Methods

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PAPER

Micellar mediated trace level mercury quantification through the rhodamine B hydrazide spirolactam ring opening process

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The micellar mediated trace level estimation of mercury in industrial effluent sample by the rhodamine B hydrazide spirolactam ring opening process is described. The catalytic reaction of mercuric ion with colorless rhodamine B hydrazide to form pink colored rhodamine B through amide bond cleavage at pH 5 forms the basis of this reaction. The dye obtained was extracted from the aqueous phase into non-ionic surfactant using Triton X-114 through cloud point formation and its absorbance was measured at 556 nm. The reaction conditions such as concentrations of rhodamine B hydrazide and Triton X-114, effect of NaCl concentration on cloud point temperature and the efficiency of extraction were optimized. The selectivity of the method towards mercury in the presence of various ions was studied. The validity of the method was examined by spiking real samples with known amounts of mercury and carrying out recovery studies. The results obtained by the proposed method were compared with the standard dithiazone method. The preconcentration and enrichment factors were found to be 5 and 12 respectively. The limit of detection and the linear range were found to be 1.4 ng ml^{-1} and 10–100 ng ml^{-1} respectively. The relative standard deviation was found to be 0.35% at 20 ng ml^{-1}.

Introduction

In recent years there has been an increased awareness amongst the general public about heavy metal pollution in water and air.[1] The toxicity of heavy metals like Pb, Cd, Hg, As and Cr in water bodies has been a very serious concern in recent years due to the excessive discharge of untreated industrial effluents. The threshold limit values of these metal ions are at ppb levels. Amongst these metals, elemental mercury is of particular interest because of its high toxicity, its reactivity, its extreme volatility and its relative solubility in water and living tissues. It causes severe damage to kidneys, and to the digestive and neurological systems.[2] The extreme toxicity of mercury and its derivatives results from its high affinity for thiol groups in proteins and enzymes leading to the dysfunction of cells and consequently causing health problems. Mercury ion can easily pass through the skin, respiratory and gastrointestinal tissues into the human body and can damage the central nervous and endocrine systems.[3] Mercury generally adopts one of three common forms: elemental mercury (Hg^0), ionic mercury (Hg^{2+}, Hg^+) and organo mercury which includes methyl mercury, dimethyl mercury, ethyl mercury and phenyl mercury *etc*. Methyl mercury (CH_3Hg) is the most toxic of all the forms of mercury in living systems.[4] Depending on environmental conditions, mercury can transform into other forms so the existence of any form of mercury in water is potentially harmful to human health.

Average mercury levels in the atmosphere are 3–6 fold higher than the pre-industrial estimates.[5–7] Due to its interesting physical and chemical properties mercury has been used in various industrial sectors like catalysis, amalgams, electrodes, lamps, batteries, thermometers, fungicides and pigments.[8] Other sources of mercury include coal mining, gold production, non-ferrous metal production, cement production, waste disposal, human crematoria, caustic soda production, pig iron and steel production.[8] Industrial processes tend to release geologically bound mercury from mercury reservoirs into the atmosphere as elemental mercury.[9] Once introduced into the environment this ion takes part in many transformations, transportation and bioassimilation processes.[10]Mercury enters into the food chain from contaminated water bodies in the vicinity of the industries where mercury has been used. The various species of fish in these water bodies accumulate mercury and then convert it to its most toxic form, methyl mercury.[11] It can be transferred into human beings and other living organisms through food consumption which results in biomagnification. According to the Environmental Protection Agency (EPA), USA, the maximum allowable level of ionic mercury in drinking water is 2 ppb.[11] In order to monitor the mercury levels at ultra trace concentrations, highly sensitive and selective methods are required for its quantification.

Although sophisticated analytical techniques, like AAS, AFS and ICP, are currently used to determine trace levels of mercury, they require expensive equipment and time consuming sample

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preparation procedures.[12–14] Therefore, there is a strong demand for inexpensive and real-time monitoring methods for the detection of mercury at trace levels. In this context, methodology based on the use of fluorescent and colorimetric chemosensors has attracted considerable attention in recent years due to their high sensitivity, selectivity, low cost and easy detection.[15] Several reagents have been used in the design of fluorescent and colorimetric chemosensors for the estimation of mercury.[16–19] Among them only a few are competitive in terms of sensitivity, selectivity and simplicity. Hence there is plenty of room for developing new chemosensors for the Hg^{2+} ion. Rhodamine is an ideal molecule for fluorescent as well as colorimetric probes because of its excellent spectroscopic properties such as large molar extinction coefficient, high fluorescence quantum yield and long absorption and emission wavelengths. It is well-known that, rhodamine derivatives with the spirolactam structure are colorless and non-fluorescent. Spirolactam ring opening results in a strong fluorescence emission molecule which has been used for the design of light "off-on" fluorescent probes and sensors.[20–27] Kim *et al.* have recently reported on the use of rhodamine B hydrazide as a chemo dosimeter for mercury quantification. In this method colorless and non-fluorescent rhodamine B hydrazide on reaction with mercury ion forms a highly fluorescent rhodamine B through the spirolactam ring opening process.[28] Measurement of the fluorescence of this compound can be correlated to the mercury ion concentrations in semi-aqueous media *i.e.* water–acetonitrile. Rhodamine B has been determined spectrophotometrically at trace level through cloud point extraction using Triton X-100 as a non-ionic surfactant by Pourreza *et al.*[29] In this method the cloud point formation takes place at 78 °C and an incubation time of 30 min. is required to get the maximum extraction efficiency. The cloud point extraction method is very useful for concentrating analytes with high recovery and high preconcentration factors.[30] It has been shown to be an effective sample preconcentration technique for improving the sensitivity and selectivity prior to flame AAS, HPLC, capillary electrophoresis and flow injection analysis.[31–34] Application of surfactant not only facilitates sensitivity enhancement but also avoids the use of toxic solvents in absorbance measurements. This aspect means that it is in agreement with "Green Chemistry" principles.[35]

