International Journal of Current Pharmaceutical Research

ISSN- 0975-7066 Vol 3, Issue 3, 2011

Research Article

SPECTROPHOTOMETRIC DETERMINATION OF COBALT IN WATER AND BIOLOGICAL SAMPLES USING 2-ACETYL-5-CHLORO THIOPHENE, THIOACETAMIDE

MD. FAKRUDDIN ALI AHMED[A] , Y. LINGAPPA[B].

[a] Professor, Department of Chemistry, FITAM, Kadapa-516001, Andhra Pradesh, [b] Professor, Department of Chemistry, Sri Venkateswara University, Tirupati – 517 502, Andhra Pradesh. Email: mfaahamed@gmail.com

Received: 29 April 2011, Revised and Accepted: 17 May 2011

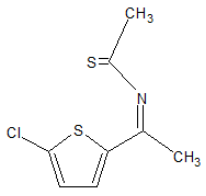
ABSTRACT

Analytical application of 2-acetyl-5-chlorothiophene, Thioacetamide (ACTTA) is described for the direct non-extractive spectrophotometric determination of Selenium. The synthesized and characterized using IR and NMR spectral data .The reagents react with Selenium, in acidic medium (pH 4.0., sodium acetate-acetic acid buffer) to form orange colored1:2 (M: L) complexes. The colour reactions are instantaneous and absorbance values remain constant for over 24 h. The molar absorptivity and Sandell's sensitivity of ACTTA methods are found to be 1.8 X 10^4 L mol^{-1} cm^4 , 0.0032 µg cm^{-2} , of Co^{II} respectively. The systems obey Beer's law in the range of 0.25-2.5 µg/ml of Co^{I}. Since ACTTA method is more sensitive it was applied for the determination of Cobalt in water and soil,and biological samples.

Key words: Spectrophotometry, Cobalt, 2-Acetyl-5-chlorothiophene, Thioacetamide , Water and biological samples.

INTRODUCTION

Cobalt traces are technically important, which are used mainly as binder in the hard metal industry and as constituents of many alloys[1] . Cobalt toxicity causes different diseases including asthma, contact dermatitis, lung cancer and bronchitis[2,3,4,5] . Cobalt(II) ions are also genotoxic and carcinogenic[3,5]. Genotoxicity follows tow mechanisms: (I) DNA breakage by cobalt metals especially hard metal particles and (II) inhibition of DNA repair by cobalt(II) ions[5] . Occupational exposure to hard metal dust, consisting of tungsten carbide (WC) and metallic cobalt particles (Co) is associated with increased lung cancer[6] . Although traces of cobalt is necessary for the synthesis of vitamin B-12, excessive administration of this trace element produces goiter and reduce thyroid activity[7]. A dietary intake of about 50 µg cobalt, of which 40 µg in the form of vitamin B-12, maintains cobalt equilibrium in the human[8] . Some death in man resulted from consumption of large amounts of beer containing 1.2-1.5 mg l-1 of cobalt that was added to the beer to promote optimal foam stabilization[9] .The normal level in human urine and blood are about 1.0 and 0.18 µg l-1, respectively[10]. Therefore, the accurate determination of cobalt at trace and ultra-trace levels using simple and rapid Spectrophotometry is essentially a trace analysis techniqueand is one of the most powerful tools in chemical analysis.The reagent 2-Acetyl-5-chlorothiophene Thioacetamide (ACTTA) has never been used as a spectrophotometric reagent for the determination of cobalt. This paper reports in its use in a very sensitive and highly specific new spectrophotometric method for the trace determination of cobalt.



ACTTA

The method is based on the reaction of non-absorbent ACTTA in a slightly acidic solution, pH 3.4-4.0 with cobalt(II) to produce a highly absorbent orange chelate product followed by the direct measurement of absorbance in aqueous solutions. With and the reagent blank solutions do not show a suitable masking, the reaction can be made highly selective ny appreciable absorbance. The method possesses distinct advantages over existing methods . [11-25]

RESULTS AND DISCUSSION

The reagents ACTTA may be easily prepared. The reagent solutions (0.001M) are found to be stable for 24 h. The absorption band from to 265 nm indicates that in solution on increasing the pH, The colour reactions of some important metal ions with ACTTA are summarized in Table1. In basic medium (above pH 4.0) coordinates the trivalent metal ion as mono anion to give neutral complexes[13].

