CHAPTER 4

SENSOR TESTING AND VALIDATION

4.1 Introduction

An assessment of the metal content in various environmental matrices is of great significance since they are a threat to the environment and human health. What makes them different from other pollutants is that they are not biodegradable and thus are retained indefinitely in the ecological systems and in the food chain, and eventually getting accumulated into the vital organs of humans. The most toxic heavy metals are lead, mercury, cadmium and thallium. Others like zinc, selenium and copper are essential biological elements. Although all of them have the potential to be toxic above a certain threshold concentration, they may also pose potential danger to humans at low levels should they be bioaccumulated into organisms residing at the lower food chain [14].

Such toxicity effects of heavy metals on the environment and humans demand a sensitive and reliable detection system. Electrochemical technique via stripping analysis is an interesting technique for this application. Such technique allows simultaneous measurement of different metals, trace level measurement making use of a preconcentration step, and while taking advantage of cheaper, versatile, reliable and portable instrumentation. One of the things that stripping analysis depends on for its success is the working electrode. While conventional electrochemical cells utilized bulky electrodes making decentralized measurements inconvenient, improvements have been made for their replacement with miniaturized and planar electrodes that could be used as disposable electrode. It is also important to note that traditional electrodes were mainly mercury based, which poses a problem due its toxicity effects. Thus alternate electrodes such as carbon (glassy carbon, carbon paste, carbon fiber and screen printed) and metal (amalgam, bismuth, gold, iridium and silver) have thus been developed. Such mercury free electrodes are highly anticipated, and thus there is ongoing research into optimizing these electrodes to match their analytical performance to the mercury based electrodes. Bismuth based electrodes have been extensively studied for measuring heavy metals, and their stripping performance is similar to that of mercury electrodes, while eliminating toxicity problems. Since

bismuth is a relatively new electrode material, it is vital to understand the factors that influence its favorable stripping performance.

4.2 Bismuth Film Sensor for Adsorptive Stripping Voltammetric Measurements of Ultratrace Beryllium

4.2.1 Introduction

Beryllium is recognized as the most toxic element without radioactivity [15] and its poisoning occurs primarily by inhalation of dust and gas. Beryllium is toxic both as a carcinogen and agent that causes the chronic beryllium disease (CBD). Environmental Protection Agency (EPA) has set a maximum allowable amount of 0.004 mg l^{-1} beryllium in drinking water. Despite the health hazards of beryllium, it is widely used by the aerospace, nuclear and defense industries. Such widespread industrial use reflects the unique properties of beryllium, including its low density, high stiffness and high melting point [16,17]. To protect workers from beryllium-related diseases it is essential to detect and monitor for the presence of trace amounts of beryllium.

Several methods for the determination of beryllium have been reported, including inductively coupled plasma-mass spectrometry [18] electrothermal atomic absorption spectroscopy [19] gas chromatography [20] or liquid chromatography with fluorescence detection [21]. In contrast to these sophisticated and expensive protocols, electrochemical (stripping) procedures offer great promise for obtaining ultra high sensitivity while meeting the portability, speed, cost and low-power demands of field detection of trace beryllium [22]. Since beryllium cannot be readily electrodeposited it has been measured at trace levels using adsorptive stripping voltammetry (AdSV), based on the interacial accumulation and voltammetric determination of its complexes [23,24]. Two complexing agents, thorin [23] and beryllon III [24] have been particularly useful for such AdSV measurements of beryllium. A limitation of these AdSV procedures, particularly for field screening applications, is their reliance on a mercury drop detector. Reliable AdSV sensors, based on preplated film electrodes should particularly benefit field measurements of beryllium.

Although these mercury electrodes offer an attractive AdSV performance, new alternative electrode materials with a similar performance are urgently desired for addressing growing concerns regarding the toxicity, handling, and disposal of mercury. The development of a reliable 'non-mercury' beryllium sensor should particularly benefit on-site (and especially in situ) measurements of beryllium. Bismuth electrodes have attracted considerable attention as an attractive alternative to mercury electrodes used in stripping analysis [25,26]. Most early stripping applications of bismuth film electrodes (BiFEs) focused on measurements of electrodeposited heavy metals. The suitability of BiFEs for AdSV has been demonstrated recently in connection to trace measurements of nickel [27], cobalt [28], uranium [29], chromium [30], molybdenum [31] or vanadium [32].

