CHAPTER 3

RESULTS

3.1 Preliminary studies of bioaccumulation of treated-*Pennisetum*

Treated-*Pennisetum* was chosen to demonstrate the preconcentration/voltammeric strategy. This plant was selected because of its strong affinity towards lead(II) [84,86]. Ability of the treated-*Pennisetum* to bioaccumulate lead(II) is given in Table 3.1.

Table 3.1 Apparent lead(II) binding capacity for treated-*Pennisetum*.

No.	Lead(II) concentration (ppm)		% Removal
	before	after	
1	100	0.85	99.15
2	100	1.02	98.98
3	100	0.74	99.26
4	100	0.65	99.35
5	100	0.52	99.48
Average	100	0.76	99.24

Table 3.1 shows the ability of treated-*Pennisetum* to bioaccumulate lead(II) evaluated by atomic absorption spectrophotometry. The percentage of lead(II) retained by treated-*Pennisetum* is between and 98.98-99.48 %. The results indicate that treated-*Pennisetum* has a strong affinity towards lead(II).

In order to study the voltammetric properties of lead(II) on the electrode surface, the 10 % treated-*Pennisetum* modified carbon paste electrode was submerged in a stirred 100 ppm lead(II) standard solution for 3, 6 and 9 min respectively. The electrode was then transferred to the measurement cell containing acetate buffer pH 5.0 and ionic strength 0.60; a potential of –0.700 V was applied for 20 sec and the cyclic voltammograms were obtained between –0.700 and –0.400 V. The results obtained are shown in Figure 3.1.

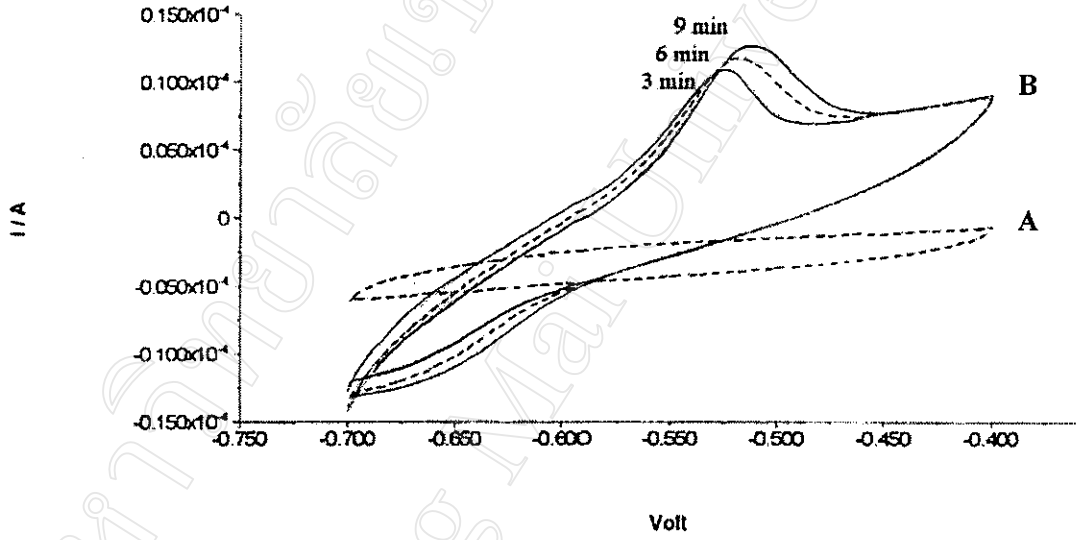


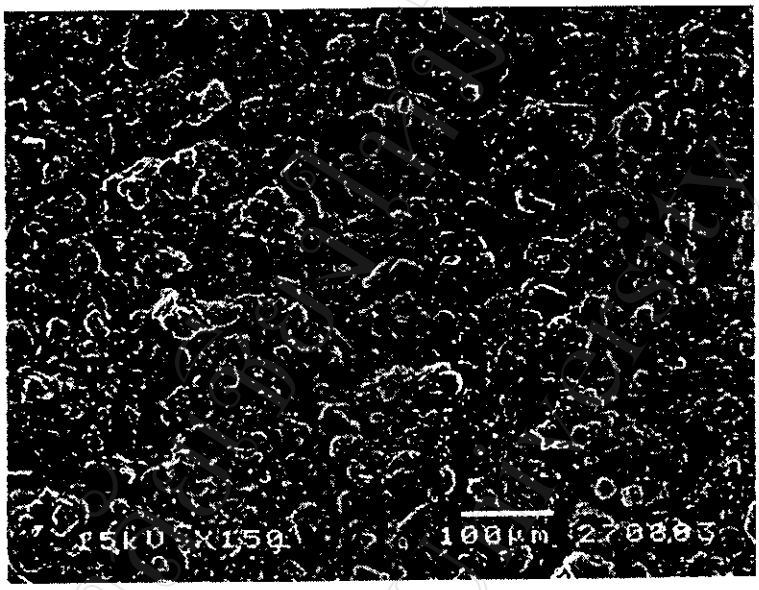
Figure 3.1 Cyclic voltammogram for 100 mg/l. lead(II) following 3, 6 and 9 min stirring at the treated-*Pennisetum* modified carbon paste electrode respectively; Scan rate, 10 mV/sec. Supporting electrolyte, acetate buffer pH 5.0 : A= Carbon paste electrode; B = treated-*Pennisetum* modified carbon paste electrode.

Figure 3.1 compares cyclic voltammograms obtained with the carbon paste electrode (A) and treated-*Pennisetum* modified carbon paste electrode (B) following 3, 6 and 9 min stirring (open circuit conditions) in a lead(II) solution. Voltammetric peaks are not observed when the unmodified electrode is used (A), as expected in the absence of lead(II) collection. At modified electrode (B) exhibits a distinct current response, corresponding to the reduction of the surface-bond lead(II). It was seen that a well-defined anodic wave at –0.600 to –0.450 V was observed. The longer the accumulation time, the more lead(II) was collected, and the larger peak current was obtained. Therefore the anodic peak was chosen for lead(II) quantification. Reoxidation is not observed upon scanning in the positive direction. In addition, no peaks are observed upon subsequent scans.

