



Trade Science Inc.

Environmental Science

An Indian Journal

Current Research Paper

ESAIJ, 2(1), 2007 [69-74]

Selenium Studies In Various Environmental Samples By Spectrophotometry



Corresponding Author

P.Chiranjeevi
Environmental Monitoring Laboratory,
Department of Chemistry, S.V. University,
Tirupati-517 502, A.P., (INDIA)
Tel.: +91 877 2250556; Fax: +91 877 2261274.
E-mail: sai_chiranjeevi@yahoo.co.in



Co-Authors

K.Kiran^a, K.Janardhanam^a, J.Dilip Kumar^b, P.Reddy Prasad^b
^aDepartment of Environmental Science, S.V. University,
Tirupati-517502 (INDIA)
^bEnvironmental Monitoring Laboratory, Department of Chemistry,
S.V. University, Tirupati-517 502, A.P., (INDIA)

Received: 18th December, 2006
Accepted: 23rd December, 2006

Web Publication Date: 27th December, 2006

ABSTRACT

A novel, rapid, highly sensitive and selective spectrophotometric method was developed for the determination of traces of selenium (IV). The method is based on direct oxidation of leuco methylene green (LMG) with selenium (IV) in acetate buffer at pH 4.0±0.2 to give blue colored derivative with λ_{\max} 624 nm and the colored product is stable for more than 7 days at 35°C. Beer's law is obeyed for selenium (IV) in the concentration range of 0.25-2.5 $\mu\text{g ml}^{-1}$ at the wavelength of maximum absorption. The optimum reaction conditions and other analytical parameters were investigated to enhance the sensitivity of the present method. The detailed study of various interferences made the method more selective. The proposed method was successfully applied to the analysis of selenium in various environmental samples. The results obtained were good agreement with the reported methods at the 95 % confidence level. The performance of proposed method was evaluated in terms of student's t-test and variance ratio f-test which indicates the significance of proposed method over reported method.

INDIA

© 2007 Trade Science Inc.

KEYWORDS

Selenium (IV);
Oxidation;
Leuco methylene
green (LMG);
Spectrophotometry;
Environmental samples.

INTRODUCTION

Selenium is an essential and most important interesting element from the point of view of its clinical

and environmental effects. The valence state of selenium is important information in the determination of selenium. Selenium(IV) paradoxical element which is reported as a double character well known

Current Research Paper

as essential as well as toxic trace element and it is widely distributed in nature, but relatively small concentrations in rocks, plants, coal, and other fossil fuels. Selenium enters natural water through seepage from seleniferous soil, chemical weathering and industrial wastes. Trace levels ingestion of selenium is not only toxic to animals but also toxic to human beings. Selenium is an essential trace nutrient to living system. Diseases due to selenium deficiency are well known in veterinary medicine. The selenium concentration of most drinking waters and natural waters is less than $10 \mu\text{g ml}^{-1}$ [1]. Several analytical techniques have been reported for the determination of selenium, which includes electro thermal atomic absorption spectrometry[2], graphite furnace atomic absorption spectrometry[3], Hydride generation inductively coupled plasma mass spectrometry[4], derivative hydrate generation atomic absorption spectrometry[5], flow-injection technique[6] and differential pulse cathode-stripping voltametry were reviewed[7]. Few reagents were available for the spectrophotometric determination of selenium in various environmental matrices such as leuco crystal violet[8], J-acid[9], 1-aminonaphthalene-7-sulfonic acid[10], sodium salt of hexamethyleneiminecarbodithioate[11], variamine blue[12] resazurin[13], DPH/4-AAP[14]. The above reported methods suffer from poor sensitivity, selectivity and stability of the color derivative. So, it is desirable to develop simple, highly sensitive, reliable and rapid method for the determination of selenium (IV) in environmental samples.