In this report we have proposed a simple procedure for the quantification of mercury at ppb levels from aqueous media through cloud point extraction at room temperature using rhodamine B hydrazide as a reagent and Triton X-114 as a non-ionic surfactant for phase separation. The rhodamine B obtained was extracted into the micellar phase and its absorbance was measured at 560 nm. The method is highly sensitive and it has been applied to measure ultra trace level mercury in industrial effluents.

Experimental

Apparatus

Absorbance measurements were made using a Shimadzu Scanning Spectrophotometer (model UV-3101PC) with 1cm quartz cuvettes. A centrifuge (REMI, Mumbai, India) with 15 ml calibrated tubes were used to accelerate the phase separation. All pH measurements were carried out using a Control Dynamics digital pH meter (model APX 175). All the infrared spectroscopic measurements were performed using a Shimadzu Spectrometer (model FTIR-8400S). NMR spectra were recorded using a Bruker-400 MHz spectrometer with chemical shifts reported as parts per million (ppm in DMSO d_6, TMS as internal standard). Mass spectral data was obtained using a Thermo Finnigin Deca QXP Mass Spectrometer.

Reagents and solutions

Rhodamine B hydrazide was purchased from Sigma-Aldrich. 1 mM of rhodamine B hydrazide was prepared by dissolving 0.0456 g in 100 ml of 1 : 10 ethanol : water. Standard Hg^{2+} solution of 1000 ppm was prepared by dissolving 0.1796 g of $HgBr_2 \cdot 2H_2O$ (Merck, Mumbai, India) in 1 : 10 ethanol : water. Working solutions were prepared by appropriate dilution on the day of use. 1 M NaCl (Merck, Mumbai, India) solution was prepared by dissolving 4 g in 100 ml of distilled water. 2% of Triton X-114 (Acros Organics, New Jersey, USA) was prepared by dissolving concentrated solution in hot distilled water. Potassium bromide FTIR grade (Sigma–Aldrich, purity 99%) was used for IR spectroscopy. Buffer solutions of pH 3–6 were prepared using sodium acetate and acetic acid and adjusting the pH by using dilute HCl or NaOH. Industrial waste water samples (chrome plating and Textile dyeing industry) were obtained from the Karnataka State Pollution Control Board, Bangalore, India.

Procedure

Aliquots of standard Hg^{2+} solutions (so that the final concentration would be in the range 10–100 ng ml^{-1}) were transferred into 10 ml volumetric flasks. To this 0.4 ml of 10 mM rhodamine B hydrazide solution and 1 ml of pH 5 buffer solution were added. The solution was left for 2 min for the development of a pink color then 2 ml of 2% Triton X-114 and 2 ml of 1 M NaCl were added and the solution was made up to the mark. The solution was transferred to a centrifuge tube and centrifuged for 10 min at 5000 rpm. The tube was then cooled in an ice bath in order to increase the viscosity of the surfactant phase so that the aqueous phase could be easily separated by decanting. The surfactant phase was dissolved in 2 ml of 1 : 10 ethanol : water to give a homogenous phase and the absorbance was measured at 556 nm. (Fig. 1)

Sample pretreatment

Industrial effluent from the chrome plating and textile industry were collected in polyethylene containers. The solutions were filtered and 50 ml of filtered solution was transferred into a beaker, 10 ml of concentrated sulfuric acid and 10 ml of 30% H_2O_2 were added and the solution was then heated in a water bath until the foaming ceased in order to oxidize any elemental and mercurous ions to mercuric ions.[36] Then the solutions were cooled and known aliquots were used for the analysis by the proposed as well as the standard method.[37]

