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Spectrophotometric Determination of Trace Hg (II) Using 6-(Anthracen-2-yl)-2, 3- dihydro -1, 2, 4- triazine-3-tione as a newly Synthesized Reagent

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ABSTRACT

A simple and accurate spectrophotometric method for Hg determination with a newly synthesized reagent, 6-(2-anthracenyl) - 2, 3-dihydro-1, 2, 4-triazine-3-thione (ADTT), as a selective analytical reagent has been developed. Determination of trace Hg (II) in environmental and industrial samples was of great importance because of its health hazard for humans. Many analytical methodologies have been proposed for determination of Hg based on atomic absorption spectroscopy and HPLC. Although these methods are selective and sensitive, they have some disadvantages, requiring expensive equipment and toxic solvents, and often involving complex sample pre-treatments. Spectrophotometric methods were suitable for routine laboratories. In this paper we reported the synthesis of a new reagent, 6-(2-anthracenyl) - 2, 3-dihydro-1, 2, 4-triazine-3-thione (ADTT), and its applicability for Hg determination. 2-acethylanthracene was synthesized selectively according to the reaction of anthracene with acethylchloride under the special condition. Use of amyl nitrite and 2-acethylanthracene under anhydrous condition 2-(anthracen-2-yl) oxoacetaldehyde oxime produced, which upon reaction with thiosemicarbazone yielded 6-(2-anthracenyl)- 2,3-dihydro-1,2,4-triazine-3-thione. The yellow complex of Hg(II) with ADTT was formed in basic media according to a 1:3 stoichiometry. Beer's law was obeyed in the Hg concentration range of 5–30µg/ ml. The linear regression equation were $A= 0.010C – 0.014$ and $A= 0.023C – 0.010$ in λ_{max} 330nm and 254nm respectively. The limit of detection was 1 µg/ml. The relative standard deviations of 10 replicate determinations were 2.53% and 0.7% for determination of 4.5 and 16 µg/ml of Hg respectively. The method was applied to the determination of Hg (II) in amalgam sample with satisfactory results.

Keywords: 6-(2-anthracenyl) - 2, 3-dihydro-1, 2, 4-triazine-3-thione; Mercury (II); UV-VIS spectrophotometry

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INTRODUCTION

Toxic metals are bio-accumulative and relatively stable as well as carcinogenic and therefore require close monitoring. Mercury pollution has been considered as a major environmental issue and health hazard for humans because it may cause kidney toxicity, neurological damage, chromosome breakage and birth defects [1]. Strict regulations and guidelines have been developed limiting its level in water, environmental and food samples. Determination of trace Hg (II) is of great importance because of its high toxicity and accumulation in the environmental and human bodies but routine techniques are often not sensitive enough [2-4].

Many analytical methodologies have been proposed for determination of Hg (II) such as AAS, ICP-AES, ICP-MS and X-ray fluorescence spectroscopy [5-8]. Although these methods are selective and sensitive, they have some disadvantages, requiring expensive equipment and toxic solvents and often involving complex sample pre-treatment [9]. UV/Vis spectrophotometry using various chelating agents had a good sensitivity and reproducibility [10-13].

It has been reported that 6-(2-naphthyl)-2,3-dihydro-1,2,4-triazine thion (NDTT) forms colored complexes with Cu(II), Ni(II) and Hg(II) in basic media [14-16]. In this investigation the reacting moiety, triazin-3-thion, was left intact and the chromogenic part of the reagent, naphthyl, has been changed to anthracene and the new reagent, 6-(2-anthracenyl)-2,3-dihydro-1,2,4-triazine thion (ADTT), was synthesized as a possible more sensitive reagent for determination of Hg(II). ADTT reacts with Hg(II) forming a yellow colored complex in basic media.

The spectrophotometric method for the determination of Hg (II) using this reagent was validated. Some parameters such as pH, nature of the complex and optimum amount of the reagent were also investigated.

MATERIAL AND METHODS

Experimental

Instrumentation

Spectrophotometric measurements were made on a Schimadzu 160A UV/Vis spectrophotometer. Melting points were determined on a Kofler hot stage apparatus.