In the present study, we reported simple, sensitive, selective and rapid method for the determination of traces of selenium (IV). The implied reaction was oxidation of leuco methylene green in the presence of Se (IV) in presence of acetate buffer at pH 4.0 ± 0.2 . The method was successfully employed for the determination of selenium in various environmental samples. This type of oxidation of reaction for the determination of selenium has not been reported for our literature review.

EXPERIMENTAL

Apparatus

A Hitachi U 2001 spectrophotometer with 1.0 cm matched quartz cell was used for all absorption

measurements. A pH meter Elico Li-129 Model glass calomel combined electrode was employed for measuring pH values.

Reagents

All chemicals used were of analytical reagent grade and deionised - double distilled water was used to prepare all solutions through out experiments. A stock solution containing 100 mg l^{-1} of selenium was prepared by dissolving 0.14 g of selenium dioxide (Merck, Mumbai, India) in 1 l of deionised - double distilled water. A working solution of selenium (IV) was prepared by further dilution. 1% Leuco methylene green (from S.D. Fine Chemicals, India) was prepared by dissolving 1.0 g leuco methylene green in water and diluted up to the mark in a 100 ml standard flask with deionised - double distilled water and the solution was refrigerated.

General procedure for determination of selenium (IV)

Stock solution containing 1 - $100 \mu\text{g}$ ($0.03 - 3.5 \mu\text{g ml}^{-1}$) of selenium (IV) (the volume of the test solution was restricted to 1 ml) were transferred into 25 ml calibrated flasks, 3 ml leuco methylene green. The mixture was allowed to stand for 2 min to complete the reaction. The content was diluted to the mark with deionised - double distilled water and absorbance was measured at 624 nm against the corresponding reagent blank and the calibration graph was constructed.

Determination of selenium (IV) in spiked water samples

The spiked water samples were prepared with 500 ml of doubly-deionised distilled water by adding known amounts of Se(IV), preconcentrate as mentioned in literature and determined the percentage recovery of Se (IV) as explained in aforesaid procedure.

Determination of selenium (IV) in natural water samples

The proposed method was employed for different natural water samples (200 ml each) collected around Tirupati area. The samples were used directly to measure the Se (IV) contents by the proposed method after filtered with cellulose membrane of

pore size 0.45 μm and determined by above general procedure.

Determination of selenium (IV) in polluted water samples

The distillation procedure for the determination of selenium (IV) in waste water as follows: To an aliquot (100 ml) of known sample taken in a distillation flask, 1 g of KBr and 10 ml of concentrated H_2SO_4 treated with 0.5 ml of saturated bromine water were added and the solution was then distilled under vacuum till copious white fumes of SO_3 vapours were evolved, and this process converts all other forms of selenium in to selenium (IV). The distillate was collected in 10 ml of 5% hydroxyl ammonium chloride solution, then made up to a fixed volume and analyzed by the general procedure.

Procedure for the determination of selenium in soil

A known weight of soil sludge sample was placed in a 50 ml beaker and extracted in 4 times with a 5 ml portion of conc. HCl. The extract was boiled for 10 min to convey any (VI) present in the soil to Se (IV), cooled and neutralized (pH~0-7) with 10% NaOH. A volume of 5 ml of 5% EDTA was added and the content were made up to 25 ml with water. For selenium determination, an aliquot (4ml) of made up to solution was directly used for color development by following the general procedure.