Scanning electron microscopy can offer insights of treated-*Pennisetum* at the modified electrode. Figure 3.2 shows scanning electron micrographs of the unmodified carbon paste electrode (A) and treated-*Pennisetum* modified electrode (B). The result confirm the appearance of *Pennisetum* tissue on the electrode surface. Thus, the electrode response to lead(II) solution is ascribed to the accumulation of lead(II) on *Pennisetum* tissue.

In view of the results obtained, the anodic peak was chosen for lead(II) quantification. The collection of lead(II) on a treated-*Pennisetum* modified carbon paste electrode can be used as an effective preconcentration step prior to the voltammetric measurements in the range of -0.700 and –0.400 V. In this work differential pulse voltammetry has been applied for lead(II) quantification. The 15 % treated-*Pennisetum* modified carbon paste electrode was submerged for 2 min in a stirred 1 ppm lead(II) solution. The electrode was then transferred to measurement cell containing, as supporting electrolyte, acetate buffer of pH 5.0 and ionic strength 0.60 and the differential pulse voltammograms were obtained between –0.700 and –0.450 V. The results obtained for each technique is shown in Figure 3.3.

A



B

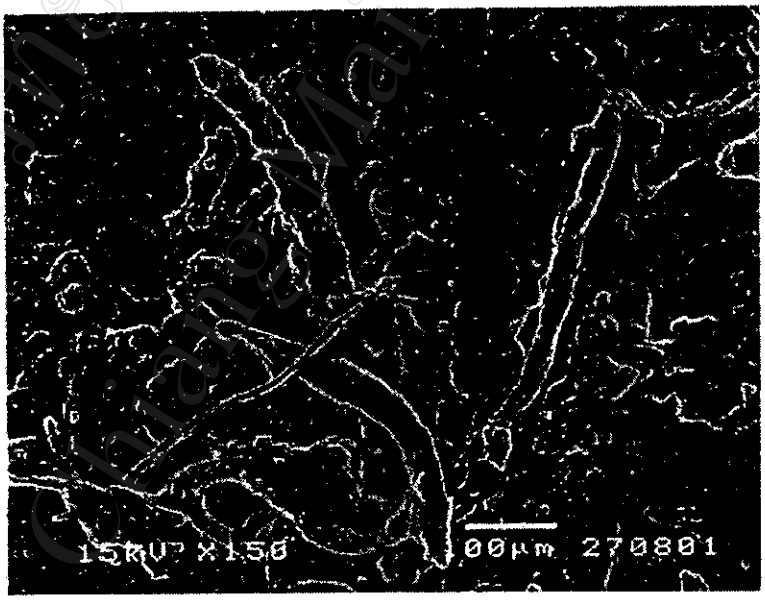


Figure 3.2 Scanning electron micrographs of the unmodified carbon paste electrode (A) and treated-*Pennisetum* modified carbon paste electrode (B).

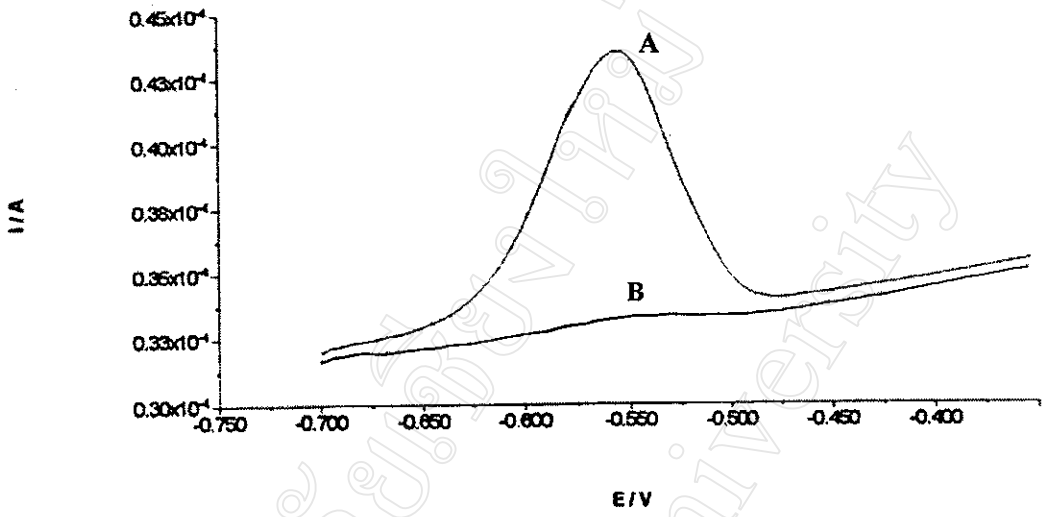


Figure 3.3 Voltammetric responses before (A) and after (B) electrode regeneration in the determination of lead(II) by differential pulse voltammetry. Support electrolyte, 0.6 M acetate buffer; scan rate, 5 mV/s; modulation amplitude, 40 mV.

Figure 3.3 shows the differential pulse voltammograms derived from 2 min preconcentration in 1 ppm lead(II) solution (A) and electrode responses after the regeneration treatment had been applied (B). Renovation of the electrode surface is easily achieved by dipping the electrode in a stirring solution of 0.05 M hydrochloric acid for 2 min. The subsequent voltammetric scan shows no lead(II) peak.

3.2 Optimization of the system

3.2.1 Effect of pH

The effect of varying the electrolyte pH on peak current for determination of lead(II) at treated-*Pennisetum* modified (15% w/w) carbon paste electrode was studied by varying the pH of electrolyte solution in the range of 3.5 - 7.0. The results obtained are shown in Table 3.2 and Figure 3.4.

