

Chapter 1

Introduction

1.1 Introduction

Inorganic arsenic is known as one of the most hazardous elements widely distributed in the earth's crust. It has received increased attention in recent years because of its high toxicity, especially its carcinogenic properties (IARC, 1987 cited in Nriagu, 1994). Arsenic contamination of drinking water has been found to be the most frequent cause. Arsenic poisoning has occurred in Thailand (Arrykul, 1996 and William *et al.*, 1996) through consumption of contaminated well water.

Determination and speciation of arsenic contamination in environmental systems is necessary, mainly because the toxicity and bioavailability of this element is species dependent. In the environment, arsenic is found in combination with other elements. One form of arsenic combined with carbon and hydrogen is referred to as organic arsenic. Another form combined with oxygen, chlorine and sulfur is called inorganic arsenic (ATSDR, 2000).

Inorganic arsenic is considered to be the most toxic form and is found in groundwater, surface water, and foods (Yamauchi, 1994 cited in Nriagu, 1994). The toxicity and mobility of arsenic is dependent on the chemical forms in which it exists, the most toxic species being arsenite As(III) and arsenate (As(V) that represent the main forms in the environment (ATSDR, 2000). Several studies have shown that inorganic arsenic can increase the risk of lung cancer, skin cancer, bladder cancer, liver cancer, kidney cancer and prostate cancer. The World Health Organization (WHO), the Department of Health and Human Services (DHHS) and the Environmental Protection Agency (EPA) have determined that inorganic arsenic is a human carcinogen (IARC, 1987 cited in Nriagu, 1994). The American Conference of Governmental Industrial Hygienists (ACGIH) classifies inorganic arsenic forms as a confirmed human carcinogen; cancer category A1. The high

potential risk makes arsenic certainly one of the most intensively studied elements in speciation analysis (WHO, 2001; ATSDR, 2000). Thus the usefulness of speciation studies can be recognized. It is necessary to develop highly sensitive methods for determination of inorganic arsenic species in different matrixes. The most popular technique for the determination of inorganic arsenic species is hydride generation (HG).

In environmental samples, this hydride generation technique has been used combined with inductively coupled plasma optical emission spectrometry (ICP-OES), since 1978 (Thompson et al., 1978a). More recently USEPA has been developing this technique and has combined it with a continuous flow system (USEPA, 1990).

In a continuous flow system, the acidified sample, blank or standard is continuously flow and mixed with a stream of reductant, usually sodium borohydride, to produce the gaseous hydride of arsenic or arsine gas. Hydrogen gas is produces as a by-product. A flow of argon is added to this mixture and the hydride is “stripped” into the gas phase. A gas/liquid separator allows the gaseous, hydride-containing phase to enter the ICP for analysis, and allows the remaining liquids to be pumped to waste. Limits of detection can generally be improved by about two orders of magnitude over simple solution nebulization using hydride generation. The hydride generator is constructed of a simple and inexpensive continuous flow hydride generator for ICP-OES analysis (Davidowski, 1993).

Hydride generation combined with inductively coupled plasma optical emission spectrometry (HG-ICP-OES) has been conducted in this study with the aim to develop a method for determination of inorganic As species such as total As, As(III) and As(V). The experimental conditions of HG-ICP-OES system were optimized. Finally, this developed method was applied to the quantitative analysis of total As, As(III) and As(V) concentration in drinking water samples.

1.2 Literature review

1.2.1 Chemistry of arsenic (As)

Arsenic (As) is the third member in Group VA of the periodic table, and its atomic number and atomic mass are 33 and 74.9216 respectively. It has four oxidation state in nature: -3, 0, +3 and +5. The oxidation number +3 and +5 of arsenic species is commonly present in a variety of complex minerals and in dissolved salts in natural waters. Arsenic compounds in nature can be released to the environment by redox reduction processes (ATSDR, 2000).