The aim of this work was to optimize and characterize an effective adsorptive-stripping voltammetric protocol for trace measurements of beryllium at a preplated bismuth film electrode (BiFE), based on the adsorptive accumulation of the arsenazo-III/Be complex. The arsenazo-III complexing agent dye has been shown useful for absorption spectrophotometric measurements of trace beryllium [33]. This dye is commonly used as an indicator for complexometric titrations of alkali earth metals [34].

It has been reported on the measurement of beryllium at mercury electrode [35]. However, there are no early reports on the voltammetric detection of arsenazo-III or related electrochemical measurements of its metal complexes on BiFE. As will be illustrated below, the adsorptive accumulation of Be–arsenazo-III complex onto the BiFE results in a highly sensitive and reproducible AdSV protocol for measuring trace levels of beryllium.

4.2.2 Materials and Methods

Ammonium chloride was obtained from Mallinckrodt Inc. Sodium acetate and stock solutions of beryllium (1000 mg l^{-1}) were purchased from Aldrich. Beryllium solutions were diluted daily as required. The 0.5 mM stock solutions of arsenazo-III (Sigma–Aldrich) were prepared by dissolving the appropriate amount of the ligand in nanopure water. The seawater sample, collected from Cha-Am Bay, Petchburi, was used without any pretreatment. The water

was adjusted to pH 9.7 with ammonium buffer (4:1 volume ratio of water:buffer) before the measurement. All experiments were carried out at room temperature.

Square-wave AdSV measurements were conducted using an Electrochemical Analyzer 621A (CH Instruments, Austin, TX) connected to a personal computer. The 10 ml electrochemical cell assembly (BAS, Model VC-2) consisted of bismuth-coated carbon-fiber working electrode, an Ag/AgCl (3 M KCl) reference electrode (Model CHI111, CH Instruments), and a platinum wire counter electrode. The carbon fibers (Alfa Aesar 10451, Johnson Matthey Co.,Ward Hill, MA) were pretreated first by a 12 h immersion in ethanol. Subsequently, the fibers were dipped into a 6 M nitric acid solution for 30 s, and rinsed with distilled water. This was followed by another wash with acetone, a thorough rinse with distilled water, and an air dry. A bundle of ca. 20 fibers was then glued to a copper wire with a silver conductive paint (SPI Supplies Inc., West Chester, PA). The bundle of carbon fibers was then inserted into a 100 μl plastic pipette tip, exposing a 3 mm length of the fibers at the narrow end of the tip. An internal copper wire provided the electrical contact. The narrow end of the pipette tip was then sealed with a nail polish. All glassware were soaked in 1 M nitric acid and rinsed several times with deionized water prior to use.

The bismuth-coated carbon-fiber electrode was prepared by a 15 min electrodeposition of bismuth at -0.8 V from a 0.1 M acetate buffer (pH 4.5) solution containing 20 mg l^{-1} bismuth. A similar film preparation (but in the presence of 100 mg l^{-1} bismuth) was employed in connection to the screen-printed carbon substrates.

The ammonium buffer (0.05 M, pH 9.7) supporting electrolyte solution contained 5 μM of the arsenazo-III complexing agent. The solution was first purged with nitrogen for 5 min to remove the dissolved oxygen. The electrode was poised at a potential of 0.0 V for 90 s for adsorbing the Be-ligand complex. The stirring was then stopped and after 15 s the square-wave voltammogram (SWV) was recorded over the 0.0 to -1.0 V range (using a step potential of 4 mV, amplitude of 25 mV and a frequency of 25 Hz). A 15 s 'cleaning' period (with stirring at -1.0 V) was employed between successive runs.

4.2.3 Results and Discussion

A comparison of a typical linear-sweep (A), square-wave (B) and differential pulse (C) AdSV signals at the bismuth-film electrode for 100 μg l^{-1} beryllium in the presence of 5 μM arsenazo-III recorded following a 90 s accumulation in an ammonium buffer medium (pH 9.7) is illustrated in Figure 4.1. The square-wave (B) and differential pulse (C) stripping modes resulted in well-defined beryllium signals of different sizes (E_p = -0.43 V for both (A) and (B)). A larger background slope and unwell-defined beryllium signal were observed using the linear scan mode. Both differential pulse and square-wave techniques corrected for the charging-current background contribution and yielded better signal-to-background characteristics.

