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Chemical Amplification Method by Spectrophotometry for the Determination of Trace Amounts of Selenium

A. Ramesh, T. V. Ramakrishna, and M. S. Subramanian*
Department of Chemistry, Indian Institute of Technology, Madras 600036, India

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A rapid and sensitive spectrophotometric method has been developed based on the extraction of the ion-association complex formed between ICl_2^- and Rhodamine $6G^+$ into benzene. The ICl_2^- is formed through iodine which is liberated during the oxidation of iodide by the selenium(IV) present in sulfuric acid medium. The method is sensitive and down to 0.1 μg selenium could be determined in an aqueous phase volume of 10 ml. The extracted Rhodamine $6G^+$–ICl_2^- complex has a molar absorptivity of 4.8×10^5 $dm^3\,mol^{-1}\,cm^{-1}$ at 535 nm. Beer's law is obeyed over the range 0.1 to 0.9 μg of selenium. The relative standard deviation is 2.4%. The method is made more sensitive and selective by adopting the aluminium hydroxide collection procedure. The interference of few ions such as Fe^{3+}, VO_3^-, TeO_3^{2-} and Sb^{5+} which were collected along with selenium on aluminium hydroxide, were overcome by adding proper masking agents. The method was applied to the determination of selenium in environmental ash sample, copper ore, bark sample, and shampoo preparations.

There is an increasing recognition that selenium is an important metalloid with industrial, environmental, biological, and toxicological significance. The principle application of selenium and its compounds in the industry are found in glass, pigments, steel, and rectifiers.[1] Selenium is an essential element in many species including humans in which it is a component of glutathione-peroxidase which is required for the metabolism and removal of hydrogenperoxide and lipid peroxidases from cells. There is a narrow range of selenium in-take that is consistent with health, outside of this range deficiency, diseases and toxicity occur.[2] It has been reported that selenium acts as an antidote for mercury, cadmium, and arsenic.[3]

In view of this the development of methods that are sensitive for the determination of micro amounts of selenium is becoming increasingly important. Of the reagents that have been used for the direct spectrophotometric determination of selenium(IV), aromatic amines containing two adjacent amino groups yielded high sensitivity allowing the formation of a selenodiazole. The analysis may then be performed by measuring either the fluorescence or the UV absorbance of the selenodiazole thus produced. As most of the direct spectrophotometric methods are less sensitive, indirect methods have been used based on the oxidizing ability of selenium(IV) followed by the spectrophotometric determination of the oxidized counter part,[4] or the reducing ability of the selenide, initially formed from selenite by conventional reductants, followed by the spectrophotometric determination of the reduced counterpart.[5]

A new spectrophotometric method for the determination of trace amounts of selenium(IV) has been developed based on its liberation of iodine from excess iodide. The liberated iodine in the presence of chloride and iodate in acidic conditions forms ICl_2^- which gets extracted as ion-association complex with Rhodamine $6G^+$ into benzene. The complex is stable for nearly 60 min and shows a maximum absorption at 535 nm. The method is very simple, sensitive and down to 0.1 μg of selenium present in 10 ml of aqueous volume can be determined.

Experimental

Apparatus. A Carl-Zeiss PMQ II spectrophotometer with 1 cm quartz cell was used for all the absorbance measurements.

Reagents. All chemicals used were of analytical reagent grade and doubly distilled water was used through out the experiments. A standard solution of selenium (1000 $\mu g\,ml^{-1}$) was prepared by dissolving 0.14 g of selenium dioxide and diluting to 100 ml with distilled water and a suitable volume of this solution was diluted to obtain working standards.

The following solutions were prepared by dissolving appropriate amounts of the reagents in distilled water.

potassium iodide solution : 0.25%
potassium iodate solution : 0.01%
sodium chloride solution : 15.0 %
Rhodamine 6G solution : 0.02 %
sulfuric acid : 2.5 M (1 M=1 $mol\,dm^{-3}$)

Benzene: Thiophene free benzene was used for the extraction purposes.

