Original Research Article

Separation, Preconcentration and determination of Hg (II) Ion in Water Samples by Cloud Point Extraction Technique Coupled with UV-VIS Spectrophotometry using a New Complexing Agent

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ABSTRACT

In this study a cloud-point extraction process using non-ionic surfactant Triton X-114 for extraction of mercury (II) from aqueous solutions was developed. The method is based on the complex formation of Hg (II) with benzyl (pyridin-2-yl) methylcarbamodithioate at pH 5.5. After preconcentration and dilution of the surfactant-rich phase, the enriched analyte was determined by spectrophotometer. The effects of different parameters such as concentration of surfactant, electrolyte, temperature and pH on the cloud point extraction were studied in detail and a set of optimum conditions were established. Under the optimum conditions, the calibration graph was linear in the range of 0.25–3 µg mL^{-1} with detection limit of 0.02 µg mL^{-1}. The precision (R.S.D. %) for 1 µg mL^{-1} of Hg (II) was 1.6% (n=5) and the preconcentration factor was found to be 20. Under the presence of foreign ions, no significant interference was observed. Finally, the proposed method was applied successfully for the determination of Hg (II) in real water samples.

Introduction

Heavy metals are of important interest because of having high toxic effects on living organisms so that more stringent regulations have been imposed for controlling their contamination in the environment in recent years [1]. The pollution of natural waters by heavy metals is a great concern nowadays due to their potentially toxic effects on living organisms. Urbanization, industrial development, and heavy traffic lead to contamination of water bodies by heavy metals [1]. One of these toxic metals is mercury, finding in low levels in natural waters in different physical and chemical forms. Mercury can cause birth defects and other undesirable effects on the human body [2-7]. It is a highly toxic element that is found both naturally and as an introduced contaminant in the environment [8,9]. Mercury has no beneficial biological function, and its presence in living organisms is associated with cancer, birth defects, and other undesirable outcomes [10,11]. The United States Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA) lists Hg and its compounds in third place on the ''Priority List of Hazardous Substances'' and the European Water Framework Directive (2000/60/EC) classifies Hg as one of 30 ''precarious dangerous pollutants'' [12]. Thus, the development of new methods for selective separation, preconcentration and determination of this metal ion in different matrices is of continuing interest. In this manner many methods are still commonly carried out using a preconcentration step to overcome the problems resulting from the low concentration levels of analytes and high levels of matrices prior to their detection [13,14].

A large number of such preconcentration techniques in recent years for mercury have been developed, including liquid–liquid extraction (LLE) [15,16], coprecipitation [17,18], microextraction [19], solidphase extraction (SPE) [20–29] and cloud point extraction (CPE)[30–35]. Cloud point extraction is based on the fact that most nonionic surfactants in aqueous solutions form micelles and become turbid when heated to the cloud point temperature or in the presence of an electrolyte. Above the cloud point, the micellar solution separates into a surfactant-rich phase with a small volume and a diluted aqueous phase [36]. In comparison with other enrichment techniques, it has large preconcentration factor and higher recovery efficiency [37]. CPE has become one of the most preferred preconcentration methodologies, which was used as a preconcentration method in simultaneous determination of some metal ions [38]. Compounds such as benzyl (pyridin-2-yl) methylcarbamodithioate (Fig 1) can act as chelating agents for metal ions by bonding through the sulfur or oxygen atom and the nitrogen atom.
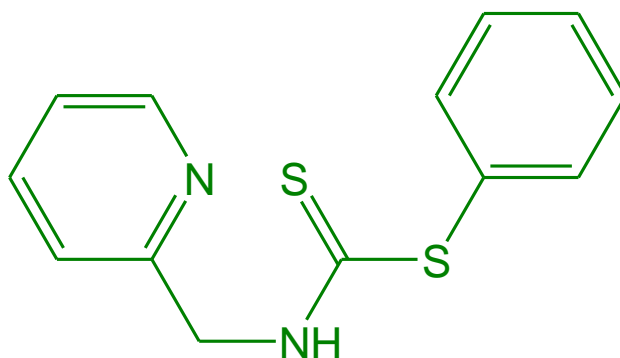

Fig. 1. Chemical structure of benzyl (pyridin-2-yl) methylcarbamodithioate

In this work, after preliminary spectrophotometric studies of mercury (II) interaction with benzyl (pyridin-2-yl) methylcarbamodithioate as a new chelating agent, we report a cloud-point extraction process coupled with UV-Vis spectrometer for preconcentration and determination of Hg^{2+} from water samples. It is important to say neither spectrophotometric studies on the complex formation of this ligand with Hg^{2+} ion nor the cloud point procedure have been reported

in the literature as being used for this mercury complex extraction, in fact both are reported for the first time.

