# CHAPTER 1 INTRODUCTION

#### 1.1 Overview

Mercury (Hg) is recognized as one of the most toxic metals. It is a nonessential metal (Mason and Jenkins, 1995). Traces of mercury are ubiquitous in soils, waters, sediments, organisms and air (Jonasson and Boyle, 1972). It occurs naturally as inorganic salts (i.e. mercuric chloride) and organic mercury. Mercury is persistent in the environment and can be biomagnified via food chains. It enters the environment either from natural or from anthropogenic origins. Elemental form of mercury is easy to volatile subsequently transport in air. It returns to the earth's surface both as wet and dry deposition (Jackson, 1991). Coal, which burned for heat or for fuel in power plants. It is known as an important source of mercury released 12,580 million worldwide during the 2040 tons year (http://www.ornl.gov/info/ornlreview/rev26-34/text/colmain.html). Other industrial sources that add mercury to the environment are paper mills (used as a slimicide), plating industries, milling and metal alloy production, chlorine and caustic soda production, fungicidal paints, preservatives, batteries, switches and relays, thermometers, barometers, medicinal uses (such as for blood pressure pills), agricultural uses (insecticides, fungicides, and in seed dressings) and dental treatments (Peterle, 1991). In 2004, the US Environmental Protection Agency estimated that dental clinics use 34 tons of mercury annually, which equals to 14% of the total annual mercury consumption in the United States of America (www.mercurypolicy.org/new/documents/whatpatientsdontknow.pdf).

The relative hazard index of mercury, calculated based on the ratio of human-made to natural occurrences of the toxic metal and the relative toxicity, is 40 to 1600. This is much higher than cadmium, lead and arsenic which are 13, 7 and 0.7, respectively (Harris and Hohenemser, 1978). Mercury has long half-life in organisms. Its half-life is about 70 days in human, 3.7 days in mice, 15 to 70 days in rat, and 20 days in dog (Berry *et al.*, 1974). A person can be exposed to mercury from

having a number of amalgams filling teeth, having skin contact with mercury, breathing in contaminated air, eating contaminated water, fishes or foods. Mercury can be excreted from the body via urine, saliva, feces and exhaled breath (Schroeder, 1995).

Silver amalgam has been used as a tooth filling material as early as the 7<sup>th</sup> century in China and since the 18<sup>th</sup> century in Europe and North America, but the composition is different from that being used today. It is a mixture of 50% of metallic mercury, 35% silver, 9% tin, 6% copper, and trace amounts of zinc (Engqvist, 1998). Dentists were concerned about mercury poisoning. This indicated by a declining of amalgam using since the 1985s, changeover from elemental mercury to prepackage dental amalgam capsules, and an increasing of a use of non-mercury filling (i.e., composite resin, glass). However, the composite resin and ceramic properties are not as good as amalgam because they are breakable and expensive. Therefor, some dentists still use amalgam as the tooth filling. This is indicated by the amount of mercury in dental purpose is increased about 40% (32 metric tons in 1995 to 44 metric tons in 2001) (www.mercurypolicy.org/document/Dentist the menace).

The over exposure of mercury will affect on human nervous system and other body systems resulted in dementia and loss of motor coordination. Mercury, which entered to the body, was collected in blood, hair, urine, teeth, nail, kidney, liver and spleen biopsy (Hac *et al.*, 2000). Hair sample is the most used biological material for monitoring of mercury exposure because it is easy to acquire and easy to store (Babi *et al.*, 2000; Morton *et al.*, 2002; Lindow *et al.*, 2003). Moreover, mercury in hair is stable for a long period of time (Holsbeek *et al.*, 1996). In this study, mercury concentration in hair samples from dental personnel of Prince of Songkla University and from Hat-Yai (Songkhla Province) residence were determined using a cold-vapor Atomic Absorption Spectrophotometric technique after hot acid digestion. The correlation of mercury content in hair between exposed persons and non-exposed persons were investigated.