General Procedure. Transfer an aliquot of the sample containing not more than 0.9 μg of selenium into a 125 ml separatory funnel, and add 1 ml of 0.25% of potassium iodide solution, 2 ml of 2.5 M sulfuric acid and dilute to 10 ml with distilled water. Extract the liberated iodine with 5 ml of benzene for 3 min and remove the aqueous layer carefully from the separatory funnel. Wash the organic layer three times with distilled water in order to remove any potassium iodide in the organic layer. To the washed organic layer add 2 ml of 0.01% of potassium iodate solution, 2 ml of 2.5 M sulfuric acid, 4 ml of 15% sodium chloride solution, 2 ml of 0.02% Rhodamine 6G solution and dilute the aqueous layer to 25 ml with distilled water. Shake the solution vigorously for 3 min and separate the organic layer into a dry test tube and add about 0.5 g of anhydrous sodium sulfate. Measure the absorbance of the pink colored benzene layer containing Rhodamine $6G^+$–ICl_2^- ion-association complex at 535 nm in 1 cm cell against the reagent blank performed under similar condition in the absence of selenium.

Calibration. 0.1 to 0.9 µg of selenium were transferred into a series of separatory funnels, and the estimation of selenium was carried out by following the above procedure. The calibration graph was linear over the concentration range studied for selenium. The molar absorptivity of the complex was established to be 4.8×10^5 dm^3 mol^{-1} cm^{-1}. The sandell sensitivity was found to be 1.6×10^{-4} µg cm^{-2}. The relative standard deviation at 0.6 µg concentration of selenium studied was found to be 2.4% (ten determinations).

Results and Discussion

Selenium(IV) in mild acidic medium oxidizes iodide to iodine and gets itself reduced to elemental selenium. The liberated iodine in acidic medium, in presence of iodate and chloride forms ICl_2^- anion, which forms an association complex with Rhodamine $6G^+$.⁶⁾ The ion associate is pink in colour and it can be extracted into benzene. The sequence of these reactions may be represented by the following steps.

$$SeO_3^{2-} + 4I^- + 6H^+ \rightleftharpoons Se(0) + 3H_2O + 2I_2 \quad (1)$$

$$2I_2 + IO_3^- + 6H^+ + 10Cl^- \rightleftharpoons 5ICl_2^- + 3H_2O \quad (2)$$

$$5ICl_2^- + 5Rh6G^+ \rightleftharpoons 5Rh6G^+ICl_2^- \quad \text{(ion associate)} \quad (3)$$

In the above reactions one mole of SeO_3^{2-} indirectly gives rise to five moles of Rhodamine $6G^+$ $-ICl_2^-$ ion associates. Therefore the determination of trace amounts of selenium can be made possible spectrophotometrically by measuring the absorbance of the resulting Rhodamine $6G^+$ $-ICl_2^-$ in benzene.

Effect of Variables. Since the first and second steps of the reactions in the procedure are acidity dependent, the variation of acidity was studied. It was found that maximum absorbance was obtained, when the overall acidity of the reaction medium was in the range 0.38—1.0 M with respect to the sulfuric acid in the first step. Likewise in the second step the overall acidity of reaction medium was found to be between 0.15—0.25 M.

Investigation of variation of potassium iodide indicated that 1 mL of 0.25% of potassium iodide solution was sufficient to provide the maximum absorbance. In the second step an optimum concentration of 2 mL of 0.01% of potassium iodate, 3 mL of 15% sodium chloride, and 1 mL of 0.02% Rhodamine 6G were necessary to get the maximum absorbance. Further increase in the concentration of the above three variables, had no effect on the absorbance value. However increase in the blank value was observed when the concentration of iodide and acid increases beyond the optimum range.

Effect of Foreign Ions. The effect of as many as 40 cations and anions at 1 mg levels on the estimation of 0.6 µg of selenium was investigated. The ions that interfered in this method can be classified according to their mode of interference like (i) those that reduce Se(IV) to Se(0) like Sn^{2+}, AsO_2^-, SO_3^{2-} (ii) those that oxidize iodide to iodine like NO_2^-, VO_3^-, Sb^{5+}, Cu^{2+}, Fe^{3+}, AsO_3^-, $Cr_2O_7^{2-}$, Ce^{4+} (iii) and those that form insoluble precipitate with iodide and get dispersed in the organic layer like Ag^+, Tl^+, Pb^{2+}, and Bi^{3+}.