The ^{1}H- NMR spectra were obtained on a Varian 400 unity plus and chemical shifts (δ) were determined in ppm relative to the internal tetramethylsilane.

Reagents and solutions

All chemicals such as anthrancene, $AlCl_3$, acetyl chloride, thiosemicarbazide, amyl nitrite, tartaric acid and chloroform were of analytical grade and prepared from Merck (Darmstadt, Germany).

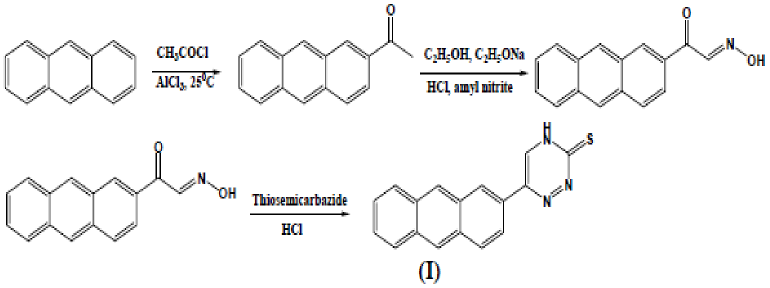
Stock solution (containing 1000 mg Hg per liter) of mercury (II) chloride, were purchased from Aldrich. The working solutions were prepared by proper dilution.

A 0.002 M solution of reagent (ADTT) was prepared freshly by dissolving suitable amount of the reagent in 1 M NaOH.

Synthesis of the reagent

6-(Anthracen-2-yl)- 2,3- dihydro-1,2,4- triazine-3-thion (ADTT) was synthesized according to our previous work except that the acethylation of anthrancene was done in our lab by the Friedel – Crafts acylation method[14].

To prepare 2- anthracenyl glyoxal aldoxim, 2-acethyl anthracene was added to a flask containing ethanol and sodium metal followed by slowly addition of amylnitrite under anhydrous condition. Then the product (2-anthracenyl glyoxal aldoxim) and thiosemicarbazide were refluxed in acidic media for 3 hours. The precipitate filtered and recrystalized from DMSO-H_2O (Scheme 1).



Scheme 1: Synthesis of 6-(anthracen-2-yl)-2,3- dihydro-1,2,4- triazine-3-thione.

General procedure

One ml of standard solution of Hg(II) (3-20 μg/ml), 1 ml of tartaric acid (1M), and 5 ml of reagent (0.002 M) were taken in a 100 ml separatory funnel and after mixing thoroughly, the resulting colored complex was extracted with 5, 3 and 2 ml of chloroform. The extracts were collected in a 10 ml volumetric flask and adjusted to the volume with chloroform. The absorbance of the solutions was recorded against reagent blank prepared in the same way but without Hg^{2+} solution.

RESULTS AND DISCUSSION

Absorption spectra of Hg- ADTT complex

6-(Anthracen-2-yl)- 2,3- dihydro-1,2,4- triazine-3-thion (ADTT) was used as a new reagent for Hg(II) determination. ADTT reacts with Hg(II) forming a yellow-colored complex in basic media which is extractable with chloroform. The spectra of Hg-ADTT complex were obtained in the wavelength region 200-600 nm. The complex shows maximum absorbance at 254 and 330 nm (Fig 1).

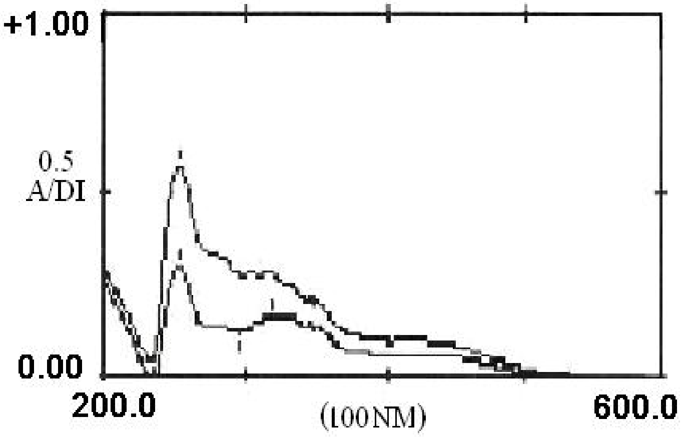


Figure 1: Absorption spectra of Hg(II)-ADTT complex in chloroform (Hg^{2+} = 15, 25 µg/ml)