# **1.2** Literature review

The biological meterials are useful for determined of mercury such as blood, urine, tissue, liver, nail and hair (Engqvist, 1998; Hac *et al.*, 2000). Hair is a suitable indicator for the monitoring of human exposure to mercury that reflects organ mercury levels as well as dietary intake. It showed a convincing relationship between the content of mercury in hair versus its content in blood. Methylmercury is incorporated into scalp hair at the hair follicle in proportion to its content in blood. The hair-to-blood ratio in human has been estimated as approximately 250:1 expressed as micrograme mercury per grame of hair to micrograme mercury to liters of blood (Diez and Boyona, 2002).

Ages, gender, weight, consumption of fish, amalgams filling are influence to determination of mercury. Mercury level in hair of dental personnel is influence of year of practice, frequency of using goggle, frequency of using gloves, frequency of using mask, self prepared of amalgam, number of patient, ventilation of work placed. Age and weight are not significant for mercury concentration in human hair (Babi *et al.*, 2000). The number of amalgam filling that dental personnel have and time of filling in the mouth found that not linear correlation with mercury concentration. These is the half life of mercury in organism is very shot whem compared with year of amalgam filling in the mouth (Babi *et al.*, 2000; Harakeh *et al.*, 2002).

Consumption to fish is factor of mercury accumulation in the body. The major portion of mercury in the diet is contained in fish in the form of methylmercury and the minor portion is in the form of inorganic mercury (Suzuki *et al.*, 1993; Holsbeek *et al.*, 1996). Mercury levels are varied due to kinds of fish; meckarel is the highest whereas gemfish is the lowest) (Gardner *et al.*, 1979). People who live in different islands in South East Asia had different mercury concentration. Ways of life is the reason of places where they live, religion, family and consumption are significant (Foo and Tan, 1998).

Harakeh *et al.* (2002) reported that the mercury concentration in Labanon dental personnel was in the range of 0.00-24.16  $\mu$ g g<sup>-1</sup>. These reported that, the number of practice, year of practice, fish consumption were significant of mercury

concentration in hair. In Thailand, study few researches reported mercury accumulation in dental personnel. The mercury level in dental personnel hair in Bangkok was in the range of 2.8-10.1  $\mu$ g g<sup>-1</sup> (Saengsirinawin and Pringsulaka, 1988). Intarasawat *et al.* (1992) reported that the average of mercury concentration in blood of dentists, dental assistants, 2-year dental student were 1.24, 1.09, 0.97  $\mu$ g per 100 mL of blood, respectively. It is shown that the mercury levels in all groups have different significant. The dental student who contact with amalgam filling when they in 3, 4, 5 and 6 years. In addition, the different year they used different amount of amalgam, for 3-year student they used less than another year. In 6-year student, they worked in the hospital so maybe they contact with amalgam less than 4 and 5 years.

# **1.3** Physical and chemical properties of mercury

Mercury (Hg) occurs naturally in the earth's crust (average abundance  $500 \ \mu g \ kg^{-1}$ ). It is found in very small amounts in ocean, rock and soil. Its chemical symbol is Hg, which derived from *Hydrargyrus* in Latin, which mean liquid silver. Natural mercury is a mixture of seven isotopes: <sup>196</sup>Hg (0.146%), <sup>198</sup>Hg (10.02%), <sup>199</sup>Hg (16.84%), <sup>200</sup>Hg (23.13%), <sup>201</sup>Hg (13.22%), <sup>202</sup>Hg (29.80%) and <sup>204</sup>Hg (6.85%) (Schroeder, 1995). Mercury is also known as quick silver because it is a silver-colored liquid at a room temperature. It is a quite inert chemical, having higher ionization potential than other electropositive elements except hydrogen. It has oxidation states of zero (Hg<sup>0</sup>), one (Hg<sub>2</sub><sup>2+</sup>) and two (Hg<sup>2+</sup>) (Jackson, 1991).

