Selective Preconcentration of Ultra Trace Mercury(II) in Aqueous Samples Using Membrane Disks Modified by Dithizone and Its Direct Spectrophotometric Determination

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A new simple and reliable method for rapid and selective extraction of mercury(II) in aqueous solution using octadecyl-bonded silica membrane disks modified with dithizone is presented. A simple modified spectrophotometric method was developed for direct determination of mercury(II) after elution of dithizonate complex from the membrane disk. Extraction efficiency and the influences of sample matrix, optimum amount of ligand, type and minimum amount of organic eluent, flow rates and type, and amount of alkaline stripping solution for free dithizone, were evaluated. The limit of detection of the proposed method is 0.03 ng/mL. The maximum capacity of the membrane disks modified with 2 mg of dithizone was found to be 397 ± 72 µg of mercury(II). Most of the cations and anions have no interfering effect on the extraction and determination of mercury. However, effects of some interfering species such as Cu^{2+}, Ni^{2+} and Bi^{3+} were decreased or eliminated using different methods. The method was successfully applied to the recovery and determination of mercury(II) in synthetic and civic-waste water samples.

Key Words: Mercury(II), Silica Membrane disk, Dithizone, Spectrophotometric method.

INTRODUCTION

Over the last two decades, the need for accurate and sensitive determination of mercury has increased and now, it is a challenging problem, because mercury is a hazardous contaminant of environment and is toxic at low levels to the biota[1]. Mercury contamination is caused by both natural and manmade sources. Natural sources include volcanic action and erosion of mercury-containing sediments. Some of the ways in which humans contaminate the environment with mercury include: mining, transporting and processing mercury ores; dumping of industrial wastes into natural waters, pulp and paper; use of mercury compounds as seed dressing in agriculture, and exhaust from metal smelters[2].

The ultimate effects of mercury in the body are inhibition of enzyme activity and cell damage. Mercury is known to affect the metabolism of essential elements such as sodium and potassium by increasing the latter's permeability. It also inhibits active transport mechanisms through dissipation of the normal gradient; destroys the mitochondrial apparatus; causes swelling of cells, leading to lysis, decreases globulins, suggesting liver dysfunction; decreases DNA content in cells, and adversely affects chromosomes and mitosis, leading to mutagenesis[1, 2]. Thus, there is a growing interest by biologists and environmentalists to establish sources of mercury contamination and to evaluate levels of pollution, and this necessitates the

development of new methods for selective separation, preconcentration and determination of mercury at ultra trace levels in different matrices.

The mercury content of various samples has been measured by different analytical methods described in the literature. These methods are based upon a wide range of analytical techniques such as inductively coupled plasma mass spectrometry[3], electrothermal atomic absorption spectrometry[4], atomic fluorescence spectrometry[5], neutron activation analysis[6], cold vapour atomic absorption spectrometry[7], electrochemical detection[8] and spectrophotometry[9].

Different methods, specially liquid-liquid extraction in the presence of various ligands such as diethyldithiocarbamate, pyrrolidinedithiocarbamate, tri-*n*-butylphosphate, tri-*n*-octylphosphine oxide, bis-(2-ethylhexyl) phosphoric acid, tenoyltrifluoroacetone, 8-hydroxyquinoline, thiocyanate, acetylacetone, dithizone and crown ethers have been applied for separation of mercury[10, 11]. Classical extraction methods have still wide analytical and industrial applications, but their use is usually time-consuming, labour-intensive and requires large amounts of high purity solvents for extraction. Nevertheless, several other techniques for the preconcentration of mercury have been proposed including cryogenic trapping, concentration in a palladium-coated graphite tube, amalgam preconcentration into a gold wire absorber and solid phase extraction[12, 13].

