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CHAPTER 1

INTRODUCTION

1.1 BACKGROUND

1.1.1 Mining and minerals

The earth's crust is made up of a relatively small group of rock-forming minerals, mainly oxygen, silicon, aluminum and iron. Smaller quantities of the other minerals are also present but are usually sparely dispersed (Lurie, 1977). However, various geological processes have brought about the concentration of specific minerals at certain places.

Since ancient times, minerals have been used by man for various purposes, ranging from the building of structures; household items (clays used for building paints, dyes, decorations, containers, roofing), for hunting and fighting, and for trading and other economic purposes. The use of minerals have increased over time, both in terms of the types of minerals that are defined as being 'useful' and the variety of uses for each mineral. At present, civilization is heavily dependent on raw materials which are obtained from the earth's crust. These include, *inter alia*, gold, silver, platinum, copper, chromium, lead, nickel, tin, vanadium, zinc, beryllium, niobium, tungsten, zirconium, boron, antimony, arsenic, germanium, carbon, sulphur and bismuth (Lurie, 1977). Economically exploitable minerals are extracted from the earth's crust through surface, underground and marine mining activities wherever sufficiently large deposits occur.

South Africa boasts an abundance of mineral resources, producing and owning a significant proportion of the world's minerals (www.southafrica.info, 2008).

South Africa's mineral wealth is found in diverse geological formations, some of which are unique and extensive by world standards. The data in Fig. 1.1 show that South Africa has nearly 90% of the platinum metals on Earth, 80% of the manganese, 73% of the chrome, 45% of the vanadium, 41% of the gold, zirconium 20% and titanium about 29%.

South Africa's position as the world's largest gold producer was usurped by China in 2007, nevertheless it accounts for over 10% of world gold production. It is also the leading producer of platinum, manganese, titanium, chrome, zirconium and vanadium. Only crude oil and bauxite are not found here (www.southafrica.info /mining, 2008).

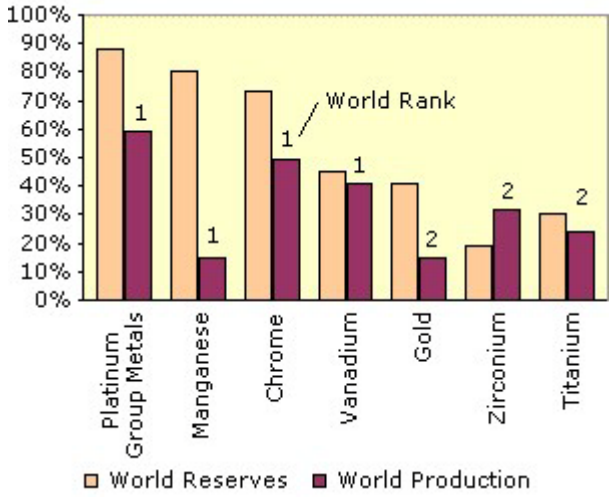

Fig. 1.1 South Africa's share of world reserves and production (www.southafrica.info, 2008)

The Limpopo Province is fast becoming the biggest mining province in South Africa due to its wide range of mineral deposits (Citizen, 2004). It is a major producer of diamonds, platinum, copper, gold magnetite, antimony, emeralds, iron ore silicon, feldspar, black granite and vermiculite. At present, mining contributes to over a fifth of the provincial economy (Citizen, 2004; City Press, 2006; Financial Mail, 2006). Major mining towns in the area include Phalaborwa in the east and Thabazimbi in the west. Two towns in Limpopo are set to boom in the near future. One is Burgersfort, in central Limpopo, where five platinum mines have been established and at least another eight are in various developmental stages. The other is Lephalale, formerly Ellisras, in North West Limpopo where coal mining has been established (Financial Mail, 2006).

1.1.2 Minerals and health

Although mining is usually associated with generating economic wealth and prosperity, many of the activities associated with mining have negative environmental impacts.

Many mineral contaminants released through mining activity are present in the environment as exchangeable ions which may adsorb onto the surface of clay, organic matter or oxides with weak bonds and are easily moved and dispersed into the ecosystem. Minerals can be transported, dispersed to and accumulated in plants and animals and can be passed across the food chain to humans (Hongyu *et al.*, 2005). Since mineral concentrations may consistently biomagnifies from one trophic level to the next, animals higher in the food chain may accumulate more toxins than their food contains (Monteiro *et al*., 1996). Some minerals, such as sodium chloride, are excreted with ease and hence are non-toxic to the body whereas the excretion rate of others is extremely low, resulting in the accumulation of some minerals, metals and metalloids, in biological tissue. These may eventually reach toxic levels (Sami & Druzynkli, 2003).

It has been established that the dietary exposure is the major route for metal bioaccumulation of many marine and terrestrial animals (Shore, 1995; Talmage & Walton, 1991; Wang & Fisher, 1999; Kormarnicki, 2000). Metals and metalloids that accumulate in biological tissue may be converted to other chemical forms and passed on via numerous pathways. Humans, as the final consumers in the food chain, are thus the likely recipients of high levels of minerals from 'contaminated' food and may accumulate high concentrations of some minerals in their tissues (Oskarson *et al.*, 1992).

Many minerals, especially heavy metals such as lead, mercury, uranium, chromium and other minerals such as arsenic are toxic. These metals affect all living organisms and have possible toxic impact on all levels of the food chain (Sami & Druzynkli, 2003). According to the Agency for Toxic Substances and Disease Registry (ATSDR) (2007), the most toxic substances that pose a significant potential threat to human health, are arsenic (As), lead (Pb) and Mercury (Hg). The effects of the toxicity may be acute (if large amounts are ingested or the element is particularly toxic) or chronic, with toxicity

only being noticed after a long time (Toens *et al.*, 1998; Bahemuka & Mubofu, 1999; Ikeda *et al.*, 2000). The latter may be the result of the bioaccumulation process across the food chain (Beretta, 1984; Lacatus *et al.*, 1996; Turkdogan *et al.*, 2003).

Mining and smelting of ores have thus increased the prevalence and occurrence of toxic elements through dust emissions, mine tailing and waste water (Hongyu *et al.*, 2005; Osher *et al.*, 2006). Mining is a major source of contamination of land surfaces as well as surface- and groundwater (Osher *et al.*, 2006). Lasat (2002) and Mc Grath *et al.* (2001) showed that exposure to toxic metals results in the reduction of microbial diversity and their activities. Increased concentrations of toxic elements result in a low fitness of animals, reproduction problems, decline in immunity and the occurrence of cancerous and teratogenic diseases (Bires *et al*., 1995). According to Magbagbeola & Oyeleke (2003), there is a significant association between the presence of heavy metals and the incidence of some human diseases.

The consumption of food contaminated by lead, mercury, arsenic, cadmium and other metals can seriously deplete the body stores of iron, Vitamin C and other essential nutrients leading to decreased immunological defenses, intra uterine growth retardation, impaired psycho-social faculties and disabilities associated with malnutrition (Iyengar & Nair, 2000). The intake of arsenic and some heavy metals may cause chromosomal aberration, genetic abnormalities and can lead to cancer. There are many studies indicating the link between heavy metals and cancer. For example, the high concentration of lead in fruits and vegetables in the Van region of Eastern Turkey were found to be related to a high upper gastrointestinal cancer rates (Turkdogan *et al.*, 2003). In Bangladesh, where there is a high concentration of arsenic in groundwater and some foods, various diseases ranging from melanosis, leukomelanosis, keratosis, hyperkeratosis, non-pitting edema, gangrene, and skin cancer are prevalent (Hindmarsh *et al*., 2002).

1.1.3 Detection of exposure to toxic minerals

In order to be in a position to detect toxicity risk, it is necessary to estimate the extent to which living organisms are exposed to contaminants (Samuillah, 1990; Shore & Douben, 1994). Selection of nutrient sources at the higher trophic levels in terrestrial ecosystems where bioaccumulation has already occurred could thus be useful in predicting risks to human health (Komarnicki, 2000). Specifically, the early detection of potential toxicity problems can be achieved by focusing on the nutrient sources of the most vulnerable members of society i.e. the very young and the old. This study focuses on the very young i.e. infants.

Patriarca *et al*. (2000) highlight the fact that substantial differences exist between the physiology, metabolism and lifestyle of young children compared with adults. For instance, infants have a higher resting metabolic rate and both food consumption and surface area per unit body weight is higher than in adults. Gastrointestinal absorption of several chemicals, including metals, is higher in the young because of the characteristics of the intestinal function and structure at this age as well as the type of diet. They also have higher percentage water in their bodies, which results in twice as large an extracellular water compartment compared to adults (Patriarca *et al*., 2000:352). Binding of toxins to plasma proteins is less because of the lower concentration of albumin found in the plasma of newborns and competition for binding sites with endogenous substances. However, this may not result in faster elimination since infants' kidneys are immature. "The renal function in newborns and infants is tailored for their special needs, thus they have higher retention of nitrogen and minerals than adults and other children" (Patriarca *et al*., 2000:353). They are thus especially vulnerable to the ingestion of any potentially harmful substance.

Milk is a basic food in the diet of infants. As an excretion of the mammary gland, milk can carry numerous xenobiotic substances (drugs, pesticide, metals and various environmental contaminants) which constitute a toxicological risk. It is one of the main sources by which animals and man are exposed to toxic elements (Licata *et al.*, 2003).

Milk is thus an obvious choice for the identification of infant exposure to toxicity. According to Miller (1971), after consumption of a toxic element, its concentration increases quickly in animal milk, while the excretion rate is much slower (Miller, 1971; Houpert *et al.*, 1997). The demand for healthy milk, rich in nutrients with high biological value calls for the determination of residual concentration of metals in milk. However, relatively little attention has been paid to the transfer of toxic substances to infants via milk (Oskarsson *et al.,* 1995).

1.2 JUSTIFICATION FOR THE STUDY

Despite its mineral wealth, the Limpopo province is one of the poorest regions in South Africa and has a strong rural basis (Human Science Research Council, 2000). The Limpopo Province, which consists of five districts, namely Waterberg, Capricorn, Vhembe, Mopani and Sekhukhune, is the northernmost of all the provinces. Figure 1.2 shows the map of Limpopo province, delineating the five districts.

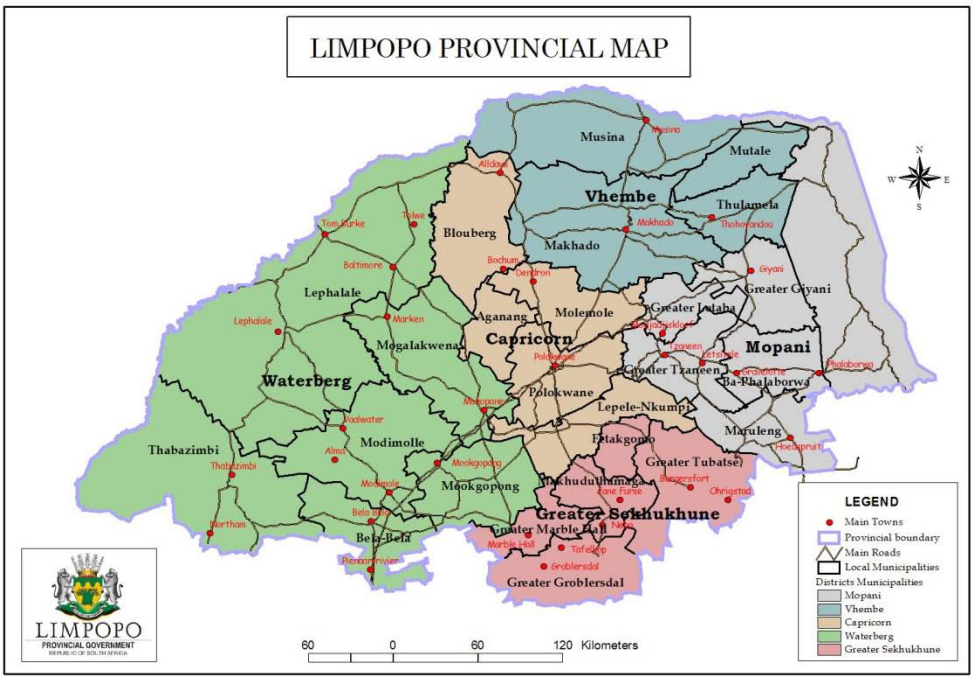

Fig 1.2 Map of Limpopo showing the districts (Data source: www.limpopo.gov.za¸2008)

Many households still have unsatisfactory access to education, health care, energy and clean water (Gyekye & Akinboade, 2003). According to the Human Science Research Council (2000), the population of Limpopo was 5 277 432 in 2000. This constituted 11.7% of South Africa's total population. According to the 2001 Census, 33.4% of the population aged 20 years or more, had no formal school education; 49% of those in the age group 15-64 years were unemployed; 33% of those who were employed were in elementary occupations. The official unemployment rate for Limpopo is 35.6% (SSA, 2003; SSA, 2006). In 2002, over six out of ten persons (61%) lived below the national poverty line (Adelzadeh & Ohiorhenuan, 2004). Almost one-quarter of households did not have access to toilet facilities and a mere 14% had a refuse removal service at least once a week. The poverty is most acute in the rural areas of Limpopo, where 72.5% of people live in poverty, compared to 32.8% in urban areas (Punt *et al.*, 2005).

Limpopo is a typically developing area, with many rural people practicing subsistence farming (South Africa Year Book, 2003/04). Many rural inhabitants keep livestock including cattle, goats, sheep, horses and donkeys. The Department of Agriculture statistics estimated that at the end of November 2003, South Africa had 6 681 000 goats of which Limpopo is the second largest goat rearing province (with 3 022 000 goats) after the Eastern Cape (SSA, 2003). The goats are used as food and for their skins. Due to the poverty levels, rural mothers often give their babies goat's milk to complement the quantity of breast milk given to them (Mafu & Masiki, 2002).

Limpopo province is a relatively low rainfall region with only a few strong perennial rivers and dams thus most of the population is dependant upon groundwater obtained from wells and boreholes (Rosewarne, 2005). However, highly toxic minerals, such as lead and mercury are mined in Limpopo. In fact, many minerals, such as gold, copper, tins and platinum with which arsenic are associated, occur throughout the Province. It is thus possible that surface and groundwater sources may be contaminated with toxic minerals. Therefore people in Limpopo, especially those living in rural areas, may be

exposed to harmful levels of mineral toxicity. If the water sources are contaminated with toxic minerals, bioaccumulation of toxic minerals will occur across the food chain.

Although a considerable amount of research has been done worldwide on toxic mineral contamination, it appears that hardly any has been conducted on these toxic minerals in Limpopo Province. Moreover, as far as is known, no research has been carried out on the level of mineral contamination to which infants are exposed in this Province.

1.2 STUDY AIMS AND OBJECTIVES

1.2.1 Aims

This research aims to establish whether toxic elements in the environment pose a threat to the health status of infants living in the rural areas of Limpopo province.

1.2.2 Objectives

The objectives of the study are

- To identify potentially contaminated areas in Limpopo Province.
- To describe the extent of environmental contamination in selected areas.
- To determine the exposure pathway to humans in contaminated areas.

The study will focus on the three most toxic minerals, namely mercury (Hg), lead (Pb) and arsenic (As) and is confined to the Limpopo Province.

1.3 RESEARCH FRAMEWORK

According to Ott *et al*. (2007: 8), traditional studies regarding toxic elements in the environment and their effects, relay on an environmental analysis approach. This comprises the identification of a source of pollution and then attempting to determine the risk it poses – without necessarily identifying the target of the risk. This study, however, follows an exposure analysis approach to risk assessment as described by Ott *et al.*

(2007), in that it works from both directions, both the source of contamination and the receptor (target). It seeks to consider the routes of exposure by which the pollutant may reach the target. Exposure is deemed to occur when any pollutant makes direct contact with the human being through air, water, soil or food (Ott *et al*., 2007: 7).

This research focuses on exposure of infants to the toxic agents, arsenic, mercury and lead. In this study the term 'infant' includes newborns (birth to 4 weeks) and infants (from 1 month to 1 year) (WHO, 1986) whose diet consists mainly of milk products. Although exposure pathways include inhalation, dermal exposure and ingestion, the present study focuses on exposure through ingestion alone. Sources of contamination include water, food, and soil.

Figure 1.3 gives a diagrammatic representation of the sources and pathways of contamination to infants.

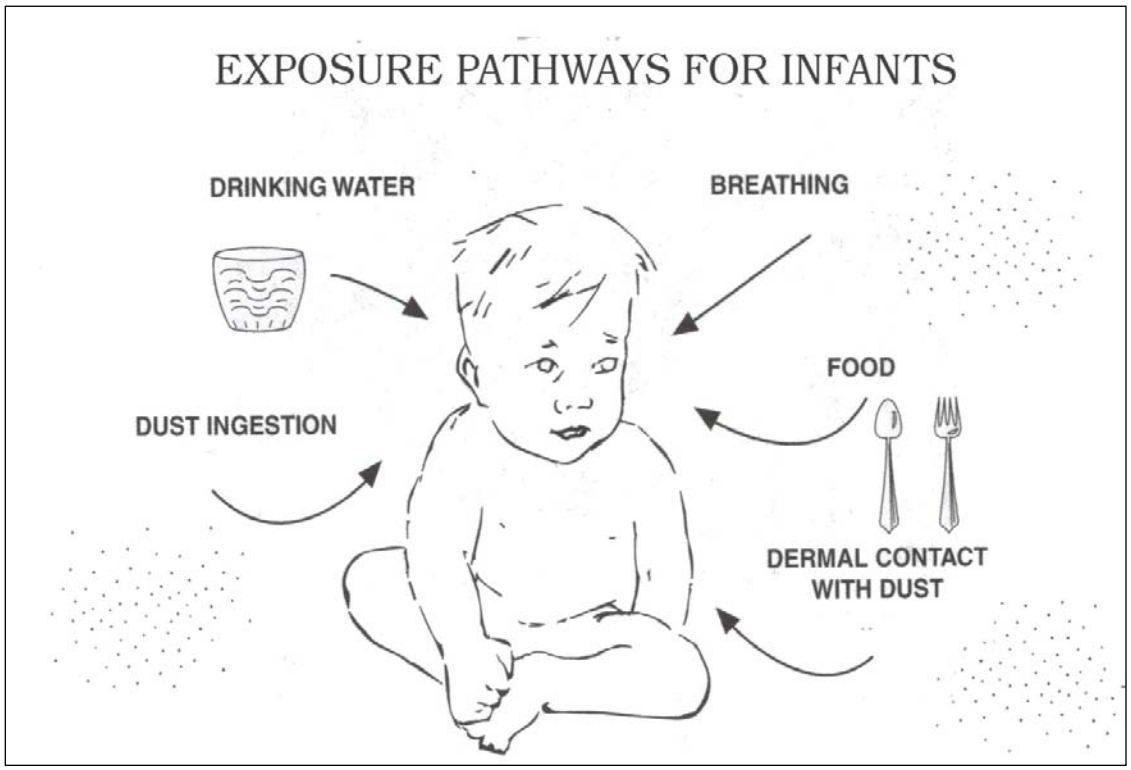

Fig 1.3 Contamination sources and pathways to infants *(Ott et al., 2007, cover page).*

It should be noted that this research does not measure the exposure dose i.e. the amount of contaminant that actually enters the body cells and fluids (body burden) by absorption or intake or the impact of the toxins on human beings.

1.4 CHAPTER LAYOUT

Chapter 2 comprises the literature review. This chapter includes literature that deals with the risks associated with lead, mercury and arsenic and their bioavailability. The existence of these three metals in plants/crops, animals and milk is reviewed. The chapter also includes the health risks that these three elements can pose to human beings.

The methodology used in the research is discussed in Chapter 3. It includes the use of geological maps to identify areas that are potentially at risk, the selection of the communities from where the samples were taken as well as the collection of the samples and their analyses.

In Chapter 4, the results of the analyses of toxic minerals in the environment are presented. The chapter focuses on the levels of arsenic, mercury and lead in groundwater, soil samples and plant material from the study areas. The concentrations found in the study areas are compared with contamination levels in other parts of the world.

Chapter 5 forms the crux of the study, in which exposure by to infants of the three most toxic elements, namely arsenic, mercury and lead are discussed for a selected study area.

The final chapter gives an overview of the research, the conclusions reached and recommendations for further research.

CHAPTER 2

LITERATURE REVIEW

There are various studies and a large volume of literature on toxic minerals; their sources and their effects on both ecosystems and humans. Toxic mineral contamination is becoming increasingly common. Worldwide, and possibly in South Africa, the effect of the toxicity ranges from simple health problems such as skin disease to serious ones such as teratogenesis and abortion even death.

This chapter gives an overview of the hazards of toxic minerals followed by sections dealing specifically with the environmental and health impacts of lead, mercury and arsenic. Since milk is the primary source of nutrient for the very young and old people, the chapter concludes with a section on the dangers of toxic minerals contamination in milk.

2.1 HAZARDS OF TOXIC MINERALS

At present, domestic and wild animals as well as man are being exposed to various aspects and factors which are foreign to the habitat in which they live. Examples are heavy metals and toxic elements. Heavy metals contamination could originate from natural sources (contaminations from the bedrock) or from anthropogenic sources. The anthropogenic source of heavy metals in the soil are either as a consequence of mining, smelting and aerosol deposition (Fergusson, 1990), or as an outcome of working on the soil such as the application of fertilizer (agriculturally induced), waste pollution via water and landfill sites. In addition, the dynamic development of industry and transportation as well as the continuing over-intensive use of various chemical compounds in agriculture cause levels of toxic heavy metals in the environment to constantly be on the increase (Topolska *et al.*, 2004).

Mining is arguably the largest anthropogenic source of mineral contamination. Geochemical processes acting upon mining wastes initiates the process of transporting heavy metals from contaminated areas and redistributing them to the surrounding soils, streams, groundwater, thus endangering the health of the surrounding ecosystem and human population (Barcan & Kovnatsky, 1998; Mc Grath *et al.,* 2001; Shi *et al.,* 2002). The presence of some metals in almost all places facilitates the increasing possibility of exposure to man and animals (Beretta, 1984).

Microbial diversity and activities of the soil are important indices of soil quality since soil microbes play a significant role in recycling of plant nutrients, maintenance of soil structure, detoxification of noxious chemicals and the control of plant pests and growth (Elsgaard *et al.,* 2001; Fillip, 2002). Heavy metals interact with physiological elements which become harmful to these living organisms (Topolska *et al*., 2004).

In higher organisms, the intake of toxic elements occurs mainly through the respiratory system or through water intake and food ingestion which goes across the food chain (Caggiano *et al.,* 2004). Food chain contamination is one of the major routes for entry of metals into the animal system and therefore, monitoring the bioavailability pools of metals in contaminated soil and ground water are of a paramount concern. Ruminants such as goats, sheep and cattle feed on grasses which have absorbed and accumulated elements from the soil over time. A number of reports have confirmed the transference of trace metals from contaminated soil to plants and from plants to livestock such as Oskarsson *et al.* (1992), Brun *et al.* (2001), and Lehmann & Rebele (2004). Recent models have been developed to predict the dynamic transfer of arsenic (As), mercury (Hg) and cadmium (Cd) to animal tissue. These have been verified through experimental studies (Beresford *et al.* 1999; 2001). There is also definite evidence of subsequent transfer through the food chain up to human. According to the information from the report of the Agency for Toxic Substance and Disease Registry on the web page (Agency for Toxic Substance Disease Registry, 1999) arsenic, lead and mercury head the priority list of toxic minerals. Thus, it is of a paramount importance to study the effect of these minerals on plants animals and humans in order to enhance good health of the population.

2.2 TOXIC MINERAL CONTAMINATION IN SOUTH AFRICA

South Africa has a rich diversity of minerals, many of them being either toxic themselves, or associated with toxic elements. However, relatively little research has been conducted on the affect of toxic minerals on humans and the environment.

The most- researched toxic mineral in South Africa is undoubtedly arsenic.
Arsenic-bearing minerals are primarily associated with geological settings where sulphide mineralization has taken place. They are thus commonly found in geological environments associated with carbonaceous shale, volcanogenic massive sulphide deposits (VMS), lead zinc deposits, hydrothermal terrains, geothermal springs, and the platinum group element seams. Arsenic is thus usually present in areas where gold, silver, copper, nickel, zinc, lead and cobalt are found (Sami & Druzynski, 2003). Such deposits are widespread throughout South Africa, especially in the Limpopo Province. Since arsenic dissolves readily through a range of pH and Eh(reduction-oxidation potential) conditions, this element has been recognized as one of the most serious inorganic contaminants in drinking water obtained from groundwater sources (Sami & Druzynski, 2003).

The major source of water supply in the rural areas of South Africa is groundwater. It is utilized for both domestic and agricultural use and it is a major pathway of arsenic mobilization (Sami & Druzynski, 2003). Analysis of 1514 boreholes in South Africa showed that 252 contained measurable arsenic concentrations, 200 of which have concentrations exceeding the WHO limits of 0.01 mg/l and over 40 had concentration exceeding 0.1 mg/l (Sami & Druzynski, 2003). Figure 2.1 below shows the levels of arsenic concentration in sampled boreholes in South Africa (Sami & Druzynski, 2003).

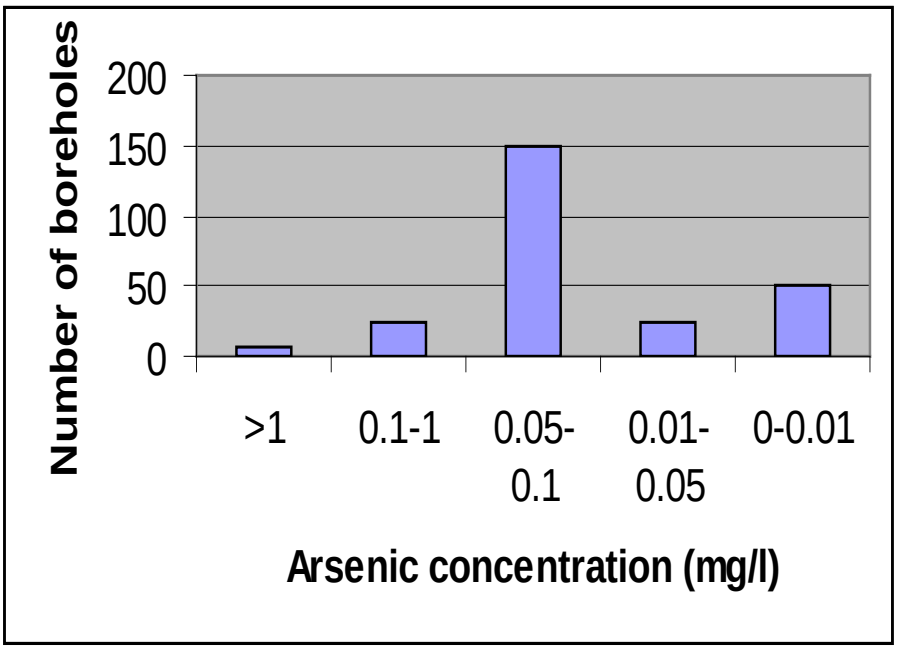

Figure 2.1 Number of boreholes with their levels of arsenic contaminants in South Africa *(Sami & Druzynski, 2003).*

Numerous lead deposits occur in the dolomitic regions of Limpopo and the North West Province while Mercury mining took place in the vicinity of Gravelotte in Limpopo. Lead and mercury are also associated with many mining activities in South Africa.

It is thus clear that all three of the most dangerous minerals, namely, arsenic, mercury and lead are found in high concentrations in parts of South Africa and it is possible that these elements might enter the food chain and affect the health of South Africans.

