An Evaluation of Clam *Amiantis umbonella* (Bivalve) as a Bio-indicator of Heavy Metal Pollution in Kuwait Marine Coastal Waters.

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ABSTRACT OF THE DISSERTATION

An Evaluation of Clam *Amiantis umbonella* (Bivalve) as a Bio-indicator of Heavy Metal Pollution in Kuwait Marine Coastal Waters.

By QAISER TARIQUE

Dissertation Director:
Dr. Joanna Burger

To evaluate the clam *Amiantis umbonella* as a bio-indicator of metal contamination, the concentrations of a number of metals (Hg, Cd, Pb, Cu, Ni, Mn, Cr, V, Zn, and Fe) were determined in clam's soft tissues, in different organs, water, and sediment in Kuwait Bay of the Arabian Gulf. The primary objective of this study was to evaluate the relationship of all metals concentrations in soft tissue and various organs of clam, water and sediment and to compare them with a reference site, and with results obtained from other geographical areas of the world. Metal concentrations and metal burdens of Pb, Hg, Cu, and Cd in the soft tissues of clam were also compared as a function of clam size.

Hg, Cd, Cu, Ni, Cr, V and Pb were significantly higher in water and sediment near point sources compared to reference sites. The concentration of all metals, except Zn were significantly higher in kidneys, gonads, mantles, gills, livers and hearts of contaminated site clams, compared to the reference site clams. The highest inter-location variability was in kidneys, compared to all other organs. The concentration of Cu, Hg, Ni and Fe in contaminated site clams was significantly higher in the gonads (15.0 ppm, 2.32 ppm, 4.3 ppm, 4.6 ppm wet weight respectively) than in kidneys, mantles and livers. The

highest Cr concentration (12.5 ppm) was in the mantles. The liver was found to be the main depository of Cu (16.7 ppm) in contaminated site clams. Among the metals, Cd body burden and concentration was positively correlated to clam's wet weight/length. However, the concentrations of Pb, Hg and Cu were not correlated with clam length or weight, indicating it may be biologically regulated.

The concentrations of Ni, Cu, Cd, Pb, Mn, V and Cr in sediment were highly correlated with those in water. The levels of metals in clams from this study are generally within the range of mean values reported in the literature for other areas of the world. Only elevated levels of Hg (6.2 ppm), Pb (2.0 ppm) and Cd (4.8 ppm) from Kuwait exceeded the levels set by FAO, WHO and FDA for human consumption.

DEDICATION

For my husband, Tarique, with much love.

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I could not have completed this project without the help of several respected people. My graduate committee members, Dr. Joanna Burger, Dr. John Reinfelder, Dr. Michael Gochfeld and Dr. Keith Cooper provided valuable direction during all stages of my graduate program.

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Research), Dr. Manaf Behbehani (Kuwait University) and Dr. Salim Al-Mohanna (Kuwait University)

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I am lucky to have friends who also assisted me through out this process. Dr. Mobeen Faqih, Dr. S.M. Ghazanfar and Dr. Deepak Malhotra were always willing to help with valuable advice.

Finally, I want to thank my entire family. My husband, Dr. Asif Tarique yielded his love and unwavering faith in me which gave me the confidence to pursue my ambition. I will forever be thankful to my daughters, Ryeha and Kahkashan for supporting me and being pillars of strength through this challenging time. They believed in me and always encouraged me when times were getting tough. And my three sons: I wish to extend my warmest thanks to my son Akif for his continued unwavering support and valuable contribution towards my Ph.D. He was always there when I needed him. I am certain that without his encouragement and constant guidance I could not have finished this dissertation. I owe gratitude to my oldest son Ibraiz for his understanding and expertise. He not only helped with my admission to the program but also was my official advisor. My youngest son Suhaib deserves unmatched praise. His computer expertise proved invaluable throughout my education. He laid the foundation for my documents and pushed me to work harder. He instilled motivation in me to achieve my dreams.

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1. General Introduction

1.1. Introduction of this Dissertation

Metal pollution of the coastal environment continues to attract the attention of environmental researchers (Amiard et al., 1986, 1987; Shulkin e al., 2003). Coastal marine ecosystems have become more extensively degraded in recent years (Goldberg et al.,1978). In spite of efforts to improve resource management, industrialized society continues to release chemicals to the environment via aquatic effluents, spillages (accidents) and atmospheric emissions. However the increase of anthropogenic inputs of metals to aquatic ecosystems has caused growing concern among the general public and resource managers, and has given rise to strategies to monitor the exposure of organisms to these metals. It is not surprising that coastal waters are under enormous environmental stress. It is also believed that ocean disposal of selected wastes can safely take place in certain areas. However, if assimilative capacities are exceeded, receiving waters soon begin to exhibit evidence of ecological stress. This continuing release of contaminants requires an ongoing assessment of chemical concentrations and their ecological effects in coastal regions (Tripp and Farrington, 1985; Widdows *et al.,* 1997).

Pollution by heavy metals is a serious problem due to their toxicity and their ability to accumulate into biota (Islam and Tanaka, 2004). Therefore, determination of metal concentration in organisms is an essential part of any assessment and monitoring program in the coastal zone. Marine pollution is generally referred to as the presence in the marine environment (water, sediment or organism) of both inorganic and organic compounds often of anthropogenic origin, which occurs in excess thus leading to alterations in the characteristics of the ecosystem (Diaz *et al.,* 2001). Metals may enter aquatic ecosystem at points near their source, or more diffusely through atmospheric deposition of industrial or automobile emissions directly onto waters or indirectly through runoff from surface waters (Bellos et al., 2005). Once in waters, metals undergo a range of physical and chemical reactions that influence their fate, including partitioning between aqueous and solid phases in the water column, and in turn, burial in sediments and accumulation and transfer in food chains (Paulson et al., 2003). Various studies have shown the impact of point sources on inter-tidal benthic communities adjacent to discharge points, resulting in mainly sub-lethal effects including their accumulation and subsequent biomagnifications through the food chain becoming detrimental to human beings (Szefer et al., 2002).

The potential threat of metals to aquatic organisms has been directly and indirectly assessed during recent decades, using several methods. As an indirect measure of the abundance and availability of metals in the marine environment the bioaccumulation of metals by the tissues of the marine organisms led to the adoption of the bio-indicator concept (Langston et al., 1998).

The use of biological indicator organisms in the study of trace metal pollution in coastal marine systems has become attractive and well accepted in the past three decades (Cosson et al., 2000). Bio-monitoring and surveillance are key methods for assessing the status and wellbeing of ecological receptors within functioning ecosystems (Burger and Gochfeld, 2001).   According to the National Academy of Science (NAS, 1993), the use of bio-indicators has proven to be effective in advancing the state of knowledge and understanding of environmental processes, supporting environmental regulation, standard setting and enforcement, and improving on existing methods of instrumentation.

The ability of tolerant species (bivalve) to concentrate pollutants hundreds or thousands of times over levels present in the ambient environment provides a reliable method of detecting a contaminant's presence. The use of bivalve (mollusks) as indicators of trace metal enrichment or biological stress in aquatic environments has gained widespread acceptance (Goldberg et al., 1978; Bryan et al., 1980; Phillips, 1980; Popham et al., 1980; Jensen et al., 1981; Thomson et al., 1984). Bivalves such as *Mytilus* have been successfully used as indicator organisms in environmental monitoring programs throughout the world to identify variation in chemical contaminants among sites and contribute to the understanding of trends in coastal contamination (O' Connor et al., 1995, Beliaeff et al., 1998, North America; Picer and Picer, 1991; Thailand; Sericano et al.,1995; Central and South America; Widdow et al ., 1995; Britain. The National Status and Trends program in the United State runs by the National Oceanic and Atmospheric Administration is an extensive bio-monitoring program (O'Connor, 1991, 2002). Similar programs have been established independently in some states, California, (Gunther et al., 1999); Maine, Gulf Watch Program, (Townsend, 1992), as well as other countries, such as France (Cossa et al., 2002), Russia (Tkalin et al., 1998), and Taiwan (Hung et al ., 2001).

1.2. Rationale of using clam as a bio-indicator of metal pollution in Kuwait Bay

Bivalves are an important component of the benthos in coastal ecosystem and are commonly used bio-sentinel organisms to assess exposures of the benthos to metal contamination (Goldberg et al., 1983; O'Connor 1992; Brown and Luoma, 1995). The criteria by which organisms are accepted as bioindicator for the assessment of contamination were proposed more than twenty five years ago and remain unchanged, such as the 'Mussels Watch Program'. The importance of bivalves in pollution impact studies is shown by the magnitude and longevity of the international Mussel Watch (Goldberg, 1978). This program originated in the late 1960s and continues to maintain momentum today (Cantillo 1992; O'Connor 1998; Jeng et al., 2000). Large-scale monitoring programs such as this provide information on both current status and long term trends which allows evaluation of species, population, and ecosystem effects (Burger, 2006).

Bivalves concentrate many metals in their soft tissues and the tissue metal levels represent a time integrated response to bio-available metal in food and water (bioaccumulation). They are ideal bio-monitors of aquatic systems, because they are tolerant of ecological variations in parameters such as salinity, and have the ability to accumulate organic and metallic pollutants at concentrations several orders of magnitude above those in the ambient environment (Bryan et al., 1977, 1983).

The factors which influence metal concentration and accumulation are bioavailability of metals, season, size, sex, hydrodynamics of the environment, changes in tissues composition and reproductive cycle (Boyden and Phillips, 1981). The following attributes have led to the use of bivalves, particularly clams and mussels as "sentinel" or "indicator" organisms in environmental monitoring programs throughout the world:

1. Bivalves, such as mussels and clams, are dominant members of coastal and estuarine communities and have a wide geographical distribution. This minimizes the problems inherent in comparing data from markedly different species (Philips, 1980; NRC, 1986).

2. Clams and mussels are sedentary and therefore better than mobile species as integrators of chemical contamination in a given area (Roesijadi, 1982).
3. Bivalves are relatively tolerant of a wide range of environmental conditions, including moderately high levels of many types of contaminants (Widdow, 1985, 1997).
4. Most bivalves are suspension feeders that pump large volumes of water (several liters per hour) and concentrate many chemicals in their tissues relative to the concentration in seawater. This often makes measurements of trace contaminants easier to accomplish in their tissues than in seawater (Farrington, 1987).
5. The measurement of chemicals in bivalve tissues provides an assessment of biological availability that is not apparent from measurement of contaminants in environmental compartments (water, suspended particulates and sediments) (NRC, 1983)
6. In comparison to fish and crustaceans, bivalves have very low levels of activity of those enzyme systems capable of metabolizing organic contaminants, such as aromatic carbons. Therefore contaminant concentrations in the tissues of bivalves more accurately reflect the magnitude of environmental contaminants (NRC, 1986; Phillips, 1980).
7. Mussels and clam populations are relatively stable and can be sufficiently large for repeated sampling, thus providing data on short and long term temporal changes in contaminant levels (O'Connor, 1992).
8. Mussels and clams can be and maintained in cages and readily transplanted to sites of interest, either in the inertial zone or sub tidally on moorings.

Mussels and clams are a commercially important seafood species on a worldwide basis and measurement of chemical contamination is of interest for public health consideration (Philips, 1980). In recent years, researchers have focused their attention on the identification of other possible bio-monitors or bio-indicators for trace metal pollution to expand current understanding of different bioaccumulation strategies for trace metals. Monitoring schemes will be most useful if they include multiple species representing different tropic levels (Burger, 2006)

Individual bio-indicators respond differently to different sources of bio-available metal (e.g., solution, sediment or in food). Element concentrations in bivalves at the same location differ between different species and individuals due to species-specific abilities and capacities to regulate or accumulate trace metals (Reinfelder et al., 1997; Otchere et al., 2003). Aquatic and related fauna such as mussels (O'Connor, 1992), clams (Burdon-Jones and Denton, 1984), barnacles (Fialkowski and Newman, 1998), colonial sea birds (Burger, 1997, Burger and Gochfeld, 2004) and fish (Denton and Burdon-Jones, 1984) are reliable bio-indicators of heavy metal-monitoring programs in marine ecosystems.

Ecological risks from metal contaminants are difficult to document because responses differ among species, threats differ among metals, and environmental influences are complex. Unifying concepts are needed to better tie together such complexities (Luoma et al., 2005). To gain a complete picture of total heavy metal bio-availability in a marine habitat it is necessary, therefore, to use a suite of bio-indicators that reflect metal bio-availabilities in all available sources (Phillip, 1990; Rainbow, 1990, 1993). Such comparative use of different bio-indicators should allow identification of the particular source of the contaminant metal (Phillip & Rainbow, 1993). As early as 1976, claims were made that mussels (*Mytilus edulis*) should not be used as indicator of copper pollution since there was a variability of copper uptake by the organism (Phillip, 1976) which could have been due to environmental factors such as temperature and salinity. A more recent report supports the claim that *M. edulis* are efficient accumulators of many metals, but do not show great changes in tissues concentrations of zinc and copper, in response to increases in metal bio-availability (Rainbow, 1995).

In recent years, researchers have focused their attention on the identification of other possible bio-monitors or bio-indicators for trace metal pollution to expand current understanding of different bioaccumulation strategies for trace metals. Monitoring schemes will be most useful if they include multiple species representing different tropic levels (Burger, 2006).

The overall goal of the study was to explore the potential use of the clam *Amiantis umbonella* (bivalve) as an effective bio-indicator species for metal contamination in the Kuwait marine environment. In addition to its use in environmental evaluations, the amount of pollutants bio-accumulated is a concern as these edible bivalves are consumed by humans and wildlife.

Clams *A. umbonella* were used as bio-indicator of metal contamination. This species is widely distributed in Asia and especially in India, Pakistan, Mauritius, Arabian Gulf, Qatar, Kuwait. This clam typically lives and feeds at a depth of 7-8 cm in fine sediments and quiet observable and easily accessible during low tide (Chapter 2, Figure 3). Clam *(A. umbonella)* as a facultative deposit feeder has the potential to accumulate bio-available contaminants from both water and sediment. In addition to using clams as bio-indicators of marine coastal contamination, it is also recognized that they are an important link for the transfer of contaminants between sediments, water and higher organisms, including man. Hence, information on the contaminant concentrations in their tissues is potentially useful in considering toxicological and public health implications of marine coastal contamination (Phillip & Rainbow, 1993). Contaminants, such as metals, can have adverse effects on the health and wellbeing of organisms, including humans. Thus there is a need of information on contaminant trends, especially for coastal regions, which are increasingly industrialized and developed (Burger and Gochfeld, 2002). Understanding the fate and effect of chemicals can be used to assess the health of ecosystems and to provide early warning of changes in the environment that might indicate adverse effects (Burger, 2002).

The primary objective of this study was to study the concentrations of ten metals (Hg, Cd, Pb, Cu, Mn, Zn, Cr, Ni, V, and Fe) in a clam population (*A. umbonella)* living adjacent to water desalination and electric power plant in Kuwait Bay (Chapter 2, Figure 3) and compared with reference sites clams at Umm-Kishish study area, 5 km away from the point source (Chapter 2, Figure 3). In addition, metal concentrations were also compared with those in marine clams from around the world. Metal accumulation in biota and organs were also compared with those in the sediment and water from their habitats. Furthermore, metal accumulation was studied as a function of age and size of clam.

This dissertation is divided into six chapters. Chapter 1 gives a general introduction (literature review) and rationale for studying the use of clam *Amiantis umbonella* as a bio-indicator of metal pollution in Kuwait marine environment. A conceptual model of metal bioaccumulation showing the potential routes of contaminant mobility and fate was developed. Chapter 2 focuses on the accumulation of ten trace metals (Hg, Cd, Pb, Cu, Mn, Zn, Cr, Ni, V, and Fe) in clams near a desalination and power plant compared to a reference sites. Chapter 3 investigates the distribution of heavy metals in the organs of the marine clam's population living along point source contaminated and reference sites in the Kuwait Bay. Chapter 4 focuses on the concentrations and body burden variations of four metals (Hg, Cd, Pb and Cu) in relation to clam shell length and total wet weight of the contaminated site clams. Chapter 6 presents the overall conclusion while Chapter 6 states the future direction. Chapter 7 contains the References while the Curriculum Vita is found in Chapter 8.

1.3. Metal Contamination and Related Questions

Metal Contamination and related questions.

1. By which pathway, water or sediment, do heavy metals enter these clams?
2. In which organs are metals accumulated and to what extent?
3. Can clam whole tissues and / or organs be used as bio-indicator of metal contamination?
4. Is metal bio-accumulation in clam tissues related to metal exposures that can either adversely affect the health of the organism, or be transferred up the food web?
5. How do metal concentrations and body burdens vary with clam length and weight?

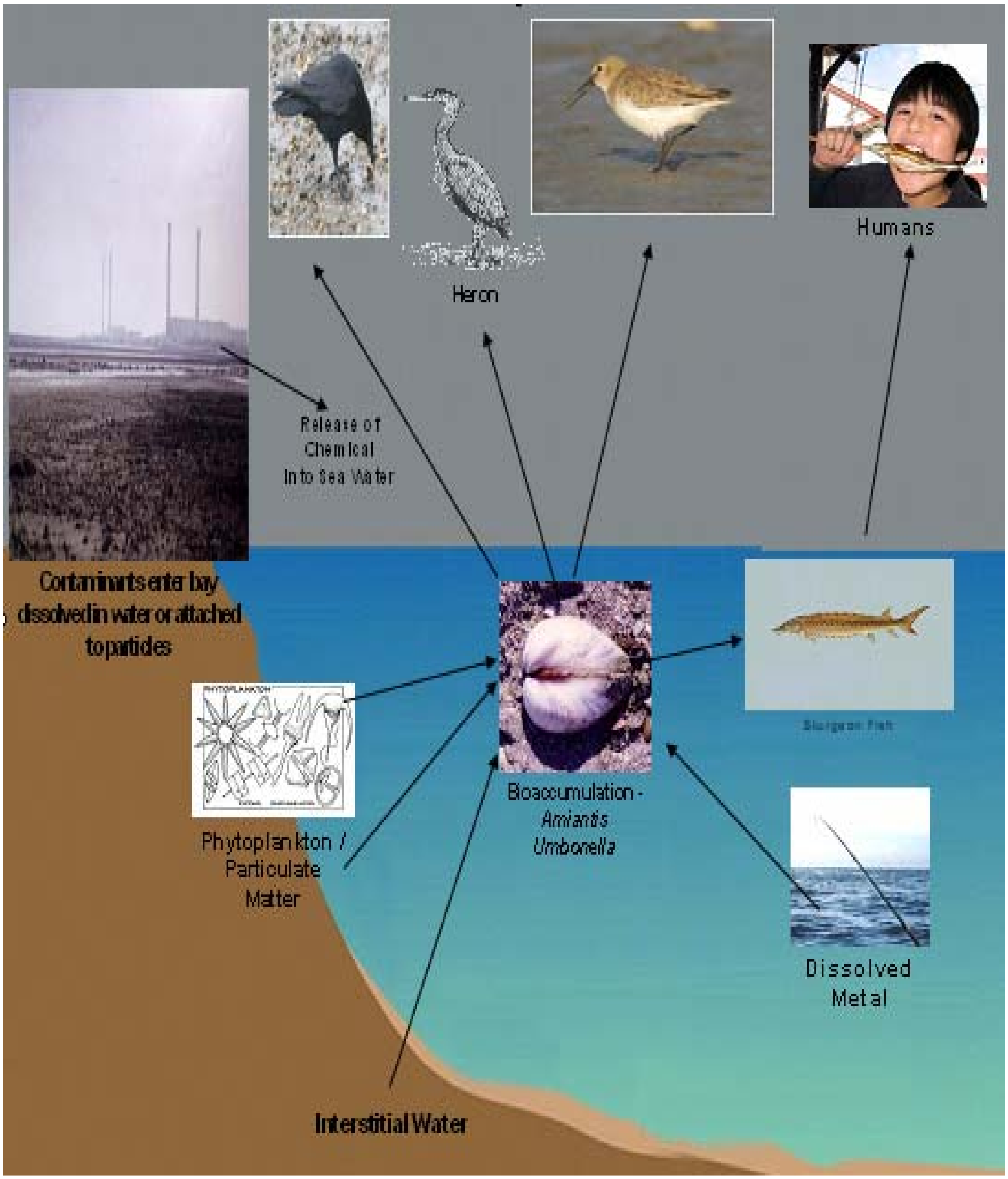

Figure 1. Conceptual Site Bioaccumulation Model.

Schematic representation of the conceptual site bioaccumulation model showing clam *A. umbonella* and potential routes of contaminants mobility and fate.

1.4. Conceptual Site Bioaccumulation Model

A conceptual site bioaccumulation model was developed (Figure 1) showing metal contaminants entering a system, their transportation, and routes of exposure to clams population and humans. The primary route for contaminant (metal) to enter any reach of the Kuwait Bay was through surface waste water discharge (thick concentrated brine and hot water) from desalination and electric power plants. Clams as a benthic facultative filter feeder received all these contaminants through different exposure pathways such as metals dissolved in water column through gills, ingestion of food (phytoplankton) and particulates matter. Clams might get contaminants from interstitial water due to stress or poor quality or quantity of suspended particulate matter that cause the switch from filter to deposit feeding. Michael et al., (2002) has previously demonstrated that pore water (interstitial) is the most sensitive indicator for controlling the metal partitioning between solid and dissolved phase and therefore their potential bioavailability. Contaminants loading from each of the routes had body burdens and metal concentrations increase in soft tissues of the clams.

2. The Accumulation of trace metals in the clam *A. umbonella* near a Desalination/Power Plant compared to a reference site in Kuwait.

2.1. Abstract

To evaluate the clam *Amiantis umbonella* as a bio-indicator of metal contamination, the concentrations of a number of metals Hg, Cd, Pb, Cu, Ni, Mn, Cr, V, Zn and Fe were determined in clams, water, and sediment in Kuwait Bay of the Arabian Gulf at varying distances (0.25 to 5 km) from desalination and power plant waste water discharges. The primary objective of this study was to evaluate the relationship of all ten metal concentrations in the soft tissue of clams, water, and sediment and to compare them with a reference site, 5 km away from the point source, and with results obtained from other geographical areas of the world. The Hg, Cd, Cu, Ni, Cr, V and Pb were significantly higher in water at all sites near desalination and power plants compared to the reference sites. All metals were also higher in sediment near the point source compared to the reference sites. However, some metals (Hg, Cu, Cd, Cr, Mn, Pb) exhibited a spatial gradient within contaminated sites near the desalination and the power plants. The concentrations of Ni, Cu, Cd, Pb, Mn, V and Cr in sediment were highly correlated with those in water. There were significant locational differences of metal concentrations in clams. The clams collected from sites ¼ km from the desalination/power plant had the highest levels of Cu, Ni, Pb, Mn and Cr compared to the other sites farther away from the point source. The Hg, Cd, Cu, Ni, Cr, and V concentrations were significantly higher in the soft tissues of clams from all stations adjacent to the desalination and power plants compared to those from the reference sites.

`In this study a statistically significant correlations (Linear regression) of Hg, Cd, Cu, Cr and Zn concentrations in clam soft tissues and sediment, (R^2= 0.63, 0.54, 0.58, 0.63, 0.59) we observed but the concentrations of these metals were weakly correlated between clams, soft tissue, and water. In contrast Fe, Mn and Pb concentrations were strongly correlated with concentrations in water (R^2= 0.40, 0.28, 0.67) and weakly with those in sediment. There were inverse correlations between V concentrations in clam soft tissues and those in sediment and in water. The levels of metals in clams from this study are generally within the range of mean values reported in the literature from all other areas of the world. Only elevated levels of Hg (6.2 ppm), Pb (2.0 ppm) and Cd (4.8 ppm) from Kuwait exceeded the levels set by FAO for human consumption.

2.2. Introduction

Metal bioaccumulation in macro-invertebrate tissues can be used for bio-monitoring of metal exposures that can either adversely affect the health of the organisms themselves, or be transferred up the food web to affect higher organisms (Luoma, 1996; Burger, 2006). Bivalves present at the second tropic level in marine ecosystems have long been known to accumulate both essential and non-essential trace elements, providing a time-integrated indication of environmental contamination (Dallinger and Rainbow, 1993). Many researchers have suggested the potential use of bivalves, especially mussels, clams, and oysters, as bio-monitors or bio-indicators for monitoring metal contamination in aquatic systems (Philip, 1980; Bryan et al., 1985; Cossa, 1988; Philips and Rainbow, 1993; Claisse et al., 2001; Szefer et al., 2002. Cunningham (1979) pointed out that by definition biological systems can only respond to what is bio-available to them, and test organisms therefore provide the best indication of bio-available pollutants.

Bivalves are an important component of the benthos in the coastal ecosystem and are commonly used bio-sentinel organisms to assess the exposures of the benthos to metal contamination (Goldberg et al., 1983; O'Connor 1992; Brown and Luoma, 1995). The criteria by which organisms are accepted as bio-indicators for the assessment of contamination were established 25 years ago and remain unchanged, such as the mussels watch program. The importance of bivalves, in pollution impact studies is shown by the magnitude and longevity of the International Mussel Watch (Goldberg, 1975). This program originated in the late 1960s and continues to maintain momentum today. (Cantillo, 1980; O'Connor, 1998; Jeng et al., 2000). These established, large-scale monitoring programs provide information on both current status and long- term trends, which allow evaluation of species, population, and ecosystem effects (Burge, 2006).

Bivalves concentrate many metals in their soft tissues (Borchardt, 1983; Fischer 1988) and the tissue metal levels represent a time- integrated response to bio-available metals in food and water (bioaccumulation). They are ideal bio-monitors of aquatic systems, because they are tolerant of ecological variations in parameters such as salinity and temperature. Bivalve has the ability to accumulate organic and metallic pollutants at concentrations several orders of magnitude above those in the ambient environment without being killed by the levels encountered (Bryan et al., 1977, 1983).

In recent years, researchers have focused their attention on the identification of other possible bio-monitors or bio-indicators for trace metal pollution to expand the current understanding of different bioaccumulation strategies for trace metals.

Monitoring schemes will be most useful if they include multiple species representing different tropic levels (Burger, 2006).

Individual bio-monitors respond differently to different sources of bio-available metals (e.g., solution, sediment, or in food). Element concentrations in bivalves at the same location differ between different species and individuals due to species-specific abilities or capacities to regulate or accumulate trace metals (Reinfelder et al., 1997; Otchere et al., 2003). Aquatic and related fauna such as mussels (O'Connor, 1992), clams (Burdon-Jones and Denton, 1984), barnacles (Fialkowski and Newman, 1998), colonizing sea birds (Burger, 1997; Burger and Gochfeld, 2004), and fish (Denton and Burdon-Jones, 1986) are reliable bioindicators of heavy metal-monitoring programs in marine ecosystems.

To understand heavy metal bioavailability in a marine habitat, it is necessary, therefore, to use a suite of bio-indicators that reflect metal bio-availabilities in all available sources (Phillip, 1990; Rainbow, 1990, 1993). Comparative use of different bio-indicators will also allow identification of the particular source of the contaminant metal (Phillip & Rainbow, 1993).

The overall goal of the study is to examine the use of the clam *A. umbonella* (bivalve) as a potential bio-indicator for metal contamination in the Kuwait marine environment. In addition to its use in environmental evaluations, the amount of pollutants bio-accumulated is a concern, as these bivalves are consumed by humans and wildlife.

Clam *A. umbonella* were used as a bio-indicator of metal contamination. This clam typically lives and feeds at a depth of 7-8 cm in fine sediments and obtains nutrition from filter and deposit-feeding. These clams are also an important link for the transfer of contaminants between sediments, water, and higher organisms, including man. Hence, information on the contaminant concentrations in their tissues is useful in considering toxicological and public health implications of marine coastal contamination (Phillip & Rainbow, 1993). Contaminants such as metals can have adverse effects on the health and wellbeing of organisms, including humans. Thus, there is a need for information on contaminant trends, especially for coastal regions, which are increasingly industrialized and developed (Burger and Gochfeld, 2002). Understanding the fate and effect of chemicals can be used to assess the health of ecosystems and to provide early warnings of changes in the environment that might indicate adverse effects (Burger, 2002)

In this study, the concentrations of the metals Hg, Cd, Cu, Ni, Mn, Pb, Cr, Iron, V, and Zn in four coastal sites were examined in sediments, water, and in the infaunal clam *A. umbonella*, a bivalve that is common in Kuwait Bay along the Arabian Gulf. In addition, metal levels in clams will be compared to the results obtained from other geographical areas of the world.

The primary objective of this study to compare metal bioaccumulation in the clam *A. umbonella* population living adjacent to a water desalination and electric power plant in Kuwait Bay (Figure/Map 2) with a reference site at the Umm-Kishish study site, 5 km away from the point source. It was expected that the reference site would support a higher biomass of the clam population with a very low concentration of metals (Figure/Map 2).

