Spectrophotometric determination of Mercury in environmental samples using 5-methylthiophene-2-carboxaldehyde ethylenediamine (MTCED)

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ABSTRACT

A simple, rapid and sensitive spectrophotometric method was developed for the determination of Hg (II) using 5-methylthiophene–2-carboxaldehyde ethylenediamine (MTCED) as an analytical reagent. The reagent has been synthesized and characterized using IR, 1H NMR and Mass spectral data. The metal ion in aqueous medium forms light yellow colored complex with MTCED at (pH = 2.0) (sodium acetate-hydrochloric acid buffer solution) showing maximum absorbance at 385 nm. Hence, analytical studies were further carried out at 385 nm. The reagent reacts with mercury in acidic medium to form light yellow colored 1:1 (M:L) complex. The color reactions are instantaneous and absorbance values remain constant for 24 hours. The composition of the Hg (II) complex with MTCED was studied by the method of Job’s continuous variation and molar ratio method. Beer’s law was obeyed in the range of 0.83-8.6 µg ml\(^{-1}\) of Hg (II). The molar absorptivity and sandell’s sensitivity of the method were found to be 5.58 x 10\(^4\) Lmol\(^{-1}\) cm\(^{-1}\) and 0.00179 µg/ cm\(^{-2}\) respectively. Since MTCED method is more sensitive, it was applied for the determination of mercury in environmental samples.

Key words: Mercury determination, non-extractive spectrophotometry, 5-methylthiophene –2-carboxaldehyde ethylenediamine (MTCED) in environmental samples.

INTRODUCTION

Mercury is one of the most toxic heavy metal in the earth and it exists in nature at trace and ultra trace amounts in three valence states [1]. Mercury (0, I, II) species and are able to combine with most inorganic and organic ligands to form various complexes, e.g. HgX\(_{4-x}\) (where X= Cl, Br and I) and methyl mercury [2,3]. Mercury can accumulate in animals and plants and also enters into human body through the food chain causing damage to central nervous system [4]. Due to the toxicological effects and potential accumulation of mercury onto human bodies and aquatic organisms, the determination of mercury (II) or organo mercury (II) has seen an upsurge of interest in the last few years [5]. According to WHO, the allowed limits of mercury in drinking water are less than (1.0 ngmL\(^{-1}\)) [6].

Mercury is a serious environmental pollutant because of its toxic effects on all living organisms [7]. Mercury and its compounds cause serious diseases such as leukemia [8]. Mercury compounds can be present as a result of anthropogenic activities in various environmental samples [9]. They are usually present in natural waters at trace levels [9] and [10]. The lakes, rivers in vicinity of the industrial areas are the important indicators for mercury pollution. So, it needs to develop new, selective, effective, cheap methods for determination of mercury [11].

A serious problem in the determination of mercury is related to low concentrations of target species. The main species of mercury in natural waters are inorganic mercury (Hg\(_2^{2+}\), Hg\(^{2+}\)) and methyl mercury (CH\(_3\)Hg\(^+\)). Recent reports estimate that total mercury concentration is in the range of 0.2-100 ng L\(^{-1}\) and methyl mercury concentrations are lower (ca. 0.05 ngL\(^{-1}\)) in natural waters[12].
Mercury compounds have been used as catalysts, fungicides, herbicides, disinfectants, pigments and for other purposes. The world production was about 10,000 t in 1973 [13] and about 6,500 t in 1980 [14]. In addition to the production of pure mercury by industrial processes, mercury is released into the environment by human activities such as the combustion of fossil fuels, waste disposal and by industry. Recent estimates of anthropogenic emissions are in the order of 2,000 to 3,000 t year\(^{-1}\) [15-18].

Mercury is one of the most toxic heavy metals. It enters the environment as metallic, inorganic and organic mercury compounds through various industries like pulp and paper industry, chlor-alkali plants, gold and silver mining, electrical industry, paints, fungicides and pharmaceuticals. Fossil fuel burning and cement manufacture cause emission of mercury in air [19–21]. The average value of mercury reported in oil is 10 ppm and sludge from sewage treatment plant is reported to contain up to 25 ppm mercury [22]. WHO has suggested intake of 0.3 mg of mercury of which not more than 0.2 mg should be in the form of methyl mercury and maximum permissible value in drinking water is 0.001 mg·l\(^{-1}\) [23]. The toxicity of mercury depends on its chemical species and it is found that organomercurials are more toxic than inorganic mercury compounds [24,25]. Mercury and its compounds are reported to be mutagenic and teratogenic in nature [26].