The following sections give an in-depth review of the environmental and health impacts of arsenic, mercury and lead. Each section discusses the occurrence of the minerals, contamination pathways, and impacts on micro-organisms, plants, animals and man. Since infants are particularly vulnerable to toxicity, and their diet consists exclusively of mother's milk, the chapter concludes with information on the dangers of toxic minerals in milk.

2.3 LEAD (Pb)

Lead is one of the most toxic metals with bioaccumulative features (Dorea, 2004). It has four isotopes: ^{204}Pb, ^{206}Pb, ^{207}Pb, ^{208}Pb and one common radioactive isotope which ^{202}Pb. Lead is formed in the earth by the decay of uranium and thorium (Iyer *et al*., 1999).

Lead is mined in form of galena (lead ore (PbS)), which is crystalline in structure with a metallic lustre. The occurrence of lead-zinc deposits are found in many places in South Africa such as the deposit in Pering mine in the North West Province and Leeuwbosch lead deposits, north of Thabazimbi in Limpopo Province (Cairncross, 2005). Figures 2.2 and 2.3 show some lead ore crystals found in some Zeerust district of North west Province and Waterberg region of Limpopo province in South Africa.

Fig. 2.2 Galena Crystals on matrix from the Zeerust in North West province

Fig 2.3 Galena Crystals from Waterberg district in Limpopo Province

(Source: Cairncross, 2005)

Lead is a pervasive and widely distributed pollutant with no beneficial biological roles (Swarup *et al*., 2005). Its negative effects range from slight biochemical or physiological disorders to serious pathological conditions in which organs and systems can be damaged or their function altered according to the degree of exposure (Francisco *et al.*, 2003). Most of the lead accumulated in humans is deposited in the bone which is a long term storage site with half life of approximately 20 years (Steenhout, 1982). Lead (Pb) affects mainly the nervous system especially the central nervous system (CNS) (Dorea & Donangelo, 2006).

Lead is distributed into the environment in various ways such as through crust weathering, industrial emissions, motor vehicle emissions and paint pigments. Lead contamination can occur naturally through the dissolution of lead from soil and earth crust. The average lead content in crustal rocks is about 16 mg/kg. The most common sedimentary rocks, shale and mudstone, have an average lead content of 23 mg/kg (Alloway, 1995).

In the United States, about two thirds of the houses built before 1940 are painted with lead-based paint and those built between 1940 and 1960, half were painted with lead-based paint (http://www.detoxamin.com/otherprod.html, 2009). Lead can also enter the drinking water through plumbing materials like the lead-water pipes, copper pipes with lead solder, and brass faucets (http://www.epa.gov/opptintr/lead). It was postulated that lead toxicity was one of the factors that led to the fall of the ancient Roman Empire because many of the wealthy and people in government could afford to drink wine. The wine in those days was usually enhanced by grape syrup called “sapa” which is simmered in a lead-lined kettle and mixed with the wine at the concentration of 250-1000 milligram per liter. This is an extremely high concentration, considering the fact that a teaspoonful of “Sapa” is able to induce lead poisoning if coupled with taking water from lead water pipes. This enormous consumption of lead was suggested to have affected the coordination of the CNS and hence to decrease decision-making capabilities (Thutmoose, 2002).

Figure 2.4 shows the pathways of lead in the environment, i.e. through the soil, water and ambient air into the plants, animals, drinking water and humans. In humans, the contaminant moves via the blood to soft tissue, the liver, kidneys and bones, eventually reaching all parts of the body.

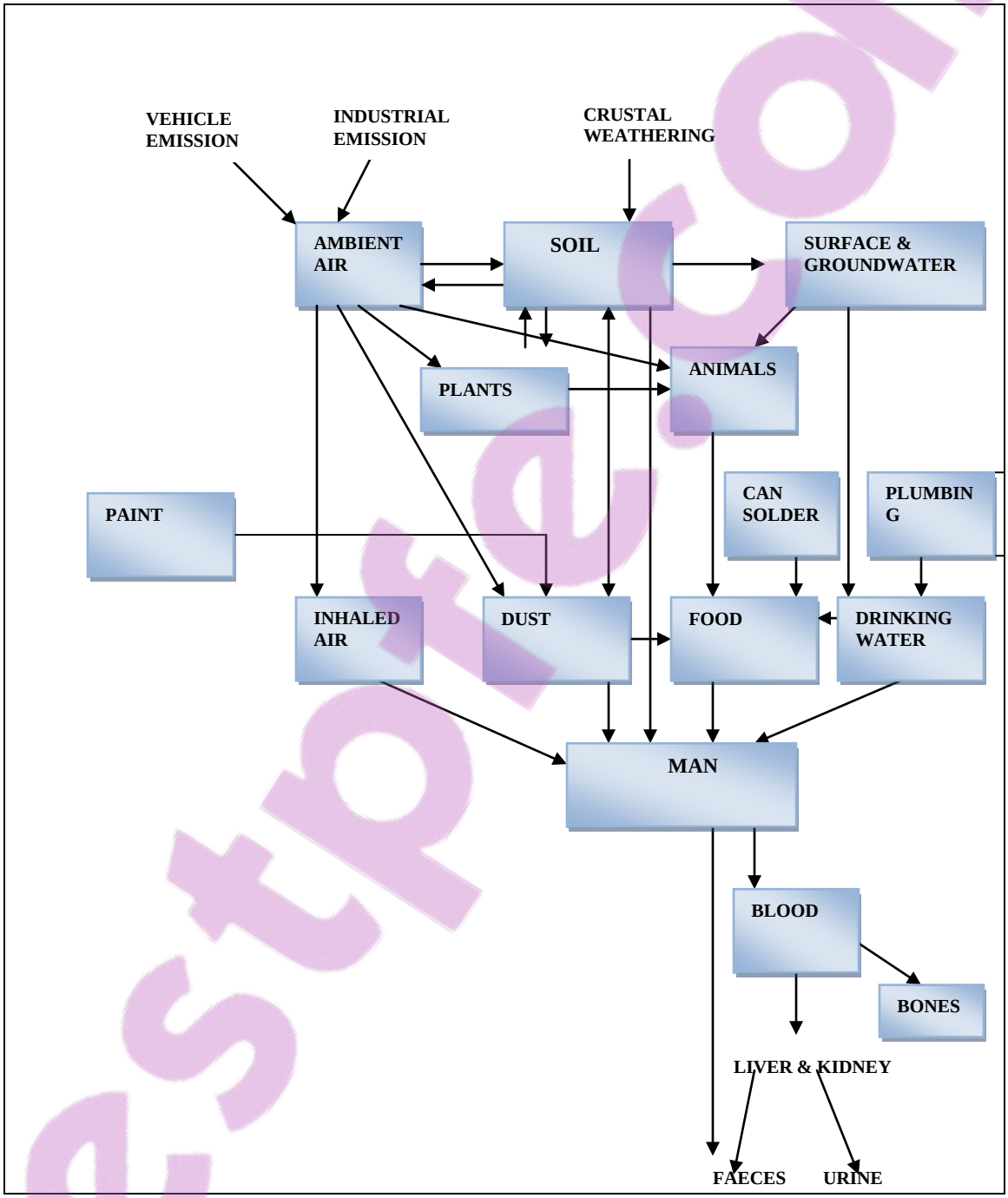

Fig. 2.4. The pathways of lead from the environment to man and body disposition of lead (Elias, 1985).

2.3.1 Lead (Pb) in plants

Urban pollution and industrial activities influence the lead content of foods as a result of air pollution. Foods can also become contaminated with lead from fertilizers which are used in the growing of the plants (Raszyk *et al*., 1996; Moline *et al.*, 2000). In humans, it was estimated that foods and drinking water account for approximately 98% of lead acquisition for those who are not exposed to lead as a result of their occupation (WHO, 1996).

Plants seldom absorb or accumulate lead. However, this could take place in soils with high lead concentration. It is considered safe to use soil with lead providing that is less than 500 ppm (Gray Environmental Inc., 2002).

2.3.2 Lead in animals

An investigation on heavy metals in polymetal dust waste products of lead (Pb) and zinc (Zn) on rodents showed both elements accumulate over time in the liver, kidney of the rodents. A high percentage of aberrant metaphases were found in the bone marrow cells which is an indication of genetic deformation of the cells (Topashka, 1993; Metcheva *et al.*, 1994).

Lead poisoning is more common in farm ruminants than in other animals (Radostits *et al.*, 2000). Animals obtain lead from the soil, water, feeds and fodders (Kohferova & Korenekova, 1995; Dwivedi *et al.*, 2001).

2.3.3 Clinical effects of lead contamination

Clinical effects are signs and symptoms of a disease that are visible and recognizable in both animals and humans. Some diseases may exhibit clinical symptoms whereas others may be sub-clinical, i.e. without any recognizable symptoms. Many of the early stages of diseases are sub-clinical (http//www.your dictionary.com, 2008).

Lead has clinical effects on human. It is a neurotic with longer lasting effects than mercury because it is stored in the bones (Parga *et al.*, 2006). Though stored in the bone,

lead could leak continuously into the blood stream leading to health hazards which may include mental deficiency in children. This particular symptom is more common in children because the barrier between their blood and the brain is not well developed (Pueschel *et al.*, 1996). Children's developing brains are thus most vulnerable to lead exposure. Lead (Pb) can also lead to brain dysfunction in adults (Clarkson, 1987; Carton, 1988; Ling-Chu *et al.*, 2006). A high percentage of the surviving children of mothers exposed to Pb contamination suffer seizures and show signs of mental retardation and anemia. (Clarkson, 1987; Carton, 1988; Mushak, 1991, Pueschel *et al.*, 1996). Lead can also lead to movement disorders, kidney dysfunction and abnormal perception during fetal growth and development. Reports show as well that prenatal and postnatal lead exposure also predisposes children to dental caries (Larkin, 1997).

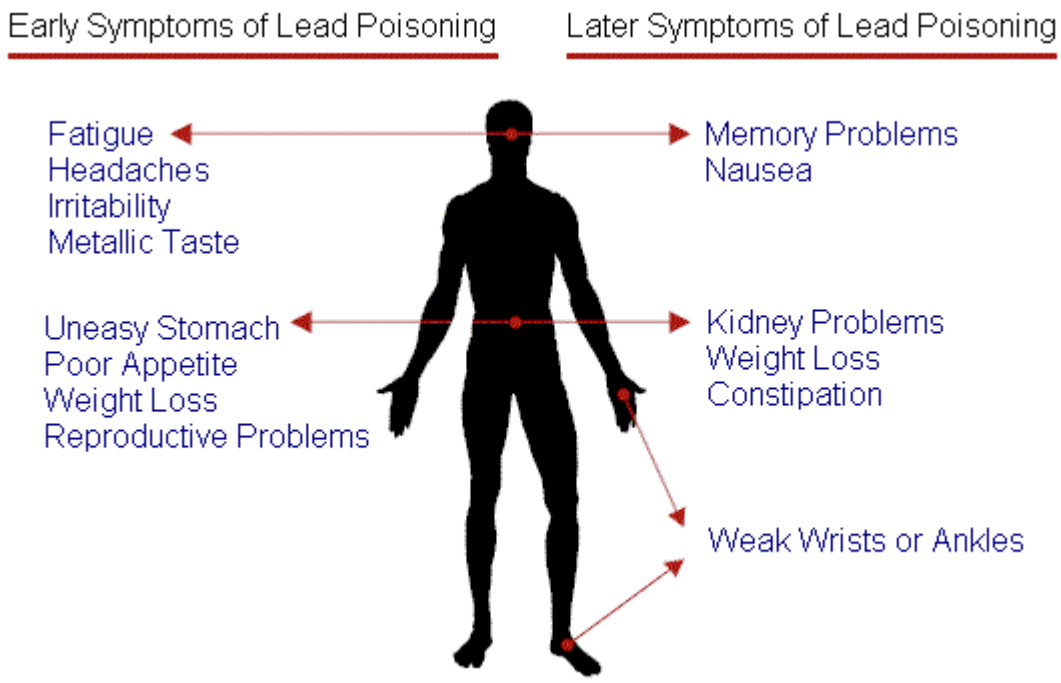

Fig 2.5 Early and late symptoms of lead poisoning at their target areas

(http://www.detoxamin.com/otherprod.html:2009)

Ethylene Diamine Tetra-acetic Acid which is also known as EDTA is one of the chelating devices or chemicals that can be used in the removal of lead from the body by attaching itself to the lead. This is used world-wide as an effective treatment for lead poisoning (http://www.detoxamin.com/otherprod.html, 2009).

2.4 MERCURY (Hg)

Mercury is a liquid metal with density of 13.596g/cm^3. It forms a rhombohedra crystal at -40^oC. It is usually found as small isolated drops associated with cinnabar, but it can also be found as large fluid masses in rock cavities. Mercury is related to vast minerals of which lead amalgam ($HgPb_2$) and potarite (PbHg) form part. Lead amalgam is a tetragonal, grey-white mineral with metallic luster while potarite is a tetragonal silver-white colored mineral (Weissman, 2007).

Like other toxic heavy metals, mercury is emitted into the environment from a number of natural as well as anthropogenic sources. It occurs naturally at trace levels in the environment by tillage of the earth's crust (Cava *et al.*, 2004).

Mining activities are one of the main anthropogenic sources of human exposure to mercury (especially inorganic mercury) (Berzas *et al.*, 2003). Gold mining emits elemental mercury vapor that may be inhaled and absorbed into the blood stream (Grandjean *et al.*, 1999). Sources of anthropogenic emission of mercury also include smelting and refining of metals.

The three most important chemical forms of mercury that occur in the environment are: elemental mercury [Hg(O)], inorganic mercury (Hg^{2+}) and methyl mercury (CH_3Hg^+). The most toxic is methyl mercury (WHO, 1991). Inorganic mercury is the most common form that is present in rain water which is deposited into the rivers. It is not considered to be harmful at low levels because it is easily excreted by animals and thus not bioaccumulated. However, kidney damage may result from exposure to high concentrations for protracted periods.

(http://people.uwec.edu/piercech/Hg/mercury_water/ drinkingwater.htm).

Figure 2.6 shows the transport pathway of mercury in the environment. Mercury in fossil fuels can be converted into a vapour through industrial activities, releasing elemental mercury vapour into the atmosphere. The elemental mercury can undergo photochemical

oxidation to form inorganic mercury which can be released into rivers or the oceans where it may settle as sediment or be converted into methyl mercury by marine or aquatic bacteria. The direct or indirect consumption of such water becomes a major supply of methyl mercury, both to the animals and water plants. Methyl mercury converting-bacteria may in turn be consumed by fish or they may adhere to the plankton, which is eaten by the fish. The fish may progressively be eaten by bigger fish until they are finally consumed by higher animals or humans.

(http://people.uwec.edu/piercech/Hg/mercury_water/drinkingwater. htm, 2008).

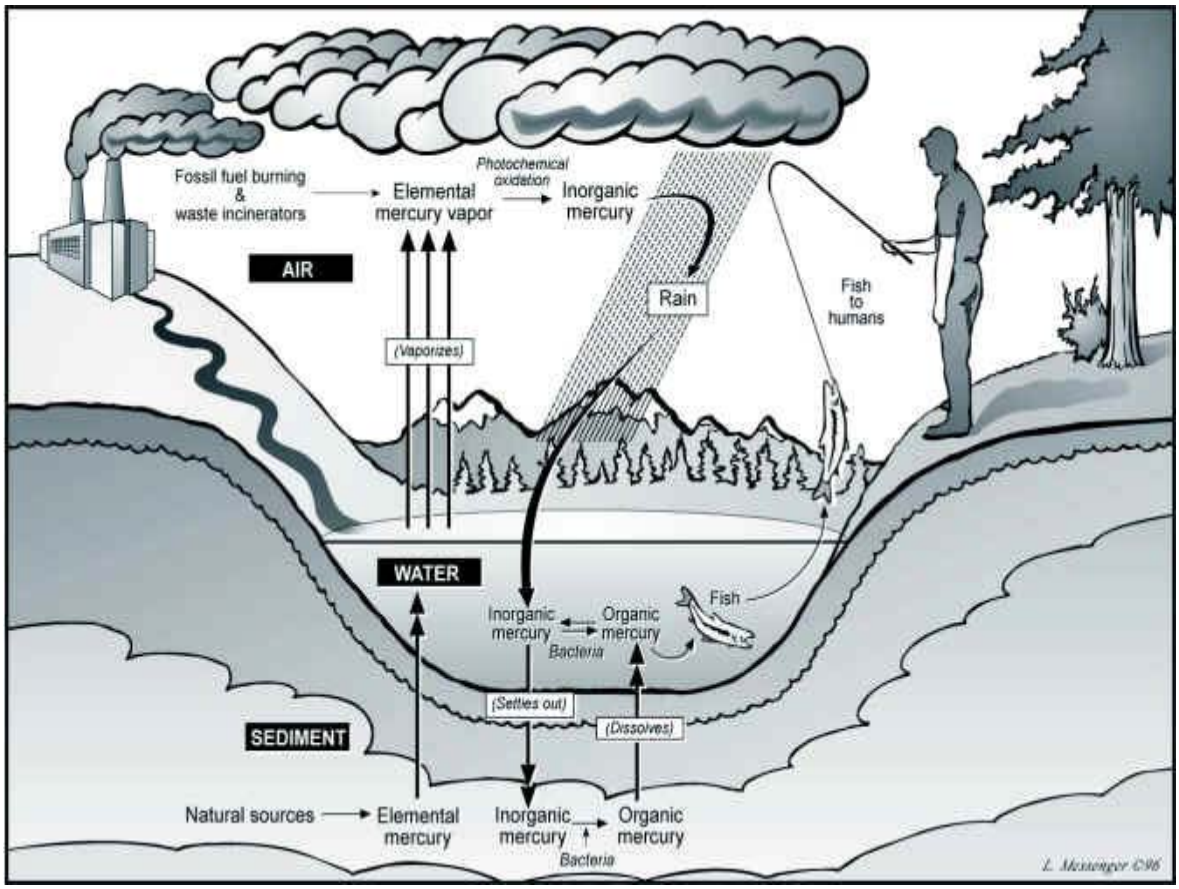

Fig.2.6 The mercury cycle (*http://people.uwec.edu/piercech/Hg/mercury_water/cycling: 2008).*

Various transport and scavenging processes return the majority of airborne mercury to the terrestrial environment close to the pollution source (Lindberg, *et al.*, 1979; Baeyens *et al.*, 1996). Deposition of the element in the surface soils and its re-emission into the atmosphere may induce elevated levels on and in plants. Surface contamination of plant leaves is particularly severe in areas located near mercury mine dumps and when herbivores (goats, sheep and cattle) consume the leaves, mercury contamination will be passed on through the food chain to humans.

2.4.1 Mercury in plants

Accumulation of mercury in plants has been studied by many authors; *inter alia*, Lodenius (1980), Weaver *et al.*, (1984), Kioski *et al.*, (1988) and Banasova & Halub (1999). They found that mercury uptake and its redistribution in plants vary from one plant to the other (Matt, 2005). The translocation of mercury from the root of the plant to the shoot and leaves is very slow and hence the metal tends to accumulate in the roots (Matt, 2005; Beauford *et al*., 2008). Studies by Gothberg *et al*., (2004) showed that the amount of mercury taken is dependent on the ratio of the concentration of the soil nutrient to the mercury in the growing medium (soil). The lower the concentration of the soil nutrients in the growing medium, the higher the metal accumulation in the plant.

2.4.2 Mercury in animals

Experiments conducted by Gnamus & Horvat (1999) indicated that the tissues of herbivores reared in a mercury active mining environment may contain up to 350 times higher concentration of mercury than those that were reared in a mercury-free environment. In addition, tissues of old animals mostly contain higher mercury levels compared to the corresponding tissues of young ones.

Marine and aquatic animals living in water bodies polluted with mercury usually contain the organic form of mercury, namely methyl mercury. Fishing communities are exposed to the mercury contamination through the marine food sources they consume (Franco *et al.*, 2006).

2.4.3 Clinical effects of mercury contaminations

Mercury is an important environmental toxicant that causes neurological and developmental impairment in both humans and animals (Jefferson *et al*., 2006). Inorganic mercury is toxic to the renal, reproductive and nervous systems (Frumkin, *et al.*, 2001). Methyl mercury is readily absorbed by the gut and easily crosses the blood – brain barrier and placenta (JECFA, 2003). This organic mercury has the capability of reaching high levels in the central nervous system (CNS) which inevitably becomes dangerous to the system (Aschner & Clarkson, 1988, Clarkson *et al*., 2003). Mercury has also been shown to alter calcium and glutamate homeostasis and cause oxidative stress in the cells (Ou *et al.*, 1999, Aschner *et al.*, 2000; Hansen *et al.*, 2006).

Some studies have shown that the exposure of pregnant women to methyl mercury can lead to direct embryo and fetal intoxication (Harada, 1995) and that prenatal exposure causes neurologically deficits in the offspring (Grandjean *et al*., 1999). Matsumoto *et al*., (1965) documented two cases of exposure of pregnant women to mercury in the Minamata Bay incident, where the mothers exhibited no clinical observable effects from their exposure to mercury, but their infants developed cerebral palsy. This seems to indicate that infants are more susceptible to the effects of toxic minerals. The aged are also susceptible to mercury poisoning, leading to decreased functioning to the central nervous system and the impairment of cognitive abilities over time (Dorea & Donangelo, 2006).

Mercury can be removed from mercury-contaminated water by coagulation/filtration, granular activated carbon, lime softening, and reverse osmosis.

2.5 THE ENVIRONMENTAL AND HEALTH IMPACT OF ARSENIC

Arsenic is the most widespread natural occurring toxic element. It is also one of the most toxic substances in the environment. Even small amounts may cause a variety of diseases, including cancer and may eventually lead to death (Toens *et al.*, 1998). According to DWAF (1996), concentrations of more than 1000 ug/l of arsenic in water could result in cancer or death in human beings.

Arsenic is a semi metal (metalloid) that occurs as part of over 200 compounds, including arsenides, arsenates, arsenites, oxides, silicates, sulphides and sulfosalts (Baur, & Onishi, 1978). It is widely distributed throughout the world (Sami & Druzynski, 2003). The average concentration of arsenic in the earth's crust is 2 parts per million, in fresh water less than 1 ug/l (ppb), and in sea water is approximately 4 ug/l (DWAF, 1996a). Arsenic has a high affinity to some metals such as gold, copper, nickel and silver and may occur in higher concentrations in areas where such metal ores are found.

Exposure to arsenic may come from both natural and anthropogenic activities which contaminate the water. It may also be released through the mining and smelting processes (Crout *et al.*, 2004).

2.5.1 Arsenic (As) in water

The contamination of groundwater by arsenic has been reported in various countries in different parts of the world. As many as 200,000 people were estimated to have been poisoned in West Bengal (India) alone and several cases were reported in Bangladesh, where geothermal activities in the Taupo volcanic zone elevated arsenic concentration in some lakes and rivers (Aggette & Spell, 1978). Groundwater As contamination has also been reported in Nepal (Tandukar *et al.*, 2001; Shresther *et al.*, 2003) and from Vietnam, where several million people consuming untreated groundwater run a considerable risk of chronic arsenic poisoning (Berg *et al*., 2001). The occurrence of arsenic in groundwater of La Comarca Lagunera in Mexico is predominantly as a result of the mineral dissolving from natural weathered rocks and soils which produce high arsenic levels in well water

(Parga, *et al.,* 2005). In Ontario, Canada, Wyllie (1937) reported that water from some deep wells contain large amount of arsenic. According to Jacobson (1998), arsenic poisoning (arsenicosis) has become the world's biggest environmental health disaster.

Table 2.1 below lists some of the international areas that have reported groundwater arsenic contamination.

TABLE 2.1 REPORTED INCIDENTS OF ARSENIC CONTAMINATION IN GROUNDWATER

LOCATION	YEAR	ARSENIC SOURCE	CONCENTRATION (µg/ℓ)
Ontario, Canada	1937	Well water	100-410
Hungary	1941-1981	Well water	60-4000
Monmte Quemado, Cordoba, Argentina	1955	Well water	100
Antofagasta, Chile	1959-1970	River water	800 (maximum)
South west Taiwan	1961-1985	Well water	10-1820
Langunera region, Northern Mexico	1963-1983	Well water	8-624
Lane county, Oregon	1962	Well water	50-1700
Halifax, Nova Scotia, Canada	1976	Well water	>3000
Xing- Jiang, PR China	1980	Well water	850 (maximum)
West Bengal, India	1983	*Tube well water	<3-3800
Bangladesh	1995	*Tube well water	<3-4730
Nepal	2001	*Tube well water	2620(maximum)
Vietnam	2001	*Tube well water	1-3050

(Source: Rahman *et al.*, 2001*).*

**A Tube well is type of water well in which a long 12.7 to 20.3 cm wide stainless steel tube is inserted in the well. This tube is then extended to the underground water below the lower surface of the well. http://en.wikipedia.org/wiki/Tube_well*

2.5.2 Arsenic in plants and crops

Arsenic in its soluble form is easily taken up by plants. Crops take up toxic elements through their roots from contaminated soil and even leaves can absorb toxic metals deposited on their surfaces. It increases in plants with increasing concentration in the growing medium. However, crops accumulate metals to different extents, depending upon the tissue and plant part (Lepp, 1981, Data *et al*., 2000). For example, a survey of watercress growing in a groundwater-contaminated region revealed arsenic concentrations that were 2 mg/kg above the World Health organization (WHO) limit for arsenic in foodstuffs (Brandsetter *et al*., 2001).

Plant accumulation of arsenic may facilitate the entry of this toxic element into the food chain. Studies by (Queirolo *et al*., 2000) have indicated that crops grown in Talabre (northern Chile) contains very high arsenic concentration in the edible parts

Figure 2.7 describes the pathways by which humans can ingest arsenic. These pathways occur from the soil to soil water to the plants. The animals feed on plant and drink ground water which is contaminated. When they are consumed, the arsenic is transferred to humans. Another pathway could be via the water that runs off to the river and sea. The water contaminates the ecosystem of the fish and sea foods. They are later eaten by humans thus acquiring some of the arsenic in the animals. Humans can also ingest some of these contaminations directly through drinking water and the use of contaminated water for cooking.