Solid phase extraction (SPE) or liquid-solid extraction is a popular and growing technique that is used for sample preparation for analysis. It is an attractive alternative to classical liquid-liquid extraction methods that reduces solvent usage and exposure, disposal costs and extraction time for sample preparation purposes[14–17]. In recent years, the octadecyl-bonded silica SPE disks have been utilized for the extraction and separation of different organic compounds from environmental matrices[18–23]. Moreover, the SPE disks modified by suitable ligands are successfully used for selective extraction and concentration of metal ions[24–26]. The aim of this work was the development of a rapid, efficient and highly sensitive method for selective extraction and concentration of ultra trace amounts of Hg^{2+} ions from aqueous media using octadecyl silica membrane disks modified by dithizone (diphenylthiocarbazone, H_2Dz) and direct spectrophotometric determination of extracted complex after elution from the membrane disk.

EXPERIMENTAL

HPLC-grade methnol, chloroform, dichloromethane and carbon tetrachloride (from Merck or Aldrich) were used without any further purification. All acids and bases were of the highest purity available from Merck and were used as received. Reagent grade mercuric chloride, hydroxylamine hydrochloride, EDTA (disodium salt, dihydrate) and the chloride or nitrate salts of used cations (from Merck or Fluka) were of the highest purity available and were dried, before using, under vacuum over P_2O_5. Reagent grade dithizone (Merck) was used as received. Doubly distilled deionized water was used throughout. The standard stock solution of mercury(II) was prepared by dissolving an appropriate amount of mercuric chloride in 1% (v/v) nitric acid solution. Working solutions were prepared by appropriate dilution of the stock solution with water.

The absorbance measurements were carried out with a Shimadzu UV-160 spectrophotometer at 485 nm. Extractions were performed with 47 mm diameter and 0.5 mm thickness Empore membrane disks containing octadecyl-bonded silica (8-μm particles, 6 nm pore size, 3M Co., St. Paul, MN) with a standard Millipore 47 mm filtration apparatus connected to a vacuum pump. The typical composition of the disks was 90% (w/w) octadecyl-bonded silica and 10% (w/w) PTFE fibres.

Preparation of SPE membrane disks

In order to remove potential interferences and to ensure optimal extraction of the analyte of interest, the disk cleaning and conditioning should be done before its use. Thus, after placing the membrane disk in the filtration apparatus, 10 mL methanol was poured on to the disk and immediately drawn through the disk by applying a slight vacuum. After all of the solvent has passed through the disk, it was dried by passing air through it for a few minutes. The disk conditioning started by pouring 10 mL methanol on to the disk. Immediately, a low vacuum was applied and the solvent was drawn through the disk until a thin layer of methanol was left on the surface of the disk. This is to ensure complete wetting of the disk with the organic solvent. Immediately thereafter 10 mL chloroform was introduced on to the disk and was drawn through the disk. The disk was then dried under vacuum for 5 min or longer if necessary. Finally, a solution of 2 mg dithizone in 2 mL chloroform (a dark-blue solution) was introduced on to the disk so that the solution was spread on the whole disk surface and was drawn slowly through the disk by applying a slight vacuum. The passed solution was collected in a test tube. The a few drops of 50% methanol-water added to the test tube (until just before appearance of a colloidal suspension), and the resulting mixture was again introduced to the reservoir and passed through the disk slowly. The filtration step was repeated until the passed solution was completely clear and colourless. Finally, the disk was washed with 10 mL 0.1 M nitric acid and dried by passing air through it. The membrane disk modified by dithizone is now ready for sample extraction.

