CHAPTER 3

RESULTS AND DISCUSSION

3.1 Automated on-line solvent extraction devices coupled with FAAS

3.1.1 An automated on-line solvent extraction and FAAS for some heavy metals determination in soil

3.1.1.1 Signal enhancement by using organic solvent instead of aqueous solution

The enhancement of signals by using organic solvent comparing with signal in aqueous solution for each metal was investigated by analysing organic and aqueous solutions of same concentrations of metals. The manual batch extractions of each metal in MIBK were done and the extraction was aspirated directly to FAAS. The results were compared with the signals obtained from those prepared in aqueous solution. The results are represented in **Table 3.1**. Signal enhancement of about 2-3 times when using organic solvent instead of aqueous solution as medium was observed.

There are several explanations for this effect. The organic solvent has a lower viscosity and a lower specific gravity than water, thus it's easier to aspirate. The lower surface tension of organic solvent brings about finer nebulisation so that more sample amount reaches the flame per time unit. Moreover, the evaporation of water is a strong endothermic reaction that reduces the flame temperature, while the combustion of organic solvent is exothermic reaction increasing the flame temperature. A higher flame temperature can lead to better atomisation of the element. Therefore, the signal was enhanced when using organic solvent [59].

Table 3.1 Signal enhancement by using organic solvent

Metals	Absorbance organic (A) and aqueous (B) ratio (A/B)
Cu	2.3
Pb	3.1
Ni	2.8
Cd	2.2
Zn	2.5

3.1.1.2 The influence of the uptake rate of organic solvent to signal

The influence of the uptake rate of organic solvent to signal was studied. The manual extraction of 1 mg l^{-1} Cu(II) standard solution (aqueous:organic ratio=1) was performed. The uptake rates in the range of 1.5-7.5 ml min^{-1} were varied by changing various sizes of nebuliser tube and also adjusting the nebuliser knob. The results are illustrated in **Figure 3.1**.

It was found that the higher uptake rate of organic solvent, the greater signal obtained. This may be due to higher amount of sample solutions were entered the flame lead to increasing of signal. However, when the uptake rate higher than 5.0 ml min^{-1}, the decrease of signal was observed. This may be due to the less efficiency of nebulisation when higher uptake rate was applied. Thus the uptake rate of 5 ml min^{-1} was selected for next experiment.

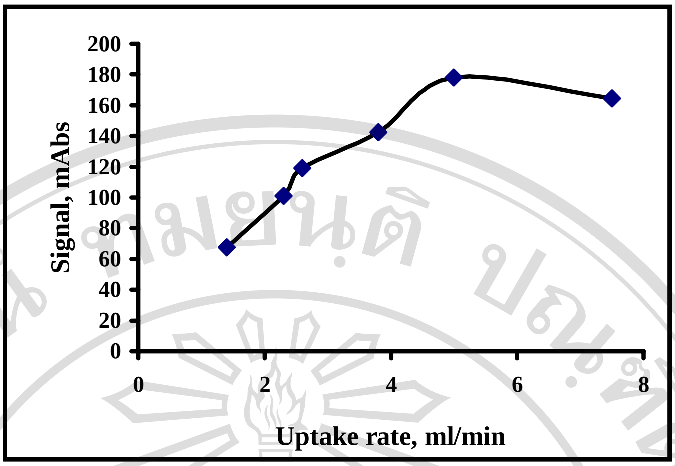

Figure 3.1 The influence of uptake rate of organic solvent on signal for 1.0 mg l^{-1} Cu(II) standard solution

3.1.1.3 Optimisation for an injection volume

The effect of injection volume was investigated. The standard solutions of Cu(II) in the range of 10-1,000 μg l^{-1} in 0.1 M HNO_3 were prepared. The results are presented in **Table 3.2** and **Figure 3.2**.

It was found that higher injection volume produced higher sensitivity (better slope of a calibration graph). The sensitivity slightly decreased when the injection volume was larger than 150 μl. The injection volume of 150 μl gave the highest sensitivity, however, peak shape was well defined and precision was the best when using injection volume of 100 μl. Smaller injection volume was preferred because less organic phase was used and more convenient to fill sample loop up. Hence, an injection volume of 100 μl was selected for next experiments.

Table 3.2 Effect of injection volume

Injection volume (μl)	**Equation of calibration graph (Y=aX+b)**	**r^2**	**%RSD (200 μg l^{-1} Cu(II)), n=5**
50	0.2159x+9.9438	0.9910	24.1
75	0.3512x+3.6890	0.9941	10.6
100	0.4396x+29.682	0.9993	1.7
150	0.4811x+22.044	0.9999	2.9
200	0.4552x+39.606	0.9976	3.3

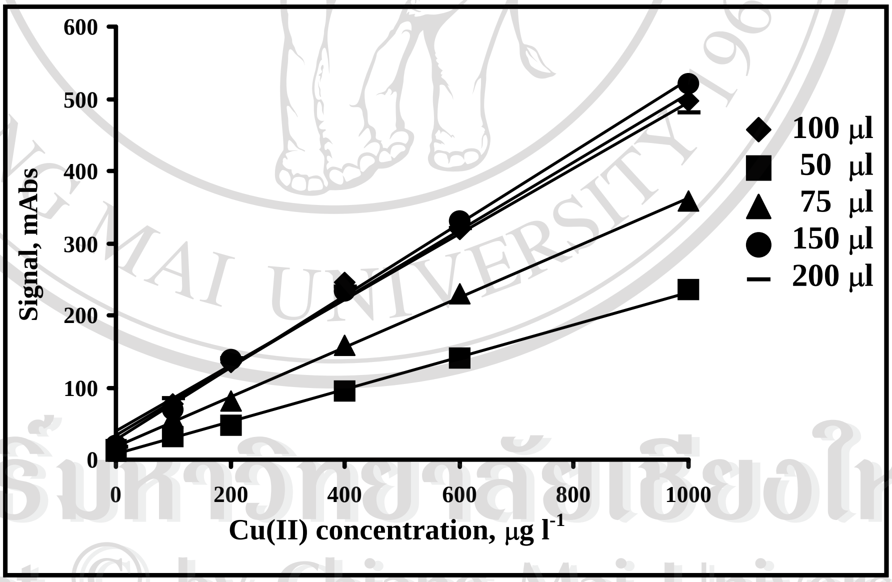

Figure 3.2 The effect of injection volume on calibration graph

3.1.1.4 Optimisation for extraction time

The standard solution of 500 μg l^{-1} of Cu(II) was used to optimise the extraction time. **Figure 3.3** presents the results of the effect of extraction time to signal. Extraction time of 30 s was selected because no signal increase was observed for the extraction time longer than 30 s. Longer extraction time provided better formation between Cu(II) and DDC and faster transferring to organic layer. In order to compromise between high signal and short analysis time, 30 s of extraction time was chosen.