Effect of pH

The pH of reaction medium plays an important role in metal- chelate formation and subsequent extraction. The effect of the pH on the complex formation and extraction of Hg-ADTT was studied by using borate buffer in the range of 5-14. The absorbance increased by increasing the pH value. The maximum extraction efficiencies were obtained at pH = 14 (Fig. 2). So the basic medium using NaOH 1 M was chosen for further extraction.

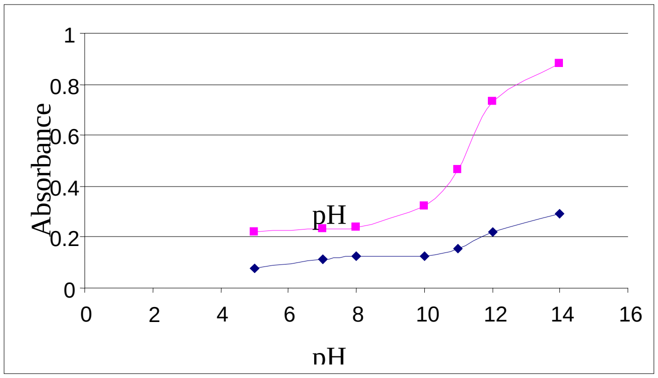


Figure 2: Effect of pH on the absorbance of Hg(II)-ADTT complex (λ = 254, 330nm)

Nature of the complex

To determine the composition of the complex the continuous variation method was used. The total molar concentration of Hg (II) and ADTT was held constant but the ratio of the components was varied. Absorbance measurements were carried out at two different wavelengths 254 and 330 nm. The maximum absorbance obtained at 0.25 mole fraction of Hg (II) indicated the ratio of (1:3) Hg (II) to ADTT in the complex (Fig. 3).

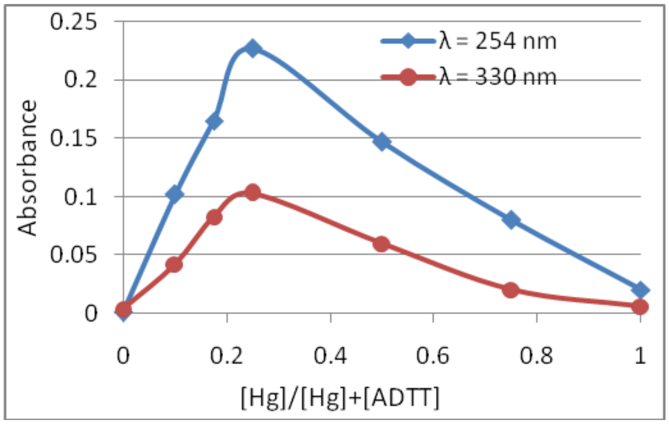


Figure 3: Composition of Hg(II)-ADTT complex at 254 and 330 nm by continuous variation method

Determination of optimum amount of ADTT

The concentration of Hg(II) was held constant (4.98 × 10^{-5} M) while the ligand concentration varied between 4.98 × 10^{-5} to 2.49 × 10^{-4} M. The absorbance of the complex was plotted as a function of the mole ratio of ligand to metal (L/M). The maximum absorbance was obtained at 1.49 × 10^{-4} M, showed ADTT/Hg (II) ratio of 3 at both wavelengths (254 and 330 nm) (fig. 4). At higher concentrations of ADTT the absorbance did not change. Therefore a little more than this ratio was used for all determinations.