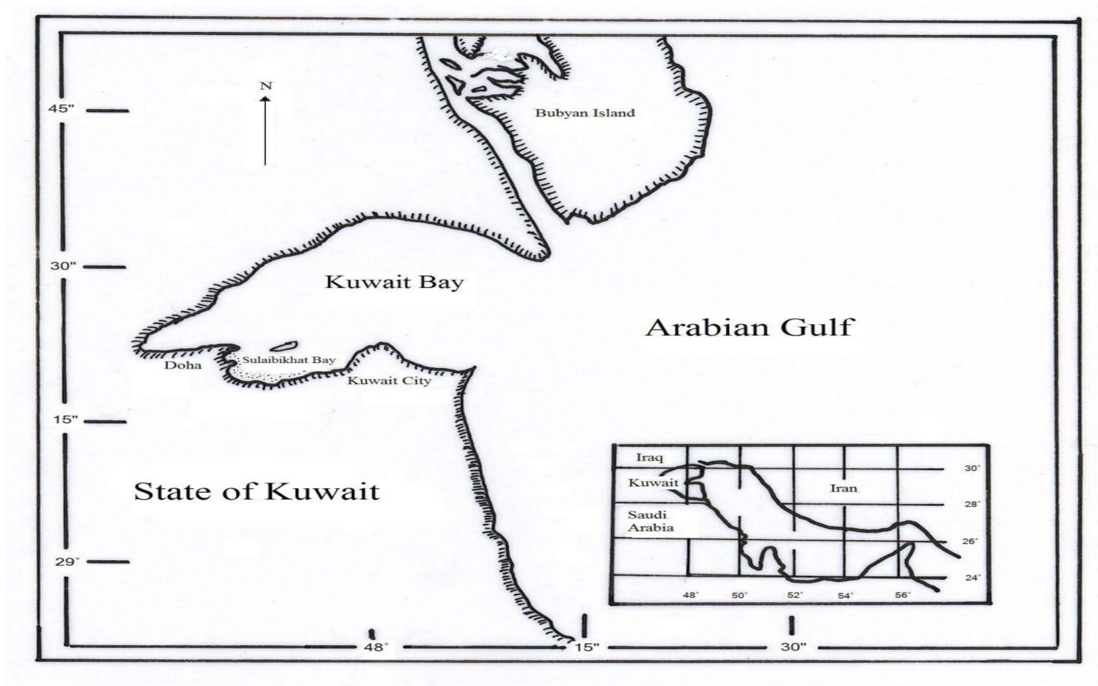

Figure 1. MAP: Geographical location of Kuwait at Arabian Gulf

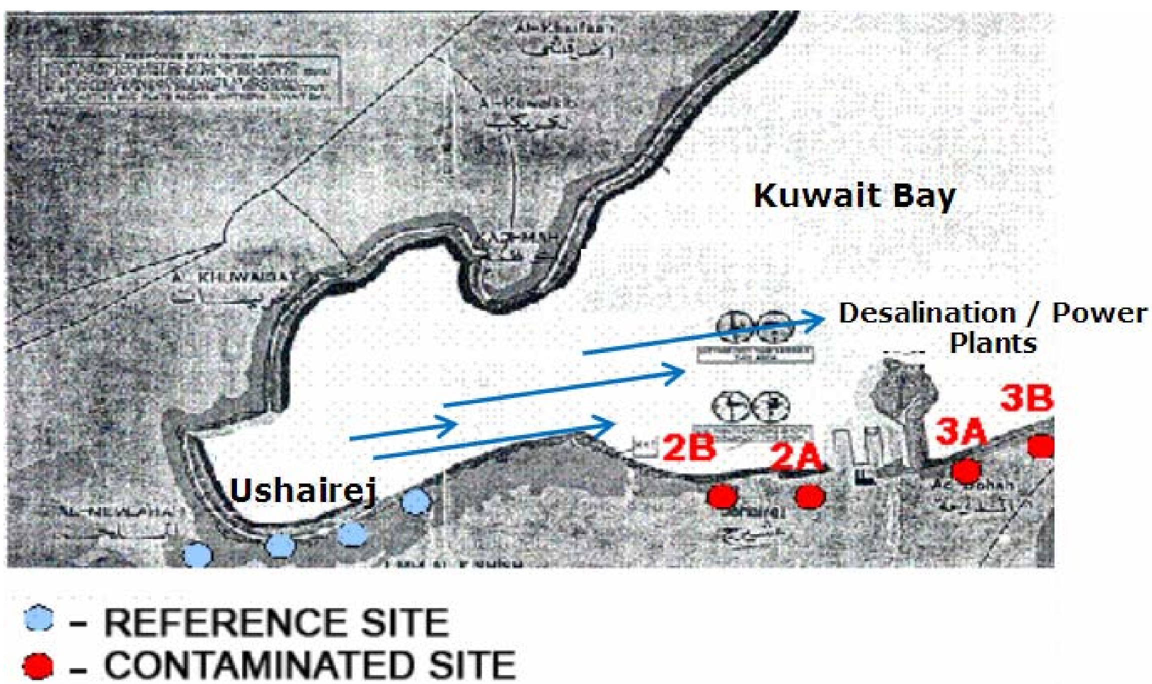

Figure 2. MAP: Kuwait Bay including sampling sites (2A, 2B, 3A, and 3B).

Figure 3. PICTURE: Desalination and Power Plants and Contaminated Coastal site.

Figure 4. PICTURE: Clams *A. umbonella* at low tide (near Desalination and Power Plants)

To determine whether the indigenous clam *A. umbonella* can serve as an effective bio-monitoring species for the waste water discharge from desalination and power plant in Kuwait Bay, the relationship between the total concentrations of Hg, Pb, Cd, V, Zn, Mn, Cu, Fe, and Ni in clams soft tissues, sediment and water from contaminated sites near desalination and power plants and compared to the reference sites.

The null hypothesis was tested that

1 There is no difference in metal concentrations in the soft body of clam populations living in marine water near water desalination and electric power plans compared to the reference sites.

2 There is no correlation between the concentrations of heavy metals present in the tissues of clams and those in water and sediment.

2.3. Materials and Methods

2.3.1. Study Sites

Kuwait's coastline, including its islands, has been estimated at about 400 km. It is mainly sandy and muddy with very few inlets other than Kuwait Bay. The northern coast from Doha northwards is bordered by extensive mudflats created by the floodwaters of the Tigris and Euphrates Rivers, which enter the north end of the Gulf through the Shatt Al – Arab Water way. Doha coast is mainly sandy. The maximum tidal range is 3.5 – 4. 0 m. The water temperature varies widely from 12 ^{o}C in January to 34 ^{o}C in July. Salinities are generally high throughout most of the year, ranging from 38 to 42 ppt.

The sampling sites for clams *A. umbonella* are shown in Figure/Map 2. The sites were selected on the basis of on the availability of the clams and their proximity to the desalination and power plants and also the distributions of suspected anthropogenic influences in the area. Station 2A was ¼ mile from the outflow of the desalination plan, and 3A was ¼ mile far from the outflow of the steam power plant (Figure/Picture 3).

These sites are evenly spread out around the Doha Harbor and Ushairej area, which lies in the northwest of Kuwait, with a relatively highly developed desalination plant and steam power plant. This is one of the largest steam power plants in the world, which can be fueled alternatively with natural gas, crude oil, and light fuel oil. It was constructed on the coast of the Gulf in the vicinity of the capital city of Kuwait.

2.3.2. Clams Collection and Preparation

A minimum of 100 clams (n=100) were randomly collected at each of four sites (2A, 2B, 3A, and 3B), during December 1999, January 2000 with a similar shell length to minimize the effect of body weight (Marina and Enzo, 1983). After collection the clams were transported to the laboratory in self-supporting tanks (capacity of 150 liters, aerated sea water continuously flowing). The clams were depurated in seawater for approximately 24 hours. The seawater salinity and temperature were measured during each sampling.

For whole clam analysis, 100 clams were divided into 3 composite samples, A, B, and C, of 33 clams each. Clams wet weights were recorded (mg). All clams were shucked and whole tissues were mixed to form a single composite sample from contaminated and reference locations for investigation of metal depositions (atomic absorption spectroscopy). The tissues were stored frozen in pre-cleaned jars for further analysis.

To investigate the variability of the metal concentrations individually, two samples of 32 clams were collected randomly from contaminated sites 2A and 3A and the reference sites.

Prior to analysis, clam tissues were freeze dried to a constant weight (±0.5%). Then the sample was ground to homogenize it and sieved in order to normalize the variation in the grain size distributed. Samples were extracted by placing 0.5 gm of the homogenized sample in a Teflon beaker and heating it (120 °C) with concentrated HNO3 until dry. 5 ml acid (HNO3+HCLO4+HF) was added to the sample and heated up to 120 °C until dry. Then the sample was cooled to room temperature and passed through filter paper and poured into the 50 ml volumetric flask, diluting with HCl 0.1% to make up the volume.

2.3.3. Sediment and Water collection

Water and sediment samples were also collected during sampling from each of the four stations near the desalination/power plans and the reference site. Three samples of sediment and water were collected from each contaminated site (2A, 2B, 3A, and 3B), and 12 samples of sediment and water were collected randomly from the reference site. Beside the sub-samples, 3 standards were digested to validate the analytical procedure. Along with the samples and standards, 3 blanks were carried for setting up the system base line. Sediments were air dried and passed though a 2 mm sieve and kept in a deep freezer (-20 ^{o}C) until processing. Then the sample was dried by a freeze dryer until all humidity and water were decanted from the sample. This step was continued for five days until the weight was reduced to less than 0.5%. Then the samples were ground to homogenize them and sieved in order to normalize them. The sample was kept in a glass bottle for further digestion.

A weight of 0.5 gm of the sediment powder was kept in a Teflon beaker that was cleaned with 20% of HNO_3. 5 ml of conc. HNO_3 +$HClO_4$ +HF with ratio of 3: 2: 1 was added to the sample and evaporated to near dryness while covering the samples continuously for 3 hours. The sample was passed through a filter paper and transferred to a 50ml volumetric flask, and the volume was made with 0.1% HCl. The final sample was ready for elemental analysis by ICP-AES (Inductivity Coupled Plasma Atomic Emission Spectrometry).

The final results were calculated by the following equation:

Metal/gm dry weight= A*V/D

A: The concentration of the sample measured

V: Final volume of the collected sample.

D: Dry weight of the sample

After the seawater samples were delivered to the lab, 5ml of HNO3 was added to the sample and filtered by a vacuum pump. The pH was adjusted between 5.0 – 5.5 by the addition of 2N HNO3 solution. The samples were then passed through the Ion-exchange column at a rate of 2ml/min, and 25ml of ammonium acetate was added. The sample was poured into a 25 ml volumetric flask, and 25ml of 2N HNO_3 was added. At this point, the samples were ready for atomic absorption.

2.3.4. Statistical Analysis

T tests were used to determine whether the mean of the 10 metals from the desalination and power plants and the reference sites differed from each other ($P<0.05$). A linear regression analysis and a coefficient of determination were used for the correlation between independent and dependent variables. At the same time, ANOVA (F test) was used to find out the significance of the regression coefficient (R^2).

2.4. Results

2.4.1. Spatial differences of metal concentrations in Water

Mean metal concentrations in water, SD, T-test, RSD (% of relative standard deviation) at the studied locations (2A. 2B, 3A, and 3B) are given in table 1, 2, 3, and 4. Bar graphs show (Figures 1, 2, and 3) the average enrichment for all metals in water samples and the percentage (%) increase at the contaminated sites compared to the reference sites from four sampling locations.

There was a great variation in metal concentrations in all four sites. Cu, Cd, Ni, Cr, Mn, Pb, and V are significantly different ($P< 0.05$) at all contaminated sites compared to reference sites.

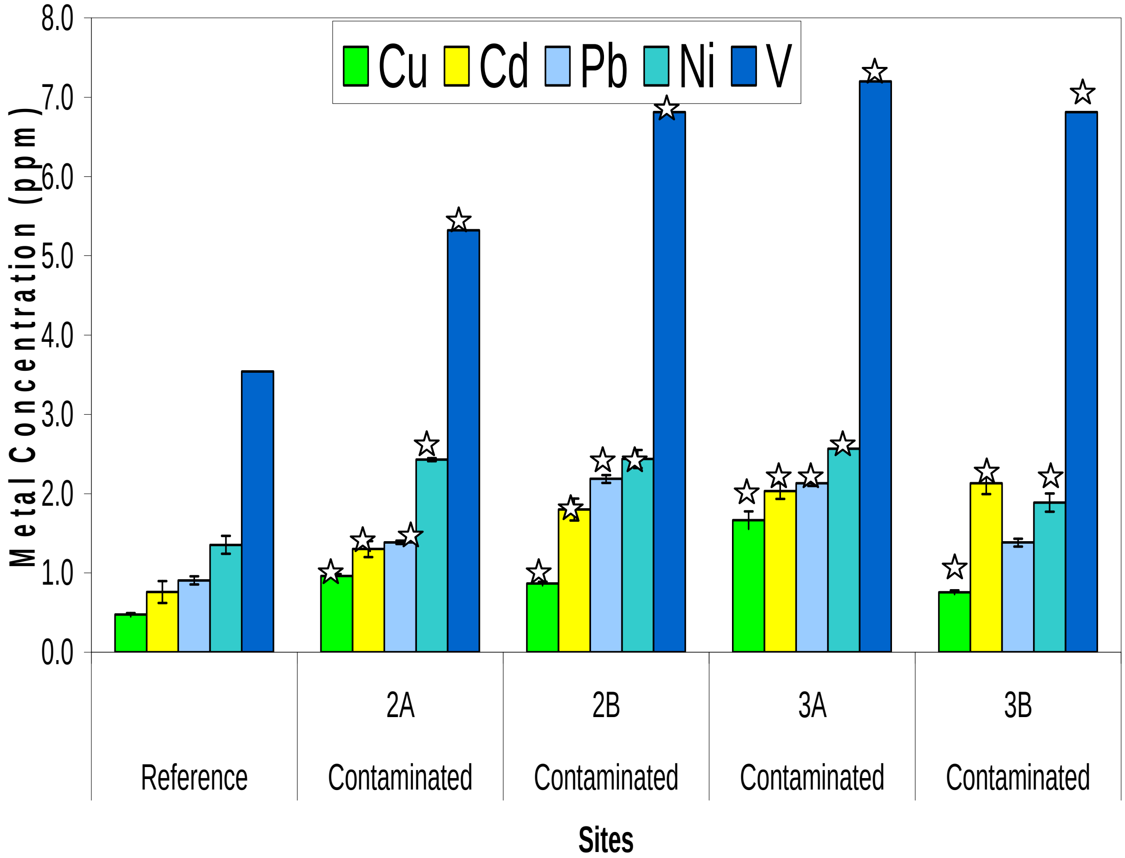

Figure 1. Average Cu, Cd, Pb, V concentrations in water (ppb) at the contaminated and the reference site.

Error bars show SD. Average values represent triplicate (n=3). The Symbol☆ represents significant difference of metal concentrations at the contaminated site as compared to the reference site.

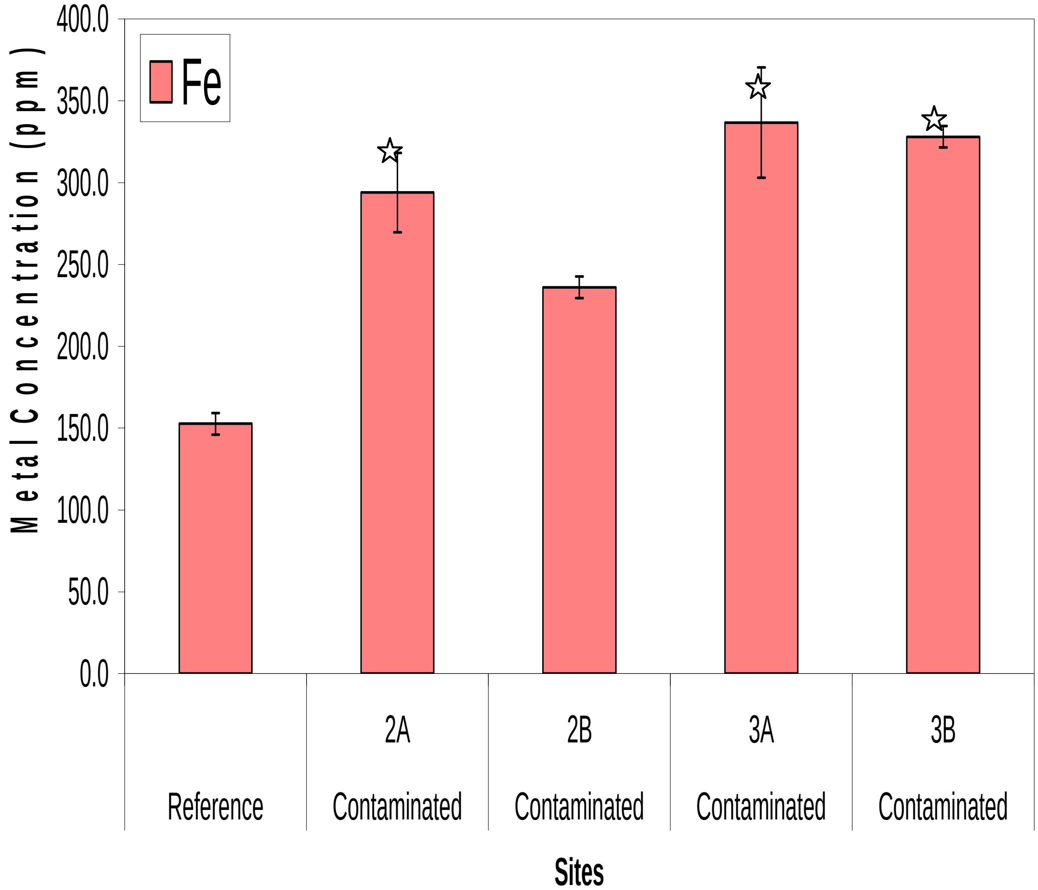

Figure 2. Average Fe concentration in water (ppb) at the contaminated and the reference site.

Error bars show SD. Average values represent triplicate (n=3). The symbol☆ represents significant difference of metal concentrations at the contaminated site as compared to the reference site.

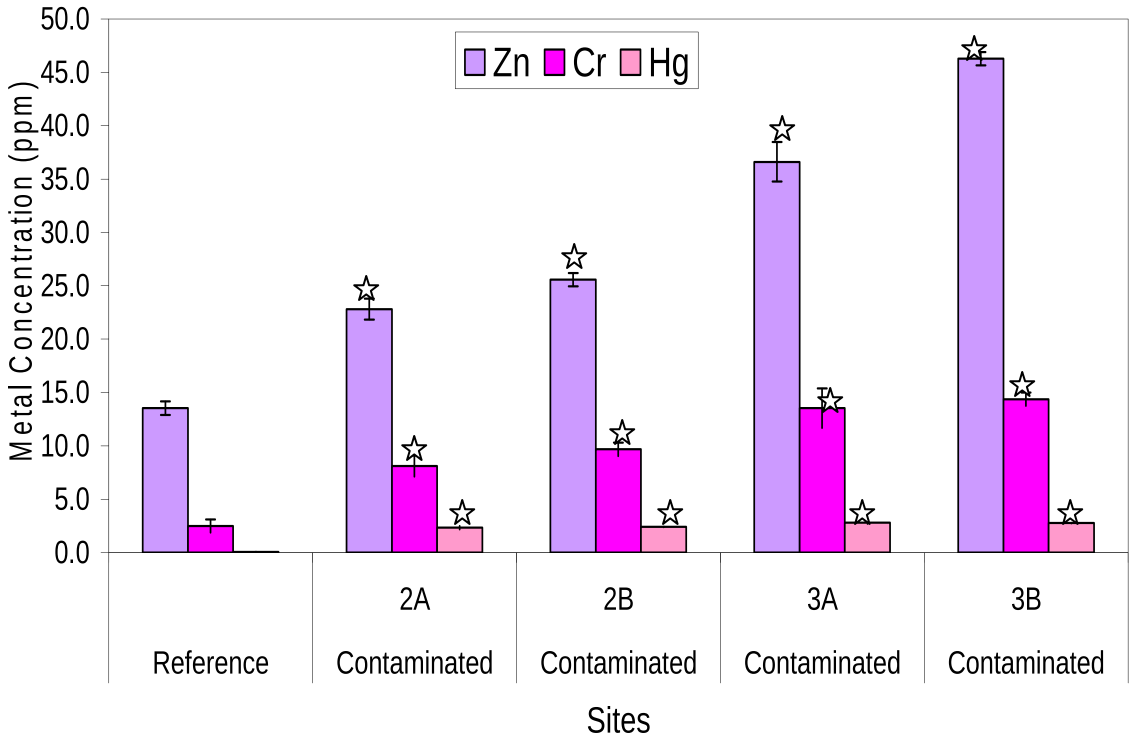

Figure 3. Average Zn, Cr, and Hg concentrations in water (ppb) at the contaminated and the reference site.

Error bars show SD. Average values represent triplicate (n=3). The symbol ☆ represents significant difference of metal concentrations at the contaminated site as compared to the reference site.

The highest Cu concentrations were found in water at station 2A near the desalination plant (Table.1). It was 2-fold higher than at the reference site. The Cd concentration was greater than at the reference site by a factor of 1.8. In comparison, Ni, Cd, Pb, V, Iron, and Zn concentrations were a factor of 1.5-1.8 higher than those at the reference site.

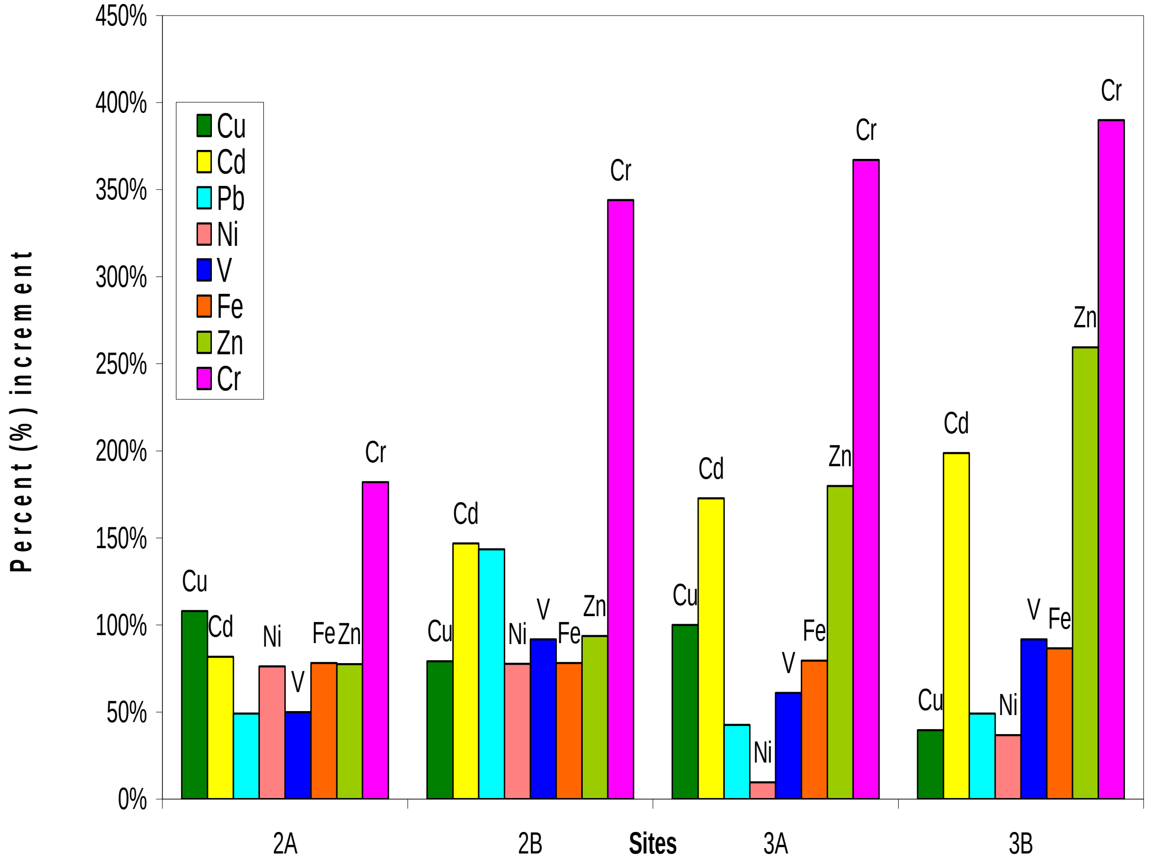

Figure 4. Relative enrichment percent (%) of metals in the water of contaminated site with respect to the reference site.

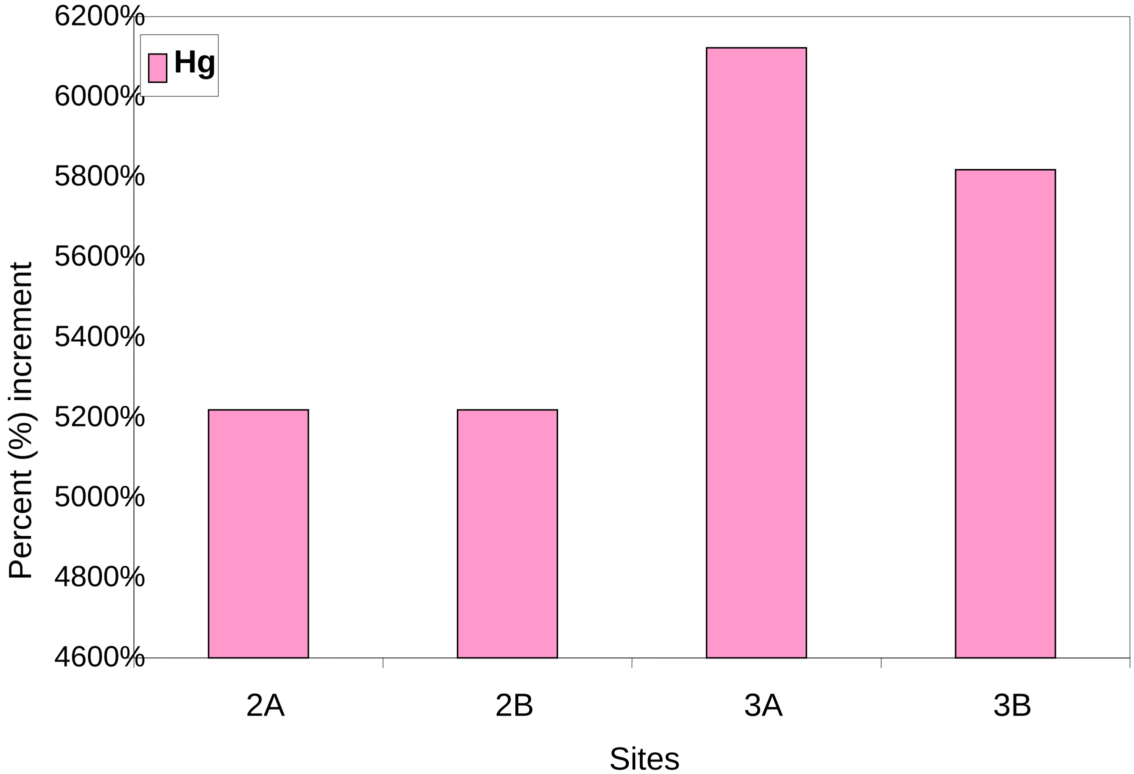

Figure 5. Relative enrichment (%) of Hg in the water of contaminated sites with respect to the reference site.

Table 1. Metal Concentrations in Water at Location 2A

Metal concentrations in water (ppb), SD (standard deviation), T-test, contamination/ reference sites, RSD (relative standard deviation) at location 2A.

Metals		Location 1			Location 2			Location 3			Location 4			Mean	SD	T-test	Contam/ Refere	RSD
	Sites	A	B	C	A	B	C	A	B	C	A	B	C					
Cu	Reference	0.45	0.48	0.53	0.46	0.47	0.49	0.47	0.48	0.49	0.45	0.46	0.45	0.5	0.0	4.96E-14	2.0	5%
	Contaminated	0.94	0.95	0.99										1.0	0.0			3%
Cd	Reference	0.6	0.8	0.7	0.6	0.8	0.9	0.7	0.8	0.9	0.5	0.9	0.9	0.8	0.1	1.33E-05	1.7	18%
	Contaminated	1.2	1.3	1.4										1.3	0.1			8%
Pb	Reference	0.85	0.89	0.88	0.88	0.98	0.94	0.83	0.92	0.96	0.98	0.89	0.86	0.9	0.1	3.76E-10	1.5	6%
	Contaminated	1.4	1.4	1.36										1.4	0.0			2%
Ni	Reference	1.25	1.32	1.27	1.37	1.43	1.29	1.29	1.23	1.39	1.45	1.64	1.32	1.4	0.1	3.36E-10	1.8	8%
	Contaminated	2.43	2.45	2.41										2.4	0.0			1%
V	Reference	3.53	3.55	3.59	3.65	3.32	3.55	3.98	3.87	3.33	3.54	3.47	3.11	3.5	0.2	4.85E-09	1.5	7%
	Contaminated	5.37	5.28	5.31										5.3	0.0			1%
Fe	Reference	148.4	145.2	145	145	149	164.1	161.4	157	158	152	153.2	153.2	152.6	6.6	2.75E-11	1.9	4%
	Contaminated	321	287	274										294.0	24.3			8%
Zn	Reference	12.6	12.8	12.9	14.3	14.8	14.3	13.9	13.5	13.6	12.7	13.4	13.5	13.5	0.7	1.60E-10	1.7	5%
	Contaminated	22.6	21.5	24.3										22.8	1.4			6%
Cr	Reference	1.9	2.05	2.7	2.1	3.6	2.5	3.6	2.2	3.1	2.1	2	2	2.5	0.6	6.04E-09	3.3	25%
	Contaminated	9.2	7.8	7.3										8.1	1.0			12%
Hg	Reference	0.05	0.04	0.03	0.05	0.04	0.06	0.04	0.06	0.03	0.05	0.06	0.05	0.0	0.0	2.04E-17	50.0	23%
	Contaminated	2.2	2.3	2.5										2.3	0.2			7%

The highest concentration of Ni was determined in location 2B, ¼ km away from the water desalination plant. The Ni concentration was higher than that at the reference sites by a factor of 2.8. The concentrations of Cd and Pb were a factor of 2.5 higher than those at the reference sites. The concentrations of Cu, V and Iron were greater than those at the reference sites by a factor of 1.8 – 1.9 (Table. 2).

Table 2. Metal Concentrations in Water at Location 2B

Metal concentrations in water (ppb), SD (standard deviation), T-test, contamination/ reference sites, RSD (relative standard deviation) at location 2B.