Experimental

Materials

All the chemicals and reagents were of the highest purity available and used as received. The solutions were prepared with triply distilled water. The nonionic surfactant Triton X-114 was from Sigma–Aldrich (St. Louis, MO, USA). Standard solution of mercury prepared by serial dilution of 1000 mg L^{-1} standards (Fluka A.G., Buchs, Switzerland) with water. Other chemicals used in this study were obtained from Merck (Darmstadt, Germany). benzyl (pyridin-2-yl) methylcarbamodithioate was synthesized according to the literature [39-40]. A stock solution of this ligand (8×10^{-4} mol.L^{-1}) was prepared by dissolving an appropriate amount of it in methanol.

Instruments

A model lambda 25 double beam UV–Vis spectrophotometer (PerkinElmer Inc., USA) with a 1 mL quartz cells was used for recording all spectra and absorbance measurements. A Metrohm 827 pH meter (Metrohm A.G., Herisau, Switzerland) equipped with a glass electrode was used for pH measurements. A Julabo model F12 water bath (Julabo Labortechnik GmbH, Germany) with ±0.1°C temperature control was used for cloud point extraction experiments and a centrifuge with 10 mL calibrated centrifuge tubes (Superior, Germany) was utilized to accelerate the phase separation process.

General CPE procedure

An aliquot of Hg (II) solution was transferred to a biker (20 mL) and 0.5 mL of a 8×10^{-4} mol L^{-1} ligand solution was added. The mixture was kept at room temperature for 30 min then; 2.5 ml of Triton X-114 (1% v/v) and 4 mL of triply distilled water were added. The pH of solution was

adjusted at 5.5 and transferred to a 10 mL centrifuge tube. Then the solution was up to the mark (10 mL) with triply distilled water. Subsequently, it was shaken and left to stand in a thermostatted water bath for 10 min at 55°C. Separation of the aqueous and surfactant-rich phase was accelerated by centrifugation for 12 min at 3500 rpm. After cooling in a salt-ice bath (10 min), the surfactant-rich phase became viscous. Then the aqueous phase was carefully removed with a pipette and 0.5 mL methanol was added to the surfactant-rich phase. The content of tube was transferred to a 1 mL quartz cell for an absorbance measurement at 330 nm.

Preparation of water samples

Water samples (tap, deionised and riever water) were filtered using a 0.45 μm pore size membrane filter to remove any suspended particulate matter and stored in a refrigerator in the dark prior to analysis. Aliquots of water (5 mL of each) samples were subjected to the CPE methodology described above.

RESULTS AND DISCUTION

Preliminary Complexation Studies

In order to obtain some information about the stoichiometry and stability of the Hg (II)-Ligand complex, in preliminary experiments, the complexation of ligand with this cation was investigated spectrophotometrically in methanol solvent. In this manner, a solution containing a constant concentration of ligand (5×10^{-5} mol L^{-1}) was titrated with a methanol solution of Hg (II). The resulted spectrum is given in Figure 2.

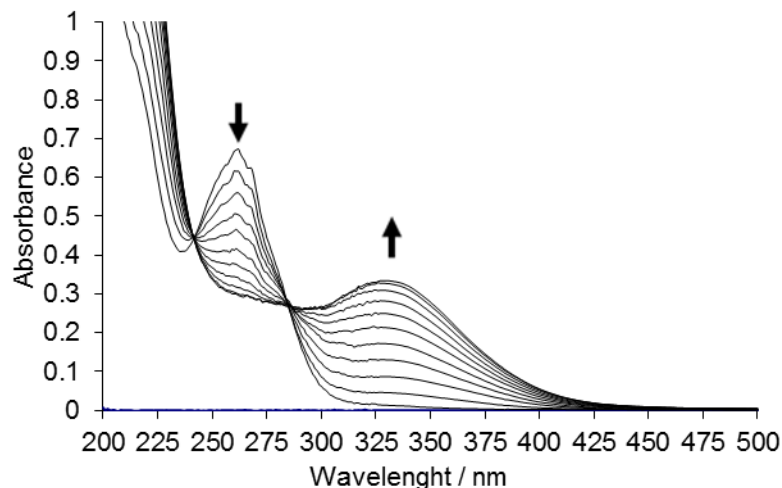**Fig. 2.** Absorption spectra of benzyl (pyridin-2-yl) methylcarbamodithioate (5×10^{-5} mol L^{-1}) in Methanol solution at 25°C in the presence of Hg^{2+} (0-1.6×10^{-4} mol L^{-1})