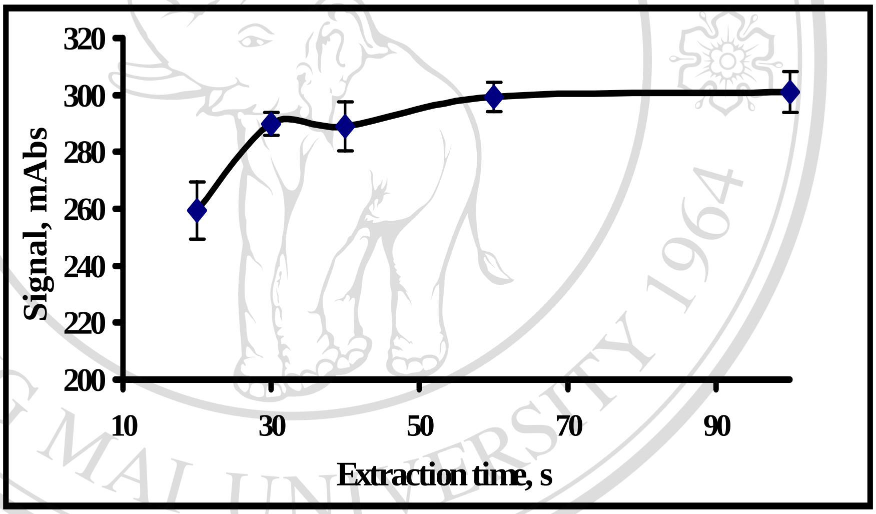

Figure 3.3 The effect of extraction time on signal

3.1.1.5 Optimisation for pause time

The standard solution of 500 μg l^{-1} of Cu(II) was used to optimise pause time. As it can be seen in **Figure 3.4**, pause time in the range of 10 s to 25 s gave no significant difference on signal. This can be implied that organic phase and aqueous phase can be separated well by using only 10 s of pause time. To ensure that organic phase can efficiently separate from aqueous phase, the pause time of 20 s was chosen for next experiments.

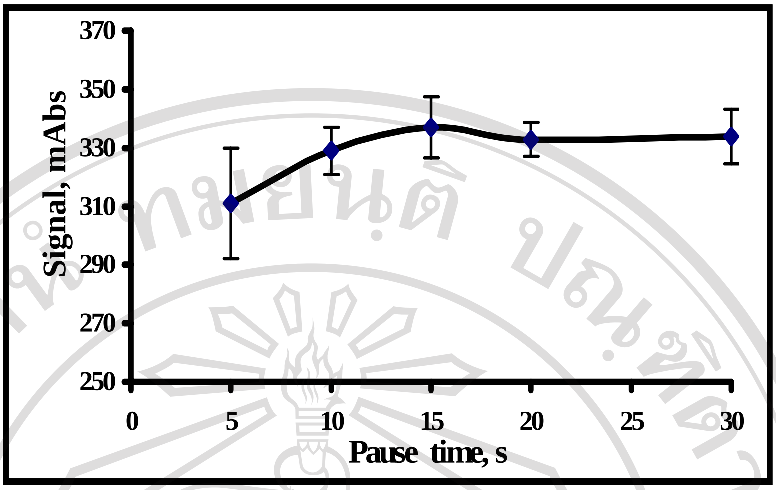

Figure 3.4 The effect of pause time on signal

3.1.1.6 Study the carry-over effect and optimisation for methanol rinsing time

The carry-over effect of the system was studied. Methanol was used as a rinsing solution. To observe this effect, the rinsing time of methanol was varied from 0-30 s. In each rinsing time, standard solution of 2 mg l^{-1} Pb(II) was determined for 3 times. The average signal was calculated. After that determination of blank (0.1 M HNO_3) was performed again. The % carry-over was calculated as **equation 3.1**. The results are presented in **Figure 3.5**. It was observed that rinsing step was necessary to avoid carry-over effect. At least 10 s of rinsing time must be applied to minimise carry over effect down to 5%. The rinsing time of 15 s was chosen to ensure less carry-over effect remained.

% Carry-over = Ax100/B **…………..equation 3.1**

where A = first signal of blank after running of 2.0 mg l^{-1} Pb,

B = average signal of 2.0 mg l^{-1} Pb.

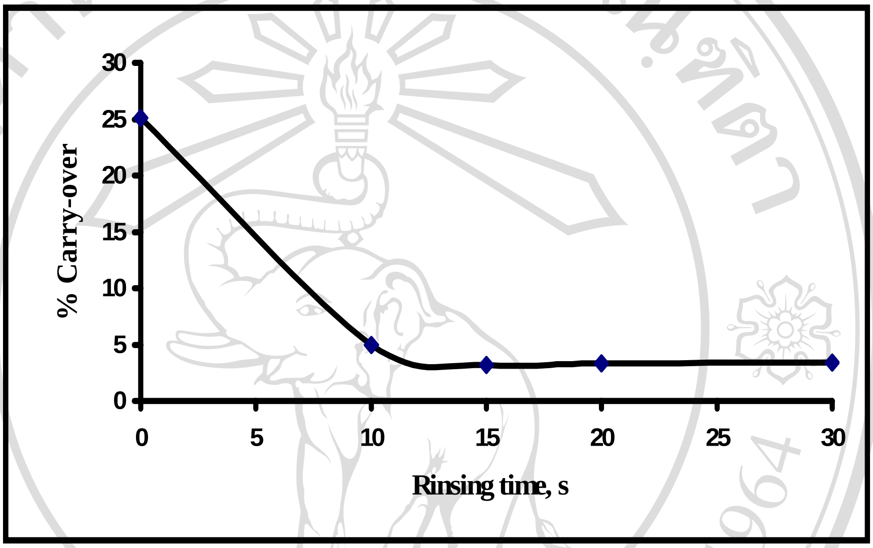

Figure 3.5 Optimisation of methanol rinsing time

3.1.1.7 The effect of pH on signal

As mentioned before in **Section 1.4.1**, pH of medium solution affects the extraction efficiency. To investigate this effect, 250 ml of 1.0 mg l^{-1} standard solutions of each metal solution were prepared. An Acetate buffer was used to control pH. The pH was varied in the range of 0-4 by adding conc. HCl. The results are shown in **Figure 3.6A** and **Figure 3.6B**.