Table 1 Common name, chemical structure, chemical formula, synonym and trade name of the arsenic, arsenite and arsenate species

Characteristic	Arsenic	Arsenite (As ⁺³)	Arsenate (As ⁺⁵)
Chemical structure	As	$\begin{array}{c} \text{HO—As—OH} \\ \\ \text{OH} \end{array}$	$\begin{array}{c} \text{O} \\ \\ \text{HO—As—OH} \\ \\ \text{OH} \end{array}$
Chemical formula	As	H ₃ AsO ₃	H ₃ AsO ₄
Synonym(s)	Arsenic black; colloidal arsenic; gray arsenic	Arsenious acid; arsenic oxide arsenious oxide; white arsenic;	Arsenic acid; orthoarsenic acid
Registered trade name	No data	Arsenolite®; Claudelite®	Dessican L-10®; Scorch®

(Source: ATSDR, 2000)

Arsenic trioxide (As_2O_3 or As_4O_6) is one of the primary intermediates used to make other forms of arsenic. Dissolved in water, it forms arsenious acid although the pure acid has not been isolated. Salts are called arsenites, and these forms represent the +3 oxidation state of arsenic, As(III). Arsenates (AsO_4^{3-}) are salts of arsenic acid, or more formerly orthoarsenic acid. This represents the +5 oxidation state of arsenic. Arsenite can be easily oxidized to arsenate, and arsenate can also be reduced to arsenite (ATSDR, 2000). The common name, chemical structure, chemical formula, synonym and trade name of the arsenic, and inorganic arsenic species of arsenite and arsenate species are shown in Table 1.

1.2.1.1 Physical and chemical properties

The physical and chemical properties of arsenic, arsenite and arsenate shown in Table 2

Table 2 Physical and chemical properties of arsenic, arsenite and arsenate species

Property	Arsenic	Arsenite (As^{+3})	Arsenate (As^{+5})
Molecular weight	74.92	131.92	150.95
Color	Gray	White	White
Odor	Odorless	Odorless	-
Physical state	Solid	Solid	Solid
Melting point	817°C at 28 atm	312.3°C	35.5°C
Boiling point	613°C at sublimes	465.5°C	160°C
Density d20	5.727 g/cm ³	3.738 g/cm ³	2.0-2.5 g/cm ³
Water(20°C)	Insoluble	37 g/L	3,020 g/L
Acid	Soluble in HNO_3	Soluble in HCl	-
Alcohol	-	Slightly soluble	Soluble
Vapor pressure	1 mm Hg (373°C)	66.1 mm Hg (312°C)	-
Valence states	0	+3	+5

(Source: ATSDR, 2000)

1.2.1.2 Application

The arsenic in used is estimated to in compound form, primarily of arsenate or arsenic trioxide. The major use for inorganic arsenic is in wood preservation. Inorganic arsenic compounds were used in medicine in the 1970s, primarily for treatment of leukemia, psoriasis, and asthma. It has been estimated that 70% of the world arsenic production is used in timber treatment as copper chrome arsenate, 20% in agricultural chemicals as arsenic-containing pesticides, and the remainder in glass, pharmaceuticals and non-ferrous alloys (USNRC, 1999).

1.2.1.3 Source and Potential Exposure

Inorganic arsenic is released to the environment from natural sources such as wind-blown dirt and volcanoes, release from industrial sources far exceed those from natural sources. Industrial sources of arsenic include nonferrous metal mining and smelting, pesticide application, coal combustion, wood combustion, and waste incineration. Thus, humans may be exposed to arsenic by eating food, drinking water, or breathing air. However, levels of inorganic arsenic, the form of most concern, are low. Levels of arsenic in various locations, weather conditions, industrial activity and urban air generally range from less than 1 to about 2,000 ng/m³ and 20 to 30 ng/m³ respectively. Of these, food is usually the largest source of arsenic. Fish and seafood contain the greatest amounts of arsenic, but this is mostly the organic form of arsenic that is less harmful. The level of inorganic arsenic intake from these sources is generally about 3.5 µg/day (ATSDR, 2000).