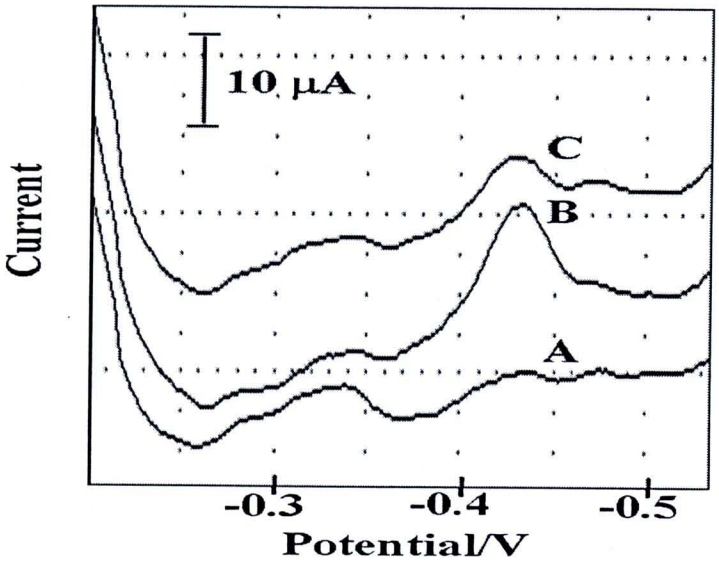
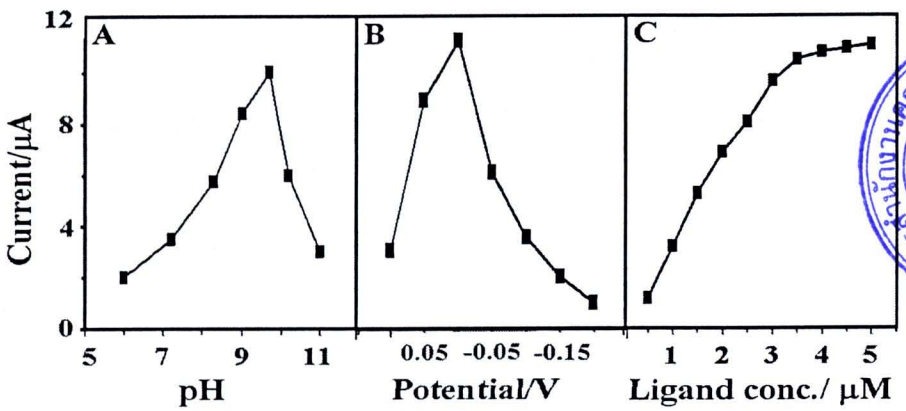


Figure 4.1 Comparison of different stripping modes: linear scan voltammetry (A); square-wave voltammetry (B) and differential pulse voltammetry; (C). Conditions: bismuth-coated carbon-fiber electrode; supporting electrolyte, 0.05 M ammonium buffer (pH 9.7) containing 100 μg l^{-1} beryllium and 5 μM arsenazo-III; nitrogen purging time, 5 min; pre-conditioning potential, -1.0 V; pre-conditioning time, 15 s; accumulation potential, 0.0 V; accumulation time, 90 s; quiet potential, 0 V; quiet time, 15 s; scanning potential window, 0.0 to -1.0 V. Scan rate, 0.1 V/s (A); amplitude, 0.025 V (B); and 0.05 V (C) potential step, 0.004 V (B and C); pulse width, 0.05 s (C); pulse period, 0.2 s (C); frequency, 25 Hz (B).

Square wave voltammetry was selected for all subsequent work due to its distinct speed and sensitivity advantages.

The influence of the pH on the Be AdSV peak current was examined over the 6.0–11.0 range (Figure 4.2A). The response increases slowly between pH 6.0 and 8.3, and very rapidly between pH 8.3 and 9.7. A sharp decrease of the signal is observed at higher pH. Such profile reflects the effect of the pH upon the complexation, adsorption and redox processes. The peak potential of the Be–arsenazo-III complex shifted gradually (from -0.43 to -0.48 V) upon increasing the pH from 6.0 to 9.7 (not shown). All subsequent work involved a solution of pH 9.7.



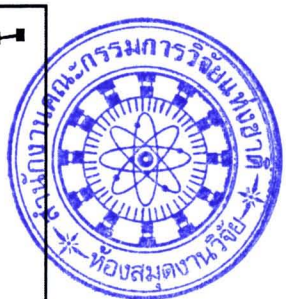


Figure 4.2 Effect of the pH (A), accumulation potential (B) and arsenazo-III concentration (C) upon the square-wave adsorptive stripping response of 100 μg l^{-1} beryllium. Other conditions, as in Figure 4.1B.

