systems.

3.3. Analytical characteristics of the proposed method

Analytical characteristics of the developed procedure were thoroughly evaluated under optimized experimental conditions. A linear calibration curve was established for Cu(II) and Ni(II) ions in the concentration range of 0.1–5.0 µg mL⁻¹ and for Cd(II) ions in the range of 0.1–2.0 µg mL⁻¹ with correlation coefficients higher than 0.99. The limits of detection (LODs) and limits of quantification (LOQs) were determined using the standard deviation obtained from blank measurements at the optimum conditions. The LODs were calculated as 3 times the standard deviation, while the LOQs were determined as 10 times the standard deviation. The LODs were calculated as 0.54, 0.34, and 1.95 µg L⁻¹ and LOQs were obtained as 1.81, 1.13, and 6.61 µg L⁻¹ for Cu(II), Cd(II), and Ni(II) ions, respectively. The precision of the method was evaluated by conducting the analysis ten times on 15 mL solutions containing 10 µg of Cd(II) and 25 µg of Cu(II) and Ni(II) ions under the optimized conditions. This allowed for a comprehensive assessment of the method precision and determination of the relative

Table 1 | Common foreign ions' effect on the recoveries of HMs (CCMBATT conc.: 0.07 mM, pH: 7.0, sample volume: 15 mL, standing time: 30 min, centrifugation rate: 3,000 rpm, centrifugation time: 5 min, *N*:3)

Ions	Added as	Conc. (mg L ⁻¹)	Recovery (%)		
			Cd(II)	Cu(II)	Ni(II)
Na ⁺	NaCl	5,000	96.0 ± 3.4	98.6 ± 0.7	99.8 ± 0.4
K ⁺	KCl	1,000	96.9 ± 0.4	92.9 ± 2.0	96.5 ± 1.4
Ca ²⁺	CaCl ₂	1,000	95.8 ± 2.4	98.8 ± 1.7	103.0 ± 5.7
Mg ²⁺	Mg(NO ₃) ₂	1,000	91.9 ± 1.1	96.4 ± 1.7	94.0 ± 1.4
SO ₄ ²⁻	Na ₂ SO ₄	1,000	98.9 ± 0.8	98.1 ± 2.0	104.0 ± 1.4
NO ₃ ⁻	NaNO ₃	1,000	94.9 ± 3.2	92.9 ± 1.3	95.3 ± 0.4
PO ₄ ³⁻	Na ₃ PO ₄	1,000	97.6 ± 3.2	91.7 ± 3.7	94.8 ± 1.5
NH ₄ ⁺	NH ₄ NO ₃	1,000	98.4 ± 2.1	99.8 ± 1.0	91.8 ± 3.9
Al(III), Mn(II), Fe(III), Zn(II)	^a	25	94.0 ± 2.7	95.2 ± 3.8	95.4 ± 2.0
Phenol	-	10	93.6 ± 0.1	91.5 ± 0.9	94.8 ± 1.4
4-chlorophenol	-	10	98.4 ± 0.3	95.4 ± 1.2	97.1 ± 1.0
Mixed ^b			104.0 ± 2.8	99.6 ± 1.8	93.4 ± 0.8

^aNitrate salts were used.^b1,395 mg L⁻¹ Na⁺, 2,897 mg L⁻¹ Cl⁻, 2,546 mg L⁻¹ NO₃⁻, 250 mg L⁻¹ K⁺, Ca²⁺, Mg²⁺, SO₄²⁻, PO₄³⁻, NH₄⁺, 10 mg L⁻¹ Al(III), Mn(II), Fe(III), Zn(II).

standard deviations (RSD %). Accordingly, the RSD % were found to be 2.1, 3.3, and 3.0% for Cu(II), Cd(II), and Ni(II) ions, respectively, indicating good method precision. Saracoglu *et al.* (2003) developed a co-precipitation procedure utilizing samarium hydroxide as an inorganic coprecipitant. In their study, LODs for Cu(II), Cd(II), and Ni(II) ions were determined as 1.1, 0.4, and 3.1 µg L⁻¹, respectively and RSD % were obtained as <10% for the analyte ions. In a method developed using aluminum hydroxide as a co-precipitating agent, the LODs were determined as 3.0 µg L⁻¹ for Cu(II) and 6.0 µg L⁻¹ for Cd(II) ions (Doner & Ege 2005). In a different method developed for the separation and preconcentration of HMs with the Bi(III)-4-methylpiperidinedithiocarbamate co-precipitation procedure, the RSD % was found to be <10% (Efendioglu *et al.* 2007). In the study in which MEFMAT (an organic co-precipitating agent) was used without using a carrier element, the LODs were determined as 1.49 µg L⁻¹ for Cu(II) and 0.45 µg L⁻¹ for Cd(II) ions and RSD % were found to be lower than 3.5% for both analyte ions (Duran *et al.* 2011). In the CEFC method, in which ICOTMA was used as an organic co-precipitant, the LODs for Cu(II) and Ni(II) were determined as 0.58 and 0.27 µg L⁻¹, respectively (Duran *et al.* 2014). Mendil *et al.* (2015) obtained the LOD and RSD % for Cu(II) ions as 0.80 µg L⁻¹ and 8%, respectively by a CEFC procedure based on 2,9-dimethyl-4,7-diphenyl-1,10-phenanthroline (BCP). In addition, a comparison of the analytical performance parameters of the current CEFC method with other separation and preconcentration methods (solid phase extraction) is given in Table 2. Considering these studies given in the literature, the superiority of the developed method in terms of low LOD and RSD % is remarkable.