The present study was planned to determine the prevalence of selected trace elements in environmental samples.

We used an ICP-OES machine which converts all lead forms into inorganic lead. Inductively coupled plasma optical emission spectrometry (ICP OES) and inductively coupled plasma mass spectrometry (ICP-MS) are very feasible techniques for quantitative determination of the Hg (II). The most important advantages of these techniques are high sample throughput, simplicity and good sensitivity in comparison to other techniques [27-28] used for Hg (II) determination.

Nowadays, due to the higher sensitivity achieved with axially viewed plasma and better spectral resolution given by high-resolution monochromators, it is expected that low concentrations of all naturally occurring lead may be directly quantified by ICP OES. Furthermore, depending on the nebulizer used to introduce the sample solution in the plasma, sensitivity improves.

This paper describes synthesis, characterization and analytical properties of new reagent viz., 5-methylthiophene–2-carboxaldehyde ethylenediamine (MTCED). Since the reagent is more sensitive, it is used for the determination of mercury in various water samples.

**MATERIALS AND METHODS**

**Apparatus**
Schimadzu 160A UV-Visible spectrophotometer (PerkinElmer Singapore Private Limited, Singapore) equipped with 10 cm quartz cell and an ELICO model L1-610 pH meter (M/s ELICO private limited, Hyderabad, India) were used in the present study. ICP-OES (Inductively Coupled Plasma-Optical Emission Spectrometry Model-7000) methods were used for the quantitative analysis of Hg (II). To determine the Hg (II), AOAC methods were used (AOAC 1986, 2003; Jorhem 1993). In this method, the samples were dissolved at 190°C and 400 psi pressure in Mars 5 apparatus (Vessel Type XKP 1500, CEM, Matthews, USA). The Hg (II) was analyzed by inductively coupled plasma-optical emission spectrometry (Varian Vista-MPX CCD Simultaneous Spectrophotometer, Mugrave-Victoria, Australia) [29-30]. (Mandal & Suzuki 2002).

**Reagent and solutions**
All chemicals used were of analytical-reagent grade of the highest purity available procured from Merck. Doubly distilled de-ionized water was used throughout the experiment. Glass vessels were cleaned soaking in acidified solutions of \(K_2Cr_2O_7\) followed by washing with conc. HNO\(_3\) and were rinsed several times with high purity de-ionized water. Stock solutions environmental water samples and medicinal leave samples were kept in polypropylene bottle containing 1ml of conc. HNO\(_3\).

**Preparation of reagent (MTCED)**
5-methylthiophene-2-carboxaldehyde (1 ml, 0.0092 mol) in 50 ml of methanol, ethylenediamine (0.55 g, 0.0092 mol) dissolved in 50 ml of methanol were taken in 250 ml round bottom flask. Suitable quantity (1ml) of 1 molar sodium acetate was added to the reaction mixture and refluxed for 10 hours. On cooling the reaction mixture wheat colored product was separated out. It was collected by filtration and washed several times with hot water followed by n-hexane. This compound was recrystallised from methanol and dried in vacuum. The ligand was stable for more than 6 months. Yield is 93; m.p.82-83 °C. The structure of MTCED as shown in the (Fig.1).
Characterization of reagent (MTCED)
The reagent has been synthesized and characterized by IR, $^1$HNMR and Mass spectral data. Infrared spectrum of MTCED shows bands at 3087, 2915, 2830, 1634, 1476, 1375, 1305, 706 and 593 respectively corresponding to $\nu$(C-H) stretch (sp$^2$---C-H) (thiophene), $\nu$(C-H) stretch (methyl and methylene), $\nu$(C = N) schiffbase, $\delta$ (-CH$_2$) symmetrical stretch , $\delta$ (-CH$_3$) symmetrical stretch, $\nu$(C - N) stretch, $\nu$(C - S) stretch and Hetero cyclic ring C – C oop bend . $^1$HNMR spectrum of MTCED (CDCl$_3$+DMSO-d$_6$) showed signals at 2.45 (3H), 6.74-7.16 (2H), 3.30(2H) due to methyl protons, thiophene protons, methylene protons. Mass spectrum of MTCED shows signal at 277 (M+1) corresponding to its molecular ion peak. The molecular formula of the reagent is C$_{14}$H$_{16}$N$_2$S$_2$ (M.Wt, 276).