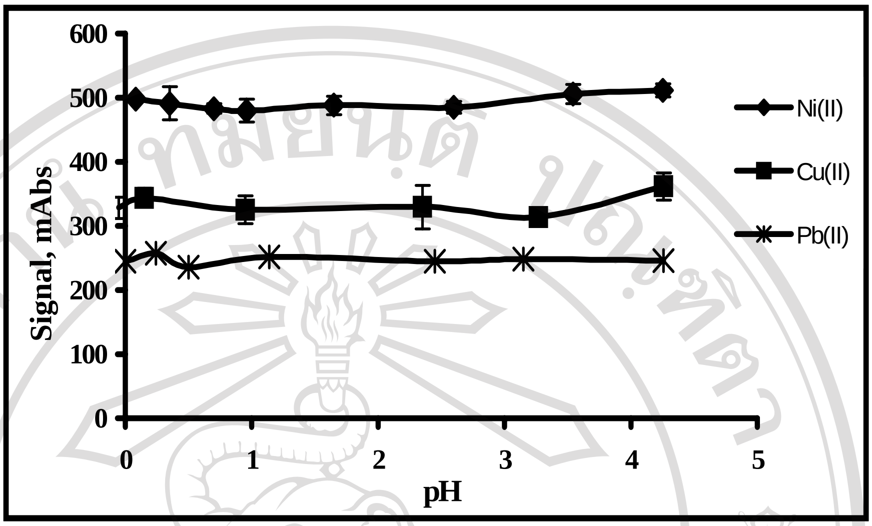

Figure 3.6A The effect of pH on signal for Cu(II), Pb(II) and Ni(II) determination

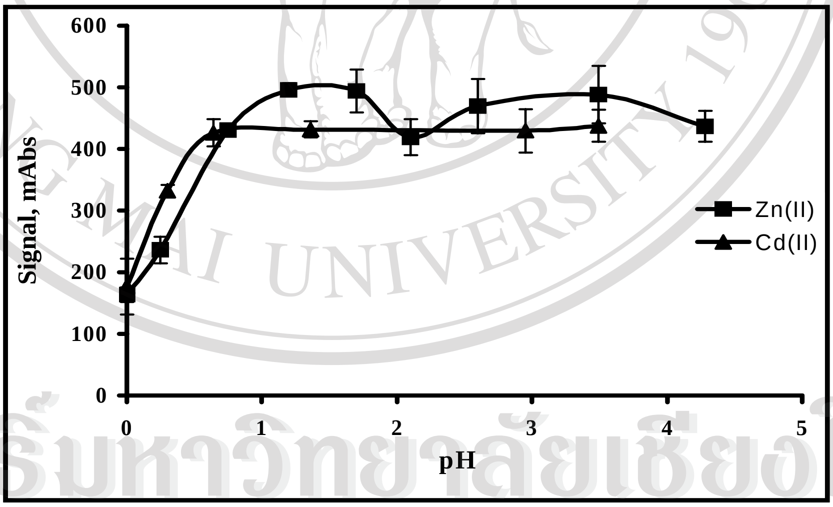

Figure 3.6B The effect of pH on signal for Cd(II) and Zn(II) determination

It is indicated that there is no significant difference on signal at different pH values for Cu(II), Pb(II) and Ni(II) determination. Therefore, it is possible to determine these metals in a digest solution without any pH adjustment. However, pH has effect to Cd(II) and Zn(II) determination obviously. Both of Cd(II) and Zn(II) could not be extracted efficiently at pH approximately lower than 1.0. It is necessary to adjust pH of sample before determination. These are corresponding to calculated extraction as presented in **Table 1.1**.

3.1.1.8 Summary of optimum conditions

The optimum conditions can be summarised as presented in **Table 3.3**.

Table 3.3 Optimum conditions for the automated on-line solvent extraction unit

Parameter	**Condition**
Manifold and components	see **Figure 2.1**
Chemicals	1.0% DDC for organic ligand, 0.1 M HNO_3 for sample medium, MIBK for organic phase and methanol for rinsing solution
Uptake rate of organic solvent, ml min^{-1}	5
Injection volume, μl	100
Extraction time, s	30
Pause time, s	20
Methanol rinsing time, s	15
pH for standard/sample	no need to adjust pH after digestion for Cu(II), Pb(II) and Ni(II) but pH 1.0 for Cd(II) and Zn(II)

3.1.1.9 Interference by Fe(II)

Normally, high amounts of Fe are presented in soil samples. Fe(II) can be co-extracted with heavy metals. The extraction efficiency of Fe(II) is influenced very much by pH. To study the effect of Fe(II) on signal, pH 0.5 was applied to study for 0.5 mg $l^{-1}$of Cu(II), 1.0 mg $l^{-1}$of Pb(II) and Ni(II), pH 1.0 for 0.1 mg $l^{-1}$of Cd(II) and Zn(II). The results are shown in **Figure 3.7**. It was observed that there is no significant difference on signal for 0.5 mg l^{-1} Cu(II) if Fe(II) up to 1,000 mg l^{-1} was presented. Unfortunately both of 1.0 mg $l^{-1}$of Pb(II) and 1.0 mg $l^{-1}$of Ni(II), the Fe(II) has effect to signal if the concentration of Fe(II) exceeds 500 mg l^{-1}. For 0.1 mg $l^{-1}$of Zn(II), and 0.1 mg $l^{-1}$of Cd(II) only 200 and 400 mg $l^{-1}$of Fe(II) could interfere to the Zn(II) and Cd(II) determination, respectively.

It was found that higher tolerance of Fe(II) was observed when performed in lower pH. Fe(II) can be co-extracted less efficient in acidic condition. Such low pH often is encountered in acidic digest of soil samples. It is possible to extract the metals directly from this digest. Thus, this can avoid the interfering Fe(II) to be co-extracted with the analyte. In the case of Fe(II) co-extracting with analyte, large amounts of DDC will be used to form complex with Fe(II) resulting in not enough DDC to form complex with the analyte. Therefore, the signal will decrease. To overcome this effect, DDC concentration should be increased to 2% of DDC for Cd(II) and Zn(II) determination.

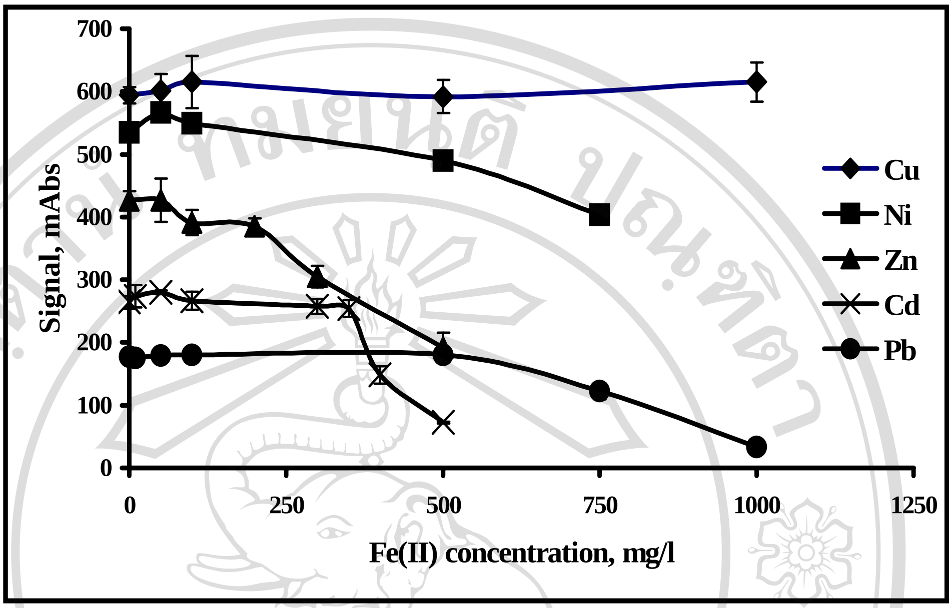

Figure 3.7 Effect of Fe(II) on signal

3.1.1.10 Precision

The precision of the proposed method was investigated by aspirating 11 replicates of standard solutions of each metal solution by using the optimum conditions as summarised in **Table 3.3**. The precision of the method for each heavy metal is not more than 5% as represented in **Table 3.6**.