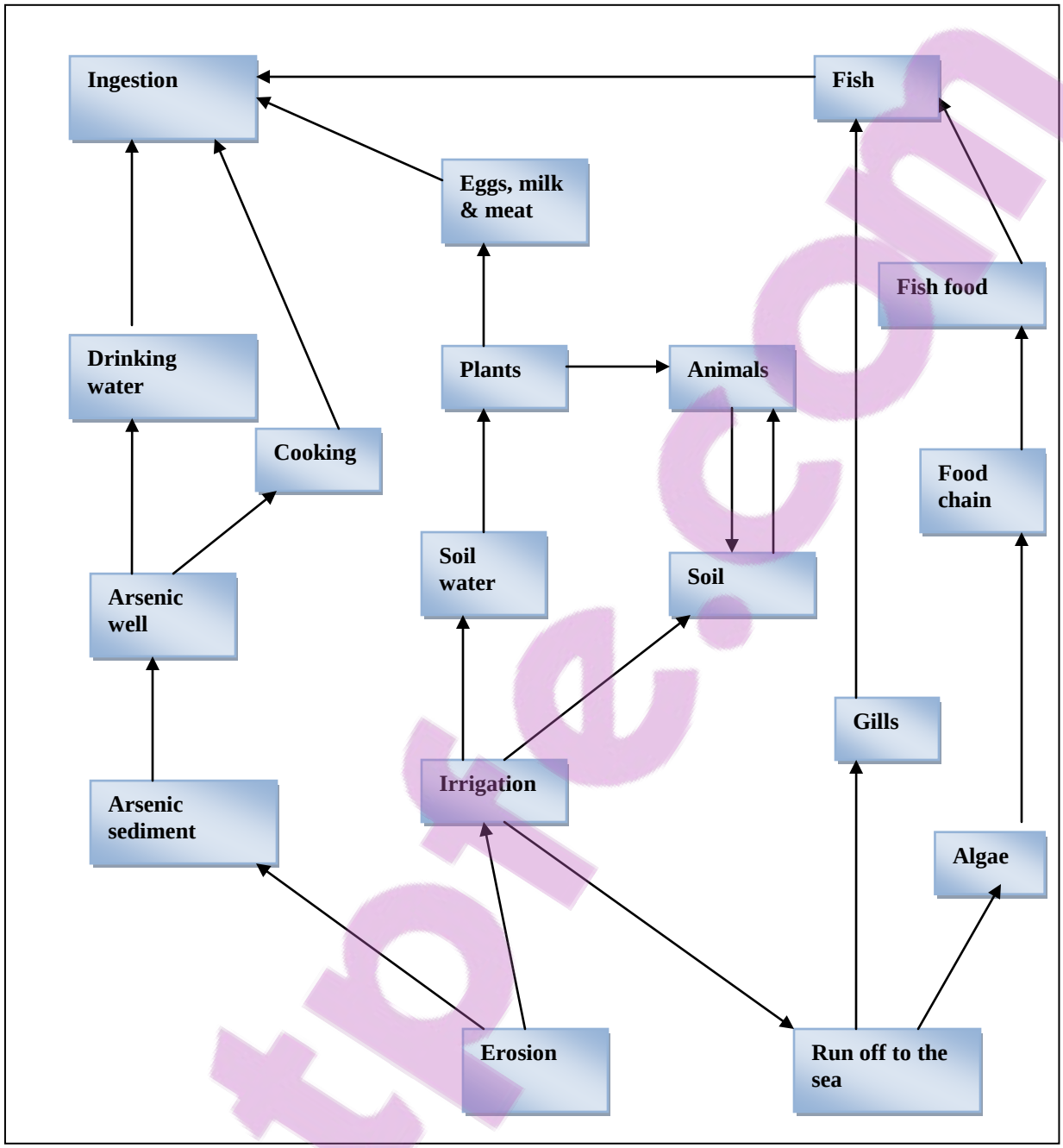

Fig 2.7 Possible pathways arsenic ingestion by humans (Correll et al., 2004)

2.5.3 Clinical effects of arsenic contaminations

Arsenic, which is absorbed by living organisms, is excreted very slowly and hence accumulation occurs easily (Toens *et al.*, 2005). Arsenic poisoning can be both chronic and acute. Chronic poisoning is characterized by skin lesions including hyper

pigmentation and cancer while acute poisoning may result in death from upper respiratory, pulmonary, gastro-intestinal and cardiovascular failure (DWAF, 1996a). Nerve damage, characterized initially by a sensory loss in the peripheral nervous system is a primary symptom of arsenic poisoning. Major clinical symptoms of arsenic contamination observed in Inner Mongolia, Argentina, Chile, India, Taiwan and Thailand include pigmentation melanosis, leucoderma and cutaneous skin lesions, often accompanied with peripheral neuritis, gastroenteritis, hypertrophy of the liver, bronchitis and cardiac infarction (Schuurmann & Markert, 1997). Epidemiological studies have shown that the cardio vascular system is particularly sensitive to exposure through long term ingestion of arsenic in drinking water. Noticeable effects include hypertension and increased cardiovascular disease mortality (NRC, 2001).

Characteristic skin lesions of arsenic toxicity may be used as an indicator of high exposure and are distinctive in contrast to other clinical manifestation of arsenic intoxication. At later stages skin cancer and gangrene may occur, especially in people who have been drinking arsenic contaminated water for more than 10 years (Astolfi *et al.*, 1981; Gao, 1997; Xiau, 1997; Mazunder *et al.*, 1998; Turker *et al.*, 2001, Parga *et al.*, 2006). Dermatological manifestations of arsenic poisoning were noted especially in children in Antofagasta in Chile (Borgono & Greiber, 1971). Peripheral vascular manifestation of arsenic contamination in these children includes Raynaud's syndrome, ischemia (the localized tissue anemia due to obstruction of the inflow of arterial blood) of the tongue and hemiplegia, which is the unilateral paralysis of the body that results from injury to the motor centers of the brain with partial occlusion of the carotid artery, mesenteric arterial thrombosis and myocardial ischemia. One death from arsenic dermatitis has been reported from Ontario, Canada (Wyllie, 1937).

An incremental trend in the rate of spontaneous abortion was observed with an increasing occupational and residential exposure to arsenic. Congenital malformations also appear to be more frequent if expectant mothers are employed in areas exposed to arsenic (Nordstorm *et al*., 1979).

Figures 2.8, 2.9 and 2.10 show some examples of the effect of arsenic poisoning.

Fig 2.8 Gangrene patient from West Bengal. (Schuurmann & Markert, 1997).

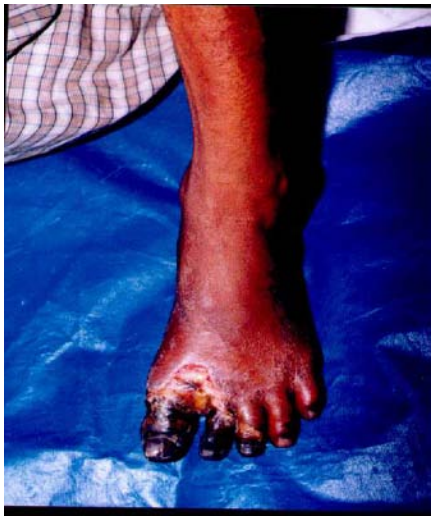

Fig 2.9 Gangrene in the foot

http://phys4.harvard.edu/~wilson/arsenic/pictures/arsenic_project_pictures2.html

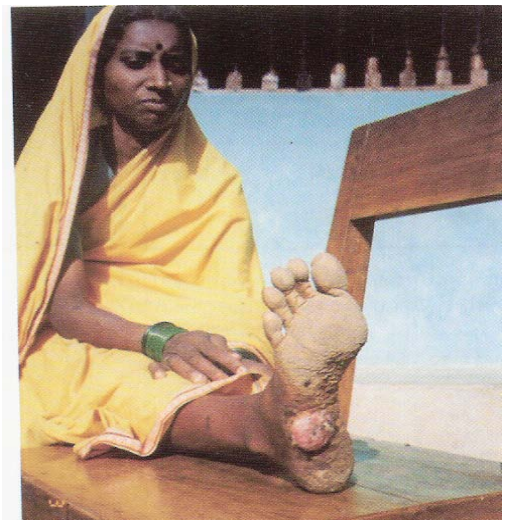

Fig 2.10Patient with a non healing ulcer (Schuurmann & Markert, 1997).

Table 2.2 shows the medical effects of different levels of arsenic contamination in water. According to the table, the range of 0-10 μg/l can be tolerated by humans without any health risks. Any concentration above this could result into various diseases ranging from skin cancer to death.

TABLE 2.2: EFFECTS OF ARSENIC RANGE ON HUMAN HEALTH

Arsenic range (ug/l)	**Effects**
Target water quality range 0-10	No health effects expected; ideal concentration range
10-200	Tolerable concentration but low risk of skin cancer in highly sensitive individuals over a long term
200-300	Increasing possibility of mild skin lesion over long term. Slight possibility of induction of skin cancer over
300-600	Possibility adverse, chronic effects in sensitive individuals; brief exposure has no effect; skin lesions, including hyper pigmentation, will appear on long term exposure
600-1000	Symptoms of chronic poisoning such as skin lesions including hyper pigmentation, will appear on long term exposure
1000-10, 000	Cancer or death will result from chronic poisoning
>10, 000	Death will result from acute poisoning

(South African water quality Vol .1 Domestic use (Department of Water Affairs and Forestry, 1996b)

Arsenic is most effectively removed from water in its pentavalent form using the process of coagulation and flocculation followed by settlement and filtration. Some other technologies include the removal of arsenic from ground water by ion exchange resin, reverse osmosis, co-precipitation and various membrane techniques (DWAF, 1996; Parga *et al.*, 2006). It occurs in three oxidation states, namely, (0), (III) and (V). In solution, arsenic can exist as arsenite, As (III) and arsenate, As(V); also as various organic complexes. The trivalent form arsenite is first converted to pentavalent form using an appropriate oxidizing agent such as chlorine or potassium permanganate (DWAF, 1996).

All other cases of chronic arsenic toxicity - barring arsenic-induced cancer - can be reversed by discontinuing the exposure to contaminated water. Acute toxicity may be reversed by administering chelation therapy using 2, 3- dimercaptopropanol (Toens *et al.*, 1998).

2.6 MILK AS A SOURCE OF CONTAMINATION

Milk is an excretion of the mammary glands and serves as the main source of nourishment for infants (Licatta *et al.*, 2003). The determination of the residual concentration of the metals in milk could be important direct indicator of the hygienic status of the milk and/or of its derived products, as well as an indirect indicator of the degree of the pollution of the environment in which the milk is produced.

Several studies have been conducted on the toxic minerals contamination in milk (Maggi *et al.*, 1975; Feller, 1983; Gregorio & Saracusano, 1997). The results differ according to the various sampling areas and the analytical method employed.

Studies have shown that toxic minerals in animal feed may be transferred to their milk (Alonso *et al.*, 2000). Various studies from Italian regions have reported high levels of cadmium and lead in milk. In some cases, processed milk product contained higher amounts of toxins than the fresh milk (Iyengar *et al.*, 1982; Jorhem *et al.*, 1991).

The consumption of milk that has been contaminated by the toxic minerals may constitute a significant source of toxic mineral in human diet (Mehennaoui *et al.*, 1999).

2.6.1 Lead (Pb) in milk

The level of lead in milk obtained from animals exposed to the environmental pollutant is a serious public health concern in India (Swarup *et al.*, 2005). A linear dose-related excretion of lead from plasma to milk was found in mice after intravenous injections of lead. However, 24 hrs after administration, the lead concentration in milk was found to be higher than that in the blood (Hallen, 1995).

A study conducted in Mexico City confirms that lead may be transferred to babies via mother's milk. Ryu *et al.* (1985) and Ettinger (2004) found a correlation between the lead concentration in the breast milk of lactating mothers and the corresponding blood lead of infants. This was already evident after only one month.

2.6.2 Mercury in milk

Experimental studies have shown that both inorganic and organic forms of mercury can leach into milk (Oskarsson *et al.*, 1996; Manfroi *et al.,* 2004; Franco *et al.*, 2006). All chemical forms of mercury can cross the placental barrier and be secreted into the milk. It is thus important to test for and control the presence of mercury in milk (Klassen *et al.*, 1999)

High amounts of mercury in human milk are attributed to both the presence of amalgam fillings in mothers' teeth and to fish consumption (Oskarsson *et al.*, 1996). Methyl mercury bioaccumulates in fish. It has been found that in communities with a high intake of sea foods, levels of 3-7 ngHg/g in blood (Galster, 1976; Skerfving, 1988). Moreover, fish meal is often given to animals in their feed. There is thus a high probability that methyl mercury may be transferred to the milk of animals that are fed with the Hg-contaminated feedstuffs (Dorea, 2004). The presence of inorganic mercury in breast milk could also originate from demethylation of methyl mercury in the body (WHO, 1990).

2.6.3 Arsenic in milk

It is known that arsenic crosses the barrier of the placenta easily thus exposing the foetus to arsenic contamination during pregnancy. There is thus a possibility of the transfer of arsenic from the mother's body to the milk to the mammary glands. However, conflicting results have been found.

On the one hand, Rana *et al.* (1996) found that the concentration of arsenic in the milk of cows reared in the areas contaminated with arsenic was 12 times higher than the milk of

the cows in the areas that were not contaminated with arsenic. Furthermore, 100 deaths were recorded among the Japanese infants in 1955 due to arsenic poisoning from powdered milk with an arsenic concentration of 4-7 mg/l (Dakeishi *et al*., 2006).

On the other hand Fängström, *et al.* (2008) found that mothers that are exposed to high arsenic concentration had a low concentration of arsenic in their breast milk. Also, in the Argentine, the indigenous women that are exposed to about 200 μg/l arsenic in the drinking water showed very low excretion in breast milk (~ 3 μg/l) (Concha *et al.*, 1998b). In Munster (Germany), a war zone with high arsenic contamination, it was discovered that the level of the arsenic in the mothers who are highly exposed to arsenic is lower than the WHO (1993) lower limit of the daily permissible intakes for adult of 15 μg/kg/wk (Sternowsky *et al*., 2002**).**

These finding suggest that the secretion of arsenic from the lactating mothers to their milk is not to any significant level. Tillette (2008) postulated that the noticeable increase in arsenic urine concentrations of infants with mothers that had low breast milk arsenic concentration might be from the water they are given rather from milk.

CHAPTER 3

STUDY AREA AND METHODOLOGY

South Africa is a country rich in minerals and Limpopo province is one of the provinces with the widest range of minerals deposits (Citizen, 2004). Due to the large number of these deposits, there is a high possibility of groundwater contamination in Limpopo province and this is one of the factors which prompted the choice of the province as a study area. The majority of the province comprises of rural areas where groundwater is the only source of water for plants, animals and humans.

This chapter describes the methodology by which the research was conducted. The study comprised of three objectives. The methods used to achieve each are discussed in the sections below.

3.1 OBJECTIVE 1: TO IDENTIFY POTENTIALLY CONTAMINATED AREAS IN LIMPOPO PROVINCE.

This objective was achieved in two stages, as outlined in sections 3.1.1 and 3.1.2.

3.1.1 Step 1: Preliminary identification of study areas

The first step was to identify the broad areas where As, Hg and Pb contamination may occur based on geological characteristics. A geological maps showing the spatial distribution of minerals and the location of mines in Limpopo is presented as Figure 3.1.

Fig 3.1 Mineral map of Limpopo Province (source: www.limpopo.gov.za)

According to Fig. 3.1, mineral deposits are widespread throughout the Limpopo Province with clusters of heavy mineralization occurring in the vicinity of Musina (L1); in an east-west band extending from the south-eastern corner of the Bochum district to Giyani (L2); around Gravelotte and Phalaborwa in the Namakgale districts (L3); around Mokopani (A4); and to the west of Bela-Bela (Warmbaths) (L5).

These five areas represent the possible study areas. Areas where all three toxic elements were located were obvious choices for selection as study areas. Where there was little or no coincidence, research areas were identified using random selection methods.

3.1.1.1 Areas with potential Hg contamination

Careful perusal of Fig. 3.1 shows only one area with mercury-containing deposits. That is the area in the vicinity of Gravelotte.

Gravelotte in area L3 is one of the rural areas in Limpopo that is very rich in minerals. Among the 57 listed minerals found in this area are gold and mercury. The presence of these minerals suggests that there could be the possibility of arsenic and lead contamination. It is thus obvious that Gravelotte should be included as a study area.

3.1.1.2 Areas with potential As contamination.

As mentioned in Chapter 2, arsenic-bearing minerals are primarily associated with geological settings where sulphide mineralization has taken place. Arsenic is usually present in areas where gold, silver, copper, nickel, zinc, lead and cobalt are found (Sami & Druzynski, 2003).

The majority of the areas of heavy mineralization shown in Fig 3.1, with the exception of those near Musina and in the Namakgale districts, coincide with those identified by Sami and Druzynski (2003) as potentially high risk areas (Fig 3.2) using borehole data. Groundwater in these areas showed As concentrations ranging from 0.01 to 0.1 mg/l. The areas so identified included the surrounds of Mokopane (L4), Leeupoort (L5), Vivo (L2), and almost the whole of the Nebo district.

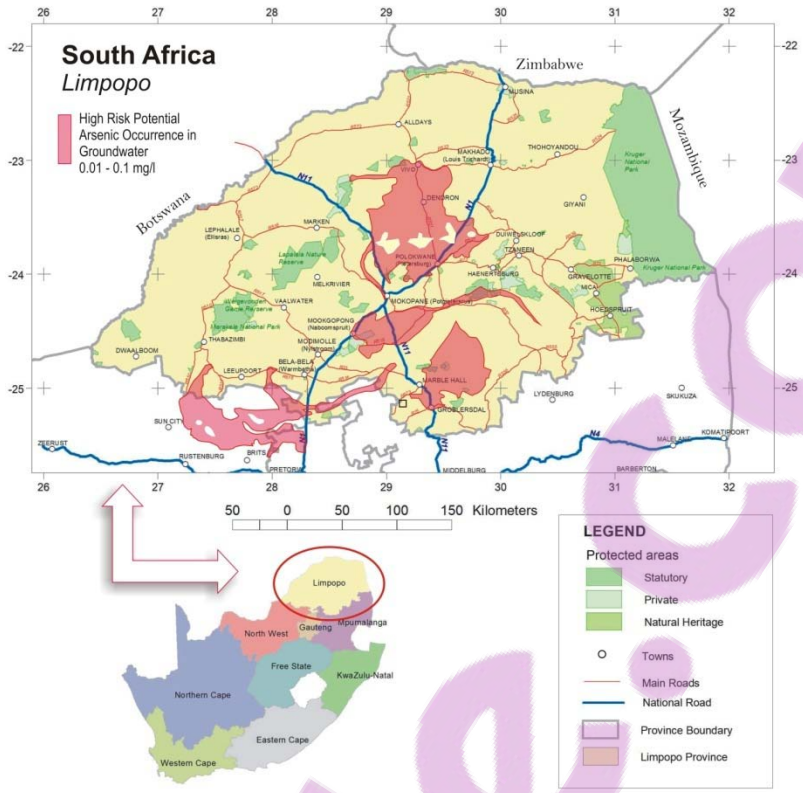

Fig 3.2 Potential arsenic risk regions in Limpopo Province (Sami & Druzynski (2003) / www.place.co.za, 2003)

In order to delineate the second study area, one of the areas shown on Fig 3.2 was selected randomly by assigning numbers to the areas and picking one number out of hat. The region so selected was L1, i.e. the region to the west of Bela-Bela, in the vicinity of Leeupoort.

3.1.1.3 Areas with potential lead contamination

According to Lurie (1977), zinc and lead minerals generally occur together in deposits. Lead ore minerals include Galena (PbS) and cerussite ($PbCO_3$). A number of old mines related to the Bushveld Complex produced Zn and Pb in the past. Numerous lead deposits also occur in Transvaal dolomite.

Perusal of Fig 3.1 indicates the presence of Pb in the area near Gravelotte as well as to the south of Leeupoort. It was therefore assumed that the two previously selected areas would also exhibit some potential for lead contamination.

3.1.2 Step 2: Final selection of study areas

In order to select the specific areas to be studied, the actual villages where the research would be conducted had to be identified. Prerequisites for this selection were that the villagers were dependent upon subsistence farming, that goats were reared in the communities and that goat's milk was consumed.

A geographical grid was superimposed over the selected regions in Fig 3.1. The "Leeupoort" grid occupies the area between 24°37'30" and 25°00'00" South and 27°42'00" to 28°00'00" East (2427DC). This area is henceforth referred to as study area A. The area around Gravelotte lies between the latitudes 23°45'00" to 24°00'00" South and longitudes 30°30'00" to 30°45'00" East (2330DC) (study area B).

In order to select sample villages, all villages in the two study areas were listed. The villages in study area A are: Buffelskraal, Rooiberg, Doornfontein, Leeupoort, Weihoek, Driefontein, Rooikrans, Koperfontein and Ruin, while the villages study area B are Kwagga, Salwana, Malete, Mpageni, Gravelotte, Mashororo, Mawa, Waterbok, Kondowi, Haartebeest, Grootfontein, Mabete, Eden, Thiergarten, Murchison, Bettie, Bosbok, Allonbridge and Leysdorp.

Two villages were selected within each of the study areas by means of random selection. The names of all the villages in the specific area were written on different piece of paper, folded and placed in container. The folded papers were drawn randomly. Two villages were selected for sampling from each study area. The two from study area A were Rooiberg and Leeuport while Gravelotte and Kwagga were selected for study area B.

The final selection was confirmed after site visits to each of the villages to ascertain that those selected actually fulfilled the four criteria namely that:

- There is high potential for contamination of groundwater due to geology.
- Rural communities are dependent on groundwater as main water source.
- Villagers are dependent on subsistence farming.
- Infants are fed on goats' milk once they are weaned from mothers' milk.

All four selected villages (Rooiberg, Leeupoort, Gravelotte and Kwagga) were found to fulfill these criteria and depend exclusively on subsistence farming. Figures 3.3 and 3.4 depict the topographical maps of the study areas showing the location of rural villages.

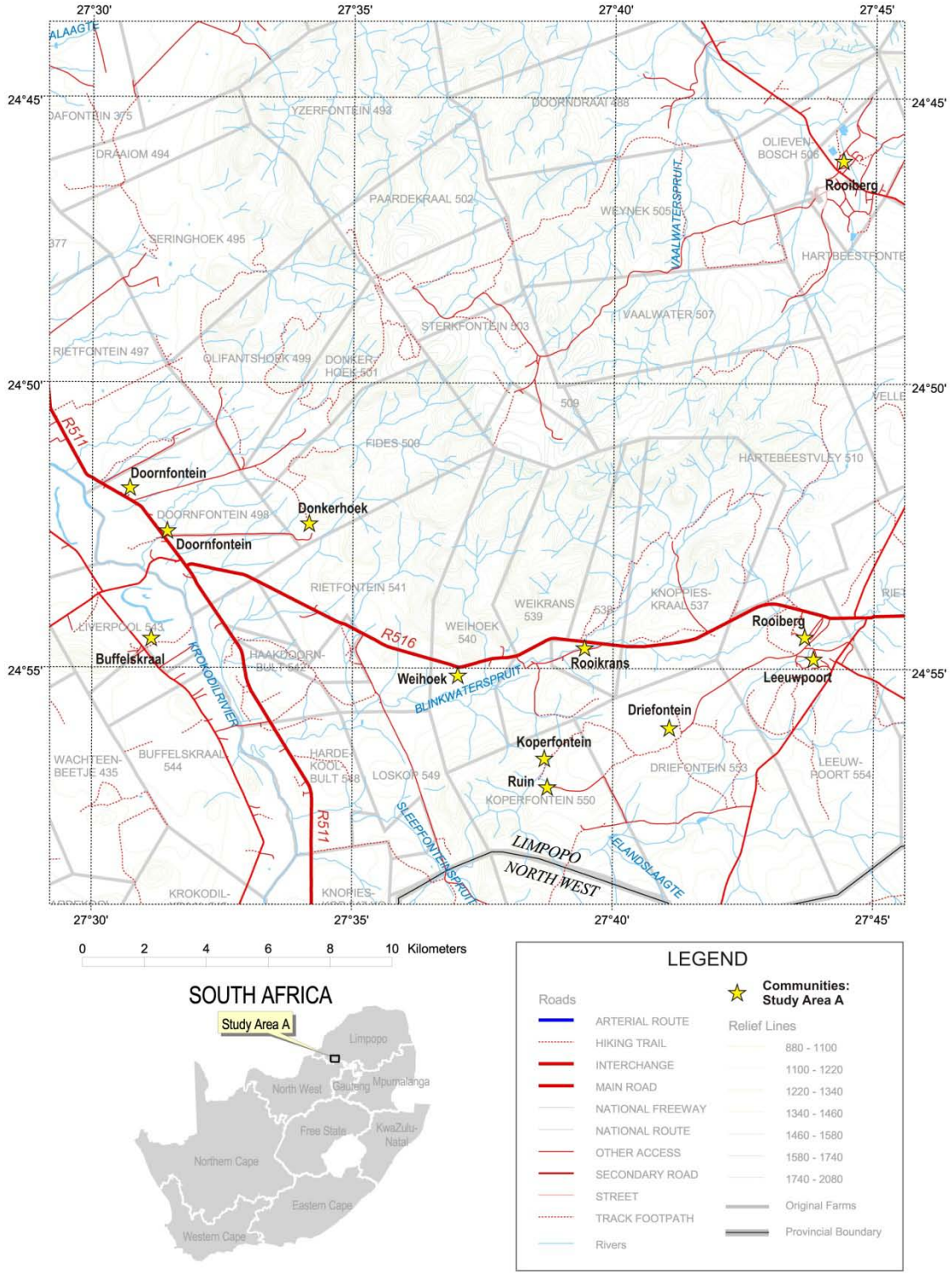

Fig 3.3 The location of rural villages in study area *A (South Africa 1:50 000 Toposheet 2427DC) Chief direct of survey & mapping 1982 (2nd Edition*)

Fig 3.4 Location of rural villages in study area B *(South Africa 1:1 000 000 Toposheet 2330 DC) Chief direct of survey & mapping 1982 (2nd Edition*)

3.1.3 Characteristics of the study areas

Tin mines were located at Rooiberg and Leeupoort. According to Lurie (1977: 94), the Rooiberg tinfield lies about 60 km west of Bela-Bela. In this area mineralization occurs in sediments of the Pretoria Group. Minerals found in the area include tourmaline, cassiterite, carbonates and a variety of sulphides (pyrrhotite, pyrite, chalcopyrite, bismuthinite, sphalerite and galena (Leube & Stumpfl, 1963). The minerals contain gold, copper, iron, tin, tungsten, lead, zinc, molybdenum, bismuth and arsenic (Lurie, 1977). In the 1970s there were three working mines over a curves strike of 20 km at Rooiberg, Nieupoort and Leeupoort. A number of dormant workings are also located in the area.

Gravelotte is located on the Murchison Range, which represents a remnant of Swaziland rocks. It is well known for its rich deposits of antimony and gold. This metal is used for pigments and detonators, matches and flame-proofing chemicals (Lurie, 1977, 83). Currently South Africa is one of the largest producers of antimony.

Antimony and related minerals are found in mineralized quartzites and schists in Archaean granites. The general geological structure consists of steeply folded parallel anticlines and synclines. The rocks form a line of hills running in an ENE direction for about 55 km from Leydsdorp and is known amongst geologists as the ‘Antimony Line’ (Lurie, 1977).

Gravelotte Mines also exploit the gold-bearing reefs of the Witwatersrand and Transvaal Supergroups. Gold mineralization was formed under conditions of partially closed systems, where various mineral-forming processes developed (metasomatism, crystallization of true solutions and gels, gel metasomatism, dispersion of crystalline phases, segregation of mineral particles, formation of early minerals, etc.) (Safonov and Prokof’ev, 2006).

Interestingly, perusal of geological and mineral maps of South Africa (Fig 3.1), indicate that mercury deposits are also found in the vicinity of Gravelotte. This is the only place

where such deposits are indicated. According to Prof Morris Viljoen in the Department of Geology, University of the Witwatersrand (Wits) (pers. Comm.), mercury was mined in the vicinity of Gravelotte during the war, but the mercury mines have subsequently closed down.

3.2 OBJECTIVE 2: TO DESCRIBE THE EXTENT OF ENVIRONMENTAL CONTAMINATION IN THE SELECTED AREAS.

This objective was achieved by collecting and analyzing groundwater, soil and plant samples in the selected areas.

A member of each community who had been working with the local government in these areas over the years was identified as the contact persons in each of the areas. In study area A, Mr. Morris was the contact person (Fig. 3.5) while in area B; Mrs Shishenga was the contact person. The purpose of the research was explained to them and they in turn conveyed it to the community. This method assisted in obtaining the cooperation of the communities.