Mercury is classified as class B character. It tends to form covalent bond and showed the ligand-binding preferences for the following anions decrease in the order S>N>P>O. Mercury occurs in several forms, including metallic mercury, inorganic mercury and organic mercury. Metallic mercury is an elemental form of mercury, it evaporates at room temperature. Mercury vapor is colorless and odorless. Physical and chemical properties of elemental mercury are summarized in Table 1-1.

| Atomic number            | 80  |
|--------------------------|---|
| Electron configuration   | $[Xe]5d^{10}6s^2$   |
| Atomic mass              | 200.59  |
| Atomic radius            | 0.150 nm  |
| Atomic volume            | 14.81 $\text{cm}^3/\text{g-atom}$                         |
| Melting point            | -38.87°C  |
| Boiling point            | 357.25°C  |
| Density                  | $13.546 \text{ g cm}^{-3} \text{ at } 20^{\circ}\text{C}$ |
|                          | $13.959 \text{ g cm}^{-3} \text{ at } 0^{\circ}\text{C}$  |
| Ionization potentials    | -   |
| 1 <sup>st</sup> electron | 10.437 eV   |
| 2 <sup>nd</sup> electron | 18.756 eV   |
| 3 <sup>rd</sup> electron | 34.2 eV   |
| Resistivity              | $95.8 \times 10^{-6}$ ohm cm <sup>-1</sup> at 20°C        |
| Vapor pressure           | 0.246 Pa at 25°C ( $1.85 \times 10^{-3}$ Torr)            |
| Heat of vaporization     | 14.67 cal/g-atom at 25°C                                  |

**Table 1-1** Selected physical and chemical properties of elemental mercury

Source: Schroeder, 1995

Inorganic mercury compounds or so-called mercury salt occur when mercury combines with elements such as sulfur or oxygen. Most inorganic mercury compounds are white powders or crystals, except mercuric sulfide (also known as cinnabar), which is red and turns black after exposure to light.

Mercury can form highly covalent bond with carbon. Organometallic Hg(II) compounds are resistant to oxidation and hydrolysis and are quite stable kinetically (though not thermodynamically) in water (Cotton and Wilkinson, 1988). There are a potentially large number of organic mercury compounds for example phenylmercury acetate or phenylmercury hydroxide, which were used as antiseptic, fungicide and germicide. The naturally occurring organometallic Hg(II) species are methyl mercury ( $CH_3Hg^+$ ) and dimethylmercury (( $CH_3$ )<sub>2</sub>Hg). The  $CH_3Hg^+$  is usually associated with anionic ligands. Both  $CH_3Hg^+$  and  $(CH_3)_2Hg$  are synthesized mostly by microbial methylation of bioavailable inorganic Hg(II) species, a reaction mediated by many different species and strains of free-living bacteria and fungi (methylating microbes, or methylators), ranging from anaerobes to aerobes, under a wide range of environmental condition (Matilainen and Verta. 1995). Dimethylmercury is now only used for some specific chemical tests and therefore any occurrence of it as a direct human source is very unusual. As part of the mercury cycle, most dimethylmercury in the environment has been transformed from inorganic

mercury by microorganisms (in soil and sediments, in air or water), most likely under alkaline pH condition. Dimethylmercury degrades to (mono)methylmercury under acidic pH condition.

## 1.4 Uses of mercury

Mercury is used for the manufacture of industrial chemicals or for electrical and electronic applications or thermometers as well as dental fillings. The consumption of refined mercury in the USA between 1985 to 2001 is given in Table 1-2. It revealed the release of mercury into the environment in 2001 is lower level than in the period from 1985 to 1996. However, total world production of mercury in 2004 is about 1,260 metric tons, which is slightly decreasing from the year 2000s. (http://www.indexmundi.com/en/commodities/minerals/mercury table %20.html).

Table 1- 2Consumption of refined mercury in the USA between 1985 and 2001<br/>(metal values are in metric tons)