3.1.1.11 Sample preparation

A Dry soil sample (1-5 g) was weighed in a blue cap bottle. Then nitric acid (7.0 M) was added. It was left to stand for 30 min to evaporate perfume causing from organic material digestion. Then the blue cap bottle was placed into autoclave and the autoclave was turned on. The autoclave was heated to 120 ^{0}C for 30 min. Then the heat was switched off and the autoclave was cooled down and then opened. The supernatant was decanted and the residue was rinsed with distilled water to get as much as possible of dissolved metals out. The content was diluted to 100 ml in a volumetric flask. Eventually, the digested sample was transferred to a plastic container.

3.1.1.12 Application

The proposed system was validated by using soil reference materials. The results are shown in **Table 3. 4**.

Table 3.4 Application of the proposed method for heavy metal determination in soil reference materials

Heavy metals	**Soil-5, n=2 (mg kg^{-1})**		**DS-259, n=2 (mg kg^{-1})**	
	Developed method	**Certified value^a**	**Developed method**	**Certified value^b**
Cu	72.0	72.0	15.0	14.0
Pb	144.6	146.0	28.5	22.0
Ni	7.5	7.3	7.6	7.6
Cd	1.13	1.16	0.19	0.17
Zn	390	360	47	55

[a] certified value of soil reference material from IAEA

[b] value for house standard, E&R, DTU. Precision better than 10%

*n= replication, Soil-5= soil reference material and DS-259 = house standard for soil, E&R, DTU

The proposed system was also applied to soil samples. The results are shown in **Table 3.5.**

Table 3.5 Application of the proposed method for heavy metal determination in soil samples

Heavy metals	**A, n=6 (mg kg^{-1})**		**B, n=6 (mg kg^{-1})**	
	Developed method	**AAS method**	**Developed method**	**AAS method**
Cu	11.0 ±0.5	9.9±0.3	14.0±1.0	13.8±0.9
Pb	7.8±0.7	8.6±1.8	10.4±0.9	13.4±1.8
Ni	1.06±0.08	1.34±0.23	17.4±1.2	19.8±1.3
Cd	0.15±0.01	<DL[a]	0.13±0.01	<DL[b]
Zn	12.1±0.9	13.0±0.7	43.9±1.4	38.9±2.0

A = soil sample collected from Lundgard located in Denmark
B = soil sample collected from Ronhave located in Denmark
DL= detection limit
By standard addition, [a]0.15±0.02 and [b]0.14±0.01
*n= replication

It was found that the results obtained by the proposed method were corresponding to those obtained from certified values for soil reference materials. For heavy metals determinations in soil samples, the results obtained by using the proposed method agreed well with those obtained by using conventional method (evaluated by t-test value at 95% confidence level).

3.1.1.13 Characteristics of the procedure

The analytical characteristics of the proposal procedure are shown in **Table 3.6.**

Table 3.6 Analytical characteristics of the automated on-line solvent extraction coupled with FAAS for the determination of the studied heavy metal ions

Characteristics	**Metals**				
	Cu	**Pb**	**Ni**	**Cd**	**Zn**
Linear range, $\mu g\ l^{-1}$	50-300	100-600	50-400	2-20	25-250
Regression equation (Y= aX+b)	0.5579x-0.3356	0.1869x-0.1355	0.4955x+5.1275	3.7964x+1.3402	1.8953x+6.3915
Correlation coefficient (r^2)	0.9991	0.9987	0.9990	0.9952	0.9976
Precision, %RSD, n=11	3.0	5.0	4.1	4.4	4.4
Detection limit[1], $\mu g\ l^{-1}$	10.0	28.8	34.3	0.7	14.4
Detection limit[2], $mg\ kg^{-1}$	0.1	2.9	3.4	0.1	1.4
Sample frequency (f), h^{-1}	23.7	23.5	23.7	22.4	21.2
Enrichment factor (EF)[3]	5.3	14.7	10.2	6.6	2.9
Concentration efficiency (CE)[4], min^{-1}	2.1	5.8	4.0	2.5	1.0

[1] Detection limit defined as 3 times of blank deviation plus blank signal [105]

[2] Detection limit, mg kg^{-1} defined as the detection limit in mg kg^{-1} for 5 g of sample in 50 ml (5 g of sample which digested with nitric acid (7.0 M) and finally adjusted volume to 50 ml is the maximum amount to make the digestion efficiency)

[3] EF= Ratio of slope between with and without preconcentration [106]

[4] CE= EFxf/60 [106]

3.1.2 Cr(III) and Cr(VI) speciation analysis in tap water and leachate samples

3.1.2.1 Initial conditions

For the initial conditions, the optimum conditions similarly as the previous work (as summarised in **Table 3.3**) were employed for this work. The initial conditions for Cr(III) and Cr(VI) are represented in **Table 3.7.**

Table 3.7 The initial conditions for Cr(III) and Cr(VI) determination

Parameters	**Value**
Uptake rate of organic solvent, ml min^{-1}	5
Sample volume, µl	100
Extraction time, s	30
Pause time, s	30
Rinsing time, s	20
pH of sample/standard	1
DDC concentration, %w/v	1

3.1.2.2 Effect of pH

Efficiency of extraction for Cr(VI) is affected by pH. To investigate this effect, 0.3 mg l^{-1} standard solution of Cr(VI) was prepared. Acetate buffer (0.5 M) was used to control pH. The pH was varied in the range of 0-4 by adding HCl (conc.). The results are illustrated in **Figure 3.8**. It indicates that pH has affected to Cr(VI) determination. It could not be extracted efficiently at pH approximately lower than 1.0. It is necessary to adjust pH before determination. pH 1.0 was selected for the experiment.