pKa values of reagents
The pKa values were determined by recording the UV-Visible spectra of 1X10$^{-4}$M solutions of the reagent at various pH values and by taking the arithmetic mean of the values obtained from the measurements at different wave lengths determined spectrophotometrically using Phillips and Merrit method. The values of deprotonation of MTCED were 9.0 ($pK_1$= $pK_2$).

MTCED solution
A (1X10$^{-3}$M) solution was prepared by dissolving 0.276 gm of MTCED in 100 ml of methanol. The reagent solution is stable for at least 24 h.

Hg (II) Standard solution
A (1X10$^{-3}$M) stock solution of mercury was prepared by dissolving 0.272 g of mercuric chloride (Hgcl$_2$) (Merck Darmstadt) in double distilled water containing few drops of conc. Hcl and made up to the mark in a 100 ml volumetric flask.

1000 ppm stock solution of mercury was prepared by dissolving 0.13535 gm of mercuric chloride in 100 ml distilled water or 1.3535 gm of mercuric chloride in 1000 ml distilled water.

Buffer Solution
1M Sodium acetate + 0.1M hydrochloric acid (pH= 0.5 – 3.0), 0.2M Sodium acetate + 0.2M acetic acid (pH = 3.5 – 6.0), 1M Sodium acetate + 0.2M acetic acid (pH= 6.5 – 7.5), 2M Ammonia + 2M ammonium chloride (pH= 8.0 – 12.0) buffer solutions are prepared in distilled water. Suitable portions of these solutions are mixed to get the desired pH.

Potassium permanganate solution
A 1% potassium permanganate solution was prepared by dissolving in de-ionized water. Aliquots of this solution were standardized with oxalic acid.

Tartrate solution
A 100 ml stock solution of tartrate (0.01% w/v) was prepared by dissolving 10 mg of ACS grade (99%) potassium sodium tartrate tetrahydrate in (100 ml) de-ionized water.

Aqueous ammonia solution
A 100 ml solution of aqueous ammonia was prepared by diluting 10 ml concentrated NH$_3$ (28–30%, ACS grade) to 100 ml with de-ionized water. The solution was stored in a polypropylene bottle.
Preparation of Sample solutions

Preparation of water samples

Different water samples were collected from various places around Tirupati, A.P., and India. The samples (150 ml) were stored at 5°C in metal free polyethylene bottles. Water samples were filtered through whatman filter paper no. 41 and collected into 250 ml beakers. All the filtered environmental water samples were evaporated nearly to dryness with a mixture of 10 ml of HNO₃ and 5 ml of H₂SO₄ in a fume cupboard and then cooled to room temperature. The sample was digested in the presence of an excess potassium permanganate solution according to the method recommended by Fifield et al. The residues were then heated with 10 ml of deionized water in order to dissolve the salts. The solutions were cooled and neutralized with dilute NH₄OH. The digest was transferred into a 25 ml calibrated flask and diluted up to the mark with deionized water.

The reaction of Hg (II) was tested with 5-methylthiophene–2-carboxaldehyde ethylenediamine (MTCED) at different pH values. The samples were prepared in 10 ml of the solution containing constant volume of 2 ml of Hg (II), 2 ml of buffer solution (pH = 2.0), 1 ml of 1x10⁻² M MTCED, 2 ml lithium chloride solution and different metal ions. The solution was then shaken with 10 ml portions of methanol for two minutes and the allowed to stand. Each time the combined organic phase of aliquots were taken in 10 ml standard flasks and made up to the mark with methanol. The absorbance was measured in 200-800 nm range against reagent blank.

Recommended procedure

An aliquot of the sample solution was taken in 25 ml standard flask containing 10 ml of buffer solution of (pH = 2.0) and 1 ml of 1x10⁻⁴ MTCED reagent solution and 1 ml of Hg (II) solution made up to the mark with distilled water. The absorbance of the complex was measured against the reagent blank at 385 nm. The absorbance values were referred to the predetermined calibration plot to compute the amount of mercury.