The influence of the accumulation potential on the Be stripping peak current was examined over the range of +0.10 to -0.20 V (Figure 2B). The peak rises rapidly between +0.10 and +0.05, and then more slowly up to 0.0 V. The response decreases sharply between 0.0 and -0.05 V and more slowly at more negative potentials. An accumulation potential of 0.0 V led to the highest degree of adsorption and was thus used for the further measurements. The effect of the arsenazo-III concentration upon the Be peak current is shown in Figure 4.2C. As expected for such adsorptive accumulation processes. The response increases rapidly up to around 3 μM arsenazo-III, more slowly up to 4 μM, and levels off thereafter.

The influence of the accumulation time upon the Be–arsenazo-III stripping peak current increases linearly with the accumulation time up to 60 s, then more slowly up to 120 s and starts to level off for longer periods (not shown). The resulting current–time dependence thus displays a curvature characteristic to AdSV measurements, reflecting saturation of the surface at longer accumulation periods.

The coupling of the effective adsorptive accumulation of the Be–arsenazo-III complex at the BiFE with the fast square-wave voltammetric scan results in a highly sensitive beryllium response. Figure 4.3 displays stripping voltammograms for increasing concentrations of beryllium in 10 $\mu g\ l^{-1}$ steps following a 90 s preconcentration time and using the optimised parameters. Such short accumulation results in well defined peaks for these low beryllium concentrations. As expected for adsorptive accumulation processes, the response increases linearly with the beryllium concentration up to 50 $\mu g\ l^{-1}$, then more slowly above 60 $\mu g\ l^{-1}$. Although a curvature, characteristic of adsorption processes, is observed above 60 $\mu g\ l^{-1}$, no leveling off is indicated even at high beryllium levels. (see inset for the resulting calibration plot; slope of the linear portion, 80 nA l μg^{-1}; correlation coefficient, 0.998). While the data of Figure 4.3 (curve a) indicate a detection limit of around 3 $\mu g\ l^{-1}$ (based on the signal-to-noise characteristics; S/N = 3), a substantially lower detection limit can be obtained in connection to longer accumulation times and a background subtraction operation. Such background-subtraction AdSV response for a 2 $\mu g\ l^{-1}$ beryllium solution following a 10 min preconcentration has been investigated (not shown). A well defined response, with favorable signal-to-noise characteristics, is observed, indicating a detection limit of around 0.25 $\mu g\ l^{-1}$ (27.8 nM) beryllium. Such detection limit meets the requirements of monitoring of contaminated sites and of most water quality applications.

The new electrochemical detection is suitable for measuring beryllium in natural water systems. The determination of beryllium in such water systems is indicative of the metal uptake through dust or gas sources [16]. Figure 4.4 demonstrates the suitability of the system for monitoring low levels of beryllium in an untreated seawater sample. Well defined peaks (Ep = -0.41 V) are observed for increasing beryllium concentrations in 20 $\mu g\ l^{-1}$ steps (a–g). The peak

height increases linearly with the beryllium concentration up to ca. 100 μg l^{-1} and then more slowly (slope of the initial linear portion, 50 nA l μg^{-1}; correlation coefficient, 0.995, not shown). The smaller slope, compared to that observed in the synthetic sample (of Figure 4.3), appears to reflect matrix effects, including coexisting calcium and magnesium ions and surface-active macromolecules. The low background response (unspiked sample; dotted line) indicates the absence of potential interferences.

