Determination of mercury in water and fish samples by cold vapor atomic absorption spectrometry after solid phase extraction on agar modified with 2-mercaptobenzimidazole

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ABSTRACT

A novel solid phase extraction method for the determination of mercury has been developed. The Hg(II) ions were retained on a mini-column packed with agar powder modified with 2-mercaptobenzimidazole at a flow rate of $6\,mL\,min^{-1}$. The retained Hg(II) ions were eluted with $3\,mol\,L^{-1}$ solution of HCl and measured by cold vapor atomic absorption spectrometry (CV-AAS). The effect of different variables such as pH, sample flow rate, amounts of 2-mercaptobenzimidazole loaded on agar and $SnCl_2$ concentration was investigated and optimum conditions were established. The calibration curve was linear in the range of 0.040–$2.40\,ng\,mL^{-1}$ with $r = 0.9994$ ($n = 8$). The limit of detection based on three times the standard deviation ($3S_b$) ($n = 10$) obtained under optimum conditions was $0.02\,ng\,mL^{-1}$. The relative standard deviation (R.S.D.) for the determination of 0.4 and $2.0\,ng\,mL^{-1}$ of Hg(II) was 2.6 and 1.9% ($n = 8$), respectively. The method was successfully applied to determine Hg(II) in water, wastewater and fish samples.

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1. Introduction

Mercury is a global pollutant and is identified as a highly toxic element because of its accumulative and persistent character in the environment and living organisms. Lethal concentrations of mercury salts range from less than $0.1\,ng\,mL^{-1}$ to more than $200.0\,ng\,mL^{-1}$ for marine species and freshwater organisms [1]. Therefore, routine monitoring and control of mercury are becoming increasingly important, especially in water system. Several analytical techniques such as spectrophotometry [2,3], inductively coupled plasma mass spectrometry (ICP-MS) [4], inductively coupled plasma atomic emission spectrometry (ICP-AES) [5], voltammetric [6], atomic fluorescence spectrometry (AFS) [7] and neutron activation analysis [8] have been developed for the determination of mercury. Nevertheless, cold vapor atomic absorption spectroscopy (CV-AAS) is the most widely used method due to its simplicity, relatively low cost of operation, high sensitivity and selectivity [9–11]. However, CV-AAS is not straightforwardly applicable to some environmental, clinical, or biological samples in view of low analyte content and matrix of the sample. Therefore, a reliable preconcentration step is essential for quanti-

tative separation and enrichment of mercury ions from the matrix of the sample. Several separation and preconcentration procedures including preconcentration on gold amalgamator [12,13], liquid–liquid extraction [14], cloud point extraction [15] and solid phase extraction [16,17] have been used prior to measurement by CV-AAS.

The application of solid phase extraction for preconcentration of trace metals from different samples results in several advantages such as the minimal waste generation, reduction of sample matrix effects, simplicity, high preconcentration factor as well as sorption of the target species on the solid surface in a more stable form.

Various organic and inorganic solid phases including chloromethylated polystyrene [18], Cyanex 923 [19], naphthalene [20,21], activated carbon [22], silica gel [23] and alumina [24] can be used as a support for the extractor compound that reacts selectively with the target species. The most successful selective extractor compounds for soft metal ions are sulfur-containing molecules, which are widely used in different analytical fields.

In this paper, agar–agar powder was used as a new solid support. Agar–agar powder is a mixture of branched and unbranched polysaccharides that has a high molecular weight and is insoluble in water at room temperature [25,26]. This article aims at developing a rapid, sensitive and efficient method for preconcentration and CV-AAS determination of Hg(II) ions in water, wastewater and

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fish samples by sorption on the agar mini-column modified with 2-mercaptobenzimidazole (2-MBI).

2. Experimental

2.1. Instrumentation

The atomic absorption measurements were performed with a Philips PU9100X (England) atomic absorption spectrometer fitted with a mercury hollow cathode lamp (Unicam, Franklin, MA). A wavelength of 253.7 nm and spectral bandpass of 0.5 nm was used throughout. A T-cell quartz tube (120-mm length and 5-mm i.d.) was placed directly on the nitrous oxide/acetylene burner equipped with T-cell tube holder. The nitrous oxide/acetylene flame was turned off throughout the process. All measurements were recoded on the height mode of atomic signal.