Metals		Location 1			Location 2			Location 3			Location 4			Mean	SD	T-test	Contam/ Refere	RSD
	Sites	A	B	C	A	B	C	A	B	C	A	B	C					
Cu	Reference	0.45	0.45	0.48	0.53	0.46	0.47	0.49	0.47	0.48	0.49	0.45	0.46	0.5	0.0	2.30E-08	1.8	5%
	Contaminated	0.98	0.76	0.86	0	0	0	0	0	0	0	0	0	0.9	0.1			13%
Cd	Reference	0.6	0.8	0.7	0.6	0.8	0.9	0.7	0.8	0.9	0.5	0.9	0.9	0.8	0.1	8.96E-09	2.4	18%
	Contaminated	1.7	1.8	1.9	0	0	0	0	0	0	0	0	0	1.8	0.1			6%
Pb	Reference	0.85	0.89	0.88	0.88	0.98	0.94	0.83	0.92	0.96	0.98	0.89	0.86	0.9	0.1	1.98E-15	2.4	6%
	Contaminated	2.15	2.19	2.22	0	0	0	0	0	0	0	0	0	2.2	0.0			2%
Ni	Reference	1.25	1.32	1.27	1.37	1.43	1.29	1.29	1.23	1.39	1.45	1.64	1.32	1.4	0.1	3.04E-10	1.8	8%
	Contaminated	2.43	2.45	2.43	0	0	0	0	0	0	0	0	0	2.4	0.0			0%
V	Reference	3.53	3.55	3.59	3.65	3.32	3.55	3.98	3.87	3.33	3.54	3.47	3.11	3.5	0.2	2.36E-12	1.9	7%
	Contaminated	6.81	6.84	6.79	0	0	0	0	0	0	0	0	0	6.8	0.0			0%
Fe	Reference	148.4	145.2	145	145	149	164.1	161.4	157	158	152	153	153	152.6	6.6	3.43E-07	1.5	4%
	Contaminated	210	224	274	0	0	0	0	0	0	0	0	0	236.0	33.6			14%
Zn	Reference	12.6	12.8	12.9	14.3	14.8	14.3	13.9	13.5	13.6	12.7	13.4	13.5	13.5	0.7	1.37E-12	1.9	5%
	Contaminated	24.5	25.7	26.5	0	0	0	0	0	0	0	0	0	25.6	1.0			4%
Cr	Reference	1.9	2.05	2.7	2.1	3.6	2.5	3.6	2.2	3.1	2.1	2	2	2.5	0.6	1.03E-08	3.9	25%
	Contaminated	9.7	7.8	11.5	0	0	0	0	0	0	0	0	0	9.7	1.9			19%
Hg	Reference	0.05	0.02	0.03	0.07	0.05	0.06	0.04	0.04	0.03	0.06	0.03	0.06	0.0	0.0	1.09E-19	53.3	35%
	Contaminated	2.3	2.4	2.5	0	0	0	0	0	0	0	0	0	2.4	0.1			4%

The highest concentration of Ni and Cd was found in location 3A respectively by a factor of 2.9 and 2.8 higher than the reference sites. The Cu concentration was 2- fold higher than the reference sites. The Pb, V and iron concentrations were a factor of 1.4, 1.6, and 1.8 greater than at the reference sites (Table 3)

The Cd concentration was 3- fold higher in location 3B than the concentration in the reference site. The V and Iron were a factor of 1.9 higher than the reference sites. The Cu and Ni were a factor of 1.4 higher than the reference site. The Pb concentration was greater than the reference site by a factor of 1.5 (Table 4). Regression between the trace metal concentrations in sediment and water were tested for clams collected from all four stations. The Mn, Cr, Iron, and Pb concentrations in sediment showed a positive regression slope with concentrations in water. There was a significant positive correlation of V concentration in sediment and water ($p<0.05$).

Table 3. Metal Concentrations in Water at Location 3A

Metal concentrations in water (ppb), SD (standard deviation), T-test, contamination/ reference sites, RSD (relative standard deviation) at location 3A.

Metals		Location 1			Location 2			Location 3			Location 4			Mean	SD	T-test	Contam/ Referen	RSD
	Sites	A	B	C	A	B	C	A	B	C	A	B	C					
Cu	Reference	0.45	0.45	0.48	0.53	0.46	0.47	0.49	0.47	0.48	0.49	0.45	0.46	0.5	0.0	4.26E-13	2.0	5%
	Contaminated	0.92	0.95	0.96	0	0	0	0	0	0	0	0	0	0.9	0.0			2%
Cd	Reference	0.6	0.8	0.7	0.6	0.8	0.9	0.7	0.8	0.9	0.5	0.9	0.9	0.8	0.1	9.12E-10	2.7	18%
	Contaminated	1.9	2.1	2.1	0	0	0	0	0	0	0	0	0	2.0	0.1			6%
Pb	Reference	0.85	0.89	0.88	0.88	0.98	0.94	0.83	0.92	0.96	0.98	0.89	0.86	0.9	0.1	4.66E-09	1.5	6%
	Contaminated	1.3	1.4	1.3	0	0	0	0	0	0	0	0	0	1.3	0.1			4%
Ni	Reference	1.25	1.32	1.27	1.37	1.43	1.29	1.29	1.23	1.39	1.45	1.64	1.32	1.4	0.1	2.68E-01	1.0	8%
	Contaminated	1.3	1.4	1.5	0	0	0	0	0	0	0	0	0	1.4	0.1			7%
V	Reference	3.53	3.55	3.59	3.65	3.32	3.55	3.98	3.87	3.33	3.54	3.47	3.11	3.5	0.2	3.03E-10	1.6	7%
	Contaminated	5.8	5.9	5.7	0	0	0	0	0	0	0	0	0	5.8	0.1			2%
Fe	Reference	148.4	145.2	144.7	144.8	149.3	164.1	161.4	157.4	158.4	151.6	153.2	153.2	152.6	6.6	3.92E-09	2.2	4%
	Contaminated	356	378	276	0	0	0	0	0	0	0	0	0	336.7	53.7			16%
Zn	Reference	12.6	12.8	12.9	14.3	14.8	14.3	13.9	13.5	13.6	12.7	13.4	13.5	13.5	0.7	8.52E-15	2.7	5%
	Contaminated	34.6	36.9	38.3	0	0	0	0	0	0	0	0	0	36.6	1.9			5%
Cr	Reference	1.9	2.05	2.7	2.1	3.6	2.5	3.6	2.2	3.1	2.1	2	2	2.5	0.6	5.30E-12	5.4	25%
	Contaminated	13.8	14.7	12.1	0	0	0	0	0	0	0	0	0	13.5	1.3			10%
Hg	Reference	0.02	0.04	0.04	0.06	0.05	0.06	0.07	0.04	0.03	0.04	0.02	0.04	0.0	0.0	1.38E-20	65.9	36%
	Contaminated	2.9	2.7	2.8	0	0	0	0	0	0	0	0	0	2.8	0.1			4%

Table 4. Metal Concentrations in Water at Location 3B

Metal concentrations in water (ppb), SD (standard deviation), T-test, contamination/ reference sites, RSD (relative standard deviation) at location 3B.

Metals		Location 1			Location 2			Location 3			Location 4			Mean	SD	T-test	Contam / Reference	RSD
	Sites	A	B	C	A	B	C	A	B	C	A	B	C					
Cu	Reference	0.45	0.45	0.48	0.53	0.46	0.47	0.49	0.47	0.48	0.49	0.45	0.46	0.5	0.0	9.78E-08	1.6	5%
	Contaminated	0.76	0.84	0.67	0	0	0	0	0	0	0	0	0	0.8	0.1			11%
Cd	Reference	0.6	0.8	0.7	0.6	0.8	0.9	0.7	0.8	0.9	0.5	0.9	0.9	0.8	0.1	5.94E-10	2.8	18%
	Contaminated	2	2.1	2.3	0	0	0	0	0	0	0	0	0	2.1	0.2			7%
Pb	Reference	0.85	0.89	0.88	0.88	0.98	0.94	0.83	0.92	0.96	0.98	0.89	0.86	0.9	0.1	3.93E-10	1.5	6%
	Contaminated	1.4	1.39	1.36	0	0	0	0	0	0	0	0	0	1.4	0.0			2%
Ni	Reference	1.25	1.32	1.27	1.37	1.43	1.29	1.29	1.23	1.39	1.45	1.64	1.32	1.4	0.1	1.32E-06	1.4	8%
	Contaminated	1.9	1.89	1.87	0	0	0	0	0	0	0	0	0	1.9	0.0			1%
V	Reference	3.53	3.55	3.59	3.65	3.32	3.55	3.98	3.87	3.33	3.54	3.47	3.11	3.5	0.2	2.36E-12	1.9	7%
	Contaminated	6.81	6.84	6.79	0	0	0	0	0	0	0	0	0	6.8	0.0			0%
Fe	Reference	148.4	145.2	145	145	149	164.1	161.4	157.4	158.4	151.6	153.2	153.2	152.6	6.6	9.73E-10	2.1	4%
	Contaminated	321	376	287	0	0	0	0	0	0	0	0	0	328.0	44.9			14%
Zn	Reference	12.6	12.8	12.9	14.3	14.8	14.3	13.9	13.5	13.6	12.7	13.4	13.5	13.5	0.7	1.16E-15	3.4	5%
	Contaminated	44.7	44.9	49.2	0	0	0	0	0	0	0	0	0	46.3	2.5			5%
Cr	Reference	1.9	2.05	2.7	2.1	3.6	2.5	3.6	2.2	3.1	2.1	2	2	2.5	0.6	6.01E-12	5.8	25%
	Contaminated	14.6	15.8	12.7	0	0	0	0	0	0	0	0	0	14.4	1.6			11%
Hg	Reference	0.09	0.04	0.05	0.07	0.05	0.06	0.02	0.05	0.04	0.06	0.03	0.06	0.1	0.0	3.06E-18	53.5	36%
	Contaminated	2.8	2.6	2.9	0	0	0	0	0	0	0	0	0	2.8	0.2			6%

2.5. Spatial differences of metal concentrations in Sediments

All sites close to the outlet of the desalination and power plants were contaminated compared to the reference sites. The Hg, Cu, Cd, Pb, Cd, V, Zn, Cr and Ni concentrations were significant in all sites near the desalination and power plants.

Metal concentrations, mean, SD, T-test, and RSD, (% Relative standard deviation) are listed in Table 5, 6, 7and 8. Bar graphs (Figure. 3 and 4) show the average metal concentrations of Cu, Cd, Cr, Hg, Mn, Pb, Ni, V, Zn and Fe from four sampling locations (reference vs contaminated sites) and the percentage increase in the contaminated site compared to the reference sites.

There were variations of metal concentrations in sediments from all contaminated sites compared to the reference sites.

The 2A location was nearest to the outlet of the desalination plant. The Hg and Cr concentrations were a factor of 21.3 and 3.1 higher at the contaminated sites than the reference sites. The Mn, Zn, V, and Cu concentrations were a factor of 2 higher than the reference site. The Cd, Pb, and iron concentrations were also a factor of 1.9, 1.8 and 2.2 more than the reference sites (table 5).

Table 5. Metal Concentrations in Sediment at Location 2A

Metal concentrations in sediment (ppm), SD (standard deviation), T-test, contamination/ reference sites, RSD (relative standard deviation) at location 2A.

Metals		Location 1			Location 2			Location 3		Location 4			Mean	SD	T-test	Contam/ Referen	RSD
	Sites	A	B	C	A	B	C	A	C	A	B	C					
Cu	Reference	5.3	5.9	5.7	5.3	5.1	5.7	5.3	5.6	5.9	5.7	6.1	5.6	0.3	1.76E-13	2.0	5%
	Contaminated	10.8	11	11.2	0	0	0	0	0	0	0	0	11.0	0.2			2%
Cd	Reference	3.9	4	3.9	3.8	3.9	4.1	2.1	2.7	2.1	2.4	2.3	3.2	0.8	2.05E-05	1.9	26%
	Contaminated	6.2	5.9	6.4	0	0	0	0	0	0	0	0	6.2	0.3			4%
Pb	Reference	45.6	43.6	46.1	48.8	47.8	46.4	45.9	45.7	47.9	45.3	44.9	46.5	1.8	1.32E-14	1.8	4%
	Contaminated	84.2	83.3	84.8	0	0	0	0	0	0	0	0	84.1	0.8			1%
Ni	Reference	12.8	14.1	13.8	21.3	21.8	21.6	28.2	30.2	22.7	21.9	21.4	21.6	5.8	4.55E-05	1.9	27%
	Contaminated	41.2	39.9	41.7	0	0	0	0	0	0	0	0	40.9	0.9			2%
V	Reference	16.2	17.1	16.8	16.7	17.2	16.9	16.3	17.6	16.9	16.3	17.2	16.9	0.4	4.61E-18	2.0	3%
	Contaminated	33.5	33.9	33.8	0	0	0	0	0	0	0	0	33.7	0.2			1%
Fe	Reference	4.9	4.8	5.2	3.7	3.6	3.8	3.5	3.6	4.8	4.2	5.2	4.3	0.7	4.42E-09	2.2	16%
	Contaminated	9.44	9.36	9.38	0	0	0	0	0	0	0	0	9.4	0.0			0%
Zn	Reference	22.8	24	23.8	24.2	25.1	31.7	32.3	23.6	24.1	25.7	23.8	26.0	3.5	1.20E-10	2.4	13%
	Contaminated	62.6	61.2	61.8	0	0	0	0	0	0	0	0	61.9	0.7			1%
Cr	Reference	21.3	19.6	19.2	29.3	30	29.2	21.7	21.6	21.2	23.1	19.5	23.0	4.1	2.71E-11	3.1	18%
	Contaminated	70.4	69.9	71	0	0	0	0	0	0	0	0	70.4	0.6			1%
Hg	Reference	0.25	0.23	0.37	0.12	0.15	0.18	0.18	0.29	0.26	0.25	0.23	0.2	0.1	6.57E-20	21.3	31%
	Contaminated	4.7	4.9	4.7	0	0	0	0	0	0	0	0	4.8	0.1			2%
Mn	Reference	27.6	27.3	28.1	32.7	32.1	27.9	31.4	30.6	28.9	28.6	30.1	27.7	0.4	1.61E-07	2.5	1%
	Contaminated	54.2	68.9	86.1	0	0	0	0	0	0	0	0	69.7	16.0			23%

Sediment samples from location 2B were enriched with higher Hg, Ni, Cu, Cd, V and Zn concentrations compared to the reference sites. The Hg concentration was a factor of 25.4 higher at the contaminated sites compared to the reference sites. The Ni concentration was higher than the reference site by a factor 2.1 (Table 6). The remaining Cu, Cd, V, and Zn concentrations were 2- fold higher than the reference sites. The Cr, Mn, Pb and iron concentrations were a factor of 1.3, 1.9, 1.5, and 1.7 higher than the reference sites respectively.

Table 6. Metal Concentrations in Sediment at Location 2B

Metal concentrations in sediment (ppm), SD (standard deviation), T-test, contamination/ reference sites, RSD (relative standard deviation) at location 2 B.

Metals		Location 1			Location 2			Location 3		Location 4			Mean	SD	T-test	Contam/ Referen	RSD
	Sites	A	B	C	A	B	C	A	C	A	B	C					
Cu	Ref	5.3	5.9	5.7	5.3	5.1	5.7	5.3	5.6	5.9	5.7	6.1	5.6	0.3	6.24E-14	2.3	5%
	Cont	12.9	13.8	12.6	0	0	0	0	0	0	0	0	13.1	0.6			5%
Cd	Ref	3.9	4	3.9	3.8	3.9	4.1	2.1	2.7	2.1	2.4	2.3	3.2	0.8	1.14E-05	2.1	26%
	Cont	5.7	6.8	7.2	0	0	0	0	0	0	0	0	6.6	0.8			12%
Pb	Ref	45.6	43.6	46.1	48.8	47.8	46.4	45.9	45.7	47.9	45.3	44.9	46.5	1.8	1.42E-11	1.5	4%
	Cont	68.1	68	70	0	0	0	0	0	0	0	0	68.7	1.1			2%
Ni	Ref	12.8	14.1	13.8	21.3	21.8	21.6	28.2	30.2	22.7	21.9	21.4	21.6	5.8	6.88E-06	2.1	27%
	Cont	44.3	44.8	45.9	0	0	0	0	0	0	0	0	45.0	0.8			2%
V	Ref	16.2	17.1	16.8	16.7	17.2	16.9	16.3	17.6	16.9	16.3	17.2	16.9	0.4	4.61E-18	2.0	3%
	Cont	33.5	33.9	33.8	0	0	0	0	0	0	0	0	33.7	0.2			1%
Fe	Ref	4.9	4.8	5.2	3.7	3.6	3.8	3.5	3.6	4.8	4.2	5.9	4.3	0.8	8.83E-06	1.7	18%
	Cont	7.28	7.55	7.31	0	0	0	0	0	0	0	0	7.4	0.1			2%
Zn	Ref	22.8	21.6	23.8	24.2	25.1	31.7	32.3	23.6	24.1	25.7	23.8	25.8	3.7	9.09E-09	2.0	14%
	Cont	52.6	51.2	54.8	0	0	0	0	0	0	0	0	52.9	1.8			3%
Cr	Ref	21.3	19.6	19.2	29.3	30	29.2	21.7	21.6	21.2	23.1	19.5	23.0	4.1	9.43E-03	1.3	18%
	Cont	29.3	30	29.2	0	0	0	0	0	0	0	0	29.5	0.4			1%
Hg	Ref	0.25	0.23	0.37	0.12	0.15	0.18	0.18	0.29	0.26	0.25	0.23	0.2	0.1	2.54E-19	25.4	31%
	Cont	5.6	5.5	5.9	0	0	0	0	0	0	0	0	5.7	0.2			4%
Mn	Ref	27.6	27.3	28.1	32.7	32.1	27.9	31.4	30.6	28.9	28.6	30.1	27.7	0.4	3.09E-11	1.9	1%
	Cont	51.8	52.1	51.8	0	0	0	0	0	0	0	0	51.9	0.2			0%

The Hg, Cr, and Cd concentrations were greater than reference sites by a factor of 24.2, 3.1, and 2.8 in sediment at location 3A near the power plant. The Mn and Fe concentrations were a factor of 2.4 and 2.2 higher when compared with the reference sites. The Cu concentration was a factor of 1.9 higher than the reference sites. The Pb, Ni, V and Zn concentrations were a factor of 1.5, 1.3, 1.3, and 1.2 higher than the reference sites respectively (Table 7).

Table 7. Metal Concentrations in Sediment at Location 3A

Metal concentrations in sediment (ppm), SD (standard deviation), T-test, contamination/ reference sites, RSD (relative standard deviation) at location 3 A.

Metals		Location 1			Location 2			Location 3		Location 4			Mean	SD	T-test	Contam/ Refere	RSD
	Sites	A	B	C	A	B	C	A	C	A	B	C					
Cu	Reference	5.3	5.9	5.7	5.3	5.1	5.7	5.3	5.6	5.9	5.7	6.1	5.6	0.3	4.95E-11	1.9	5%
	Contaminated	9.9	11.6	11	0	0	0	0	0	0	0	0	10.8	0.9			8%
Cd	Reference	3.9	4	3.9	3.8	3.9	4.1	2.1	2.7	2.1	2.4	2.3	3.2	0.8	1.59E-08	2.8	26%
	Contaminated	8.9	8.8	8.9	0	0	0	0	0	0	0	0	8.9	0.1			1%
Pb	Reference	45.6	43.6	46.1	48.8	47.8	46.4	45.9	45.7	47.9	45.3	44.9	46.5	1.8	1.18E-11	1.5	4%
	Contaminated	68	71	70	0	0	0	0	0	0	0	0	69.7	1.5			2%
Ni	Reference	12.8	14.1	13.8	21.3	21.8	21.6	28.2	30.2	22.7	21.9	21.4	21.6	5.8	5.44E-02	1.3	27%
	Contaminated	26.3	27.2	29.3	0	0	0	0	0	0	0	0	27.6	1.5			6%
V	Reference	16.2	17.1	16.8	16.7	17.2	16.9	16.3	17.6	16.9	16.3	17.2	16.9	0.4	2.21E-11	1.3	3%
	Contaminated	21.9	22.8	22.3	0	0	0	0	0	0	0	0	22.3	0.5			2%
Fe	Reference	4.9	4.8	5.2	3.7	3.6	3.8	3.5	3.6	4.8	4.2	5.9	4.3	0.8	2.72E-08	2.2	18%
	Contaminated	9.4	9.3	9.9	0	0	0	0	0	0	0	0	9.5	0.3			3%
Zn	Reference	22.8	24	23.8	24.2	25.1	31.7	32.3	23.6	24.1	25.7	23.8	26.0	3.5	8.89E-03	1.2	13%
	Contaminated	31.7	32.3	30.9	0	0	0	0	0	0	0	0	31.6	0.7			2%
Cr	Reference	18.3	19.6	19.2	29.3	30	29.2	21.7	21.6	21.2	23.1	19.5	22.7	4.3	5.63E-11	3.1	19%
	Contaminated	71.8	69.9	68.7	0	0	0	0	0	0	0	0	70.1	1.6			2%
Hg	Reference	0.25	0.23	0.37	0.12	0.15	0.18	0.18	0.29	0.26	0.25	0.23	0.2	0.1	4.05E-18	24.2	31%
	Contaminated	5.3	5.2	5.7	0	0	0	0	0	0	0	0	5.4	0.3			5%
Mn	Reference	27.6	27.3	28.1	32.7	32.1	27.9	31.4	30.6	28.9	28.6	30.1	27.7	0.4	1.22E-13	2.4	1%
	Contaminated	64.9	66.5	64.1	0	0	0	0	0	0	0	0	65.2	1.2			2%

The Cd concentrations were 3-fold higher at contaminated site 3B, compared to the reference sites. The Hg concentration was higher by a factor of 27.1 than the reference site. The Cu, Cr, Mn, Ni, Zn, and iron concentrations were 2- fold higher than the reference site. The Pb and V concentrations were greater than the reference site by a factor of 1.6 and 1.4 respectively (Table 8).

Table 8. Metal Concentrations in Sediment at Location 3B

Metal concentrations in sediment (ppm), SD (standard deviation), T-test, contamination/ reference sites, RSD (relative standard deviation) at location 3B.

Metals		Location 1			Location 2			Location 3		Location 4			Mean	SD	T-test	Contam/ Refere	RSD
	Sites	A	B	C	A	B	C	A	C	A	B	C					
Cu	Reference	5.3	5.9	5.7	5.3	5.1	5.7	5.3	5.6	5.9	5.7	6.1	5.6	0.3	5.32E-11	2.0	5%
	Contaminated	9.9	11.6	11.2	0	0	0	0	0	0	0	0	10.9	0.9			8%
Cd	Reference	3.9	4	3.9	3.8	3.9	4.1	2.1	2.7	2.1	2.4	2.3	3.2	0.8	3.08E-09	3.1	26%
	Contaminated	9.9	9.8	9.5	0	0	0	0	0	0	0	0	9.7	0.2			2%
Pb	Reference	45.6	43.6	46.1	48.8	47.8	46.4	45.9	45.7	47.9	45.3	44.9	46.5	1.8	1.78E-12	1.6	4%
	Contaminated	72.5	75	76.6	0	0	0	0	0	0	0	0	74.7	2.1			3%
Ni	Reference	12.8	14.1	13.8	21.3	21.8	21.6	28.2	30.2	22.7	21.9	21.4	21.6	5.8	6.88E-06	2.1	27%
	Contaminated	44.3	44.8	45.9	0	0	0	0	0	0	0	0	45.0	0.8			2%
V	Reference	16.2	17.1	16.8	16.7	17.2	16.9	16.3	17.6	16.9	16.3	17.2	16.9	0.4	3.56E-13	1.4	3%
	Contaminated	24.1	23.9	24.6	0	0	0	0	0	0	0	0	24.2	0.4			1%
Fe	Reference	4.9	4.8	5.2	3.7	3.6	3.8	3.5	3.6	4.8	4.2	5.9	4.3	0.8	2.02E-09	2.5	18%
	Contaminated	10.9	10.5	10.8	0	0	0	0	0	0	0	0	10.7	0.2			2%
Zn	Reference	22.8	24	23.8	24.2	25.1	31.7	32.3	23.6	24.1	25.7	23.8	26.0	3.5	9.06E-11	2.4	13%
	Contaminated	63	62	62.9	0	0	0	0	0	0	0	0	62.6	0.6			1%
Cr	Reference	21.3	19.6	19.2	29.3	30	29.2	21.7	21.6	21.2	23.1	19.5	23.0	4.1	2.41E-07	2.0	18%
	Contaminated	46	45.2	44.9	0	0	0	0	0	0	0	0	45.4	0.6			1%
Hg	Reference	0.25	0.23	0.37	0.12	0.15	0.18	0.18	0.29	0.26	0.25	0.28	0.2	0.1	3.04E-18	27.1	31%
	Contaminated	6.1	6.5	5.9	0	0	0	0	0	0	0	0	6.2	0.3			5%
Mn	Reference	27.6	27.3	28.1	32.7	32.1	27.9	31.4	30.6	28.9	28.6	30.1	27.7	0.4	1.40E-08	2.3	1%
	Contaminated	54.9	63.2	76.6	0	0	0	0	0	0	0	0	64.9	10.9			17%

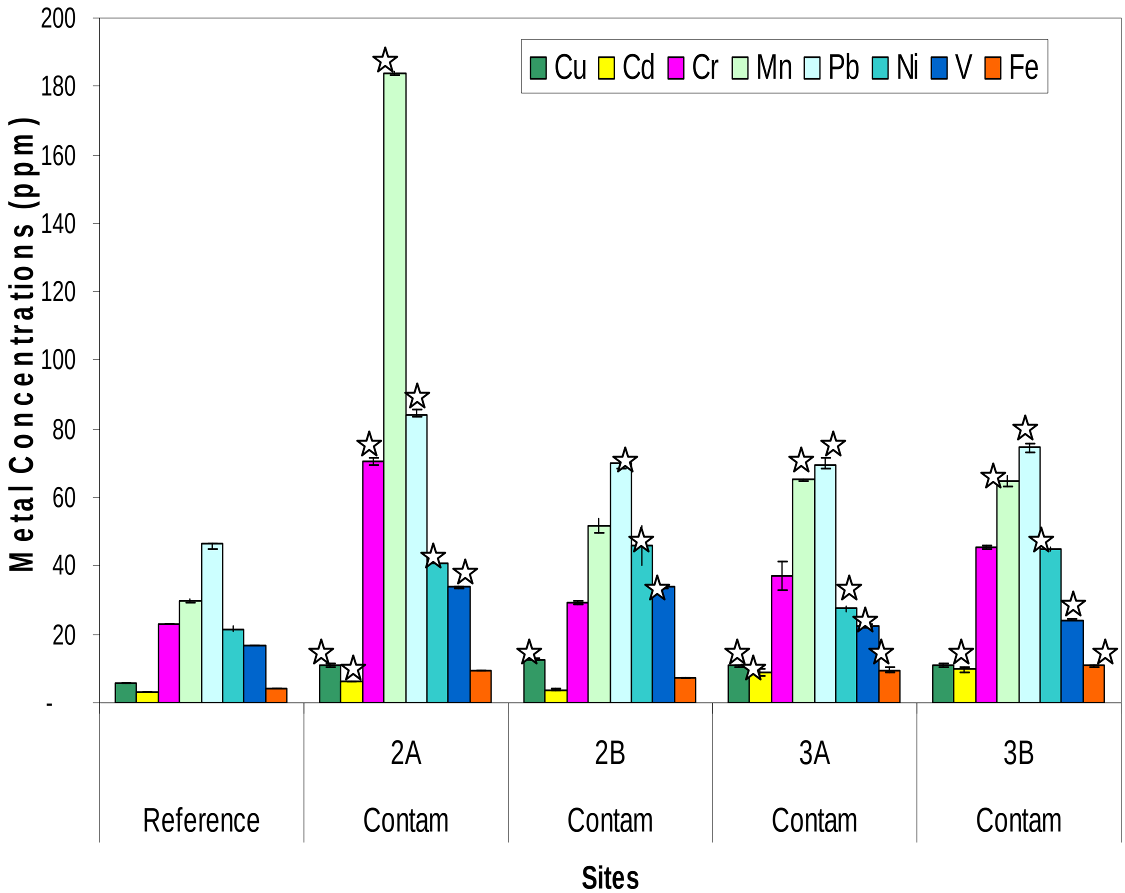

Figure 6. The average metal concentrations (ppm) in sediment at the contaminated and the reference site. The symbol ☆ represents a significant difference of metal concentrations at the contaminated site compared to the reference sites.

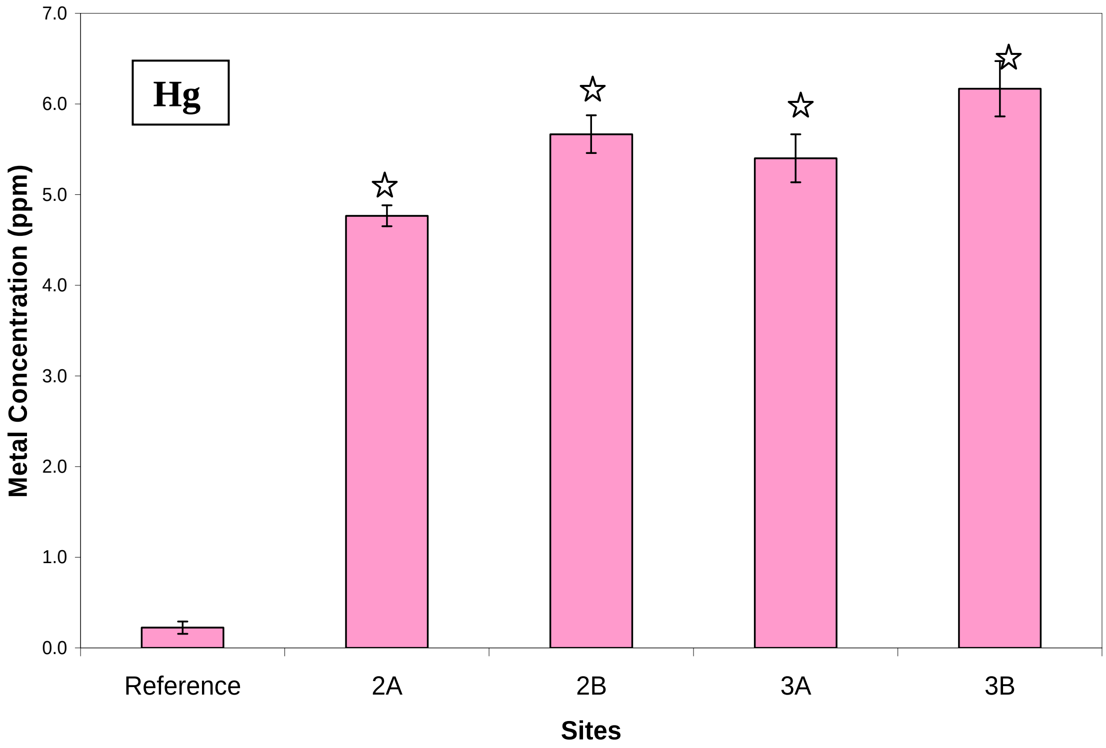

Figure 7. The average Hg concentration in sediment (ppm) at the contaminated and the reference site.