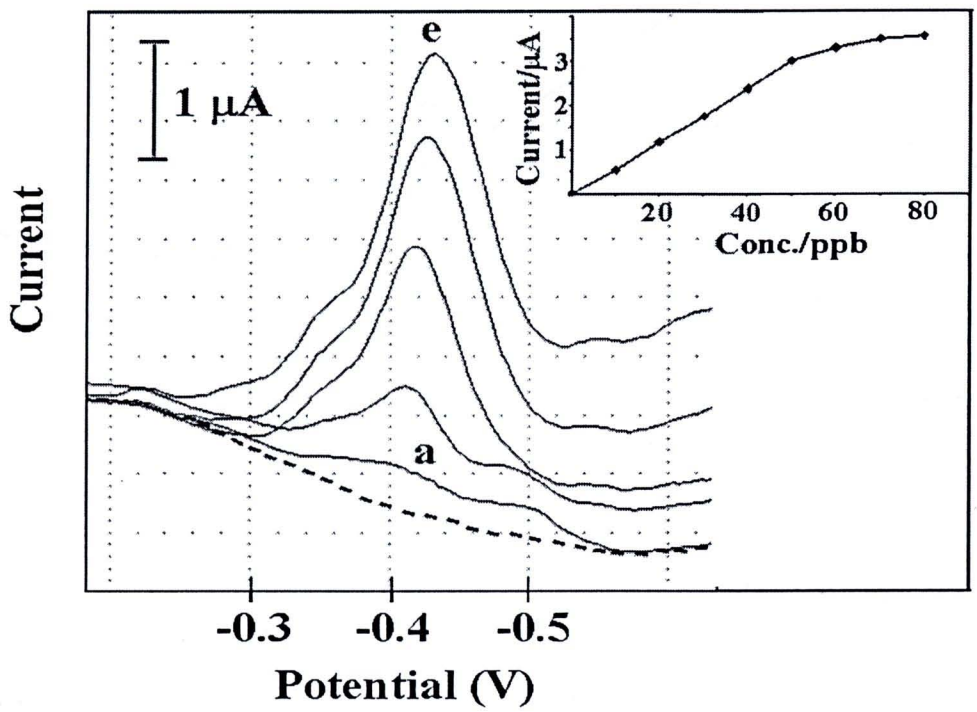


Figure 4.3 Adsorptive stripping square wave voltammograms for increasing levels of beryllium in 10 μg l^{-1} steps (curves a–e) along with the background response (dotted line). Also shown (inset) is the resulting calibration plot. Other conditions, as in Figure 4.1B.

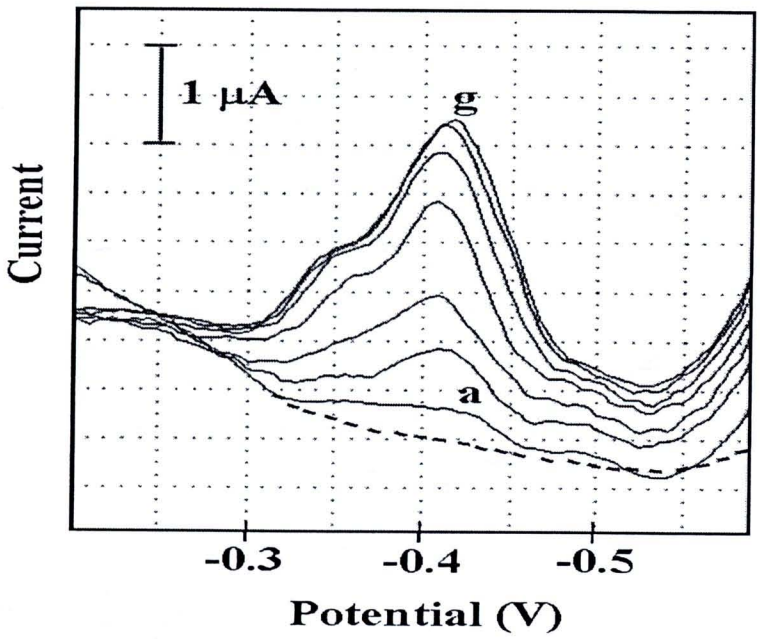


Figure 4.4 Adsorptive stripping square-wave voltammograms for a seawater sample spiked with increasing levels of beryllium in 20 μg l^{-1} steps (a–g) along with the response for the unspiked sample (dotted line). The water sample was adjusted to pH 9.7 with ammonium buffer (4:1 volume ratio of water:buffer) before the measurement. Other conditions, as in Figure 4.1B.

The long-term stability of 40 repetitive voltammograms recorded, for a seawater sample containing 100 μg l^{-1} beryllium, at 3 min intervals over a prolonged (120 min) period is illustrated (not shown). A highly stable response, with a mean peak current of 14.6 μA and a relative standard deviation of 3.9%, is observed for these 40 runs. Such stability indicates no apparent surface fouling by surface-active substances of the seawater matrix.

4.2.4 Conclusions

We have demonstrated a highly sensitive cathodic stripping protocol for detecting trace beryllium based on the adsorptive accumulation of the Be-arsenazo-III complex at a bismuth film electrode. Because of the toxicity, handling, and disposal of mercury, the new procedure obviates the need for the large mercury-drop electrode, mercury film electrode and related mercury

disposal issues. The same pre-plated bismuth film could thus be employed for multiple measurements of beryllium. The new electrochemical protocol offers great promise for meeting the portability, sensitivity, speed, cost and low-power demands of field detection beryllium. Future efforts in this direction will focus on developing single-use screen printed electrode (SPE) for on-site measurements of beryllium.

4.2.5 Acknowledgements

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