As most ions excepting Al^{3+}, TeO_3^{2-}, Zn^{2+}, Cd^{2+}, Ba^{2+}, Mg^{2+}, Ca^{2+}, F^-, Cl^-, Br^-, MoO_4^{2-}, citrate, tartrate, and EDTA, interfered in this procedure it was decided to explore the possibility of selectively separating selenium from other interfering elements. A collection procedure was adopted for the efficient separation of selenium from other interfering ions.

A patented method⁷⁾ for the collection of selenium by adsorption on aluminium hydroxide has been reported. Since the details of such collection procedure were not available, studies were carried out for the quantitative collection of selenium on aluminium hydroxide. A slurry of aluminium hydroxide was prepared as reported in the literature.⁸⁾ A 3 mL of 5% slurry was taken in a centrifuge tube to which was added 0.6 µg of selenium, 2 mL of 0.1 M EDTA solution and the collection procedure was carried out using 2 mL sodium acetate–acetic acid buffer adjusted to different pH values from 2 to 8. The final volume was maintained at 10 mL. The solution was digested on a water bath for 2 min, cooled, centrifuged, and the centrifugate was discarded. The precipitate was dissolved in a minimum quantity of sulfuric acid and transferred into a separatory funnel and the recovery of selenium was determined using the developed procedure.

Figure 1 shows the effect of variation of pH on the collection of SeO_3^{2-} by aluminium hydroxide slurry. Figure 2 shows the variation of percentage recovery of selenium with varying aluminium hydroxide slurry volume. Using 3 mL of 5% slurry the effect of variation of sample volume was next investigated and the results are shown in the Fig. 3. The influence of other metal ions on the recovery of selenium using aluminium hydroxide slurry was next studied. Of the ions studied only Fe^{3+}, VO_3^-, TeO_3^{2-}, and Sb^{5+} were found to collect along with SeO_3^{2-}. The interference of Fe^{3+} and Sb^{5+} which were collected along with selenium were masked by adding 2 mL of a 1% solution of sodium fluoride in the estimation procedure before the addition of potassium iodide. Vanadate interference was overcome by reducing it with Fe^{2+} and masking Fe^{3+} formed, by fluoride. TeO_3^{2-} upto one milligram level concentration studied did not cause any significant interference.

Application. The developed procedure was applied to determine the selenium content in an analyzed sample of incineration ash taken from coalmine in Germany. The applicability of this method was also tested for the estimation of selenium content in bark and copper ore samples. The incineration ash sample was decomposed using hydrofluoric acid and sulfuric acid,⁹⁾ and the bark sample using sulfuric acid and per-

Table 1. Determination of Selenium in Various Materials.

Sample	wt/vol	Selenium found by ICPAES	Selenium added	Selenium found by proposed method [a)]		Recovery
		%	µg	µg	%	%
Incineration ash (CRM 176)	0.5 g/25ml	0.0041	—	20.9	0.004	—
			10	30.8		99.0
			15	35.5		97.3
Bark	1.0 g/25ml	0.0016	—	17.5	0.002	—
			10	27.5		100.0
			15	37.5		100.0
Copper ore	1.0 g/50ml	0.0062	—	63.5	0.006	—
			40	103		98.8
			60	123		99.2
Shampoo						
	0.25 g/100ml	0.97	—	2.41 mg	0.96	—
			0.5 mg	2.89 mg		96.0

a) Average of three determinations.