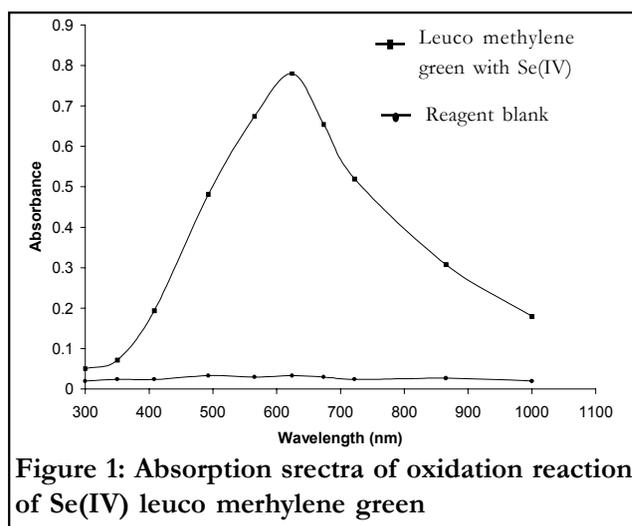
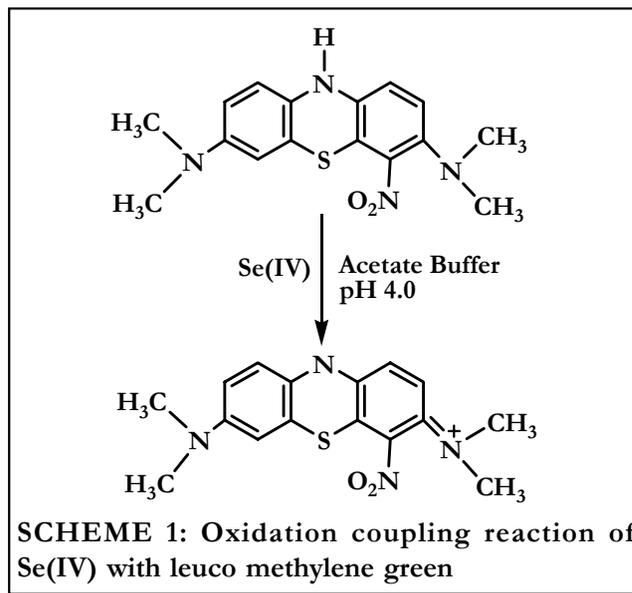
Determination of selenium (IV) in plant materials

Finely chopped fresh vegetable samples (5 g each) were placed in a 250 ml beaker, and a solution of concentrated $\text{H}_2\text{SO}_4 / \text{HNO}_3$ 1:1 (v/v) (10 ml) was added. This mixture was heated until the solution is clear. The solution was filtered off and concentrated to 5 ml then cooled and diluted to 50 ml with deionised double-distilled water as recommended in reported methods^[15,16]. Then the general procedure was applied to the 1 ml of the solution.

RESULTS AND DISCUSSION

Absorption spectra of oxidation reaction

The proposed method involves the formation of the blue colored derivative in presence of Se (IV) as



shown in SCHEME 1 with λ_{max} 624 nm against the corresponding reagent blank and the calibration graph was constructed as shown in figure 1.

Optimum conditions for color product formation

In order to establish the optimum conditions necessary for a rapid and quantitative formation of the colored product with maximum stability and sensitivity, the investigators measured the absorbance of a series of solutions by varying one and fixing the other parameters at 624 nm.

It was found that a 1 % solution of leuco methylene green in the range of 2.0-4.0 ml, was necessary to achieve the maximum color intensity of the oxidative coupling product figure 1. The color intensity decreased below the lower limit and above the upper limit, and a colored product was unstable.

Current Research Paper

Therefore, 3.5 ml of leuco methylene green was recommended for all measurements.

Effect of temperature on colored product

The reaction between leuco methylene green with Se (IV) was found to be instantaneous. The effect of the temperature on the product was studied at different temperatures; it was found that the colored product was stable for more than 15 days in the temperature range of 25-40°C. So, the temperature of 35°C was selected as optimum for maximum color development.

Effect of pH on absorption maximum

The effect of pH on the peak height of selenium (IV) at different concentrations was investigated with a fixed reagent concentration (1.0 % Leuco methylene green) in the pH range of 3.0-8.0 and the peak height was measured for each concentration level of selenium (IV). At all concentration levels of Se (IV), maximum peak heights were found between pH 3.0-6.0. Therefore, a pH 4.0 was selected for further studies.