Table 3.2 Effect of varying pH on voltammetric response of treated-*Pennisetum* modified (15% w/w) carbon paste electrode.

pH	Peak Current (μA)			
	no.1	no.2	no.3	Average
3.5	8.511	8.345	8.448	8.435
4.0	11.435	11.668	11.532	11.545
4.5	14.249	14.454	14.326	14.343
5.0	15.501	15.436	15.443	15.460
5.5	13.263	13.574	13.368	13.402
6.0	9.910	10.456	10.179	10.180
6.5	7.092	7.173	7.214	7.160
7.0	6.288	6.052	6.112	6.151

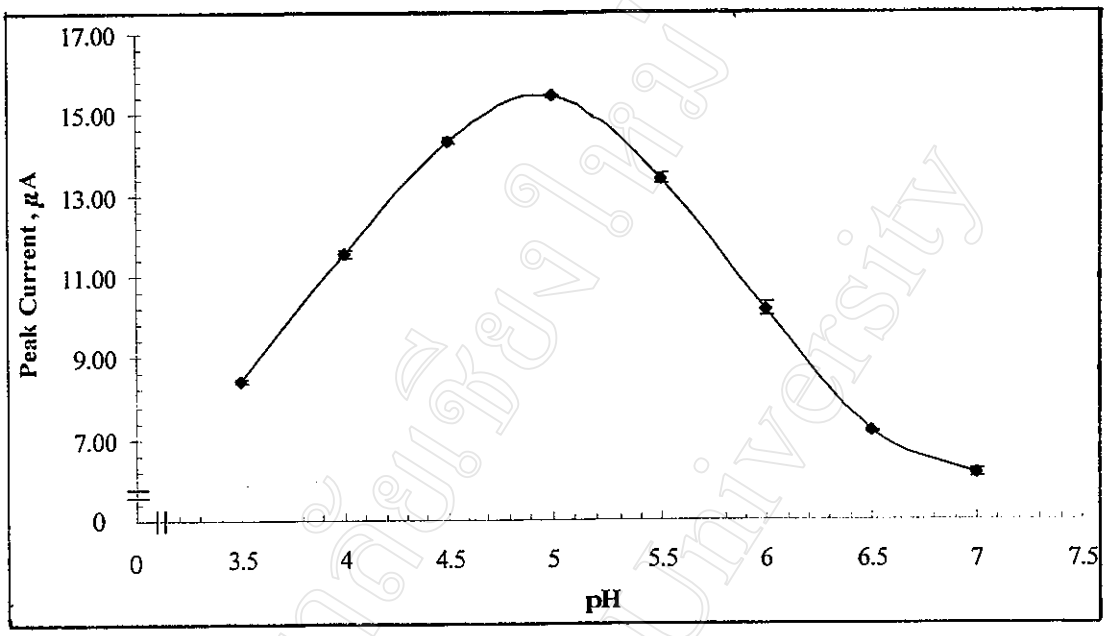


Figure 3.4 Effect of varying the pH of acetate buffer for the detection of lead(II) at treated-*Pennisetum* modified carbon paste electrode following accumulation of 1 ppm aqueous solution of lead(II) for 2 min. Detection conditions: electrolyte, 0.6 M acetate buffer; scan rate, 5 mV/s; pulse amplitude, 40 mV.

The results show that maximum peak current was obtained when the pH of electrolyte solution was 5.0.

3.2.2 Effect of accumulation time

The effect of varying the accumulation time for 0.05, 0.1 and 1 ppm lead(II) solution at the treated-*Pennisetum* modified (15% w/w) carbon paste electrode was studied by varying the accumulation time in the range of 1-10 min. The results obtained are shown in Table 3.3 and Figure 3.5.

Table 3.3 Effect of accumulation times on voltammetric response of treated-*Pennisetum* modified (15% w/w) carbon paste electrode.

Time (min)	Peak Current (μA)		
	0.05 ppm Pb^{2+}	0.1 ppm Pb^{2+}	1 ppm Pb^{2+}
1	0.411	1.046	10.147
2	0.809	2.257	15.660
3	1.439	3.481	18.542
4	2.069	4.460	19.775
5	2.582	5.899	21.051
6	2.358	7.021	22.112
8	3.980	9.031	24.124
10	5.022	10.561	25.271