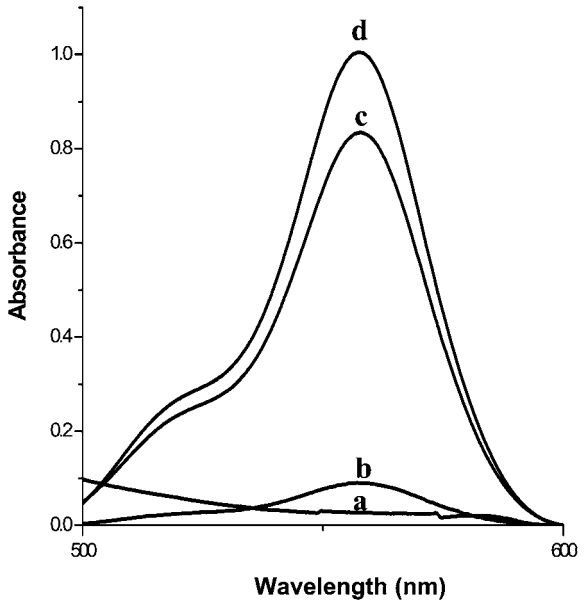
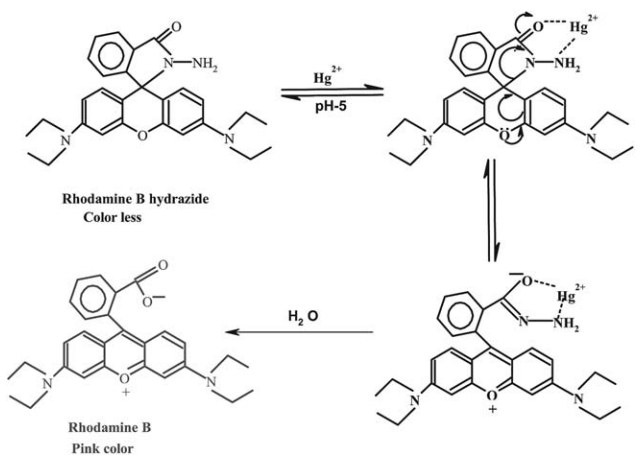


Fig. 1 UV-Vis absorption spectra of (a) rhodamine B hydrazide (reagent blank–aqueous phase) (b) rhodamine B hydrazide in the presence of Hg^{2+} (aqueous phase) (c) rhodamine B hydrazide in presence of Hg^{2+} (micellar phase) (d) commercially procured rhodamine B (micellar phase).

Results and discussion

Mercury ion induces the ring opening of colorless rhodamine B hydrazide to form a pink colored rhodamine B in stoichiometric quantities. This is due to the fact that in the presence of mercury ion hydrolysis of the amide bond occurs forming the basis of the spirolactam ring opening process of rhodamine B hydrazide (Scheme 1).[28] The stoichiometric mole ratio of the reaction between Hg^{2+} ion and rhodamine B hydrazide was obtained by a Job plot and was found to be 1 : 1. The binding constant K of the rhodamine B hydrazide–Hg^{2+} complex was calculated according to the spectroscopic method and was found to be 1.6×10^3.[38] Hydrolysis of the amide bond assisted the ring opening of colorless rhodamine B hydrazide to form a pink colored rhodamine B in the case of mercury in a similar manner to that of copper.[19] The rhodamine-B obtained from the above process is hydrophobic in nature, hence it can be preconcentrated using Triton X-114 as a micellar medium at room temperature. The rhodamine-B obtained was subjected to cloud point extraction before its spectrophotometric measurement. The use of non-ionic surfactant like Triton X-114 in place of Triton X-100 not only facilitates the cloud point extraction at room temperature but also enhances the sensitivity of the method more than nine times over an aqueous procedure.



Scheme 1 Mechanistic pathway of rhodamine B formation by Hg^{2+} ions through the spirolactam ring opening process of rhodamine B hydrazide.

Evidence for the spirolactam ring opening

The catalytic activity of Hg^{2+} ion on the colorless rhodamine B hydrazide to transform it into a pink colored rhodamine B through the spirolactam ring opening process was confirmed by UV-Vis, FTIR, NMR and mass spectral studies.

UV-Vis study

Rhodamine B hydrazide is colorless and doesn't absorb in the visible region. When it was treated with Hg^{2+} ions a pink colored compound was obtained with an absorption maximum at 560 nm. Then commercially procured rhodamine B absorption spectrum were recorded and the spectrum obtained was compared with the absorption spectrum of the compound formed by the catalytic activity of mercuric ions with rhodamine B hydrazide. The two absorption spectra match exactly and both of them have the same $\lambda_{max,}$ *i.e.* 560 nm (Fig. 1c and 1d). This study clearly indicates that mercury quantitatively reacts with rhodamine B hydrazide to form a pink colored rhodamine B compound (Scheme 1).

FTIR study

The pink colored rhodamine B compound formed by the catalytic reaction between rhodamine B hydrazide and mercuric ions was separated by evaporating the solution in a water bath. The solid obtained was recrystallized with methanol and the FTIR spectrum was recorded through a KBr disc. The IR spectra of rhodamine B hydrazide was compared with the IR spectra of rhodamine B which was extracted from the solution. Strong stretching frequencies at 3500 and 1690 cm^{-1} were observed in the IR spectra of rhodamine B hydrazide which are due to N–H stretching of NH_2 (primary amine) and NCO (amide) stretching respectively. However these stretching frequencies were not present in the spectra of rhodamine B that was extracted from the reaction mixture. A strong OH stretching frequency corresponding to COOH at 3500 cm^{-1} was observed (Fig. 2a and 2b). This study shows that mercuric ions have catalyzed the rhodamine B hydrazide to form rhodamine B which is a pink colored compound (Scheme 1). The color intensity of the rhodamine B formed is directly proportional to the mercuric ion concentration.