Fig 3.5 Conversation with Mr. Morris at Rooiberg

3.2.1 Collection of water samples

The major source of water for Leeupoort, Gravelotte and Kwagga are boreholes while at Rooiberg the source of water is a dam.

The samples (water, plants and milk) were collected in plastic containers which had been washed with detergent, rinsed and then re-rinsed again with distilled water to avoid any external contamination.

Plastic containers were used in order to prevent the adherent of the element unto the container which could affect the integrity of the samples.

The first set of water samples were collected in the winter season (July), 2007. These were collected from the dam at Rooiberg and the borehole at Leeupoort, respectively, since these are the major water supplies for domestic purposes in the villages. One liter samples were collected from each village. The water sample was collected at a depth of 10 cm depth from the dam in Rooiberg after the container has been rinsed with the same water from the dam to avoid contamination at about. At Gravelotte and Kwagga water samples are taken from the boreholes that were located outside the villages using the same procedure as that of Leeupoort. The samples were taken to the Agricultural Research Council (ARC), Pretoria, for analysis.

The second set of water samples were taken in the summer time (November, 2007). This was the rainy season and all the water sources were full to capacity. The same procedure as the first collection was adopted for the collection of the second samples. Samples were also analysed at the ARC laboratory, in Pretoria. Fig. 3.7 below shows the dam at Rooiberg and Fig 3.8 illustrates the collection of the water.

Fig 3.6 The dam site at Rooiberg

Fig 3.7 Collection of water samples at Rooiberg

3.2.2 Collection of soil samples

The soil samples were collected from the area surrounding the dam at Rooiberg. Not only is dam water consumed by the locals, but goats forage on the vegetation surrounding the dam. The soils were collected in two different areas. Samples of the top soil plus A and the B horizons to a depth of around 30 cm, were collected and stored in separate bags. The samples were taken to the ARC in Pretoria for analysis.

3.2.3 Collection of plant samples

The commonest plants and shrubs eaten by the goats were identified through the information given by the goats' owners. These plants were collected in the morning in the month of February, 2006 (during summer) by cutting the foliage of the plants. Plant samples were placed in plastic bags, were labeled and were taken to the ARC laboratory for treatment and analysis.

The second set of plant samples were taken in the summer time (November, 2007). This was during the rainy season.

Another set of water, soil and plant samples were collected from Rooiberg during August 2009 using the same procedures.

3.3 OBJECTIVE 3: TO DETERMINE THE EXPOSURE PATHWAY TO HUMANS IN CONTAMINATED AREAS.

In order to achieve this objective, all the sources and pathways had to be identified by which As, Pb and Hg reach infants in the study area.

Fig 1.3 in Chapter 1 indicated that the main sources of contamination of infants by ingestion are dust from the soil, water and food. The collection of water and soil samples has been discussed in the previous section. The following sections describe the collection and analysis of goat's milk and mother's milk. With this information, it is possible to trace the path of contamination through the food chain from groundwater to humans and

thus to determine whether mercury arsenic and lead pose a health hazard to human in the selected parts of Limpopo.

3.3.1 Collection and analysis of goat's milk

In the study areas, goats are reared intensively i.e. they are confined to a specific area. In the mornings, the goats are allowed out of their pens to forage and drink from any available water sources.

All the female goats with kids were numbered and two were randomly selected for milking. The udders of the goats were first washed thoroughly with alcohol and water. With the assistance of the owner of the goats, they were milked. A 20 ml sample of milk was taken from each of the goats. The samples of the milk were taken to the ARC laboratory for analysis.

Figure 3.8 and 3.9 shows the herd from which the milk were taken and the milk of the selected goats respectively.

Fig 3.8 Herds of goats from which samples were taken

Fig 3.9 Milking the selected goat

3.3.2 Human milk sampling

The human milk samples were obtained from the clinic that was closest to the villages and which is attended by the pregnant women. The consent of the mothers was obtained via the village contact person who explained the purpose of the research and the possible environmental benefit for the entire communities to the mothers and the nursing sisters at the clinics. The milk was collected from nursing mothers who had just given birth and who originated from the villages. The breasts of the mothers were washed with water before the extraction of the milk into a plastic container. This was done with the assistance of Dr. Arajuwa Peterson of outpatient department in Thabazimbi district Hospital. The milk sample was taken to the ARC for analysis.

3.4 ANALYSIS OF SAMPLES

Samples were analyzed by the ICWS at the ARC laboratories in Pretoria. The total metal as well as the arsenic, lead and mercury content were determined using inductively coupled plasma source atomic emission spectroscopy (ICP-AES).

The results of the analyses are presented in Chapters 4 and 5.

CHAPTER 4

ENVIRONMENTAL ASSESSMENT

4.1 INTRODUCTION

Elements, either toxic or non-toxic, have a specific way of expressing themselves in nature and follow definite pathways in the ecosystem. A simplified diagrammatic representation of the pathways of elements in the food chain that is relevant to this study is given below (Fig. 4.1).

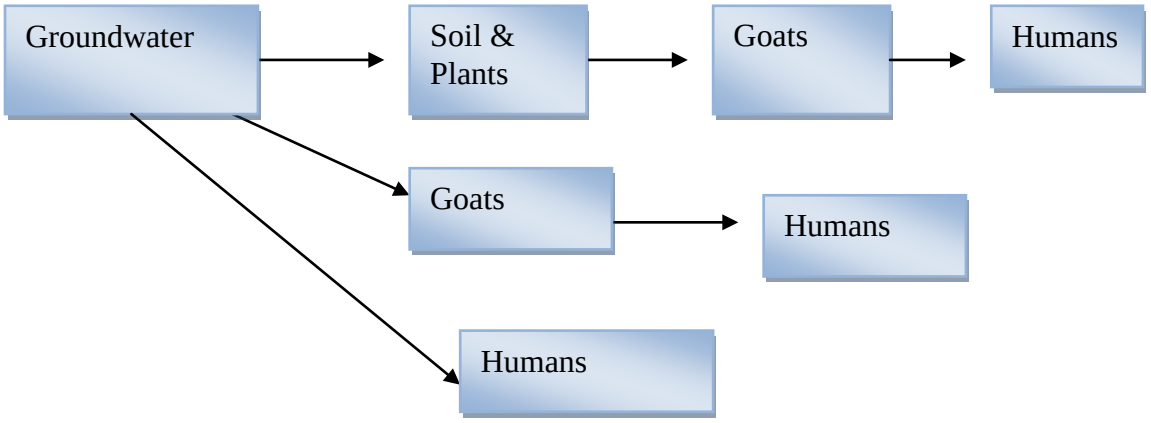

Fig. 4.1 The pathways of elements in the food chain

Water is essential for all living organisms and any contamination of water sources can be transferred to all components in the ecosystem. Similarly, contaminants in the soil may be taken up by plants or may settle on the leaves of plants as dust. Crop plants form the stable diet of the major proportion of the world's population. Leaves of plants also serve as food for animals such as goats and livestock feed, as well as to many insects, which in turn, are eaten by birds. The accumulation of toxic elements in the food chain is, therefore, a potential problem that may affect all life on earth.

An important question that arises is: when is something contaminated? Many governmental and international organizations have attempted, and are attempting, to answer this question. This obviously differs from one type of organism to another. Most research has been conducted on defining the concentration of potentially harmful substances in plants and animals that serve as food for humans, domesticated animals and livestock. Broad consensus has been reached world-wide on what is considered to be dangerous to health with regard to water quality. Two organizations in South Africa have defined water quality guidelines, namely the Department of Water Affairs (previously the Department of Water Affairs and Forestry), DWAF) and the South Africa Bureau of Standards (SABS). Internationally the World Health Organisation (WHO) is the directing and coordinating authority for health within the United Nations system.

This chapter focuses on the concentrations of arsenic, mercury and lead in the water, soil and plant samples collected from the two study areas (study area A which includes the two villages Rooiberg and Leeupoort and study area B represented by Gravelotte and Kwagga). The mineral content in the study areas are compared with those from other countries so as to obtain an idea of the relative levels of contamination present in Limpopo. For this reason, each of the following section of this chapter commences with a short discussion of global patterns of As, Hg and Pb. Thereafter the results of the analyses and presented, followed by a comparison with the global values or with quality guidelines where applicable.

4.2 OVERVIEW OF ARSENIC (As) CONTAMINATION IN THE ENVIRONMENT

4.2.1 Global environmental As patterns

According to Bissen & Frimmel (2003), the total amount of As in the upper crust is 4.01 mil. kg with an average of 6 mg/kg. It is not surprising that traces of As is found in all parts of the environment, including ground water, soils, plants and animals.

In natural waters, the As concentration varies between a few ug/l to many hundreds of mg/l. Arain *et al*. (2009) summarised literature from a wide variety of sources and compiled a list of As levels in different countries. Part of their table is reproduced as Table 4.1.

TABLE 4.1 CONCENTRATION OF As IN GROUNDWATER IN VARIOUS COUNTRIES (Source: Arain *et al*., 2009: 245)

COUNTRY	**CONCENTRATIONS (ug/l)**
United States of America	100 - >500
Canada (Nova Scotia)	18 - 146
Hungary	25 - >50
Ghana	2 - 175
SW Finland	17 - 980
W Bengal, India	<1 - 1300
Calcutta, India	<50 – 23,080
China	<100 - 1860
Thailand	1.25 - 5114
Taiwan	Up to 1800
Japan, Fukuoka	0.001 - 0.293
Bangladesh, Pakistan	<2 - >900

Countries with the highest levels of As are India, Thailand and China, whereas water in Japan is the ‘cleanest’.

The arsenic contents in soils and sediments also vary widely from one place to another depending on the local geology and anthropogenic activities. A list of some of these values is given in Table 4.2, with China having the highest measured values. The mean values in Bangladesh and Italy exceed those of the other countries.

TABLE 4.2 ARSENIC CONCENTRATIONS IN SEDIMENT AND AGRICULTURAL SOILS (mg/kg). (Source: Arain *et al.*, 2009:245).

COUNTRY	RANGE/VALUE	MEAN
West Bengal, India	10 - 196	-
Bangladesh	9.0 - 28	22.1
Argentina	0.8 - 22	5
China	0.01 - 626	11.2
Germany	2.5 - 4.6	3.5
Italy	1.8 - 60	20
Japan	0.4 - 70	11
Mexico	2 - 40	14
South Africa	3.2 - 3.7	-
Switzerland	2 - 2.4	2.2
USA	1.6 - 72	7.5

One of the highest concentrations of As found was in Zimbabwe where 20 g/kg was measured on gold ore deposits. Other mining countries, such as Thailand, Ghana, South Africa, England, Greece, Mexico, Canada and the USA have also recorded very high levels of As in soils, probably due to the fact that As is a by-product of mining and smelting of Cu, Pb, Co and Au ores (Bissen & Frimmel, 2003).

In an aqueous environment, inorganic As (lll) (arsenite) and As (V) (arsenate) are the most abundant species. The mobility of these species is influenced by the pH value, the redox potential, the presence of absorbents (such as oxides and hydroxides of Fe(lll), Al(lll) and Mn(lll/lV), humic substances and clay minerals (Bissen & Frimmel, 2003).

Since arsenate has a strong affinity for iron oxides and hydroxides in soil, the concentration of arsenate in soil solutions is usually low. However, Zhao *et al*. (2008) found that when arsenate was added to nutrient solutions, plant roots rapidly converted it to arsenite. Plant roots are capable of taking up arsenite from the external environment. Thus, the chemical and biological processes taking place in the rhizosphere influences the speciation of As and its bioavailability. According to Woolson *et al*. (1973) in Walsh *et al*. (1977), there is a linear correlation between the levels of As in soils and in plants.

Arsenite uptake shares the highly efficient Silicon pathway of entry into root cells and efflux towards the xylem (Zhao *et al*., 2008). In root cells, any arsenate is reduced to arsenite, effluxed to the external medium, complexed with thiol peptides and sequestered in the vacuoles. Some As, albeit relatively little, is translocated to the shoots. It is therefore not surprising that the highest levels of As occur in plant roots, intermediate levels in vegetative tissue and lowest levels in reproductive tissue (Mpheil *et al.*, 1960) in Walsh *et al*., 1977). While humans eat mainly reproductive tissue, goats eat vegetative tissue. The US Public Health Service tolerance level for As in edible plant material is 2.6 ppm (Walsh *et al.*, 1977). Methylated As species have also been found in plant samples. These may originate from soil, but *de novo* methylathion of As also occurs in plants (Zhao *et al*., 2008). However it is the inorganic As which is the most toxic.

Elevation of As levels in the environment causes considerable concern with respect to plant uptake and subsequent entry into domestic animals, wildlife, and the human food chain (Meharg & Hartley-Whitaker, 2002). An objective of this study was to determine the extent of toxic mineral contamination in the environment. The results of chemical analyses of water, soil, plant and plant material from the study areas in Limpopo, are discussed below. Limits of 0.05 ug/l for water (Pillay *et al*., 2003); 2.5 mg/kg with a range of 0.4 – 7 mg/kg for soil (Pillay *et al*., 2003); and 80 mg/kg dry weight, corresponding to a range of 3 – 300 mg/kg, for plant material (Ward, 2000), will be used to evaluate the extent of contamination in soils, water, and plant materials collected in the study areas.

4.2.2 Arsenic levels in the study areas

Tables 4.3a & b display the results on the analyses of water, soil, plant material and goat's milk in the study areas. The soils in Rooiberg were collected from two different horizons (A and B) of the soil. The horizon A is dark; it is the top soil which contains humus and organic component. The next level to it is the horizon B which is much lighter in colour. Due to leaching from the top soil, however, part of the organic component may have drained into this horizon.

TABLE 4.3a ARSENIC LEVELS IN STUDY AREA A

Source	**Rooiberg**					**Leeupoort**	
	Concentration	**Date**	**Concentration**	**Date**	**Concentration**	**Concentration**	**Date**
Water (ug/l)	13.68	Nov. 2007	11.92	Aug. 2009	12.89	21.75	Nov. 2007
Soil (mg/Kg) A	18.09	Nov. 2007	13.3	Aug. 2009			
B	18.27	Nov. 2007	15.44	Aug. 2009			
Plants – absorbed (mg/Kg)	1.06	Nov. 2007	1.11	Aug. 2009	0.23	0.49	Nov. 2007
Plant material – dust deposition(mg/Kg)	60.71	Nov. 2007		Aug. 2009			
Goat's milk (mg/l)	0.16	Nov. 2007	0.01	Aug. 2009		0.20	Nov. 2007

TABLE 4.3b ARSENIC LEVELS IN STUDY AREA B

Source	**Gravelotte**					**Kwagga**	
	Concentrations	**Date**	**Concentrations**		**Date**	**Concentrations**	**Date**
Water (ug/l)	17.63	Feb. 2008	0	0	Aug 2009	0.40	Feb. 2008
Soil (mg/Kg)	1.74	Feb. 2008				0	Feb. 2008
Plants – absorbed (mg/Kg)	0.1576	Feb. 2008				0.9962	Feb. 2008
Goat's milk	0.01	Feb.	0.03	0.03	Aug	0.00109	Feb.

(mg/L)		2008			2009		2008

4.2.2.1 *Arsenic in water*

Comparison of Tables 4.3a and b reveals that Leeupoort, Gravelotte and Rooiberg have relatively high values of As. However, compared to many other countries (as seen in Table 4.1), these values are low. Nevertheless, they may pose a health hazard, seeing that they exceeded the limits for drinking water of 10 ug/l by WHO (1996). The concentration at Leeupoort (which is the highest of all) exceeds the WHO limit by a factor of 2. Water at Kwagga, on the other hand, appears to be of acceptable quality.

Water samples at Rooiberg were obtained from a dam, while the other samples were all from boreholes. While the possibility exists that the dam water could have been contaminated by mining activities when the mine was inactive, the As in the water obtained from boreholes is probably geologic in origin. It is likely that the As occurs as the inorganic arsenate species.

4.2.2.2 *Arsenic in soil*

Due to logistical reasons, soil samples were only collected from Rooiberg and Gravelotte. It appears from Table 4.3 that the soil at Rooiberg has considerably more than at Gravelotte. Comparison with soil mean As values from other countries (Table 4.2) indicates that the amount of As in soils at Rooiberg is higher than in countries such as Germany, Switzerland, Argentina, China, France, Japan, Mexico, the USA and other parts of South Africa. The concentration at Rooiberg far exceeds the permissible limits (2.5 mg/kg), falling outside the acceptable range of 0.4 – 7 mg/kg (Pillay *et al*., 2003).

The first sample for Rooiberg was collected from a site very close to the dam. These soils were grayish and possibly waterlogged during the wet season. Two clear horizons were evident.

It is possible that the origin of the As is due to the local geology. However, leaching of As has obviously taken place at Rooiberg since the B horizon contains more As than the A horizon.

The relatively high values of As in the natural water and soils – especially at Rooiberg - suggest that this potentially toxic element may enter the biotic components of the food chain.

4.2.2.3 *Arsenic in plant material*

Plant material from the study areas contain surprisingly low amounts of As. Rooiberg has by far the most, but even this amount is much lower than the permissible limits of 80 mg/kg dry weight as given by Pillay *et al*. (2003). Most of the plant material in samples 1 and 2 were composed of grasses while the third sample contained a mixture of herbaceous weeds and woody material. Unfortunately, the types of plants in the other study areas were not identified. It was interesting to note that there was no difference in the plant materials collected during summer and winter (1.06 vs 1.11 mg/kg).

Extremely high concentrations of As were found in the dust on the plant material (60.7 mg/kg vs 1.11 mg/kg inside plants). This is probably due to mining activities. Goats are allowed to roam free in this area and feed on any kind of material that is available, including As-containing dust.

Unfortunately no information is available on the As content of other vegetation in the area – or for that matter - for other parts of South Africa. It is thus not possible to determine whether plants bioaccumulate As from the water soils in the area.

4.2.2.4 *Arsenic contamination in the food chain*

Potential toxicity problems occur at Rooiberg, Leeupoort & Gravelotte due to the high levels of As in water. Kwagga does not appear to have any As contamination problem. The higher amount in water, soils, plant in study area A is probably due to the combination of geology and mining activities.

Water at Rooiberg is accessible to goats. At the other research sites, water from boreholes and rainwater are source of water for the goats. It is possible that the rainwater dilutes the concentration of As in the water consumed by goats.

4.3 PATHWAYS OF MERCURY IN THE ENVIRONMENT.

4.3.1 Overview of global mercury contamination patterns in the environment

Mercury occurs in the environment naturally and in many diverse forms. Generally, mercury contamination is boosted by the activities of humans but generally, the surface water mercury levels in are usually less than 5 parts of mercury per trillion parts of water which is about a thousand times lower than "safe" drinking water standards. According to Fan (1987), mercury in water ranges between below 0.001 to 0.003 ug/l. Normal soil levels range from 0.05 to 0.08 mg/kg (World Bank Group, 1998). Most often, mercury in soils are immobile but chemical and biological activities can transform it to other forms that are toxic and mobile (Heaton, *et al*, 1998). According to literature, levels of mercury in rainwater are in the range 5–100 ng/litre, but mean levels as low as 1 ng/litre have been reported (IPCS, 1990). Ingestion of 500 mg of mercury (II) chloride causes severe poisoning and sometimes death in humans (Bidstrup, 1964). DWAF (1996) guideline for mercury in drinking water is 1 ug/l.

Soil guidelines were established for various types of land use. Table 4.4 gives the general view of the United Kingdom soil guideline values for residential, allotment (open spaces owned by government) and commercial areas (Environmental Agency, 2002).

Table 4.4 Soil Guideline Values for compounds of inorganic mercury as a function of land-use (Environmental Agency, 2002).	
Standard land-use	**Soil Guideline Value (mg/kg dry weight soil)**
Residential with plant uptake	8
Allotments	8
Residential without plant uptake	15
Commercial/industrial	480

The mercury concentration in aquatic bodies differs. There is accumulation of mercury in water bodies when there is inflow of water from soils with high mercury concentration into a basin. Table 4.5 shows the values of mercury in various water bodies as compiled by Stein *et al.* (1996).

Table 4.5 Mercury concentrations in aquatic environments (source: Stein *et al.*, 1996).	
Aquatic environment	**Concentrations**
Open sea	0.5-3.0
Coastal Ocean	2.0-15
Rainwater	2.0-10
Rivers and lakes	1.0-5.0
Remote lakes	0.2-5.3
Urban lakes	1.7
Great lakes	0.9-3.9
Mining contaminated lakes	3.6-104
Chlor-alkali contaminated lakes	5.0-80
Swedish lake	1.35-15
Minnesota lakes	0.9-7.0
Groundwater	2.0-4.0

Clearly, nay water bodies that occur in close proximity to human activities have higher Hg levels than those located in a natural environment.

Plants accumulate mercury through the uptake of water from the soil or from the atmosphere through gaseous exchange (Stein *et al.,* 1996). Table 4.6 shows the concentration of some groups of plants studied by Palusova and his colleagues (1991).

Table 4.6 Mercury concentrations in food (source: adapted Palusova *et al.*, 1991).	
	Concentration in ug/kg
Leafy vegetables	1-76
Fruiting vegetables	0-53
Root vegetables	0-63
Root	1-20

These values are contrary to expectation since it was assumed that metals are not translocated readily through the plant. It was therefore expected that roots would have higher mercury levels than other parts of the plant.

4.3.2 Mercury levels in the study areas

Tables 4.7a & b display the results on the analyses of water, soil, plant material and goat's milk in the study areas.

TABLE 4.7a MERCURY LEVELS IN STUDY AREA A							
Source	**Rooiberg**					**Leeupoort**	
	Concentration	**Date**	**Concentration**		**Date**	**Concentration**	**Date**
Water (µg/l)	0.1062	Nov 2007	0	0	Aug 2009	0.1262	Nov. 2007
Soil (mg/kg) A	0.09	Nov 2007	0.09	-	Aug 2009	-	
B	0.10	Nov 2007	0.07		Aug 2009		
Plants – absorbed (mg/kg)	0.05	Nov 2007	0.05	0.04	Aug 2009	0.04	Nov. 2007
Plant material – dust deposition (mg/kg)	1.464	Nov 2007					
Goat's milk (mg/l)	0.05	Nov 2007	0.01		Aug 2009	0.015	Nov. 2007

TABLE 4.7b MERCURY LEVELS IN STUDY AREA B								
Source	**Gravelotte**					**Kwagga**		
	Conc.	**Date**	**Conc.**		**Date**	**Conc.**	**Date**	**Conc.**	**Date**
Water (µg/l)	2.104	Feb 2008	0		Aug 2009	0.1954	Feb 2008	0	
Soil (mg/kg)	0.021	Feb 2008	0.067		Aug 2009	-		-	
Plants – absorbed (mg/kg)	0.3608	Feb 2008	0		Aug 2009	0.1616	Feb 2008		
Goat's milk (mg/l)	0.011	Feb 2008	0.01	0.006	Aug 2009	0.00561	Feb 2008		
Conc.(concentration)									

4.3.2.1 *Mercury in water*

The mercury concentration of water in the Gravelotte is higher than those from Leeupoort, Rooiberg and Kwagga. This is only to be expected since this is the only mercury-mining area. The concentration in Gravelotte is above the standard set by both the DWAF and SABS value. Thus, the mercury concentration in Gravelotte is possible to pose a health hazard on the people in the community.

4.3.2.2 *Mercury in soil*

Table 4.7 indicates that the soil at Rooiberg more mercury in it than at Gravelotte. The mercury concentration in the soil samples from all the communities in both study areas is higher than the values given by Sloan *et al.*, (2001) and estimated to range from 0.03 to 0.05 mg/kg mercury concentration at a soil depth of 0 - 15 cm. The soil mercury in both communities was lower than the South African permissible mercury in agricultural soils which is 0.5 mg Hg/kg (Vivier *et al*., 1988)

4.3.2.3 *Mercury in plant material*

The mercury in the plant material from the study areas ranges from 0.04 - 0.368 mg/kg. The plant mercury is higher in the study area B (Gravelotte and Kwagga) which also has the highest mercury concentration in the groundwater. The mercury concentration levels in study area B (Gravelotte and Kwagga): 361 μg/kg and 161 ug/kg respectively are more than 100 times higher than the identified mercury level by Palusova *et al.* (1991) in Table 4.6.

4.3.3.4 *Mercury pathways in the food chain*

Deductions made from the information shown in Table 4.7 indicated that there is high level of mercury in the groundwater in study area B which may pose a health problem for the inhabitants of the communities. The sampled plants are those generally eaten by the goats. The plants in these areas differ due to the type of vegetation. This may account for the difference in the heavy metal uptake ability of the plants in the study areas.

4.4 LEAD IN THE ENVIRONMENT

4.4.1 Overview of lead contamination in the environment

Lead is one of the heavy metals that rarely exist naturally as a metal (ATSDR, 1992). Most source of drinking water contains less than 0.005 parts of lead per million parts of water (ppm). Thus the major source of lead contamination to humans is from the soil or dust (ATSDR, 1992). The Environmental Protection Agency (EPA) in United States 1991 has established a limit that the household water lead content should not be more than 0.015 mg/l. The drinking water standard established by Department of Water and Forestry (DWAF, 1996) and the South African Bureau of Standards (SABS, 1999) are 10 μg /l and 50 μg/l, respectively.

Various laws have been enacted by the environmental agencies so as to reduce the concentration of lead in the environment. Such actions include the Clean Air Act Amendments (CAAA) of 1990 banned the sale of leaded gasoline as of December 31, 1995. The EPA and United states department of housing and development sets a

standard that paint with lead content exceeding 1.0 mg/cm^2 cannot be used in housing. The general natural concentration of lead in the soil in United States ranges from 2-200 mg/kg with exemption of areas with lead deposits (Faust and Aly, 1981). Lead concentration has drastically increased due to human activities especially mining and the use of vehicles (ATSDR, 1992).

The maximum allowable limits for lead in soil are given for various countries in Table 4.8.

Table 4.8 Maximum allowable limits (M.A.L.) for Pb in soil (mg/kg) used in different countries (adapted Kabata-Pendias, 1995)	
Countries	**MAL for lead in soil (mg/kg)**
Australia	100
Canada	200
Poland	100
Japan	400
Great Britain	100
Germany	500

4.4.2 Lead levels in the study areas

The table 4.5a and b illustrates the lead concentrations in the soil, water, plants and goats' milk in the sampled community.

TABLE 4.9a LEAD LEVELS IN STUDY AREA A							
Source	**Rooiberg**					**Leeupoort**	
Water (μg/l)	0.7731		0	0		0	
Soil (mg/kg) A	29.52		24.27				
B	25.36		26.29				
Plants – absorbed (mg/Kg)	1.81		0.99	0.92		0.85	
Plant material – dust deposition(mg/kg)	115.1						
Goat's milk (mg/l)	0.27		0.01			0.348	

TABLE 4.9b LEAD LEVELS IN STUDY AREA B								
Source	**Gravelotte**					**Kwagga**		
	Concentration	**Date**	**Concentration**		**Date**	**Concentration**		**Date**
Water (ug/l)	0.1862	Feb. 2008	0	0	Aug 2009	0.276	0	Feb. 2008
Soil (mg/kg)	4.227	Feb. 2008	4.252	-	Aug 2009	0	-	Feb. 2008
Plants – absorbed (mg/kg)	0.4791	Feb. 2008	-	-	Aug 2009	2.574	-	Feb. 2008
Goat's milk (mg/L)	0.00908	Feb. 2008	0.008	0.009	Aug 2009	0.0093	0.0063	Feb. 2008

4.4.2.1 *Lead in water*

The 0.7731 µg/l lead concentration in Rooiberg is the highest of the four communities. It is lower than the WHO 0.01 mg/l and 0.04 mg/l of the German drinking water standard (Zietz *et al.,* 2000) land the 20 µg/l that was recorded by Dabeka *et al.,* (1987) in five Canadian cities. It is lower than the permissible South African drinking water standards of 10 µg /l (DWAF, 1996) and is less than the of 20.6 µg/l lead concentration in early morning tap water in Thohoyandou, South Africa (Okonkwo & Maribe, 2005).