Gaseous mercury atoms were generated using homemade glassware called reaction cell-gas liquid separator (RC-GLS) (Fig. 1). The characteristics were as follows: internal diameter, 10 mm, length, 150 mm, an inlet for N_2 gas and the outlet for N_2 and Hg gas mixture. In each experiment, Hg(II) sample solution and reductant were introduced to the RC-GLS from the removable cap and placed directly on the frit glass that was fitted to this glassware.

2.2. Reagents and solutions

All chemicals used were of analytical reagent grade and the presence of mercury was not detected within the working range. Double distilled water was used throughout the work.

The Hg(II) stock solution ($1000\,\mu g\,mL^{-1}$) was prepared by dissolving 0.1349 g of $HgCl_2$ (Merck, Darmstadt, Germany) in 1 mL of concentrated HNO_3 (Merck, $d = 1.4$, 70%) and diluting to 100 mL with water. The working solutions of Hg(II) were prepared daily by appropriate dilution of the $10\,\mu g\,mL^{-1}$ mercury solutions which was prepared weekly, with water. A 2% (w/v) $SnCl_2 \cdot 2H_2O$ (Merck) used as reducing agent was prepared daily by dissolving the appropriate amount of $SnCl_2 \cdot 2H_2O$ in HCl (Merck) and diluting with water.

Agar–agar powder (Merck) of mesh 60 size and 2-mercaptobenzimidazole (Merck) were purchased for solid phase preparation. All containers were soaked in 20% of HCl and HNO_3, respectively, and then cleaned thoroughly with water [27].

A citrate–citric acid buffer solution was prepared by adding $0.2\,mol\,L^{-1}$ of NaOH to 50 mL of $0.2\,mol\,L^{-1}$ citric acid (Merck) solution and adjusting the pH to 2.5 using a pH meter (model 632, Metrohm, Herisau, Switzerland).

2.3. Sample pre-treatment

Drinking water sample supplied from Water Safe Company and Abadan Petrochemical wastewater samples were collected in a 2.5-L PTFE bottle and filtered through a filter paper (Whatman no. 40) before use.

For digestion of fish samples, 500 mg of dried sample was placed in a digestion vessel and 5 mL of HNO_3 (70%) as well as 6 mL of H_2O_2 (30%) were added. The vessel was immediately assembled, gently swirled and placed in the pre-heated oven at 180 °C for about 1.5 h [27]. Then 6 mL of $1\,mol\,L^{-1}$ of $K_2S_2O_8$ was added and heated for 30 min. The digested fish sample was cooled at room temperature [28,29]. Appropriate amounts of $2\,mol\,L^{-1}$ NaOH was added to neutralize the excess of HNO_3 and then 4 mL of the citrate–citric acid buffer solution (pH 2.5) was added to adjust the pH at the optimized value.

300 mg of DORM-3 certified reference material, was refluxed with 10 mL of H_2SO_4 (98%) and 10 mL of HNO_3 (70%) at 250 °C for 2 h. Then 10 mL of $K_2S_2O_8$ (5%) and 10 mL of $KMnO_4$ (5%) was added and digested for 2 h. It was cooled, filtered, neutralized with sodium hydroxide and diluted to 500 mL in a volumetric flask. 50 mL of this solution was treated under the general procedure.

2.4. Mini-column preparation

For the preparation of the adsorbent, 250 mg of 2-mercaptobenzimidazole was added to 15 mL of acetone (Merck) in a 50-mL beaker and heated up to 45 °C for 10 min. Then 5 g of agar–agar powder was added gently to the solution and mixed thoroughly. The mixture was placed at 4 °C for 15 min and then left to dry at room temperature for about 30 min. 200 mg of this dried adsorbent was packed in a mini-column (8-mm i.d. and 80-mm length) for preconcentration procedure. The dried adsorbent can be stored and used for 4 months after the preparation.

