

Differential Pulse Cathodic Stripping Voltammetric Determination of Trace Level of Arsenic in Blood and Urine of A Poisoned Patient: A Case Study

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Abstract

A simple, rapid and sensitive method is described for determination of arsenic in blood and urine of a poisoned patient at the level of $\mu\text{g/l}$ based on differential pulse cathodic stripping voltammetry. Only arsenic (III) is deposited on mercury electrode in the presence of copper and selenium in hydrochloric acid medium. The total arsenic is performed by reducing As (V) to As (III) using sodium metabisulphite/sodium thiosulphate reagent stabilized with ascorbic acid. The As (III) concentration in blood and urine were found to be $9\mu\text{g/l}$ and $841\mu\text{g/l}$ respectively after about twenty five days of homicidal poisoning through food and treatment as outpatient in hospital.

Keywords: Arsenic; Differential Pulse Cathodic stripping Voltammetry; Microwave Digestion System; Homicidal Poisoning.

Introduction

Arsenic is a common trace element with toxic properties known for centuries. Lethal acute intoxication by arsenic has become rare, although arsenic is still sometimes used for homicidal purposes in India. Such patients are brought to emergency room. Merely by medical examination the nature of poison cannot be decided and hence proper treatment or antidote cannot be given in time, which sometimes results in death. To overcome this problem the concerned medical officer collects the stomach wash, gastric lavage, blood, urine etc., of the poisoned patient and sent to forensic clinical toxicology division. The samples are analyzed on top priority and on the basis of report of analysis the line of treatment; an antidote is decided by the concerned medical officer and thus life of the person can be saved.

Several instrumental methods such as spectrophotometry [1], atomic absorption spectrometry [2-4], inductively coupled plasma mass spectrometry [5-6], neutron activation analysis [7] etc., are reported in literature for quantitation of arsenic in forensic case work. The objective of this work was to develop a simple, rapid and sensitive method for determination of traces of arsenic in blood and urine by differential pulse cathodic stripping voltammetry (DPCSV). One major advantage of this technique is that the running cost of instrument is quite low compared to any other technique.

Materials and Methods

All chemicals used were of analytical reagent grade. Milli Q water (from Millipore apparatus) was used throughout. Aqueous As(III) stock solution (1000ppm) was prepared by dissolving 0.131 gm As_2O_3 (Merck, India) in 100ml volumetric flask and two pellets of sodium hydroxide were added to it and made to 100ml. 0.1 ml of this solution was diluted to 100ml to give 1ppm solution. 0.142 gm of selenium dioxide (Qualigens Mumbai) was dissolved in 100ml MilliQ water. Then 0.1 ml of this solution (1000ppm) was diluted to 100 ml to give 1ppm. 0.397 gm copper nitrate (Merck, India) was dissolved in 100 ml to give 1000 ppm solution. Aqueous solutions of 14 % (w/v) sodium meta-bisulfite, 1.4 % (w/v) sodium thiosulphate were prepared.

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The reducing agent was prepared in 50 ml flask by slowly adding 5ml of 10% (v/v) sulphuric acid to 10ml of sodium meta bisulphate solution with vigorous shaking (allow to remove the gas), followed by addition of the 10ml sodium thiosulphate solution and 0.2 gm solid L-ascorbic acid, as antioxidant (Qualigens, Mumbai) and volume was made to 50 ml with Milli Q water.

Case History

A 51 year male patient (with puffiness eyes) was brought to hospital for nausea and vomiting since three days. The investigation of blood revealed among others serum creatinine, serum bilirubin, serum albumin and serum globulin in normal except SGOT (serum glutamic oxaloacetic transaminase) and SGPT (serum glutamic pyruvic transaminase) 180 and 219 respectively; higher than their normal ranges 5.0-49.0 U/L. The chemical examination of urine revealed sugar, bile pigment, urobilinogen in normal range. The microscopic examination showed pus cells, red cells, and epithelial cells within normal range. The ultrasonography of abdomen revealed all normal or-

gans except fatty liver. The patient was under treatment for 25 days as outpatient with no relief. He was having nausea, pain in abdomen with loose motion. A significant weight loss was observed with numbness/tingling sensation, in upper and lower limbs. The patient was feeling difficulty in walking. The patient was also referred to neurologist, where the nerve conduction studies were normal. In the mean time the patient told concerned physician that, he has dispute with his wife and claimed that she might have given him arsenic in food.