The author has introduced a new sensitive reagent MTCED for the extractive spectrophotometric determination of trace amounts of Hg (II).

The proposed method when compared with other spectrophotometric methods is more sensitive and selective. It also offers advantages like reliability and reproducibility in the addition to its simplicity instant color development and less interferences. The results obtained through UV-Visible spectrophotometer have been compared with those obtained through the ICP-OES.

RESULTS AND DISCUSSION

Mercury reacts with 5-methylthiophene–2-carboxaldehyde ethylenediamine (MTCED) in sodium acetate-hydrochloric acid buffer solution of pH 2.0 and gives 1:1 light Yellow colored complex. The complex has a maximum absorbance at 385 nm. The optimum reaction conditions for the quantitative determination of the metal-ligand complex was established through a number of preliminary studies, such as the effect of pH, reagent concentration, interference of foreign ions, in order to develop a rapid, selective and sensitive extractive spectrophotometric method for the determination of Hg (II) at microgram levels.

Absorption spectra of the reagent and Hg (II)-MTCED complex

Absorption spectra of Hg (II)-MTCED complex and reagent show maximum absorbance at 385 nm and 340 nm respectively (Fig.2). The reagent showed minimum absorbance at the wavelength of maximum absorbance of the complex. Hence, all the spectral measurements of the complex have been carried out at 385 nm.

The study of the effect of pH on the color intensity of the reaction mixture showed that the constant and maximum color is obtained in the pH range 1.0- 3.0, the complex has maximum absorbance in buffer solution of (pH = 2.0). The analytical studies were therefore, carried out at (pH = 2.0).

Different molar excess of MTCED were added to Hg (II) concentration and the absorbances were measured adopting the standard procedure. It was observed that 5 fold molar excess of reagent with respect to metal ion is necessary to get maximum absorbance. Hence, a 5 fold molar excess of reagent was used for further experimental studies.

The absorbance of the solution was measured at different time intervals to ascertain the time stability of the color complex. It was observed that the color development was instantaneous and remained constant for more than 24 hrs. Physicochemical and analytical properties of Hg (II) complex of MTCED were summarized in Table 1.
Table 1. Physico-chemical and analytical characteristics of Hg (II) – MTCED complex

<table>
<thead>
<tr>
<th>S.No.</th>
<th>Characteristic Property</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Absorbance Maximum ( \lambda_{\text{max}} ) (nm)</td>
<td>385</td>
</tr>
<tr>
<td>2</td>
<td>pH – range (optimum)</td>
<td>1.0 – 3.0</td>
</tr>
<tr>
<td>3</td>
<td>Mole of reagent required mole of metal ion for full color developed</td>
<td>5 Fold</td>
</tr>
<tr>
<td>4</td>
<td>Beer’s law validity range (µg/ml)</td>
<td>0.83-8.6</td>
</tr>
<tr>
<td>5</td>
<td>Molar absorptivity (L mol(^{-1}) cm(^{-1}))</td>
<td>5.58 x 10(^4)</td>
</tr>
<tr>
<td>6</td>
<td>Specific absorptivity (ml g(^{-1}) cm(^{-1}))</td>
<td>0.11710</td>
</tr>
<tr>
<td>7</td>
<td>Sandell’s sensitivity (µg/cm(^2))</td>
<td>0.00179</td>
</tr>
<tr>
<td>8</td>
<td>Composition of complex as obtained in Job’s and molar ratio methods (M:L)</td>
<td>1 : 1</td>
</tr>
<tr>
<td>9</td>
<td>Stability constant of the complex</td>
<td>1.308 x 10(^6)</td>
</tr>
<tr>
<td>10</td>
<td>Relative standard deviation (RSD)%</td>
<td>0.27052</td>
</tr>
<tr>
<td>11</td>
<td>Y-intercept</td>
<td>+0.01827</td>
</tr>
<tr>
<td>12</td>
<td>Angular coefficient (m)</td>
<td>0.09993</td>
</tr>
<tr>
<td>13</td>
<td>Correlation coefficient(( \psi ))</td>
<td>0.99876</td>
</tr>
</tbody>
</table>

Fig. 2. Absorption spectra of (a). Hg (II) – MTCED complex (\( \lambda_{\text{max}} = 385 \) nm) in aqueous solution, (b). MTCED Vs Water blank (1 x \( 10^{-4} \) M)