2.5. General procedure

For the preconcentration of mercury ions, 250 mL solutions containing mercury in the range of 0.04–2.40 $ng\,mL^{-1}$ and 4 mL of citrate buffer at pH 2.5 was passed through a mini-column packed with 200 mg of the adsorbent at a flow rate of $6\,mL\,min^{-1}$ (five preconcentration operations were performed simultaneously). The mercury ions were eluted from the mini-column by 2.5 mL of $3\,mol\,L^{-1}$ of HCl at a flow rate of $6\,mL\,min^{-1}$. The eluted mercury solution was placed in the RC-GLS and 4 mL of 2% of $SnCl_2$ solution was added. After 1 min, N_2 gas at a flow rate of $350\,mL\,min^{-1}$ was passed through the RC-GLS to take the mercury vapors to the T-Cell quartz tube. The measurements were performed on the height mode of atomic signal.
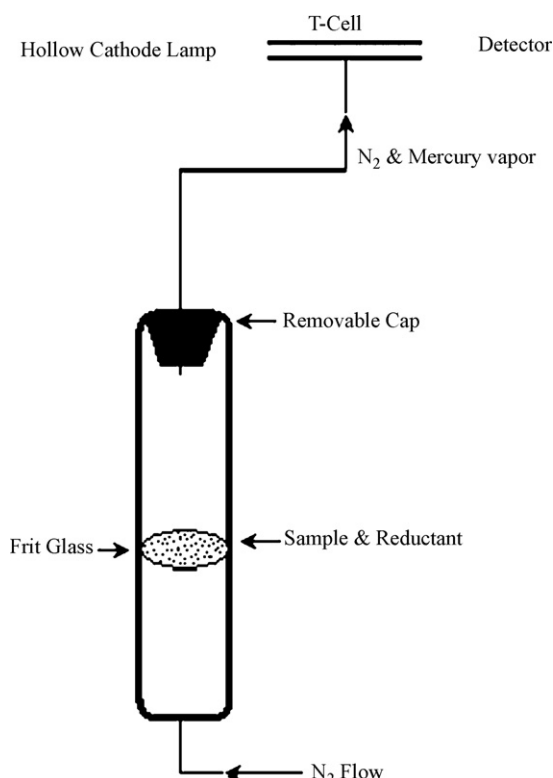

Fig. 1. Schematic diagram of the reaction cell-gas liquid separator (RC-GLS).
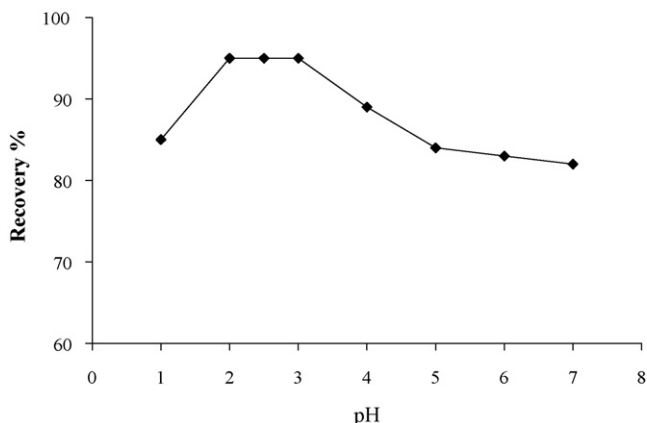
Fig. 2. Effect of pH on the recovery of 0.4 ng mL^{-1} of Hg(II).

Fig. 4. Effect of amounts of ligand immobilized on agar powder on the retention of 0.1 μg of Hg(II).

3. Results and discussion

2-Mercaptobenzimidazole is a well-known analytical reagent for mercury. 2-MBI was immobilized on agar–agar powder and used as a new adsorbent for retention and preconcentration of mercury. The effect of different variables was investigated in order to achieve highest possible sensitivity.

3.1. Effect of pH

The reaction between mercury ions and 2-MBI reagent can be influenced by the pH of the solution. Therefore, in this experiment the effect of pH on the preconcentration of 0.4 ng mL^{-1} of Hg(II) was investigated. Fig. 2 shows that the sorption of Hg(II) ions increased with increasing the pH of the solution up to 2 and remained constant in the range of 2–3. Thus, the pH 2.5 was selected as the optimum value for the sorption of Hg(II) ions and 4 mL of citric acid–citrate buffer solution pH 2.5 was used to maintain this pH.