1.2.1.4 Health effects

Short-term (acute) poisoning with inorganic arsenic has been recognized since ancient times, and large oral dose (above 60,000 µg/L in food or water) can produce death, and swallowing lower levels (ranging from about 300 to 30,000 µg/L in food and water) may case stomach ache, nausea, vomiting and diarrhea. Other effects include decreased production of red and white blood cells which may cause fatigue, abnormal heart rhythm, blood-vessel damage resulting in

bruising, and impaired nerve function causing a “pins and needles” sensation in hands and feet.

Long-term (chronic) oral exposure to inorganic arsenic causes a pattern of skin changes. These include a darkening of the skin and the appearance of small “corn” or “warts” on the palms, soles, and torso. A small number of the corns may ultimately develop into skin cancer. Swallowing arsenic has been reported to increase the risk of cancer in the liver, bladder, kidneys, prostate and lungs. Several reported have classified inorganic arsenic as a known human carcinogen (ATSDR, 2000).

1.2.1.5 Maximum Contaminant Level (MCL) and Guidelines

The WHO guideline for arsenic in drinking water was provisionally reduced in 1993 from 50 µg/L to 10 µg/L. The new recommended value is based largely on analytical capability. If the standard basis for risk assessment applied to industrial chemicals were applied to arsenic, the maximum permissible concentration would be lower still. The USEPA limit was also reduced from 50 µg/L to 10 µg/L in January 2001 following prolonged debate over the most appropriate limit. The European Community’s maximum admissible concentration (MAC) for As in drinking water is now also reduced to 10 µg/L. The Japanese limit for drinking water is 10 µg/L, and the interim maximum acceptable concentration for Canadian drinking water is 25 µg/L (Smedly and Kinniburgh, 2001).

1.2.1.6 Arsenic contamination in Thailand

In the September 1987, human health problems in Ron Phibun District (Figure 1) were exposed to the public when the first serious case of keratosis and hyperpigmentation was diagnosed on residents who suffered from arsenical skin cancer. A clinical survey during 1987-1988 the showed that more than 1000 people between the age 4 months and 85 years were affected (Chooprapawan, 1995). In 1992 a joint Thai-Japan clinical survey of blood taken from students was studied. Over 85% of students had high levels of arsenic in their blood, with a 22% incidence of skin lesions and hyperkeratosis (Paijitprapapon and Ramnarong, 1994). Many residents in the mining area suffered from the same problem; skin lesions and

hyperkeratosis that was related to consumption of contaminated surface and groundwater. Milintawisamai *et al.*, (1997 ; JICA, 2000) found arsenic at concentrations of up to 100 times more than is recommended by the World Health Organization (WHO) for potable water: 10 µg/L.

Analytical methods are of critical importance because the inorganic arsenic species occur at very low concentrations in the environment. The aim of the analytical methods that are available for detecting, measuring, and/or monitoring inorganic arsenic, its metabolites, and other biomarkers. So that we can define more clearly exposure and effects.