Solid-phase extraction and quantification of Hg^{2+}

After completeness of modification, 500 mL of sample solution (0.2 M HNO_3) containing 5 μg of Hg^{2+} ions was passed through the disk at a flow rate of 40–50 mL/min. After the extraction, the disk was dried completely by passing air through it. A proper test tube was then placed under the extraction funnel. The extracted mercury-dithizone complex and the excess dithizone were eluted from the disk using chloroform (5 + 5 mL) at a flow rate of 5–10 mL/min. Then the eluent was transferred into a 50 mL separating funnel that contained 15 mL of 1.0 M NaOH solution and it was shaken for 1 min. This process ensures complete stripping of excess dithizone by alkaline solution so that the colour of the complex solution is changed from dark-blue to orange-yellow, that means no free dithizone has been left in the chloroform phase. Then a proper volume of chloroform phase (lower phase) was taken and transferred to a UV-Vis cell. The mercury concentration was then determined at 485 nm against a reagent blank (linear calibration range 0.1–2.0 μg/mL, r = 0.999).

Determination of Hg^{2+} ions in civic-waste water samples

After sampling, the samples were acidified and stored in a refrigerator using PVC containers to the time when the determination possibility was brought about. Then, a 1000 mL aliquot of every water sample was passed through bare membrane disk to remove all particles and organics that usually exist in such water samples. Then, the samples were passed through a modified membrane disk and the recommended procedure for mercury(II) ions determination was carried out.

RESULTS AND DISCUSSION

The Hg^{2+} ion as a soft acid possesses a coordination number of four and exhibits a high tendency to form complexes with ligands containing S or N donor atoms with soft and intermediate base properties respectively. Thus, the existence of a donating S atom as well as N atoms in dithizone was expected to increase both the stability and selectivity of its mercury(II) complex over other metal ions[27-29]. Mercury(II) ions in an acidic medium react with excess of dithizone to form the dithizonate Hg(HDz)$_2$, which is soluble in CCl$_4$ or CHCl$_3$[10, 27].

Some preliminary experiments were carried out in order to investigate the quantitative retention of mercury ions by the membrane disks in the absence and presence of dithizone. It was found that the disks modified with 5 mg of the ligand are capable to quantitatively retain Hg^{2+} ions from 1.0 M HNO$_3$ solution (5 μg of Hg^{2+} in 50 mL solution), while the bare membrane disk shows no tendency to extraction of mercury ions.

Effect of sample matrix, eluent and stripping solution for free dithizone

To investigate the influence of the matrix on the extraction of mercury(II) ion, HNO$_3$, HCl, H$_2$SO$_4$ and CH$_3$COOH were tested. 500 mL Solutions containing 5 μg of mercury(II) and varying concentrations of the acids were passed through the membrane disk modified with 10 mg of dithizone (Fig. 1). Mercury(II) ion is
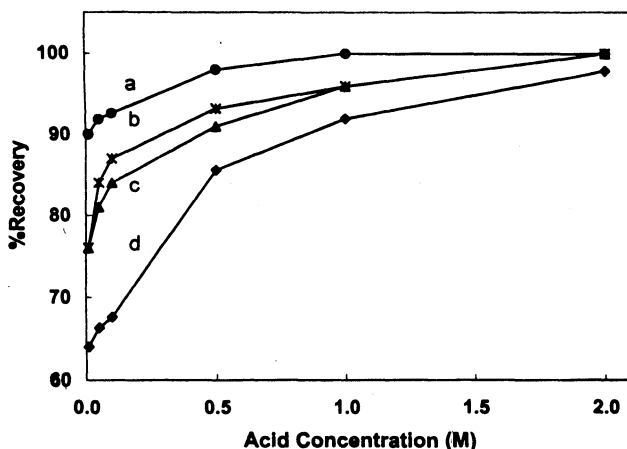

Fig. 1.　Recovery of 5 μg of mercury from 500 mL solutions containing different acids of varying concentrations by the membrane disk modified with 5 mg of dithizone: (a) HNO$_3$, (b) H$_2$SO$_4$, (c) HCl, and (d) CH$_3$COOH

quantitatively extracted by concentrations higher than 0.5 M HNO$_3$ and higher than 1.0 M HCl, H$_2$SO$_4$ and CH$_3$COOH. Acid concentrations more than 1.0 M reduce the efficient lifetime of the disk, while concentrations of HCl greater than 1.0 M impede the formation of mercury dithizonate, owing to the formation of stable mercury chloride complexes[11]. Thus, 1.0 M HNO$_3$ was chosen as sample matrix.