The symbol☆represents a significant difference of metal concentrations at contaminated site compared to the reference site. Error bars show SD.

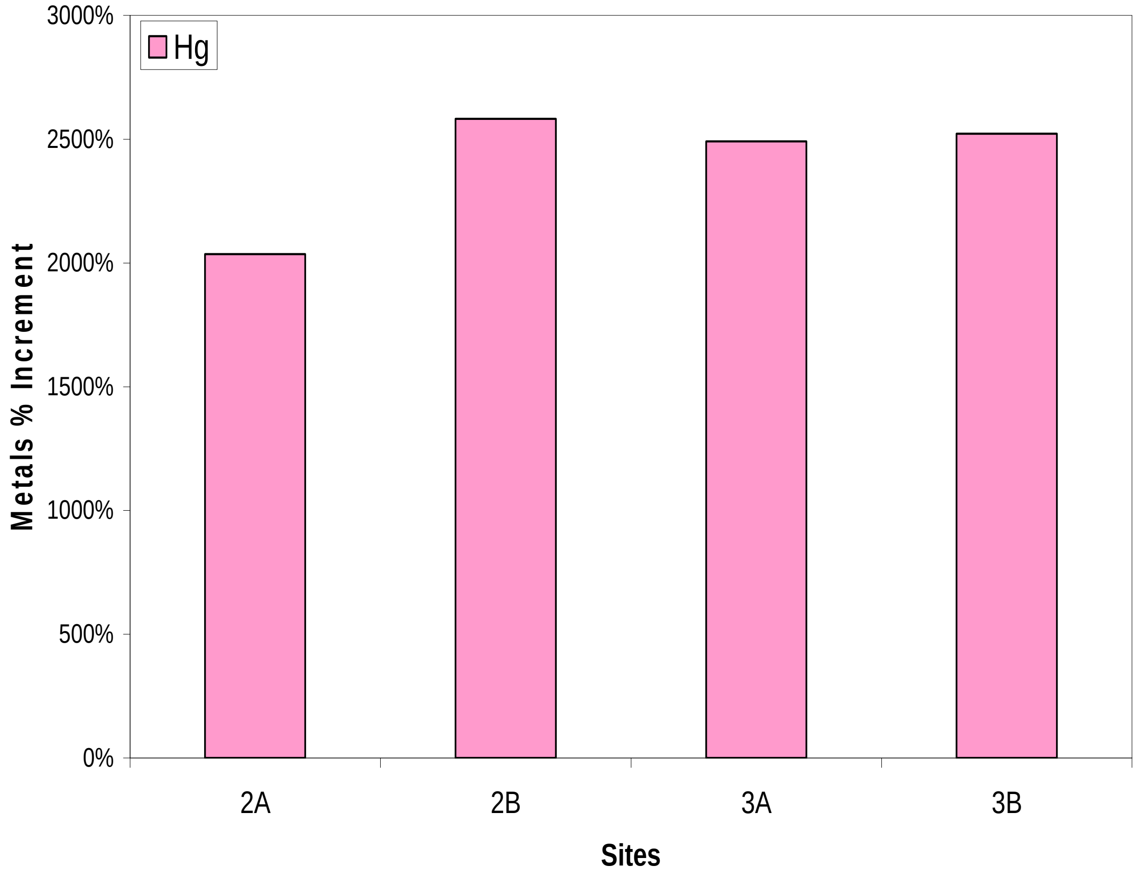

Figure 8. Relative enrichment (%) of Hg in the sediment of the contaminated sites with respect to the reference site.

2.6. Spatial differences in metal concentrations among clam

The results suggest that all sites (2A, 2B. 3A, 3B) close to the desalination and power plants were more contaminated compared to the reference sites (Figure 5). There were variations in metal concentrations in all locations adjacent to the desalination/power plant. The Hg, Cd, Cr, and Cu are significantly higher($p<0.05$) in all four stations near the desalination/power plant compared to the reference sites.

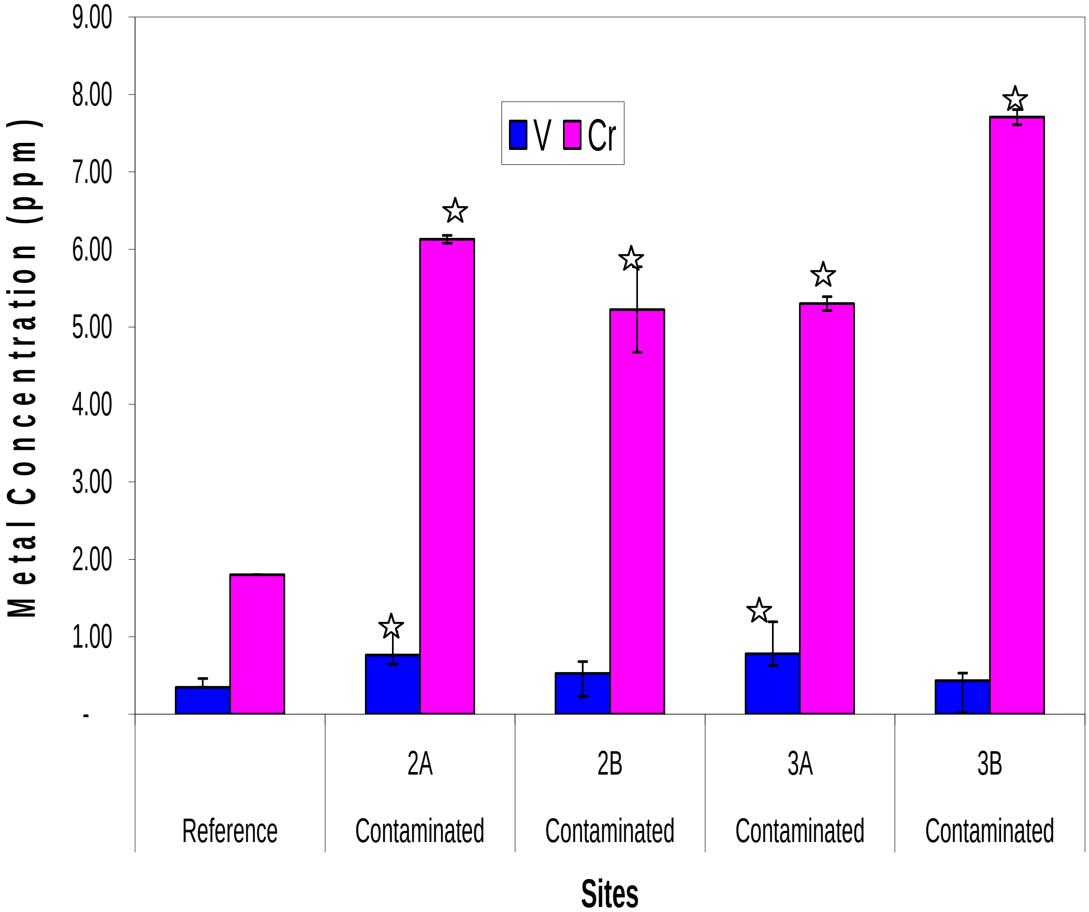

Figure 9. The average V and Cr concentrations in soft tissues (ppm) at the contaminated and the reference Site. Error bars show SD.

The symbol☆represents a significant difference between contaminated and the reference site. 100 clams were collected from each station and divided into three groups of 33 each (n=3). Each composite sample consists of 33 clams.

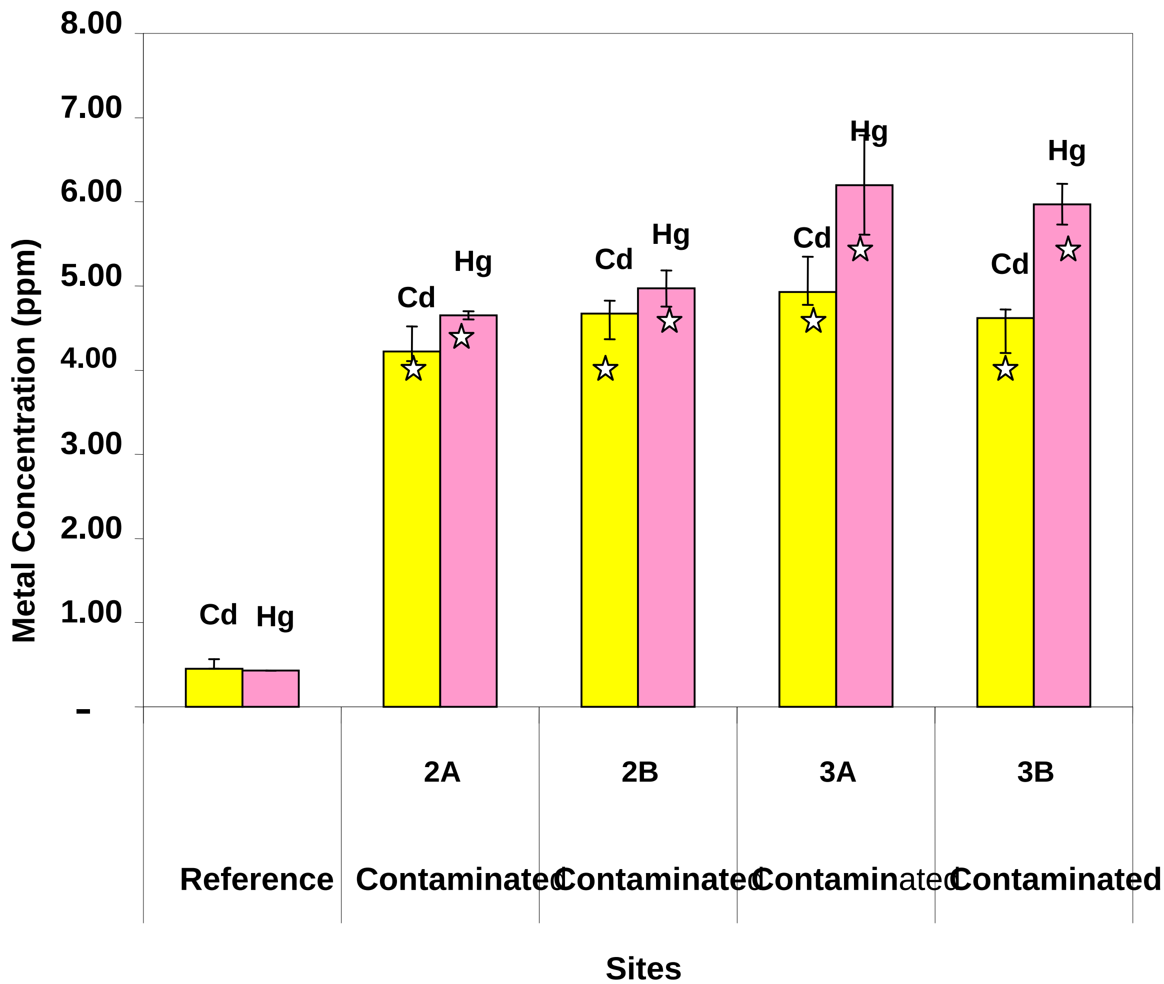

Figure 10. The average Cd and Hg concentrations in soft tissues (ppm) at the contaminated and the reference site.

The symbol☆represents a significant difference between contaminated and the reference sites. Error bars show SD.

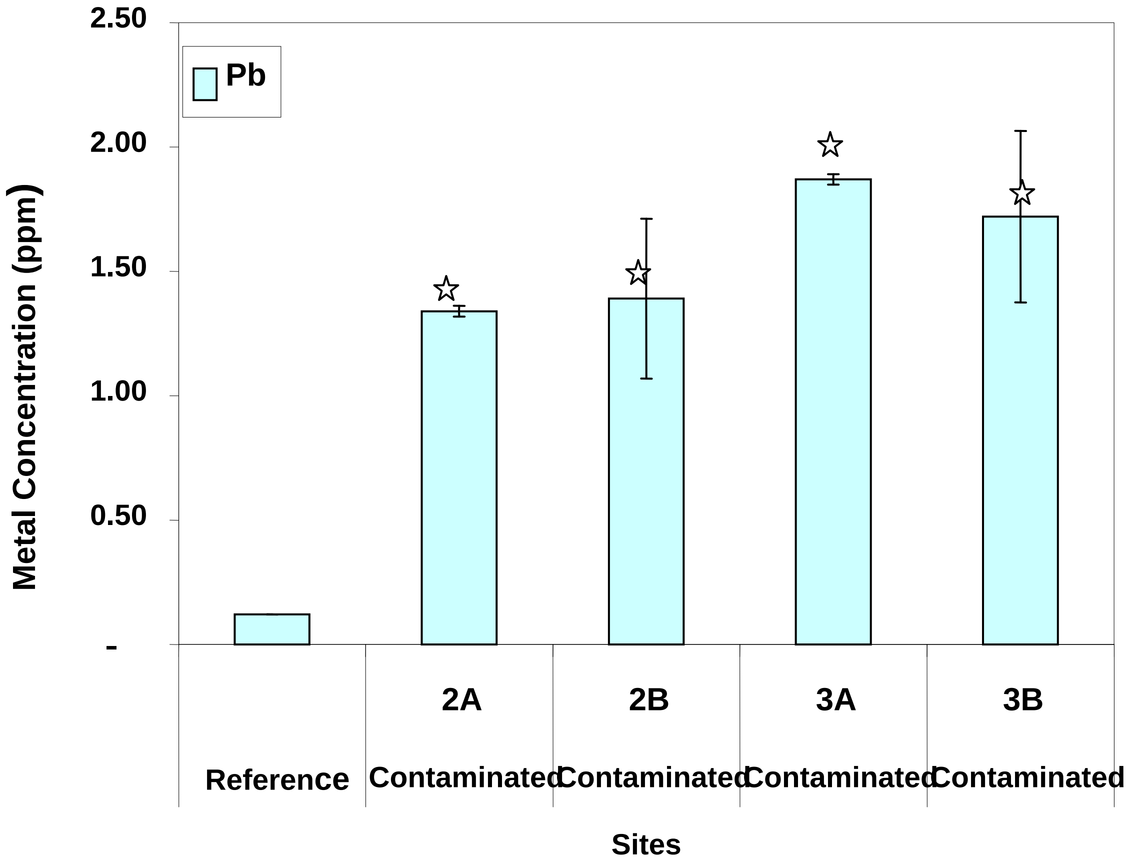

Figure 11. The average Pb concentrations in soft tissues of clam *Amiantis umbonella* (ppm) at the contaminated and the reference Site.

The symbol ☆ represents a significant difference between the contaminated and the reference sites. Error bars show SD.

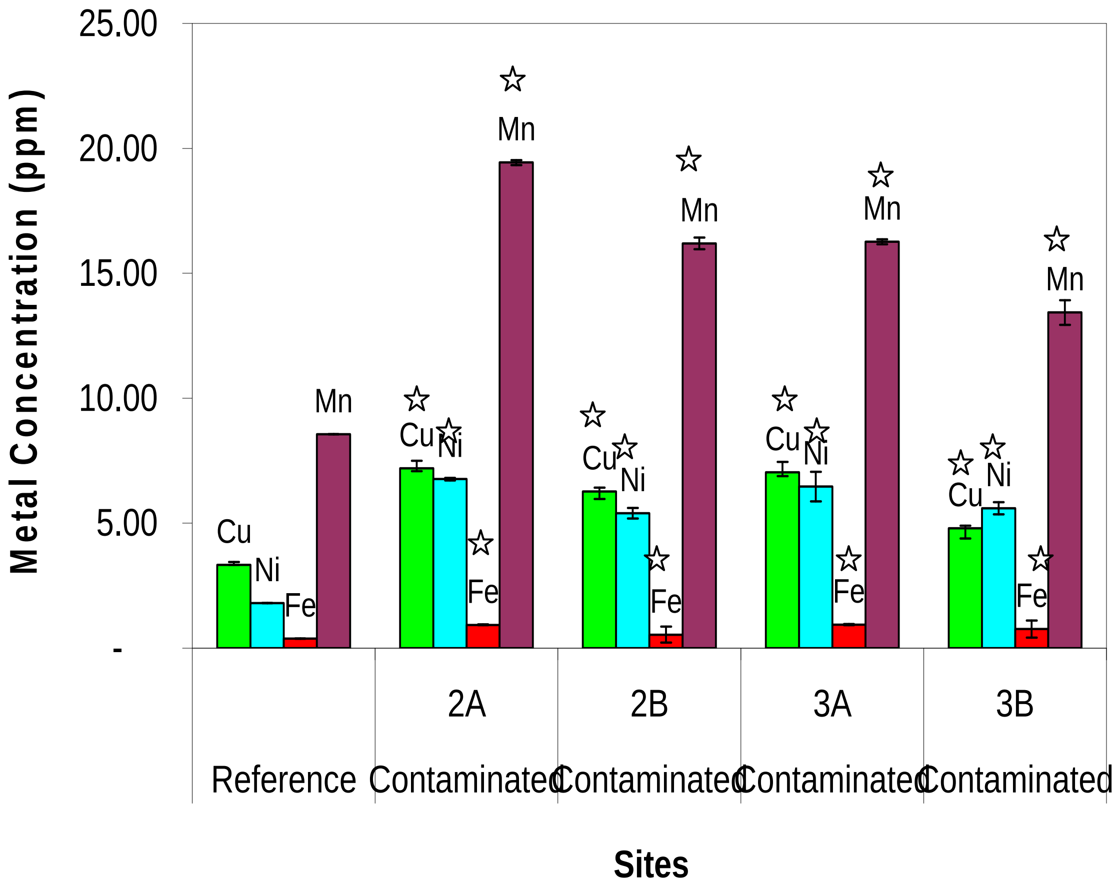

Figure 12. The average Cu, Ni, Fe and Mn concentrations in soft tissues of clam *A. umbonella* (ppm) at the contaminated and the reference Site.

The symbol☆represents a significant difference between the contaminated and the reference site.

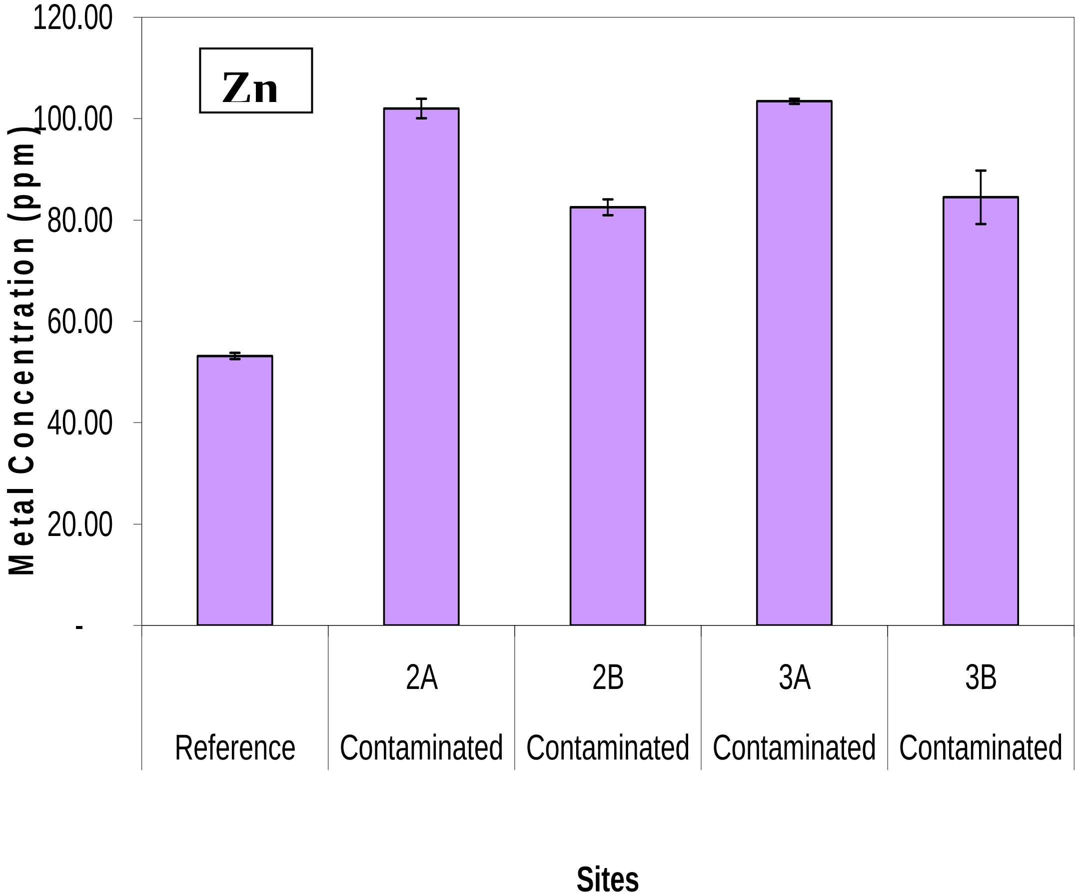

Figure 13. The average Zn concentration in soft tissues of clam *A. umbonella* (ppm) at the contaminated and the reference Site.

The observed mean metal concentrations in clams collected from all four contaminated and the reference site with mean, SD, RSD (% relative standard deviation) T-test, and percent (%) more than the reference site are shown in Table 9, 10, 11, 12. Bar graphs (Figure. 5, 6, 7, 8) show the average metal concentrations of Cu, Cd, Cr, Hg, Mn, Pb, Ni, V, Zn, Fe from four sampling locations (contaminated vs. reference site).

Table 9. Metal Concentrations in Soft Tissue at Location 2A

Metal concentrations (ppm) in soft tissues of clam A. umbonella, Mean, SD (standard deviation), T-test, contaminated/reference sites, percent (%)increase at the contaminated sites compared to the reference sites, RSD (relative standard deviation) at location 2A.

Metals		Location			Mean	SD	T-test	Contam/ Refere	% Increase in Contam	RSD
	Sites	A	B	C						
Cu	Reference	3.2	3.4	3.4	3.3	0.1	1.57E-05	2.2	103%	3%
	Contaminated	7.2	7.5	6.9	7.2	0.3				4%
Cd	Reference	0.45	0.4	0.5	0.5	0.1	2.04E-06	10.8	896%	11%
	Contaminated	4.98	4.61	4.98	4.9	0.2				4%
Pb	Reference	0.25	0.29	0.28	0.3	0.0	6.81E-04	6.4	657%	8%
	Contaminated	1.54	1.59	2.12	1.8	0.3				18%
Ni	Reference	1.8	1.7	1.9	1.8	0.1	2.18E-06	3.8	263%	6%
	Contaminated	6.9	6.5	6.9	6.8	0.2				3%
V	Reference	0.37	0.29	0.38	0.3	0.0	1.20E-04	2.2	103%	14%
	Contaminated	0.73	0.79	0.77	0.8	0.0				4%
Fe	Reference	0.34	0.45	0.37	0.4	0.1	2.52E-04	2.4	162%	15%
	Contaminated	0.98	0.85	0.97	0.9	0.1				8%
Zn	Reference	53.9	52.9	52.9	53.2	0.6	9.73E-07	1.9	95%	1%
	Contaminated	103.1	99.8	103.2	102.0	1.9				2%
Cr	Reference	1.8	1.9	1.7	1.8	0.1	9.66E-08	3.4	261%	6%
	Contaminated	6.12	6.13	6.14	6.1	0.0				0%
Hg	Reference	0.1	0.09	0.08	0.1	0.0	4.24E-06	65.6	6438%	11%
	Contaminated	4.65	4.98	5.23	5.9	0.3				5%
Mn	Reference	8.1	8.3	7.9	8.1	0.2	2.37E-06	2.4	149%	2%
	Contaminated	18.8	19.8	19.7	19.4	0.6				3%

The Hg concentration was a factor of 32.1 higher than the reference site. The Cd concentration was 4- fold higher in clams collected at the station 2A contaminated location compared to the reference sites. The Ni and Cr were a factor of 3.8 and 3.4 higher than the clams from the reference sites. The V and Cu concentrations were 2-fold higher than the reference sites. The Fe concentration was a factor of 2.4 higher than the clams from the reference sites. The Pb, Mn and Zn concentrations were greater than the reference sites by a factor of 1.8, 2.4, and 1.9 respectively (Table 9).

The Ni and Cr concentrations in the clam tissues at the station 2 B were 3 – fold higher than the reference sites. The Hg concentration was a factor of 28.1.6 higher than the reference sites. The Cd concentration was a factor of 2.9 higher than the reference sites. The Cu concentration was greater than the reference site by a factor of 2.6. The Pb, and V concentrations were a factor of 2.1 and 2.3 higher than the reference sites respectively. The concentrations of Mn, Zn and Fe in clams at the 2B contaminated sites were a factor of 2, 2, and 1.5 higher compared to the reference site (Table 10).

Table 10. Metal Concentrations in Soft Tissues at Location 2B

Metal concentrations (ppm) in soft tissues of clam *A. umbonella*, Mean, SD (standard deviation), T-test, contaminated/reference, percent (%) increase at contaminated sites compared to the reference sites, RSD (relative standard deviation) at location 2B.

Metals		Location 1			Mean	SD	T-test	Contam/ Referen	% Increase in Contam	RSD
	Sites	A	B	C						
Cu	Reference	2.2	2.5	2.4	2.4	0.2	3.12E-06	2.6	163%	6%
	Contaminated	6.4	6.1	6.3	6.3	0.2				2%
Cd	Reference	0.3	0.2	0.3	0.3	0.1	1.31E-04	16.7	1537%	22%
	Contaminated	3.78	4.65	4.91	4.4	0.6				13%
Pb	Reference	0.17	0.12	0.21	0.2	0.0	1.64E-03	2.1	57%	27%
	Contaminated	0.37	0.34	0.33	0.3	0.0				6%
Ni	Reference	1.9	1.7	1.8	1.8	0.1	7.91E-07	3.0	194%	6%
	Contaminated	5.5	5.4	5.3	5.4	0.1				2%
V	Reference	0.23	0.21	0.25	0.2	0.0	7.99E-06	2.3	112%	9%
	Contaminated	0.53	0.52	0.53	0.5	0.0				1%
Fe	Reference	0.34	0.33	0.37	0.3	0.0	4.40E-04	1.5	54%	6%
	Contaminated	0.51	0.53	0.57	0.5	0.0				6%
Zn	Reference	42.2	3.9.7	41.6	41.9	0.4	2.74E-05	2.0	100%	1%
	Contaminated	83.5	80.7	83.3	82.5	1.6				2%
Cr	Reference	1.8	1.9	1.7	1.8	0.1	9.70E-07	2.9	201%	6%
	Contaminated	5.23	5.32	5.12	5.2	0.1				2%
Hg	Reference	0.1	0.09	0.08	0.1	0.0	1.86E-05	63.1	7288%	11%
	Contaminated	4.98	5.47	5.91	5.7	0.5				8%
Mn	Reference	8.1	8.3	7.9	8.1	0.2	5.45E-03	2.0	82%	2%
	Contaminated	14.4	19.8	14.4	16.2	3.1				19%

Table 11. Metal Concentrations in Soft Tissues at Location 3A

Metal concentrations (ppm) in soft tissues of clam *A. umbonella*, Mean, SD (standard deviation), T-test, contaminated/reference, percent (%) increase at the contaminated sites compared to the reference sites, RSD (relative standard deviation) at location 3A.

Metals		Location			Mean	SD	T-test	Contam/ Refere	% Increase in Contam	RSD
	Sites	A	B	C						
Cu	Reference	1.9	1.8	1.5	1.7	0.2	1.95E-05	4.1	360%	12%
	Contaminated	6.7	7.5	6.9	7.0	0.4				6%
Cd	Reference	0.24	0.25	0.29	0.3	0.0	2.92E-06	18.1	1455%	10%
	Contaminated	4.65	4.98	4.51	4.7	0.2				5%
Pb	Reference	0.2	0.1	0.1	0.1	0.1	4.14E-04	14.7	2030%	43%
	Contaminated	1.56	2.18	2.13	2.0	0.3				18%
Ni	Reference	2.5	2.4	2.4	2.4	0.1	7.41E-05	2.7	179%	2%
	Contaminated	5.9	6.8	6.7	6.5	0.5				8%
V	Reference	0.2	0.3	0.2	0.2	0.1	4.29E-05	3.3	290%	25%
	Contaminated	0.77	0.79	0.78	0.8	0.0				1%
Fe	Reference	0.31	0.33	0.45	0.4	0.1	1.78E-04	2.6	98%	21%
	Contaminated	0.98	0.96	0.89	0.9	0.0				5%
Zn	Reference	42.2	39.9	38.8	40.3	1.7	2.22E-07	2.6	166%	4%
	Contaminated	103.1	104	103.2	103.4	0.5				0%
Cr	Reference	1.8	1.9	1.7	1.8	0.1	5.50E-06	2.9	212%	6%
	Contaminated	5.5	5.1	5.3	5.3	0.2				4%
Hg	Reference	0.1	0.09	0.08	0.1	0.0	1.14E-03	67.5	9525%	11%
	Contaminated	5.76	4.76	7.7	6.1	1.5				25%
Mn	Reference	8.1	7.9	7.7	7.9	0.2	4.57E-03	2.1	88%	3%
	Contaminated	19.8	14.5	14.5	16.3	3.1				19%

Table 12. Metal Concentrations in Soft Tissues at Location 3B

Metal concentrations (ppm) in soft tissues of clam *A. umbonella*, Mean, SD (standard deviation), T-test, contaminated/reference, percent (%) increase at contaminated sites compared to the reference sites, RSD (relative standard deviation) at location 3B.