Ringbom plot for red colored derivative

Ringbom's plot is the established standard adopted to know the optimum range of concentration for a system that obeys Beer's law. The plot is drawn between log C of Selenium (IV) and (1-T) (where T is the transmittance). The plot has a sigmoid shape with a linear segment at intermediate absorbance values (0.25-0.65) and concentration values (0.35-4.5 µg ml⁻¹). The slope of Ringbom's plot figure 2 is 0.131. Hence, the ratio between the relative error in concentration and photometric error is 0.17, for a concentration of 1.0, for 99.30 to 100.50 % photometric error.

Optical parameters

Beer's law range, molar absorptivity, Sandell's sensitivity, and other parameters of the oxidative coupling mixture were given in (TABLE 1). The precision and accuracy of the method was studied by analyzing the coupling solution containing known amounts of the cited reagents within Beer's law limit. The low values of the standard deviation (%) and the percentages of error indicated the high accuracy

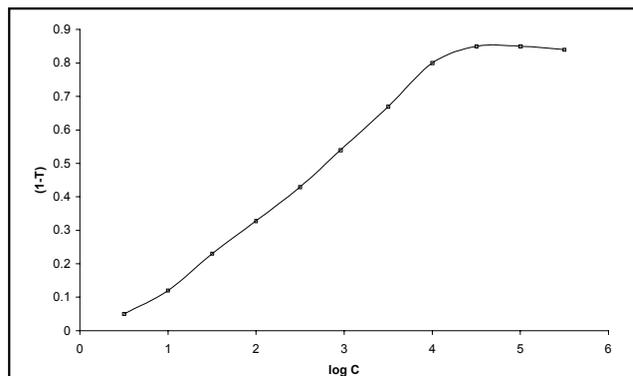


Figure 2: Ringbom plot of oxidation of Se(IV) with Leuco methylene green

TABLE 1: Optical characteristics of oxidative coupling mixture and precision data

Optical characteristics of present method	
Color	blue
λ_{\max} [nm]	624
Stability	More than 7 days
Beer's law range [$\mu\text{g ml}^{-1}$]	0.25 - 2.5
Molar absorptivity [$\text{l mol}^{-1} \text{cm}^{-1}$]	3.125×10^4
Sandell's sensitivity [$\mu\text{g cm}^{-2}$]	0.00380
Regression equation (Y) ^b	
Slope ^a	0.392
Intercept ^b	0.00061
Correlation coefficient ^c	1.129
Relative standard deviation [%] ^c	0.809
Range of error (95 % confidence level)	± 0.496
Detection limit [$\mu\text{g mL}^{-1}$]	0.0195
% error (at 95% confidence level) ^c	0.6092

^a Experiments performed under optimized conditions (see text) with 1 ml of 0.014% aqueous SeO₂ solution.

^b Y=ax+b, where x is the concentration of selenium in $\mu\text{g ml}^{-1}$, ^cn=4.

TABLE 2: Effect of non-target ions on the determination of 3.0 $\mu\text{g ml}^{-1}$ selenium (IV)

Species	Tolerance limit [$\mu\text{g ml}^{-1}$]
EDTA	10600
Na ⁺ , Mg ²⁺ , Cl ⁻ , NO ₃ ⁻ , F ⁻ , CHCOO ⁻ , CO ₃ ²⁻ , K ⁺	2090
Ba ²⁺ , SO ₄ ²⁻ , CN ⁻ , SCN ⁻ , Tartarate	1100
PO ₄ ³⁻ , Al ³⁺ , Cd ²⁺ , NO ₂ ^{-a}]	800
Cu ^{2+b}), Ni ²⁺ , Co ²⁺ , Ca ²⁺	84
Zn ²⁺ , Pb ²⁺ , SO ₃ ²⁻ , NO ₃ ⁻ , Cr ³⁺ , As ⁵⁺	50
Fe ²⁺ , S ²⁻	36

^aCan be masked up to 810 $\mu\text{g ml}^{-1}$ by the addition of 3 ml of 2 % sulfamic acid. ^bCan be masked up to 86 $\mu\text{g ml}^{-1}$ by the addition of 3 ml of 5% EDTA.