3.2. Effect of reducing agent concentration

The influence of the SnCl$_2$ concentration on the cold vapor generation was evaluated within the range of 1.0–5.0% (w/v). Fig. 3 shows that the maximum absorbance is obtained at concentrations between 2.0 and 3.0% (w/v). Thus, SnCl$_2$ concentration of 2.0% was selected for further experiments.
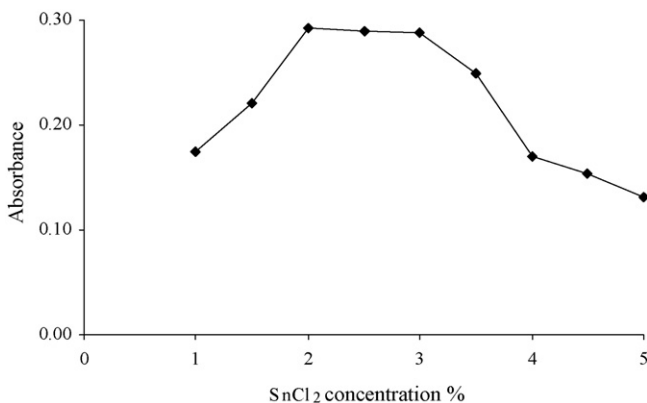
3.3. Effect of amount of ligand

For a proper modification of the agar adsorbent, the effect of the amount of 2-mercaptobenzimidazole on the retention of Hg(II) ions was studied. Various amounts (25–450 mg) of 2-mercaptobenzimidazole were added to 5 g of agar–agar powder and 200 mg of the mixture was used as a mini-column. As shown in Fig. 4, the agar mini-column modified with 20–90 mg g^{-1} of 2-MBI ligand is capable of retaining Hg(II) ions with quantitative recoveries. Thus, 200 mg of the adsorbent containing 50 mg g^{-1} of the ligand was used for further investigations.

3.4. Effect of elution parameters

The nature of the eluent is of prime importance and should meet three criteria; efficiency, selectivity and compatibility [24]. Several eluent solutions such as thiourea, HNO$_3$, H$_2$SO$_4$ and HCl were tested. As it is observed in Table 1 a solution of HCl at concentration of 3 mol L^{-1} is a proper solvent to elute Hg(II) from the column with 96% recovery. Higher concentrations of HCl have no effect on the recovery. The effectiveness of HCl as an eluent may come from both its ability to decompose the complex of Hg(II) and 2-MBI on the mini-column and produce stable complexes of HgCl$_4$$^{2-}$ or HgCl$_3$$^-$ that is not retained on the mini-column. To achieve the highest recovery for mercury ions, the effect of volume of the eluent was also tested. The minimum volume of 3 mol L^{-1} HCl required for quantitative elution of the retained analyte complexes was 2.5 mL.

3.5. The effect of flow rate

In order to ensure quantitative recovery (>95%), the effect of flow rate of mercury sample solution was studied in the range of 1–12 mL min^{-1} under above optimum conditions. As the results in Fig. 5 show, a sample solution containing 0.4 ng mL^{-1} of Hg(II) can be retained on the column quantitatively with flow rates in the

Fig. 3. Effect of SnCl$_2$ concentration on the absorbance of 0.4 ng mL^{-1} of Hg(II).

Table 1
Recovery of 0.4 ng mL^{-1} Hg(II) from the modified agar mini-column using different stripping solutions

Stripping solution	Recovery (%)[a]
Thiourea (1 mol L^{-1})	1
H$_2$SO$_4$ (3 mol L^{-1})	62
HNO$_3$ (3 mol L^{-1})	76
HCl (3 mol L^{-1})	96

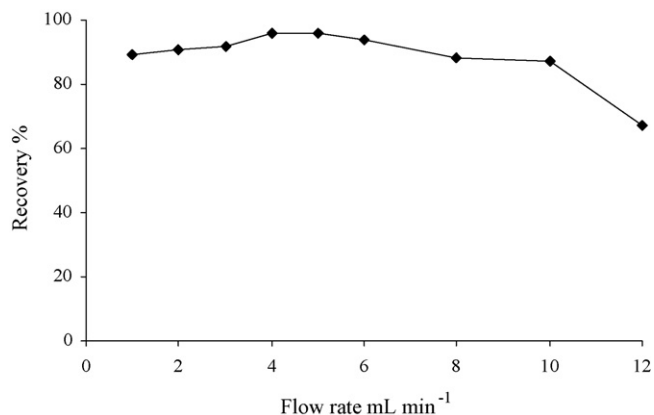[a] Recoveries are based on three replications.