Metals		Location			Mean	SD	T-test	Contam/ Refere	% Increase in Contam	RSD
	Sites	A	B	C						
Cu	Reference	2.8	2.9	2.1	2.6	0.4	5.20E-04	1.8	124%	17%
	Contaminated	4.8	4.9	4.7	4.8	0.1				2%
Cd	Reference	0.26	0.32	0.25	0.3	0.0	7.50E-06	16.7	1812%	14%
	Contaminated	4.81	4.28	4.78	4.6	0.3				6%
Pb	Reference	0.06	0.05	0.03	0.0	0.0	7.02E-05	38.5	6000%	33%
	Contaminated	1.99	1.57	1.83	1.8	0.2				12%
Ni	Reference	1.7	1.9	1.8	1.8	0.1	3.07E-05	3.1	217%	6%
	Contaminated	5.9	5.2	5.7	5.6	0.4				6%
V	Reference	0.2	0.3	0.2	0.2	0.1	6.62E-03	1.9	100%	25%
	Contaminated	0.4	0.5	0.4	0.4	0.1				13%
Fe	Reference	0.31	0.33	0.45	0.4	0.1	1.77E-03	2.1	76%	21%
	Contaminated	0.67	0.83	0.79	0.8	0.1				11%
Zn	Reference	45.7	52.7	42.5	47.0	5.2	4.69E-04	1.8	86%	11%
	Contaminated	89.4	85.2	78.9	84.5	5.3				6%
Cr	Reference	1.8	1.9	1.7	1.8	0.1	4.15E-06	4.3	359%	6%
	Contaminated	7.33	7.98	7.81	7.7	0.3				4%
Hg	Reference	0.1	0.09	0.08	0.1	0.0	3.06E-06	70.3	8300%	11%
	Contaminated	6.12	6.13	6.72	6.3	0.3				5%
Mn	Reference	7.8	7.4	8.5	7.9	0.6	7.41E-04	1.7	71%	7%
	Contaminated	12.3	13.5	14.5	13.4	1.1				8%

The Cd concentration in clams collected at the station 3A ¼ km far from the outlet of the power plant was 10 – fold higher than the reference site. The Hg concentration in clam soft tissues was by a factor of 55.9 higher than the reference sites. The Cu concentration was 4 – fold higher at the contaminated sites than the reference sites. The V concentration in clams collected at Station 3 A was 3 – fold higher than the reference sites. The Cr, Mn, Zn concentrations were a factor of 2.9, 2.1, and 2.6 greater compared to the reference sites (Table 11).

The Cd concentration was 7 – fold higher than the reference sites in clams collected from station 3B near the outlet of power plant. The Hg concentration was higher than the reference site by a factor of 46.7. The Ni and Cr concentrations were 3-4 folds higher than the reference sites. The Pb concentration was greater than the reference sites by a factor of 5.8. The Mn, Cu, V, Zn, and iron concentration in clams were a factor of 1.7, 1.8, 1.9, 1.1, and1.5 higher compared to the reference sites respectively (Table 12).

2.7. Metal Interactions and Relationship among metals

There were several trace metal interactions were recorded. Metal interactions and inter-metal correlation among clams (n=32) sediment and water were determined.. There was a strong positive correlation between Hg concentration in soft tissues of clam *A. umbonella* and concentration in sediment and water (Figure 14, 15). There was a weak correlation of Hg concentration between sediment and water (Figure 15). There was also positive correlation ($p<0.05$) between Ni concentration in clam soft tissues and concentrations in sediment and water (Figure 17, 18). There was also a positive correlation of Ni concentrations between sediment and water. (Figure 19).

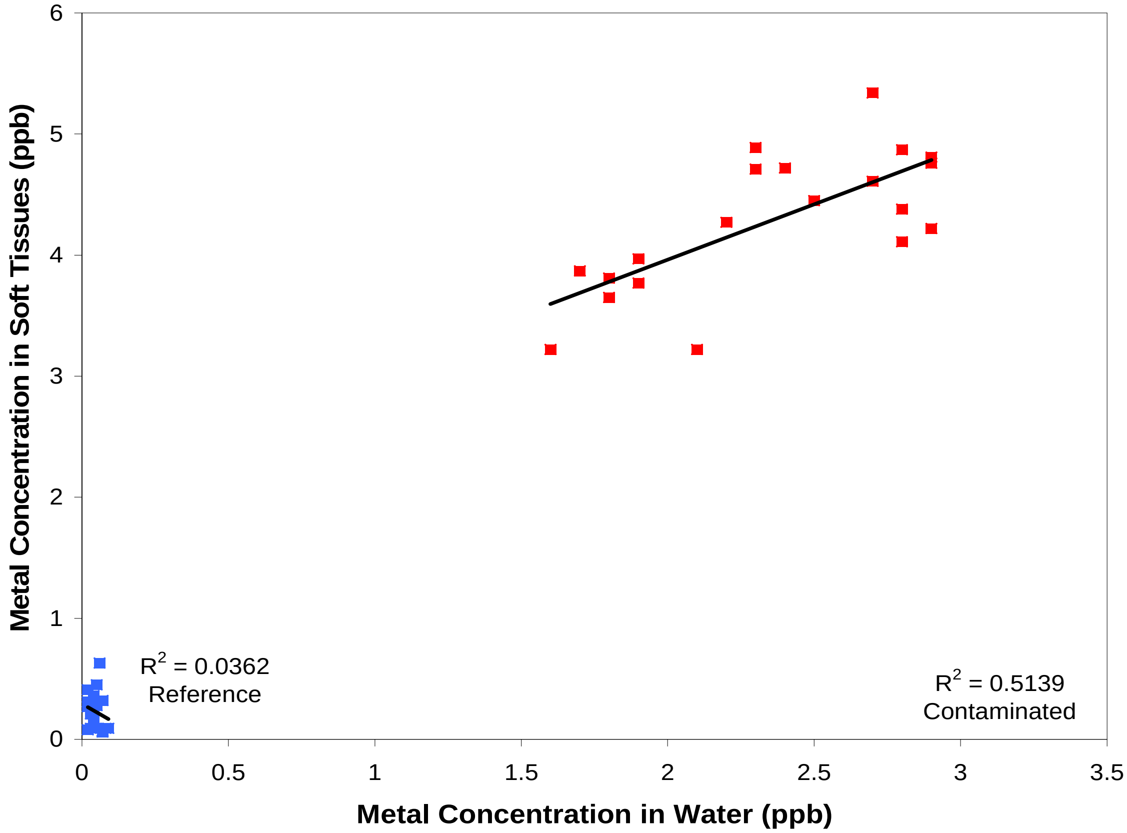

Figure 14. **Correlation of the Hg concentration in soft tissues (ppm) of clam *A. umbonella* and water (ppb) at the contaminated and the reference sites.**

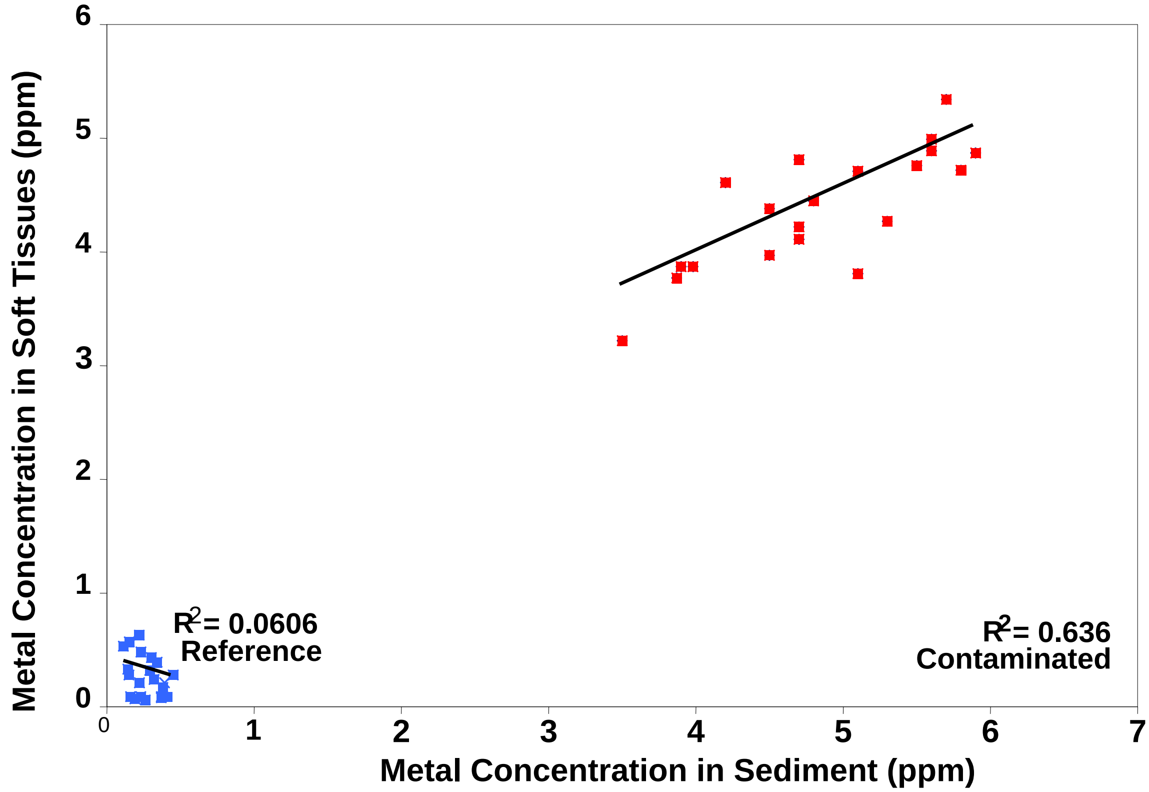

Figure 15. Correlation of the concentration of Hg in soft tissue of clam *A. umbonella* (ppm) and sediment (ppm) at the contaminated site (p=0.0418)) and the reference site.

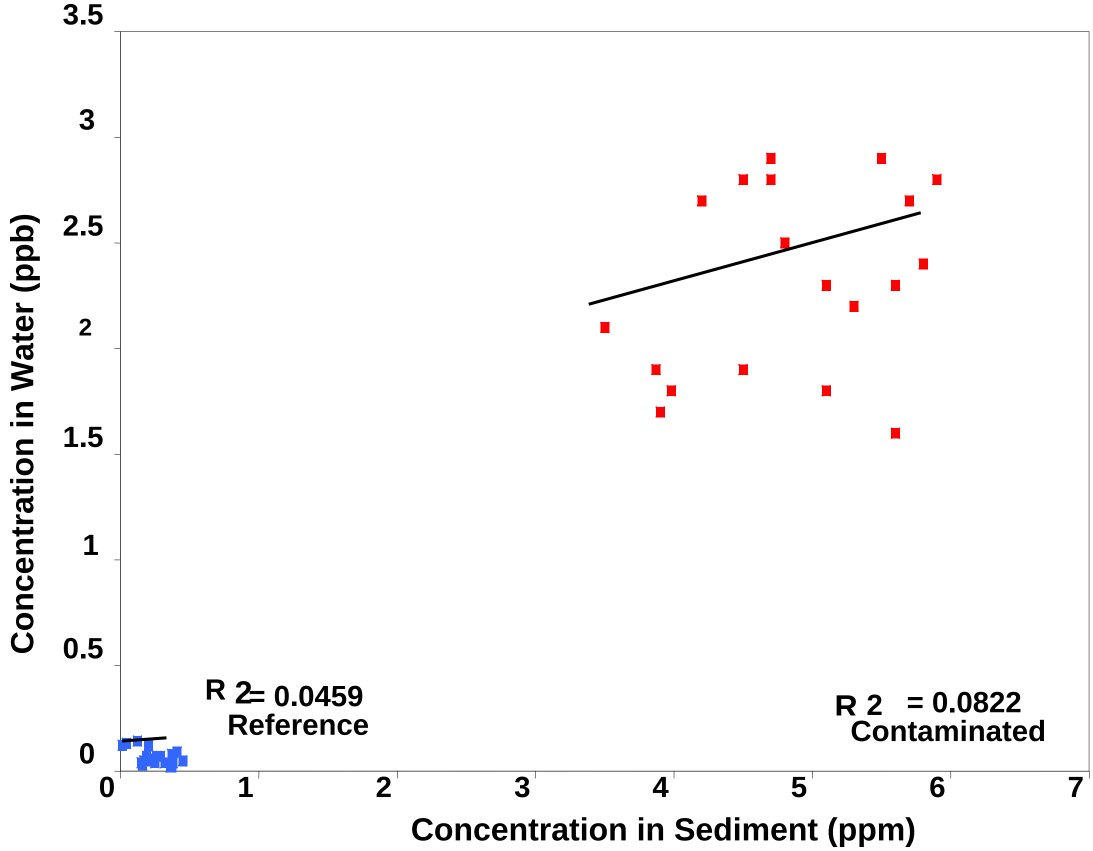

Figure 16. Correlation of the concentration of Hg in sediment (ppm) and water (ppb) at the contaminated (p=0.208) and the reference sites.

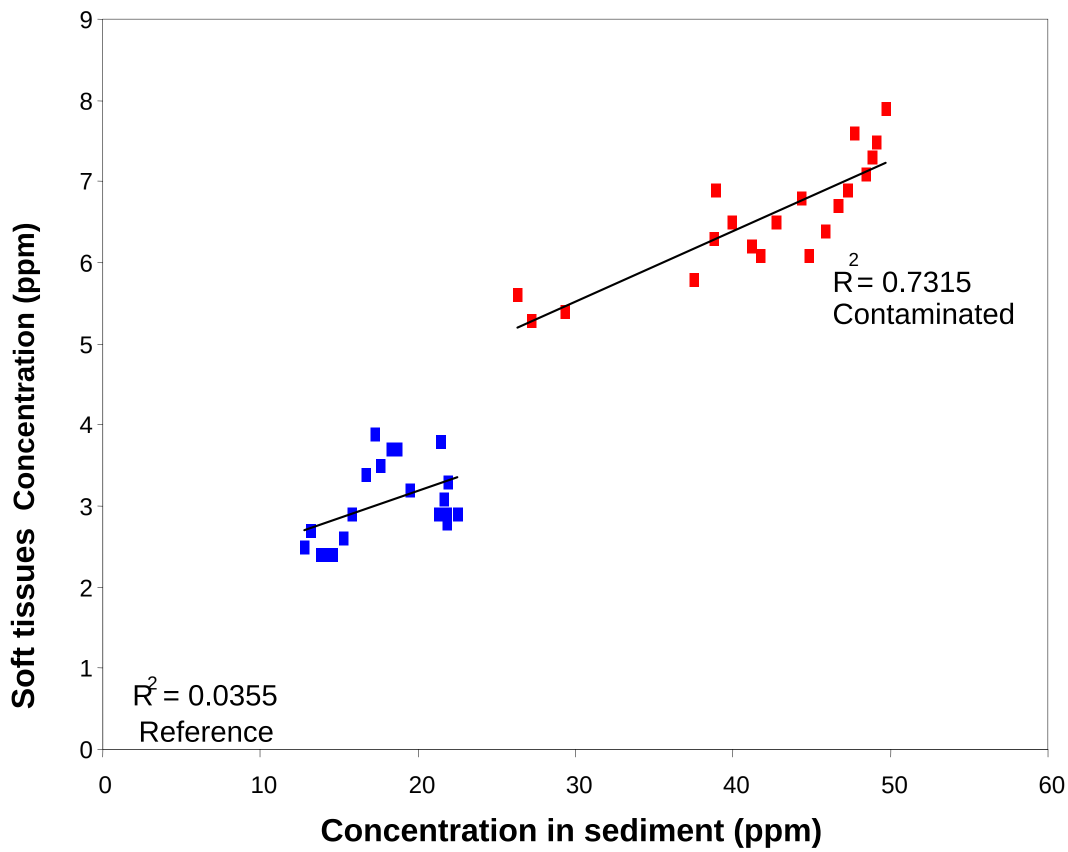

Figure 17. Correlation of the concentration of Ni in soft tissues of clam *A. umbonella* and sediment (ppm) at the contaminated (p=6.39E-08) and the reference sites.

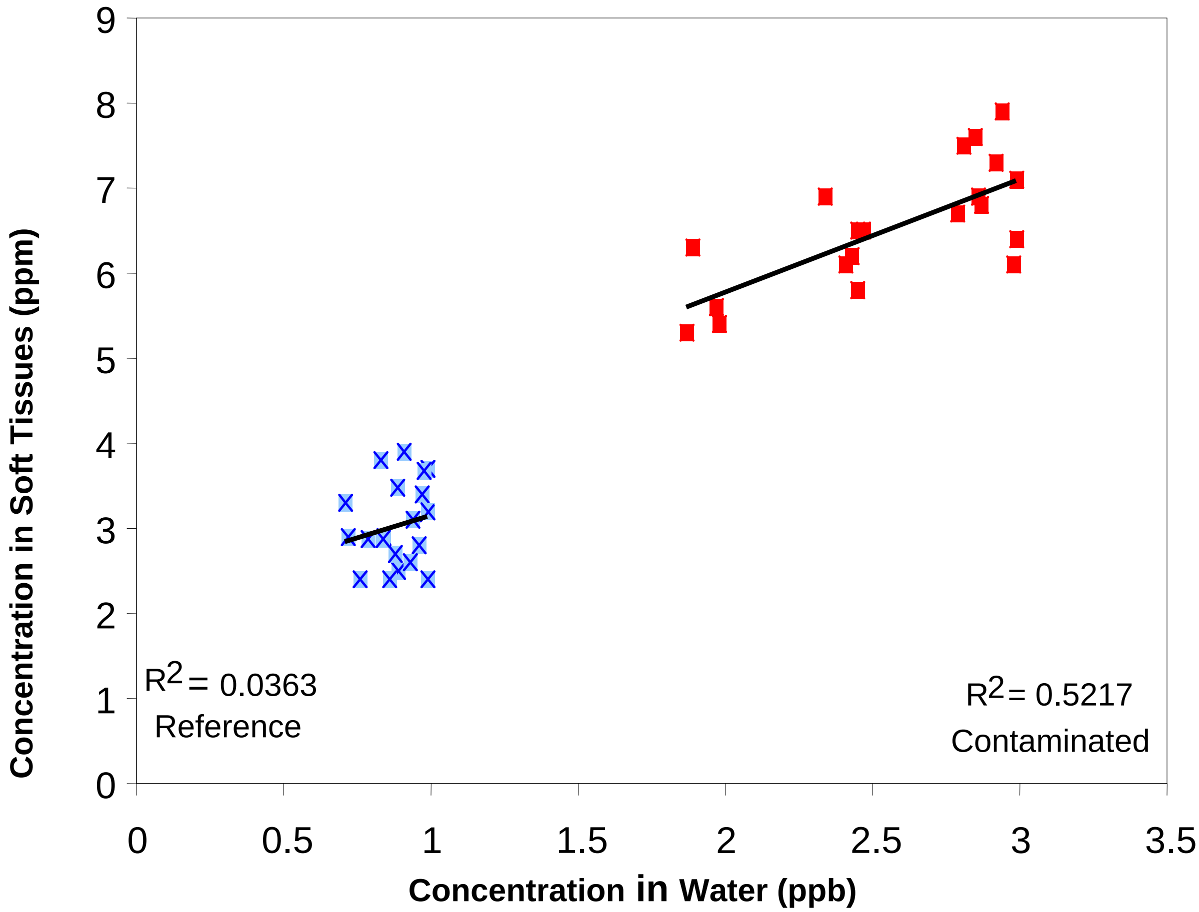

Figure 18. Correlation of the concentration of Ni in soft tissues of clam *A. umbonella* (ppm) and water (ppb) at the contaminated (p=0.0003) and the reference sites.

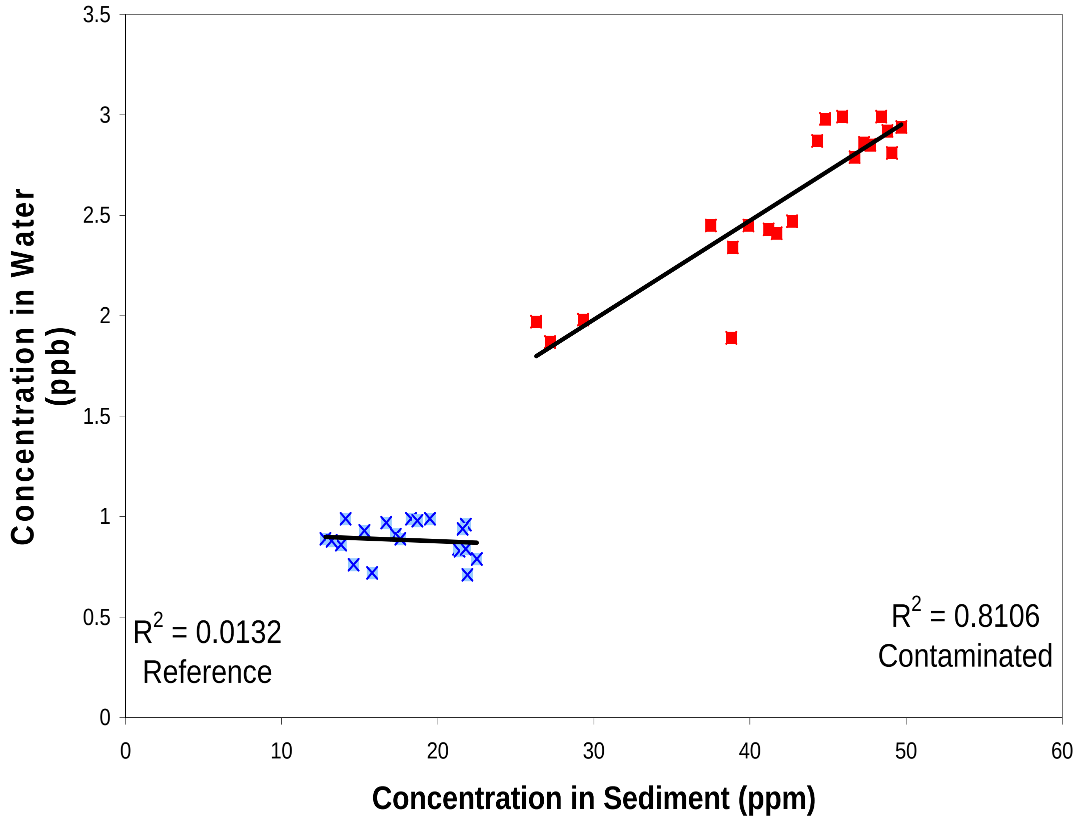

Figure 19. Correlation of the concentration of Ni in water (ppb) and sediment (ppm) at the contaminated (p=1.55E-06) and the reference sites.

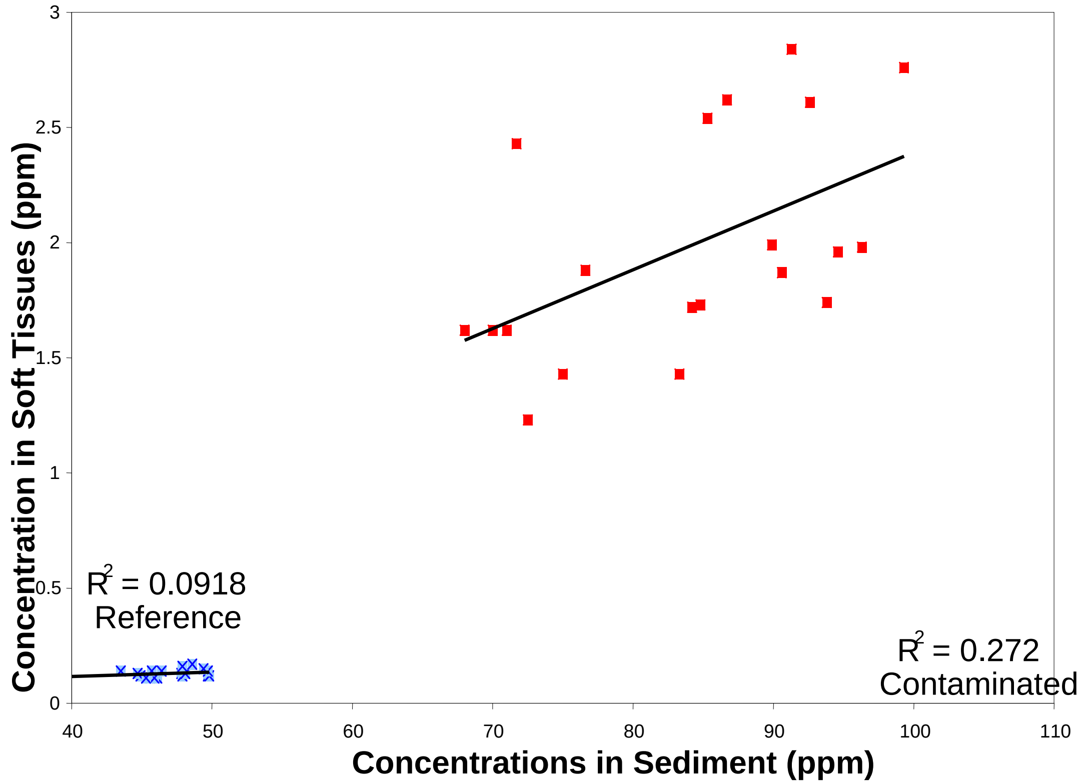

Figure 20. Correlation of the Pb concentration (ppm) in soft tissues of clam *A. umbonella* and sediment at the contaminated (p=0.0185) and the reference sites.

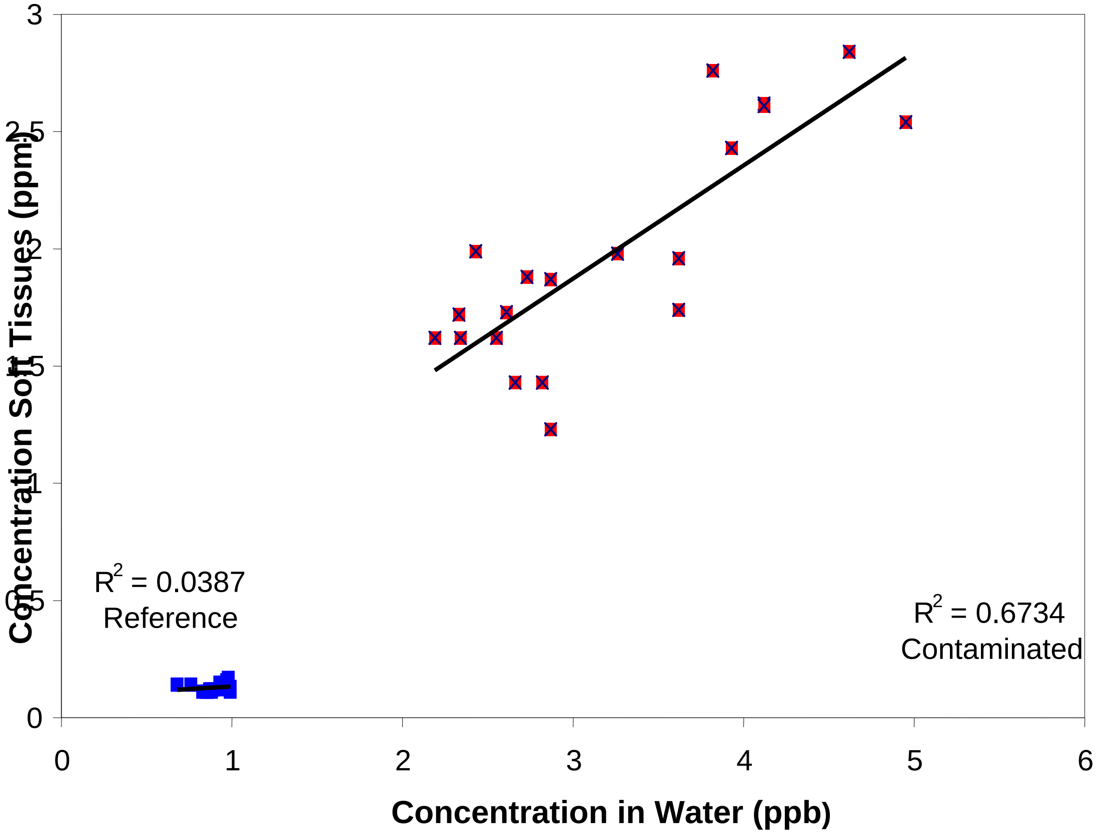

Figure 21. Correlation of the Pb concentration in soft tissues (ppm) of clam *A. umbonella* and water (ppb) at the contaminated (p=9.33E-06) and the reference sites.