Current Research Paper

of the present investigation.

Reaction mechanism

Under the reaction condition, one mole of leuco methylene green solution reacts with one mole of Se (IV) in 2ml of acetate buffer at pH 4.0 ± 0.2 to loses $2 e^-$ and 2 protons develops the chromatic methylene green due to oxidation of Se(IV) into Se(II). This reaction mechanism was shown in SCHEME 1.

Interference of non-target ions

The effect of various species on the determination of Se (IV) was investigated. The tolerance limit was taken as the amount that caused $\pm 2\%$ absorbance error in determination of $3.0 \mu\text{g ml}^{-1}$ of Se (IV). The results are shown in (TABLE 2).

Applications and comparison of proposed method with reported method

TABLE 3: Determination of selenium in various environmental samples

Sample ^a /ml	Added selenium $\mu\text{g/ml}$	Proposed method		Reference method ^[12]		F-test, t-test
		Selenium found / μg^b	Recovery (%) ^a	Selenium found / μg^b	Recovery (%) ^a	
Spiked water samples	2.00	1.98	99.00 \pm 0.12	-	-	-
	4.00	3.97	99.25 \pm 0.28	-	-	-
	6.00	5.99	99.83 \pm 0.15	-	-	-
Natural water ^b	3.00	2.97	99.00 \pm 0.16	3.00	99.7 \pm 0.11	F=0.26,t=0.13
	6.00	5.99	99.83 \pm 0.23	5.99	100.2 \pm 0.12	F=0.08,t=0.06
	9.00	8.99	99.88 \pm 0.11	9.00	99.8 \pm 0.09	F=0.61,t=0.35
Polluted water ^c	-----	3.25	-	3.26	-	-
	3.00	6.24	99.89 \pm 0.25	6.26	99.8 \pm 0.09	F=0.08,t=0.04
Soil sludge ^d (1g)	-----	3.75	-	3.73	-	-
	2.25	5.98	99.81 \pm 0.11	5.99	99.7 \pm 0.10	F=0.87,t=0.45
Plant materials, ^e						
1. Cabbage (5g)	-----	3.78	-	0.38	-	-
	2.50	6.26	99.95 \pm 0.17	6.28	99.7 \pm 0.13	F=0.42,t=0.25
2. Cruciferac plant(5g)	-----	4.26	-	4.29	-	-
	2.80	7.03	99.89 \pm 0.11	7.05	99.6 \pm 0.18	F=0.19,t=0.08

^aMean \pm standard deviation(n=4), ^bCollected from Swarnamuki River belt near Sri kalahasthi, ^{c,d}Collected from Renigunta Industrial area, ^e Collected from local market, ^f* Collected from local shop.

TABLE 4: Comparison of spectrophotometric method with present method for the determination of selenium in various environmental

Reagents	Sensitivity ($\mu\text{g mL}^{-1}$)	Remarks	Reference
MBTH/Dopamine	4.0	Less sensitive, stable	[1]
Leuco crystal violet	-	Less stable, selective and high solvent consumption	[8]
J-acid	5.80	Highly commercial, high reagent consumption and 10 min is needed for color development	[9]
1-naphthylamine-7-sulphonic acid	5.32	Less selective, more reagent consumption and metal ions are interfered seriously	[10]
Sodium salt of hexamethylene imine carbodithioate	5.50	Needs extraction, less stable, high solvent consumption	[11]
Variamine blue	20	Less sensitive, low detection limit, maintenance of pH conditions	[12]
Resazurin	-	It require some catalytic reactions, high solvent consumption	[13]
DPH/4-AAP	3.5	Less sensitive, stable	[14]
Congored	5.5	More sensitive, rapid, facile, non extractive, economical reagent, highly stable(more than 15 days), metal ions do not interfere.	This work

Current Research Paper

The proposed method for the analysis of selenium (IV) has been employed for various environmental samples were presented in (TABLE 3). The obtained results were compared with reported method in terms of Student's t-test and Variance ratio f-test. The analytical data summarized in (TABLE 3) suggest that the percentage of selenium recovery from environmental samples ranges from 99.50 to 100.50 %, which is more reliable and sensitive than the other methods.