^{1}H NMR study

The transformation of colorless rhodamine B hydrazide to pink colored rhodamine B through the spirolcatam ring opening process by the catalytic activity of mercuric ions was confirmed by ^{1}H NMR studies. After treating rhodamine B hydrazide with Hg^{2+} ions at pH 5, the pink colored solution was evaporated to

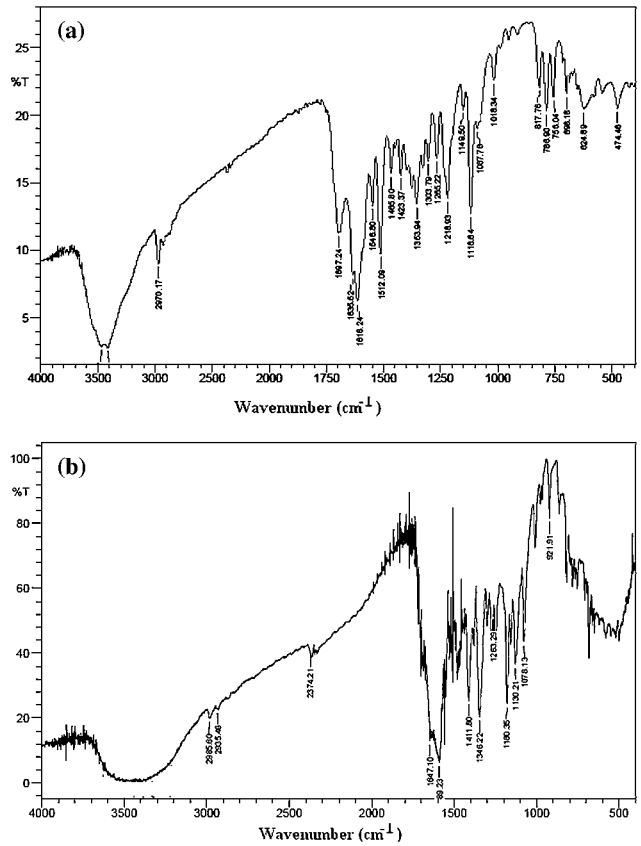


Fig. 2 FTIR spectra of (a) rhodamine B hydrazide and (b) rhodamine B generated by the spirolactam ring opening process.

dryness and the experiment was repeated several times to get a substantial quantity of the rhodamine B compound. The residue was dissolved in a minimum quantity of methanol and recrystallised. The compound obtained was dissolved in DMSO before recording the ^{1}H NMR spectrum (Fig. 3b). The obtained spectrum was compared with the spectrum of authentic rhodamine B compound (Fig. 3c).

The peaks at 1.17 ppm and 3.4 ppm are due to diethyl amine groups of rhodamine B hydrazide (Fig. 3a). However these peaks are slightly shifted to 1.24 ppm and 3.62 ppm when rhodamine B hydrazide transformed into rhodamine B compound (Fig. 3b).The peak at 6.6 ppm is due to the xanthene group of rhodamine B hydrazide which gets slightly shifted to 7.3 ppm when it transforms into rhodamine B by the catalytic activity of mercury ion. The peak at 7.01 ppm is due to the lactam ring of the rhodamine B hydrazide which shifts to 7.85 ppm during transformation. The spectrum of the authentic rhodamine B compound (Fig. 3c) exactly matches that of the rhodamine B compound formed during the reaction. All these observations reveal that rhodamine B hydrazide undergoes transformation to rhodamine B in the presence of mercury ion under aqueous conditions at pH 5.0 through the spirolactam ring opening process.

Mass study

Rhodamine B generated through the spirolactam ring opening process was examined by electron spray ionization mass

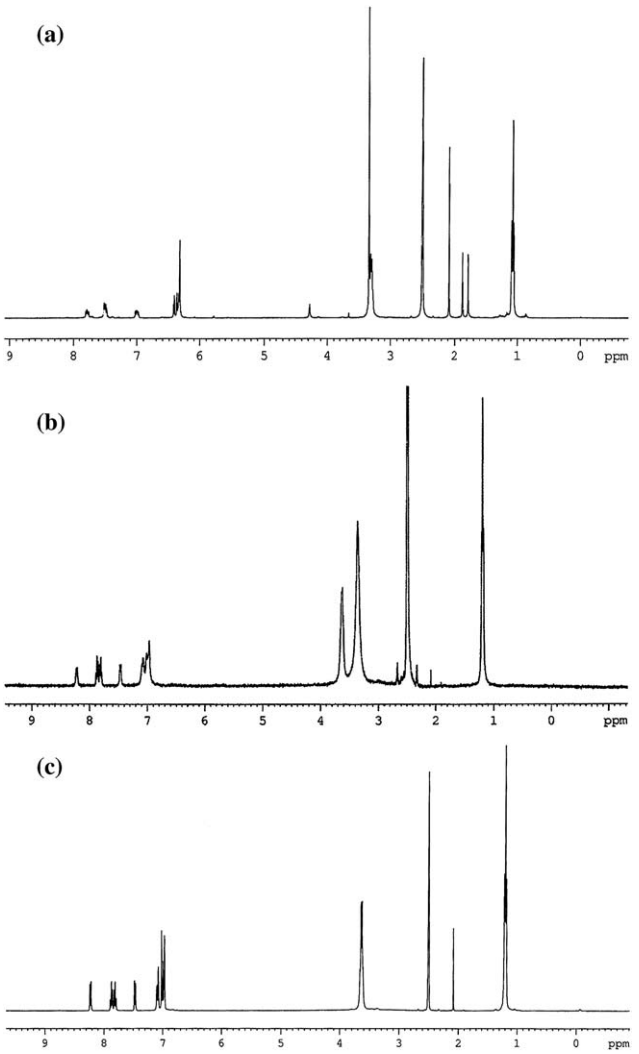


Fig. 3 ^{1}H NMR spectra of (a) rhodamine B hydrazide, (b) rhodamine B generated in the reaction and (c) rhodamine B commercially procured.

spectrometry (ESI-MS) to determine whether or not the reaction product is the same as the authentic sample. The appearance of a molecular ion peak at $m/z = 443.47$ $[M + H]^+$ and the disappearance of the rhodamine B hydrazide peak at $m/z = 4577.6$ $[M + H]^+$ in the generated rhodamine B revealed that the rhodamine B hydrazide transforms into rhodamine B when it is treated with mercury ionic solution at pH 5 (Fig. 4a and 4b).