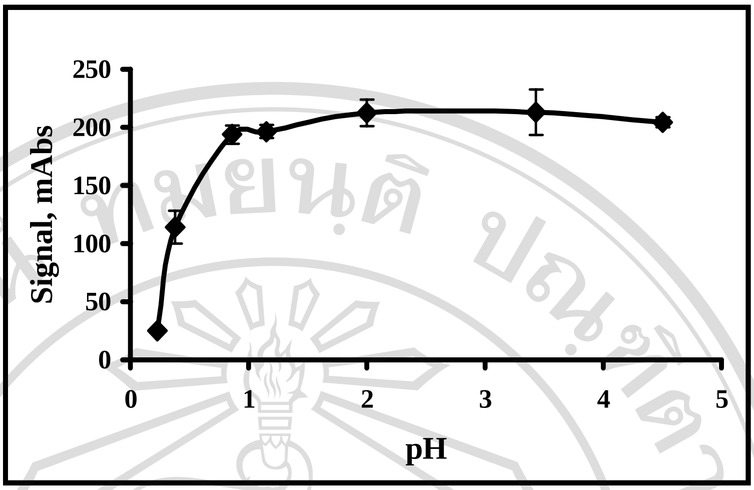

Figure 3.8 The effect of pH on signal for Cr(VI) determination

3.1.2.3 Effect of Cr(III)

Normally, high amounts of Cr(III) are presented in samples. To study the effect of Cr(III) for Cr(VI) determination, pH 1.0 was applied to study for 0.1 mg $l^{-1}$of Cr(VI). The results are presented in **Figure3.9**. It was observed that there is no significant difference on signal for 0.1 mg l^{-1} Cr(VI) if Cr(III) up to 400 mg l^{-1} presented. That means Cr(III) does not interfere for Cr(VI) determination even high concentration of Cr(III) contained in the samples.

3.1.2.4 Oxidation procedure

An aliquot of the sample solution (80 ml), 15 ml of 1% (w/v) $Na_2S_2O_8$ and 5 ml of 0.1 M of $AgNO_3$ were taken (the pH was adjusted to 1.0 by using nitric acid). Then mixed solution was heated (put in the oven at 80 °C) for approximately 15 min. Then it was cooled down before measurement. The reactions of the oxidation procedure can be described in **equation 3.2 and equation 3.3** [107].

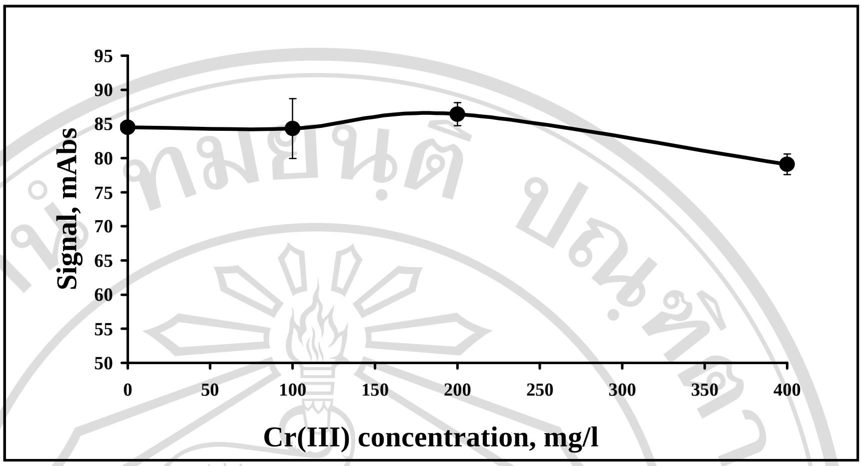

Figure 3.9 The effect of Cr(III) on signal of Cr(VI) determination

$S_2O_8^{2-} + Ag^+ = 2SO_4^{2-} + Ag^{3+}$ …………**equation 3.2**

$3Ag^{3+} + 2Cr^{3+} + 7\ H_2O = Cr_2O_7^{2-} + 3Ag^+ + 14H^+$ ……….. **equation 3.3**

3.1.2.5 Ionic strength effect

High concentration of various ions might be presented in sample, especially leachate samples. In this work, the leachate samples from incineration fly ash (a fine grey powder with very high contents of ions and some heavy metals) were studied. **Table 3.8** shows the main elements and their concentration ranges, which could be presented in the samples.

Efficiency of extraction for Cr(VI) is also affected by ionic strength. Hence, this effect was investigated. The solution containing 15 g K^+, 12 g Cl^-, 11 g Ca^{2+}, 10 g Na^+, 2 g S^{2-} and 0.5 g Zn^{2+} in 1,000 ml of DI-water was prepared (the ionic strength equal to 0.9). This solution is assumed to obtaine the highest value of ionic strength, which could be possible in these leachate samples. Other ionic strength solution was obtained by diluting this solution with DI-water. Standard solutions of Cr(VI) of 0.1 and 0.2 mg l^{-1} in various ionic strength solution were analysed. It can be seen from **Figure 3.10** that ionic strength up to 0.4 has not affected to Cr(VI) determination. It could not be extracted efficiently when ionic strength higher than 0.4.

Table 3.8 The major elements and concentration presented in leachate samples

Elements	Concentration, g l^{-1}
K	1-15
Cl	0.3-12
Ca	0.2-11
Na	0.1-10
S	0.1-2
Zn	0.003-0.5

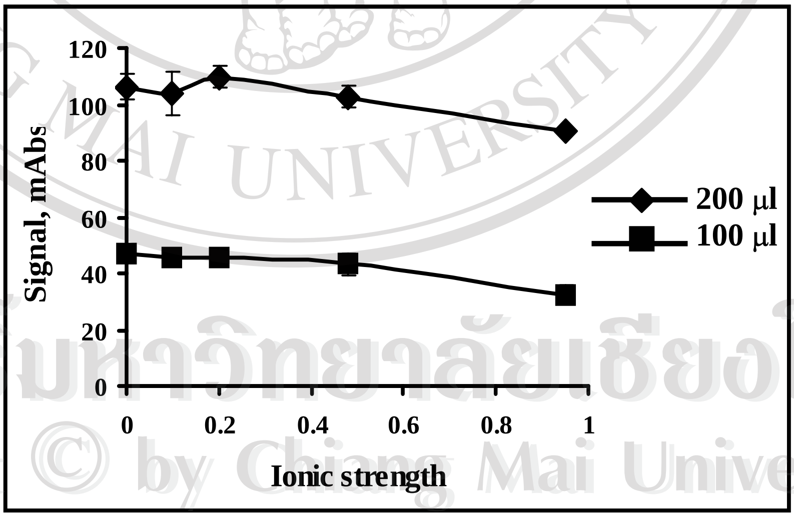

Figure 3.10 The effect of ionic strength on signal for Cr(VI) determination

3.1.2.6 Application

The method was tested by analysing several synthetic mixtures containing standard Cr(III) and Cr(VI). The recoveries obtained by spiking Cr(III) and Cr(VI) into distilled water (adjust to pH 1) are summarised in **Table 3.9**. The method was also applied to the determination of Cr(III) and Cr(VI) in tap water as shown in **Table 3.10** and leachate samples in **Table 3.11.**

Table 3.9 Determination of Cr(III) and Cr(VI) in various mixtures of standard solutions of Cr(III) and Cr(VI) in different ratios

Mixtures	**Cr(VI) added $\mu g\ l^{-1}$**	**Cr(III) added $\mu g\ l^{-1}$**	**Cr(VI) found $\mu g\ l^{-1}$**	**Cr(III) found $\mu g\ l^{-1}$**	**%Recovery**	
					Cr(VI)	**Cr(III)**
1	25	25	25	30	101	120
2	50	50	53	44	106	88
3	25	125	25	160	99	108
4	125	25	130	32	104	127
5	50	500	49	540	97	108
6	50	1,000	45	1176	89	113
7	50	2,000	46	2136	92	105