Fig. 5. Effect of sample flow rate on the recovery of 0.4 ng mL^{-1} of Hg(II).

range of 4–6 mL min^{-1}. Therefore, to achieve the speed of operation, a sample rate of 6 mL min^{-1} was selected as the optimum value. The effect of flow rate of the eluent solution was also studied in the range of 1–10 mL min^{-1} and it was found that up to flow rates of 6 mL min^{-1} the recovery remained constant. Therefore, a flow rate of 6 mL min^{-1} was selected for eluting the retained mercury ions from the column.

3.6. Effect of sample volume

The effect of the sample solution volume on mercury sorption was studied by passing 100–500 mL sample solutions containing same amounts of mercury through the mini-column at a flow rate of 6 mL min^{-1}. For the sample solutions containing 0.4 ng mL^{-1} of mercury ions, the maximum sample volume that can be passed with quantitative recovery (>95%) was 250 mL. Above 250 mL the percent sorption decreased for the analyte.

3.7. Capacity of the adsorbent

In order to determine the maximum amount of Hg(II) ions retained on the modified agar adsorbent 100 mL of a solution containing 100 μg of Hg(II) was passed through the column at optimized flow rate and pH. Then 2.5 mL of 3 mol L^{-1} HCl at a flow rate of 6 mL min^{-1} was passed to elute mercury ions from the column. Because of high mercury concentration, the solution was diluted to 100 mL with distilled water and then 400 μL of this solution was analyzed by CV-AAS. The maximum capacity was found to be 49 ± 2 μg g^{-1} of the adsorbent.

3.8. Analytical performance

A linear calibration curve was obtained in the range of 0.04–2.4 ng mL^{-1} of Hg(II) in the initial solution. The linear regression equation was $A = 0.1617\,C + 0.0488$, with $r = 0.9994$ ($n = 8$), where A and C are the absorbance and concentration of mercury in ng mL^{-1}, respectively. The limit of detection (LOD) of the proposed method based on three times the standard deviation ($3S_b$) and for 10 replicate measurements of blank solution at optimized conditions was 0.02 ng mL^{-1}. The relative standard deviation (R.S.D.) for the determination of 0.4 and 2.0 ng mL^{-1} of Hg(II) was 2.6 and 1.9% ($n = 8$), respectively.

Table 2
Effect of foreign species in the determination of 0.4 ng mL^{-1} of Hg(II)

Species	Tolerance ratio (w/w)	Recovery (%)
Na$^+$, K$^+$, NH$_4^+$, Ca^{2+}, Cu^{2+}, Pb^{2+} and Ni^{2+}	50,000	97
Fe^{2+}, Fe^{3+}, Mg^{2+} and Zn^{2+}	25,000	95
Al^{3+}, Cr^{3+}, Bi^{3+} and Mn^{2+}	5000	97
Sb^{3+}	250	96
As^{3+} and Ag$^+$	50	98
NO$_3^-$, HCO$_3^-$, CO$_3^{2-}$ and SO$_4^{2-}$	50,000	100
NO$_2^-$, H$_2$PO$_4^-$, Br$^-$,SO$_4^{2-}$ and PO$_4^{3-}$	25,000	95
Cl$^-$	5000[a]	95
F$^-$ and I$^-$	500	98
EDTA, thiourea acetic acid	25,000	98

[a] After addition of 6 mL 1 mol L^{-1} K$_2$S$_2$O$_8$ [28].

3.9. Interference studies

The effect of diverse cations, anions and organic substances on the preconcentration and determination of 0.4 ng mL^{-1} Hg(II) by the proposed method was studied. Each ion or organic substance was considered to be an interferent when it caused an error greater than ±5% in the determination of mercury. As shown in Table 2 the proposed method was relatively selective for Hg(II). However, Cl$^-$ interfered in the preconcentration procedure. This was attributed to the formation of chlorocomplexes of mercury such as HgCl$_4^{2-}$. The interference of this anion in water and fish samples can be easily eliminated by adding 6 mL of 1 mol L^{-1} potassium peroxidisulfate (K$_2$S$_2$O$_8$) during digestion procedure [29,30].