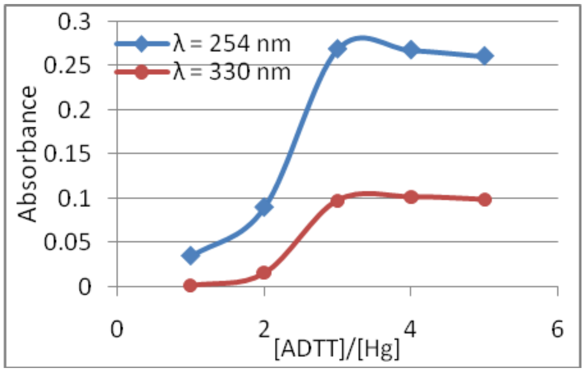


Figure 4: The effect of ADTT amount on the absorbance at 254 and 330 n ([Hg^{2+}] = 4.98 × 10^{-5})

Verification of Beer's law Validity

The calibration data for Hg (II) was constructed at 254 and 330 nm under optimum experimental condition. The plots reveal that the Beer's Law is obeyed in the range of 3-20 µg/ml at 254 nm and 5-20 µg/ml at 330 nm of Hg (II) with a correlation coefficient of $R^2 = 0.999$. The regression equations were $Y = 0.0239X – 0.011$and $Y = 0.010 X – 0.0 14$ for 254 and 330 nm respectively. The limit of quantification of Hg (II) was found to be 3 µg/ml. The detection limit (LOD) expressed as the concentration which is 3 times as large as that of the background was 1 µg/ml. Molar absorptivity was measured at 254 and 330 nm. The results are presented in Table 1.

Table 1: Analytical data of calibration curve and molar absorptivity of Mercury (II)-reagent

Parameters	λ (nm): 254	λ (nm): 330
Linearity range	3-20 µg/ml	5-20 µg/ml
Regration equation	$Y = 0.023 X – 0.010$	$Y = 0.010 X – 0.014$
Correlation coefficient (R$^{2)}$	0.999	0.999
Molar absorptivity	4.76×10^3	1.85×10^3

Accuracy and precision

The accuracy and precision of the method were determined in two sets of ten identical samples with a final content of 4.5 and 16 µg/ml of Hg (II). The concentration of Hg (II) was determined according to the recommended procedure. The results are illustrated in Table 2.

Table 2: Accuracy and precision data for determination of two sets of identical concentration of Mercury

Sample	**Added (µg)**	**Recovered (µg)**	**Added (µg)**	**Recovered (µg)**
1	4.50	4.60	16.00	16.10
2	4.50	4.40	16.00	15.90
3	4.50	4.60	16.00	16.20
4	4.50	4.60	16.00	16.20
5	4.50	4.30	16.00	16.10
6	4.50	4.40	16.00	16.10
7	4.50	4.40	16.00	16.20
8	4.50	4.40	16.00	16.00
9	4.50	4.50	16.00	16.30
10	4.50	4.60	16.00	16.10
Mean		4.48		16.12
SD		0.11		0.11
%CV		2.53		0.70
Recovery (%)		98.22		100.75

Effect of Foreign Ions

The effect of foreign ions was determined using a standard solution of Hg (II) containing 20 μg/ml. As it was shown before, the complexing moiety of the ligand is triazine-3-thion [10]. It was shown that most of the cations ($Fe2^+$, Ca^{2+}, $Sr2^+$, Cd^{2+}, Co^{2+}, Mg^{2+}, Ba^{2+}, $Mn2^+$, Ba^{2+}) had not interference on the determination of Hg(II) with ADTT. Nickel, copper and Pd (II) forms complexes with ADTT. To analyze Hg (II) in presence of these cations, derivative spectrophotometric method can be used.

CONCLUSION

It was concluded that the developed spectrophotometric method using the new reagent, 6-(anthracen-2-yl)- 2,3- dihydro-1,2,4- triazine-3-thion (ADTT), was simple, accurate, sensitive, and reproducible. It could be directly and easily applied for Hg (II) determination except in the case of interfering Cu (II) and Ni (II) ions. In this situation derivative spectrophotometry is recommended.

ACKNOWLEDGMENT

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