Table 3.10 Determination of Cr(III) and Cr(VI) in tap water

Cr(VI) added $\mu g\ l^{-1}$	**Cr(III) added $\mu g\ l^{-1}$**	**Cr(VI) found $\mu g\ l^{-1}$**	**Cr(III) found $\mu g\ l^{-1}$**	**%Recovery**	
				Cr(VI)	**Cr(III)**
0	0	<DL*	<DL	-	-
5	5	4.9	4.7	98	93
10	10	9.5	9.7	95	97
10	20	9.0	23.2	90	116

*** <DL=** Lower than detection limit (4 $\mu g\ l^{-1}$)

Table 3.11 Determination of Cr(III) and Cr(VI) in leachate samples

Samples	**Cr(VI) added μg l^{-1}**	**Cr(III) added μg l^{-1}**	**Cr(VI) found μg l^{-1}**	**Cr(III) found μg l^{-1}**	**%Recovery**	
					Cr(VI)	**Cr(III)**
Leachate1	0	0	67.2	99.4	-	-
	25	25	27.3	26.0	109	104
	25	50	28.1	61.9	112	124
	50	25	46.7	22.7	93	91
Leachate2	0	0	41.7	12.2	-	-
	25	25	24.2	26.4	97	105
	25	50	24.7	45.3	99	91
	50	25	41.0	29.7	82	119

3.1.2.7 Characteristics of operation procedure

The analytical characteristics for the developed method are shown in **Table 3.12.**

Table 3.12 Analytical characteristics of the proposed method for Cr speciation

Analytical characteristics	**Value**
Linear range, μg l^{-1}	5-100
Regression equation (Y= aX+b)	0.4734x-1.9875
Correlation coefficient	0.9939
Precision, %RSD, n=11 (25 μg l^{-1} and 50 μg l^{-1} Cr(VI))	6%
Detection limit[1], μg l^{-1}	4
Sample frequency (f), h^{-1}	20

[1] Detection limit defined as 3 times of blank deviation plus blank signal [105]

3.2 On-line preconcentration and preseparation by using a column extraction procedure coupled with FAAS

3.2.1 FI on-line preconcentration for lead determination

The set up is depicted in **Figure 2.2**. All optimisations were performed using a standard solution using the initial conditions as shown in **Table 3.13** by taking from previous work [18].

Table 3.13 The initial conditions for lead determination by FI-FAAS

Parameters	**Conditions**
Lead solution, M HNO_3	0.1
Eluent (EDTA), M	0.005
Eluent flow rate, ml min^{-1}	4.0
Loading time, s	15-120
Loading flow rate, ml min^{-1}	4.0

3.2.1.1 Optimisation of flow rate of eluent

The flow rate of eluent was optimised by using standard of Pb(II) 0.5 mg l^{-1}. The flow rates of eluent in the range of 1.5-5.2 ml min^{-1} were varied while the loading flow rate of standard was fixed at 4.0 ml min^{-1}. The loading time in the range of 15-90 s was applied. The single standard graph was plotted in μg of Pb (0.5-3.0 μg) against peak height at each flow rate of eluent (**μg Pb** was calculated as shown in **equation 2.1, p.40**). The results are presented in **Table 3.14**.

It indicates that higher flow rate of eluent offered higher sensitivity (referring a better slope of a single standard calibration). However, when the uptake rate was higher than 4.1 ml min^{-1}, the decrease of signal was observed. This may be due to the less efficiency of nebuliser when higher uptake rate was applied. Therefore, a flow rate of approximately 4.0 ml min^{-1} was selected for next experiment.

Table 3.14 The effect of flow rate of eluent on signal

Flow rate, ml min^{-1}	**Regression equation (Y= aX+b)**	**r^2**
1.5	0.9321x + 0.0375	1
2.6	1.0229x + 0.2191	0.9984
4.1	1.0112x + 0.1693	0.9989
5.2	0.7631x + 0.1392	0.9973
6.6	0.7802x + 0.178	0.9946

3.2.1.2 Optimisation of HNO_3 concentration as medium solution Concentration of HNO_3 was optimised. Both flow rates of eluent and standard/sample solution were fixed at 4.0 ml min^{-1}. Pb(II) standard of 0.15 mg l^{-1} was used with loading times varied from 30-300 s (this should be 0.3-3.0 μg Pb, respectively). The HNO_3 concentration was varied in the range of 0.001-2.0 M.

From **Table 3.15**, not much different in sensitivity could be observed, by considering slope of a single standard calibration, for HNO_3 concentration from 0.1-2.0 M. However the sensitivity decreased when HNO_3 concentration was lower than 0.1 M. Therefore, HNO_3 concentration of 0.5 M which gave highest sensitivity was chosen for next experiment.

Table 3.15 The effect of HNO_3 concentration on signal

Concentration of HNO_3 (M)	Regression equation (Y=aX+b)	r^2
0.001	0.5904x + 0.3358	0.9669
0.1	0.8427x + 0.1997	0.9938
0.5	0.9965x + 0.2487	0.9929
1.0	0.8891x + 0.2052	0.9942
2.0	0.8558x + 0.1693	0.9943

3.2.1.3 Single standard calibration

A single standard calibration for lead determination was possible in the range, which had been tried. The advantages of single standard calibration over normal calibration should be gained. In order to see possibility of using the single standard, the same value of **μg Pb** obtained from various loading times and concentrations of standard solution would not provide the significant difference in signal.

By using a set of conditions (as shown in **Table 3.16**), the same value of **μg Pb** obtained from various loading times and concentrations of standard Pb(II) gave no significant difference in signal. Therefore, a single standard calibration (**Figure 3.11**) should be possible in the range of 0.4-1.6 μg Pb for the solutions of 50-200 μg l^{-1} under the conditions employed. Hence, single standard can be employed instead of normal calibration.

Table 3.16 A study on a single standard calibration for lead determination

μg Pb	Set of conditions		Signal, n=3 (cm)
	Conc. of Pb(II) (μg l^{-1})	Loading time (min)	
0.4	50 100	2 1	2.1±0.2 2.2±0.3
0.6	50 150	3 1	3.4±0.3 3.1±0.2
1.2	100 150	3 2	5.7±0.4 6.2±0.1
1.6	100 200	4 2	7.0±0.5 7.5±0.2