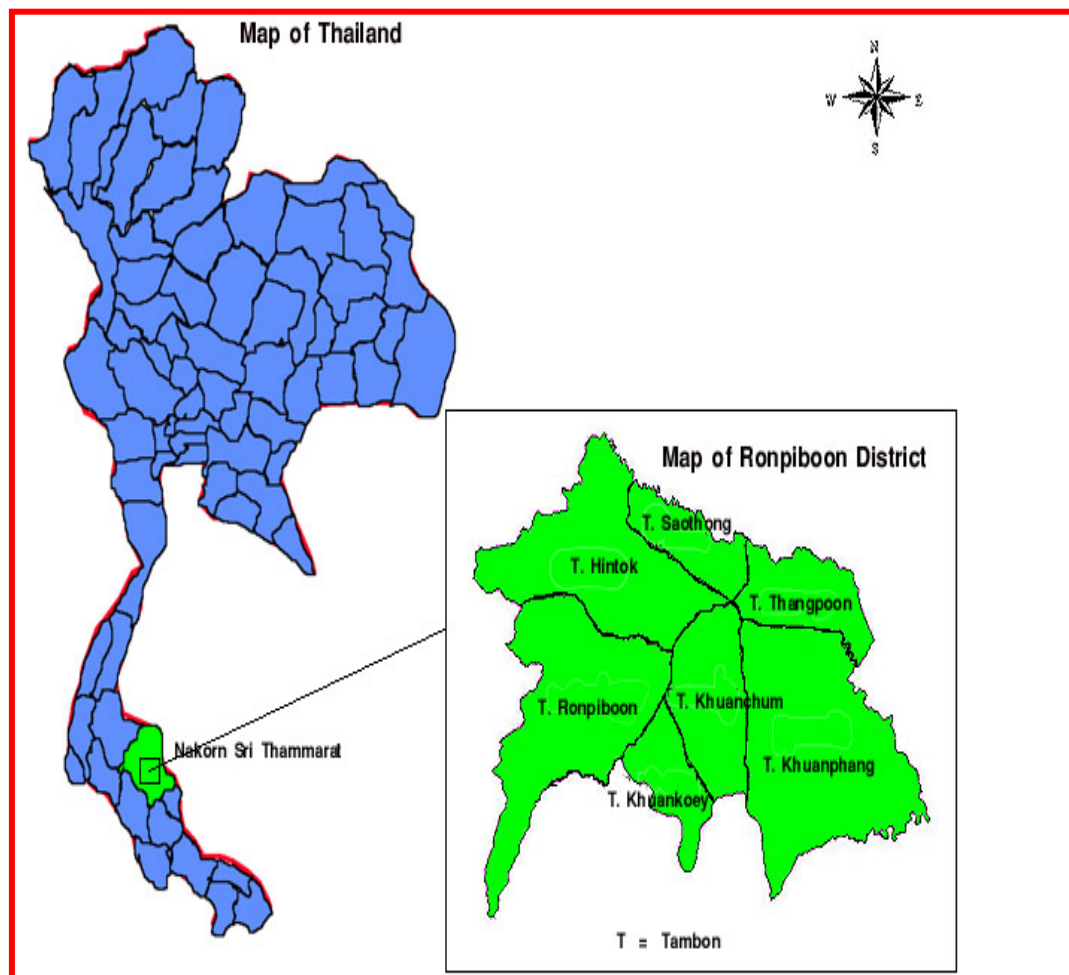


Figure 1 Map of Ron Phibun District, Showing the arsenic contamination
(Source: Wiliam et al., 1996)

1.2.2 Inductively coupled plasma-optical emission spectrometry (ICP-OES)

An ICP source consists of a flowing stream of argon gas ionized by an applied radio frequency field typically oscillating at 40 MHz. This field is inductively coupled to the ionized gas by a water-cooled coil surrounding a quartz “torch” that supports and confines the plasma. A aerosol sample is generated in an appropriate nebulizer and spray chamber and is carried into the plasma through an injector tube located within the torch. The aerosol sample is injected directly into the ICP, subjecting the constituent atoms to temperatures of about 6000 to 8000 °K. Because this results in almost complete dissociation of molecules, significant reduction in chemical interferences is achieved. The high temperature of the plasma excites atoms producing an ionic emission spectra. The ICP provides an optically “thin” source that is not subject to self-absorption except at very high concentrations. Thus linear dynamic ranges of four to six orders of magnitude are observed for many elements (B. Boss and J. Fredeen, 1997).

The widely used analytical method for the determination of trace elements, ICP-OES marked its fortieth anniversary in 2004. This method is based on OES coupled with an ICP source. The ICP source, as described by Green field *et al.* and Wendt and Fessel (Greenfield *et al.*, 1964 Fessel, 1969) has a range of attractive properties for analytical optical emission spectrometry. Many advantages including low detection limits, good precision, fast sample throughput and short integration time, a wide linear calibration, negligible interference effects resulting from condensed-phase chemical reactions in the high-temperature discharge.