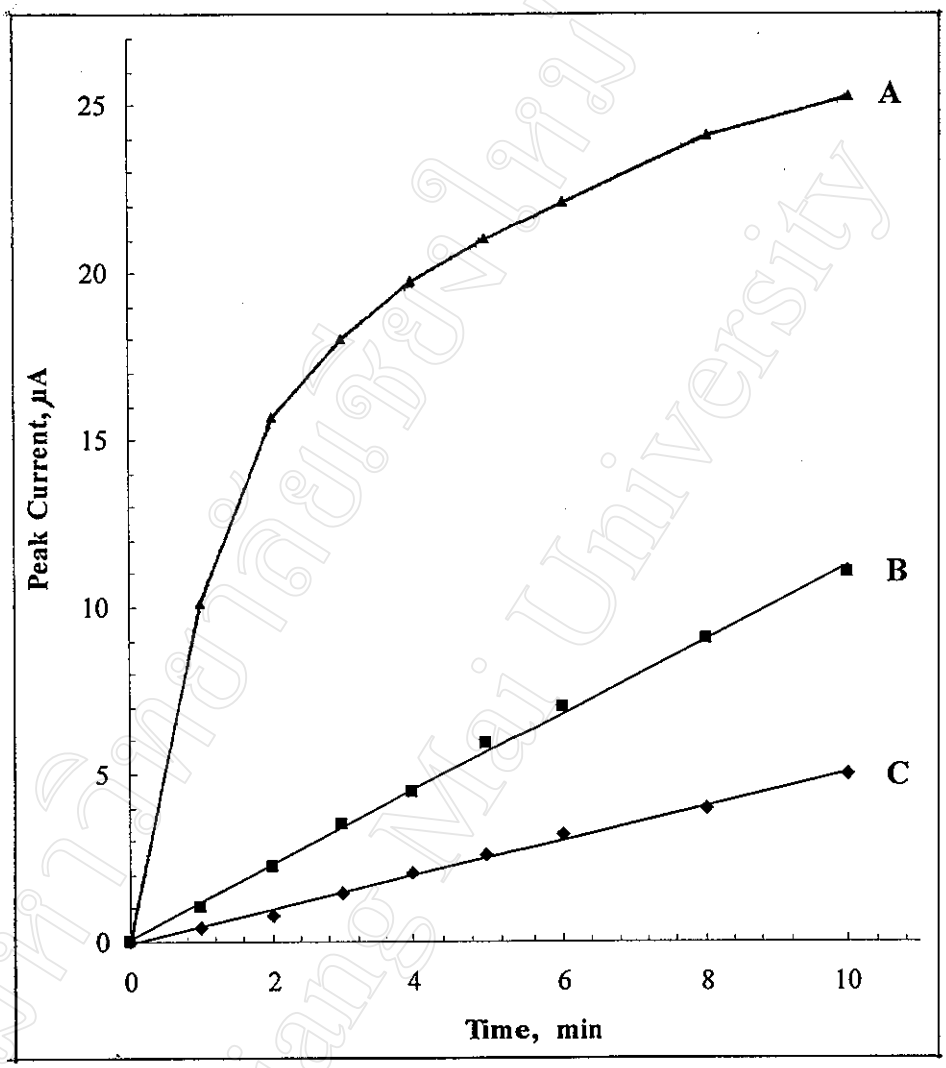


Figure 3.5 Effect of varying the accumulation time on voltammetric response. Lead(II) concentration, (A) 1.00 ppm, (B) 0.10 ppm, (C) 0.05 ppm; electrolyte, acetate buffer pH 5.0 ; scan rate, 5 mV/s; pulse amplitude, 40 mV.

Figure 3.5 shows the influence of preconcentration time on peak current for various lead(II) concentrations. A linear dependence of peak current on accumulation time is established for all concentrations. Electrode saturation for accumulation times longer than 10 min occurs for a 1 ppm lead(II) solution.

3.2.3 Effect of ionic strength

The effect of various ionic strength on peak current for determination of 0.05 ppm lead(II) at treated-*Pennisetum* modified (15% w/w) carbon paste electrode was studied by varying the ionic strength of electrolyte solution in measurement cell over the range of 0.2-1.0. The results obtained are shown in Table 3.4 and Figure 3.6.

The results indicate that the high peak current was obtain when ionic strength of electrolyte solution in measurement cell was 0.6.

Table 3.4 Effect of ionic strength on voltammetric response of treated-*Pennisetum* modified (15% w/w) carbon paste electrode.

Ionic Strength	Peak Current (μA)			
	no.1	no.2	no.3	Average
0.2	1.350	1.361	1.305	1.339
0.4	1.540	1.528	1.539	1.536
0.6	2.593	2.581	2.558	2.577
0.8	1.735	1.728	1.713	1.725
1.0	1.454	1.497	1.424	1.458

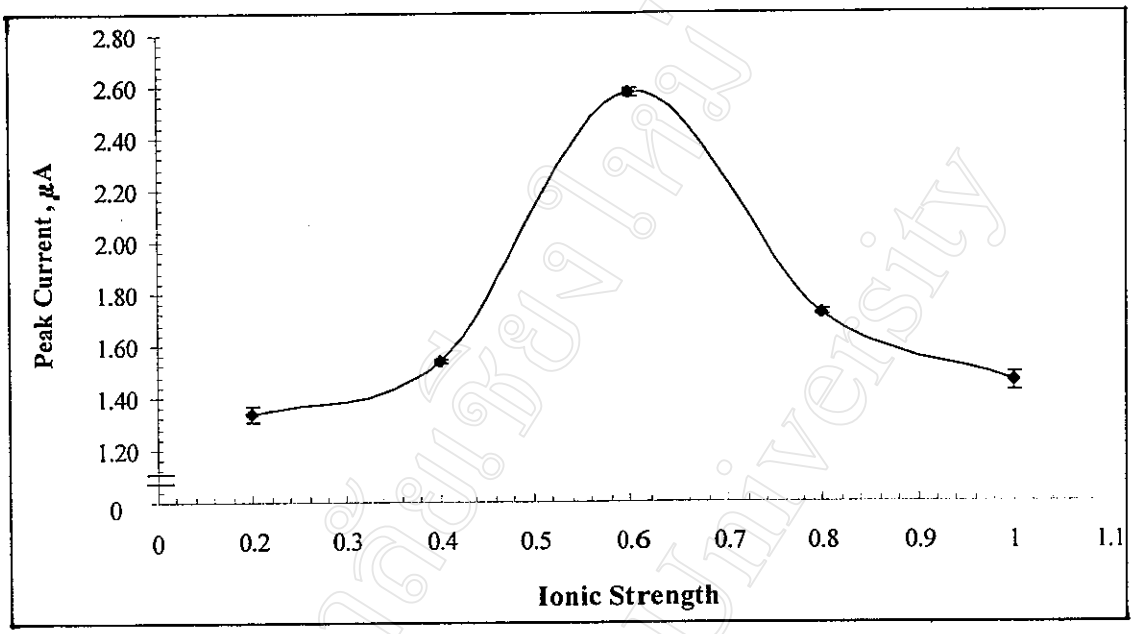


Figure 3.6 Effect of ionic strength on the peak current. Supporting electrolyte, acetate buffer pH 5.0; lead(II) concentration, 0.05 ppm; preconcentration time, 5 min; scan rate, 5 mV/s; pulse amplitude, 40 mV.

3.2.4 Effect of paste composition

The effect of various paste composition on the peak current for determination of 0.05 ppm lead(II) at treated-*Pennisetum* modified carbon paste electrode was studied by varying the percentage of treated-*Pennisetum* on paste composition in the range of 0-20 % (w/w). The results obtained are shown in Table 3.5 and Figure 3.7.

The results show that the best analytical response was obtained with the one containing 10 % (w/w) of treated-*Pennisetum*.

Table 3.5 Effect of paste composition on voltammetric response.