As it is obvious, an increase in the absorption band at 325 nm and a decrease in the absorption band at 263 nm were apparent as the concentration of complex is increased and the concentration of free ligand is decreased. In order to study the stoichiometry of complex Job method at 325 nm was carried out in 0.0 to 1.0 mole fractions of Hg (II). Fig. 3 illustrates absorbance of different Hg (II) mole fractions in complex and as can be seen, maximum absorbance obtained in mole fraction of 0.5 which indicates resulted complex has 1:1 (metal-to -ligand) stoichiometry.
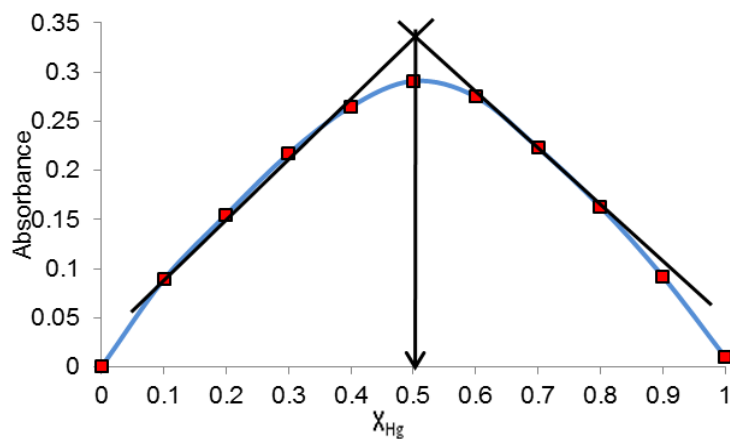

Fig. 3. Variations of Hg (II) complex absorbance in different Hg (II) mole fractions (325 nm)

The formation constant (Log k) of the resulting complex was evaluated 4.8, by computer fitting of the absorbance–mole ratio data using KINFIT [41] at the same wavelengths.

Method development

At first and before analysis of standards and real samples, different variables, which affect the absorption intensity of extracted Hg, were optimized using a standard solution of this element.

Optimization of the system

Effect of pH

The separation of metal ions by cloud point method involves prior formation of a complex with sufficient hydrophobicity, which can be extracted by surfactant micelles. pH plays a unique role on metal-chelate formation and then extraction. So it was the first variable optimized in the extraction procedure. Fig. 4 shows the effect of pH on the Hg recoveries. As can be seen, higher recoveries were achieved at pH range of 5.5–7.5. Therefore a pH value of 5.5 was selected as optimum value. At higher pHs, absorbance decreases sharply because of mercury hydroxide formation. In lower pHs, complex formation is decreased because the ligand is protonated.
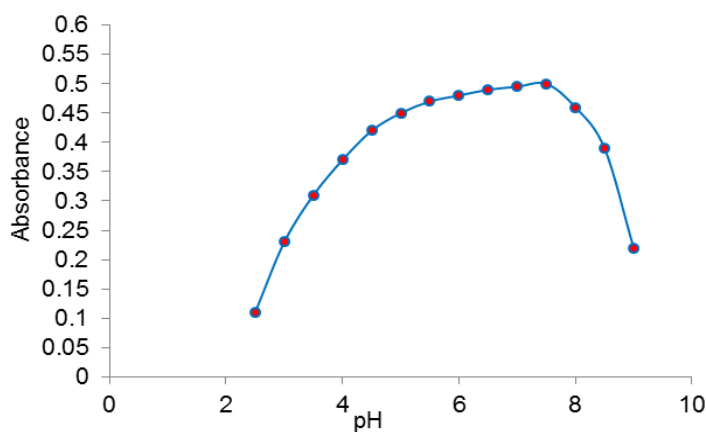

Fig. 4. Effect of pH on the preconcentration of 10 mL of a Hg solution (1.5 μg mL^{-1}). Condition: Ligand, 4×10^{-5} mol L^{-1}; Triton X-114, 0.25% (v/v); equilibrium temperature, 55°C for 10 min; centrifugation time, 12 min in 3500 rpm.