Optimization study

Initial studies were done by extracting the rhodamine B, obtained by treating rhodamine B hydrazide with mercury ions in aqueous solutions at pH 5, into non-ionic surfactant phase Triton X-100. In order to separate the aqueous and surfactant phases the solution was heated to 70 °C in the presence of 1 M NaCl as a salting out agent. We then tried to extract rhodamine B into the non-ionic surfactant phase Triton X-114, the cloud point temperature of this surfactant is at room temperature therefore heating was not required. In order to quantify trace levels of mercury in natural water samples all the parameters influencing

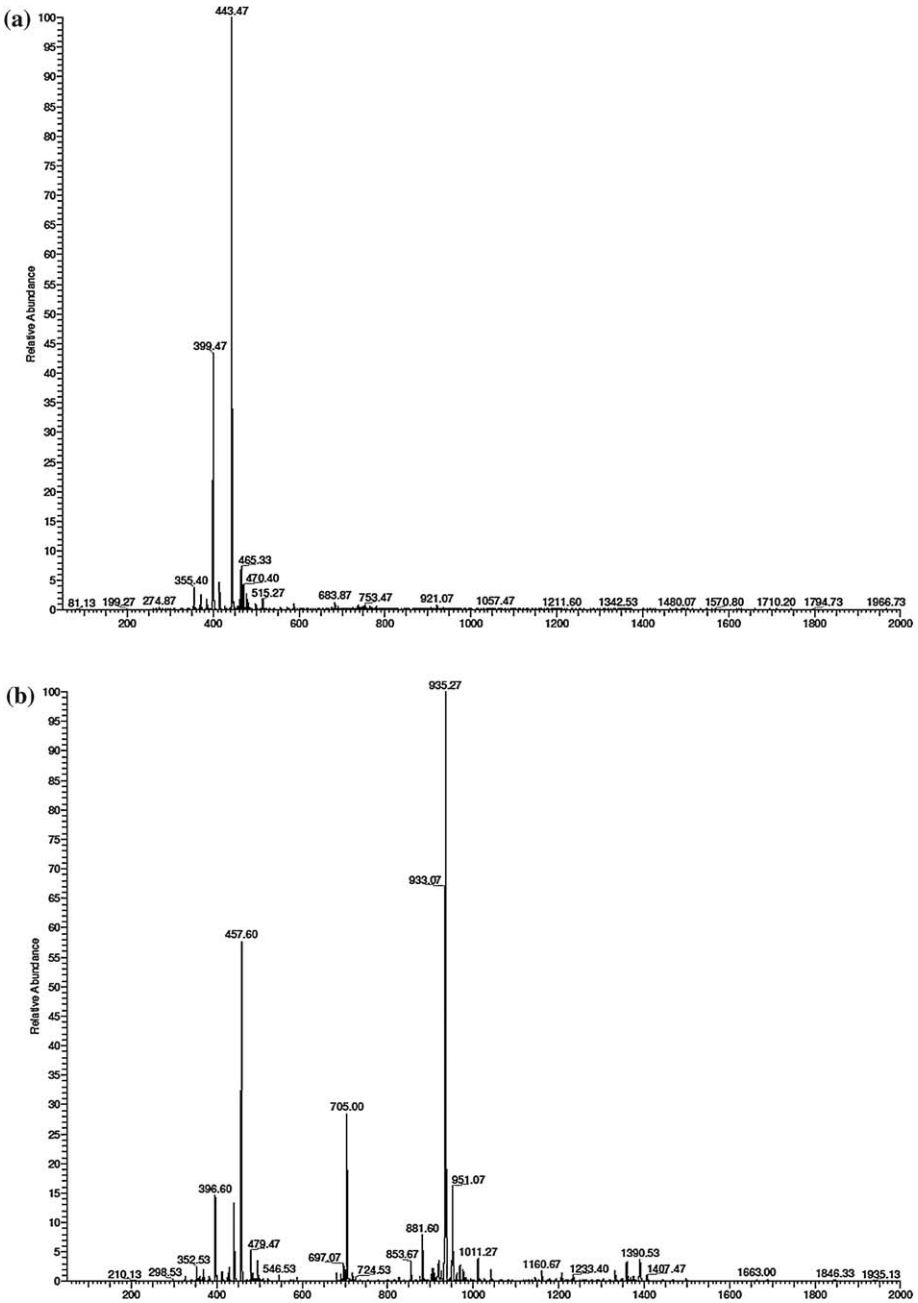


Fig. 4 ESI-MS of (a) rhodamine B generated in the reaction and (b) rhodamine B hydrazide.

the complex formation between the ligand and the metal ion, and the factors affecting the cloud point extraction like Triton X-114 concentration, NaCl as salting agent and centrifugation time were studied. The reaction variables like ligand concentration, pH, reaction time, surfactant concentration, salting agent and centrifugation time were optimized to achieve maximum absorbance for the sample and minimum absorbance for the blank.