4.4.2.2 *Lead in soil*

The soil lead concentrations in study area A are higher than the maximum permissible concentration of 6.6 mg/kg set by Water Research Commission for South African agricultural soils but is lower than the M.A.L. for other countries (Table 4.10).

4.4.2.3 *Lead in plant material.*

The concentration of lead in the plant material in Gravelotte (4971 µg/kg) is the highest followed by that at Rooiberg (1810 µg/kg). The other two study sites have a low plant lead concentrations**.** The concentration in the plants material is higher than the recorded lead concentration in brown rice in Northern Taiwan (Zueng-sang Chen, 1991)

4.4.3 *Lead pathways in the study area*

From the Table 4.12, it shows that Rooiberg has the highest lead concentration in its environment than the others. All the environmental indicators (water, plant and dust) used in the study confirms this. Although, the Pb water concentration in Rooiberg (which was the highest) does not exceed the South African drinking water limit, the soil (Rooiberg) exceeds the maximum permissible concentration set for South African agricultural soil by the Water Research Commission (Smith and Vasiloudis, 1991).

4.5 SYNTHESIS.

The mineral content of plant material exceeds that of water. This may be due to bio-accumulation. Plants absorb the elements as they carried along the nutrients from the soil. Considering the fact that As is higher in the all the environmental components at Rooiberg, confirms the supposition by Sámi & Druzynski (2003) that there is possibility of arsenic toxicity in study area A. The groundwater in the borehole from Leeupoort is also higher than at Gravelotte. This indicates that there is possibility that the As in Study area A is of a geological origin. This also is evident in the high concentration of the As in the soils in study area A compared with B. The water Hg from Gravelotte is higher than the acceptable Hg in South African drinking water. This agrees with the initial preliminary identification of study areas in this study (which was with the use of map: region G mineral) of the possibility of Hg contamination in Gravelotte. This may be a threat to the community in the area.

CHAPTER 5

EXPOSURE PATHWAYS OF ARSENIC, MERCURY AND LEAD TO INFANTS

5.1 INTRODUCTION

This chapter deals with the exposure pathways of As, Hg and Pb from the environment to humans. Only one study site was chosen, namely Rooiberg, since it had the most complete of results. It was also the area that was shown to have high levels of arsenic in the ground water, thus posing a possible health hazard, both directly and indirectly (through the environment) to infants, the most vulnerable segments of society.

5.1.1 Vulnerability of infants to contaminants

As mentioned in Chapter 1, infants have a higher fluid intake per body weight to replace their relatively larger surface area, higher metabolic rate and inability of the kidneys to concentrate urine. This implies that xenobiotics and toxic minerals which can be stored in tissues may accumulate to a greater extent in infants than in adults (Hooper and McDonald, 2000).

There are four main sources of exposure of toxic minerals to infants. They are: water, food (mother's milk and goat's milk) and dust. During the first months of life, an infant's diets consist mainly of breast milk, cow or goat's milk or formulas. These might be additional sources of toxic substances. For children on formula feeds, the water used to make it up can also be a source of exposure.

Another source of exposure is soil or house dust. Oral exploration constitutes an important part of normal development between 6 and 36 months. It has been found that the ingestion of soil and house dust through hand-to-mouth activities of children below

the age of 5 years is typically 100 mg/d (Patriarca *et al*., 2000:353). These values were determined for European conditions. Infants in rural areas of South Africa are probably exposed to more dust, but the value of 100 mg/d is used as the standard in the analyses.

The above-mentioned factors enhance the vulnerability of infants to the noxious effects of chemicals. The differences between children and adults also modify the patterns of exposure. Importantly, the above factors serve to limit the applicability of data obtained from adult populations.

5.1.2 Assumptions

A number of assumptions were made in this chapter. Although some of these are based on scientific evidence, others are based on knowledge from experienced doctors.

- For the purposes of this study, the development of infants was divided into four stages, namely, newborns, breastfeeding infants, crawling infants and toddlers. It was assumed that the development rate and growth of infants in Rooiberg follow the general growth curves milestones as set out by Health24, as shown in Fig 5.1.

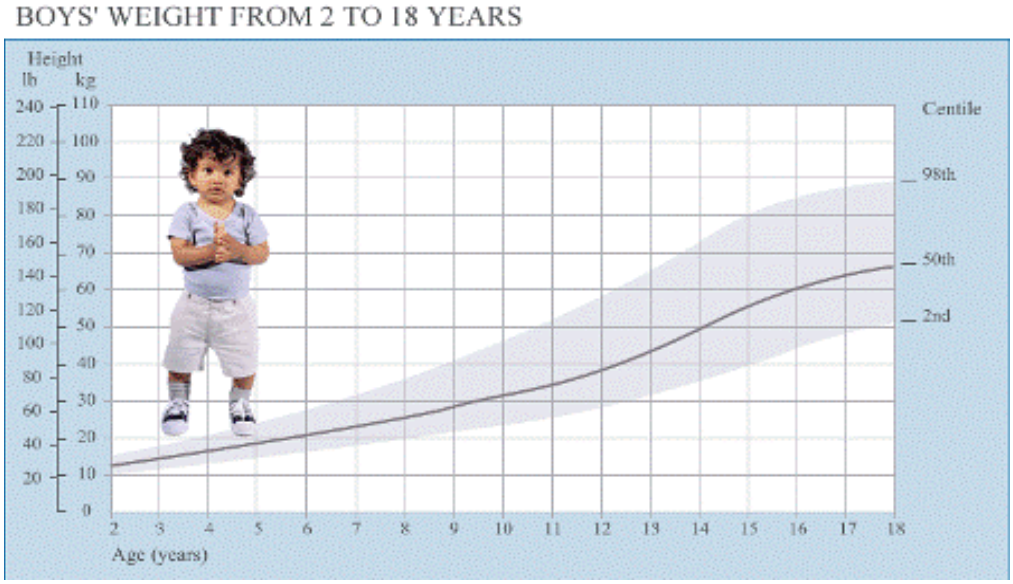

Fig 5.1 Boys' weight from birth to 2 years of age *(Health24, 2009)*

- Breast feeding usually continues up to the age of two years but women in rural areas are generally considered to breast-feed their offspring for a longer period than their urban counterparts.

- Breast milk is the ideal food for a baby. Breast milk is rich in a range of nutrients that help a baby to grow, develop and stay healthy. The health benefits of breastfeeding are widely acknowledged, but the duration for exclusive breast feeding has not been ascertained. There are diverse opinions on this. According to American Academy of Pediatrics (AAP, 2005), exclusive breastfeeding is recommended up to 6 months of age, with continued breastfeeding along with appropriate complementary foods up to two years of age or beyond. The Anthropologist, Katherine Dettwyler in Australia Breastfeeding Association's (ABA) magazine 'Essence' stated that the estimated the natural age of weaning to be between two and a half and seven years, based on developmental factors and comparisons with other mammals.

- Cow's and goat's milk is usually used as a substitute for mother's milk once weaning commences. Since no cows were seen in the vicinity of the study areas it was assumed that goat's milk is used. The villagers confirmed this to be the case.
- The types and amounts of foods ingested vary from one stage to another, as does the exposure to dust.

The following assumptions were made in this study:

- **Newborns** (birth – 1 month) are fed exclusively on breast milk or formulae (made up with locally obtained water). The average feeding rate for 3-day old babies is about 100 ml every four hours. This is 600 ml/d (pers comm. Dr Prinsloo, 2009). Newborns were assumed to weigh 4 kg.
- **Breast feeding stage:** At age 3 months, babies are still breast-fed and obtain all their nutrients from mother's milk. The consumption rate at 3 months is 150 ml/4 hrs, or 900 ml/d (pers comm. Dr Prinsloo, 2009). For this study, the average age of breast feeding babies was assumed to be 5 months. At this stage the baby should weigh around 8 kg and consume 1 litre of mother's milk per day.

It was further assumed that babies of this age would be exposed to around 50 mg of dust/d.

- **Crawling infants**: Babies start to crawl at 6 months and attempt standing at around 8 months. For the purposes of the study, the age of crawling babies was assumed to be 7 months. In rural areas, infants of this age are still largely dependent on breast milk, although weaning does start to take place. It was assumed that the total consumption of fluids for crawling infants is 1 l/d of which 750 ml comprises of breast milk and the remaining 250 ml, water. The average weight of a crawling infant was assumed to be 8.5 kg. Crawling infants are exposed to at least 100 mg dust per day.
- **Toddlers:** Babies usually start walking at 9 to 14 months of age. The average age of a toddler was taken to be 12 months. At this stage, toddlers are slowly weaned from breast milk, which is replaced by goat's milk. It was assumed at one year-old babies weigh approximately 10 kg and consume around 250 ml water, 500 ml breast milk and 250 ml goat's milk, and are exposed to 100 mg dust per day.
- Lastly, It was also assumed that the formula-feeds used for infants do not contain any arsenic, mercury or lead.

5.1.3 Limitations

The most serious limitation to this part of the study was the small number of samples analysed. This was due to logistical problems experienced in reaching the far-rural areas. Language barriers also caused difficulties.

The major problems were encountered when attempting to obtain mother's milk. Despite obtaining prior consent from the clinic and the breast-feeding mothers, upon arrival at the study area, the husbands would not give permission for milk samples to be taken. It appears that the men-folk were afraid that the milk would be used to test for HIV/AIDS, or that the researchers had some dark muthi-related ulterior motives. In the few cases where it was possible to obtain mother's milk, information on the actual stage of lactation could not be obtained. This is an important shortcoming since Rossipal *et al.* (1998)

found that the composition of mother's milk changes over time. The concentrations of essential trace elements, such as Cu, Mn, Mo and Zn, decrease significantly during the lactation period. Toxic elements such as Cd and Hg in early mature milk (42 – 60 days past partum) are only one tenth of that found in colostrum (1-3 days post partum). The concentration of minerals in milk differs even during a breast feeding event.

A further complication was that the two samples of breast milk obtained from Rooiberg were mixed and analysed as a single sample at the ARC laboratory.

Problems were also encountered with obtaining milk from the goats. Goats are usually milked early in the morning or late in the afternoon. It was not always possible to be at the research areas at these times due to their remoteness. During some trips no goat milk was obtained, and during others, very small amounts. Notwithstanding this, the ARC was able to analyse even these meager samples.

Other information not obtained were:

- The speciation of the minerals.
- The exact age at which the babies were weaned from mother's milk.
- The proportion of goat's milk, mother's milk and water-based formulae/food used for toddlers.
- The exact amount of dust to which infants are exposed.

5.1.4 Procedure followed in the chapter

In contrast to Chapter 4 which dealt with the relative abundance of As, Pb and Hg found in the different environmental components of the food chain, this chapter focuses on the exposure of infants to toxic elements. It therefore compares the quality of water, soil, goat's and mother's milk from Rooiberg with known standards or quality guidelines as determined by various authorities.

The quality of the water samples were compared with the guidelines for drinking water standards as set by the World Health Organisation (WHO), the South African Department of Water and Forestry (DWAF) and the South Africa Bureau of Standards (SABS). These standards give the permissible levels i.e. the upper limit of contamination that is allowed for potability.

In order to assess whether the infants at Rooiberg are at risk of exposure to As, Hg and Pb, the ingestion of these elements was determined and compared with the respective Provisional Tolerable Weekly Intake (PTWI) as set by the United Nation's Food and Agricultural Organisation (FAO) and the WHO. Minimal Risk Levels (MRLs) were also used to determine the extent of exposure. The Agency for Toxic Substances and Disease Registry (ATSDR) established chronic, sub-acute and acute limits for each of the toxic elements in terms of their health effect on humans.

Hazard Quotients (HQs) were calculated to quantify the extent of potential risk to which infants are exposed during their different developmental stages. The Hazard Quotient is the ratio between the estimated exposure and the tolerable daily intake for that mineral. It is a method used to estimate ecological risk to the harmful effect of a contaminant on the object of concern. It is useful in the estimation of risk because it is quick and easy to use. It is also simple to interpret since the level of risk can be assessed using a single value of (< 1 or > 1). If the HQ exceeds 1, the toxicant may produce an *adverse effect*; a hazard quotient of less than one indicates that no adverse effects are likely over a lifetime of exposure (Lemly, 1996).

The EPA (US Environmental Protection Agency) and the FAO (Food and Agricultural Agency) use the following formula to calculate the HQ:

$$HQ_{(RfD)} = \frac{\text{Exposure Estimate}}{\text{RfD}}$$

or

$$HQ_{(MRL)} = \frac{\text{Exposure Estimate}}{\text{MRL}}$$

Where:

HQ= Hazard quotient

Exposure estimate = total estimated exposure through ingestion

RfD = Reference Dose for chronic oral exposure (EPA, 2010). (The RfD is an estimate (with uncertainty spanning perhaps an order of magnitude) of a daily exposure to the human population (including sensitive subgroups) that is likely to be without an appreciable risk of deleterious effects during a lifetime (EPA, 2010).

MRL = Minimal Risk Levels.

The MRL and RfD values are the same for As and Hg, namely, As = 0.3 ug/kg_{bw}/d (FAO/WHO, 1983; 1988); and Hg = 0.3 ug/kg_{bw}/d (US EPA, 1995).

Due to the high toxicity of Pb, no RfD or MRL values are available for Pb. This implies that the relative risk could not be compared for the three minerals. Therefore, another HQ was calculated using the formula:

$$HQ_{(TDI)} \quad \frac{\text{Exposure Estimate}}{\text{PTDI}}$$

Where:

PTDI and PTWI are the Potential Tolerable Daily and Weekly Intake levels, respectively. PTWI and hence PTDI are available for all three minerals.

PTDI As = 2.1 ug/kgbw/d;
PTDI Hg = 0.7 ug/kgbw/d
PTDI Pb = 3.6 ug/kgbw/d (FAO/WHO, 1986).

In order to distinguish between the HQs, HQ_{MRL} and HQ_{TDI} are used.

The exposure to each of the three toxic elements is discussed in sections 5.2 to 5.4. The layouts of these sections were as follows:

- Each section commences with a summary of the results of the analyses and a general comparison of levels at Rooiberg with those reported from other parts of the world.
- Thereafter exposure from each of the *sources* – i.e. water, mother's milk, goat's milk and dust were calculated for each of the development levels of infants (newborns, suckling, crawling infants and toddlers). This is necessary since the sources of ingestion and the amount ingested differ from one development phase to another.
- In the sections following this, the extent of total exposure experienced by infants was determined for each of the different development phases. Here the weight of the infant is the important issue, since PTDIs and MRLs are expressed per kilogram body weight (kg_{bw}). Comparison of total exposure during each of the development phases with these standards give an indication of the extent of the risk to which infants are exposed at Rooiberg. High levels of exposure to more than one toxin will obviously increase the risk of adverse health effects and could have a significant impact on the well-being of individuals throughout their lives.

5.2 EXPOSURE TO ARSENIC

5.2.1 Findings

Table 5.1 shows the levels of As found in the water, soil, goat's milk and mother's milk at Rooiberg. Where available, results from the milk samples obtained from the other research areas (Leeupoort (L), Gravelotte (G) or Kwagga (K)) have been included.

TABLE 5.1 ARSENIC LEVELS IN SAMPLES FROM ROOIBERG

	Sample 1	Sample 2	Sample 3	Date	Other
Water (ppb)	13.68	11.92	12.89	Aug. 2009	21.75 (L); 17.63 (G); 0.40 (K)
Soil A horizon (ppm)	18.27	18.09		Aug. 2009	1.74 (G)
Goat milk (ppm	0.16	0.01		Aug. 2009	0.20(L); 0.02(G); 0.001(K)
Mother's Milk (ppm)	0.05			Aug. 2009	0.01(G)

Ppb = ug/l; ppm = mg/kg or mg/l
G = Gravelotte; K = Kwagga; L = Leeupoort

As indicated in Chapter 4, the arsenic concentration of both water and soil is higher at Leeupoort than at Rooiberg. Although the concentration of As found in the study area was relatively high, the waters did not nearly reach the concentration of 12 mg/l which was found in some well water in South Africa McKee & Wolf (1963) in Luh *et al*., (1973: 2). Unfortunately the original document could not be located and hence the location of this well could be determined. Nevertheless, arsenic in water from for both Rooiberg and Leeupoort exceed the DWAF limits of 10 ug/l.

The average goats' milk arsenic is also higher in Leeupoort than in Rooiberg. According to Anke (1986) in Casey (1995: 654), milk from cows and goats usually contain 20 to 60 ug As/l. Most of the milk obtained from goats in the study areas had As levels of only 0.01 to 0.03 mg/l i.e. 10 - 30 ug/l. This falls within the 'normal' range when compared with world averages. However, one of the samples from Rooiberg contained 0.16 mg/l i.e. 160 ug/l. This may be due to the goats ingestion of plant material covered in dust (see

Table 4.3a). This is between 2 and 8 times the amount found in goat's milk from other countries. This may be due to the goats ingestion of plant material covered in dust (See Table 4.3a).

Casey (1995) conducted a survey of literature on the levels of As found in breast milk. She reported that in countries such as India, Yugoslavia, Guatemala, Hungary and Sweden, As levels were between 0.2 and 0.7 ng/ml (i.e. 0.2 – 0.7 ug/l). Very high levels were found in Northern Chile (5 ug/l), Greece (6 ug/l) and the Philipines (19 ug/l). The concentration of As in the Rooiberg mother's milk was 0.05 mg/l i.e. 50 ug/l). This is more than twice the amount found in the Philippines and more than 70 times the average in most countries. The above indicates that infants are exposed to high levels of As from all four contamination sources – water, dust and soil, goat's milk and mother's milk.

5.2.2 Exposure through the ingestion of water

From Table 5.1, it is clear that water at Rooiberg exceeds the WHO (1996) recommended limit of 10 ug/l, but it does not exceed the Mandatory Limit of 50 ug/l (Luh *et al*., 1973).

The extent of exposure depends on a number of factors such as the metal speciation, the level and period of exposure and the physical and nutritional state of the infant (Dorea & Donangelo, 2006). The amount of water taken in by the infant depends upon its age. A newborn's dietary requirements are met by mother's milk, except where some problem arises and the infant is given a formula (using local water). Otherwise, it is assumed that infants start taking in water (either as is or in formula) at the crawling stage.

Exposure to As is thus:

- *For newborns on formula feeds*:
  Consumption of newborns = 600 ml/d. The total amount of As taken in = 600 ml/d x 13.68 ug/l = 8.2 ug/d.
- *Breastfeeding and crawling infants on formula feeds:* Consumption = 1 l/d.
  As intake = 13.68 ug/d.

- *Crawling infants and toddlers not on formula feeds:* Consumption of water = 250 ml. As intake from water = 250 ml x 13.68 ug/l = 3.42 ug/d.
- *Toddlers on formula feeds:* Consumption = 750 ml/d. As intake = 750 ml/d x 13.68 ug/l = 10.26 ug/d.

5.2.3 Exposure through dust and soil

According to the EPA (1997), children under the age of 5 years ingest 100 mg soil per day due to oral exploration by suckling (Holson *et al*., 2000; Patriarca *et al.*, 2000) and during the crawling and toddler stages. This is the default value used for crawling infants and toddlers. Breast-feeding (suckling) infants probably ingest slightly lower amounts of dust. The assumption here is 50 mg/d (0.00005 kg/d).

Exposure to As:

- *Suckling/Breast feeding babies*: 5×10^{-5} kg/d x 18270 ug/kg = 0.910 ug/d.
- *Crawling infants and toddlers*: 10^{-4} kg/d x 18270 ug/kg = 1.827 ug/d.

According to the ATSDR, a concentration of 44.0845 mg/kg gives a hazard quotient of 1. The amount of As to which infants are exposed via dust, is thus very low.

5.2.4 Exposure through goat milk

The assumption was made that goat's milk is consumed only once the infant reaches the toddler stage. Exposure to As through goat's milk is 250 ml x 0.16 mg/l = 0.04 mg/d = 40 ug/d.

5.2.5 Exposure through mother's milk

It is assumed that mother's milk intake varies between 500 ml for older infants and 1 litre during the breast feeding stage.

Exposure to Arsenic via mother's milk:

- *Newborns*: 600 ml x 0.05 mg/l = 0.03 mg/d = 30 ug/d.
- *Breast feeding infants*: 1 l x 0.05 = 0.05 mg/d = 50 ug/d.
- *Crawling infants*: 750 ml/d x 0.05 mg/l = 0.04 mg/d = 40 ug/d.
- *Toddlers:* 500 ml/d x 0.05 mg/l = 0.025 mg/d = 25 ug/d.

5.2.6 Discussion

5.2.6.1 Exposure to newborns

The total amount of As ingested originates from mother's milk or water used to make up the formula.

For babies ingesting mother's milk, the total As ingested is 0.03 mg/d = 30 ug/d. Assuming the baby weighs 4 kg, the potential exposure level = 30/4 = 7.5 ug/kg_{bw}/d. If using water to make up formula feeds, ingestion is 8.2 ug/d i.e. 8.2/4 = 2.05 ug/kg_{bw}/d.

According to the Joint FAO/WHO (1989) the PTWI is 15 ug inorganic As/kg_{bw}/week i.e. 2.1 ug/kg_{bw}/d. The speciation of the arsenic is not known, but a worst case scenario is assumed. This implies that a breast fed newborn in Rooiberg might be exposed to more than twice the PTWI. Babies being fed on formula foods are less exposed to As. The exposure of 2.05 ug/kg_{bw}/d is just below the FAO/WHO limits.

It is unclear whether the threshold values apply to newborns whose entire diet is composed of mother's milk or to adults. As indicated, alternate threshold values for As were obtained from the ATSDR (1999). The MRL values differentiate between acute and chronic poisoning. According to the ATSDR (1999), MRL the threshold value for chronic poisoning is 0.3 ug/ kg_{bw}/d. Since babies are exposed to daily ingestion of toxins, chronic threshold values apply. Using this value, the As intake for breast fed newborns

Rooiberg exceeds the chronic MRL levels by a factor of 7.5/0.3 = 25 and those on formula feeds, 7 times (2.05/0.3) (as shown by the HQ values).

5.2.6.2 *Exposure by suckling infants*

Breast feeding infants are assumed to weigh an average of 8 kg at 5 months of age. If the babies receive all their nutrients from mother's milk, they ingest 1 litre milk and hence 50/8 = 6.25 ug As/kg_{bw}/d. This exceeds the PTDI (2.1 ug/kg/d) by a factor of almost 3. If the baby is on formula feeds, the water used to prepare the food contains 13.68 ug/l. The daily intake of As from this is 13.68/8 = 1.71 ug As/kg_{bw}/d. This is below the PTDI.

However, at this stage it must be assumed that the baby is exposed to some dust. If exposure is 50 mg dust per day, the As contribution of dust is 18270 ug/kg x 5 x 10^{-5} kg/d i.e. 0.914 ug/d or 0.115 ug/kg_{bw}/d.

The total As exposure of breast feeding infants is thus 6.37 ug/kg_{bw}/d and that of a formula fed baby, 1.29 ug/kg_{bw}/d. The former value is 3 times the PTDI and 21 times more than the stipulated chronic MRL level. Formula-fed infants are at lower risk, since the As ingested is below the PTDI. Nevertheless, the exposure exceeds the MRL values by a factor of 4.

5.2.6.3 *Exposure by crawling infants*

It was assumed that the average age and weight of crawling infants are 7 months and 8.5 kg, respectively. Crawling infants ingest about 750 ml breast milk, 250 ml water-based drinks and 100 mg dust per day. The amount of As ingested is: 37.5 ug/d from milk; 3.42 ug/d from water and 1,827 ug/d from dust. This is a total of 42.75 ug/d. The intake per kg body weight is 42.75/8.5 = 5.03 ug/kg_{bw}/d. This is more than twice fold the PTDI and 16 times the MRL.

Formula-fed babies are much better off, since the food component contributes 1.61 ug/kg_{bw}/d to the total ingestion of As, in comparison to the 4.81 ug/kg_{bw}/d in breast-fed

infants. Although the food component is below the limits given by the FAO/WHO, it is still 5 times the MRL.

5.2.6.4 *Exposure by toddlers*

Twelve month-old toddlers weigh approximately 10 kg and consume a mixture of mother's milk, goat's milk and water- based foods as well as being exposed to dust. The breast milk contributes 25 ug; the goat's milk, 40 ug, the water, 3.42 ug and dust 1.827 ug As per day. Equivalent daily ingestion rates per kilogram body weight are 2.5; 4; 0.34 and 0.1827 ug/kg_{bw}/d, respectively. This gives a total of 7.03 ug/kg_{bw}/d for breast-milk fed infants and 5.21 ug/kg_{bw}/d for formula-fed babies. Once again, this vastly exceeds the PTDI and MRL values. The food component alone contributes between 5 and 7 ug/kg_{bw}/d i.e. around three times the WHO (1983) limit.

5.2.6.5 *Summary: Exposure of arsenic to infants*

Table 5.2 gives a summary of relevant values of As ingestion by infants from the various sources of contamination, during the different development periods. In all cases, the dust component of As is very small. It is also clear that the greater the water component in the diet, the lower the risk of contamination. Both mother's milk and especially goat's milk are contaminated by arsenic and these contribute to the As intake risk by infants. Therefore the lower the amount of goat's milk consumed at an early age, the better for the health of the infant.

It is evident Table 5.2 that all the HQ values of infants following a normal diet, exceeded 1. This indicates that all the stages of the infants at Rooiberg are exposed to hazardous levels of As which may leave a harmful health effect.

TABLE 5.2 SUMMARY: EXPOSURE TO ARSENIC VIA INGESTION

DATA & INFO	WATER	MOTHER'S MILK	GOAT MILK	DUST/SOIL	TOTAL (ug/kg_{bw}/d)	HQ
Rooiberg	13.68 ug/l	50 ug/l	160 ug/l	18270 ug/kg		Total exposure/0.3
DWAF limits	10 ug/l					

PTWI = 2.1 ug/kgbw/day **ATSDR chronic limit = 0.3 ug/kgbw/d**						
INFANT AGE & WEIGHT	**WATER**	**MOTHER'S MILK**	**GOAT MILK**	**DUST/SOIL**	**TOTAL ($ug/kg_{bw}/d$)**	**HQ**
NEWBORNS Weight = 4 kg	600 ml	600 ml	0	0		
on mothers' milk	-	**7.5 $g/kg_{bw}/d$**			7.5	25
on formula feeds	**2.05 $ug/kg_{bw}/d$**				2.05	6.83
SUCKLING INFANTS 8 kg	1 litre	1 litre		0.00005 kg		
On mother's milk		**6.25 $ug/kg_{bw}/d$**		**0.115 $ug/kg_{bw}/d$**	6.37	21.2
On formula feeds	**1.17 $ug/kg_{bw}/d$**			**0.115 $ug/kg_{bw}/d$**	**1.29**	**4.3**
CRAWLING INFANTS 8.5 kg	For mother's milk – 250 ml; For formula feeds = 1 litre	For mother's milk = 750 ml; For formula feeds = 0		100 mg = 10^{-4} kg		
On mother's milk	**0.40 $ug/kg_{bw}/d$**	**4.41 $ug/kg_{bw}/d$**		**0. 215 $ug/kg_{bw}/d$**	5.03	16.77
On formula feeds	**1.61 $ug/kg_{bw}/d$**			**0.215 $ug/kg_{bw}/d$**	**1.83**	**6.1**
TODDLERS 10 kg	For mother's milk = 250 ml	500 ml	250	100 mg = 10^{-4} kg		
	For formula 750 ml		250 ml	10^{-4} kg		
On mother's milk	**0.342 $ug/kg_{bw}/d$**	**2.5 $ug/kg_{bw}/d$**	**4 $ug/kg_{bw}/d$**	**0.183 $ug/kg_{bw}/d$**	**7.03**	**23.43**
On formula feeds	**1.026 $ug/kg_{bw}/d$**		**4 $ug/kg_{bw}/d$**	**0.183 $ug/kg_{bw}/d$**	**5.21**	**17.37**

5.3 EXPOSURE TO MERCURY

5.3.1 Findings

The concentration of mercury found in the sampled soil, water, plant, goats' milk and human milk in Rooiberg are presented in Table 5.2 below.