Effect of ligand concentration

The extraction efficiency as a function of the ligand concentration is shown in Figure 5. For this study, 10 mL of a solution containing Hg ion with various amounts of ligand ((3-12) $\times 10^{-5}$ mol L^{-1}) was subjected to the CPE method.
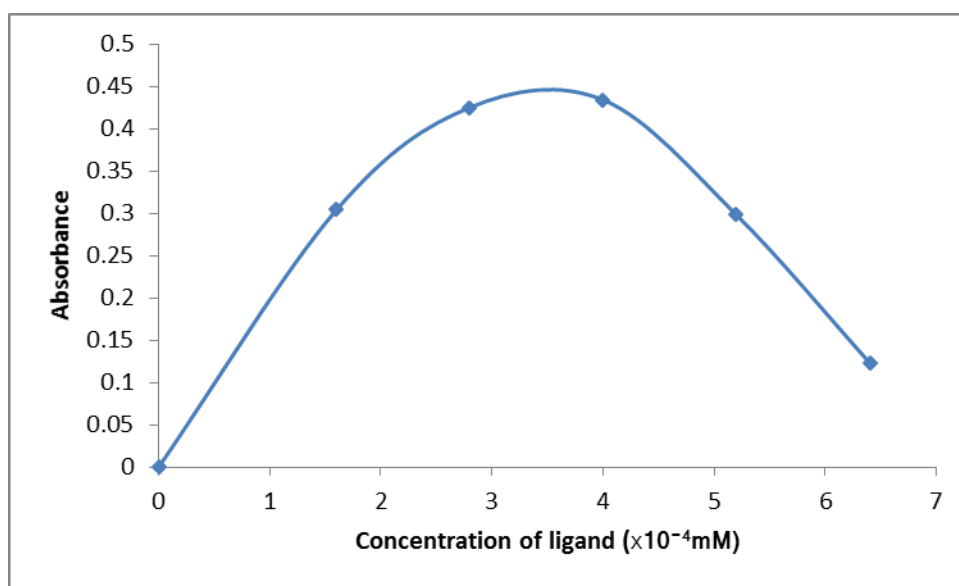

Fig. 5. Effect of ligand concentration on the preconcentration of 10 mL of a solution containing Hg(II). Condition: pH = 5.5; Triton X-114, 0.25% (v/v); equilibrium temperature, 55°C for 10 min extraction time; centrifugation time, 12 min in 3500 rpm.

The results revealed that the extraction recovery increases by increasing ligand concentration up to 4×10^{-5} mol L^{-1} and remained nearly constant at higher concentrations. Therefore, this concentration was selected as optimum value.

Effect of Triton X-114 concentration

A successful cloud point extraction should be able to maximize the extraction efficiency through minimizing the phase volume ratio (V surfactant-rich phase/V aqueous phase), so as to improve the preconcentration factor. Triton X-114 was chosen as the non-ionic surfactant due to its lower

cloud point temperature (22–25°C) and high density of the surfactant-rich phase, which facilitates phase separation by centrifugation. The effect of Triton X-114 concentration on the metals recovery percent in the range of 0.05–0.5% (v/v) was investigated (Fig. 6).
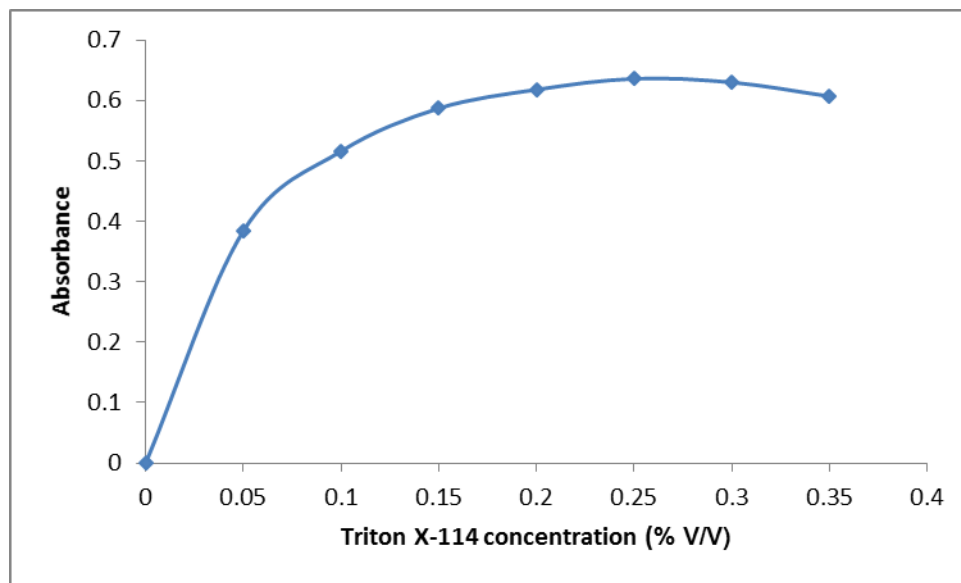

Fig 6. Effect of Triton X-114 concentration on the preconcentration of 10 mL a solution containing mercury. Condition: pH = 5.5; equilibrium temperature, 55°C for 10 min extraction time; centrifugation time, 12 min in 3500 rpm; ligand, 4×10^{-5} mol L^{-1}.