It is evident from the above data that the proposed method is simple, highly sensitive and rapid than the reported method in literature as shown in (TABLE 4).

CONCLUSION

The proposed method is simple, highly sensitive and rapid for the spectrophotometric determination of selenium (IV) in various environmental samples. The limit of detection of the proposed method is superior and compared with previously reported methods. The method has additional advantages over reported methods owing to it's: oxidative reagent employed in the present method, i.e., leuco methylene green is fairly soluble in water and very cheap. The color derivatives (SCHEME 1) formed by oxidation process by Se(IV), is highly stable (more than 7 days) when compared to that of the reported method^[12] and shown in (TABLE 4). Less interference, more accuracy, avoidance of lengthy extraction steps and statistical analysis which made present method to be placed among the most sensitive ones for the determination of Se(IV) in various environmental samples. The proposed method can be employed as additional method for the continuous monitoring of Selenium in various environmental samples.

ACKNOWLEDGEMENT

The authors are grateful to Head, Department of Biotechnology, S.V. University, Tirupati, for providing instrumental facility.

REFERENCES

- [1] M.S.Hemant Kumar, P.Nagaraja, H.S.Yathirajan, J.S.Prakash; *Fresenius Environ.Bull.*, **11**, 396 (2002).
- [2] R.M.Paulo, Correia, Elisabeth de Oliveira Pedro V.Oliveira; *Talanta*, **57**, 527 (2002).
- [3] H.C.Wang, H.W.Peng, M.S.Kuo; *Anal.Sci.*, **17**, 527 (2001).
- [4] Amauri A.Menegario, Maria Fernanda Gine; *Spectrochimica Acta Part B*, **55**, 355 (2000).
- [5] Han Wan Sun, Jing Ha, De Qiang Zhang, Li Li Yang, Jian Min Sun; *Analytical Sciences*, **18**, 603 (2002).
- [6] J.Fan, H.X.Lu, C.L.Ye, F.L.Cui; *Guang Pu Xue Yu Guang Pu Fen Xi*, **25**, 263 (2005).
- [7] Tommaso ferri, Silva Rossi, Pool Sangiorgio; *Analytica Chimica acta*, **361**, 113 (1998).
- [8] D.Agarwal, G.Sunitha, V.K.Gupta; *J.Indian Chem.Soc.*, **73**, 151 (1998).
- [9] R.Manish, K.N.Ramachandran, V.K.Gupta; *Talanta*, **41**, 1623 (1994).
- [10] K.Pyrzynsaka; *Anal.Sci.*, **13**, 629 (1997).
- [11] M.N.Pathare, A.D.Sawant; *Anal.Lett.*, **28**, 317 (1995).
- [12] H.D.Revanasiddappa, T.N.Kiran Kumar; *Anal.Sci.*, **17**, 1309 (2001).
- [13] A.Safavi, A.Afkhami; *Anal.Lett.*, **28**, 1095 (1995).
- [14] K.Suresh, K.Suvaradhan, L.Krishnaiah, P.Chiranjeevi; *Helvetica Chimica Acta*, **88**, 343 (2005).
- [15] M.Tuzen, M.Soylak, L.Elci; *Anal.Chim.Acta*, **548**, 101 (2005).
- [16] M.Tuzen, K.Parlar, M.Soylak; (2005) *J.Hazard.Mater.*, **121**, 79 (2005).