Organic solvents such as chloroform, dichloromethane and carbon tetrachloride were tested as eluent for the extracted mercury dithizonate complex from the disk (Fig. 2). Chloroform was found to be the best eluent not only because of the complete recovery with the least amount of solvent but also because of the improvement of direct spectrophotometric determination of eluted complex and development of faster stripping process for excess dithizone from the eluent.
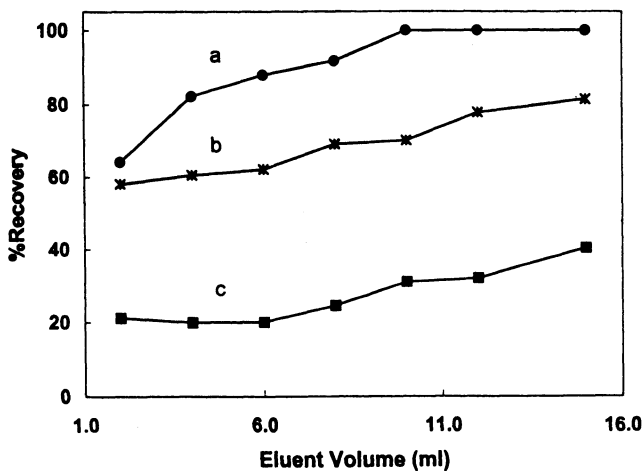

Fig. 2.   Recovery of mercury from the modified disk using different volumes of eluent organic solvents: (a) chloroform, (b) carbon tetrachloride, and (c) dichloromethane

In order to remove the free dithizone from the eluent, different volumes of NH$_3$ and NaOH solutions at varying concentrations were tested. The Hg(HDz)$_2$ formed is resistant to the action of alkaline solutions. Thus, 125 mL of 1.0 M NaOH solution was chosen as the appropriate washing solution for the free dithizone, which provides its complete stripping at a shaking time less than 1 min.

Effect of amount of ligand and flow rates

The optimal amount of dithizone for the proper modification of the membrane disks was investigated (Fig. 3). The membrane disks modified with 0.6–5.0 mg dithizone retain 5 μg of mercury quantitatively. In order to prevent the washing of dithizone from the disk at high sample volumes and to achieve higher concentration and capacity factors, 2 mg of dithizone was used for further studies. The influence of the flow rates of the sample solution and chloroform though the membrane disks on the retention and recovery of Hg^{2+} ion was investigated. It was found that, in the range of 1–50 mL/min the retention of mercury by the modified membrane disk is not affected by the sample flow rate significantly. On the other hand, quantitative elution of dithizonate complex from the disk was achieved in a flow rate range of 1–16 mL/min.
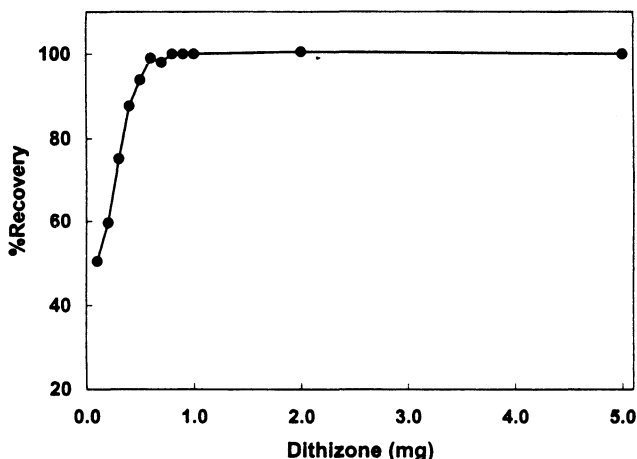
Fig. 3.   Recovery of 5 µg of mercury from 500 mL 1.0 M HNO$_3$ solution by the membrane disk modified with varying amounts of dithizone

Analytical performance

When 5 µg mercury in 10, 50, 100, 500, 1000, 2000 and 3000 mL sample solutions was passed through the modified disks, under optimal experimental conditions, the Hg^{2+} was retained quantitatively. Thus, the breakthrough volume for the method should be greater than 3000 mL, providing a concentration factor higher than 300.