The concentration of 0.25% (v/v) was chosen as optimum for the quantitative recovery of complexes. At lower concentrations, the extraction efficiency of complexes is low, probably because of the inadequacy of the assemblies to entrap the hydrophobic complexes quantitatively. Therefore, it was chosen as an optimum value.

Effect of ionic strength

The cloud point of micellar solutions can be controlled by addition of salts, alcohols, non-ionic surfactants and some organic compounds (salting-out effects). To date, most of the studies conducted have shown that ionic strength has no appreciable effect on the extraction efficiency

[42]. It was observed that the addition of inorganic salts including NaCl, KNO_3 and Na_2SO_4 in the range of 0–2 mol L^{-1} had no significant effect on the cloud point extraction efficiency.

Effects of the equilibration temperature, equilibration time and centrifugation time

The dependence of metals recoveries percent upon equilibration and incubation times was studied within the range of 5–30 min. Time of 10 min was chosen as optimal time for completion of the clouding process and analyte extraction. It was also observed that a temperature of 55 ˚C is sufficient for the maximum recovery of the analyte. Centrifugation time hardly ever affects micelle formation but accelerates phase separation in the same sense as in conventional separation of a precipitate from its original aqueous environment [43]. A centrifuge time of 8 min was selected as optimum in further experiments.

Selectivity studies

Although preliminary spectrophotometric studies of ligand with some metal ions (Cd^{2+}, Co^{2+}, Cr^{3+}, Cu^{2+}, Mg^{2+}, Ni^{2+}, Sn^{2+}, Zn^{2+}, La^{2+}, Ca^{2+}, Mn^{2+}, Ag^+, Tl^+ and Ga^{3+}) did not show any interaction other than mercury and very negligible with silver but some more investigation about selectivity was carried out. In this manner, a series of solutions (10 mL0) containing analyte ion and different concentrations of interfering ions were subjected to the complete procedure. The results were in agreement with those obtained from spectrophotometric studies. Therefore, the mercury recovery was almost quantitative in the presence of other ions.

Analytical characteristics

Table 1 summarizes the analytical characteristics of the optimized method, including the regression

equation, linear range, limit of detection, reproducibility and preconcentration factor. The limit

of detection which defines as $C_L=3S_B/m$ (where C_L, S_B, and m are the limit of detection, standard

deviation of the blank, and the slope of calibration graph, respectively) and preconcentration

factor was calculated to be 0.02 µg mL^{-1} and 20 respectively.

Table 1. Analytical characteristics of the proposed method

Reproducibility (%RSD)	1.6
Linear range (µg mL^{-1})	0.25 – 3
Regression equation (n=6) (C, µg mL^{-1})	y = 0.0375x − 0.0003
Correlation coefficient (R^2)	0.9993
Limit of detection (µg mL^{-1})	0.02
Preconcentration factor	20

Application

In order to evaluate the analytical applicability of the proposed method, it was applied to the

determination of mercury in real water samples. The results are given in Table 2. The recoveries

for the addition of different concentrations of mercury to water samples 97% - 103%. The results

showed that the proposed method could be successfully applied to the determination of mercury

in water samples.

Table 2. Determination of mercury in water samples by proposed method

Sample	Added (mg L^{-1})	Found (mg L^{-1})	Recovery (%)
River water	0	0.38	-
	0.1	0.10	102.1
	0.5	0.49	97.18
	1	1.04	103.35
	1.5	1/50	100/2
	2	1/95	97/25
Distillated water	0	0	-
	0.1	0.10	98
	0.5	0/49	97/6
	1	0/99	98/7
	1.5	1/47	97/8
	2	2/00	100/2
Tap water	0	0.00	-
	0.1	0.10	103.5
	0.5	0.50	99/9
	1	1.01	101/2
	1.5	1.48	99.0
	2	1.99	99.9

Conclusions

Spectrophotometric studies on benzyl (pyridin-2-yl) methylcarbamodithioate indicated that it forms a complex with Hg (II) having a stoichiometry of 1:1. Based on this complex formation, a cloud point extraction method coupled with UV-Vis spectrometer was developed for the separation, preconcentration and determination of this cation at μg L^{-1} levels. The proposed method provides a good reproducibility and gives a precise and sensitive procedure for the determination of mercury that is applicable to the water samples containing trace amounts of this element. It is important to say neither Spectrophotometric studies on the complex formation of this complexing reagent with $Hg2^{+}$ ion nor the cloud point procedure have been reported in the literature as being used for mercury determination, in fact both are reported for the first time.

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