3.4. Analysis of water samples

To verify the accuracy and precision of the method, various amounts of analyte ions were spiked into sea and stream waters. The relative recovery (RR %) values for water samples ranged from 90.8 to 97.6% for Cu(II) ions, from 91.8 to 101.0% for Cd(II) ions, and from 91.2 to 98.4% for Ni(II) ions (Table 3). These spike/recovery tests demonstrated that the CEFC method based on CCMBATT is an accurate method, capable of determining analyte ions in water samples without considerable matrix interference. Furthermore, the suggested procedure was successfully applied to assess the levels of Cu(II), Cd(II), and Ni(II) ions in real water samples (Table 4). The maximum tolerable Cu(II), Cd(II), and Ni(II) ion levels in drinking water recommended by World Health Organization (WHO) are 2.0, 0.003, and 0.07 mg L⁻¹, respectively (Braga *et al.* 2017). The concentrations of Cu(II) determined in both stream and sea waters using the developed process were obtained to be lower than the tolerable limits set by the WHO for drinking water. Ni(II) and Cd(II) concentrations were found to be considerably higher than the allowable limit in stream water 4 obtained from the area close to the mining activities. Cd(II) levels are also above the tolerable limit in seawater and stream water 1 samples. It is important to take the required precautions about the

Table 2 | Comparison of the present CEFC system with different solid phase extraction methods in literature

SPE adsorbent	Analyte	LOD ($\mu\text{g L}^{-1}$)	RSD (%)	Ref.
MPAEMA-co-DVB-co-AMPS ^a	Cd	0.9	<2	Daşbaşı <i>et al.</i> (2016)
	Cu	1.9		
Fe ₃ O ₄ @ Diaion HP-2MG resin	Ni	15.0	3–5	Soylak <i>et al.</i> (2023)
SP70-benzoin oxime chelating resin	Cd	4.2	5.3	Narin <i>et al.</i> (2007)
Fe ₃ O ₄ @polythiophene	Cd	1.1	1.7	Sodan <i>et al.</i> (2020)
	Cu	1.4	2.4	
Magnetic Fe ₃ O ₄ <i>Alnus glutinosa</i> sawdust biochar/SiO ₂ /CTAB	Cd	0.62	3.69	Duran & Ozdes (2023)
	Cu	0.24	1.97	
3-mercaptopropyl trimethoxysilane-functionalized graphene oxide–magnetic nanoparticles	Cd	0.9	5.04	Lamaiphan <i>et al.</i> (2021)
Magnetite-coated polyaniline-polythiophene copolymer	Ni	2.8	1.9	Elci (2022)

^aPoly [2-(4-methoxyphenylamino)-2-oxoethyl methacrylate-co-divinylbenzene-co-2-acrylamido-2-methyl-1-propanesulfonic acid].

Table 3 | Spiked recoveries of analyte ions from water samples (Sample pH: 7.0, sample volume: 15 mL, concentration of CCMBATT: 0.07 mM, final volume: 4.0 mL, *N*:3)

Element	Added (μg)	Stream water 1		Stream water 2		Stream water 3		Stream water 4		Sea water	
		Found (μg)	Recovery (%)	Found (μg)	Recovery (%)	Found (μg)	Recovery (%)	Found (μg)	Recovery (%)	Found (μg)	Recovery (%)
Cd(II)	0	ND	–	ND	–	ND	–	ND	–	ND	–
	5	4.76 ± 0.16	95.2	4.59 ± 0.23	91.8	4.93 ± 0.19	98.6	4.73 ± 0.19	94.6	5.05 ± 0.20	101.0
	10	9.45 ± 0.35	94.5	9.60 ± 0.06	96.0	9.29 ± 0.08	92.9	9.36 ± 0.13	93.6	9.75 ± 0.35	97.5
Cu(II)	0	ND	–	ND	–	ND	–	ND	–	ND	–
	12.5	12.0 ± 0.1	96.0	11.9 ± 0.3	95.2	12.2 ± 0.5	97.6	11.8 ± 0.6	94.4	11.6 ± 0.4	92.8
	25	23.9 ± 1.0	95.6	24.2 ± 0.4	96.8	24.3 ± 0.9	97.2	22.7 ± 0.8	90.8	23.5 ± 0.8	94.0
Ni(II)	0	ND	–	ND	–	ND	–	ND	–	ND	–
	12.5	11.8 ± 0.5	94.4	11.4 ± 0.4	91.2	12.0 ± 0.4	96.0	11.7 ± 0.3	93.6	12.3 ± 0.3	98.4
	25	24.4 ± 0.3	97.6	24.3 ± 0.6	97.2	23.3 ± 0.5	93.2	22.8 ± 1.1	91.2	24.2 ± 0.8	96.8

ND, not detected.