The maximal capacity of the membrane disk modified by 2 mg of dithizone was determined by passing 500 mL protions of the sample solution containing 3000 µg mercury, followed by spectrophotometric determination of the retained mercury. The maximal capacity of the membrane disk obtained from three replicate measurements was 397 ± 72 µg of mercury on the disk. The limit of detection (LOD) of the proposed method was studied under the optimal experimental conditions. The LOD obtained from $C_{LOD} = k_b S_b m^{-1}$ for a numerical factor $k_b = 3$[12] is 0.03 ng/mL.

The influence of several cations and anions on the solid phase extraction and determination of mercury(II) ion (5 µg of mercury in 500 mL of 1.0 M HNO$_3$ solution) was studied. The results are summarized in Table-1. The results show that the mercury(II) ions in the binary mixtures are retained almost completely by the modified disk, even in the presence of up to about 50 mg of diverse ions. However, some of the species tried such as Bi^{3+}, Ni^{2+}, Zn^{2+} and especially Cu^{2+} interfere with the determination of mercury(II) ion. These interferences were eliminated or reduced considerably in the presence of proper masking, reducing and stripping agents (Table-2). It should be noted that the quantitative separation of Hg^{2+} from some of the interfering cations could be achieved by washing the disk with 10–50 mL of 2.0 M HNO$_3$ solution, prior to the elution of the extracted mercury dithizonate complex from the disk.

To assess the applicability of the proposed method for samples with different matrices containing varying amounts of a variety of diverse ions, it was applied to the separation and determination of Hg^{2+} ions from two synthetic samples as well as a natural water sample (Table-3). The results show that the recovery of mercury is almost quantitative.

TABLE-1

SEPARATION OF 5 µg OF MERCURY FROM 500 mL 1.0 M HNO_3 SOLUTION IN THE PRESENCE OF DIFFERENT DIVERSE IONS BY THE MEMBRANE DISK MODIFIED WITH 5 g OF DITHIZONE

Diverse ion	Amount taken (mg)	% Recovery of Hg^{2+} ion	
Na^+	51.0	103.2	$(0.9)^a$
K^+	52.0	97.1	(1.3)
Ca^{2+}	26.5	99.1	(2.7)
Mg^{2+}	25.4	100.1	(0.6)
Ba^{2+}	3.5	102.4	(1.9)
Sr^{2+}	3.5	102.1	(2.2)
Mn^{2+}	4.5	98.7	(3.2)
Co^{2+}	3.0	104.0	(3.1)
Cu^{2+}	0.02	97.0^b	(1.7)
Ni^{2+}	0.5	97.9^b	(3.0)
Zn^{2+}	0.3	103.2^b	(3.0)
Ag^+	0.1	99.7^b	(0.7)
Cd^{2+}	0.5	103.1	(1.9)
Pb^{2+}	0.5	101.9	(2.2)
Al^{3+}	2.0	103.2	(0.5)
Bi^{3+}	0.1	97.0^b	(3.7)
Fe^{3+}	0.1	99.4	(1.0)
Cr^{3+}	3.0	98.0	(1.0)
Ce^{4+}	3.5	99.0	(2.1)
EDTA	37.0	100.6	(1.5)
NO_3^-	51.0	103.0	(2.3)
Cl^-	52.0	98.1	(1.8)
CO_3^{2-}	2.7	99.7	(2.1)
SO_4^{2-}	3.5	99.0	(0.8)

[a] Values in parentheses are % RSDs based on three replicate analyses.
[b] After interference removing.