4. Application

4.1. Analysis of certified reference material

In order to evaluate the accuracy of the developed procedure, mercury was determined in a certified reference material (DORM-3). It was found that there is no significant difference between results obtained by the proposed method and the certified value (Table 3).

4.2. Analysis of fish samples

The proposed method was applied to the determination of Hg(II) ions in Persian Golf fish samples, Benni (*Barbus sharpie*) and Biyaah (*Liza abu*) which were purchased from Abadan local fish market. 500 mg of each dried sample was initially digested as described in Section 2.3 and then subjected to the proposed procedure. The recovery of the spiked standard solutions as shown in Table 4 was in the range 95–103% which demonstrates the good recovery of the method.

4.3. Analysis of water samples

The drinking water supplied from Water Safe Company and Abadan petrochemical wastewater samples were passed through the mini-column at optimum conditions, after adding appropriate amounts of K$_2$S$_2$O$_8$ and HNO$_3$ for digestion and removal of chloride ion and organic compounds. The recovery test was also conducted

Table 3
Determination of mercury in a certified reference material

Certified reference material	DORM-3
Certified[a] (μg g^{-1})	0.409 (±0.027)
Found[a] (μg g^{-1})	0.405 (±0.078)
Relative error (%)	0.98

[a] Values in parentheses are confidence limit (95%) based on five replications.

Table 4
Determination of Hg(II) in fish samples

Fish sample	Added (ng mL^{-1})	Found (ng mL^{-1})	Recovery (%)
Liza abu[b]	–	0.06 ($\pm$0.01)[a]	–
	0.4	0.46 ($\pm$0.02)	100.0
	0.8	0.86 ($\pm$0.03)	100.0
Barbus sharpie[c]	–	0.38 ($\pm$0.01)	–
	0.4	0.77 ($\pm$0.02)	98.7
	0.8	1.16 ($\pm$0.04)	99.1

[a] Values in parentheses are confidence limit (95%) based on five replications.
[b] Amount of mercury in *Liza abu* was 31.5 ng g^{-1}.
[c] Amount of mercury in *Barbus sharpie* was 189.2 ng g^{-1}.

Table 5
Determination of Hg(II) in water samples

Sample	Added (ng mL^{-1})	Found (ng mL^{-1})	Recovery (%)
Water Safe Company	–	ND	–
	0.4	0.38 ($\pm$0.01)[a]	95.0
	0.8	0.78 ($\pm$0.02)	97.5
Abadan Petrochemical Wastewater	–	0.35 ($\pm$0.02)	–
	0.4	0.74 ($\pm$0.03)	98.7
	0.8	1.14 ($\pm$0.02)	99.1

ND: not detected.
[a] Values in parentheses are confidence limit (95%) based on five replications.

to evaluate the feasibility of the method. Each water sample was spiked with two standard solutions. The results are listed in Table 5. According to the results, the concentration of Hg(II) in the Abadan petrochemical wastewater sample is out of linear calibration range of the proposed method, but because of its complex matrix, this sample was also diluted and analyzed by the proposed method.

5. Conclusion

The proposed method offers a simple, inexpensive and selective method for the enrichment of Hg(II). Up to our knowledge, this is the first application of modified agar as an adsorbent for solid phase extraction. The reaction cell-gas liquid separator used in this procedure provides high sensitivity, which is comparable or better than some of the previously reported methods (Table 6). Since the agar–agar powder is a neutral and natural product, it has no side effects on the human health and the environment, in comparison with solid phases that have been used in other similar preconcentration procedures [18,20,30]. Additionally, low volumes of acetone were consumed for the preparation of modified agar mini-column. The preconcentration factor of this method is 100, which is better

Table 6
Comparison of the proposed method with some of the methods reported in literature

System	Linear range (ng mL^{-1})	DL (ng mL^{-1})	Ref.
Diphenylthiocarbazone complex on cellulose column	0–2000	2	[3]
Imprinted diazoaminobenzene vinylpyridine copolymers	0.13–25	0.05	[30]
Silica gel modified With diaminothiourea	–	0.28	[31]
C$_{18}$-catridge and *p*-sulfobenzylidenerhodanine	10–1200	0.02	[32]
Cation-exchanger Dowex 50Wx4	–	0.027	[33]
Sol–gel doped with Cyanex 301	0.1–2	0.033	[34]
Agar powder modified by 2-mercaptobenzimidazole	0.04–2.4	0.02	Present work

than some of the similar procedures. The proposed method can be successfully applied for separation and preconcentration of Hg(II) ions from water, wastewater and fish samples.