During inductively coupled plasma-optical atomic emission spectrometry, the sample is usually transported into the instrument as a stream of liquid sample. Inside the instrument, the liquid is converted into an aerosol through a process known as nebulization. The aerosol sample is then transported to the plasma where it is desolvated, vaporized, atomized, and excited and or ionized by plasma. The excited atoms and ions emit their characteristic radiation which is collected by a device that converts this data into concentration information for the analyst. A representation of the layout of a typical ICP-OES instrument is shown in Figure 2.

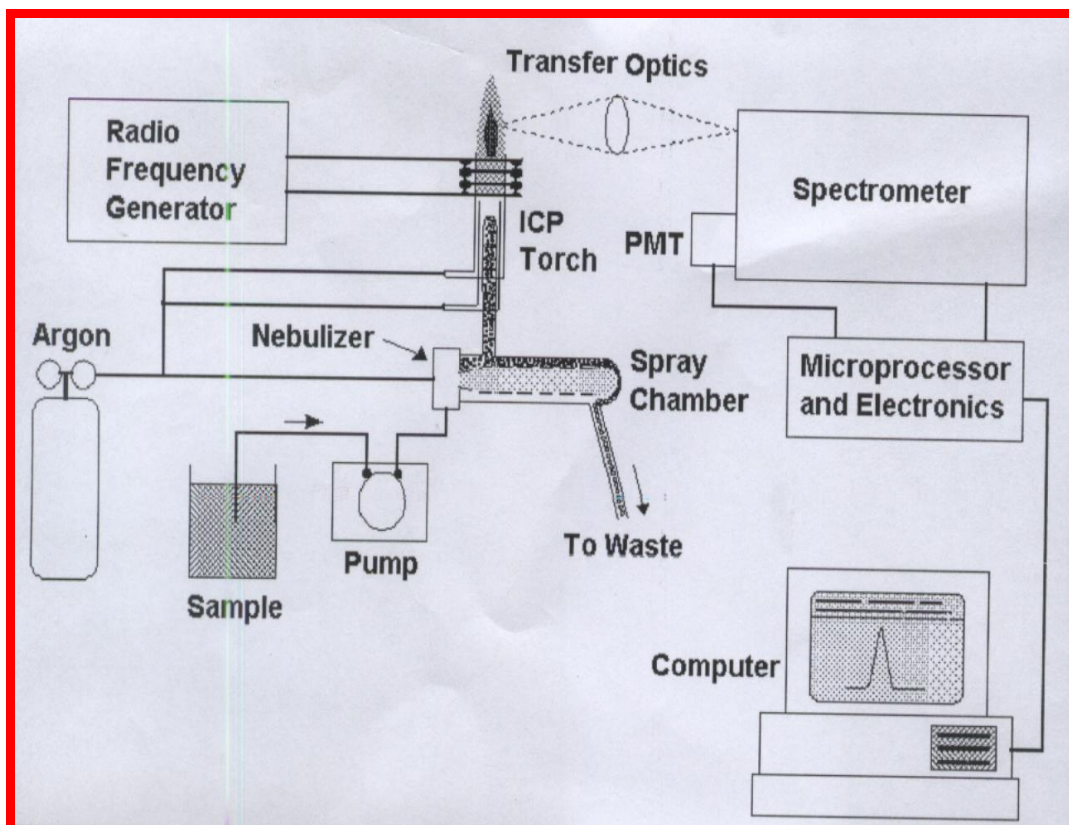


Figure 2 Major components and layout of a typical ICP-OES instrument
(Source: Boss *et al.*, 1997)

The detection limits using ICP-OES with conventional aqueous-solution pneumatic nebulization techniques are not enough for inorganic arsenic analysis and moreover, have troublesome spectra interferences from the real samples. Then, the most widely used alternative sample introduction technique is hydride generation into the plasma. This increases the sensitivity considerably increased, and in addition achieves a useful separation from the sample matrix.