Adherence of the Hg (II) – MTCED complex system to Beers law

For the possible determination of Hg (II) at micro level, the absorbance of the solution containing different amounts of the metal ion is measured at 385 nm. The linear plot between the absorbance and the amount of Hg (II) is drawn and the straight line obtained with the equation \( A_{385} = 0.09993C + 0.01827 \) (Fig.3). Further Beers law is obeyed in the range of 0.83-8.6 µg/ml, the molar absorptivity and sandell’s sensitivity were found to be 5.58 x 10\(^4\) L.mol\(^{-1}\)cm\(^{-1}\) and 0.00179 µg/cm\(^2\) respectively. The standard deviation of the method for ten determinations of 1.91 µg/ml is ±0.00158. The results showed that standard deviation of the method was not more than 0.00158 and relative standard deviation was less than 0.27052. These results indicate that the method has good precision, besides being accurate.

Interference of Foreign Ions

The effect of various cations and anions which are generally associated with the metal ion in the determination of Hg (II) was studied by measuring the absorbance of mercury. The complex contains 1.91 µg/ml of Hg (II) in solution. The colour of the reaction was developed as described in the standard procedure. An error of ±2% in the absorbance reading was considered tolerable. The tolerance limit (TL) values in ppm for various anions and cations in the MTCED methods respectively were presented in Table 2. Higher amounts of Fe\(^{2+}\) do not interfere in the presence of 70 ppm of fluoride. Larger amounts of Hg\(^{2+}\) do not interfere in the presence of 600 ppm of iodide.
The present method 5-methylthiophene–2-carboxaldehyde ethylenediamine (MTCED) was applied for the determination of mercury when present alone and present in water samples.

The present ligands containing thiophene ring are found to be potential and cost effective for the determination of Hg (II) without the need for extraction using the toxic solvents. Further the reagents are easy to synthesize using commercially available precursors. Moreover the present method is simple, rapid and very sensitive for non-extractive spectrophotometric determination of Hg (II) in aqueous medium.

![Fig.3. Calibration plot for Hg (II) determination](Image)

**Effect of foreign ions on the extraction of the Hg (II) –MTCED complex**

The effect of foreign ion is studied by measuring the absorbance of the reaction mixture containing 1.91 µg/ml of Hg (II) in the presence of different amounts of foreign ions. The results presented in the Table 2. An error of ± 2% in the absorbance value caused by foreign ions is considered as a tolerable limit.

<table>
<thead>
<tr>
<th>Ion Added</th>
<th>Tolerance limit µg/ml</th>
<th>Ion Added</th>
<th>Tolerance limit µg/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tartrate</td>
<td>594</td>
<td>W(V)</td>
<td>365</td>
</tr>
<tr>
<td>Iodate</td>
<td>509</td>
<td>Mn (II)</td>
<td>2.3</td>
</tr>
<tr>
<td>Urea</td>
<td>288</td>
<td>Pb (II)</td>
<td>8.3</td>
</tr>
<tr>
<td>Citrate</td>
<td>386</td>
<td>Cr (VI)</td>
<td>1.2</td>
</tr>
<tr>
<td>Bicarbonate</td>
<td>245</td>
<td>Ti (III)</td>
<td>0.41</td>
</tr>
<tr>
<td>Thiocyanate</td>
<td>234</td>
<td>Cd (II)</td>
<td>0.23</td>
</tr>
<tr>
<td>Sulphate</td>
<td>386</td>
<td>Hg(II)</td>
<td>0.40</td>
</tr>
<tr>
<td>Oxalate</td>
<td>351</td>
<td>Ni (II)</td>
<td>0.23</td>
</tr>
<tr>
<td>Thiourea</td>
<td>303</td>
<td>Fe (II)</td>
<td>0.21</td>
</tr>
<tr>
<td>Nitrate</td>
<td>249</td>
<td>Au (III)</td>
<td>0.42</td>
</tr>
<tr>
<td>Acetate</td>
<td>236</td>
<td>Pt (IV)</td>
<td>0.38</td>
</tr>
<tr>
<td>Phosphate</td>
<td>38</td>
<td>Pd(II)</td>
<td>0.24</td>
</tr>
<tr>
<td>Bromide</td>
<td>34</td>
<td>Ag (I)</td>
<td>0.21</td>
</tr>
<tr>
<td>Chloride</td>
<td>15</td>
<td>V (V)</td>
<td>0.11</td>
</tr>
<tr>
<td>Fluoride</td>
<td>7.9</td>
<td>Cu (II)</td>
<td>0.13</td>
</tr>
</tbody>
</table>