References

[1] R. Eisler, Mercury Hazards to Living Organisms, 1st ed., Taylor & Francis, 2006, pp. 155–156.
[2] S.M. Park, H.S. Choi, Sensitized spectrophotometric determination of trace Hg(II) in benzalkonium chloride media, Anal. Chim. Acta 459 (2002) 75–81.
[3] N. Rajesh, M.S. Hari, Spectrophotometric determination of inorganic mercury(II) after preconcentration of its diphenylthiocarbazone complex on a cellulose column, Spectrochim. Acta A 70 (2008) 1104–1108.
[4] D. Karunasagar, J. Arunachalam, S. Gangadharan, Development of a 'collect and punch' cold vapor inductively coupled plasma mass spectrometric method for the direct determination of mercury at nanograms per litre levels, J. Anal. Atom. Spectrom. 13 (1998) 679–682.
[5] X. Zhu, S.D. Alexandratos, Determination of trace levels of mercury in aqueous solutions by inductively coupled plasma atomic emission spectrometry: elimination of the 'memory effect', Microchem. J. 86 (2007) 37–41.
[6] N.L.D. Filho, D.R. do Carmo, Study of an organically modified clay: selective adsorption of heavy metal ions and voltammetric determination of mercury(II), Talanta 68 (2006) 919–927.
[7] X. Li, Z. Wang, Determination of mercury by intermittent flow electrochemical cold vapor generation coupled to atomic fluorescence spectrometry, Anal. Chim. Acta 588 (2007) 179–183.
[8] P.R. Devi, T. Gangaiah, G.R.K. Naidu, Determination of trace metals in water by neutron activation analysis after preconcentration on a poly(acrylamidoxime) resin, Anal. Chim. Acta 249 (1991) 533–537.
[9] D.L. Tsalev, Vapor generation or electrothermal atomic absorption spectrometry?—both!, Spectrochim. Acta B 55 (2000) 917–933.
[10] E.T. Ritschorff, N. Fitzgerald, R.L. Mcluaughlin, I.D. Brindle, The use of a modified multimode sample introduction system for the simple and rapid determination of cadmium by chemical vapor generation atomic absorption spectrometry, Spectrochim. Acta B 60 (2005) 139–143.
[11] M. Tuzen, M. Soylak, Mercury contamination in mushroom samples from Tokat, Turkey, Bull. Environ. Contam. Toxicol. 74 (2005) 968–972.
[12] M. Wilhelm, F. Muller, H. Idel, Biological monitoring of mercury vapour exposure by scalp hair analysis in comparison with blood and urine, Toxicol. Lett. 88 (1996) 221–226.
[13] M.V.B. Krishna, D. Karunasagar, S.V. Rao, J. Arunachalam, Preconcentration and speciation of inorganic and methyl mercury in waters using polyaniline and gold trap-CVAAS, Talanta 68 (2005) 329–335.
[14] M.S. Hosseini, H.H. Moghadam, Sensitized extraction spectrophotometric determination of Hg(II) with dithizone after its flotation as ion-associate using iodide and ferroin, Talanta 67 (2005) 555–559.
[15] M. Garrido, M.S. Di Nezio, A.G. Lista, M. Palomeque, B.S. Band, Cloud-point extraction/preconcentration on-line flow injection method for mercury determination, Anal. Chim. Acta 502 (2004) 173–177.
[16] B.Y. Spivakov, G.I. Malofeea, O.M. Petrukhin, Solid-phase extraction on alkylbonded silica gels in inorganic analysis, Anal. Sci. 22 (2006) 503–519.
[17] E. Zolfonoun, A. Rouhollahi, A. Semnani, Solid-phase extraction and determination of ultra trace amounts of lead, mercury and cadmium in water samples using octadecyl silica membrane disks modified with 5,5′-dithiobis (2-nitrobenzoic acid) and atomic absorption spectrophotometry, Int. J. Environ. Anal. Chem. 88 (2008) 327–336.
[18] M.A.H. Hafez, I.M.M. Kenawy, M.A. Akl, R.R. Lashein, Preconcentration and separation of total mercury in environmental samples using chemically modified chloromethylated polystyrene-PAN (ion-exchanger) and its determination by cold vapor atomic absorption spectrometry, Talanta 53 (2001) 749–760.
[19] T. Duan, X. Song, J. Xu, P. Guo, H. Chen, H. Li, Determination of Hg(II) in waters by on-line preconcentration using Cyanex 923 as a sorbent—cold vapor atomic absorption spectrometry, Spectrochim. Acta B 61 (2006) 1069–1073.
[20] N. Pourreza, M. Behpour, Column preconcentration of mercury as HgI$_4$$^{2-}$ using methyltrioctylammonium chloride–naphthalene adsorbent with subsequent anodic stripping-differential pulse voltammetric determination, Anal. Chim. Acta 481 (2003) 23–28.
[21] N. Pourreza, K. Ghanemi, Determination of copper by flame atomic absorption spectrometry after solid-phase extraction, Spectrosc. Lett. 39 (2006) 127–134.
[22] M. Soylak, I. Narin, M. Dogan, Trace enrichment and atomic absorption spectrometric determination of lead, copper, cadmium and nickel in drinking water samples by use of activated carbon column, Anal. Lett. 30 (1997) 2801–2810.
[23] M.E. Mahmoud, M.M. Osman, M.E. Amer, Selective pre-concentration and solid phase extraction of mercury(II) from natural water by silica gel-loaded dithizone phases, Anal. Chim. Acta 415 (2000) 33–40.