Effect of rhodamine B hydrazide

The cloud point extraction efficiency depends on the hydrophobic nature of the complex formed between the analyte and chelating agent. In this case the chelating agent is rhodamine B hydrazide which forms a complex with mercury in a 1 : 1 stoichiometric ratio (Scheme 1) and then hydrolyses to hydrophobic rhodamine B. The rhodamine B obtained was entrapped in a micellar medium of Triton X 114 and its absorbance was measured. The optimum concentration of rhodamine B hydrazide required to give a maximum absorbance was studied by keeping all the other parameters constant and varying the concentration of rhodamine B hydrazide from 1–7 mM. Constant absorbance values were obtained above 4 mM as shown in Fig. 5. Hence 0.5 ml of 1 mM rhodamine B hydrazide was used to maintain an overall concentration of 5 mM in all further studies.

Effect of pH

Rhodamine B exhibits different structures under different conditions including pH, solvent, temperature *etc.* and it also undergoes molecular aggregation at particular concentrations. So it is very important to study the effect of pH. The pH effect from 1–7 was studied by keeping all other parameters constant. The absorbance values were found to be maximum at a pH range of 4–5 and were found to decrease beyond 5 as shown in Fig. 6.This is because the extraction of rhodamine B was found to be maximum under acidic conditions only.[29] The control of pH at 5 facilitates the selectivity towards mercury, hence the medium pH was maintained by using acetic acid–acetate buffer. Under controlled pH conditions only mercury induces the reaction, therefore interference by copper can be completely eliminated from the reaction.

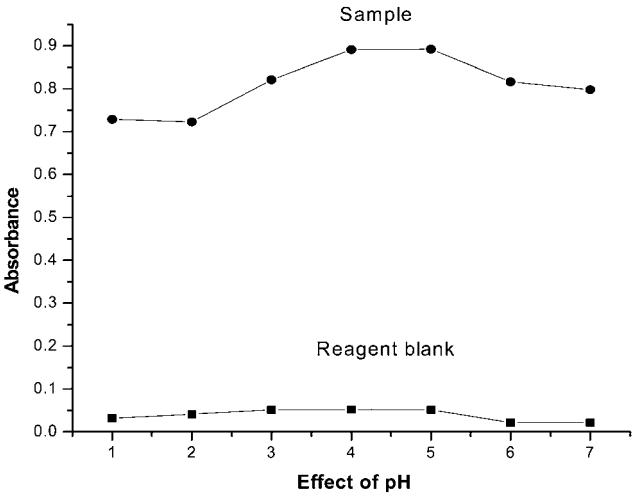


Fig. 6 Effect of pH.

Effect of Triton X-114

The effect of surfactant is crucial in cloud point extraction procedures because the cloud point phenomenon of non- ionic surfactant is reversible and can be influenced by many factors like its concentration and number of ethylene oxide units in the molecule *etc*.[39] Extraction efficiencies of these procedures depend upon the hydrophobic nature of the extractable species. In our protocol rhodamine B produced by hydrolysis of amide bond assisting spirolactam ring opening of rhodamine B hydrazide is the hydrophobic species. Quantitative extraction of rhodamine B is obtained by using Triton X-100 as well as Triton X-114. But in the case of Triton X-100, heating is required to attain the cloud point whereas in the case of Triton X-114 no heating is involved and the clouding takes place at room temperature.[40] Hence Triton X-114 was used in place of Triton X-100 to extract the formed rhodamine B by the catalytic activity of mercury with rhodamine B hydrazide. The optimum concentration of Triton X-114 required was studied by varying the surfactant concentration in the range 0.25 to 1.0% (v/v) and all other variables were kept at optimum conditions. Maximum absorbance values were obtained between 0.6 to 1.0% (v/v). Hence 2.5 ml of 2% Triton X-114 was used to maintain an overall concentration of 0.6% in all further studies (Fig. 7).

Effect of NaCl

The presence of electrolytes decreases the cloud point temperature and increases the extraction efficiency by the salting out effect.[39] The electrolytes induce the dehydration of poly(oxyethylene) groups thereby resulting in the desorption of ions from the hydrophilic parts of the micelles and inducing van der Waals forces between the micelles which leads to clouding of the surfactant.[41] The different electrolytes like KCl, NaCl and $CaCl_2$ was used as additives to induce the above mentioned effect. Even though all three electrolytes increase the extraction efficiency of the rhodamine B into the Triton X-114 phase the best results were obtained in the case of NaCl. Therefore the concentration of NaCl required was studied by keeping all the other parameters constant. Maximum absorbance values for the sample and

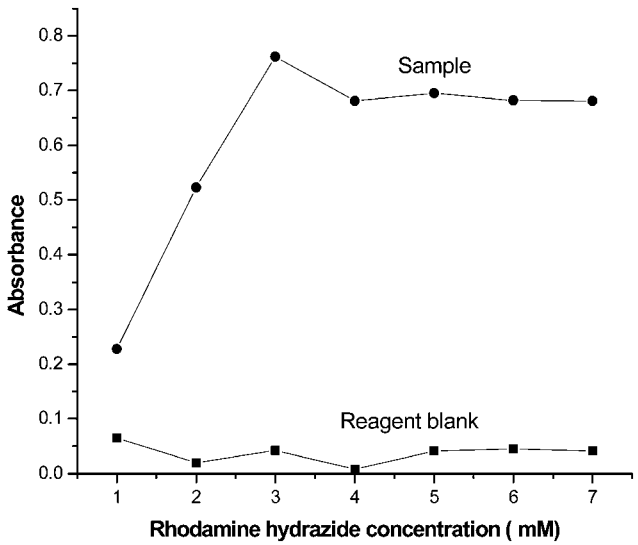


Fig. 5 Effect of rhodamine B hydrazide concentration.