In order to rule out possibility of arsenic poisoning patient's blood, urine, hair and nail samples were sent to toxicology laboratory of the hospital, so that the line of treatment could be chalked out.

Microwave digestion system

Microwave digestion system (Aurora Instruments, Canada) equipped with a rotor for six teflon digestion vessels was used for the digestion of sample. Vessel of microwave digester was cleaned up by 50% (v/v) nitric acid and dried. One ml of blood/urine sample was transferred into the linear vessel and 1 ml

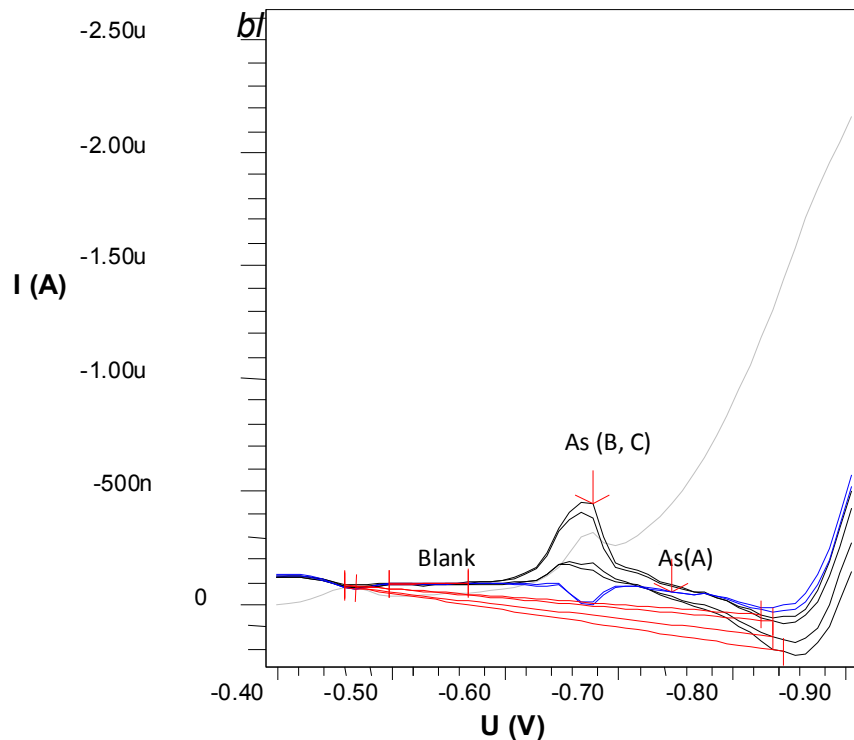


Fig.1: Voltammogram of Arsenic in blood sample obtained from standard addition technique with number of replication being two.