1.2.3 Continuous flow hydride generation method (HG)

The used of hydride generation for ICP-OES has been widely reported (Ding and Sturgeon, 1997; Muller, 1999; Do *et al.*, 2000; and Gettar *et al.*, 2000).

The continuous flow hydride generator, introduced recently, offers the advantages of simplicity in operation, excellent reproducibility, low detection limits, and high sample volume throughput for inorganic arsenic analysis. This method is applicable to determination of arsenic by conversion to arsine gas by sodium borohydride reduction and transported into a plasma of ICP-OES for analysis (Figure 3).

Arsenite, As(III) oxidation state is instantaneously converted by sodium borohydride in acid solution to arsine. The arsine is purged continuously by argon into a plasma of ICP-OES and converted to gas-phase atom. Arsenate, As(V) oxidation state of arsenic is reduced relatively slowly by sodium borohydride to As(III), which is then instantaneously converted to arsine. The arsine atoms emission peaks commonly are decreased by one-fourth to one-third for As(V) when compared to As(III). Determination of total arsenic requires that all inorganic arsenic compounds be in the As(III) state. Organic and inorganic forms of arsenic are first oxidized to As(V) by acid digestion. The As(V) then is quantitatively reduced to As(III) with sodium or potassium iodide before reaction with sodium borohydride.

The hydride generation technique is utilized for separation of the analyte arsenic from the matrix by conversion to its volatile hydride called arsine (AsH_3). This technique offers a route to the trace analysis of several important arsenic species that have specific problems when analyzed by conventional methods. Conventional methods of ICP-AES for arsenic determination is poor due to problems

associated with concentration by the sample matrix. HG-ICP-OES is an alternative method for arsenic analysis because of its sensitivity. (Tian *et al.*, 1998).