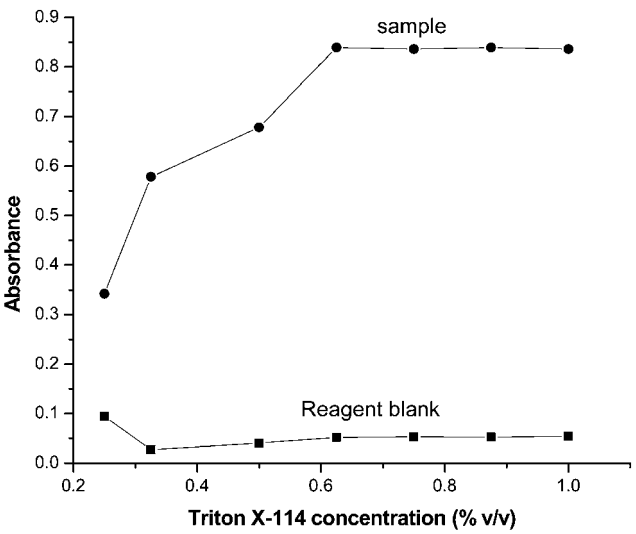


Fig. 7 Effect of Triton X-114.

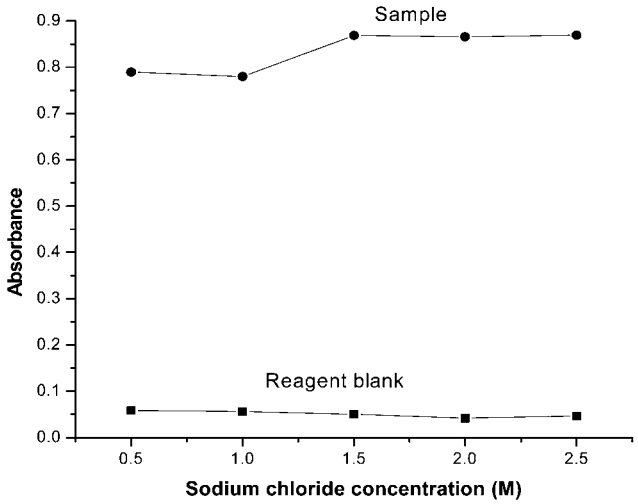


Fig. 8 Effect of NaCl.

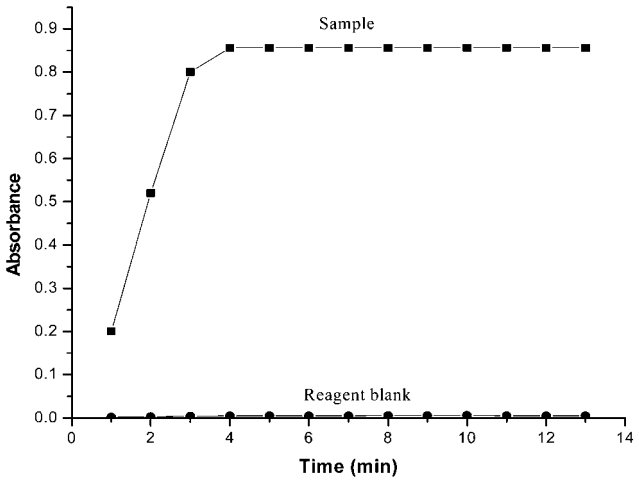


Fig. 9 Effect of reaction time.

minimum absorbance values for the blank were obtained at a concentration of 1.5 M NaCl. In order to get this concentration 2 ml of 1 M NaCl was fixed as an optimum concentration in all further studies (Fig. 8).

Effect of reaction time and temperature

The entire reaction, *i.e.* both the reactions of mercury with rhodamine B hydrazide and cloud point extraction of rhodamine B, was carried out at room temperature. Pourreza *et al.* have determined rhodamine B quantitatively through cloud point extraction using Triton X-100 as a non-ionic surfactant at elevated temperature from colorant substrate materials. However the rhodamine B formed by the catalytic reaction between mercury and rhodamine B hydrazide can be quantified using Triton X-114 at room temperature. Although the reaction between mercury and rhodamine B hydrazide has been found to be instantaneous the reaction was allowed to occur for two minutes to ensure completion. This was confirmed by studying the variation in the absorbance measurements against time (Fig. 9). Cloud formation takes place instantaneously after adding the surfactant to the produced rhodamine B. The cloud formed was centrifuged for about 5 min at 5000 rpm which is sufficient for phase separation. Increasing the centrifugation time beyond 5 min doesn't change the absorbance value. Hence 5 min centrifugation was used as the optimum time for phase separation of rhodamine B in the reaction mixture by cloud point extraction. This method provides a simple and efficient phase separation in the cloud point extraction of mercury at room temperature.