|                                | 1985  | 1990 | 1992 | 1993 | 1994 | 1995  | 1996 | 2001 |
|--------------------------------|-------|------|------|------|------|-------|------|------|
| Chlor-alkali                   | 235   | 247  | 209  | 180  | 135  | 154   | 136  | 46   |
| Paint                          | 169   | 22   | 0    |      |      |       |      |      |
| Laboratory                     | 14    | 32   | 18   | 26   | 24   |       |      |      |
| Other chemical/allied products |       |      | 18   | 18   | 25   |       |      |      |
| Electric lighting              | 10    | 33   | 55   | 38   | 27   | 30    | 29   | 28   |
| Wiring devices and switches    | 95    | 70   | 69   | 83   | 79   | 84    | 49   | 60   |
| Batteries                      | 952   | 106  | 16   | 10   | 6    | < 0.5 |      |      |
| Measuring Instrument           | 79    | 108  | 52   | 65   | 53   | 43    | 41   | 22   |
| Dental                         | 50    | 44   | 37   | 35   | 24   | 32    | 31   | 44   |
| Other uses                     | 84    | 58   | 148  | 103  | 110  | 93    | 86   |      |
| Total                          | 1,718 | 720  | 622  | 558  | 483  | 436   | 372  | 200  |

Source: www.altcorp.com/dentalinformation/hgwastes.htm

#### **1.5** Amalgam filling and chemical reactions





Figure 1-1 Amalgam Filling

Dental amalgam is the most widely used for the restoration of premolar and molar teeth. It is a mixture of alloy powder and mercury, which after setting forms a high strength solid mass. The alloy powder comprises of 45-70% silver, 12-30% tin, 5-30% copper and 0-2% zinc depending on types of alloy. Set silver amalgam consists of 43-50% mercury which is combined with either silver or tin. In the unset alloy, silver and tin are presented as  $Ag_3Sn$  (gamma phase). In set amalgam mercury reacts with both silver and tin to form  $Ag_2Hg_3$  (gamma-one) and  $Sn_{7-8}Hg$ (gamma-two). A chemical reaction of setting amalgam is shown

$$Ag_{3}Sn + Hg \rightarrow Ag_{2}Hg_{3} + Sn_{7-8}Hg$$
(1)  
gamma gamma-one gamma-two

 $Sn_{7-8}Hg$  corrodes easily in the oral environment and releases tin ions and mercury (Mateer and Reitz, 1970). Some of this mercury may react with residual  $Ag_3Sn$  to form new  $Ag_2Hg_3$  and  $Sn_{7-8}Hg$  phases, and some mercury and metal ions may penetrate dentin<sup>1</sup> (Horsted-Bindslev *et al.*, 1991). The residual mercury in the restoration may be released to the oral cavity as elemental mercury which is dissolved in the saliva or leaves in the breath. The total amount of mercury released from dental amalgam depends upon total numbers of fillings and surface areas of each filling,

<sup>&</sup>lt;sup>1</sup> The dense, yellowish calcareous tissue that forms the main part of tooth, between the enamel layer and the pulp cavity

chewing, grinding motions, eating habits of the person, corrosion and other chemical conditions in the mouth (Horsted-Bindslev *et al.*, 1991; Gainsford *et al.*, 1992).

New formulation of high-copper amalgam is introduced. The chemical reaction of setting amalgam is shown below.

$$Ag_{3}Sn + Cu + Hg \rightarrow Ag_{2}Hg_{3} + Cu_{6}Sn_{5}$$
<sup>(2)</sup>

 $Cu_6Sn_5$  is less sensitive to corrosion and resulted in less emission of mercury from amalgam. However, it is not in common use because of poor physical characteristics.

## **1.6** Bioavailability, mercury absorption metabolism and health effect

The bioavailability of trace metal is strongly influenced by their speciation. Hg has high stability constant with organic ligands. It can form organometallic compounds such as methylmercury. The interaction of Hg with biological component depends on physicochemical variables and biological activities, which control the speciation, binding, release and pathway of Hg. Metal biosorption is described as passive metal uptake, which occurs owing to the chemical composition of living cells and metal properties. The protein and structural polysaccharides of the cells contain a series of available metal binding sites. These binding groups include carboxyl, thiol, phosphate, sulphydril and amine groups. They are components of cell wall and cytoplasmic membrane and also external layers (da Costa, 1999). Hg has a high affinity for biomolecules such as amino acids, proteins (amino acid sequences), pyrimidines and nucleic acids (Simkiss and Taylor, 1995). The  $Hg^{2+}$  ion presents low affinity for phospholipids while alkyl mercuric chlorides promptly interact with phospholipid layers (Bharathi et al., 1990). Mercury affects body metabolism by inactivating many enzymes such as alkaline phosphatase, lactate dehydrogenase and glucose-6-phosphataseby binding to the sulfhydryl group of catalytic cysteine groups in the protein backbone (Mason and Jenkin, 1995). The elimination of inorganic mercury from the body differs from different tissue, time and concentration of exposure to mercury. The route of excretion after short-term exposure is fecal (50%), followed by exhalation (37%) and urinary excretion (13%) (Horsted-Bindslev, 1991).