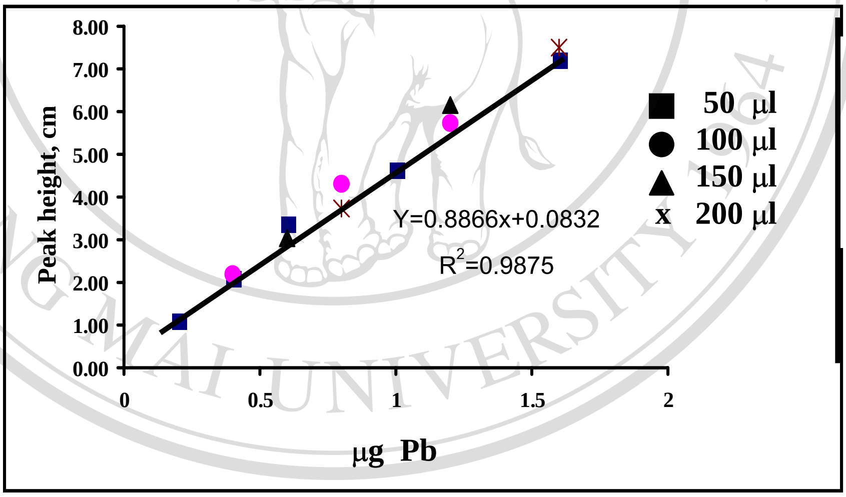

Figure 3.11 Single standard calibration for FI on-line preconcentration for lead determination

3.2.1.4 Application to water samples

The method can be utilized for lead determination in water samples from an industrial estate. The water samples were acidified with 1 M HNO_3 after collection. It was found that the results obtained by this method agreed well with those obtained by ICP-AES (t=1.09). The results are exhibited in **Table 3.17.**

Table 3.17 Determination of lead in water samples from an industrial estate (triplicate results)

Samples	Lead content found, mg l^{-1}	
	FI-FAAS	**ICP-AES***
1	0.54 ± 0.03 (±0.02)**	0.51± 0.05
2	0.72± 0.02 (±0.02)**	0.72± 0.03
3	1.62± 0.00 (±0.02)**	1.52± 0.02
4	2.52± 0.03 (±0.02)**	2.51± 0.05
5	0.19± 0.02 (±0.02)**	0.22± 0.02
6	0.11± 0.00 (±0.02)**	0.12± 0.02

* Analysis by Dr. Ponlayuth Sooksamiti, Mineral Resource Region 3 (Chiang Mai), Chiang Mai, Thailand

** ± the standard deviation taken from the 6x3 replications

3.2.2 SI- column preconcentration for iron determination

Possibility of column preconcentration using SI system for better automation was investigated. Fe(II) determination was chosen as a model for this study. Dowex-50Wx8 was used as a packing material. It is a strong acid cation resin containing 8% divinylbenzene. Its fine mesh resins are based on a microporous copolymer of styrene and that results in maximum resistance to oxidation, reduction, mechanical wear and breakage. Dowex-50Wx8 contains sulfonic acid groups that are bonded to the surface. The sulfonic acid group is strongly acidic, and attracts or exchanges cationic species in contacting. The system is depicted in **Figure 2.3** and operation procedure is described in **Table 2.2.** The conditions taken from previous work [108] were employed as in **Table 3.18.**

Table 3.18 The initial conditions for study SI-column preconcentration for iron determination

Parameters	Value
Iron solution, M HNO_3	0.1
Eluent, M HNO_3	4.0
Eluent flow rate, ml min^{-1}	4.0
Loading time, s	40
Loading flow rate, ml min^{-1}	2.4

Using the above conditions, a calibration was made by using various concentrations of Fe(II) (**Figure 3.12**).

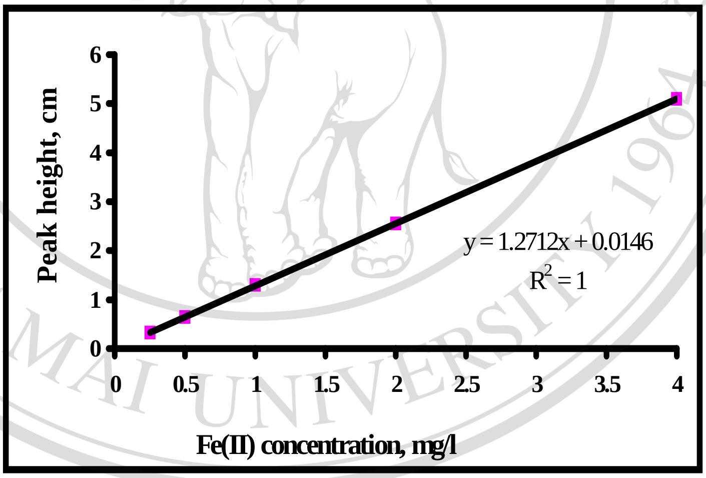

Figure 3.12 Calibration for SI- column preconcentration using Fe(II) standard solution

3.2.2.1 Carry-over effect

The carry-over effect of the system was considered. The column has to be washed with sufficient eluent volume to get rid of memory effect. To observe this effect, the volume of 4 M HNO_3 as eluent was varied from 0-3 ml. In each eluent volume, standard of 3 mg l^{-1} Fe(II) was determined for 3 times. The average signal was calculated. After that determination of blank (0.1 M HNO_3) was performed again. The % carry-over was calculated as **equation 3.1** (**p. 52**). The results are presented in **Table 3.19.**

It was observed that the proposed system without washing step showed the carry-over effect. Therefore, an extra eluent volume was required to clean the column. At least 3 ml of eluent volume must be introduced in washing step to minimise carry over effect down to less than 5%.

Table 3.19 Optimisation of eluent volume to minimised the carry-over effect

Eluent volume, ml	%Carry-over
0	31
1.5	17
3.0	5
4.5	0

As the preliminary work, it can be concluded that there is possibility to adopt SI-column preconcentration for iron determination. However, the system is time consuming because of operation procedure of SI-system, approximately 8 min for a cycle. The possible way to reduce analysis time is to reduce washing time. By using shorter column (1 cm-length), less eluent volume might be employed resulting in less washing time. It was found that this could reduce only a min of analysis time. The analytical characteristics of the developed system can be summarised in **Table 3.20**.

Table 3.20 Characteristics of performance for SI-column preconcentration for iron determination

Characteristics	**Value**
Linear range, mg l^{-1}	0.25-4.0
Analysis time, min	7.5
Sample frequency, h^{-1}	8.0
Precision, %RSD (n=4, 2 mg l^{-1} Fe(II))	3.10
Detection limit, mg l^{-1} [1]	0.19
Enrichment factor (EF) [2]	1.4
Concentration efficiency (CE) [3], min^{-1}	0.19

[1] Detection limit defined as 3 times of blank deviation plus blank signal [105]

[2] Ratio of slope between with and without preconcentration [106]

[3] CE=EF*f/60 [106]

3.3 Preliminary study for SI-ASV for arsenic speciation

The conditions for arsenic speciation preliminary study were adapted from the previous work [90], which reported a batch procedure [see **Appendix E**] as described in **Section 2.3.4**.

3.3.1 As(III) calibration

As(III) determination in the range of 25-100 μg l^{-1} was carried out. The voltammograms of As(III) determination are depicted in **Figure 3.13**.