Interference studies

In order to adopt the present method for the determination of mercury ions in waste water samples the selectivity of the method towards the mercury ions over other metal ions and anions present in these waste water samples was checked. The method shows high selectivity towards Hg^{2+} ions. No other cations and anions induce the spirolactam ring opening of rhodamine B hydrazide to give rhodamine B except cupric ion which interferes only above pH 5. Hence the interference from the Cu^{2+} ions can be overcome by the control of the medium by maintaining the pH value at 5. Further the effect of extraction efficiency of rhodamine B in the presence of other ions was studied by adding other metal ions and anions that are commonly present in waste water samples. The extraction efficiencies of rhodamine B were found to be unaltered for the metal ions and anions studied as shown in Table 1. The tolerance limits for different metal ions and anions that are commonly present in waste water samples are found to be less than 1000 μg so the masking of these metal ions in the mercury estimation is undesirable.

Analytical merits of the method

Analytical merits of the optimized method are summarized in Table 2. The limit of detection and linear range were found to be 1.4 ng ml^{-1} and 10–100 ng ml^{-1}, respectively. The RSD was found to be 0.35% for 20 ng and 0.5% for 60 ng respectively. The preconcentration factor and improvement factor were found to be 5 and 12, respectively.

Table 1 Effect of foreign ions

Interferent	Tolerance limit (μg)
Ca^{2+}, Zn^{2+}, Ni^{2+}, Co^{2+}, Fe^{2+}	2000
Cd^{2+}, K^{+}, Na^{+}, Pb^{2+}	1000
Na^{+}, Mg^{2+}, Ag^{+},Hg^{+}	800
Cu^{2+}	500
I^{-}, Cl^{-}, NO_2^{-}, NO_3^{-}, SO_4^{2-}	>1000

Table 2 Analytical merits of the proposed method

Linear range (ng ml^{-1})	10–100
Detection limit (ng ml^{-1})	1.4
Relative Standard Deviation, % ($n = 5$)	0.35
Maximum preconcentration factor	5
Improvement factor	12

Table 3 Determination of mercury in industrial effluent and water samples[a]

Sample	Hg^{2+} originally found (ng ml^{-1})		Hg^{2+} added (ng ml^{-1})	Total Hg found (ng ml^{-1})		% Recovery	
	Proposed method	Standard method		Proposed method	Standard method	Proposed method	Standard method
1. Chromplating industry effluent	0.31 ± 0.01	0.31 ± 0.03	0.10	0.41 ± 0.01	0.39 ± 0.06	95.1	100
2. Textile industry effluent	0.1 ± 0.02	ND	0.01	0.02 ± 0.06	0.019 ± 0.05	95.0	105
3. Lake water	ND	ND	20.00	19.0 ± 0.01	19.0 ± 0.02	99.5	99.5
4. Tap water	ND	ND	25.00	24.9 ± 0.04	24.90 ± 0.03	99.6	99.6

[a] ND, not detected.

Table 4 Comparison of proposed method with other methods[a]

Method	Linear range (μg l^{-1})	Detection limit (μg l^{-1})	Precocentration factor	Improvement factor	Reference
CV-ICP OES	—	0.45	—	13	40
CV-AAS	—	0.12	20	—	41
HPLC-CV-AFS	0.002–4	2.00	10	29	42
Spectrophotometry	4–240	0.53	—	—	43
Spectrophotometry	10–100	1.40	5.0	12	Proposed method

[a] CV-ICP OES, cold vapor-inductively coupled plasma emission spectrometry. CV-AAS, cold vapor-atomic absorption spectrometry. HPLC-CV-AAS, high performance liquid chromatography-cold vapor atomic absorption spectrometry.

Application study

The validity of the proposed method was checked by applying it in the determination of mercury concentration in various water samples collected from environmental matrices. The waste water samples collected from the chrome plating and textile dyeing industries were analysed for the presence of mercury by the proposed method. Recovery studies were also carried out after spiking the samples with known concentrations of mercury. The proposed method was also applied in the estimation of the levels of mercury present in potable water samples like lake and tap waters. The obtained results of the proposed method were validated by comparing the results obtained with the standard dithizone method.[36] (Table 3)

Conclusion

A simple and sensitive procedure has been proposed for the determination of mercury based on the reaction between mercury and rhodamine B hydrazide to form rhodamine B and its cloud point extraction using a non-ionic surfactant. The formed rhodamine B absorbance was measured at 560 nm after extracting into surfactant phase through cloud point formation at room temperature. This procedure allows the determination of mercury in nanogram levels and is highly selective to divalent mercury, other metal ions and anions present in water do not interfere. The method can be applied in the determination of mercury in industrial effluents and the results are in good agreement with the results obtained by the standard dithizone method. This method is simple and more sensitive than conventional spectrophotometric methods because it involves cloud point extraction, and it involves the use of an ecofriendly non-toxic surfactant rather than the organic solvents which are conventionally used. The sensitivity of the method has been compared with some of the reported micellar mediated cloud point extraction procedures (Table 4).[42–45] The proposed method can serve as an alternative to the existing methods.

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