Treated-*Pennisetum* content (%w/w)	Peak Current (μA)			
	no.1	no.2	no.3	Average
0	0.000	0.000	0.000	0.000
5	0.976	0.992	0.982	0.983
10	4.901	4.801	4.770	4.824
15	2.573	2.571	2.589	2.581
20	0.513	0.524	0.500	0.512

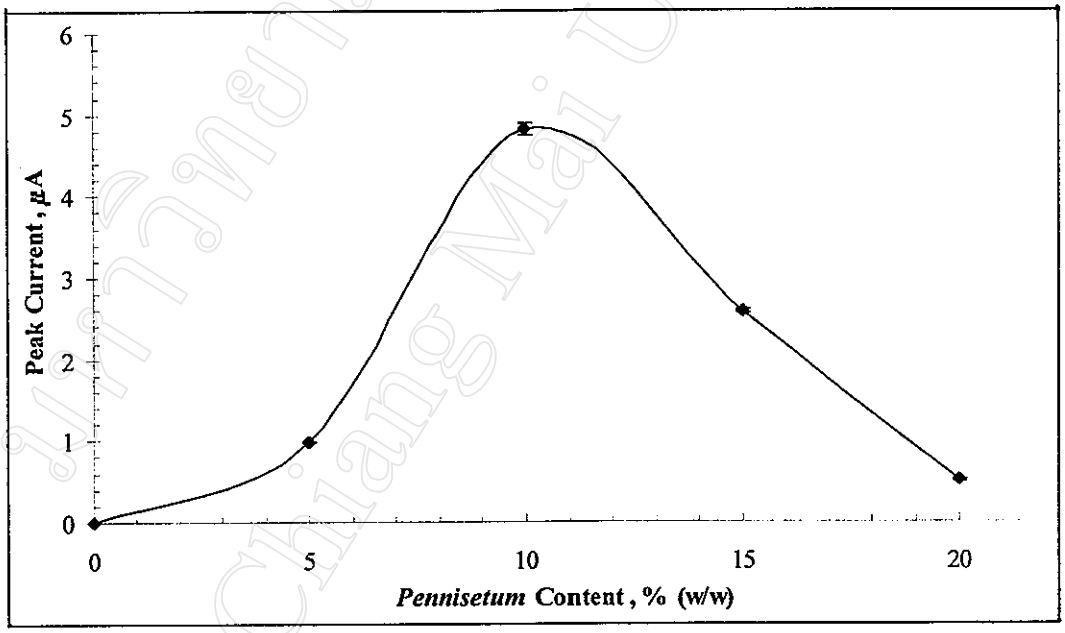


Figure 3.7 Effect of paste composition on the peak current. Supporting electrolyte, acetate buffer pH 5.0; ionic strength, 0.6; lead(II) concentration, 0.05 ppm; preconcentration time, 5 min; scan rate, 5 mV/s; pulse amplitude, 40 mV.

3.2.5 Effect of modulation amplitude

To obtain the best sensitivity of this method, effect of various modulation amplitudes on peak current for determination of 0.05 ppm lead(II) at treated-*Pennisetum* modified (10% w/w) carbon paste electrode was studied by varying the modulation amplitude in the range of 20-160 mV. The results obtained are shown in Table 3.6 and Figure 3.8.

The results indicate that high peak current was obtained when modulation amplitude was 120 mV.

Table 3.6 Effect of modulation amplitude on voltammetric response of treated-*Pennisetum* modified (10 % w/w) carbon paste electrode.

Modulation amplitude (mV)	Peak Current (μA)			
	no.1	no.2	no.3	Average
20	1.478	1.529	1.393	1.467
40	4.932	4.935	5.036	4.968
60	7.765	7.402	7.423	7.530
80	9.926	9.849	10.042	9.939
100	12.270	12.600	12.016	12.295
120	13.579	13.448	13.633	13.553
140	12.546	12.907	12.775	12.743
160	11.850	11.782	11.201	11.611

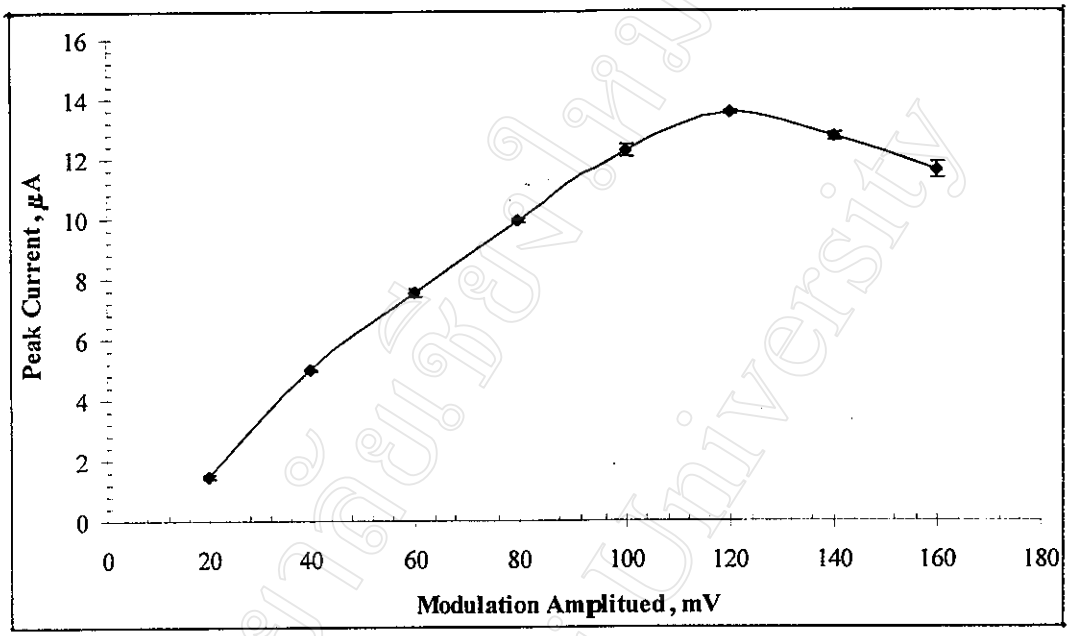


Figure 3.8 Effect of modulation amplitude on the response of treated-*Pennisetum* modified (10% w/w) carbon paste electrode. Supporting electrolyte, acetate buffer pH 5.0; ionic strength, 0.6; lead(II) concentration, 0.05 ppm; preconcentration time, 5 min; scan rate, 5 mV/s.