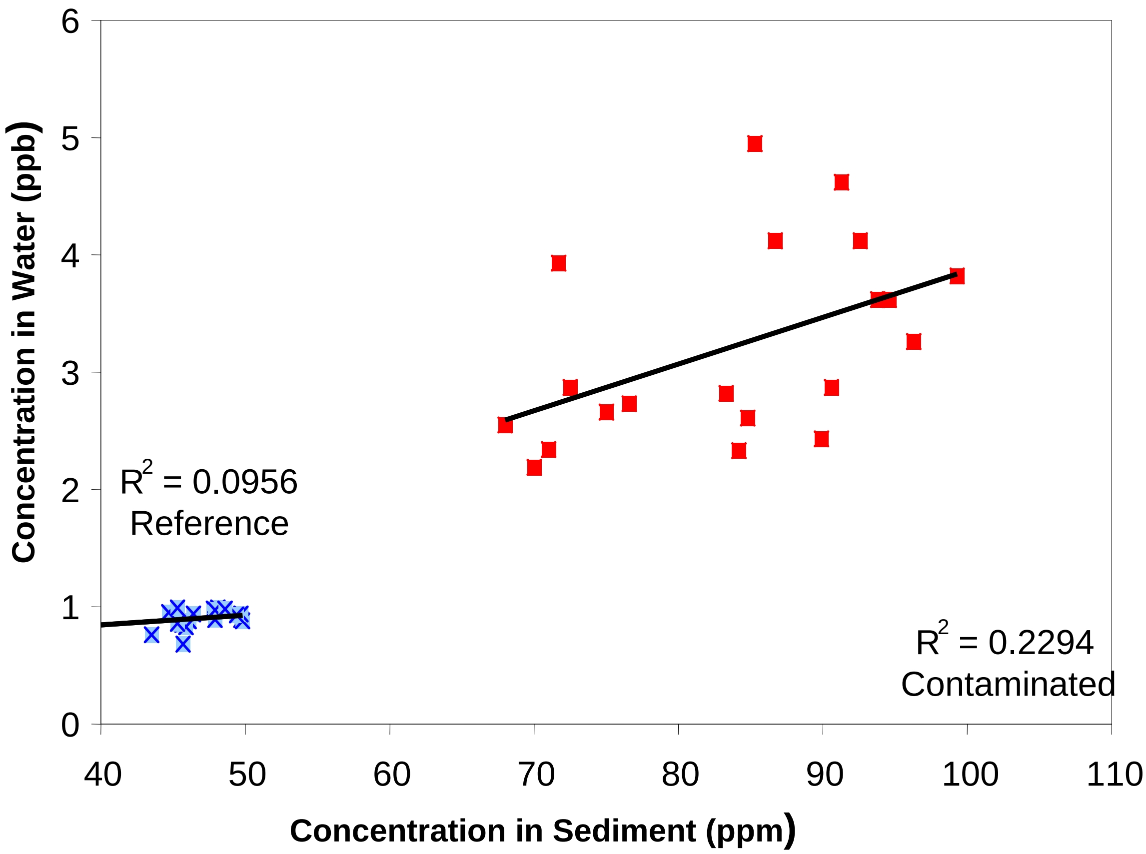

Figure 22. Correlation of the Pb concentration in water (ppb) and sediment (ppm) at the contaminated (p=0.033) and the reference sites.

There was a strong correlation of the Pb concentration (Figure 21, R^2= 0.67) between clams soft tissues and water and weak correlation between clams soft tissues and sediment (Figure 20, R^2=0.27). There was also a positive correlation of the Pb concentration ($p<0.05$) between water and sediment (Figure 22, R^2= 0.22).

There was a positive correlation of the Mn concentrations in clams soft tissues and water (Figure 24), and also a strong positive ($p<0.01$) correlation of the Mn concentrations between sediment and water (Figure 23, R^2= 0.48).

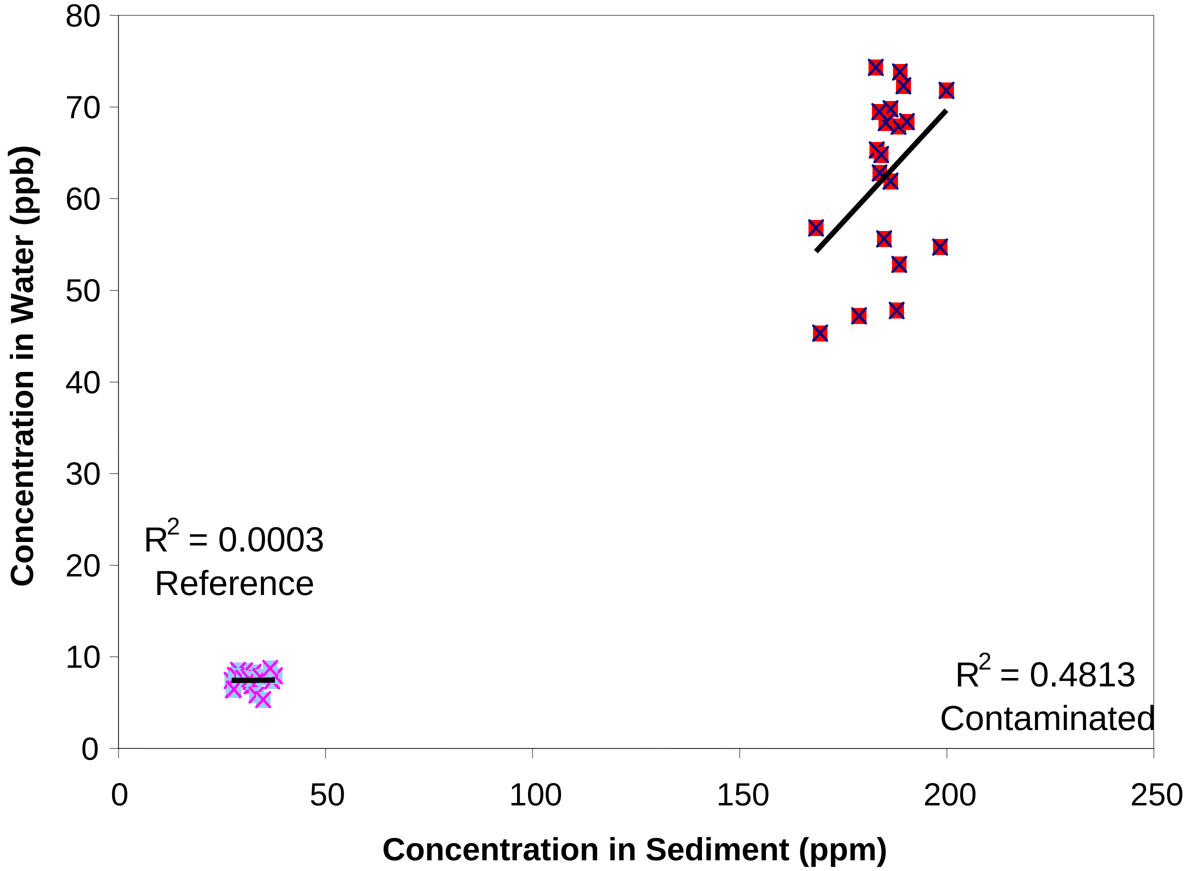

Figure 23. Correlation of the Mn concentration in water and sediment at the contaminated (p=0.086) and the reference sites.

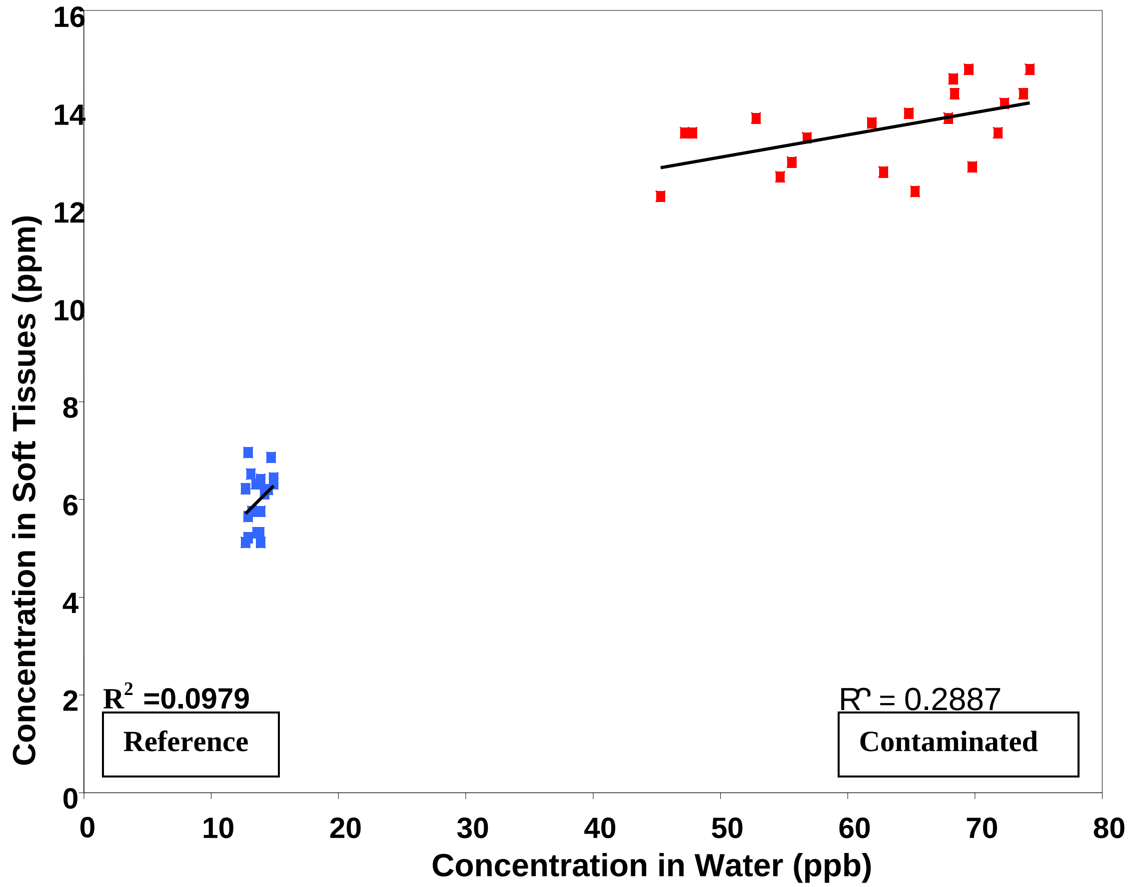

Figure 24. Correlation of the concentration of Mn in soft tissues of clam *A. umbonella* and water at the contaminated (p=0.015) and the reference sites.

The Cu concentration was also positively correlated with the concentration in water and sediment at the contaminated site (Figure 25, 26). There was a strong correlation between Cu concentration in soft tissues and sediment (Figure 25, $p<0.05$).

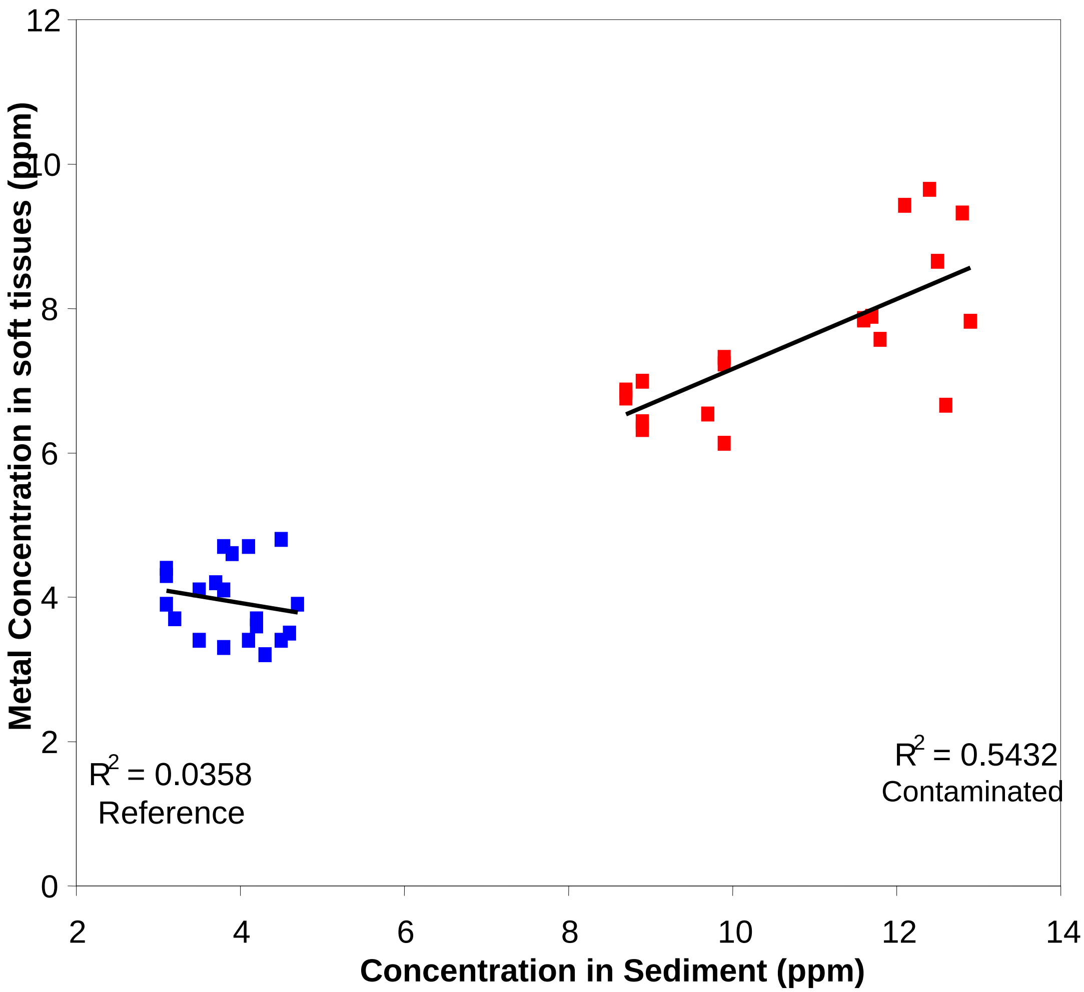

Figure 25. Correlation of the Cu concentration in soft tissues and Sediment (ppm) at contaminated (p=0.0002) and the reference sites.

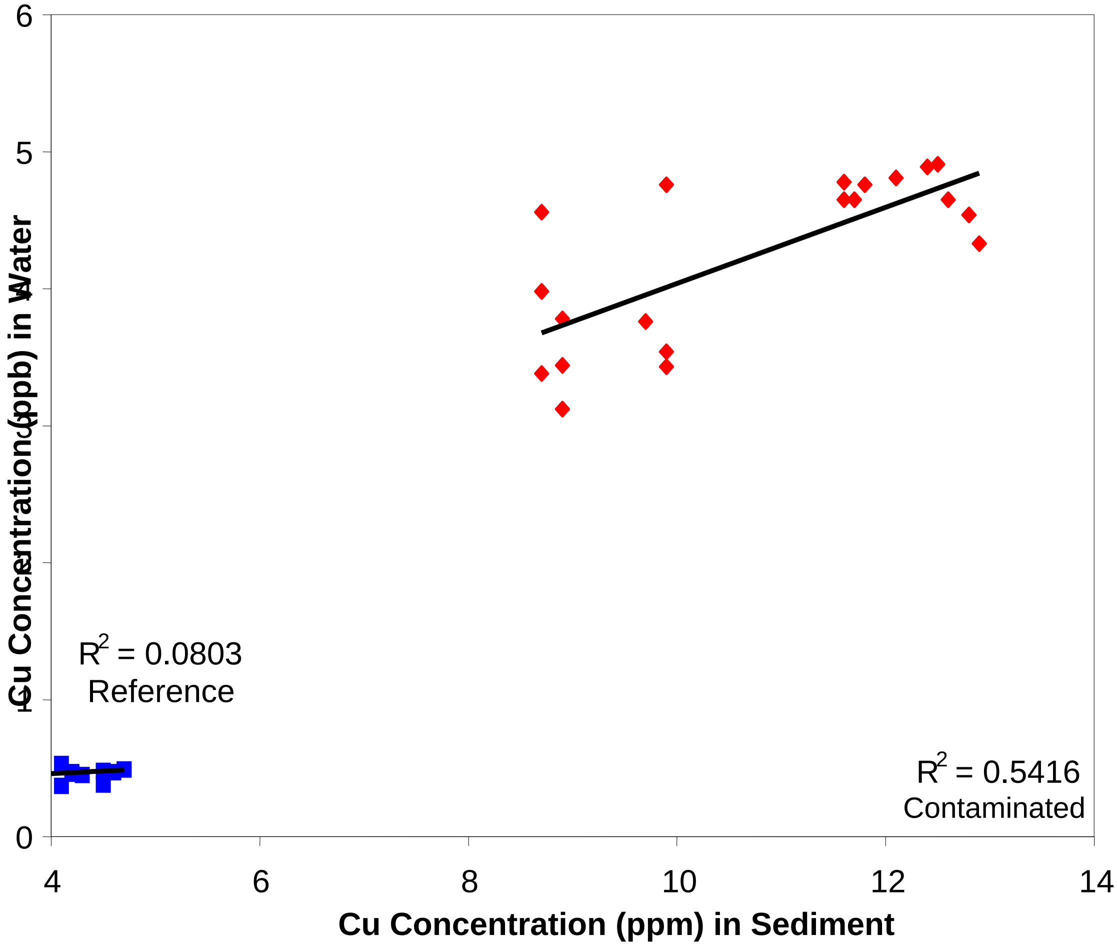

Figure 26. Correlation of Cu concentration in sediment (ppm) and water (ppb) at the contaminated (P=0.0128) and the reference site.

There was a strong positive correlation of the V concentration between sediment and water (Figure 27), but there was a negative regression slope of the V concentration between soft tissues and sediment and also between the concentration in soft tissues and water. There was a positive correlation of the Zn concentration between soft tissues of clam *A. umbonella* and sediment (Figure 28). The Fe concentration was positively correlated with the concentration in water (Figure 29).

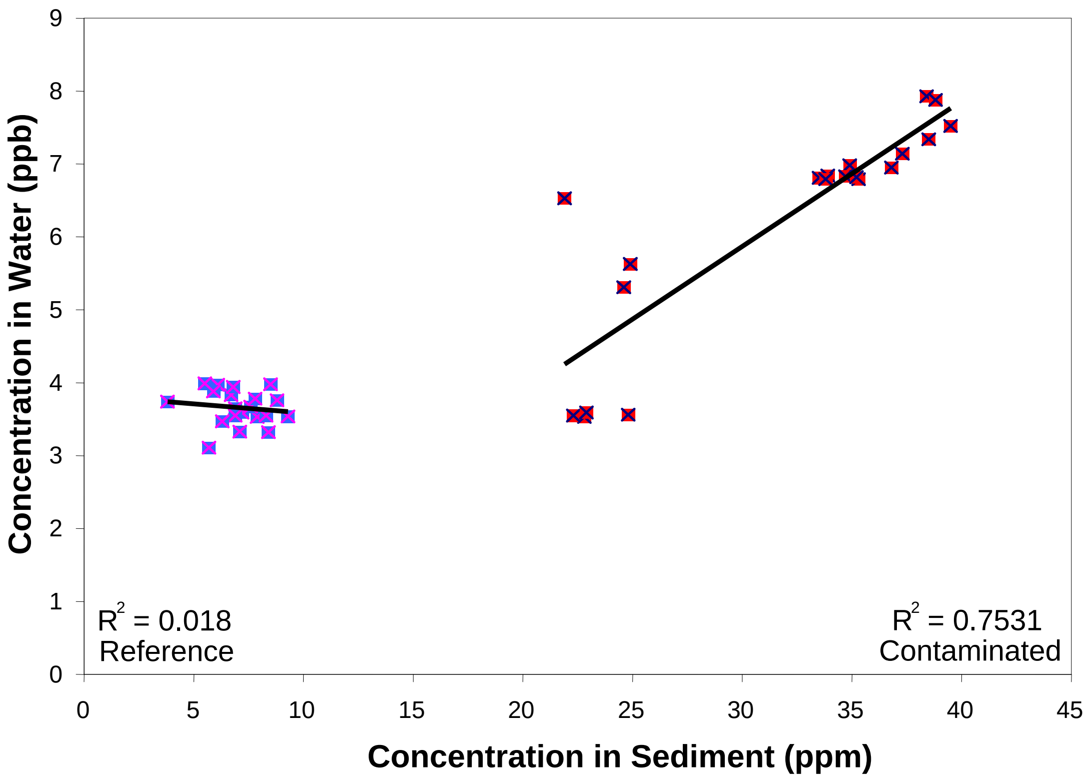

Figure 27. Correlation of the V concentration in sediment (ppm) and water (ppb) at the contaminated (p=7.16E-07) and the reference sites.

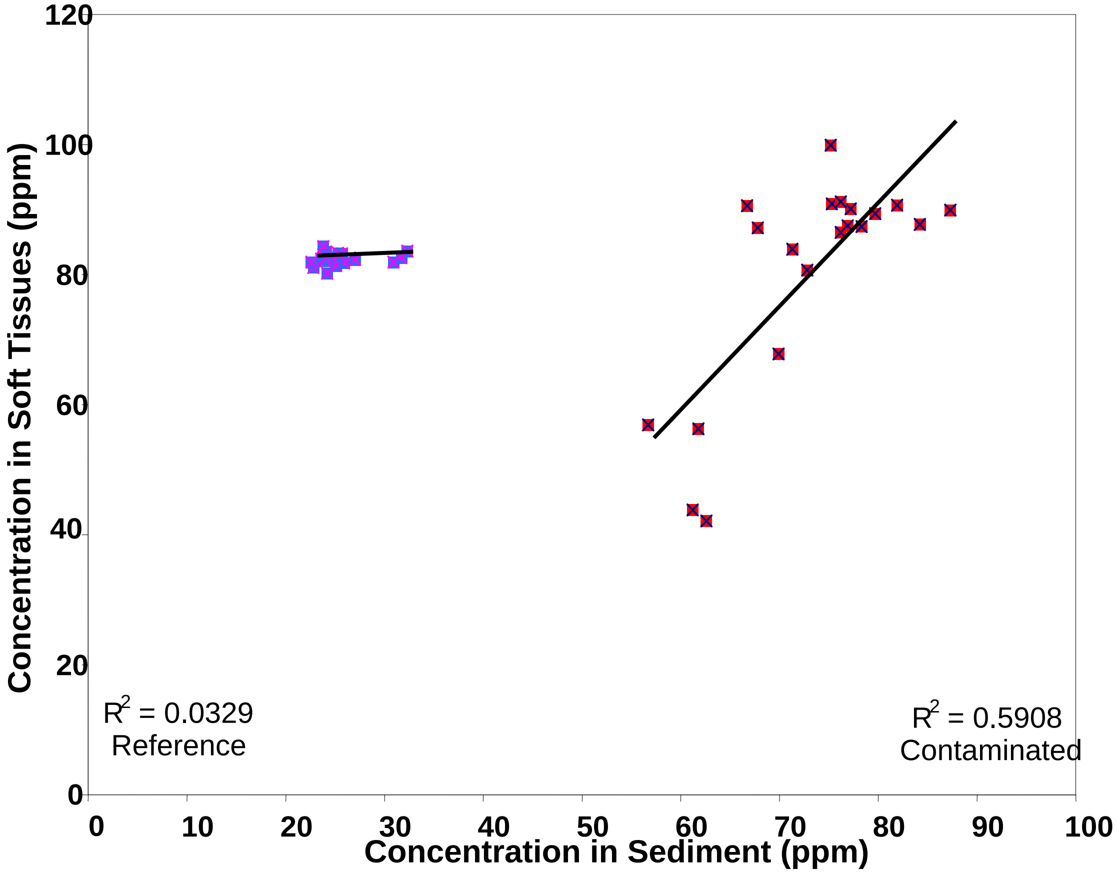

Figure 28. Correlation of the Zn concentration in soft tissues of clam *A.umbonella* and sediment (ppm) at the contaminated (p=7.52E-05)) and the reference sites.

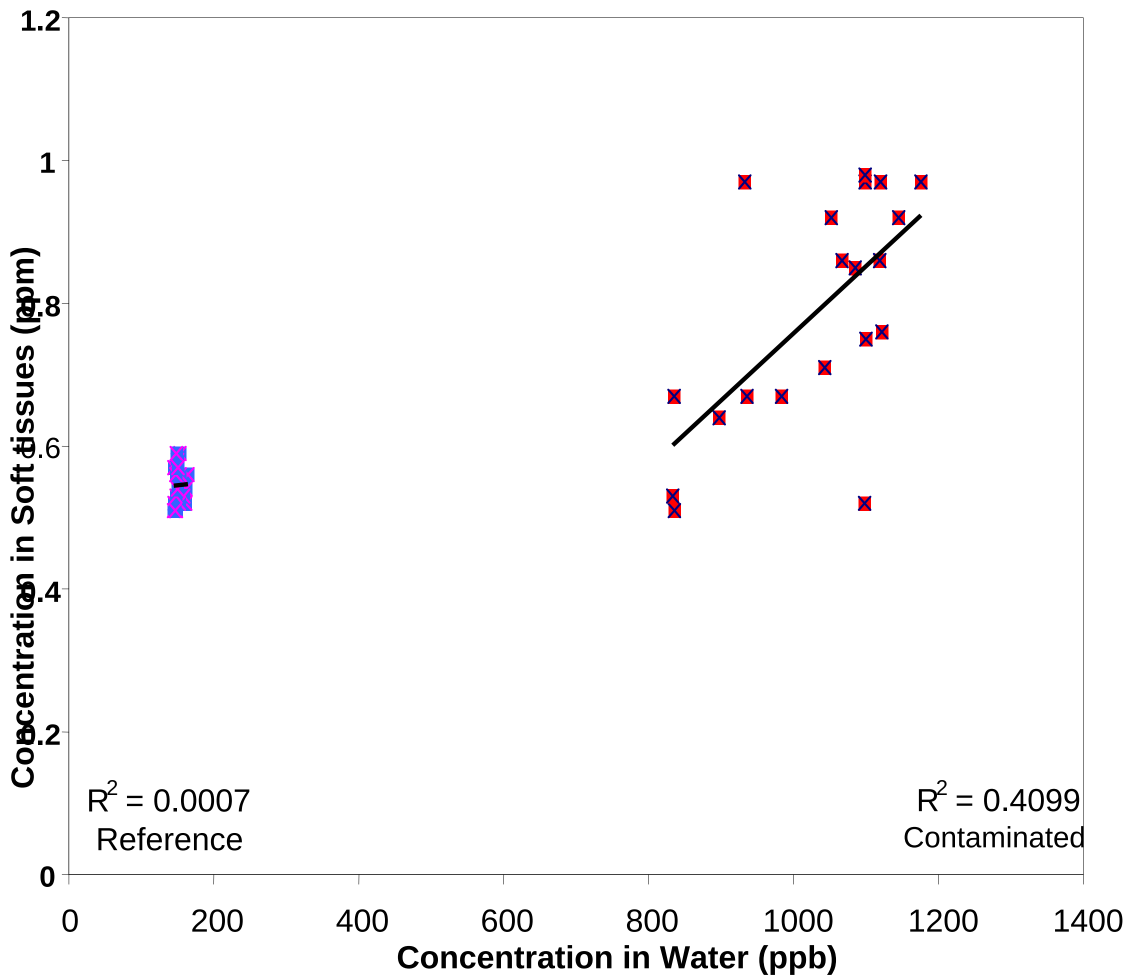

Figure 29. Correlation of the Fe concentration in soft tissues (ppm) of clam *A. umbonella* and water (ppb) at the contaminated (p=0.0033) and the reference sites.

There was a positive regression slope between the Cr concentration in clams soft tissues and sediment (Figure 30) and also between soft tissues and water (Figure 32, $p<0.05$). The Cr concentration in sediment was also positively correlated with concentration in water (Figure 31).

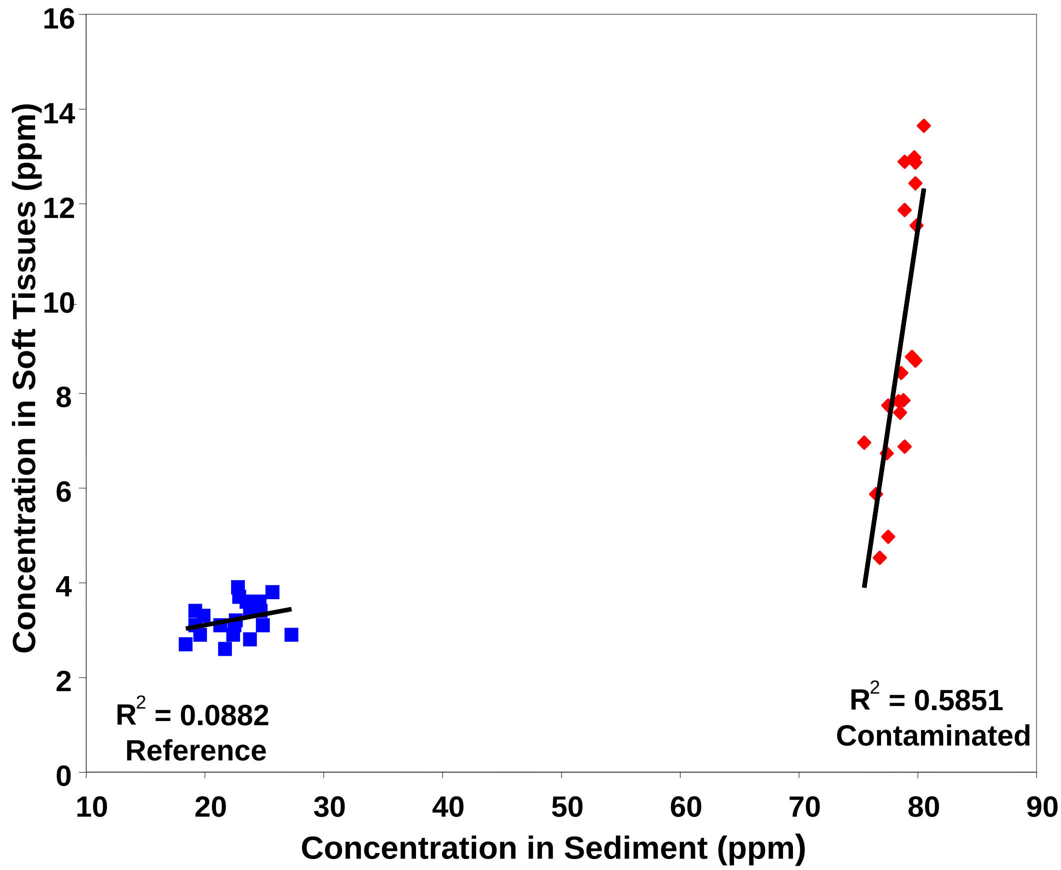

Figure 30. Correlation of the Cr concentration in soft tissues of clam *A. umbonella* and sediment (ppm) at the contaminated (p=8.54E-05) and the reference sites.