Table 4 | Analyte levels of water samples (Sample volumes: 100 mL, final volumes: 4.0 mL, *N*:3)

Element	Liquid samples ($\mu\text{g L}^{-1}$)				
	Sea water	Stream water 1	Stream water 2	Stream water 3	Stream water 4
Cd(II)	4.80 ± 0.05	3.60 ± 0.02	0.70 ± 0.02	2.18 ± 0.08	57.8 ± 1.9
Cu(II)	15.4 ± 0.1	40.9 ± 1.2	7.68 ± 0.18	26.4 ± 1.0	416 ± 9
Ni(II)	1.70 ± 0.01	1.40 ± 0.02	1.06 ± 0.04	3.82 ± 0.22	149 ± 6

Ni(II) and Cd(II) levels in sea and stream water to protect the aquatic environment. Kumar *et al.* (2023a) have reported the Cu(II), Cd(II), and Ni(II) concentrations in the range of 2.12–5.05 $\mu\text{g L}^{-1}$, 0.04–9.86 $\mu\text{g L}^{-1}$, and 1.64–4.63 $\mu\text{g L}^{-1}$, respectively, in water samples collected from East Kolkata Wetlands. During the analysis of water samples collected over a period of 12 months from the Büyük Menderes River, Türkiye, the average concentrations of Cu(II) and Ni(II) were determined to be 3.5 and 24.7 $\mu\text{g L}^{-1}$, respectively (Minareci *et al.* 2018). The levels of Cu(II), Cd(II), and Ni(II) detected in

the Gomti River, India, were found to be significantly higher compared to the findings of our current study. Specifically, the mean concentrations recorded were 0.09 mg L^{-1} for Cu(II), 0.87 mg L^{-1} for Cd(II), and 1.12 mg L^{-1} for Ni(II) (Kumar *et al.* 2023b). According to a study by Baltas *et al.* (2017), the concentrations of Cu(II), Cd(II), and Ni(II) in sea water samples collected from Black Sea, Rize Harbour, Türkiye were reported as 45.0, 2.0, and $7.0 \mu\text{g L}^{-1}$, respectively. Gouda *et al.* (2022) have reported the Cd(II), Cu(II) and Ni(II) levels of sea water obtained from Red Sea, Egypt as 3.8, 12, and $2.7 \mu\text{g L}^{-1}$, respectively. In the same study, although the levels of Cd(II) and Ni(II) ions were below the detection limit in river water collected from Shobra, Egypt, Cu(II) was detected as $8.3 \mu\text{g L}^{-1}$. Indeed, variations in the reported results can arise due to various pollution sources and their intensity. Factors such as industrial activities, agricultural practices, urbanization, and wastewater discharges can contribute to the differences in pollutant concentrations observed in different regions or studies. These variations highlight the importance of considering local conditions and pollution sources when interpreting and comparing research findings related to water pollution.

4. CONCLUSIONS

The quantification of HMs in water samples is crucial for environmental protection, safeguarding public health, complying with regulatory standards, implementing water treatment measures, and identifying pollution sources for effective remediation. Therefore in the present research, a new co-precipitation procedure was introduced utilizing an organic co-precipitant, CCMBATT, without the need for a carrier element. The procedure was successfully employed for separation, preconcentration, and quantification of trace levels of Cu(II), Cd(II), and Ni(II) ions in water samples. Several critical experimental parameters, including pH, CCMBATT concentration, standing time, sample volume, centrifugation time, and rate, were extensively evaluated and optimized to ensure reliable results. Notably, the presence of interfering ions (Na^+ , K^+ , Ca^{2+} , Mg^{2+} , SO_4^{2-} , NO_3^- , PO_4^{3-} , NH_4^+ , Fe(III), Al(III), Mn(II), Zn(II), phenol, and 4-chlorophenol) at high concentrations did not affect the recoveries of the target analyte ions, demonstrating the method's robustness and selectivity. The accuracy of the developed method was assessed through spike/recovery experiments, wherein the recovery percentages of all spiked samples exceeded 90%, indicating excellent accuracy. An essential advantage of this method is the elimination of contamination risks associated with the carrier element, which commonly occurs in conventional co-precipitation procedures. Consequently, the reliability of the analysis for the target analyte ions is significantly improved. Furthermore, the method offers several additional benefits, including its environmentally friendly nature, rapid execution, simplicity, high sensitivity, and exceptional efficiency for monitoring Cu(II), Cd(II), and Ni(II) ions in complex water samples.

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DATA AVAILABILITY STATEMENT

All relevant data are included in the paper or its Supplementary Information.

CONFLICT OF INTEREST

The authors declare there is no conflict.

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