Cobalt(II), reacts with ACTTA in acidic to give water soluble complexes. The colour reactions are instantaneous at room temperature. The change in the order of addition of metal ion, reagent (ACTPDA), and buffer has no effect on the absorbance of complexes. Analytical characteristics of the complexes are summarized in Table 1. The stiochiometry of the complexes (M:L = 1:2) was determined by job's continuous variation and molar ratio methods. Sodium acetate (0.2M)-acetic acid (0.2M) buffer solution (pH 4.0, T=300 K) and equimolar (1.8 X 10^4) solutions of Co(II) AC TTA were used in the calculation of stability constants of the complexes.

The effect of various cations and anions in 1.66 µgmL $^{-1}$ of Co(II). which are generally associated with the metal ion in the determination was studied Cobalt(II) by measuring the absorbance of COBALT the a complexes containing 1.66 µgmL $^{-1}$ of selenium (IV) in solution. The colour reaction is developed as described in the standard procedure. An error of ±2% in the absorbance reading was considered tolerable. The tolerance limit (µgmL^{-1}) values in µgmL $^{-1}$ for for various anions and cations in ATT methods respectively are as follows: citrate (1152,1152); tarrate (888,888); ascorbate (752,752); iodate (761,761); iodide(612,761), thiocyanate (507,507); phosphate(465,465); urea (384,384); bromide (317,317); sulphate (252,384); thiosulphate (246,246), nitrate (212,244); oxalate (281,281); fluoride (75,75); Ba^{2+} (675,800); Mn^{2+} (275, 325); Mg^{2+} (125,150); Sr^{2+} (100,125); W^{6+} (110,110); Sn^{2+} (47,47); Pb^{2+} (41,47); Cd^{2+} (23,26); Mo^{6+} (19,23); Tl^{3+} (14,14); Fe^{2+} (12,10); Cr^{6+} (10,12); Zn and Pd^{2+} (10,10), Pt^{4+} (8,8); Fe^{3+}(5,4); Au (4,4); Ag^{+} (2,5); Ni^{2+} (1,1.2); Cu^{2+}(1,1) . Higher amounts of Fe^{3+} (13,17) do not interfere in the presence of 70ppm of fluoride. Larger amounts of Hg^{2+} (40, 48) do not interfere in the presence of 600ppm of iodide.

Table 1: Physico-chemical and analytical properties of Se^{IV} complexe with ATCPDA

S.No.	Characteristics	Co-ACTTA
1.	λmax (nm)	470
2.	pH range (optimum)	4.0
3.	Mole of reagent required per mole	10-fold
4.	of metal ion for full colour development	
	Time stability of the complex (in hours	24
	Beer's law validity range (µg/ml)	0.25-2.48
5.	Molar absorptivity (L $mol^{-1}cm^{-1}$)	1.8 X 10^{4}
7.	Specific absorptivity (ml g^{-1} cm^{-1})	0.305
8.	Sandell's sensitivity (µg of Se^{IV} mL^{-1})	0.0033
9.	Composition of the complex as obtained	
	in Job's and molar ration methods (M:L)	1 : 2
	Stability constant of the complex	2.9 X 10^{11}
11.	Standard deviation	0.0097
12.	Relative standard deviation (RSD)	0.6

Table 2: Concentration of Cobalt in Blood and Urine Samples

Sample	Co(II) µg/ml				
	ACTTA (n=5)		Proposed method (n=5)		Sample source
	Found	RSD (%)	Found	RSD (%)	Normal
Blood	17.8	1.2	18.3	1.3	adult(male)
Urine	6.7	0.8	7.2	1.0	
Blood	45.6	1.5	44.5	1.7	Anaemia patient (male)
Urine	16.7	1.4	16.5	1.5	
Blood	550.7	1.6	558.3	1.1	Paralysis patient(male)
Urine	137.7	1.8	140.6	1.6	
Blood	152.6	1.6	158.4	1.3	Pulmonary patient(male)
Urine	46	1.5	48.2	1.2	

[a]Samples were collected from **FIMS** Hospital,Kadapa,(A.P) India.

Table 3: Determination of Cobalt in some Environmental Water Samples

Sample	Cobalt (µg/g)[a]		Recovery
	Added	Found	±S.D.%
Tap water[a]	0	4.0	
	100	104.02	98.9 ± 0.25
	500	505.10	100.1 ± 0.5
Well water[b]	0	11.2	
	100	112.0	100.4 ± 0.4
	500	515.0	100.5±0.19
River[c] Water	0	9. 50	
	100	110.8	100.5 ± 1.1
	500	512.0	100.4 ± 0.7
Bay of Bengal (Chennai) [d]	0	6.5	
	100	105 .03	99.0 ± 0.5
	500	509.0	100.3 ± 0.4

[a]Average of five determinations. [a] Mean ± Relative Standard Deviation (*n* = 5)

[b,c]. Palamanaru ground water-Chittour,A.P.india., Bay of Bengal (Chennai) [d] ,Tamilanadu

The present method (ACTTA) was applied for the determination of selenium when present alone and present in water and biological sample (Table 2), (Table 3). The present ligands containing heterocyclic ring are found to be potential and cost effective for the determination of Cobalt(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 spectro-photometric determination of Cobalt(II) in aqueous medium.