[24] V. Camel, Solid phase extraction of trace elements, Spectrochim. Acta B 58 (2003) 1177–1233.
[25] M. Fuchigami, A. Teramoto, Y. Jibu, Texture and structure of pressure-shift-frozen agar gel with high visco-elasticity, Food Hydrocolloids 20 (2006) 160–169.
[26] S.S. Singh, V.K. Aswal, H.B. Bohidar, Structural studies of agar–gelatin complex coacervates by small angle neutron scattering, rheology and differential scanning calorimetry, Int. J. Biol. Macromol. 41 (2007) 301–307.
[27] A.G. Howard, P.J. Statham, Inorganic Trace Analysis, 1st ed., John Wiley, 1997, pp. 82–84.
[28] F.A. Silva, I.L. Alcantara, P.S. Roldan, Florentino, P.M. Padilha, Determination of Hg in water by CVAAS using 2-aminothiazole modified silica, Eclet. Quim. 30 (2005) 47–55.
[29] K. Helrich, Official Methods of Analysis, vol. 1, 15th ed., 1990, pp. 263–266.
[30] Y. Liu, X. Chang, D. Yang, Y. Guo, S. Meng, Highly selective determination of inorganic mercury(II) after preconcentration with Hg(II)-imprinted diazoaminobenzene–vinylpyridine copolymers, Anal. Chim. Acta 538 (2005) 85–91.
[31] Q. Wu, X. Chang, Q. He, Y. Zhai, Y. Cui, X. Huang, Silica gel modified with diaminothiourea as selective solid-phase extractant for determination of Hg(II) in biological and natural water samples, Int. J. Environ. Anal. Chem. 88 (2008) 245–254.
[32] Y. Zhang, G. Yang, Q. Hu, Y. Jiayuan, Z. Li, Solid phase extraction and spectrophotometric determination of mercury in tobacco and tobacco additives with p-sulfobenzylidenerhodanine as chromogenic reagent, Microchim. Acta 146 (2004) 297–302.
[33] A. Krata, K. Pyrzyńsk, E. Bulska, Use of solid-phase extraction to eliminate interferences in the determination of mercury by flow-injection CV AAS, Anal. Bioanal. Chem. 377 (2003) 735–739.
[34] F. Mercader-Trejo, E.R. San Miguel, J. Gyves, CVAAS determination of naturally occurring levels of mercury in sediments after on-line preconcentration with a sol–gel sorbent doped with CYANEX-301, J. Anal. Atom. Spectrom. 20 (2005) 1212–1217.