The excretion half-time of mercury from the whole body was approximately 42 days (Rahota *et al.*, 1973).

Mercury is a health hazard in all its various forms. In poisoning incidents that occurred in some countries such as mercury contaminated in seafood from Minamata Bay, Japan in 1956 and approximately 459 deaths resulted from the use of methylmercury fungicide for wheat treated in Iraq in 1971, mercury contaminated fish in Canada (Hartung and Dinman, 1972; Diez and Bayona, 2002). The symptoms of chronic mercurialism are paresthesia (abnormal sensation), ataxia (failure of muscular coordination), hearing deterioration, tremor and erethism<sup>1</sup>. Short exposure to high concentration of mercury vapor (1-3 mgHg m<sup>-3</sup>) leads to acute intoxication manifested by the signs of pulmonary inflammation (Lien *et al.*, 1983).

# **1.7** Occupational exposure

The professionals who expose to mercury by the release from amalgam are dental personnel, employees from dental industry and employees at crematories. The dental team is exposed to mercury vapor daily in connection with operative silver amalgam treatment. The exposure can also be very high when inserting new filling, during polishing and when drilling out old amalgam filling (Engqvist, 1998). The threshold limit value (TLV) for mercury vapor has been established and is use as a guideline for recommendations and occupational safety in many countries. WHO recommended an occupational long-term exposure limit of 25  $\mu$ g m<sup>-3</sup> (WHO, 1980). It has been suggested that dental clinics should have good ventilation and dentists should not handle the material with their hands (Harakeh *et al.*, 2002). Employees in the dental industry which manufacture dental material can be exposed as well as dental technicians. Employees at crematories can also be exposed to mercury vapor during the cremation of corpses having amalgam.

<sup>&</sup>lt;sup>1</sup> Erethism is a mental disturbance, characterized by acute irritability, abnormal shyness, indecision and over reaction to criticism

For non-occupational people, they can be exposed to mercury from their amalgam filling. It enters human body as vapor mercury or be dissolved in saliva. The amount of mercury release depends on number of fillings, surface areas of each filling, the chewing and eating habits, grinding teeth during sleep or when stressed and other chemical conditions in the mouth (Taskinen *et al.*, 1989; Barregard *et al.*, 1995; Engqvist, 1998). Estimates of the amount of mercury released from dental amalgam range from 3 to 17  $\mu$ g day<sup>-1</sup>

(http:// www.atsdr.cdc.gov/toxprofiles/phs46.html). Figure 1-2 show the mercury partwat to the dental personnel.



Figure 1-2 Mercury pathway in dentistry (Horsted-Bindslev, et al., 1991)

#### 1.8 Human hair

## **1.8.1** Hair struture and binding with mercury

Hair is a cross-linked, partially crystalline, oriented, polymeric network. It carries various functional groups (e.g. acidic, basic and peptide bonds) which are capable of binding small melanin, which determines the hair color. Hair is composed primarily of proteins 88% on average. These proteins are of a hard fibrous type known as keratin. This keratin has varying amounts of disulphide bridges (S-S) and it is comprised of polypeptide chains (Holsbeek *et al.*, 1996). The high affinity of hair for metal is due mainly to the presence of cystine, which up approximately 14% of human hair. Many metals found in hair are bound to sulfur atoms in cystine or to sulphydryl (SH) groups presented in amino acids. Melanin is polyanionic polymer containing negatively charged carboxyl groups and semiquinones at physiological pH and as a result can bind cations by ionic interaction. Other forces such as Van de Waal's attraction can also enhance the ionic binding. Uncharged metals, e.g. elemental mercury, may also bind to the hydrophobic core of the melanin polymer in the hair structure (Horsted-Bindslev *et al.*, 1991; Holsbeek *et al.*, 1996).