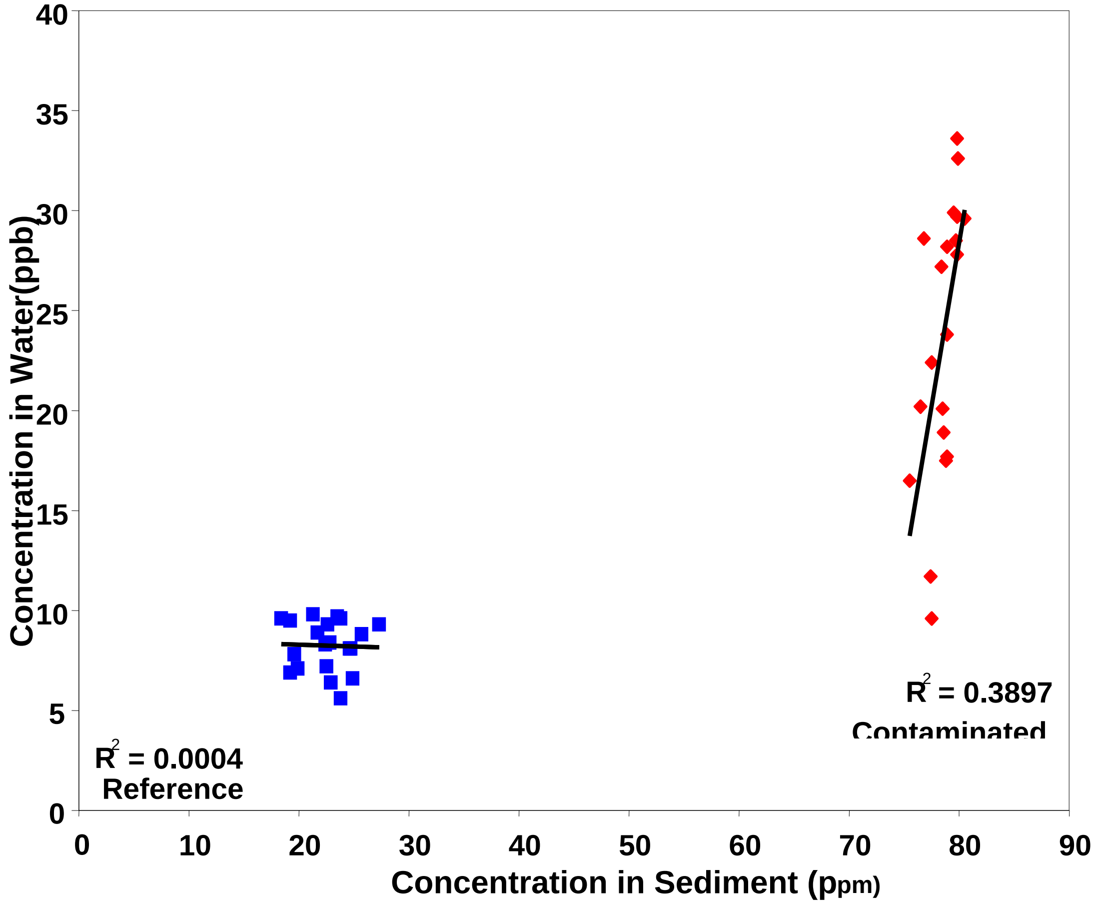

Figure 31. Correlation of the Cr concentration in sediment (ppm) and water (ppb) at the contaminated (p=0.003) and the reference sites.

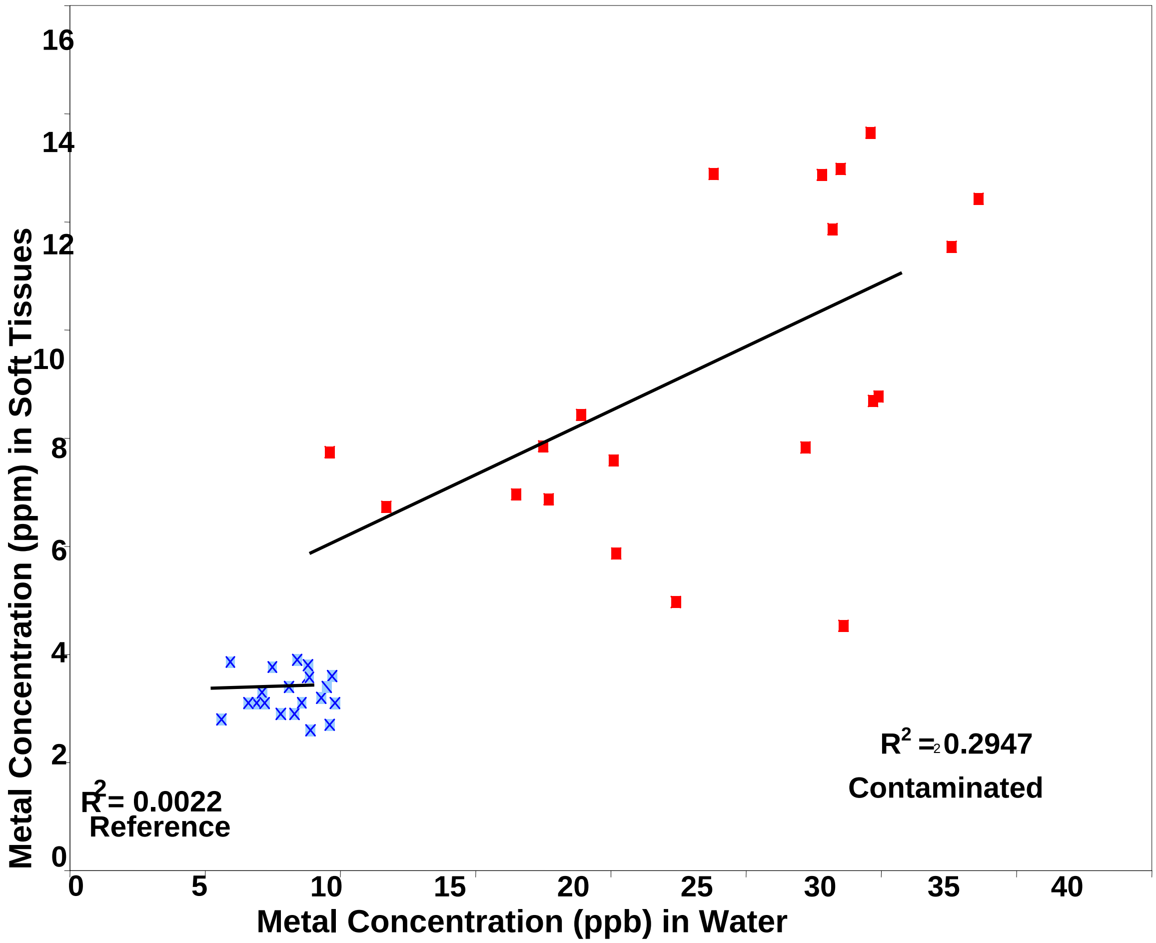

Figure 32. Correlation of the Cr concentration in clam's soft tissues (ppm) and water (ppb) at the contaminated (p=0.0134) and the reference sites.

There was a strong positive correlation of the Cd concentration in soft tissues of clam *A. umbonella* and the concentration in sediment (Figure 33) and in water (Figure 34). There was also a positive correlation of the Cd concentrations between sediment and water (Figure 35).

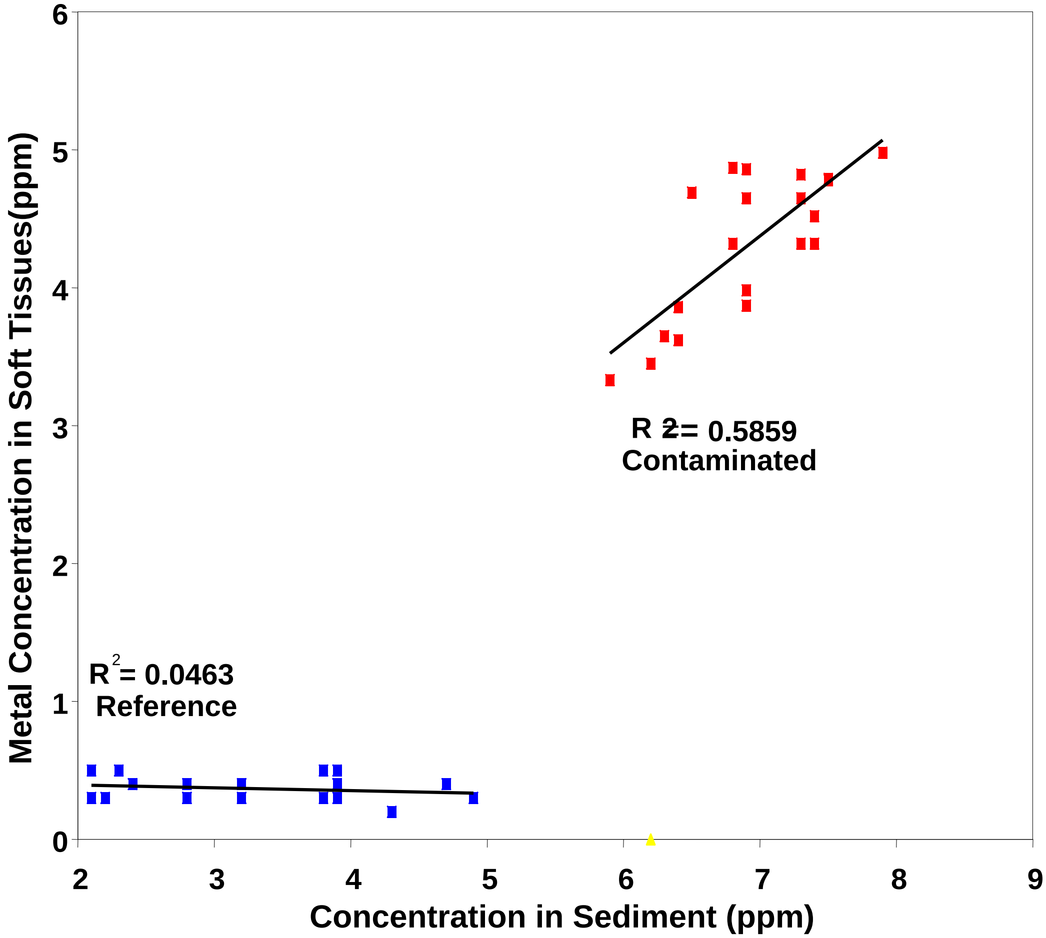

Figure 33. **Correlation of the concentration of Cd in soft tissues of clam *A. umbonella* and sediment (ppm) at the contaminated (p=84E-05) and the reference sites.**

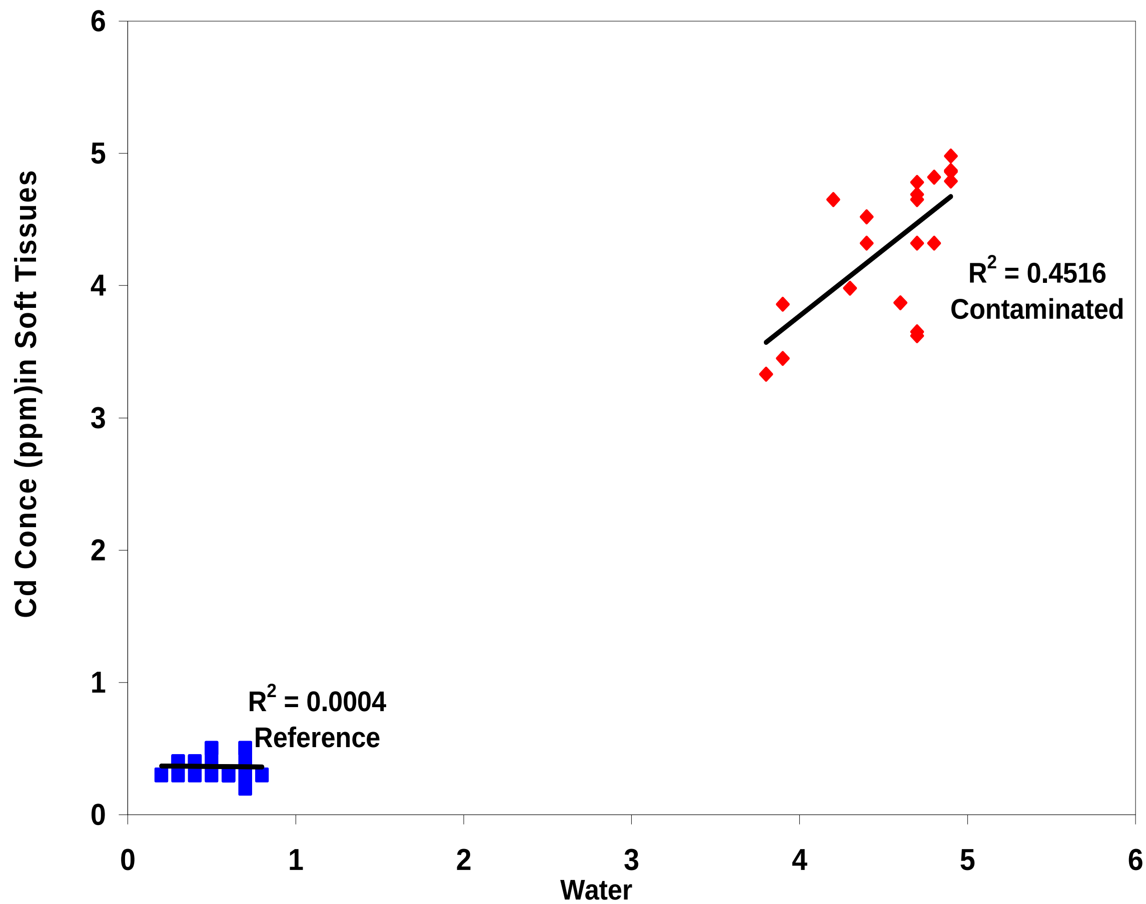

Figure 34. Correlation of the Cd concentration in soft tissues of clam *A. umbonella* (ppm) and water (ppb) at the contaminated (p=0.0001) and the reference site.

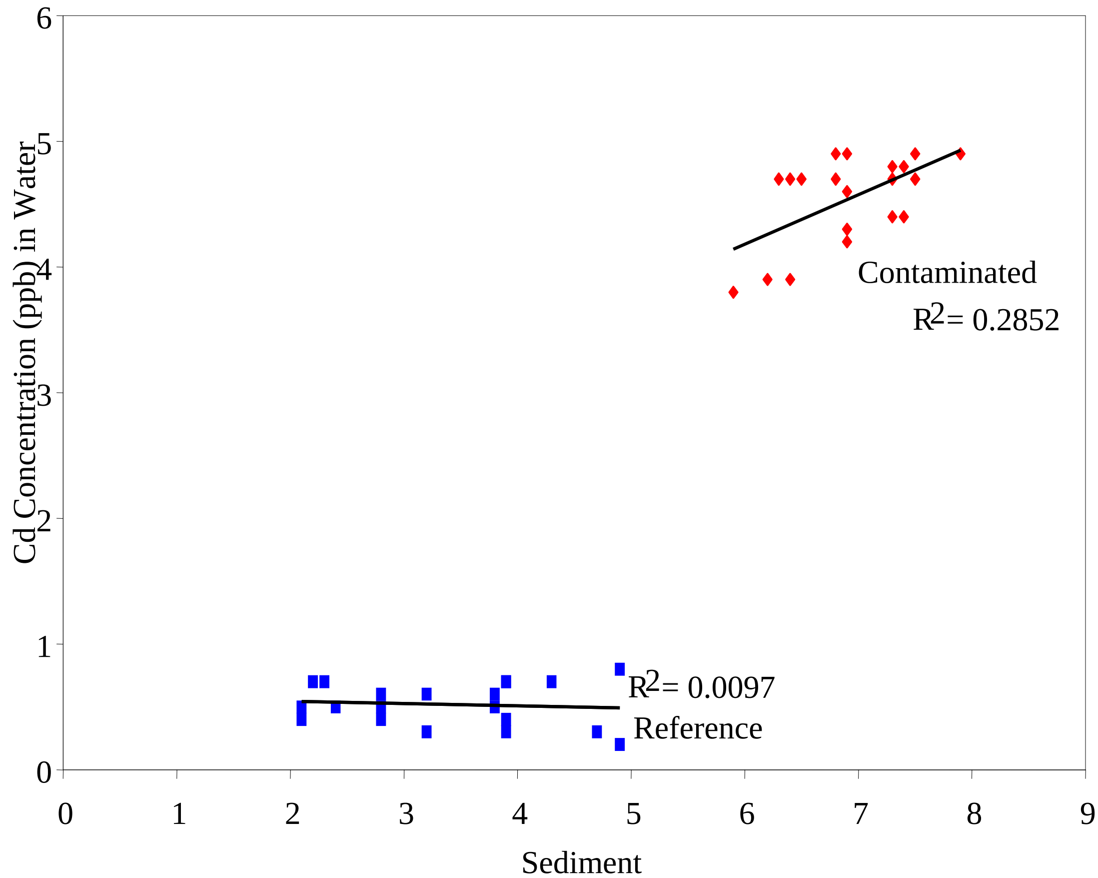

Figure 35. Correlation of the Cd concentration in water (ppb) and sediment (ppm) at the contaminated (p-0.007) and the reference sites.

2.8. Discussion

2.8.1. Metals in Water

The average concentrations of all metals in the water column were significantly higher at the sites of desalination and power plants compared to the reference sites. In addition the variability of the concentrations of Pb, Cd, Ni, Hg, Cr, V, Zn and Fe were higher at the contaminated sites than the reference sites (Table. 1). Trace metals may be discharged from the desalination plant with brine as observed by Abu –fayed, (2003) that reverse osmoses brines typically contain traces of iron, Ni, Cu, Cr and Zn. There are adverse impacts of desalination technology on the marine environment, mainly in the vicinity of the concentrated brine discharge pipe, because at times it accounts for much of the mortality of susceptible organisms in discharge waters, leaving their mark on the flora and fauna around the pipeline outlet (Aly Mohammed Aly Abdallah, 2003; Chouikhi, 2003). (Hoepner, (2002) indicated that marine ecosystem in the vicinity of a desalination plant will be affected directly as a consequence of changes in the physical, chemical and biological characteristics of the ambient waters.

Metals may also leach from treatment components from the discharged brine; therefore desalination effluents are considered a major source of pollution in the vicinity of a desalination plant (UNEP, 1999; Al-Awadhi, 1999; Subba Rao and Al-Yamani, 2000).

2.8.2. Metals in sediment

The concentration of trace metals (Hg, Cd, Pb, Cr, Cu, V, Mn, Ni and Fe) in sediment near the desalination and power plants were four to ten-fold higher compared to the reference sites. Sediments are often a major repository for contaminants introduced into surface waters (Luoma et al., 1990). Sediment accumulates concentrations of contaminants several orders of magnitude higher than those in the water (Marcos and Scott, 1990). Contaminated sediment can be directly toxic to aquatic life or can be a source of contaminants for bioaccumulation in the food chain. The significant role that sediment plays in aquatic ecosystems is well known. They serve as both as one of the primary sinks of anthropogenic trace metals in an aquatic system and source of organic and inorganic matters and the critical elements such as, C, N, P, and S (Burton et al., 1992) .

The spatial variability of metal concentrations was also significantly higher among sediment samples from the contaminated sites than the reference sites (Table 5). The highest concentrations of Cu, N, Pb and Cd in sediment were measured at the site nearest (1/4 km) the desalination plant. This could be due to the location being at the mouth of the outlet discharged from the desalination plant, an area with high interchange of water from outlet and inlet pipes.

2.8.3. Spatial differences in metal levels among clams

There were significant spatial differences of metal concentrations in clams. The clams collected from the two sites ¼ km from desalination/power plant had the highest levels of Cu and Cr and Ni compared to the other contaminated sites, ½ km far from point source. As observed by Abufayed, (2003), the reject stream from desalination and power plant is characterized by elevated temperatures and residual chemicals such as Cu, Ni, and Cr (representative of the manifold corrosion products) that can exert multiple effects on water quality, sediments and marine organisms. The enrichment of metals in clams near the desalination plant could be due to the exposure of these clams to reject stream water and concentrated brine, which is due to the density, sinks to the sea floor without prior mixing. As observed by Abufayed et al; (2003) heavy metal suspensions sinks to the bottom and tend to deposit especially in semi-stagnant areas affecting benthic invertebrates feeding on suspended or deposited sludge. Several authors have suggested that marine bivalves accumulate levels of Cr in excess of the surrounding sea water concentration (Phillips and Rainbow, 1993; Riget et al, 1996; Paez-Osuna, F et al., 1995). Discharges from power plants contain high concentration of Cu resulting in elevated Cu concentrations (8.0-14 ppb) in coastal receiving waters near power plant effluents (Dorgham, 1990). However the interpretation of Cu accumulation in bivalves is quite complex because it is also internally regulated (Shulkin et al., 2002, 2003).

Cd concentrations in clams were significantly higher ($P<0.05$) in all contaminated sites and 3- folds higher than Zn concentration compared to the reference sites. A possibility for this could be the Cd and Zn inhibit each other's uptake rates, but the mechanism is unclear (Vercauteren and Blust, 1999).

The average Pb and Cd concentration in clam tissues was significantly higher ($p<0.05$) in the contaminated sites compared to the reference sites. Since clam *A. umbonella* is a filter feeder, there would be stronger correlation between metal concentration in water and metal concentration in clam's tissues, than that between clams soft tissues and sediment. But there was a positive stronger correlation of Cd and Pb concentrations in clam's tissues and concentrations of these metals in sediment. The feeding behavior of these clams is such that they can switch from suspension feeding to feed on the newly deposited particles in the sediment. Although the clam *A. umbonella* used in this study is benthic suspension feeder, sedimentary particles can be a potentially important source for metal uptake by the clam due to sediment re-suspension as a result of tidal currents, particularly in shallow regions such as at Doha bay. Hence, it is possible that this feeding behavior of clams led to the higher concentration of Pb and Cd in its tissues.

Increases in ambient water temperature and salinity and depletion of DO (dissolved oxygen) are the major determinants of a typical brine discharges (Aly Mohammed Aly Abdallah, 2003). The higher water temperature in the power plant effluent and discharge could be partially responsible for the higher metal concentration in clam *A. umbonella*. Metal accumulation by marine bivalves can increase with a rise in temperature (Phillip and Rainbow, 1993). Higher temperature generally implies increased absorption through cellular membranes, and higher absorptions due to higher metabolic rates (Maclnnes &Calabrese, 1978; Bodek et al., 1988). Thus at higher temperatures bivalves suffer greater toxic effects of metals due to increased siphoning and sensitivity to toxicants (Belanger et al, 1990).

2.9. *Metal Interactions and Relationship among metals*

There were several correlations between metal levels in soft tissues, water and sediment. For example there were positive correlations of Ni and Cr concentrations in soft tissues of clams and sediment and also between soft tissues concentration and water. Lattemann Sabine, (2003), reported that chemicals leached from treatment components mostly through erosion and corrosion increase levels of metals such as NI and Cr in water and in fine sediments. The accumulation of metals from water and fine sediments led to elevated concentration of Ni and Cr in soft tissues of clams. The increased accumulation of Ni and Cr in clams near the desalination plant is likely due to the presence of elevated levels of Ni and Cr in reverse osmosis brine.

There was also strong correlation ($P<0.05$) of Pb concentration in clams soft tissues and water. Pb exists in water in particulate form (Balls, 1985) and bivalves take up Pb from the water and food particles in similar rates and therefore reflect environmental pollution effectively (Rainbow, 1995).

There was a positive strong correlation ($P<0.05$) of Cd concentration between clam's soft tissues and water, and also between soft tissues and sediment. The Cd is found in marine waters mostly in dissolved form (Balls, 1985). There was a strong correlation of Cu concentration ($P<0.05$) between clam's soft tissue and water, and also between soft tissues and sediment. It was expected since clam *A. umbonella* is a filter feeder; there would be a stronger correlation between metal concentration in soft tissues and metal concentration in water. However, these results indicate a statistically strong correlation of Cu concentration ($P<0.05$) between clam's soft tissues and sediment rather than Cu concentration in soft tissues and water. The ability of many in- faunal benthic species to switch between deposit feeding and suspension feeding depend on the flow regime (Taghon et al, 1980). Dauer et al, (1981) used the term "interface" feeders to refer species that were not obligatory deposit or suspension feeders. Hence, the strong correlation ($R^{2}=$ 0.58, 0.54) of Cd and Cu concentration between clam's soft tissues and sediment rather than concentration in soft tissues and water, could be due to stress or poor quantity of suspended particulate matter that caused the switch from filter feeding to deposit feeding.

Although total concentrations of some metals in sediment may be generally correlated with concentrations in organisms (Luoma and Bryan, 1978, 1979, 1982), predictive relationships are rare. Metal exposure is not the only process that affects metal bioaccumulation (Luoma and Jenne, 1977; Sunda and Guillard, 1976; Luoma, 1983). Physico-chemical reactions in receiving waters (which could vary with the time) can also affect bioavailability (Luoma, 1983). In addition to the influence of metal concentration, bioaccumulation can be complicated by external environmental factors such as salinity, pH, temperature, competing ions (Luoma, 1989; Rainbow et al; 1990). In this study due to the concentrated discharge of brine from the desalination plant directly into the marine sensitive ecosystem, that increasing temperature, salinity, depletion of DO (dissolved oxygen) and fluctuating in pH, could be affecting synergistically on the population of clam *A. umbonella* at Kuwait Bay.

2.10. Geographical differences in metal levels

Many studies have used clams to compare contaminated with uncontaminated sites to examine point source pollution (EPA, 1974; Zorba Mazin et al., 1992; Genest and Hatch, 1981;Bryan et al., 1980,1985; Gagne et al., 2006; Philip Rainbow, 2006; Chong and W. X. Wang, 2001)

Most of the previous studies focused on investigating the characteristics and pollution of the coastal waters but did not evaluate the physico-chemical characteristics and pollution level within the intertidal zone (Bakri Al Dhia and Kittaneh Wajeh, 1998; Paulson et al., 2003). In addition, many reports do not indicate where samples were collected with respect to intertidal or sub tidal zones. Most published results are for composite samples rather than individual organism (Mora de Stephen et al, 2001; Diaz et al., 1997; Chong and Wang. 2000). This does not allow direct comparison of contaminant levels among studies. These studies are not dependable because element concentrations in bivalves at the same location differ between different species and individuals due to species-specific abilities and capacities to regulate or accumulate trace metals (Reinfelder et al., 1997; Otchere et al., 2000). Further there is a limited comparative data for clams in the Arabian Gulf, although there have been some studies that examine contaminants including heavy metals in clams, oyster, algae and in some crustacean (Buo-Olayan and Subhramanyam, 1996; Campanella et al., 2001; Al-Mohanna and Subrahmanyam, 2001). Clams have been rarely used as bio-indicators of heavy metal contamination in marine coastal waters in Arabian Gulf. Hence the clam's data from this study can be serving as a basis for future studies on metal contaminations in clams to evaluate the adverse impacts of desalination technologies on the surrounding environment. These impacts near Kuwait area will be magnified because this water body partially enclosed and contains highly sensitive ecosystems.

Table 13 summarizes published data on the levels of the metals in clams from marine coastal waters in various parts of the world. It is apparent that the mean levels of Ni, Pb, Cr, Mn, Zn and V found in clams from most part of the Kuwait bay along the desalination/power plant are within the ranges of means commonly reported in the literature, including both contaminated and the reference sites. However, the Cd levels from all sites adjacent to desalination plant from this study (Kuwait) were highest (4.7-4.8 ppm) among the Cd levels reported in the literature. The Cd level reported from McMurdo Sound in Antarctica is also towards higher side (Negri Andrew et al., 2002). In addition, the Hg level of 5.9-6.2 ppm from all the contaminated sites in Kuwait Bay of this study is the highest mean value reported in the literature. Al-Majed and RaJab, 1998; Fowler, 2002, reported highest Hg concentrations in near shore surface sediments in Kuwait Bay and United Arab Emirates. According to Campbell et al., (1986) Hg concentrations increase with increasing salinity at values greater than 25 psu. This might have been the reason for increased level of Hg in clams from Kuwait Bay in this study, because the salinity in Kuwait bay was between 36 – 45 psu. There is a need to identify a point source responsible for this elevation. Further the Cr level of 7.9 ppm in this study is the higher mean value compared to Qatar in the same geographical area of the Arabian Gulf. The V level range from 0.3 to 1.0 ppm in clam's soft tissues from all contaminated sites in Kuwait bay is also a higher mean value compared to the V level, 0.76 ppm in Qatar (Mora de Stephen et al., 2001). As indicated by Al-Yamani et al., (2001) the unregulated discharge of waste water from desalination and power plants into the marine environment over many years has resulted in high concentrations of Hg, Pb, V, and Nibeing found in the sediments of Kuwait Bay.   However the Mn mean level is almost the same in both areas of Kuwait bay and Qatar.

2.11. *Metal levels and risk*

Three metals had high enough levels in the clams from Kuwait to suggest that they pose a risk to people who eat them. Bivalves are capable of accumulating extremely high levels of Cd in edible portions and therefore represent a greater hazard to human consumers than other marine organisms (EPA, 1978). For human consumption, according to (FAO) Food and Agricultural Organization (2005), the limit of Cd in bivalves for human ingestion is 4.0 ppm and the limit of Pb is 1.7 ppm in mollusks. The highest levels of Cd in clams from Kuwait were from 0.77 to 4.8 ppm, which exceeded the consumption levels set by FAO and (FDA) Food and Drug Administration, (4.00 ppm), which could affect the marine environment and humans (Hosch, 1996). Pb level in clams from Kuwait was 1.9 to 2.00 ppm that could be hazard to human consumers. World Health Organization (WHO) sets a limit of 0.5 ppm for Hg in shellfish for human consumption; the U.S. level is 1 ppm (FAO, 2005). The mean levels of Hg in clams from Kuwait were from 0.6-6.2 ppm, which also exceeded the limits established by WHO and FDA.