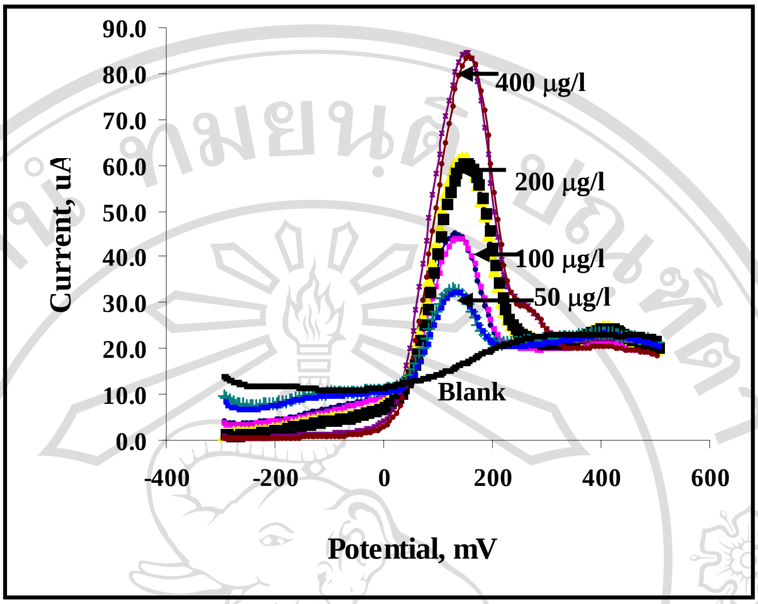

Figure 3.13 Voltammograms of As(III) determination by SI-ASV (concentrations of As(III) in the range of 50-400 μg l^{-1}, deposition time: 120 s, deposition potential: -0.3 V vs. Ag/AgCl).

3.3.2 As(V) determination

Basically, the determination of total inorganic arsenic requires a preliminary step to reduce As(V) to As(III), followed by the determination of As(III). In this work, As(V) was reduced to As(III) by off-line method as described in **Section 2.3.4.** Then total As can be measured as the same way as As(III) determination. The different amount between total As and As(III) is As(V) content.

3.3.3 Investigation of reducing agent

The reducing step is not a simple one and quite often there are problems related with the reducing agent used. In some case, the solution has to be heated to complete the conversion of As(V) into As(III) and the results are inaccurate and not precise owning to losses of volatile compounds containing As [103]. Generally, the situation is more complicated because the excess reductant interferes with the determination of As(III) and has to be removed. There are various reducing agents used for this purpose, for example, sulfur dioxide [90], titanium tri-chloride ($TiCl_3$) [109], sodium thiosulfate ($Na_2S_2O_3$) [110] and including the mixture of potassium iodide (KI) and ascorbic acid [111].

Some reducing agents, namely, $Na_2S_2O_3$, $TiCl_3$, KI and a mixture of KI and ascorbic acid were investigated for the purpose.

It was found that, $Na_2S_2O_3$ and KI cannot convert As(V) to As(III) effectively. In addition $Na_2S_2O_3$ can react with gold (from working electrode) resulting in less deposited As(III) [112]. Therefore, the conditions with and without this reducing agent for As(III) determination gave the difference in signals, peak shapes and peak positions. For $TiCl_3$, it interferes As(III) determination by producing high broad peak of the reagent over As(III) peak. Among these reducing agents, a mixture of KI and ascorbic acid should be the most suitable reducing agent with acceptable reduction performance.

3.3.4 Effect of reducing agent concentration

The effect of reducing agent concentration presented in the As(III) standard solution 100 $\mu g\ l^{-1}$ was studied. Equivalent concentration of mixture between KI and ascorbic acid was varied from 0-0.1 M. The results are shown in **Figure 3.14**.

As it was observed from **Figure 3.14,** KI-ascorbic acid reductant presented in the standard has effect to As(III) determination. A higher concentration of the reductant can cause lower signal of As(III). This may be due to the formation of a complex between gold (from working electrode) and iodine (from KI) as shown in **equation 3.4-3.8**. Therefore, the sensitivity becomes worse [113]. The concentration of 0.075 M of reducing agent was selected for conversion As(V) to As(III). Even though, this concentration can cause lower signal of As(III), the reduction efficiency is reasonable (approximately 80% conversion).

$Au + 2I^- = AuI_2^- + e^-$ ....**equation 3.4**

$I_3^- + 2e^- = 3I^-$ .... **equation 3.5**

Overall reaction: $2Au + I_3^- + I^- = 2AuI_2^-$ .... **equation 3.6**

The I_3^- in **equation 3.5** is derived from **equation 3.7**, together with **equation 3.8**:

$As^{5+} + 2e^- = I_2 + As^{3+}$ .... **equation 3.7**

$I_2 + I^- = I_3^-$ .... **equation 3.8**

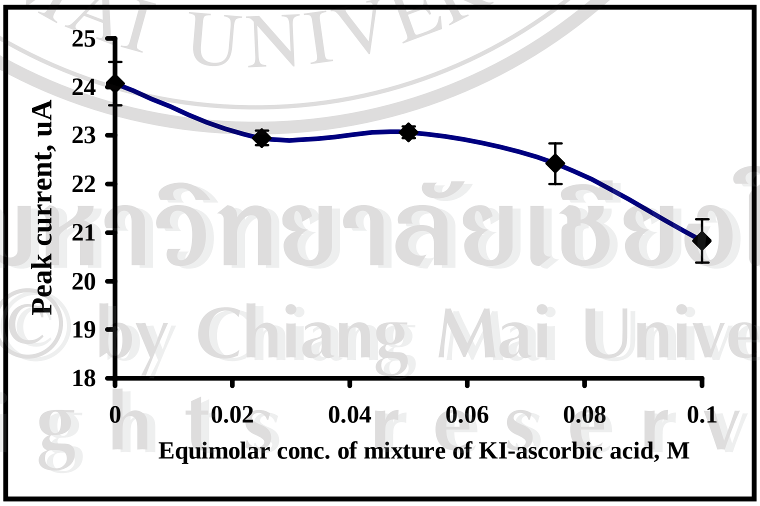

Figure 3.14 Effect of reducing agent concentration to signal of 100 μg l^{-1} As(III) determination.

3.3.5 Optimisation of reduction time

In order to convert As(V) to As(III) as much as possible, the reduction time, one of the important factor, should be optimised. As(V) standard solution of 100 μg l^{-1} was used to study. The reduction time was varied from 10-120 min.

As it can be seen from **Table 3.21**, a reduction time of 60 min or longer resulted in conversion of approximately 90%.

Table 3.21 Optimisation of reduction time

Reduction time, min	% Conversion*
10	68
30	70
60	93
90	87
120	92

* %Conversion = A*100/B
where A= signal of As(V) with reducing agent
B= signal of As(III) without reducing agent

3.3.6 Further study

Due to the time limitation for this Ph.D. study, this work could be performed only for preliminary study. The further investigation should be performed in order to complete the work. Optimisation of parameters should be planned. They would include Au plating step, the parameters, for example, plating potential, concentration of Au, platting flow rate, plating time and stability of gold film. According to deposition/stripping steps, effect of deposition potential, concentration of electrolyte (HCl), deposition flow rate and deposition time should also be investigated.