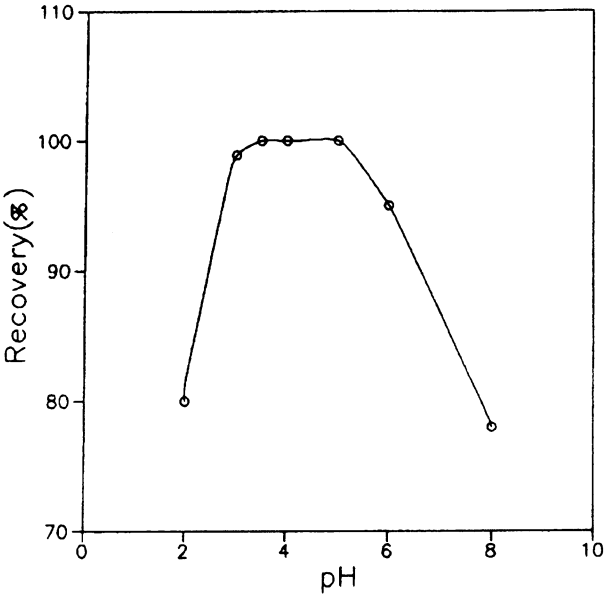

Fig. 1. Variation of pH.

chloric acid.[10)] Copper ore was decomposed with nitric acid and sulfuric acid and evaporated until dense fumes of sulfur trioxide were evolved.[11)] The shampoo sample dissolution was carried out by the procedure described by Belarra et al.[12)] The residue in each instance was dissolved in the minimum volume of dilute hydrochloric acid, filtered if necessary and diluted to known volume. Suitable aliquots were taken and analyzed for selenium as described above. Known amounts of selenium were then added to the sample before the decomposition and the above procedure was repeated. The results obtained were compared with those obtained by inductively coupled plasma atomic emission spectrometry (ICPAES). The results given in Table 1 show a close agreement between the proposed method and the ICPAES method.

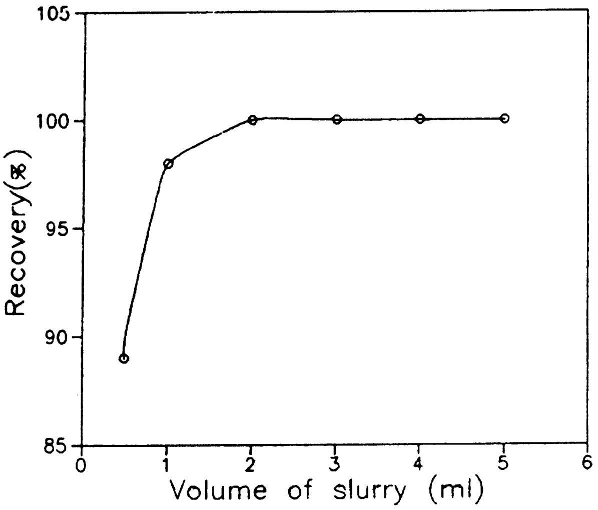

Fig. 2. Variation of slurry volume.

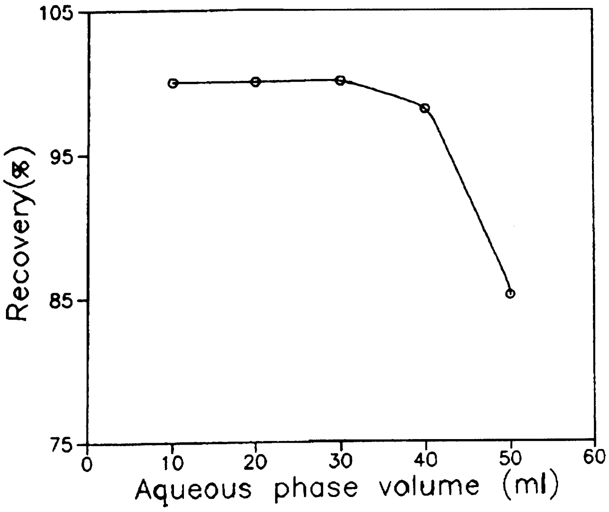

Fig. 3. Variation of aqueous phase volume.

Conclusion. Amplification reactions in their various forms already find widespread use in analytical chemistry. Successful application relies on sustained experimental skill in order to produce accurate and precise results. The proposed method can be used to determine Se(IV) with good precision and accuracy. In most of the methods reported tellurium interferes in the determination of selenium. But the present method is free from the interference of tellurium. The selectivity of the method is improved by incorporating the aluminium hydroxide collection procedure. The method allows the determination of selenium in copper ore, bark, environmental ash samples, and shampoo preparations. The results obtained clearly shows that the developed method works satisfactorily for the determination of trace amounts of selenium, in these materials.

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