**Composition and stability constant of the complex**

Job’s method of continuous variation and molar-ratation methods were applied to ascertain the stoichiometric composition of the complex. It was found that MTCED forms 1:1 complex with Hg (II) as shown in the (Fig.4).
The proposed extractive spectrophotometric method is applied for the determination of Hg (II) in water samples. A known aliquot of the above sample solutions were taken into a 25 ml separating funnel and the lead content was determined as described is given in the general procedure. The results were checked with parallel determinations by direct ICP-OES. The data obtained in the analyses of water samples were given in Table 3.

Table – 3. Determination of trace amount of Hg (II) in water samples.

<table>
<thead>
<tr>
<th>Sample Name</th>
<th>Hg (II) found µg/ml</th>
<th>ICP-OES</th>
<th>S.D</th>
<th>R.S.D(%)</th>
<th>Proposed Methoda</th>
<th>S.D</th>
<th>R.S.D(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sea waterb</td>
<td>1.07</td>
<td>0.00114</td>
<td>0.10640</td>
<td>1.013</td>
<td>0.00011</td>
<td>0.01085</td>
<td></td>
</tr>
<tr>
<td>River waterc</td>
<td>1.26</td>
<td>0.00011</td>
<td>0.00885</td>
<td>1.242</td>
<td>0.00011</td>
<td>0.00885</td>
<td></td>
</tr>
<tr>
<td>Drain waterd</td>
<td>3.44</td>
<td>0.00008</td>
<td>0.00240</td>
<td>3.328</td>
<td>0.00011</td>
<td>0.00330</td>
<td></td>
</tr>
<tr>
<td>Tap watere</td>
<td>1.03</td>
<td>0.0001</td>
<td>0.00777</td>
<td>1.023</td>
<td>0.00011</td>
<td>0.01075</td>
<td></td>
</tr>
<tr>
<td>River waterf</td>
<td>0.963</td>
<td>0.00011</td>
<td>0.01155</td>
<td>0.952</td>
<td>0.00008</td>
<td>0.00840</td>
<td></td>
</tr>
</tbody>
</table>

a. Average of the five determination; b. Collected at Bay of Bengal (Chennai), India.; c. Collected at Tungabhadra river (Kurnool), A.P., India.
d. Collected at Anantapur town, A.P., India.; e. Collected at Anantapur Municipal Storage tank, A.P., India.; f. Collected at Swarnamukhi (Srikalahasti), A.P., India.

CONCLUSION

The proposed spectrophotometric method is simple, highly sensitive and selective for the determination of Hg (II) in water samples. The proposed method is simple, rapid and common metal ions such as Fe³⁺, Pb²⁺, Co²⁺, Ni²⁺, Mn²⁺, Zn²⁺, Cr³⁺ do not interfere. Urea, bicarbonate, citrate, sulphide, SO₄²⁻, and PO₄³⁻ also do not interfere. The proposed method is more sensitive than some reported methods in the literature and has been successfully applied for the determination of mercury in various environmental samples.

In this paper a new simple, sensitive, selective, and inexpensive method with the Hg (II)–MTCED complex was developed for the determination of mercury in industrial, environmental samples for continuous monitoring to establish the trace levels of mercury in difficult sample matrices. It offers also a very efficient procedure for speciation analysis. Although many sophisticated techniques such as pulse polarography, HPLC, AAS, ICP-AES, and ICP-MS are available for the determination of lead at trace levels in numerous complex materials, factors such as

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as the low cost of the instrument, easy handling, lack of requirement for consumables and almost no maintenance have caused spectrophotometry to remain a popular technique, particularly in laboratories of developing countries with limited budgets. The sensitivity in terms of molar absorptivity and precision in terms of relative standard deviation of the present method are very reliable for the determination of mercury in real samples down to ng g⁻¹ levels in aqueous medium at room temperature (25±5°C).

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