TABLE 5.3 MERCURY LEVELS IN SAMPLES FROM ROOIBERG					
	Sample 1	**Sample 2**	**Sample 3**	**Date**	**Other**
Water (ppb)	0.1062	0	0	Aug 2009	0.1262(L); 2.1004(G); 0.0454 (G); 0.1954 (K)
Soil A horizon (ppm	0.09	0.09		Aug 2009	0.021 (G)
Goats milk (ppm)	0.05	0.01		Aug 2009	0.015 (L); 0.011 (G); 0.01 (G); 0.006(G); 0.005 (G); 0.006(G); 0.0056(K)
Mother's Milk (ppm)	0			Aug 2009	0 (G)
Ppb = ug/l; ppm = mg/kg or mg/l G = Gravelotte; K = Kwagga; L = Leeupoort					

According to Dorea and Donangelo (2006), the level of mercury in the human milk is dependent on various factors such as diet and the stage of lactation. Chien *et al.,* (2006) mention that the stored methyl mercury (MeHg) in the body can be mobilized into the milk during lactation. Accumulated inorganic mercury can also be passed to the breast-fed babies by their mother's milk (Risher, 2003).

The average concentration of mercury in milk has been found to range from 0.15 to 13.9 ug/l in other parts of the world. It was estimated that the total dietary intake of mercury by the breast-fed infants ranges from 0.21 to 3.1 ug/l (Galal-Gorchev (1993) in Dorea and Donangelo (2005). In Saudi Arabia the breast milk concentration differs with location; in Riyald the concentration is 4.15 ug/l whereas it is 2.19 ug/l in Al-Ehssa (Al-Seleh *et al*., 2003). In Poland, the mercury concentration in goat's milk in some industrial region is found to be 5.1 ug/l.

At Rooiberg, the amount of Hg in mother's milk is below detectible limits while there is 50 ug Hg per litre of goat's milk. This value is very high in comparison with that found elsewhere.

5.3.2 Exposure to Hg through the ingestion of water

Both The EPA's and the SABS's recommended limit for Hg in water is 2 ppb while the DWAF (1986) stipulate a limit of 1 ppb. The Hg concentration in the water in Rooiberg is less than these values and does thus not pose a danger to health.

The mercury exposure to infants through water is:

- *Newborns:*

  Newborn babies on mothers' milk do not consume any water.

  New born babies on formula: The consumption of the newborn is 600 ml/d. Therefore, the mercury consumed through water in Rooiberg is:

  600 ml/d x 0.1062 ug/l = 0.064 ug/d.

- For *breastfeeding (suckling) and crawling infants on formula feeds:*

  The water consumption is 1 litre/day. Therefore, the total mercury consumed is 0.1062 ug/d.

- *Crawling infants and toddlers not on formula feeds:*

  Consumption of water = 250 ml.

  Thus, intake from water is 250 ml x 0.1062 ug/l

  = 0.0266 ug/d.

- *Toddlers on formula feeds:* Consumption = 750 ml/d.

  Mercury intake: 750 ml/d x 0.1062 ug/l

  = 0.0797 ug/d

5.3.3 Exposure through dust and soil

The soil mercury concentration usually varies according to the depth of the soil, with the highest levels occurring in the surface layers of the soil (ATSDR, 1999). The Rooiberg mercury concentration is within the concentration that is naturally present in soils at concentrations (0.003 and 4.6 mg/kg) (Arbestain *et al.*, 2009).

There is 0.09 mg/kg i.e. 90 ug/kg mercury in the dust at Rooiberg. The exposure to mercury in this area is:

- *Breast feeding babies*:
  5×10^{-5} kg/d x 90 ug/kg = 0.0045 ug/d.
- *Crawling infants and toddlers*:
  10^{-4} kg/d x 90 ug/kg = 0.0090 ug/d.

5.3.4 Exposure through goat milk

The presence of the rumen in the ruminants enhances the demethylation of mercury, thus, lowering the mercury concentration in their milk and meat.

The exposure to the toddlers through the goats' milk is 0.25 l/d x 0.05 mg/l
=0.0125 mg/d = 12.5 ug/d.

5.3.5 Exposure through mother's milk

The amount of Hg in the mother's milk is not detectible. Thus, the babies in Rooiberg are not exposed to mercury through the mothers' milk.

5.3.6 Discussion

5.3.6.1 *Exposure by newborns*

The formula-fed newborns are possibly exposed to mercury through the water used in the preparation of the formula. However, since there is no Hg in breast milk, breast-fed infants are not exposed to mercury at all.

The formula-fed newborns exposure is calculated as follows:
Total mercury ingested is 0.064 ug/d. Assuming that the baby weighs 4 kg, the exposure level will be 0.064/4 = 0.016 ug/kg_{bw}/d. The daily exposure is lower than the EPA reference dose (RfD) and the MRL for mercury which is 0.3 ug/kg_{bw}/d (EE/CA Report, 2008). There is thus no likelihood of health risk due to mercury exposure for this group of babies.

5.3.6.2 *Exposure by suckling infants*

This category of infants is assumed to averagely weigh 8 kg at 5 months. The babies' liquid intake is 1 litre per day. Total mercury ingested from formula food therefore is 0.1062 ug/d. For an 8 kg baby the exposure is thus:
0.1062/8 ug/kg_{bw}/d. = 0.01328 ug/kg_{bw}/d.

At this stage of development, it is assumed that the baby is exposed to dust. Assuming that the exposure to dust is 50 mg. The total exposure through dust is thus 5 x 10^{-5} kg/d x 90 ug/kg = 0.0045 ug/d. For an 8 kg baby dust exposure per weight is: 0.0006 ug/kg_{bw}/d. The total mercury exposure of suckling babies on mothers' milk is thus 0.0006 ug/kg_{bw}/d and for formula-fed babies, it is 0.0133 ug/kg_{bw}/d.

The daily exposure is much lower than the reference dose mentioned by Dorea and Donangelo (2006).

5.3.6.3 *Exposure by crawling infants*

The average body weight of a 7 months old baby is assumed to be 8.5 kg. This age group ingests 750 ml of mother's milk, 250 water based drinks and 100 mg of dust. The mercury intake through the breast milk is not significant. The daily mercury intake through water by the breast-fed baby at this stage will be 0.25 l x 0.1026 ug/l = 0.0265 ug/d. The ingestion per body weight is therefore, 0.0031 ug/kg_{bw}/d.

However, the formula-fed babies of 8.5 kg will take in 1 litre of water based foods. Thus, the mercury ingested will be 0.1062 ug/d. Hence, 0.0125 ug/kg_{bw}/d.

The exposure through dust is 0.009 ug/d thus, for a 8.5 kg baby; the amount of Hg ingested will be 0.0121 ug/kgbw/d, for breast-fed infants and 0.0135 ug/kgbw/d for formula-fed infants. Both values are far below the MRL values for Hg and therefore these infants are not at risk from Hg.

5.3.6.4 *Exposure by toddlers*

A toddler's approximated weight is 10 kg. This category of infants is exposed to mercury from the goats' milk, water and dust. The mercury ingestion from water, goats' milk and dust are 0.0797 ug/d, 12.5 ug/d and 0.009 ug/d, respectively. The exposure per body weight is 0.00797 ug/kg_{bw}/d, 1.25 ug/ kg_{bw}/d and < 0.0001 ug/ kg_{bw}/d for water, goats' milk and dust respectively. Therefore, the total exposure for the infants on mother's milk is 1.2527 ug/ kg_{bw}/d while those on formula ingest 1.2589 ug/ kg_{bw}/d. The total exposure is higher than the RfD and MRL value of 0.3 ug/kg_{bw}/d. Toddlers are thus the most exposed group of infants to Hg. The goat's milk sample from Rooiberg contains 50 ug Hg/l compared to the mother's milk which contains no mercury. Thus, the consumption of goat's milk increases the level of mercury intake in this group of infants.

5.3.6.5 *Summary: Exposure of mercury to infants*

Table 5.4 gives overall values of Hg ingestion by infants from different sources of contamination that has been identified, through the developmental stages. The goats' milk makes the largest contribution towards the infants' Hg exposure. Since mother's milk does not contain Hg, it enhances the safety of the infants against excessive Hg exposure. The $HQ_{(MRL)}$ analysis for Hg indicated that only the toddlers will possibly be affected by the hazardous effect of Hg as it was the only group with HQ>1.

TABLE 5.4 SUMMARY: EXPOSURE TO MERCURY VIA INGESTION						
DATA & INFO	**WATER**	**MOTHER'S MILK**	**GOAT MILK**	**DUST/SOIL**	**TOTAL (** $ug/kg_{bw}/d$)	**HQ Exposure/0.3**
Rooiberg	0.1062	0 mg/l = 0 ug/l	0.05 mg/l = 50 ug/l	0.09 mg/kg = 90 ug/kg		
DWAF	1 ug/l					
PTWI = 5 ug/kg_{bw}/week.						
EPA chronic limit = 0.3 $ug/kg_{bw}/d$						
INFANT AGE & WEIGHT	**WATER**	**MOTHER'S MILK**	**GOAT MILK**	**DUST/SOIL**	**TOTAL**	
NEWBORNS Weight = 4 kg	600 ml	600 ml	0	0		
On mother's milk	-	0			0	-
on formula feeds	0.016 $ug/kg_{bw}/d$	0			0.016	0.053
SUCKLING INFANTS 8 kg	1 litre	1 litre		50 mg = 5 x 10^{-5} kg		
On mother's milk		0 $ug/kg_{bw}/d$		0.0006 $ug/kg_{bw}/d$.	0.0006	0.002
On formula feeds	0.01328 $ug/kg_{bw}/d$.			0.0006 $ug/kg_{bw}/d$.	0.0138	0.04
CRAWLING INFANTS 8.5 kg	For mother's milk – 250 ml; For formula feeds = 1 litre	For mother's milk = 750 ml; For formula feeds = 0		100 mg = 10^{-4} kg		
On mother's milk	0.0031 $ug/kg_{bw}/d$	0		0.00105 $ug/kg_{bw}/d$	0.00405	0.0138
On formula feeds	0.0125 $ug/kg_{bw}/d$	0		0.00105 $ug/kg_{bw}/d$	0.0135	0.045
TODDLERS 10 kg	For mother's milk = 250 ml +	500 ml	250	100 mg = 10^{-4} kg		
	For formula 750 ml	500 ml	250 ml	10^{-4} kg		
On mother's milk	0.0027 $ug/kg_{bw}/d$	0	1.25 $ug/kg_{bw}/d$	0.0009 ug/kgbw/d	1.2536	4.17
On formula feeds	0.00797 $ug/kg_{bw}/d$	0	= 1.25 $ug/kg_{bw}/d$	0.0009 ug/kgbw/d	1.2589	4.2

5.4 EXPOSURE TO LEAD

5.4.1 Findings

The table 5.4 below indicates the concentration of lead in the sampled water, soil, goats' milk and breast milk from Rooiberg. The others indicated are samples from Leeupoort (L), Gravelotte (G) and Kwagga (K).

TABLE 5.5 LEAD LEVELS IN SAMPLES FROM ROOIBERG

	Sample 1	**Sample 2**	**Sample 3**	**Date**	**Other**
Water (ppb)	0.7731 ug/l	0	0	Aug 2009	0(L); 0.1862(G); 0(G); 0(G); 0.276(K)
Soil A horizon (ppm)	29.52 mg/kg	24.27		Aug 2009	4.227(G); 4.252(G)
Goats milk (ppm)	0.27 mg/l	0.01		Aug 2009	0.348(L); 0.008(G); 0.006(G); 0.009(G); 0.009(G); 0.0093(K)
Mother's Milk (ppm)	0.08 mg/l			Aug 2009	0.004(G)

Ppb = ug/l; ppm = mg/kg or mg/l
G = Gravelotte; K = Kwagga; L = Leeupoort

According to the United States EPA (1986), the world production of lead is more than the commercial production of any toxic heavy metal. The concentrations of Pb in most top soils in the United States range from 10 to 30 ppm (USEPA, 1986). The Pb in soil at Rooiberg is close to the highest of these values.

Goats' milk from areas with industrial activities in Poland showed concentration of 50.1 ug/l to 157.9 ug/l (Gajewska *et al.,* 1990). The amount at Rooiberg far exceeds this value.

Lactose, a form of sugar found in milk including human milk enhances the absorption of lead in the gastrointestinal tract (Stephen and Waldron, 1975). As recorded by Galal-Gorchev (1993), the world estimated total weekly dietary intake by suckling infants ranges from 2 to 28 ug/kg_{bw}.

The concentration of mercury recorded in the sampled mothers' milk (80 ug/l) is higher than the reference dose of 20 ug/l indicated by Dorea and Donangelo (2006). It was also higher than the recorded average breast milk lead in Australia which is 0.55 ug/kg (Gulson *et al.*, 2001).

5.4.2 Exposure through the ingestion of water

The lead concentration in the sampled water in Rooiberg as indicated in the Table 5.5 is 0.77 ug/l. This is lower than the standards set by the DWAF (1996) and SABS (1999) of 10 ug/l and 50 ug/l, respectively. It thus appears that the water at Rooiberg is of good quality with respect to Pb.

Exposure to Pb is thus:

- *For newborns on formula feeds*:

  Consumption of newborns = 600 ml/d. The total amount of Pb taken in = 600 ml/d x 0.77 ug/l = 0.462 ug/d.

- *Breastfeeding and crawling infants on formula feeds:* Consumption = 1 l/d. Pb intake = 0.77 ug/d.

- *Crawling infants and toddlers not on formula feeds:* Consumption of water = 250 ml. Pb intake from water = 0.25 l/d x 0.77 ug/l = 0.1925 ug/d.

- *Toddlers on formula feeds:* Consumption = 750 ml/d. Pb intake = 0.75 l/d x 0.77 ug/l = 0.5775 ug/d.

5.4.3 Exposure through dust and soil

According to Roberts and Ott (2007), Pb contamination can be tracked into the house from outside. Breastfed infants are assumed to be exposed to 50 mg/d while the crawling/toddlers are exposed to 100 mg/d.

Exposure to Pb:

- *Breast feeding babies*: 5 x 10^{-5} kg/d x 29520 ug/kg = 1.476 ug/d.
- *Crawling infants and toddlers*: 10^{-4} kg/d x 29520 ug/kg = 2.952 ug/d.

5.4.4 Exposure through goat milk

The goats' milk is used as a supplementary food at the toddler stage for the infants. The lead intake through goats' milk is 0.25 l x 270 ug/l = 67.50 ug/d.

5.4.5 Exposure through mother's milk

Lead intake through the mother's milk is as follows:

- *Newborns*: 600 ml x 0.08 mg/l = 0.48 mg/d = 48 ug/d.
- *Breast feeding infants*: 1 l x 0.08 = 0.08 mg/d = 80 ug/d.
- *Crawling infants*: 750 ml/d x 0.08 mg/l = 0.06 mg/d = 60 ug/d.
- *Toddlers:* 500 ml/d x 0.08 mg/l = 0.04 mg/d = 40 ug/d.

5.4.6 Discussion

5.4.6.1 *Exposure by newborns*

The newborns are exposed to lead intake through the breast milk of their mother or through the water used in constituting the formula for the formula-fed babies. For infants feeding solely on the mothers' milk, the lead intake is 48 ug/d. For a 4 kg newborn baby is 12 ug/kg_{bw}/d. In the case of formula fed babies, the exposure is 0.464 ug/d and the exposure per body weight is 0.464 ug/d / 4 kg = 0.116 ug/kg_{bw}/d.

5.4.6.2 *Exposure by suckling infants*

The weight of the babies in this category as assumed by this study is 8 kg. The ingested lead concentration through the mothers' breast milk per body weight is (80/8 ug/kg_{bw}/d) = 10 ug/kg_{bw}/d for the consumption of 1 litre of mother's milk.

For the formula fed babies the lead exposure per body weight from the water (1 litre) used in making the formula is 0.7731/8 ug/kg_{bw}/d = 0.10 ug/kg_{bw}/d. The infants in this category however, are exposed to dust and the concentration of lead in the 5 x 10^{-5} kg of dust that the infants addressed (8 kg) here is exposed to is: 1.476/8 ug/kg_{bw}/d = 0.1845 ug/kg_{bw}/d.

The total lead ingested by the breastfed infant is (10 + 0.1845) = 10.1845 ug/kg_{bw}/d. For formula fed infants, the total exposure is (0.097 + 0.1845) = 0.1915 ug/kg_{bw}/d.

5.4.6.3 *Exposure by crawling infants*

The crawling infant is assumed to have the average weight of 8.5 kg. The exposure routes are from the breast milk, water-based feed and dust. It was assumed that they consume 750 ml of the mother's milk, 250 of water based drinks and are exposed to 100 mg of dust per day. Thus the lead consumed daily is: lead from mother's milk is 60 ug/d; from the water based drinks: 0.1925 ug/d and from the dust, 2.952 ug/d. The intakes per body weight of mother's milk, water based drinks and dust are: 7.059 ug/kg_{bw}/d, 0.0226 ug/kg_{bw}/d and 0.3473 ug/kg_{bw}/d respectively. Thus, the total exposure by crawling infants on mother's milk is 7.4289 ug/kg_{bw}/d. The total exposure for formula-fed suckling infant is 0.4373 ug/kg_{bw}/d.

5.4.6.4 *Exposure by toddlers*

Toddlers are infants who are in their 12^{th} month. The average weight assumed for these infants is 10 kg. They are exposed to Pb through the mothers' milk, goats' milk, water based foods and dust. The concentration of lead toddlers will be exposed to in the water intake is 0.19 ug/l, mothers' milk is 40 ug/d, in the goats' milk is 67.50 ug/d, water based foods dust is 2.952 ug/d. The intakes per body weight for water, mothers' milk, goats' milk, water based foods and dust are therefore, 0.019 ug/l; 4 ug/kg_{bw}/d, 6.75 ug/kg_{bw}/d, 0.2952 ug/kg_{bw}/d, respectively. The total lead exposure to toddler on mother's milk is 11.0642 ug/kg_{bw}/d and that on formula is 7.103 ug/kg_{bw}/d.

5.4.6.5 *Summary: Exposure of lead to infants*

The total lead intake by the infants daily are high especially in the older infants that are exposed to goats' milk and dust. The dust has the highest lead concentration followed by the goats' milk. It is of interest that the daily intake of the newborn on mothers' milk is much higher than the formula-fed newborns. There is no RfD or MRL value for Pb because of its high toxicity. Thus, there is no value for $HQ_{(MRL)}$.

However, values for $HQ_{(TDI)}$ were calculated. These were not compared to the standard value of 1 since this only applies to HQs calculated using MRL or RfD values.

At Rooiberg, mother's milk contributes most to the Pb ingestion by infants, except in toddlers where goat's milk also plays a major role. The contributions of Pb in water and dust are not significant. It is not surprising that breast-fed infants are at greater risk than formula-fed infants. In general then, breast-fed groups are more exposed to Pb than formula-fed crawling infants.

Table 5.6 SUMMARY: EXPOSURE TO LEAD VIA INGESTION						
DATA & INFO	**WATER**	**MOTHER'S MILK**	**GOAT MILK**	**DUST/SOIL**	**TOTAL ($ug/kg_{bw}/d$)**	$HQ_{(TDI)}$ **Exposure/3.6**
Rooiberg	0.7731ug/l	0.08mg/l = 80 ug/l	0.27mg/l = 270 ug/l	29.52 mg/kg = 29520 ug/kg		
DWAF	10 ug/l					
PTWI = 25 ug/l bw/week						
INFANT AGE WEIGHT	**WATER**	**MOTHER'S MILK**	**GOAT MILK**	**DUST /SOIL**	**TOTAL ($ug/kg_{bw}/d$**	$HQ_{((TDI)}$ **Exposure/3.6**
NEWBORNS Weight = 4 kg	600 ml	600 ml	0	0		
on mother's milk	-	12 ug/kgbw/d	0	0	12 ug/kgbw/d	3.3
on formula feeds	=0.116 ug/kgbw/d	0			0.116 ug/kgbw/d	0.03
SUCKLING INFANTS 8 kg	1 litre	1 litre		50 mg = 5×10^{-5} kg		
On mother's milk		10 ug/kgbw/d		0.1845 $ug/kg_{bw}/d$.	10.1845 $ug/kg_{bw}/d$.	2.83
On formula feeds	0.10 $ug/kg_{bw}/d$	0	0	0.1845 $ug/kg_{bw}/d$.	0.2845 ug/kgbw/d	0.079
CRAWLING INFANTS 8.5 kg	For mother's milk – 250 ml; For formula feeds = 1 litre	For mother's milk = 750 ml; For formula feeds = 0		100 mg = 10^{-4} kg		
On mother's milk	0.023 ug/kgbw/d	7.07 ug/kgbw/d		0.3473 ug/kgbw/d	7.44 ug/kgbw/d	2.06
On formula feeds	0.091 ug/kgbw/d	0	0	0.3473 ug/kgbw/d.	0.4383 ug/kgbw/d	0.12
TODDLERS 10 kg	For mother's milk = 250 ml	500 ml	250	100 mg = 10^{-4} kg		
	For formula 750 ml		250 ml	10^{-4} kg		
On mother's milk	0.02 ug/kgbw/d	= 4 ug/kgbw/d	6.75 ug/kgbw/d	0.2952 ug/kgbw/d	11.064 ug/kgbw/d	3.07
On formula feeds	0.058 $ug/kg_{bw}/d$	0	6.75 $ug/kg_{bw}/d$	0.2952 ug/kgbw/d	7.103 ug/kgbw/d	1.97

5.6 SUMMARY OF THE THREE TOXIC ELEMENTS HAZARD EFFECT

Table 5.7 below gives the overview of the hazard quotients of the three toxic elements. The values with the superscript 'a' indicates a high possibility of toxic effect of the contaminant on the infants, the values with the superscript 'b' indicates that there are is no possibility of the effect of toxicity of the contaminants on the infants.

Of the three minerals, exposure to As is the most significant. Infants on mother's milk may also be exposed to high levels of Pb. Table 5.7 also indicates that only the toddlers will possibly experience health problems due to exposure to Hg.

TABLE 5.7 THE HAZARD QUOTIENTS FOR ARSENIC, MERCURY AND LEAD

STAGE	**STAGE**	**ARSENIC**		**MERCURY**		**LEAD**		**TOTAL**
		HQ (RfD) Exposure /RfD RfD = 0.3 ug/kg/d	HQ (TDI) = Exposure /PTDI PTDI = 2.1 ug/kg/d	HQ (RfD) Exposure /RfD RfD = 0.3 ug/kg/d	HQ (TDI) = Exposure /PTDI PTDI = 0.7 ug/kg/d	HQ (RfD) Exposure /RfD RfD = no value	HQ (TDI) = Exposure /PTDI PTDI = 3.6 ug/kg/d	HI
Breast feeding	MM	**25[a]**	**3.43**	0	0		**3.3**	6.73
	FF	**6.83 [a]**	0.98	0.053 [b]	0.023		0.03	1.033
Suckling	MM	**21.23 [a]**	**3.03**	0.002 [b]	0.0008		**2.83**	5.86
	FF	**4.3 [a]**	0.61	0.04 [b]	0.0197		0.079	0.7087
Crawling	MM	**16.77 [a]**	**2.40**	0.0138 [b]	0.005		**2.06**	4.465
	FF	**6.1 [a]**	0.87	0.045 [b]	0.019		0.12	1.009
Toddlers	MM	**23.43 [a]**	**3.35**	**4.17 [a]**	**1.790**		**3.07**	8.21
	FF	**17.37 [a]**	**2.48[a]**	**4.2 [a]**	**1.798**		**1.97[a]**	6.248

[a] – HQ >1

[b] – HQ <1

In each of the growth stages of the infants in Rooiberg, the formula-fed indicated less likely to be effected by As, Hg and Pb. As described by the Natural Resource Defense Council (NRDC, 2005), metals rarely accumulate in fatty tissues. Their excretion rates into the milk are therefore low (http://www.nrdc.org/breastmilk/lead.asp, 2009). Nevertheless, unacceptably high levels of Pb are found in mother's milk. Goat's milk contains high concentrations of both Pb and Hg. Arsenic is not a metal and clearly bio-accumulates in mother's and goat's milk.

CHAPTER 6

SUMMARY, CONCLUSION AND RECOMMENDATIONS

6.1 SUMMARY AND CONCLUSION

Arsenic (As), mercury (Hg) and lead (Pb) are widely accepted as being the most toxic minerals known to man.

The Limpopo province of South Africa is noted for it rich mineral wealth, *inter alia*, gold, copper, antimony, zinc and iron. The toxic minerals, arsenic, mercury and lead, are associated with these minerals and with mining activity. It is therefore possible that the environment and human beings in such areas might be at risk of contamination. This study was concerned with establishing the extent of exposure of various components of the environment - water, soil, plants, animals and humans - to the three toxic minerals. The ultimate aim of the study was to assess whether arsenic, mercury and lead pose a threat to the health status of infants living in the Limpopo Province. The study focused on infants since they are the most vulnerable sector of the population. The exposure pathway was limited to ingestion, and the potential sources of contamination were water, food and dust. The food component comprised mother's milk as well as goat's milk and water used as basis for formula feeds.

Two potentially contaminated areas were identified based on their geological characteristics. One such study area was located in the vicinity of old tin mines near Leeupoort in the extreme south of the Province and the other, near a mercury mine, close to Gravelotte in the east. The villages where the study was conducted were identified by random selection. They were Rooiberg and Leeupoort in the south (Study area A) and Gravelotte and Kwagga in the east (study area B). Samples of water, soil, plants, goat's milk and mother's milk were collected and sent for analysis.

In order to assess whether the infants at Rooiberg are at risk of exposure to arsenic, mercury and lead, the ingestion of these elements was determined and compared with the respective Provisional Tolerable Weekly Intake (PTWI) as set by the United Nation's Food and Agricultural Organisation (FAO) and the WHO. Minimal Risk Levels (MRLs) were also used to determine the extent of exposure. Hazard Quotients were calculated to quantify the extent of potential risk to which infants are exposed during their different developmental stages.

A number of assumptions were made in the study. They were: That the development rate and growth of infants in Rooiberg follow the general growth curves milestones as set out by Health24, as shown in Fig 5.1; goat's milk is usually used as a substitute for mother's milk once weaning commences; the formulas used to replace or augment mother's milk is free of arsenic, mercury and lead; and that the types and amounts of foods ingested vary from one stage to another, as does the exposure to dust. Here is was assumed that newborns (3 days) weigh 4 kg and consume 600 ml mother's milk (MM) per day; suckling infants (5 months) weigh 8 kg and consume 1 litre mother's milk per day and are exposed to 50 mg dust/day; Crawling infants (7 months) weigh 8.5 kg and consume 750 ml MM/d, 250 ml water and are exposed to 100 mg dust per day. Lastly, toddlers (12 months) weigh 10 kg; drink 500 ml MM, 250 ml water and 250 goat's milk per day while being exposed to 100 mg dust every day. It should also be noted that due to the limited number of samples collected the results only provide insight into the possible exposure to the infants and are not irrefutable evidence of risk. This research can only be seen as a preliminary study.