#### 1.8.2 Advantages of analysis of mercury in human hair

Analysis of mercury in human hair offers several advantages. Firstly, the level of total mercury in hair is about 300-times larger than in other human materials. Secondly, hair sample is easier to acquire and store. Thirdly, hair grows slowly approximate 1 cm each month, so even hair closest to the scalp is several weeks old and thus may not reflect current body conditions for purposes of health diagnosis. Finally, the material is relatively inert as well as homogeneous (Salmela *et al.*, 1981; Holsbeek *et al.*, 1996; Wasiak, *et al.*, 1996; Dolbec *et al.*, 2001; Chen *et al.*, 2002; Morton *et al.*, 2002). Thus, determining trace elements in human hair has importance in biological, medical and environmental studies, as human hair represents an interesting biological indicator. The normal mercury value in human hair ranged 0.4-6.0  $\mu$ g g<sup>-1</sup> and the mercury content of > 50  $\mu$ g g<sup>-1</sup> is poison (WHO, 1972). Range of mercury concentration in human hair from various countries is presented in Table 1-3.

| Region    | Countries          | Range of Hg $(\mu g g^{-1})$ | Method     | References                          |
|-----------|--------------------|------------------------------|------------|-------------------------------------|
|           | Bangladesh         | 0.02-1.0                     | ICP-MS     | Holsbeek et al., 1996               |
|           | China              | 1.2-5.5                      | GFAAS      | Chen et al., 2002                   |
|           | Hong Kong          | 2.4-3.8                      | ICP-MS     | Dickman et al,., 1999               |
|           | Indonesia          | 1.9-2.9                      |            | Ohno et al., 1984                   |
|           | Japan              | 1.9-9.6                      | A small    | Takeuchi et al., 1982               |
|           |                    | 9.6-14.2                     | size       | Sera et al., 2002                   |
|           |                    |                              | cyclotron, |                                     |
|           |                    |                              | X-rays     |                                     |
|           |                    | 0.2-7.5                      |            | Nakagawa, 1995                      |
| Asia      | Singapore          | 4.0-5.1                      |            | Foo <i>et al.</i> , 1988            |
|           |                    | 1.1-35.5                     | CVAAS      | Foo and Tan, 1998                   |
|           | Thailand( Bangkok) | 2.8-10.1                     | CVAAS      | Saengsirinawi and                   |
|           |                    |                              |            | Pringsulaka, 1988                   |
|           | Kuwait             | 4.1-5.5                      | CVAFS      | Bou Olayan and Al Yakoob,           |
|           |                    |                              |            | 1994                                |
|           |                    | 25-1192.5                    |            | Al-Majed and Preston, 2000          |
|           | Papua New Guinea   | 0.8-21.9                     |            | Abe, 1995                           |
|           | Lebanon            | 0.3-24.2                     |            | Harakeh et al., 2002                |
|           | Egypt              | 0.2-0.3                      |            | Mortada <i>et al.</i> , 2002        |
| Africa    | Australia          | 0.05-32.4                    | Injection  | Gardner and Dunne, 1979             |
|           |                    |                              | manifold-  |                                     |
|           | ~                  |                              | AAS        |                                     |
| Australia | Germany            | 0.5-1.0                      |            | Wilhelm <i>et al.</i> , 1996        |
|           | Poland             | 0.06-0.7                     |            | Hac <i>et al.</i> , 2000            |
|           | Spain              | 0.200.9                      |            | Moreda-Pineiro <i>et al.</i> , 2002 |
|           | UK                 | 0.2-0.6                      | ICP-MS     | Lindow <i>et al.</i> , 2003         |
|           | UK                 | 0.5-2.1                      | ICP-MS     | Morton <i>et al</i> , 2002          |
|           | UK                 | 0.02-0.07                    | CVG-AFS    | Rahman <i>et al.</i> , 2000         |
| -         | Albania            | 0.2-2.0                      | CVAAS      | Babi <i>et al.</i> , 2000           |
| Europe    | USA                | 0.7-0.8                      | arts sa    | Creason <i>et al.</i> , 1982        |
| America   | Brazil             | 2.9-27.0                     | CVAAS      | Dolbec <i>et al.</i> , 2001         |
|           |                    | 4.0-20.0                     | <b></b>    | Passos et al., 2003                 |
|           |                    | 0.9-24.0                     | CVAAS      | Leino and Lodenius, 1995            |