3.2.6 Effect of scan rate

The effect of varying scan rate on peak current for determination of 0.05 ppm lead(II) at treated-*Pennisetum* modified (10% w/w) carbon paste electrode was studied by varying scan rate in the range of 1-20 mV/sec. The results obtained are summarized in Table 3.7 and Figure 3.9.

Table 3.7 Effect of scan rate on voltammetric response of treated-*Pennisetum* modified (10 % w/w) carbon paste electrode.

Scan rate (mV/sec)	Peak Current (μA)			
	no.1	no.2	no.3	Average
1	7.060	8.966	8.651	8.230
5	14.410	13.068	13.526	13.668
10	17.606	17.856	18.053	17.838
15	18.918	19.889	19.650	19.486
20	20.797	20.012	20.435	20.415

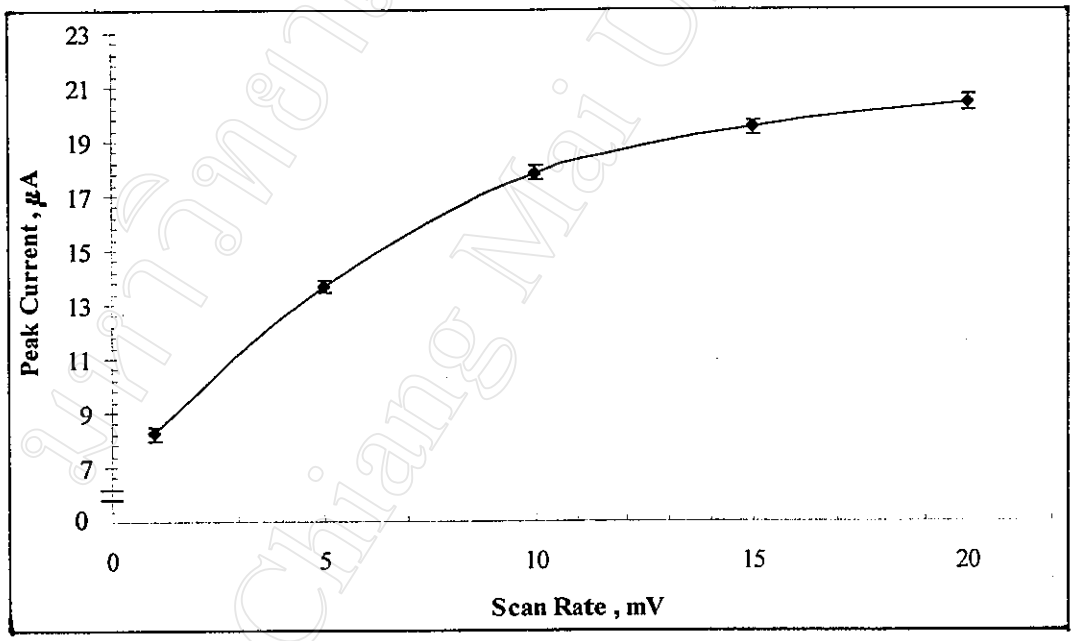


Figure 3.9 Effect of scan rate on the response of treated-*Pennisetum* modified (10% w/w) carbon paste electrode. Supporting electrolyte, acetate buffer pH 5.0; ionic strength, 0.6; lead(II) concentration, 0.05 ppm; preconcentration time, 5 min; moduration amplitude 120 mV.

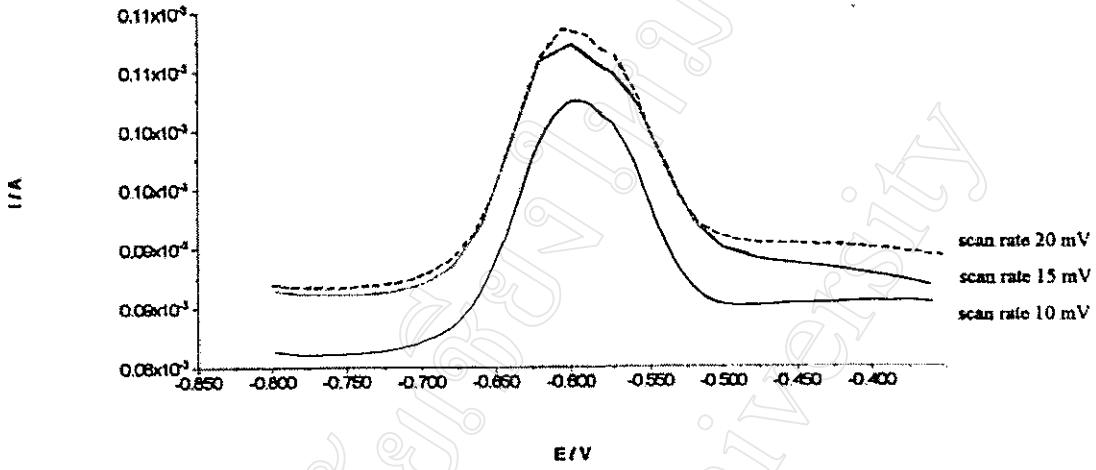


Figure 3.10 Differential pulse voltammograms for 0.05 ppm lead(II) following different scan rates. Supporting electrolyte, acetate buffer pH 5.0; ionic strength, 0.6; preconcentration time, 5 min; moduration amplitude, 120 mV.

The results show that the best analytical signal was obtained when scan rate was 10 mV/sec.

3.3 Analytical characteristics of the method

3.3.1 Linear range

The linear range of the proposed method was studied by measurement lead(II) standard solution (0.02-0.14 ppm) under the suitable condition (Table3.8). The results obtained are shown in Table 3.9 and Figure 3.11. It was found that a linear working calibration curve ranging from 0.02 to 0.08 ppm of lead(II) solution was obtained.

Table 3.8 Optimum condition for lead(II) determination.