Experimental

Preparation of ACTTA

The reaction mixture containing 2-acetyl-5-chlorothiophene, (2g, 0.01245 mol in 20ml of methanol) phenylenediamine (0.9354g, 0.01245 mol in 20ml of methanol dissolved in hot condition) was taken in 250-ml round bottom flask and refluxed for 8h. On cooling the reaction mixture, dark yellow coloured product was formed. It was collected by filtration and washed with hot water and 50 percent cold methanol. This compound was recrystallised from ethanol and dried in vacuo, yield 0.64. g ; m .p . 86 °C.

Characterisation of ACTTA:

The reagents have been characterized by IR and ^{1}H NMR spectral data. Infrared spectrum of ACTTA shows bands at [3296(s) 3091(m),3083(m),2942(m)2677(S);1650(S); 1591(s),1435(s); 1303(s); 1123(S) ,974(S), 709(δ); cm^{-1} respectively corresponding to υNH-symmetric, υNH-asymmetric, υ (C-H) aromatic stretch, υ (C=S) stretching, υC=N symmetric, υ (C-C) aromatic ring, δ(C-H) of Thiophene ring, (ACTTA) and δ (C-H)-oop bend (aromatic) and δ (C-C)-oop bend aromatic ring vibrations. ^{1}H NMR spectra of TDATSC ($CDCl_3$ + DMSO-d6) showed signals at 2.54(3H,S)due to -CH_3, 3.4,(2H,s), 4.8 (2H),due to aromatic ring, , C_4H_2S (Thiophene).

pK_a values of reagents

The pK_a values were determined by recording the UV-Visible spectra of 4 X 10^{-5} 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 ACTTA were 9.0 (pK_1)

The reagent (ACTTA) solution (0.01 M) was prepared by dissolving 50 mg of the compound in dimethylformamide (DMF) in 25-ml standard flask. The reagent solution is stable for at least 24 h. Hydrochloric acid (1 M)-sodium acetate (1 M) (pH 0.5-3.5); 0.2 M NaOAc-0.2 M AcOH (pH 4-6) solutions were used .

A stock solution (1 mg L^{-1}) was prepared by dissolving 493 mg of $Co(No_3)_2.6H_2O$. (E.Merck preanalysis) in 100 ml de-ionized wate. Hydrolosis of cobalt was prevented by adding 2ml of 2M HNO_3. Dilute standard solutions were prepared from this stock solutions as and when required.

Recommended procedure

An aliquot of the solutions containing 0.023-0.070 -µgmL $^{-1}$ of C) ,10 ml of cobalt (II)NaOAc-AcOH buffer solution (pH 6.0) and 1.0 ml of 0.01 M ACTTA were mixed in a 25-ml volumetric flask and resulting solution was diluted to the mark with distilled water. The absorbance of this solution was measured at 295 nm against respective reagent blank. The measured absorbance is used to compute the amount of selenium present in the samples using predetermined calibration plot. Schimadzu 160A UV-Visible spectrophotometer equipped with 10. cm quartz cell and an ELICO model LI-610pH meter were used in the present study.

Determination of cobalt in biological samples

Human blood (2-4 ml) or urine (5-7 ml) sample was taken into a 100-ml micro-Kjeldahl flask. A glass bead and 10 ml of

concentrated nitric acid were added, and the flask was placed on a digester under gentle heating. When the initial brisk reaction was completed, the solution was removed and cooled,and digested following a recommended method[27] . A 1 ml of volume of concentrated sulfuric acid was carefully added followed by the addition of 1 ml of 70% perchloric acid, and heating was continued to dense white fumes, while repeating nitric acid addition if necessary. Heating was continued for at least half an hour and then cooling was applied. The content of the flask was filtered and neutralized with NH4OH in the presence of 1-2 ml of a 0.01% (w/v) EDTA solution. The resultant solution was then filtered and transferred quantitatively into a 10-ml calibrated flask and made up to the mark with deionized water. A suitable aliquot (1-2 ml) of the final solution was pipetted out into a 10-ml calibrated flask and the cobalt content was determined as described under procedure using EDTA as masking agent