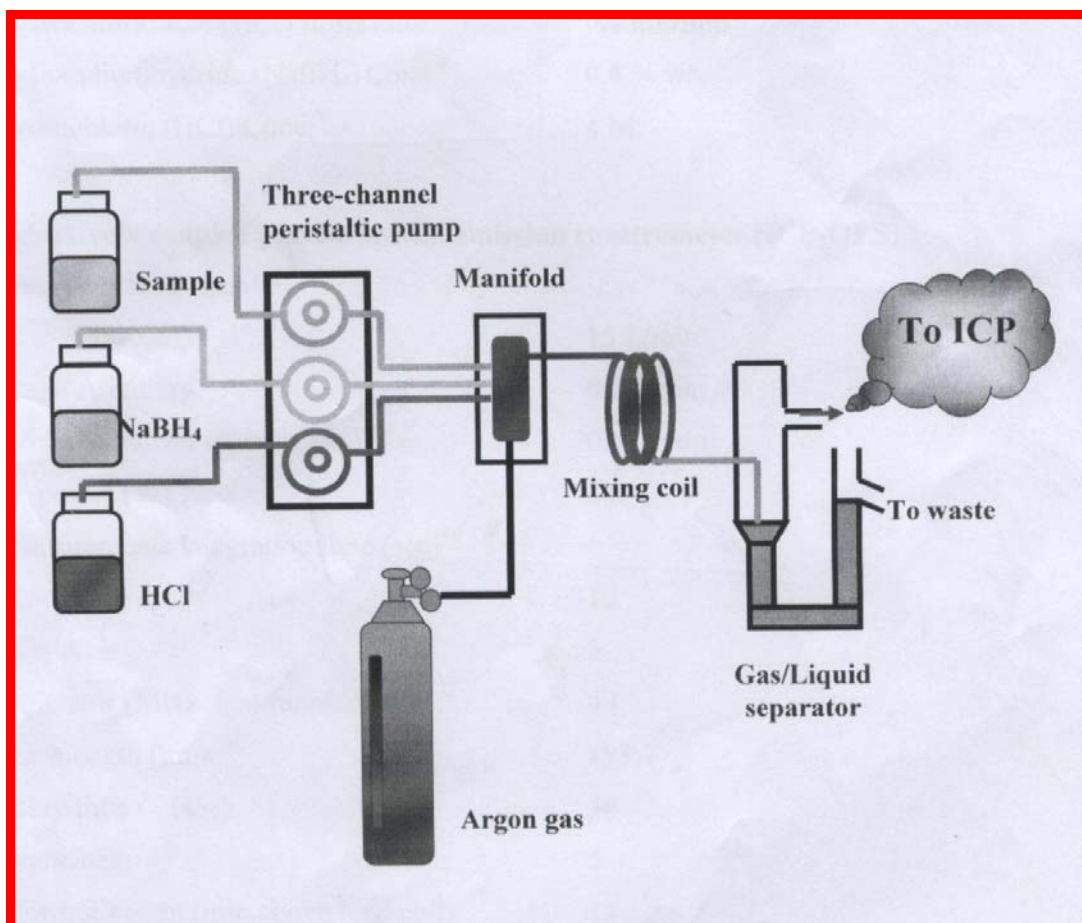


Figure 3 Schematic design and flow path of hydride generation system coupled online to ICP-OES

(Source: Davidowski, 1993)

1.2.4 Determination of inorganic arsenic

In 2000, Gettar *et al.* reported the determination of inorganic arsenic and organic arsenic species in water by ion chromatography separation, coupled online to post-column generation of the gaseous hydrides by reaction with NaBH₄ in an acid medium. Detection and measurement were performed by ICP spectrometry operating in the atomic emission mode. Arsenic emission was monitoring at 193.7 nm. Linear calibration curves were obtained in the 0.05-2 $\mu\text{g mL}^{-1}$ range of As(III)

and As(V). Results of the analyses of natural samples, such as river and ground water spiked with the studies, indicated that the analyte recoveries might be dependent on the sample composition. In addition, Muller *et al.*, (2000) develop method for determination of inorganic arsenic(III) in ground water using hydride generation coupled to ICP-AES (HG-ICP-AES) under variable sodium borohydride (NaBH_4) concentrations. The As(III) species shows significantly higher signal intensities at low NaBH_4 concentrations than the As(V) species. The NaBH_4 concentration used for the determination of As(III) cause very little considerable interference of As(V). The interference of As(V) during the As(III) measurements were very small, the interference were smaller than 2%. An amount of As(III) higher than 10% of the total arsenic amount could be determined. The linearity of calibration reaches from $2 \mu\text{g L}^{-1}$ up to $1000 \mu\text{g L}^{-1}$ with the detection limit routinely of about $1 \mu\text{g L}^{-1}$ for each species. The advantages of the linear calibration range and a higher sensitivity are major advantages of this method. Additional merits of the developed method are easy handling and high sampling rates.

On the other hand, Barra *et al.*, (2000) developed atomic fluorescence method the for determination of inorganic arsenic in soils after microwave-assisted distillation. After reduction of As(V) to its As(III) with potassium iodide, inorganic arsenic was distilled as AsCl_3 and this was finally determined by atomic fluorescence spectrometry after hydride generation with NaBH_4 in HCl medium. The methodology developed has a detection limit of $0.015 \mu\text{g L}^{-1}$, which corresponds to a concentration of $0.006 \mu\text{g g}^{-1}$, and a relative standard deviation of 3% at $8.7 \mu\text{g g}^{-1}$ of arsenic. The recovery percentages of As(III) and As(V) were 103 ± 4 and $106 \pm 4\%$, respectively. Addition, Shraim *et al.* (2000) develop by used perchloric acid in the reduction medium for speciation of arsenic by hydride generation atomic absorption spectrometry. In these methods perchloric acid as a reduction medium, L-cysteine was used as a pre-reducing agent for a certain contact time between its addition and analysis, and NaBH_4 was used as a reducing agent. The methods developed for the determination of inorganic arsenic species in environmental water samples were as follows. As(III): 5 M acid and 0.08% NaBH_4 in the absence of L-cysteine. Total inorganic arsenic [As(III)+As(V)]: 8 M acid and 0.6% NaBH_4 . As(V): by difference. Detection limits of added As spikes for all analyses were found to be between 0.5-0.7