TABLE-2

INFLUENCE OF MASKING AGENTS AND WASHING ON THE ERRORS PRODUCED BY INTERFERING IONS IN RECOVERY AND DETERMINATION OF 5 µg OF MERCURY IN 500 mL 1.0 M HNO_3 SOLUTIONS BY THE MODIFIED MEMBRANE DISKS

Interfering ion	(µg)	Masking agent (M)		%Error
Cu^{2+}	(50)	—		100
	(50)	Rinsing[a]		100
	(50)	Rinsing + EDTA	(1.0×10^{-3})	100
	(20)	Rinsing + EDTA	(1.0×10^{-3})	89
	(20)	$NH_2OH \cdot HCl$	(1.0×10^{-2})	81
	(20)	Stripping[b]		3
Ni^{2+}	(500)	—		100
	(500)	EDTA	(1.0×10^{-3})	49

Interfering ion	(μg)	Masking agent (M)		%Error
Zn^{2+}	(500)	Rinsing + EDTA	(1.0×10^{-3})	2
	(300)	—		100
	(300)	EDTA	(1.0×10^{-3})	73
	(300)	Rinsing + EDTA	(1.0×10^{-3})	0
Ag^+	(100)	—		100
	(100)	Rinsing + EDTA	(1.0×10^{-3})	82
	(100)	Stripping[c]		−2
Bi^{3+}	(100)	—		100
	(100)	Rinsing		0

[a] Washing the disk with 50 mL 2 M HNO_3 solution prior to the elution of complex.
[b] After the elution, the extract was shaken with an aqueous solution containing KI (0.1 M) and HCl (1.0 M) for stripping of $Cu(HDz)_2$.
[c] After the elution, the extract was shaken with an aqueous solution containing HCl (1.0 M) for stripping of AgCl.

TABLE-3

RECOVERY OF 5 μg OF MERCURY FROM 1000 mL SOLUTIONS OF SYNTHETIC AND REAL WATER SAMPLES

Sample	% Recovery of Hg^{2+} ions	
Synthetic 1: (Na^+, K^+, Ca^{2+} and Mg^{2+}, 15 mg of each cation)	93.1	(2.1)[a]
Synthetic 2: (Composition of sample 1 in addition to 0.01 M EDTA)	101.1	(1.0)
Synthetic 3: (Na^+, K^+, Ca^{2+}, Mg^{2+}, 15 mg of each cation and Sr^{2+}, Zn^{2+}, Pb^{2+}, Cd^{2+}, Bi^{3+}, Al^{3+}, 2 mg of each cation)	79.8	(1.9)
Synthetic 4: (Composition of sample 3 in addition to 0.01 M EDTA)	98.3	(2.4)
Tap water (Khorram-Abad, Feb 2002)	97.1	(1.3)

[a] Values in parentheses are % RSDs based on three replicate analyss.

The proposed method was also applied to determination of Hg^{2+} ions in the three river waters, taken from different civic-waste sites in Khorram-Abad. The results are given in Table-4. It is noteworthy that satisfactory agreements exist between the results obtained by proposed method and the results reported by Bandar Imam Petrochemical Co. (Bacharach 50B, Mercury-Analyzer).

TABLE-4

DETERMINATION OF MERCURY IN THREE WATER SAMPLES FROM CIVIC-WASTE SITES IN KHORRAM-ABAD[a]

Sample sites	Proposed method (ng/mL)		Mercury analyzer[b] (ng/mL)
Parsilon Co.	11.7	(0.9)[c]	9.0
Lorestan Industrial Husbandry Co.	1.3	(1.7)	0.0
Gilevaran village	4.9	(2.1)	4.0

[a] 1000 ml of each sample was taken.
[b] Mercury Analyzer (Bandar Imam Petrochemical Co.).
[c] Values in parentheses are % RSDs based on three replicate analyses.

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