Determination of Cobalt in environmental water samples

Each filtered samples (1000 ml) was evaporated nearly to dryness with a mixture of 5 ml of concentrated H_2SO_4 and 10 ml of concentrated HNO_3 in a fume cupboard, following a method recommended by Greenberg and then cooled down to room greenberg[26] . The residue was then heated with 10 ml of deionized water in order to dissolve the salts. The solution was then cooled and neutralized with dilute NH4OH in the presence of a 1-2 ml of 0.1% (w/v) EDTA solution. The resulting solution was then filtered and quantitatively transferred into a 25-ml calibrated flask and made up to the mark with deionized water. An aliquot (1-2 ml) of this preconcentrated water sample was pipetted into a 10-ml calibrated flask and then cobalt content was determined as described under the procedure using EDTA as a masking agent.

REFERENCE

1. A.A. Jensen, F. Tuchsen, Crit. Rev. Toxicol. 20 (1990)427.
2. M. De Boeck, S. Lardau, J.P. Buchet, M.K. Volders, D.Lison, Environ. Mol. Mutagen. 36 (2000) 151.
3. M. De Boeck, K. Volders, D. Lison, Mutal. Res. 548 (2004) 127.
4. V. Verougstraete, A. Mallants, J.P. Buchet, B. Swennen, D. Lison, Am. J. Res. and Crit. Care Med. 170 (2004) 162.
5. D. Lison, M. De Boeck, V. Verougstraete, M.K. Volders, Occup. Environ. Med. 58 (2001) 619.
6. M. De Boeck, N. Lombaert, S. De Backer, R. Finsy, D. Lison, M.K. Volders, Mutagenesis 18 (2003) 177.
7. D.G. Barceloux, J. Toxicol. Clin. Toxicol. 37 (1999) 201.
8. B. Venugopal, T.D. Luckiy, Metal Toxicity in Mamals- 2, Plenum Press, New York, 1999, p. 284.
9. Remington, Remington's Pharmaceutical Sciences, 17th ed., Mack Publishing Company, Easton, Pannsylvania- 18042, p. 1032.
10. G.A. Goyer, in: C.D.K. Laassen (Ed.), Casarett and Doull's Toxicology, Mc Graw-Hill, New York, 2001, p.
11. H. Eskandari, G.H. Karkaragh, A.G. Saghseloo, G.B. Dehaghi, Anal. Sci. 20 (2004) 227.
12. A. Afkhami, M. Bahram, Spectrochim. Acta A Biomol. Spectrosc. 60 (2004) 181.
13. M.C. da Silva, L.H. da Silva, F.J. Paggioli, Anal. Sci. 21 (2005) 933.
14. C. Sivani, G.R. Naidu, J. Narasimhulu, D. Rekha, J.D. Kumar, P. Chiranjeevi, J. Hazard. Mater. 146 (2007) 137.
15. A.K. Malik, K.N. Kaul, B.S. Lark, W. Faubel, A.L.J. Rao, Turk. J. Chem. 25 (2001) 105.
16. A.K. Malik, V. Kaur, N. Verma, J. Chin. Chem. Soc. 54 (2007) 715.
17. H.S. Ghaziaskar, H. Eskandari, A.A. Ensafi, Anal. Sci. 17 (2001) 327.
18. B.R. Reddy, P. Radhika, J.R. kumar, D.N. Priya, K. Rajgopal, Anal. Sci. 20 (2004) 345.
19. A.S. Amin, I.S. Ahmed, M.E. Moustafa, Anal. Lett. 34 (2001) 749.
20. L.S. Sarma, J.R. Kumar, C.J. Kumar, A.V. Reddy, Anal. Lett. 36 (2003) 605.
21. A. Safavi, H. Abdollahi, Anal. Lett. 34 (2001) 2817.
22. Y. Guangyu, D. Xuechang, H. Qiufen, Y. Jiayuan, Anal. Lett. (2002) 1735.
23. H. Eskandari, G.H Karkaragh, Anal. Sci. 19 (2003) 1549.
24. A. Afkhami, M. Bahram, A.R. Zarei, Microch. Acta 148 (2004) 317.
25. M.J. Ahmed, M.N. Uddin, Chemosphere 67 (2007) 2020.
26. M.L. Jackson, Soil Chemical Analysis, Prentice-Hall, Englewood Cliffs NJ, 1965, p. 326.
27. P. Job, Ann. Chim. 9 (1928) 113.