Analytical characteristics	Parameter
pH of electrolyte solution	5.0
Preconcentration time	5 min
Ionic strength	0.60
treated-*Pennisetum* loading	10 % (w/w)
Modulation amplitude	120 mV
Scan rate	10 mV/sec

Table 3.9 Study of linear range.

Lead(II) concentration (ppm)	Peak Current* (μA)
0.00	0.000
0.02	5.869
0.04	12.772
0.06	19.125
0.08	23.523
0.10	24.998
0.12	25.686
0.14	26.214

* Average of triplicate result.

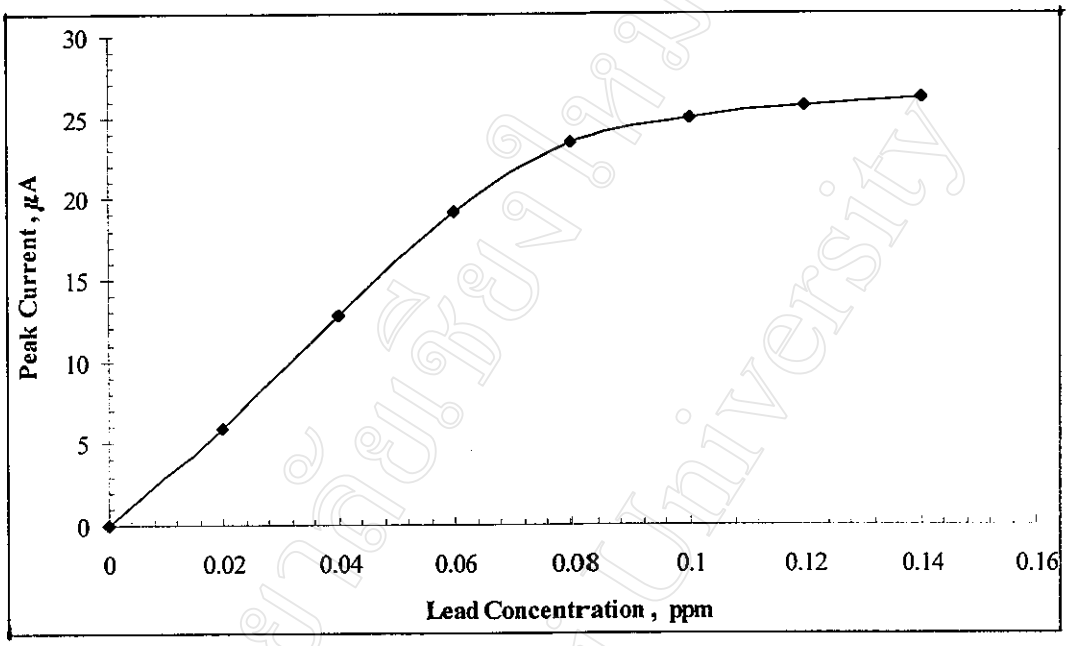


Figure 3.11 Relationship between peak current (μA) and lead(II) concentration (ppm).

3.3.2 Detection limit

The detection limit is defined as the least concentration of analyte which just gives the signal above the background signal. The results are given in Table 3.10. Based on such a definition, the detection limit of the proposed method was found to be 0.005 ppm of lead(II) solution.

Table3.10 Peak currents obtained from low concentrations of lead(II) determination.

[lead(II)] (ppm)	Peak Current* (μA)
0.008	1.533
0.007	1.292
0.006	0.769
0.005	0.530
0.004	0.000

* Average of triplicate result.

3.3.3 Calibration curve

The calibration curve of the proposed method was studied by measurement lead(II) standard solutions (0.02-0.06 ppm) using treated-*Pennisetum* and untreated *Pennisetum* modified carbon paste electrodes under the best suitable condition (Table 3.8). The results obtained are summarized in Table 3.11 and Figure 3.12.

The calibration curve shown in Figure 3.9 was established by plotting peak height versus the various lead(II) concentrations. The plots for both of the treated-*Pennisetum* and untreated-*Pennisetum* modified carbon paste electrodes gave a straight line over the entire range examined. The linear regression of the former plots was

$$Y = 305.14\,X + 0.0276 \quad (r^2 = 0.9975) \qquad (1)$$

whereas the linear regression of the latter plot was

$$Y = 185.32\,X - 0.2400 \quad (r^2 = 0.9966) \qquad (2)$$

(where Y is Peak Current in μA and X is concentration of lead(II) in ppm).

By comparison, it was found that the treated-*Pennisetum* modified electrode yields a higher response and sensitivity than the untreated-*Pennisetum* does.

Table 3.11 Calibration curve obtained from untreated and treated–*Pennisetum* modified carbon paste electrodes.

[Lead(II)] (ppm)	Peak Current* (μA)	
	untreated-*Pennisetum*	treated-*Pennisetum*
0.00	0.000	0.000
0.02	3.112	5.756
0.04	6.987	12.521
0.06	11.350	18.993
0.08	14.411	23.895

* Average of triplicate result.