It was found that both the groundwater and the surface water were contaminated with arsenic at Rooiberg, Leeupoort and Gravelotte and with mercury at Gravelotte. Lead concentrations did not exceed water quality standards at any of the sites. Arsenic was the most prevalent of the minerals in all the soils and is probably of natural (geological) origin. Both the arsenic and lead concentrations were highest at Rooiberg. The high concentrations of the minerals in the dust on plants point to an anthropogenic origin, probably related to mining activity. Although plants contain much lower amounts of

minerals than soils, the lead and mercury levels are high when compared to those available from other countries. It should be kept in mind that leaves of plants also serve as food for animals such as goats, as well as for many insects, which in turn, are eaten by birds. Most animals are exposed to the dust on the leaves of plants. Toxic minerals may thus enter the food chain and bioaccumulate from one trophic level to the next. The presence of arsenic, mercury and lead in goat's milk is a clear indication of contamination in the food chain. The amount of arsenic in goat's milk is between 2 and 8 times higher than that in other countries. It also has higher mercury than found elsewhere. The lead concentration is more than five times that of goats in highly industrialised and polluted parts of Poland.

The amount of arsenic in mother's milk at Rooiberg is more than twice the amount found in arsenic contaminated countries such as the Fillipines and more than 70 times the average in most countries. Lead levels in breast milk are also much higher than elsewhere. It is only with regard to mercury, that the breast milk is free of contamination.

Infants go through different development stages, from newborns, to breast feeding infants, to a crawling phase and finally to toddlers – each having different dietary requirements, dietary intakes and hence potential exposure levels. In this study, the exposure was determined separately for each development stage for each of the three toxic minerals.

The results of the analyses showed that dust played a minor role as a source of exposure to the three toxic minerals. Furthermore, the greater the water component in the diet, the lower the risk of exposure to the toxic minerals. Goat's milk is a significant source of contamination with regards to arsenic, mercury and lead while mother's milk is contaminated with arsenic and lead and is hence as source of toxicity. Therefore infants on formula feeds have less chance of ingesting high levels of arsenic and lead, provided that the formula food itself does not contain these minerals. Paradoxically, it is the babies on mother's milk who are protected from exposure to mercury.

In general, breast feeding infants are most at risk of exposure to arsenic and lead. There are only small differences in the levels of risk experienced during the different infant development stages, with the risk levels of toddlers and newborns being slightly higher than for suckling and crawling infants. This is an important finding since breast feeding is encouraged by most health workers. Although breast feeding is indeed important, helping the infant to develop immunity against many diseases, cognisance should be taken of the possible toxic mineral content of the milk.

The study has indicated that arsenic, lead and mercury may be potentially harmful to plants and animals in the study areas, especially in study area A. Arsenic and lead may cause health problems to infants at Rooiberg. This confirms findings by Sami and Druzynski (2003) regarding the potential of arsenic contamination in this area. Other tin-mining areas may also be at risk.

The most serious limitation of the study was the small number of samples analysed as well as the lack of information on the actual stage of lactation, the speciation of the minerals, the exact age at which the babies were weaned from mother's milk, the proportion of goat's milk, mother's milk and water-based formulae/food used for toddlers and the amount of dust to which at different stages are exposed.

6.2 RECOMMENDATIONS

Due to the severe limitations in the number of samples obtained for analysis, this study must be seen as a pilot for a much more detailed and comprehensive research project.

However, as with most research, the answering of the research question leads to a myriad of other questions to be answered and research to be conducted.
The following present some of these:

- More detailed work should be carried out in the study areas and more samples collected and analysed.

- Other potentially toxic minerals such as nickel and chromium, should be included in such a study.
- Similar studies should be carries out in other areas with mining activities, especially those identified as being potentially hazardous.
- The synergistic effect of more than one toxic mineral should be determined.
- A comprehensive Human Health Risk Assessment should be conducted considering other exposure route such as inhalation and the dermal contact
- The impact of arsenic, lead and other mineral toxins on animals, notably goats, should be assessed.

If further studies confirm the results obtained here, several mitigating measures should be instituted to limit any potential health problems. For example, children should not be given goat's milk at an early age. Consumption of this milk would, however, be considered safe for adults.

It is vital that more tests be conducted on the trace elements content of mother's milk at Rooiberg without delay - either to confirm or refute these findings. The appropriate health authorities should be informed of the results without delay so that measures can be instituted to reduce be impact of the toxic elements on the Rooiberg community.

REFERENCES

Abballe A., Ballard T.J., Dellatte E., di Domenico A., Ferri F., Fulgenzi A.R., Grisanti G., Iancovella N., Ingelido A.M., Malisch R.M., Porposa M.G., Risica S., Ziemacki G. and De Felip E.,2008. Persistant environmental contaminants in human milk: Concentrations and time trends in Italy. *Chemosphere,* 73(1), 220-227.

Adelzadeh, A., & Ohiorhenuan, J. 2004. The Challenge of Sustainable Development in South Africa: Unlocking Peoples Creativity (Report No. 2003). Cape Town: Oxford University Press.

Agency for Toxic Substance Disease Registry, 1990, 'Toxicological Profile for Lead,' ATSDR/TP-88/17. Atlanta, GA: Agency for Toxic Substances and Disease Registry.

Agency for Toxic Substance Disease Registry (ATSDR) 1999. Toxicological Profile. Retrieved November 12, 2009 from http://www.atsdr.cdc.gov/tfacts46.html.

Agency for Toxic Substances and Disease Registry (ATSDR). 1999. Toxicological profile for mercury. Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service.

Agency for Toxic Substances and Disease Registry, 1992. Toxicology profile for cadmium U.S department of health and human service. Public Health Service. Atlanta GA: ATSDR

Agency for Toxic Substances and Disease Registry, 2007. CERCLA Priority List of Hazardous Substances. Retrieved January 8, 2008 form http://www.atsdr.cdc.gov/cercla/07list.html.

Aggette, J. & Spell, A.C. 1978. Arsenic from geothermal sources. *New Zealand Research and Development report.* No 35. New Zealand: Department of Agriculture.

Alloway, B. J. 1995. *Heavy Metals in Soils.* London: Springer.

Alonso, E., Callejon, M., Jimenez, J.C. & Ternero, M. 2000. A voltametry procedure with medium exchange for the speciation of heavy metals in waste water of a sewage treatment plant. *Electroanalysis*, 13, 917-920.

Al-Saleh I, Khalil MA, Taylor A. Lead, erythrocyte protoporphyrin, and hematological parameters in normal maternal and umbilical cord blood from subjects of the Riyadh region, Saudi Arabia. Arch Environ Health 1995; 50:66–73.

American Academy of Pediatrics, 2005. Breastfeeding and the use of human milk. *Pediatrics.* 115(2), 496-506.

Anke, M. 1986: *Arsenic. Trace Elements in Human and Animal Nutrition.* (5th ed.). Orlando: Academic Press.

Antunović, Z., Bogut, I., Senčić, 1.D., Katić, D. & Mijić, P. 2005. Concentrations of selected toxic elements (Cadmium, lead, mercury and arsenic) in ewe milk in dependence on lactation stage. *Czech. J. Anim. Sci.,* 50(8), 369–375.

Arain, M.B., Kazi, T.G., Baig, J.A., Jamali, M.K., Afridi, H.I., Shah, A.Q., Jalbani, N. & Sarfraz, R.A., 2009. Determination of arsenic levels in lake water, sediment, and foodstuff from selected area of Sindh, Pakistan: Estimation of daily dietary intake. *Food and chemical toxicology*, 47, 242-248.

Arbestain, M.C., Rodríguez-lado, L., Bao, M. & Macías, F. 2009. Assessment Of Mercury-polluted Soils Adjacent to an Old Mercury-Fulminate Production Plant. *Applied and Environmental Soil Science*. 8. Retrieved February 3, 2010 from http://www.hindawi.com/journals/aess/2009/387419.html.

Aschner, M. & Clarkson, T.W. 1988. Distribution of mercury 203 in pregnant rats and their fetuses following systemic infusions with thiol- containing amino acids and glutathione during late gestation during late gestation. *Tetratology,* 38, 145-155.

Aschner, M., Yao, C.P., Allen, J.W. & Tan, K.H. 2000. Methylmercury alters glutamate transport in astrocytes. *Neurochem. Int.,* 37, 199-206.

Astolfi, E., Meccagno, A., Fernandez, J.C.G., Vaccaro, R. & Stimola, R. 1981. Relationship between arsenic in drinking water and skin cancer. *Biol. Trace Elements Res*., 3(2), 133-143.

Baeyens, W., Ebinghaus, R. & Vasilien, O. (eds). 1996. *Global regional mercury cycles: sources, Flexes and mass balances*. Kluwer: Dordrecht.

Bahemuka, T.E. & Mobufu, E.B. 1999. Heavy metals in edible green vegetables grown along the site of the Simza and Msimbazi River in Dar es Salam in Tanzania. *Food Chem*., 66, 63-66.

Banasova, V. & Holub, Z. 1999. The impact of the exploitation and processing of mercury ore on the vegetation under long term influence of lead – copper emissions. *Ekologia,* 6, 101-111.

Barcan, V. & Kovnatsky, E. 1998. Soil surface geochemical anomaly around the copper-nickel metallurgical smelter. *Water, Air & Soil Pollut.,* 130, 197-218.

Barrett J.R. 2002. The Breast Milk Biomarker: Samples Provide Clues to Exposures. *Environmental Heath Perspective*, 110(6), 311.

Baur, W.H. & Onishi, H. 1978. Arsenic. In Wedepohl (ed), 1978, *Handbook of Geochemistry*. Berlin: Springer.

Beauford, W., Barbar, J. & Barringer, A.R. 2008. Uptake and Distribution of Mercury within Higher Plants. *Physiologia Platarum.*, 39(4), 261-269.

Beresford, N.A., Crout, N.M.J. & Mayes, R.W. 2001. The transfer of arsenic to sheep tissues. *Journals of Agricultural Science.* 136, 331-344.

Beresford, N.A., Crout, N.M.J., Mayes, R.W., Maceachern, P.K., Dodd, B.A., Barnett, C.L. & Lamb, C.S. 1999. Transfer of Cadmium and Mercury to sheep tissues. *Environ. Sci. Tech.*, 33, 2395-2402.

Berg, M., Trans, H.C., Nguyeu, T.C., Pham, M.V., Scherteuleib, R. & Giger, W. 2001. Arsenic contamination of groundwater and drinking water in Vietnam: a human health threat. *Environ. Sci. Technol.*, 35, 2621-2626.

Berzas, N.J., Garcia-Bermejo, L.F., Rodriguez, M., & Doimeadios, R.C. 2003. Distribution of mercury in aquatic environment at Almaden, Spain. *Environ. Pollut.*, 122, 261-271.

Bidstrup, F.L. 1964. *Toxicity of mercury and its compounds*. Amsterdam: Elsevier.

Bires, J., Dianovsky, J., Bartko, P. & Juhasova, Z. 1995. Effects on enzymes and the genetic apparatus of sheep after administration of samples from industrial emissions. *Biol. Metals*, 8, 53–58.

Bissen, M. & Frimmel, F.M. 2003. Arsenic - a review. Part 1: occurrence, toxicity, speciation, mobility. *ActaHydrochim. Hydrobiol.*, 31, 9-18.

Borgono, J.M. & Greiber, R. 1997. Epidemologic study of arsenic poisoning in the city of Antofagasta. *Revista Medica De Chile*, 99(9), 702-707.

Brandsetter, A., Lombi, E., Wenzel, W.W. & Andriano, D.C. 2001. Arsenic contaminated soils: In D.C. Andriana (ed) *Risk assessment in trace elements in terrestrial environments: biogeochemistry bioavailability and risk of metals* (715-737). New York: Springer.

Brun, L.A., Maillet, J. & Hinsinger, P.M. 2001. Evaluation of copper availability to plants in copper contaminated vineyard soils. *Environ. Pollut*., 111, 293-302.

Caggiano, R., Sabia, S., D'Emilio, M., Anastasio, A., Ragosta, M. & Paino, S. 2004. Metal level in fodder, milk, dairy products and tissue sampled in ovine farms of southern Italy. *Environmental Research*, 99, 48-57.

Cairncross, B. 2005. *Field guide to rocks and minerals of Southern Africa*. South Africa: Struik.

Carton, J.A. 1988. Satunismo. *Med. Clin*., 91, 538-540.

Casey, C.E., Smith, A. & Zhang, P. 1995. Micro-minerals in human and animal milks. In Lawrence R.A. & Friedman L.R. *Contaminants in milk: Handbook of milk composition.* New York: Academic Press.

Cava, M. P., Rodenas, T.E., Morales, R.A., Cervera, M.L. & Guardia, M. 2004. Cold vapour atomic fluorescence determination of mercury in milk by slurry sampling using multi commutation. *Analytical Clinical acta.,* 506, 145-153.

Chien S., Bialer, M., Solanki, B., *et al.,* (2006. Pharmacokinetics interaction study between the new antiepileptic and CNS drug RW333369carbamazepine in healthy adults. *Epilepsia*, 47, 1840-40.

Chronic Arsenic Poisoning: Pictures of Sufferers retrieved August 8, 2008 from http://phys4.harvard.edu/~wilson/arsenic/pictures/arsenic_project_pictures2.html.

Clark, A. 1993. *Hey Mineral Index of Mineral*. London: Chapman & Hall.

Clarkson, T.W. 1987. Metal toxicity in the central nervous system. *Environ. Health perspectives*, 75, 59-64.

Clarkson, T.W., Magos, L. & Mayers, G.J. 2003. The toxicology of mercury – current exposures and clinical manifestations. *N. Engl. J. Med.*, 349, 1731-1737.

Concha, G., Vogler, G., Nermell, B. & Vahter, M. 1998. Low-level arsenic excretion in breast milk of native Andean women exposed to high levels of arsenic in the drinking water. *Int. Arch Occup. Environ. Health*, 71(1), 42–46.

Coni, E., Bocca, A., Coppolelli, C., & Trabalza, M. 1996. Minor and trace element content in sheep and goat milk and dairy products. *Food Chem.*, 57, 253-260.

Correll, R.L., Smith, E., Naidu, R., Huq, S.M.I. & Barnes, M.B. 2004. Estimation of human arsenic uptake via the food chain in Bangladesh (Report No. 04/194), Adelaide: CSIRO.

Counter, S.A. & Buchanan, L.H. 2004. Mercury exposure in children: a Review. *Toxicology and Applied Pharmacology*, 198, 209-230.

Crout, N.M.J., Beresford, N.A., Dawson, J.M., Soar, J. & Mayes, R.W. 2004. The transfer of As, Cd, and Hg to the milk and tissues of Dairy cattle. *Journal of Agricultural Science*, 142, 203-212.

Dabeka, R.W., Karpinski, K.F., McKenzie, A.D., & Bajdik, C.D. 1986. Survey of lead, cadmium and fluoride in human milk and correlation of levels with environmental and food factors. *Food Chem. Toxic*, 24, 913-921.

Dabeka, R.W., McKenzie, A.D. & Lacroix, G.M.A. 1987. Dietary intakes of lead, cadmium, arsenic and fluoride by Canadian adults: a 24-hour duplicate diet study. *Food additives and contaminants,* 4, 89-101.

Dakeishi, M., Murata, K. & Grandjean, P. 2006. Long-term consequences of arsenic poisoning during infancy due to contaminated milk powder. *Environ. Health,* 5, 31.

Dalvie, M.A. & Ehrich, R. 2006. Community mercury levels in the vicinity of peri-urban waste disposal sites ad fossil fuel burning operations. *Environment International*, 32(4), 493-499.

Data, S.P., Biswas, D.R., Sahara, N., Ghosh, S.K. & Rattan, R.K. 2000. Effect of long term application of sewage effluent on organic carbon, bioavailability Phosphorus, potassium and metals status of soil and uptake of heavy metals by crops. *J. Indian Soc. Soil Sci*., 48, 836-839.

Davis, B.E. 1995. Lead. In B.J. Alloway (ed.) *Heavy metals in soil* (2nd ed.). Glasgow: Blackie Academic and Professional.

Department of Water Affairs and Forestry (DWAF), 1995. Draft report on Radioactivity in water sources, a preliminary survey. *Domestic use.* 2nd edition. Pretoria: DWAF.

Department of Water Affairs and Forestry (DWAF), 1996. South African Water Quality Guidelines (2nd ed.). Pretoria: DWAF.

Department of Water Affairs and Forestry, 1996a. *Water quality guidelines aquatic ecosystem use* (1st ed.). Pretoria: DWAF.

Detoxamine chelating therapy. Proven by science. Retrieved November 13, 2008 from *www.detoxamin.com/otherprod.html*.

Dettwyler, K. nd. Breastfeeding a toddler. *'Essence' magazine*, 39, 1.

Dorea, J.G. & Donangelo, C.M. 2006. Early (in uterus and infants) exposure to mercury and lead. *Clinical Nutrition*, 25, 369-376.

Dorea, J.G. 2004. Vegetarian diets and exposure to organochlorine pollutants, lead and mercury. *Am. J Clin. Nutr.*, 80, 237-8.

Drexler, H. & Schaller, K.H. 1998. The mercury concentration in breast milk resulting from amalgam filling and dietary habit. *Environ. Res.*, 77, 124-129.

Dwivedi, S.K., Swarup, D., Dey, S. & Patra, R.C. 2001. Lead poisoning in cattle and buffalo near primary lead-zinc smelter in India. *Vet. Hum. Toxicol*., 43, 74-75.

Elias, R.W. 1985. *Lead Exposures in the Human Environment*. Boston: Elsievier.

Elsgaard, L., Peterson, S.O. & Debosz, K. 2001. Effects and risks assessment of linear alkyl benzene sulfonates in agricultural soil I. short term effect on soil microbiology. *Environ. Toxicol. Chem*., 20(80), 1656-1663.

Environment Agency, n.d. *Information on Land Quality in England including BackgroundLevels of Organic and Inorganic Contaminants* (R&D Technical Report P291). Swindon, Wilts: WRC

Environmental Agency, 2002. Microbiology of drinking water (200) part I water quality and public health. Retrieved January 8, 2008 from http://www.environment-agency.gov.uk/static/documents/Research/mdwpart1.pdf

Environmental Protection Agency, 1997. Relative bioavailability of arsenic in mining wastes (Document control no. 4500-88-AORH). Denver, CO: U.S. Environmental Protection Agency.

Ettinger, A.S., Téllez-Rojo, M.M., Amarasiriwardena, C., Bellinger, D., Peterson, K., Schwartz, J., Hu, H. & Hernández-Avila, M. 2004. Effect of Breast Milk Lead on Infant Blood Lead Levels at 1 Month of Age. *Environmental Health Perspective*, 112(14), 245-275.

Falomir, P., Alegria, A., Barbera, R., Farre, R. & Lagarda, M.J. 1999. Direct determination of lead in human milk by electro thermal atomic aborption spectrometry. *Food Chem*., 64, 111-113.

Fan, A.M. 1987. "Mercury." In L. Fishbein, A. Furst, & Mehlman, M.A. (eds.). *Genotoxic and Carcinogenic Metals: Environmental and Occupational Occurrence and Exposure.* Princeton, N.J.: Princeton Scientific Publisher.

Fängström, B., Moore, S., Nermell, B., Kuenstl, L., Goessler, W., Grandér, M., Kabir, I., Palm, B., Arifeen, S., & Vahter, M. 2008. Breast-feeding protects against arsenic exposure in Bangladeshi infants. *Environ. Health Perspect*., 116(7), 963–969.

FAO/ World Health Organization, 1983. Arsenic. *WHO Food Additive Series 18*. Geneva: WHO.

FAO/ World Health Organization, 1986. Lead (evaluation of health risks to infants and children). *WHO Food Additive Series 21*. Geneva: WHO. Retrieved January, 8 2009, 2009 from http://www.inchem.org/documents/jecfa/jecmono/v21je16.htm.

FAO/ World Health Organization, 1988. *Arsenic. WHO Food Additive Series 24*. Geneva: World Health Organization.

Faust, S.D. & Aly, O.M. 1981. *Chemistry of Natural Waters.* Boston: Ann Arbor Science.

Feller, E. 1983. Indagine preliminare sul contenuto in piobo Del latte. *Conferto alla provincial di Trento*, 9(6), 446-448.

Fergusson, J.E. 1990. *The heavy element: Chemistry*. Oxford: Peregamon press.

Fillip, Z. 2002. International approach to assessing soil quality by ecologically related biological parameters. *Agric. Ecosyst. Environ.*, 88(2), 169-174.

Francisco, N., Ruiz-Troya, J.D. & Aguera, E. 2003. Lead and lead toxicity in domestic and free living birds. *Avian Pathology*, 32, 3-13.

Franco, J.L., Teixeira, A., Meotti, F.C., Ribas, C.M., Stringari, J., Pomblum, S.C.G., Moro, Â.M., Bohrer, D., Bairros, A.V., Dafre, A.L., Santos, A.R.S. & Farina, M. 2006. Cerebellar thiol status and motor deficit after lactational exposure to methylmercury. *J. Environ. Res*., 2(3): 22-28.

Frumkin, H., Letz, R., Williams, P.l., Gerr, F., Pierce, M., Sanders, A., Elon, L., Mannings, C.C., Woods, J.S., Hertzberg, V.S., Mueller, P. & Taylor, B.B. 2001. Health effects of long term mercury exposure among chloralkali plants workers. *Am. J. Ind. Med*., 39, 1-18.

Gajewski R., Nabrzyski, M., Ganowiak, Z. & Kucharska, D. 1996. Evaluation of the health aspects of goat milk. *Rocz Panstw Zakl Hig.*, 47, 295-301.

Galal-Gorchev H. 1993. Dietary intake, levels in food and estimated intake of lead, cadmium, and mercury. *Food Addit. Contam*., 10, 115–28.

Galal-Gorchev, H. 1991. Dietary intake of pesticide residues, cadmium, mercury, and lead. *Food Addit. Contam*., 8(6), 793-806.

Galster, W.A. 1976. Mercury in Alaska Eskimo mothers and infants. *Environ. Health Perspect.*, 15, 135-140.

Gao, C. 1997. The association of the geological collaboration in Japan. *Chigaku dantai kenkyukai*, 11, 28-33.

Gnamus, A. & Horvat, M. 1999. *Mercury in terrestrial food webs of Idrija mining area*. Ebinghaus, R., Turner, R. Lacerda, L.D., Vasiliev, O. & Salomons, W. (eds). Heidelberg: Springer.

Gothberg, A., Greger, M., Holm, K. & Bengtsson, B. 2004. Influence of Nutrient Levels on Uptake and Effects of Mercury, Cadmium, and Lead in Water Spinach. *J. Environ. Qual*., 33, 1247-1255.

Goyer, R.A. 1997. Toxic and essential metal interactions. *Ann. Rev Nutr*., 17, 37-50.

Grandjean, D., White, R.F., Nielsen, A., Cleary, D. & Santos, E.C. 1999. Methylmercury neurotoxicity in Amazonian children downstream from gold mining. *Environ. Health perspect*., 101, 587-591.

Gray Environmental, Inc. 2002. Lead in Garden Soils and Plants. Retrieved August 4th, 2009 from *http://www.grayenvironmental.com/lead_in_garden_soil .htm*

Gregorio, P & Saracusano, C. 1997. Contaminazione da piombo nel latte vaccine. *Ig Mod,* 69, 296-305.

Guler, Z. 2007. Levels of 24 minerals in local goat milk, its strained yoghurt and salted yoghurt. *Small Ruminant Res.*, 71(1-3), 130-138.

Gulson, B.L. n.d. Fact sheet for medical professionals: Lead in breast milk. *Lead Action News,* 6: 2.

Gulson, B.L., Gray, B., Mahaffey, K.R., Jameson, W., Mizon, K.J., Patison, N. & Korsch M.J. 1999. Comparison of the rates of exchange of lead in the blood of newly born infants and their mothers with lead from their current environment. *J. Lab. Clin. Med.*, 133, 171-178.

Gundacker, C. & Zödl, B. 2008. Heavy metals in breast milk: implications for toxicity. In V.R. Preedy & R. Watson (eds.). *Reviews in food and nutrition toxicity.* Australia: Taylor & Francis.

Gyekye, A.B. & Akinboade, O.A**.** 2003. A Profile of Poverty in the Limpopo Province of South Africa. *Eastern Africa Social Science Research Review,* 19(2): 89-109.

Hallen, I.P. 1995. *Lead excretion in breast milk and transfer to suckling offspring.* Uppsala Biomedical centre, Sweden. (MSc-Thesis).

Hansen, J.M., Zhang, H. & Jones, D.P. 2006. Differential oxidation of thioredoxin -1, thioredoxin-2 and glutathione by metal ions. *Free Radic. Biol. Med*., 40, 138-145.

Harada, M. 1995. Minamata disease: Methylmercury poisoning in Japan caused by environmental pollution. *Crit. Rev. Toxicol.*, 25, 1-24.

Heaton, A.C.P., Rugh, C.L. Wang, N. & Meagher, R.B**.** 1988. Phytoremediation of mercury- and methylmercury-polluted soils using genetically engineered plants. *Journal of Soil Contamination*, 7(4), 497-509.

Hindmarsh, J.T., Abernethy, C.O., Peters, G.R. & McCurdy, R.F. 2002. Environmental Aspects of Arsenic Toxicity. In B. Sarkar (ed). *Heavy Metals in the Environment*. 217-229. New York: Marcel Dekkar, Inc.

Holson, J.F., Desesso, J.M., Jacobson, C.F. & Farr, C.H. 2000. Appropriate use of animal models in the assessment of risk during pre-natal development: an Illustration using inorganic arsenic. *Teratology*, 62, 51-71.

Hongyu, L., Anne, P & Bohan, L. 2005. Metal contamination of soils and crop affected by the Cehenzhou lead/ zinc mine spill (Hunan, China). *Sci. Total Environ*., 339: 153-166.

Houpert, P., Mehennaoui, S., Federspiel, B., Kolf-Clauw, M., Joseph-Enriquez, B. & Milhaud, G. 1997. Transfer of cadmium from feed to ewe food products: variations in transfer induced by lead and zinc. *Environ. Sci*., 5, 127–138.

Human Science Research Council (HSRC) 2000. *A demographic, socio-economic and cultural profile of South Africa.* Pretoria: HSRC Press.

Ikeda, M, Zhang, Z.W, Shimbo, S, Watanabe, T, Nakatsu, H. & Moon, C.S. 2000. Urban population exposure to lead (Pb) and cadmium in East and South East Asia. *Sci. Total Environ.*, 249, 373-384.

International Programme on Chemical Safety (IPCS), 1990. *Methylmercury*. (Environmental Health Criteria 101) Geneva: WHO.

Iyengar, G.V., Kasperek, K., Feinendengen, L.E., Wang, Y.X. & Weese, H. 1982. Determination of Co, Cu, Fe, Hg, Mn, Sb, Se and Zn in milk samples. *Sci. Total Environ*., 24, 267-279.

Iyengar, V. & Nair, P. 2000. Global outlook on nutrition and the environment meeting the challenges of the next millennium. *Sci. Total Environ*., 249, 331-346.