$\mu\text{g L}^{-1}$ and with recoveries of 90-112%. In 2000, Do *et al.* developed the HG-ICP-AES for determination of arsenic in the eluate from a high-performance liquid chromatography (HPLC) system. Arsenite; As(III) and arsenate; As(V) present in urine samples of patients treated intravenously with As(III), were analyzed separately by HPLC-HG-ICP-AES using a non-polar C_{18} column. This analytical method allowed the sensitivity determination of the arsenic species in the submicrogram per liter range. The measured concentrations obtained were $385 \mu\text{g mL}^{-1}$. 1M HCl with 1% NaBH_4 was used in the HG system. The signal responses of arsenic with HG were at least ten time more intense compound to the ones obtained without HG.

Then, the develop method for determined of As(III) and total inorganic arsenic in water samples by using flow injection hydride generation atomic absorption spectrometry (FI-HG-AAS), by without pre-reduction of As(V). The flow system was operated in the merging zones configuration, where sample and NaBH_4 are simultaneously injected into two carrier streams. HCl and H_2O , respectively. The sample and reagent injected volumes were $250 \mu\text{L}$ with a flow rate of 3.6 mL min^{-1} for hydrochloric acid and de-ionised water. When the NaBH_4 concentration was maintained at 0.1%, it was possible to perform arsine selective generation from As(III) and on-line arsine generation with 3.0% NaBH_4 to obtain total arsenic concentration. As(V) was calculated as the difference between total As and As(III). With the proposed procedure detection limits of 0.3 ng mL^{-1} for As(III) and 0.5 ng mL^{-1} for As(V). were achieved. The relative standard deviations were 2.3% for 0.1 ng mL^{-1} As(III) and 2% for 0.1 ng mL^{-1} for As(V). The method was shown to be satisfactory for determination of trace arsenic in water samples. (Wrobel *et al.* 2002).

Moreover, inorganic and organic arsenic speciation have been detected in fish tissue using coupled high performance liquid chromatography-microwave digestion-hydride generation-atomic absorption spectrometry. Conditions for hydride generation were 3.5 M HCl, 1% NaBH_4 , 90 mL min^{-1} was chosen for the carrier gas. Detection limits achieved were 0.3 and 1.1 ng with 95% recovery for all species (Villa-Lojo *et al.*, 2002).

Ferreira *et al.* (2002) develop method for determination of As(III) and arsenic(V) in natural waters by cathodic stripping voltammetry at a hanging mercury drop electrode. For determination of As(III) the quantification limit was $0.2 \mu\text{g L}^{-1}$

for a deposition time of 40s, and the relative standard deviation (RSD) was calculated to be 6% (n=13) for a solution with $8 \mu\text{g L}^{-1}$ of As(III). For As (total), the quantification limit was $2 \mu\text{g L}^{-1}$ for a deposition time of 3 min, and the RSD was calculated to be 3% (n=10) for a solution with $8 \mu\text{g L}^{-1}$ of As(V). The method was validated by application of recovery and duplicate tests in measuring of As(III) and As (total) in natural spring and mineral waters. For As (total), the results from the literature were compared with the results obtained by optical emission spectrometry with ICP coupled to hydride generation (OES-ICP-HG) and good correlation was observed.

In addition, Goessler and Pavkov (2003) developed anion-and cation-exchange chromatography to separate arsenic compounds and determined them with inductively coupled plasma mass spectrometer as an arsenic-specific detector. As(III) was completely oxidized to As(V) at $100 \text{ }^\circ\text{C}$. and other species required $>280 \text{ }^\circ\text{C}$. Nevertheless, accurate results are obtained with the hydride generation technique of incompletely mineralized samples when an appropriate calibration is performed.

1.3 Objectives

1.3.1 To modify and optimize methods for determination of inorganic arsenic species by using an online HG-ICP-OES technique.

1.3.2 To apply this method to real sample analysis such as drinking water samples.