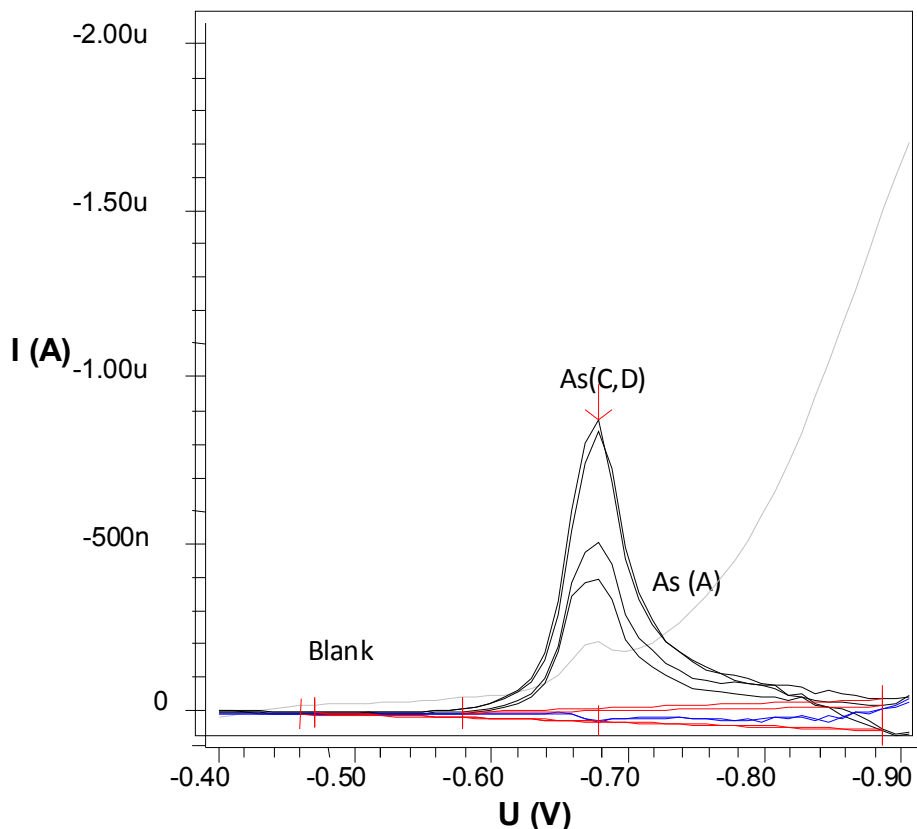


Fig.2: Voltammogram of Arsenic in urine sample obtained from standard addition technique with number of replication being two.

of water was added in reference vessel. 15 ml of 35% aqueous nitric acid was added to each vessel. The vessel carousel was loaded in microwave digestion oven and digestion machine was run in three steps of temperature range of 28-100, 100-160 and 160-170 °C for 210 sec, 300 sec, and 300 sec respectively. After cooling, the vessel was opened and the digested material was completely transferred in volumetric flasks and made to 50 ml with milli Q water in a standard flask.

Voltammetric trace metal analyzer

Trace metal analyzer (Model 797 VA Computrace from Metrohm AG Limited Switzerland) was used. The voltammetric vessel is a three electrode system. A hanging mercury drop electrode (HMDE) as working electrode, a platinum- auxiliary electrode and Ag/AgCl/3MKCl double-junction reference electrode were used.

Voltammetric determination

In all voltammetric measurements each test solu-

tion was stirred and nitrogen was purged for 5 min. 10 ml of water is taken in the measuring vessel to which 0.35 ml of reducing agent, 1 ml of conc. HCl, 80 µl of 1 ppm of selenium standard and 50µl 1000ppm copper standard solution were added and blank voltammogram is recorded from -400 mV to 900mV (Fig1 and 2). After completion of blank voltammogram 2 ml of digested sample of blood (or 0.5 ml of digested sample of Urine) was added in volumetric vessel and voltammogram is recorded under same condition (A). After completion of sample voltammogram 100 µl of 1 ppm As (III) standard was added and the voltammogram was recorded (B). Again 100 µl of 1ppm Arsenic (III) standard was added in the same vessel and voltammogram was recorded second time(C).

Results and Discussion

The voltammograms of blood and urine are given in Fig.1 and 2 respectively. All measurements were done by standard addition technique to avoid the

sample matrices effect. The sensitivity was calibrated by standard additions to the sample and the initial metal concentrations are calculated by extrapolation. Consequently the linear calibration range was automatically obtained as being related to quantitative mode of volumetric unit. The ultimate blank correction feature of the instrument was used to subtract the blank contribution due to chemicals, water, etc. A further increase in sensitivity of peak current was achieved by increasing deposition time to 60 sec. Under these conditions the concentration of Arsenic (III) in blood and urine sample of patient was found to be $9\mu\text{g/l}$ and $841\mu\text{g/l}$ respectively. However no arsenic was detected in nails and hair sample of patient. The symptoms of patient under study were same as described by Clarke [8] in arsenic poisoning cases. The advantages of proposed voltammetric method over other known techniques include its sensitivity, rapidity, cost effectiveness and sophistication. It was therefore concluded that DPCSV could be used rapidly and conveniently in the field of forensic toxicology in arsenic poisoning cases.

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