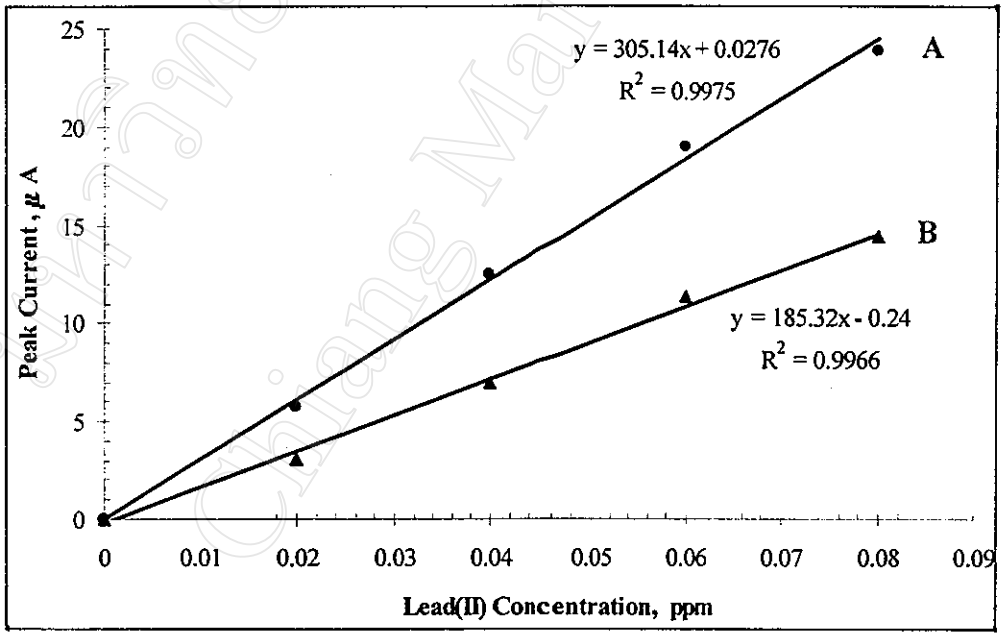


Figure 3.12 The calibration curves of lead(II) determination, using treated-*Pennisetum* modified carbon paste electrode (A) and untreated-*Pennisetum* modified carbon paste electrode (B).

3.3.4 Reproducibility of the system

The reproducibility of the system was determined by repeating measurements of 0.05 ppm lead(II) solution for 16 replicates. The results obtained are summarized in Table 3.12.

Table 3.12 Reproducibility of replicate determination of lead(II).

Experimental no.	Peak Current (μA)
1	19.296
2	17.224
3	18.233
4	16.622
5	17.754
6	16.165
7	17.039
8	19.460
9	17.596
10	17.474
11	18.612
12	17.863
13	18.052
14	17.695
15	18.761
16	16.452
Average	17.769
Standard deviation	0.958
% RSD	5.39 %

3.3.5 Repeatability of the system

The repeatability of the system was pursued by preparing 10 solutions of 0.05 ppm lead(II). All these solutions were then singly measured. The results obtained are summarized in Table 3.13.

Table 3.13 Repeatability of replicate determination of lead(II).

Experimental no.	Peak Current (μA)
1	16.477
2	19.425
3	18.015
4	19.686
5	17.548
6	16.957
7	17.586
8	18.231
9	17.899
10	16.362
Average	17.819
Standard deviation	1.109
% RSD	6.225

3.3.6 Effect of interference

The effect of some possible interferences was studied by adding known amounts of each interference to 0.05 ppm lead(II) standard solution. The electrode response for those solutions were then measured. The results obtained are presented in Table 3.14.

Table 3.14 Effect of possible interferences.

Interference	Pb(II):Interference (ppm) : (ppm)	Ratio	Peak Current* (μA)	% Relative error
Pb^{2+}	0.05 : 0.00	1 : 0	17.586	-
Fe^{3+}	0.05 : 0.50	1 : 10	17.106	- 2.73
	0.05 : 1.00	1 : 20	16.456	- 6.43
	0.05 : 2.00	1 : 40	9.648	- 45.14
	0.05 : 5.00	1 : 100	3.112	- 82.30
Cu^{2+}	0.05 : 0.05	1 : 1	16.360	- 6.97
	0.05 : 0.25	1 : 5	4.880	- 72.25
	0.05 : 0.50	1 : 10	2.671	- 84.81
Zn^{2+}	0.05 : 0.25	1 : 5	15.327	- 12.84
	0.05 : 0.50	1 : 10	12.348	- 29.78
	0.05 : 2.00	1 : 20	5.497	- 68.74
	0.05 : 5.00	1 : 100	4.753	- 72.97
	0.05 : 10.00	1 : 200	3.124	- 82.23
Hg^{2+}	0.05 : 0.50	1 : 10	17.737	0.86
	0.05 : 1.00	1 : 20	15.032	- 14.52
	0.05 : 5.00	1 : 100	12.569	- 28.53
	0.05 : 10.00	1 : 200	10.557	- 39.97
Al^{3+}	0.05 : 0.50	1 : 10	8.364	- 52.44
	0.05 : 1.00	1 : 20	4.715	- 73.19
	0.05 : 5.00	1 : 100	1.423	- 91.91
	0.05 : 10.00	1 : 200	0.633	- 96.23
Ba^{2+}	0.05 : 0.50	1 : 10	18.053	2.65
	0.05 : 1.00	1 : 20	16.211	- 7.82
	0.05 : 5.00	1 : 100	4.551	- 74.12
	0.05 : 10.00	1 : 200	2.916	- 83.42
Na^{+}	0.05 : 0.50	1 : 10	17.728	0.80
	0.05 : 1.00	1 : 20	17.389	- 1.12
	0.05 : 5.00	1 : 100	14.510	- 17.49
	0.05 : 10.00	1 : 200	13.657	- 22.34
K^{+}	0.05 : 0.50	1 : 10	18.562	5.55
	0.05 : 1.00	1 : 20	17.932	1.97
	0.05 : 5.00	1 : 100	16.105	- 8.42
	0.05 : 10.00	1 : 200	9.946	- 43.44

* Average of triplicate result.

3.4 Determination of lead(II) in mineral water samples

The method was applied to determine lead(II) in mineral waters. Primary determination of these samples revealed non-detectable lead(II) concentration. As a consequent, standard addition method was utilized instead. This can be carried out by spiking the samples with 0.05 ppm of lead(II) solution. To certify correctness of the analytical method, comparison the data obtained between the described method and atomic absorption spectrometry was made. The data obtained are summarized in table 3.15.

Table 3.15 Comparison the data obtained between the proposed voltammetric method and atomic absorption spectrophotometry (AAS) for determination of non spike Pb(II) and spike 0.05 ppm Pb(II) in mineral water samples.

Sample	Lead(II) concentration*, ppm			
	Voltammetric method		AAS	
	non spike	spike	non spike	spike
No. 1	nd.	0.056	nd.	0.054
No. 2	nd.	0.053	nd.	0.050
No. 3	nd.	0.056	nd.	0.054
No. 4	nd.	0.061	nd.	0.058
No. 5	nd.	0.052	nd.	0.051

* Average of triplicate result.