 Table 1-3
 Mercury concentration in human hair from various countries

# **1.9** Analytical method

In trace element analyses of human hair, the pretreatment and digestion are important procedures The hair sample must be washed because grease and dust on the surface of hair may contribute most of the element concentration. Different laboratory uses different washing procedure and this is no consensus on how the washing should be done (Salmela *et al.*, 1981). The preliminary digestion procedures can be classified broadly into wet oxidation (digestion) and dry oxidation (combustion or pyrolysis). Wet oxidation procedures involving one or more oxidizing agents in an acidic medium are commonly employed to liberate mercury from sample matrices. Reagents that have been used in wet oxidation procedures are HF, HCl, HClO<sub>3</sub>, HClO<sub>4</sub>, HBr, HBrO<sub>3</sub>, H<sub>2</sub>SO<sub>4</sub>, HNO<sub>3</sub>, H<sub>2</sub>O<sub>2</sub>, KMnO<sub>4</sub>, K<sub>2</sub>Cr<sub>2</sub>O<sub>7</sub>, and V<sub>2</sub>O<sub>5</sub>. Mixtures of various reagents have been used as well (Schroeder, 1995). Incomplete destruction of the matrix (e.g. biological material, food stuffs, plant material, soils, rocks) can lead to erroneous analytical results. A disadvantage of these procedures is their time-consuming and tedious nature. However, the uses of microwave digestion has been gaining wider acceptance for its rapid decomposition, reduced risk of contamination, small volume of acid required and no loss of volatile elements (Loring and Rantala, 1995).

The most commonly used methods for mercury analysis are Cold Vapor Atomic Absorption Spectrometry (CVAAS), neutron activation (NA) Inductively Couple Plasma Mass Spectrometry (ICPMS). Some other techniques are Graphite Furnace Atomic Absorption Spectrometry (GF-AAS) equipped with a Zeeman background corrector was used to study total mercury and methylmercury in hair samples (Chen *et al.*, 2002); direct Cold Vapor Generation-Electrothermal Atomic Absorption Spectrometry (CVGE-AAS) was performed for mercury in aqueous slurries environmental samples. The sample was reduced by sodium borohydride (NaBH<sub>4</sub>) and vapor mercury was trapped on Iridium treated graphite tube and atomization at 2600°C (Moreda-Pineiro *et al.*, 2002). Cold Vapor Atomic Fluorescence Spectrometry (GC-CV-AFS) has been utilized for the detection of methylmercury in biological materials (Diez and Bayona, 2002).

CVAAS is one of the most popular techniques for determining mercury in a wide variety of samples. It has the advantages of its high sensitivity, absence of spectral interferences, relatively low operation costs, simplicity, and speed. Generally, mercury in aqueous samples was reduced by sodium borohydride and vapor mercury is purged from solution by a carrier gas, such as, nitrogen or argon. After passing through a gas-liquid separator, it is introduced into the path of an atomic absorption spectrometer (Malm *et al.*, 1995; Holsbeek *et al.*, 1996).

#### 1.10 Research Objectives

The objectives of this study are (i) to optimize pretreatment and analytical procedures for mercury analysis in hair samples; washing reagents, drying temperature, digestion acid, digestion time and analytical parameters for mercury analysis by CVAAS; (ii) to determine total mercury concentration in hair of dental personnel at Prince of Songkla University and unexposed Hat-Yai residents; and (iii) to investigate correlation factors of mercury content in the hair of dental personnel and in non-exposed persons by using statistical analysis (SPSS).

# 1.11 Anticipated Outcome

This study will provide the database concerning mercury burden in dental personnel hair for considering the mercury hygiene in the dental clinic and the environment and establish the background level of mercury in non-occupationally exposed individual.