Iyer, S.S., Babinski, M., Marinho, M.M., Barbosa, J.S.F., Sato, I.M. & Salvador V.L. 1999. Lead isotope evidence for recent uranium mobility in geological formations of Brazil: implications for radioactive waste disposal. *Applied Geochem*., 14(2), 197-221.

Jacobson, G. (1998, April). Arsenic poisoning from groundwater in Bengal. *Aus. Geo. News,* p. 45.

Jefferson, L., Campos, B.H., Andrian, K.C., Camila, M. 2006. Lactational exposure to inorganic mercury: evidence of neurotoxic effects. *Neurotoxicology and tetatology,* 28, 30.

Joint FAO/WHO Expert Committee on Food Additives (JECFA), 2003. Sixty first meeting of the joint FAO/WHO experts committee on food Additives (JECFA). Summary and conclusions. *Annex 4. Rome* 10-14th June 2003.

Jorhem, L., Slorrach, S., Sundstrom, B. & Ohlin, B. 1991. Lead, cadmium, arsenic and Mercury in meat, liver and kidney of Swedish pigs. *Food Audit. Contam*., 2, 201-12.

Kabata-Pendias, A. 2001. *Trace Elements in Soils and Plants* (3rd edn.). Boca Raton, FL: CRC Press.

Kioski, E., Venalaine, M. & Nuorteva, P. 1988. The influence of the forest type topographic location and seasons of levels of Al, Fe, Zn, Cd and Hg in some plants in Southern Finland. *Ann. Bot. Fenninci,* 25, 365-370.

Klassen, C.D., Watkind, J.B. & Casarette, D. 1999. *The basic science of poisons*. *Toxicology*. New York: Mc Graw Hill.

Kohferova, J. & Korenekova, B. 1995. The effect of emission of heavy metal concentration in cattle from the area of an industrial plant in Slovakia. *Arch. Environ. Contam. Toxicol*., 29, 400-405.

Komarnicki, G.J.K. 2000. Tissue, sex and age specific accumulation of heavy metals (Zn, Cu, Pb, Cd) by populations of the mole (*Talpa europaea* L.) in a central urban area. *Chemosphere*, 41(10), 1593-1602.

Korsch, M.J. 1999. Comparison of the rates of exchange of lead in the blood of newly born infants and their mothers with lead from their current environment, *J. Lab. Clin Med*., 133, 171-178.

Lacatus, R., Rauta, C., Carstea, S. & Ghelase, I. 1996. Soil – plant- man relationship in heavy metal polluted rear in Romania. *Appl. Geochem*., 11, 105-107.

LaKind, J.S., Wilkens, A.A. & Berlin, C.M. 2004. Environmental chemicals in human milk: A review of levels, infant exposure and health, and guidance for future research. *Toxicology and Applied Pharmacology,* 19, 184-208.

Larkin, M. 1997. Lead in mothers' milk could lead to dental caries in children. *Science and Medicine.* 35: 789.

Lasat, M.M. 2002. Phytoextraction of toxic metals: a review of biological mechanism. *J. Environ. Qual.* 31, 109-120.

Lehmann, C. & Rebele, F. 2004. Evaluation of heavy metals tolerance in *Calamagrostitis epigejos and Elymus repens* revealed copper tolerance in a copper smelter population of *C.epigejos. Environ. Exp. Bot.*, 51(3), 199-213.

Lemly, A.D. 1996. Evaluation of the Hazard Quotient method of risk assessment of selnium. *Ecotoxicology and Environmental Safety*, 35, 156-162.

Lepp, N.W. 1981. Effect of heavy metal pollution on plants. Englewood, N.J: Applied Science Publishers.

Leube, A. & Stumpfl, E.F. 1963. The Rooiberg and Leeupoort tin mines, Transvaal, South Africa. *Economic Geology*, 58(3), 391-418.

Licata, P., Trombeta, D., Critani, M., Giofre, F., Matino, D., Calo, M. & Naccari, F. 2003. Levels of toxic and essential metals in samples of bovine milk from various dairy farms in Calabria, Italy. *J. Environ. Int.*, 30, 1-6.

Limpopo will be top mining province. (2004, May 08). *Citizen*, p. 11.

Lindberg, S.E., Jackson, D.R., Huckabee, J.W., Jazen, S.A., Levin, M.J. & Lund, J.R. 1979. Atmospheric emission and plant uptake of mercury from the agricultural soil near the Almaden mercury mine. *J. Environ. Qual*., 8, 572-578.

Lindqvist, O. and Schroeder, W.H. 1989. Cycling of mercury in the environment with emphasis on the importance of the elements in acid rain studies. In J.M. Pacyna & B. Ottar (eds.). *Control and fate of atmospheric trace elements*. Norwell: Kluwer academics.

Ling, C., Ching-ying, Y., Hung-Chang, L., Hsing–Jasine, C., Ming-Jers & Bor- Cheng, H. 2006. Effect of mother's consumption of traditional Chinese herbs on estimated infant daily intake of lead from breast milk. *Sci. Total Environ*., 354, 120-126.

Liu, H., Probst, A. & Liao, B. 2005. Metal contamination of soils and crops affected by the Chenzouh lead/zinc mine spill (Hunan, China). *Sci. Total Environ*., 339, 153-166.

Lodenius, M. 1980. Aquatic plants and littoral sediments as indicators of mercury pollution in some areas of *Ann. Bot. Fenn.*, 17, 336-340.

Luh, M-D., Baker, R.A. & Henley, D.E. 1973. Arsenic analysis and toxicity- a Review, *Sci.Total Environ*, 2, 1-12.

Lurie, J. 1977. *South African Geology*. Johannesburg: McGraw-Hill.

Mafu, J.U. & Masika, P.J. 2002. Milk production from local goats, is it viable option. ARDRINEWS.

Magbagbeola, N.O, Oyeleke, O. 2003. Heavy metal poisoning and fish depopulation in Nigeria: a case study of the Lagos Lagoon. *J. African Fish and Fisheries,* 8: 192-193.

Maggi, E., Campanini, G., Bracchi, P.G. & Bagnoli, R. 1975. Determinazione dellarsenico in alcuni productti di origine animale. *Antti. Sis. Vet*., 34, 577-80.

Manfroi, C.B., Schwalm, F.D., Cereser, V., Abreu, F., Oliveira, A., Bizarro, L., Rocha, J.B.T., Frizzo, M.E.S., Souza, D.O. & Farina, M. 2004. Maternal milk as methyl mercury source for suckling mice: neurotoxic effects involved with the cerebella glutamatergic system. *Toxicol. Sci*., 81, 172-178.

Matsumoto, H., Koya, G. & Takeuchi, T. 1965. Fetal Minamata disease, a neuropathological study of two cases of intra uterine intoxication by methylmercury compound. *J. Neuropathol. Exp. Neurol.*, 24, 563-574.

Matt, K.J. 1972. Mercury uptake from soil by various plant species. *Earth and Environmental Science.* 8(2): 77-80.

Mazunder, D.N., Haque, R., Ghosh, N., De, B.K., Santra, A., Charkraborty, D. & Smith, A. 1998. Arsenic levels in drinking water and the prevalent lesions in West Bengal., India. *Int. J. Epidemiol*., 27, 871-887.

Mc Grath, S.P., Zhao, F.J. & Lombi, E.L. 2001. Plant and rhizosphere process involved in phytoremediation of metal contaminated soils. *Plant Soil*, 232, 207-214.

McKee, J.E. & Wolf, H.W. 1963. Water quality criteria 2nd edition (Pub. No. 3-A). California: California State Water Resource Control Board.

Meharg, A.A. & Hartley-Whitaker , J. 2002. Arsenic uptake and metabolism in arsenic resistant and non-resistant plant species: Tansley Review. *New Phytologist***,** 154, 29-43.

Mehennaoui, S., Delacroix-buchet, A., Duche, A., Enriquez, B., Kolf, C. M & Milhaud, G. 1999. Comparative study of cadmium transfer in Ewe and Cow milks during rennet and lactic curds preparation. *Archives of environmental contamination and toxicology* 37, 389-395.

Metcheva, R., Atanasova, N. & Topashka-Ancheva, M. 1994. Bioaccumulation de mataux toxiques de l'usine de metallurgie non ferreuse pres de plovdiv dan l'organisme du campagnol Mediterranean (microtus) Guentheri dan et als. 1880 (Microtinae-rodentia). *Acta. Zool. Bulg*., 47, 54-65.

Milhaud, G. 1997. Transfer of cadmium from feed to ewe food products: variations in transfer induced by lead and zinc. *Environ. Sci*., 5, 127–138.

Miller, W.J. 1971. Cadmium nutrition and metabolism in ruminants: relationship to concentrations in tissues and products. *Feedstuffs*, 43, 24–26.

Mining and minerals in South Africa. Retrieved Sat. April 5, 2008 from www.southafrica.info.

Mishra, K., Upadhyay, A.R., Pathak, V. & Tripathi, B.D. 2008. Phytoremediation of Mercury and Arsenic from Tropical Opencast Coalmine Effluent through Naturally Occurring Aquatic Macrophytes virendra. *J. Water, Air, & Soil Pollution*, 192(1-4): 154-156.

Moline, J., Carrillo, L., Sanchez, L.T., Goldbold, J. & Todd, A. 2000. Lactation and lead body burden turnover: A plot study in Mexico. *J. Occup. Environ. Med.*, 42, 1070 - 1075.

Monteiro, L.R., Costa, V., Furness, R.W. & Santos, R.S. 1996. Mercury concentrations in prey fish indicate enhanced bioaccumulation in mesopelagic environments. *Marine Ecol. Prog Ser.*, 141, 21-25.

Mushak, P. 1991. Gastro intestinal absorption of lead in children and adults. Overview of biological and biophysical aspect. *Chem. Spec. Bioavail*., 3, 87-104.

National Research Council, 2001. *Arsenic in drinking water: update*. Washington DC: National Academy Press.

Neathery, M.W., & Miller, W.J. 1975. Metabolism and Toxicity of Cadmium, Mercury, and Lead in Animals. *Journal of Dairy Science.* 58 (12), 1767-1781.

Nordstrom, S., Beckman, L. & Nordenson, I. 1979. Occupational and environmental risks in and around a smelter in Northern Sweden V: Spontaneous abortion in female employees. *Hereditas*, 90(2), 291-6.

Nriagu JO 1978. *The biogeochemistry of lead* J.O. Nriagu (ed.). Amsterdam: Elsevier Biomedical Press.

Okonkwo, J.O. & Maribe, F. 2005. Assessment of Lead Exposure in Thohoyandou, South Africa. *The Environmentalist*, 24, 171–178

Osher, L.J, Leclerc, L., Wiersma, G.B., Hess, C.T. & Guiseppe, V.E. 2006. Heavy metal contamination from historic mining in upland soil and estuarine sediments of Egypt Bay, Maine, USA. *Estuarine, Coastal and Shelf Science,* 70(1-2), 169-179.

Oskarson, A.L., Jorham, L, Sindberg, J., Nilson, N. & Abanus, L. 1992. Lead poisoning in cattle- Transfer of Lead to milk. *Sci. Total Environ.*, 111, 83-94.

Oskarsson, A., Hallén, I.P. & Sundberg, J. 1995. Exposure to toxic elements *via* breast milk. *Analyst***,** 120, 765 – 770.

Oskarsson, A.L., Schutz, A., Skerfving, S., Palming, H.I., Ohlin, B.J. & Son Langerkvist B. 1996. Total and inorganic mercury in breast milk and blood in relation to fish consumption and amalgam fillings in lactating women. *Arch. Environ. Health*, 51, 234-241.

Ott, W.R., 2007. Exposure analysis: a receptor-oriented science. In J.W. Roberts and W.R. Ott *Exposure analysis.* W.R. Ott, , A.C. Steinemann & L.A. Wallace (eds). London: Taylor & Francis Group.

Ou, Y.C., White, C.C., Krejsa, C.M., Ponce, R.A., Kavanagh, T.J. & Faustman, E.M. 1999. The role of intracellular glutathione in methyl mercury-induced toxicity in embryonic neuronal cells. *Neurotoxicology*, 20, 793-804.

Palache, C., Berman, H., & Frondel, C. 1951. *The System of Mineralogy of James Dwight Dana and Edward Salisbury Dana, Yale University 1837-1892*. New York: John Wiley and Sons, Inc.

Palusova, O., Ursinyova, M., & Uhnak, J. 1999. Mercury levels in the components of the environment and diets. *Sci. Total Environ.*, 101, 79-80.

Parga, J.R., Cocke, D.L., Jesus, L.V., Gomes, J.A., Kesmez, M., George, I., Moreno, H. & Weir, M. 2006. Arsenic removal via lector coagulation of heavy metal contaminated ground water in La Comarca Lagurera, Mexico. *J. hazardous Materials*, 124, 247-254.

Park, Y.W., Juarez, M., Ramos, M. & Heanlein, G.F.W. 2007. Physico-chemical characteristics of goat milk and sheep milk. *Small Ruminant Research,* 68, 88-113.

Patriacia, M., Menditto, A., Rossi, B., Lyon, T.D.B. & Fell, G.S. 2000. Environmental exposure to metals of newborns, infants and young children. *Microchemical*, 67, 351-361.

Pitkin, R.M., Bahns, J.A., Filer, L.J. & Reynolds, W.A. 1976. Mercury in human

Platteland towns; after the coal rush (2006, May 08). *Financial Mail* p. 76.

Poor will gain when gold mine re-opens. (2006, May 08). *City press,* p. 10.

Pueschel, S.M., Linakis, J.G. & Anderson, A.C. 1996. *Lead poisoning in childhood.* Baltimore: Paul brooks.

Punt, C., Pauw, K., Schoor, M., Nyhodo, B., McDonald, S., Chant, L. & Valente, C. 2005. A profile of the Limpopo province: Demographics, poverty, inequality and unemployment. Background Paper: 1(9). Elsenburg: Government press.

Queirolo, F., Stegen, S., Restovic, M., Paz, M., Ostapczuk, P., Schwuger, M. J. & Muñoz, L. 2000. Total arsenic, lead, and cadmium levels in vegetables cultivated at the Andean villages of northern Chile. *J. Total Environ.*, 255(1-3), 75-84.

Radostits, O.M., Blood, D.C., Gay, C.C. & Hinchcliff, H.E. 2000. *Veterinary medicine. A text book of diseases of cattle, sheep, pigs' goats and horses*. London: Saunders.

Raghunath, R., Tripathi, R.M., Sastry, V.N. & Krishnamoorthy, T.M. 2000. Heavy Metals in maternal and cord blood. *Sci. Total Environ.* 250, 135-141.

Rahman, M.M., Chowdhury, U.K., Mukherjee, S.C., Mondal, B.K., Paul, K., Lodh, D., Chanda, C.R., Basu, G.K., Biswas, B.K., Saha, K.C., Roy, S., Das, R., Palit, S.K., Quamruzzamman, Q. & Chakraborti, D. 2001. Chronic arsenic toxicity in Bangladesh and West Bengal, India: a review and comentry. *J. toxicol. Clin. Toxicol*., 39(7), 683-700.

Rana, T., Sarkar, S., Mandal, T.K., Bhattacharyya, K., Roy, A., Kol, L. 2008. Contribution of arsenic from agricultural food chain to cow milk in highly arsenic prone zone in Nadia District of West Bengal in India. *The Internet Journal of Thoracic and Cardiovascular Surgery*. 4(2). Retrieved January 8, 2008 from http://www.ispub.com/journal/the_internet_journal_of_veterinary_medicine/volume_4_number_2_2/article/contribution_of_arsenic_from_agricultural_food_chain_to_cow_milk_in_highly_arsenic_prone_zone_in_nadia_district_of_west_bengal_in_india.html.

Raszyk, J., Gajduskova, V. & Ulrich, R. 1996. Occurrence of harmful pollutants in fattened pigs. *Vet Med.*, 41, 261-266.

Reilly, C. 1999. Heavy metals Toxicology. In M.J. Sandler, J.J. Strain, B. Caballero (eds). *Encyclopedia of Human Nutrition.* San Diego: Academic Press.

Rosewarne, P. (2005, March/April). Managing ground water for tomorrow. *The Water Wheel.* Retrieved January, 6th 2007 from www.wrc.org.za .

Rossipal, E. & Krachler, M. 1998. Patterns of trace elements in human milk during the course of lactation. *Nutrition Research,* 18(1), 11-24.

Roychhowdhury, T., Tokhunga, H. & Ando, M., 2002. Survey of arsenic and other heavy metals in food composites and drinking water and estimation of dietary intake by the villagers from an arsenic-affected area of West Bengal, India. *Sci. Total Environ.,* 308, 15-35.

Ryu, J.E., Zieglar, E.E., Nelson, S.E. & Fomon, S.J. 1985. Dietary and environmental exposure to lead and blood during early infancy. In K.R. Mahaffey (ed.). *Dietary and environmental lead: human health effects*. Amsterdam: Elsevier.

Safonov and Prokof'ev, 2006) Safonov, Y. & Prokof'ev, V. 2006. Gold-bearing reefs of the Witwatersrand Basin: A model of synsedimentation hydrothermal formation. *Geology of Ore Deposits,* 48(6), 415-447

Sami, K. & Druzykli, A.L. 2003. *Predicated spatial distribution of naturally occurring arsenic, selenium and uranium in groundwater in groundwater in South Africa Reconnaisance survey* (Report No 1236/1/03). Pretoria: Water Research Commission.

Samuillah, Y. 1990. *Biological monitoring: Animal techchnical report* (Report No. 37), London: MARC.

Schuurmann, G. & Markert, B. 1997. *Ecotoxicology: Ecological Fundamentals, Chemical Exposure, and Biological Effects*. New York: John Wiley & Sons.

Shi, W., Bischoff, M., Turco, R. & Konopka, A. 2002. Long term effect of Chromium and lead upon the activities of the soil microbial communities. *Appl. Soil Eco.*, l21, 169-177.

Shore, R.F. & Douben, P.E.T. 1994. Predicting ecotoxicological impacts of environmental contamination on terrestrial small mammals. *Environ. Contam. Toxicol.*, 134, 49-89.

Shore, R.F. 1995. Predicting cadmium, lead and fluoride levels in small mammals from soil residues and by species-species extrapolation. *J. Environ. Pollut*., 88(3), 333-340.

Shresther, R.R., Upadhyay, N.P., Pradhan, R., Khadka, R., Maskey, A., Maharjan, M., Tuladhar, S., Dahal, B.M. & Shresther, K. 2003. Groundwater arsenic contamination, its health impact 7 mitigation program in Nepal. *J. Environ. Sci. health*, A38(10), 185-200.

Skerfving, S. 1988. Mercury in women exposed to methylmercury through fish consumption and in their new born babies and breast milk. *Bull. Environ. Contam. Toxicol*., 41, 475-482.

Smith, R. & Vasiloudis, H. 1991. Importance determination and occurrence of inorganic hemical contaminants and nutrients in South African municipal sewage sludges. *Water SA*, 9(1), 19-30.

South Africa Yearbook 2003/04. Pretoria: *Government Communication and Information System (GCIS) and STE Publishers.* Retrieved January 10, 2009 from http://www.gcis.gov.za/docs/publications/yearbook.htm.

South African Statistics 2003a. *Census 2001,* Pretoria: Government Printer.

South African Statistics 2003b. *Gross Domestic Product, Statistical Release P0441, 25 November 2003.* Pretoria: Government printer.

South African Statistics 2003c. *Census 2001: Census in Brief.* Pretoria: Government Printer.

South African Statistics 2006. *Labour Force Survey*. Pretoria: Government Printer.

Steenhout, A. 1982. Kinetics of lead storage in teeth and bones: an epidemiologic approach. *Arch. Environ. Health*, 37, 224-31.

Stephens, R & Waldron, H.A. 1975. The influence of milk and related dietary constituents on lead metabolism. *Food Cosmet. Toxicol.* 13, 555–63.

Sternowsky, H.J., Moser, B. & Szadkowsky, D. 2002. Arsenic in breast milk during the first 3 months of lactation. *International Journal of Hygiene and Environmental Health*, 205(5), 405-409.

Steven, J.B. 1991. Disposition of toxic metals in agricultural food chain. 1. Steady- state Bovine milk bio transfer factors. *Environ. Sci. Technol.*, 25: 1289-1294.

Swarup, D., Patra, R.C., Naresh, R., Kumar, P. & Shekhar, P. 2005. Blood lead levels in lactating cows reared around polluted localities; transfer of lead into milk. *Sci. Total Environ*., 349, 67-71.

Talmage, S.S. & Walton, B.T. 1991. Small mammals monitors of environmental contaminants. *Environ. Contam. Toxicol.*, 119, 47-145.

Tandukar, N., Bharttacharya, P. & Mukherjee, A.B. 2001. Managing arsenic for our future. In proceedings of the international conferences on arsenic in the Asia pacific region. Adelaide: Aris .

Mao, J., Rao, B.L., McAdams, J.Y., Xuan, M., & Li, X Z. 1989. Relation of arsenic exposure to lung cancer among tin miners in Yunnan Province, China. *Br. J Ind Med.*, 46(12), 881–886.

Terephthalate, M.P. 2000. Arsenic in Asia. Water at its worst. *Environ. Health Perspect.*, 5(8), 224.

Thutmoose, C. 2002. *Lead poisoning and the fall of The Roman Empire*. Retrieved June 2, 2009 from www.everything.com.

Tiller, K.G. 1986. Essential and toxic heavy metals in soils and their ecological relevances. *Soil Sci*., 1, 29-44.

Tillett, T. 2008. Arsenic “Lactation Intolerant”? Study Indicates Low Excretion in maternal and cord blood, placenta and milk. *Proc. Soc. Exp. Biol Med.*, 151, 565-567.

Toens, P.D., Stadler,W. & Wullschleger, N.J. 1998. The association of groundwater chemistry and geology with a typical lymphocytes (as a biological indicator) in the Pofadder area, (Report No. K5/839) .North Western Cape: WRC.

Topashka, A.M. 1995. The influence of polymetal industrial dust on chromosomes and immune reactivity of laboratory mice. *International Symposium Ecologia*, 95, 34-37.

Topashka, A.M., Atanasova, N., Metcheva, R. & Monkova, V. 1993. Effect of heavy metals on guenthers vole, Micro guentheri. *International Symposium Ecology*, 93(2), 345-351.

Topolska, K., Sawicka, K. & Cieslik, E. 2004. The effect of the Krakow region on heavy metal contamination in the organs of bank voles. *J. Environ. Studies*, 13, 103-109.

Tripathi, R.M., Raghunath, R., Sastry, V.N. & Krishnamoorthy, T.M. 1999. Daily intake of heavy metals by infants through milk and milk products. *Sci. Total Environ.*, 227: 229-235.

Turconi, A., Guarcello, M., Livieri, C., Commizzoli, S., Pietri, A., Piva, G. & Roggi C., 2004. Evaluation of xenobiotics in human milk and ingestion by the newborn. *Eur. J Nutr.*, 43, 191-197.

Turkdogan, M.K., Kulicel, F., Kara, K., Tuncer, I. & Uyang, I. 2003. Heavy metals in soils, vegetable and fruit in the endermic upper gastro intestinal cancer region of Turkey. *Environ. Toxicol. Pharm.*, 13(3), 175-179.

Turker, S.B., Lamm, S.H., Li, F.X., Wilson, R., Byrd, D.M., Lai, S.T. & Lo, L. 2001. *Relationship between consumption of arsenic contaminated well water and skin disorders in hushot*. Inner Mongolia: Cambridge.

U.S. Environmental protection Agency (n.d). *Lead in drinking water*. Retrieved June 24, 2009 from www.epa.gov/opptintr/lead.

U.S. Environmental Protection Agency, 1997. Exposure Factors Handbook (Final Report). Washington: U.S. EPA.

US EPA 1995. *Mercuric Chloride. Oral reference dose and carcinogenicity assessment.* Retrieved January 29, 2009 from http://www.epa.gov/iris/index.html.

USEPA, 1986 "Air quality for lead" U. S. Environmental Protection Agency, Office of Research And Development Office of Health And Environmental Assessment Research Triangle Park. NC EPA 600/8-83-028F

Vivier, F.S., Pieterse, S.A. & Aucamp, P.J. 1988. Guidelines for the use of sewage sludge. Presented at a symposium of sludge handling. Pretoria. DWAF.

Wang, W. & Fisher, N.S. 1999. Delineating metal accumulation pathways for marine invertebrates. *Sci. Total Environ.*., 238, 459-472.

Weaver, R.W., Melton, J.R., Dshin, W. & Dubble, R.L. 1984. Uptake of Arsenic and mercury from soil by Bermuda grass (*cynodon dactylon*). *Environ. Pollut.*, 33, 133-142.

Westoo, G. 1973. Mercury and Methylmercury levels in Swedish cows' milk and human milk. *Var Foda.*, 25, 122-123.

Whanger, P.D. 1982. Factors affecting the metabolism of nonessential metals in foods. *Nutritional toxicology.* 1, 164-208.

Willet, L.B., Becker, J.J. & Bromund, C.J. 1994. Distribution of lead in lactating cows. *Ohio Agric Res Dev Cent*., 145, 9-11.

Williams, R.D. 1995. Breast-feeding best bet for babies. *FDA Consumer*, 29. Retrieved on August 25, 2009 from http://www.questia.com/googleScholar.qst;jsessionid=L96TXb9GtSRhb0l1TJLJM2yx55z5DCJ12Q07MCXjGvTFks3NKk4V!1398020991!1823011750?docId=5002239075

Woolson, E.A., Axley, J.H. & Kearney, P.C. 1973. The Chemistry and Phytotoxicity of Arsenic in Soils: II. Effects of Time and Phosphorus. *J. Soil Science Society of America*, 37, 254-259.

World Bank Group, 1998. *Pollution Prevention and Abatement Handbook.* 219-222 Effective.

World Health Organisation (WHO) 1986. Principles for evaluating health risks from chemicals during infancy and early childhood: the need for a special approach. *Environmental Health Criteria*, 59, Geneva: WHO.

World Health Organization (WHO), 1991. Inorganic mercury. *Environmental health criteria*, 118, Geneva WHO.

World Health Organization *(WHO), 1993. The WHO recommended classification of pesticides by hazard and guidelines to classification, 1992-93. Geneva*, WHO

World Health Organization (WHO), 1996. Lead, cadmium and mercury. In Trace elements in human nutrition and health. Geneva: WHO

World Health Organization, 1990. International Program on chemical safety, Environmental health criteria 101 methylmercury, *HIPCS*, Geneva: WHO.

Wyllie, J. 1937. An investigation of the source of arsenic in well water. *Can. Public Health J*., 28, 128-138.

Xiau, J.G. 1997. 96% of well water undrinkable. *Asia Arsenic Network. Japan Newsletter N*o 2, 7-9.

Your dictionary. Retrieved on August 6, 2008 from www.your dictionary.com.

Zhao, J., Ratcliff, W., Lynn, J. W., Chen, G. F, Luo, J. L., Wang, N. L., Hu, J., & Dai, P. 2008. Spin and lattice structures of single-crystalline $SrFe_2As_2$. *American Physical Society*, 78(14), 205-213.

Zietz, B., De Vergara, J.D., Kevekordes, S. & Dunkelberg, H. 2000. Lead contamination in tap water of households with children in lower Saxony, Germany. *J. Sci. Total Environ*., 125 (1-3), 19-26.

Zueng-sang C. 2004. Cadmium and lead contamination of soils near plastic stabilizing materials producing plants in Northern Taiwan. *J. Water, Air, & Soil Pollution*, 57-58(1), 203-205.

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