2.12. Summary

There is a need to critically assess the present quality of the marine ecosystem, especially the connection between ecosystem change and threats to human health. In this study an indigenous clam *A. umbonella* has been used as an effective bio-indicator species for desalination and power plants, waste water discharge in Kuwait. In addition to its use as a bio-indicator, the high levels of some metals are a concern as these bivalves are consumed by humans and wildlife. This study would provide for an early detection of potential marine-based contaminants, thus allowing for the protection of marine ecosystems and the prevention of associated human exposure. It would also provide baseline data for future sustainable monitoring programs of the coastal environment, especially for Hg, Pb and Cd.

Table 13. Ranges of mean values of metal concentrations in clam

Ranges of mean values of metal concentrations in clam *A. umbonella* by geographic region. All values are ppm (parts per million, dry weight basis). * = Composite samples.

Geographic Region	Species Clam	Ni	Hg	Cd	Pb	Cu	Cr	Mn	Zn	V	Ref
Kuwait bay, Kuwait	*Clam *Amiantis umbonella* n=3	5.4-6.8	0.6-5.9	0.77-3.9	0.27-0.48	4.8-7.5	5.2-7.71	13.4-19.4	84.5-103	0.4-0.8	This paper 2001
Kuwait bay Kuwait	Clam *Amiantis umbonella* n=32	5.3-8.3	0.5-6.2	0.47-4.8	0.51-2.0	4.7-8.8	5.1-7.9	12.2-20.9	82.2-110	0.3-1.0	This paper 2001
Hong Kong	**Clam Ruditapes Philippinarum* n=*7*			0.06-0.069			0.028-0.029		0.23-0.27		Chong et al., 2000
	**Macoma balthica* n=*7*			0.032-0.08			0.006		0.091-0.20		
	**Potamocorbula amurensis* n=*7*			0.125			0.028		0.425		
Guaymas Bay, Gulf of California, Mexico	**Chione gnidia, *Laevicardiumelatum* n=85	4.68-23.65		0.21-1.67	0.51-4.03	4.78-23		1.59-26.9	105-246		Mendez et al., 2002
San Antonio Bay, Chile I	*Surf Clam, *Mesodesma donacium*. n=10		0.11-.39		0.09-0.33						Diaz et al., 1997
Morocco	**Scrobocularia plana*			0.15-1.64		16.2-20		14.6-39.7	142-222		Cheggour et al., 1999

Geographic Region	**Species Clam**	**Ni**	**Hg**	**Cd**	**Pb**	**Cu**	**Cr**	**Mn**	**Zn**	**V**	**Ref**
Qatar	**Clam, Circentia callipyga* n=18						0.97	17.7		0.58 - 0.76	Mora de Stephen et al., 2000-2001
Tropical South America (N. Brazil)	Clam *Anomalocardia brasiliana* n=*1*	1.79		1.81		7.18		33.1	62.1		Carlos et al.,2006
Quebec, Canada	Clam *Mya arenaria* n=*1*	6-11.70.0	0.005-0.02	0.06-0.8	0.05 - 0.17	1.2-2	7.8-16	3.6-10.3	7-9.1	0.38 - 0.91	Gangne et al., 2003
Indian River Lagoon, Florida	Clam *Mercenaria mercenaria* n=3	0.5-10	0.023-0.12	0.11-0.86	0.7-12	6.3-26	0.17-0.40	4-1160	12-353	0.25-3.7	Robert et al.,1992
Chinese Bohai Sea	Clam *Ruditapes philippinarum* n=1	0.72-2.97		0.14-0.63	0.13 - 0.33	1.28 - 4.37			9.95-20.06		Liang et al., 2004

3. Distribution of Heavy Metals in the organs of the clam *A. umbonella* living near a desalination / power Plant in Kuwait

3.1. Abstract

The concentrations of Hg, Cd, Cu, NI, Mn, Pb, Cr, Fe, V, and Zn were measured in different organs of the clam *A. umbonella* collected in Kuwait Bay in an area where desalination and power plant waste water discharges are located. A comparative reference point was five kilometers away from the source. The objectives were: 1) to determine the organs in which specific metals concentrate, 2) to examine the relationship between metal enrichment in organs and metal concentrations in sediment and water; and 3) to compare metal concentrations in different organs of clams from the point source impacted and the reference sites. This study found that the concentration of all metals, except Zn, were significantly higher in the kidneys, gonads, mantles, gills, livers and hearts of the contaminated site clams, compared to the reference site clams. Further, concentrations of Cd and Pb (106.7 ppm, 97.5 ppm, respectively) were found to be significantly higher ($p < 0.05$) in kidneys than in all other organs. The highest inter-location variability was in the kidneys, compared to all other organs. The concentration of Cu, Hg, Ni and Fe in the contaminated site clams was significantly higher in the gonads (15.0 ppm, 2.32 ppm, 4.3 ppm, and 4.6 ppm wet weight) than in the kidneys, mantles, or livers. The highest Cr concentration (12.5 ppm) was in the mantles. The concentrations of Zn in organs did not differ between the contaminated sites and the reference sites. The liver (digestive gland) was found to be the main depository of Cu (16.7 ppm) in the contaminated site clams. There were several strong positive correlations between metal concentrations in clam organs and sediment as well as water

at the contaminated sites. For Hg there was a significant correlation in water and gills (R^2= 0.72), and also a strong positive correlation in sediment and gonads (R^2 = 0.53). The Cd concentration in water and mantles were positively correlated (R^2 =0.65) and Cu concentration was positively correlated between sediment and kidneys (R^2 = 0.54).

3.2. Introduction

The Kuwaiti mainland coast, which extends for about 400 Km is of vital importance for development in the country. Most of the urban, commercial, industrial, and recreational activities in Kuwait are concentrated within 15 km of the shore lines Furthermore, the coastal water is virtually the only source of fresh water and energy in the country; several desalination/power plants were established along the shoreline to meet the country’s need for drinking water and electricity (Bu-Olayan et al., 2006). The seawater used for cooling the power plants is also discharged to the sea. The discharge of salt and cooling water may increase temperature and salinity of the coastal water (Al Bakri et al., 1998). The coastal area is considered a valuable natural resource containing important ecosystem and supporting many organisms, whose extinction may affect the whole marine environment. The coastal zone is also a very important nesting and feeding ground for many resident and migratory birds (Al Bakri et al., 2000). There has been only a single study thus far on the ecology, biology and allometric relationship of intertidal clams in the Arabian Gulf (Jassim et al ., 2006).

Marine pollution is a global environmental problem because human activity on land, in the water and in the air contributes to contamination of sea water, sediments and the organisms. The potential threat of metals to the aquatic organisms has been directly and indirectly assessed recently using several methods. Assessing the bioaccumulation of metals by the tissues of marine organisms has been studied as an indirect measure. A wide range of organisms have served as bio-indicators to monitor the environmental effects and bioavailability of pollutants in the aquatic environment (Langston, 1990). In particular, bivalves have been recognized as useful sentinel organisms of contamination in aquatic ecosystem because of their abundant population, wide geographical distribution, sedentary habit, mode of feeding, and capacity to accumulate xenobiotic substances above background levels. The "Mussel Watch Program" successfully exploits this ability of filter-feeding bivalves, to identify coastal marine areas with elevated levels of heavy metals (Goldberg et al., 1978; Sericano et al., 1993; O'Connor, 1998, 2002). Bio-monitoring and surveillance are key methods for assessing the status or well being of ecological receptors within functioning ecosystems (Burger and Gochfeld, 2001). The use of bivalve molluscs as bio-indicators of trace metal enrichment or biological stress in aquatic environments has gained widespread acceptance (Goldberg et al., 1978; Bryan et al., 1980; Phillips, 1980, a, b ; Popham et al., 1980; Jensen et al., 1981; Thomson et al., 1984). Bivalve mollusks are known to tolerate high metal concentration without harming themselves (Ahn et al, 1996). The response to the metal concentration increase also includes metal redistribution among organs, characterizing the balance between metal accumulation and elimination (Podgurskaya et al., 2004). Several studies have indicated that the variability of metal concentrations among the different soft tissues and metal concentrations in the kidneys and livers (digestive gland) were usually significantly higher than in any other soft tissues (Nigro et al., 1994; Ahn et al., 1996).

This study was designed to compare metal accumulation in different organs of marine clam *A. umbonella* populations living along point-source contaminated and uncontaminated sites in Kuwait Bay. The concentrations of Hg, Cd, Cu, Ni, Mn, Pb, Cr, Fe, V, and Zn were measured in different organs of this facultative suspension feeding clam *A. umbonella,* collected near desalination and power plant industrial sites and a reference sites 5 km away from point source at Kuwait Bay. The objectives of this study were therefore to: 1) determine the organs in which specific metals concentrate, 2) examine the relationships between metal enrichment in organs and metal concentrations in sediment and water to which the clams are exposed, and 3) compare metal concentrations in different organs of clam *A. umbonella* from the point-source impacted and the reference sites.

It is hypothesized that:

1. Organs rich in protein (liver, kidney and gonads) will accumulate more Hg, Cu and Cd than other organs.

2. The bioaccumulation of certain metals including Hg, Cu, and Cd results in decreased organ and soft tissues weight of clams at the contaminated sites compared to the reference sites.

3. There will be a strong relationship (correlation) between the concentrations of metals in the organs of clam and those in its environment (water and sediment).

4. There will be strong relationship between metal concentration in specific organs and water than those in sediment.

3.3. Materials and Methods

3.3.1. Study Sites

The sampling sites for clams *A. umbonella* is shown in Figure/Map 2 (Chapter 2).

3.3.2. Clams Collection and Preparation

For organ analysis, thirty three clams were randomly collected from two contaminated Sites 2A, 3A and from reference sites during December 1999, January 2000 with similar shell length to minimize the effect of body weight (Marina and Enzo 1983). These sampling sites are shown in Figure/Map 2. After collection clams were transported to the laboratory in self-supporting tanks (capacity of 150 liters, aerated sea water continuously flowing). Clams were depurated in seawater for approximately 24 hours. Seawater salinity and temperature were measured during each sampling.

Each clam opened carefully using stainless steel scalpel blades. Soft organs such as gonads, stomach, digestive glands, gills, mantles, foot, hearts, and kidneys were removed. The fresh weight of each organ was recorded (mg).

The following weights were recorded:

1. FW: Total flesh weight
2. GW: gonads weight
3. DG: digestive glands weight
4. SW: both shells weight
5. MW: mantle weight
6. St W: stomach weight
7. Kd W: kidney weight
8. Ft W: foot weight
9. HW: heart weight

All these organs were transferred to different vials and stored at -20 ^{o}C for further investigation of metal depositions using atomic absorption.

Prior to analysis, clam tissues were freeze dried to a constant weight (±0.5%). Then the sample was ground to homogenize it and sieved in order to normalize the variation in the grain size distributed. Samples were extracted by placing 0.5 gm of the homogenized sample in a Teflon beaker and heating it (120 °C) with concentrated HNO3 until dry. 5 ml acid (HNO3+HCLO4+HF) was added to the sample and heated up to 120 °C until dry. Then the sample was cooled to room temperature and passed through filter paper and poured into the 50 ml volumetric flask, diluting with HCl 0.1% to make up the volume.

Water and sediment samples were also collected from each of the two contaminated sites and reference sites (Chapter 1).

3.4. Statistical Analysis

To test the hypotheses, all analytical tests can be found in Chapter 2.

3.4.1. Length and gonad wet weight of clam A. umbonella

The mean total wet weight of gonads, mantles and gills were significantly reduced in clams collected from the contaminated site compared to the reference sites (Figures 3, 4).

The linear regression was performed between clams length (cm) and mean gonads wet weight (mg) at the reference and the contaminated site (Figures 5, Figure 6). Gonads weight were significantly decreased (R^2 =0.07) as clams length increased at the contaminated site (Figure 6). It was observed at the reference site that there was a positive correlation (R^2=0.65) between clams length and mean gonads wet weight (Figure 5) compared to the contaminated sites.

3.4.2. Metal concentrations in organs

The Hg, Cu, Cd, Pb, Cr, V, Zn and Fe concentrations in various organs of clam *A. A. umbonella*, at three contaminated and reference sites (A,B and C), average, SD (standard deviation), T-test, contaminated/reference sites, RSD (relative standard deviation), CI (confidence interval) are listed in tables 1, 2,3 4,5, 6, 7,8 and 9. Bar graphs (Figures 5, 7, 10, 12, 14, 16, 18, 19, 21 and 23) show the mean metal concentrations of Hg, Cu, Cd, Pb, V, Zn and Fe in the various organs of clam *A. umbonella* between contaminated and the reference sites. Bar graphs (Figures 6, 8, 11, 13, 15, 17, 20, 22 and 24) show the increment enrichment percent (%) of metals such as Hg, Cu, Cd, Pb, Cr, Ni, V, Zn and Fe in the various organs at the contaminated sites compared to the reference sites. Bar graphs (Figure. 25, 26, 27 and 28) show the mean metal concentration (ppm) in clam kidneys, mantles, livers and gonads. Pie charts (Figure 33, 35, 37 and 40) show the percent (%) of Cd, Pb, Cu and Hg concentrations in gonads, mantles, kidneys, gills and livers.

The mean concentrations of all metals at the study sites except Zn were significantly higher in the kidneys, gonads, mantles, gills, livers and hearts compared to the reference sites. The mean concentrations of Pb, Cd, and V were significantly higher ($p<0.05$) in the kidneys (Figure 25) than in any other organs such as livers, mantles and gonad. The concentrations of Cd and Pb (106.7 ppm, 97.5 ppm, respectively) were higher in the kidneys than other metals. The concentrations of Hg, Ni, and Fe at the contaminated sites clams were significantly higher in the gonads (2.32 ppm, 4.3 ppm, 5.72 ppm respectively) than in the kidneys, mantles and livers (Figure 28);where as the highest Cr concentrations (12.5 ppm) were found in the mantles (Figure 26). The gonads, mantle, livers, kidneys, hearts and gills have significantly higher ($p<0.05$) concentrations of Hg, Cu, Cr, Ni, Pb and Cd in clams collected from the contaminated sites compared to those from the reference sites. The liver (digestive gland) of clam (Figure 27) was the depository for the highest concentration of Cu (16.7 ppm) compared to the kidneys, gonads and mantles. The Zn concentration in organs was not significantly differing in clams from the reference and the contaminated sites.

3.5. Relationship between metal concentrations in water, sediment and organs

Several inter metal correlations between the organs and water as well as the organs and sediment were determined (n =16) by using regression analysis. There was a strong positive correlation of Cd concentrations between water and mantles (Figure 32, R^2 =0.65). There was also a positive correlation of Cu, Cd and Pb concentration between sediment and the kidneys

(Figure 34, 31, 33: R^2 = 0.54, R^2 = 0.51, R^2 = 0.49). There was also a positive correlation of Hg concentration between sediment and the gonads (Figure 36; R^2 =0.53), between water and kidneys (Figure 38; R^2 = 0.44) and strong positive correlation between Hg in gills and water (Figure 37; R^2 =0.72). A strong positive correlation ($p<0.05$) was observed of Cu levels between water and gonads (Figure 35; R^2 = 0.52); between sediment and the kidneys (Figure 36; R^2 =0.54).

The highest concentration of Hg was in the gonads (42%); where as concentrations of Pb and Cd was highest in kidney (86%, 90% respectively). The highest elevated level of Cu was in liver (26%).

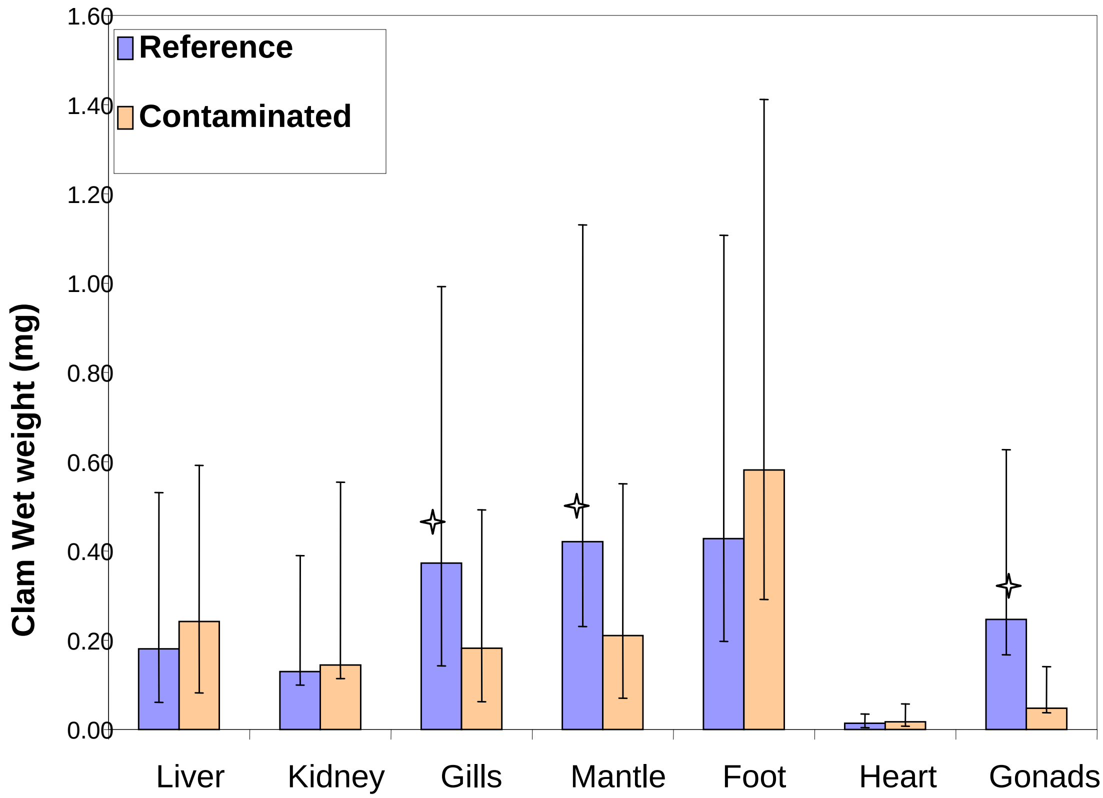

Figure 1. Mean organ weight (mg) of clam *A. umbonella* at the contaminated site and the reference site.

✧ = significantly different. Error Bars show weight max &min range.

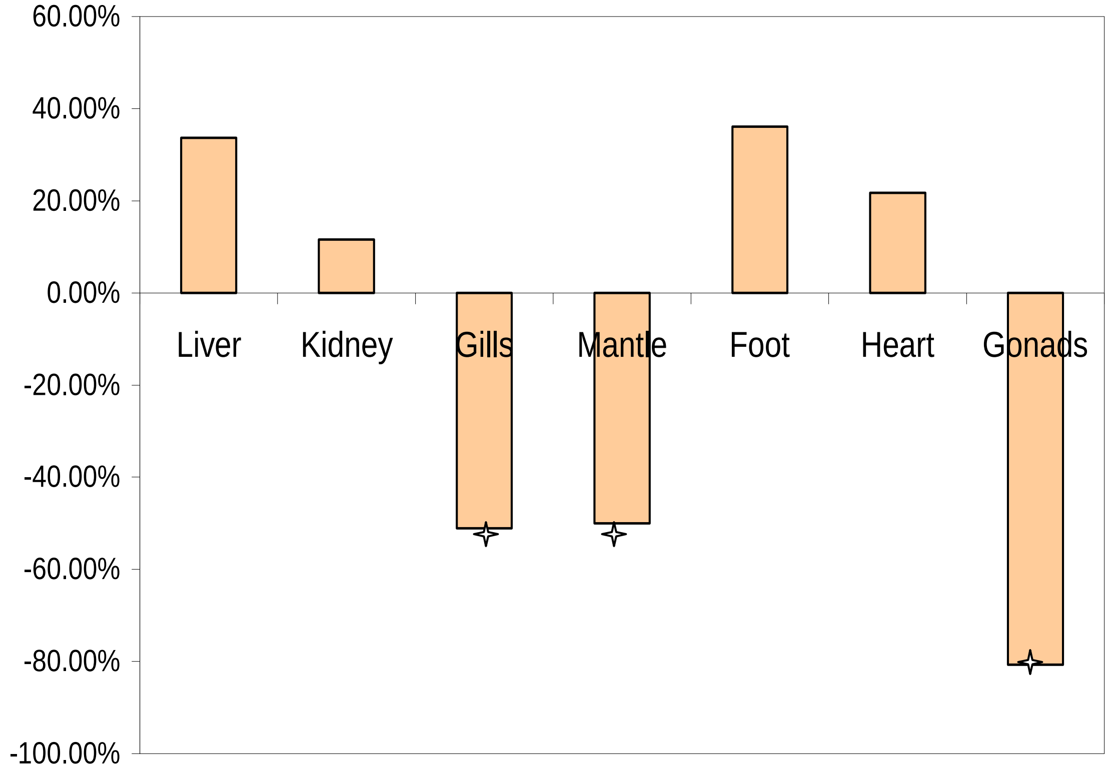

Figure 2. Relative increase (%) of wet weight of various organs of clam *A. umbonella* at the contaminated sites with respect to the reference site.

✧ **= Significantly different**

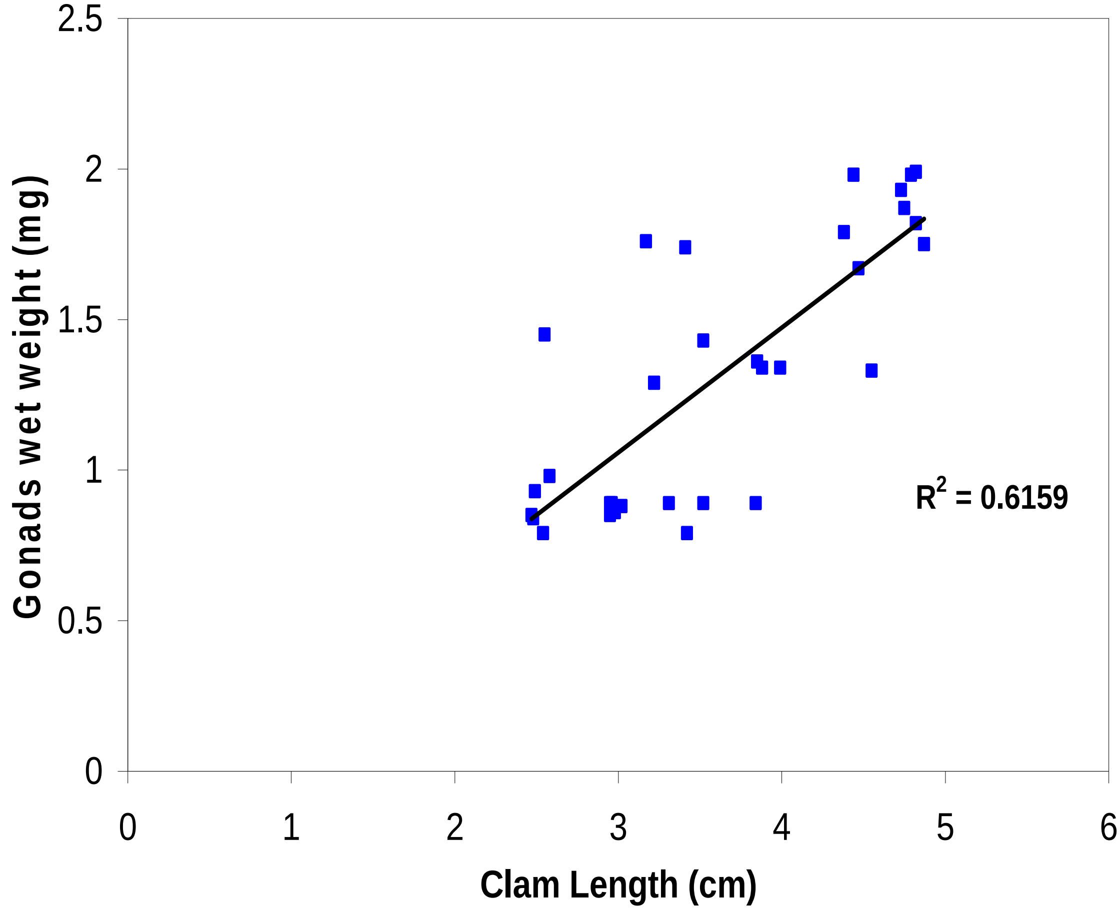

Figure 3. Correlation between clam *A. umbonella* length (cm) and gonads wet weight (mg) at the reference sites. N = 32

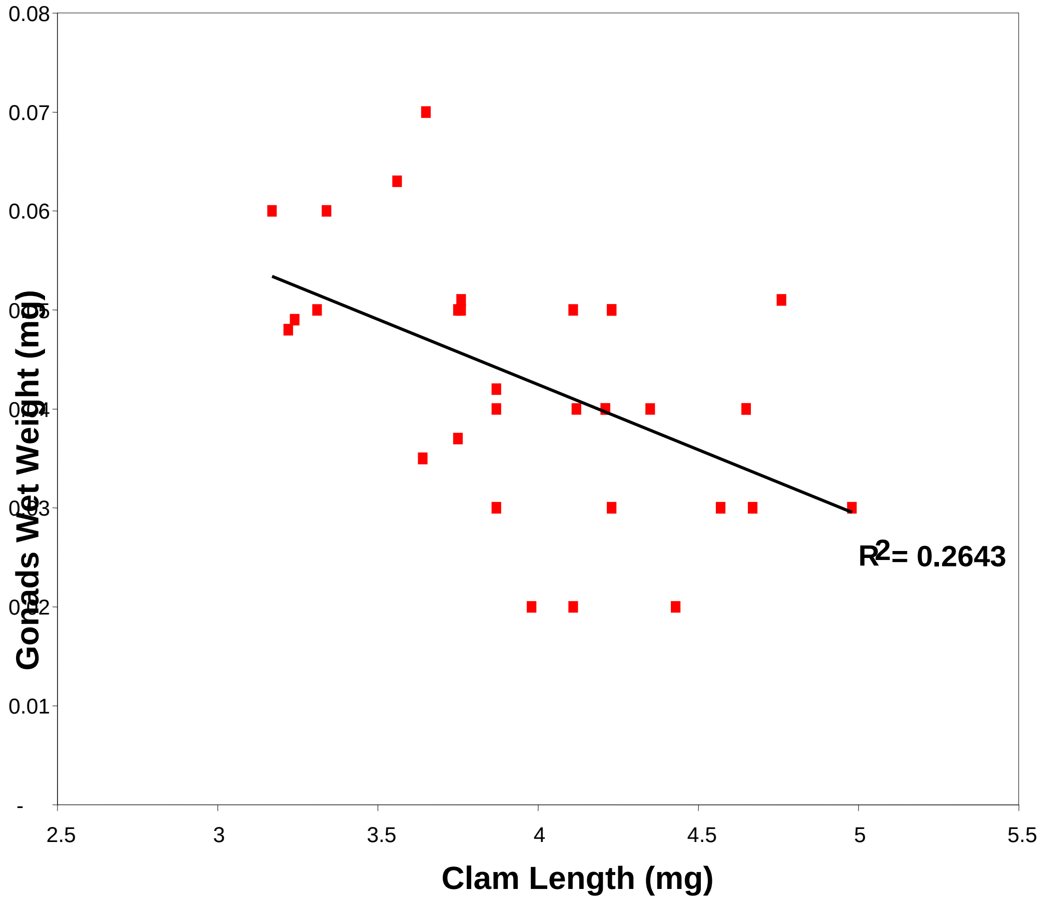

Figure 4. Correlation between clams *A. umbonella* length (cm) and gonads wet weight (mg) at the contaminated Sites.

Table 1. Mean Hg concentration in various organs of clam

Mean Hg concentration in various organs of clam *A. umbonella* , SD (standard deviation),T-test, Contamination/Reference site, RSD (Relative standard deviation), CI (Confidence interval). A, B, C = Triplicate composite sample of 33 clams each.

Metals	Sites	A	B	C	Average	SD	T-test	Contam/ Reference	Relative SD	CI
Soft Tissues	Reference	0.09	1.1	0.08	0.4	0.6	8.33E-04	7.9	138%	66%
	Contaminated	3.45	2.98	3.62	3.4	0.3			10%	38%
Foot	Reference	0.12	0.16	0.19	0.2	0.0	8.60E-04	4.2	22%	4%
	Contaminated	0.54	0.76	0.68	0.7	0.1			17%	13%
Mantle	Reference	0.05	0.09	0.06	0.1	0.0	1.56E-05	13.6	31%	2%
	Contaminated	0.89	0.98	0.85	0.9	0.1			7%	8%
Gills	Reference	0.06	0.08	0.09	0.1	0.0	2.38E-03	10.0	20%	2%
	Contaminated	0.56	0.76	0.98	0.8	0.2			27%	24%
Liver	Reference	0.08	0.09	0.01	0.1	0.0	6.02E-03	5.4	73%	5%
	Contaminated	0.32	0.23	0.42	0.3	0.1			29%	11%
Kidney	Reference	0.1	0.2	0.3	0.2	0.1	2.76E-05	9.6	50%	11%
	Contaminated	1.99	1.98	1.76	1.9	0.1			7%	15%
Gonads	Reference	0.2	0.3	0.2	0.2	0.1	2.41E-05	9.2	25%	7%
	Contaminated	2.11	1.99	2.32	2.1	0.2			8%	19%
Heart	Reference	0.1	0.21	0.22	0.2	0.1	3.61E-04	4.9	38%	8%
	Contaminated	0.98	0.88	0.76	0.9	0.1			13%	12%

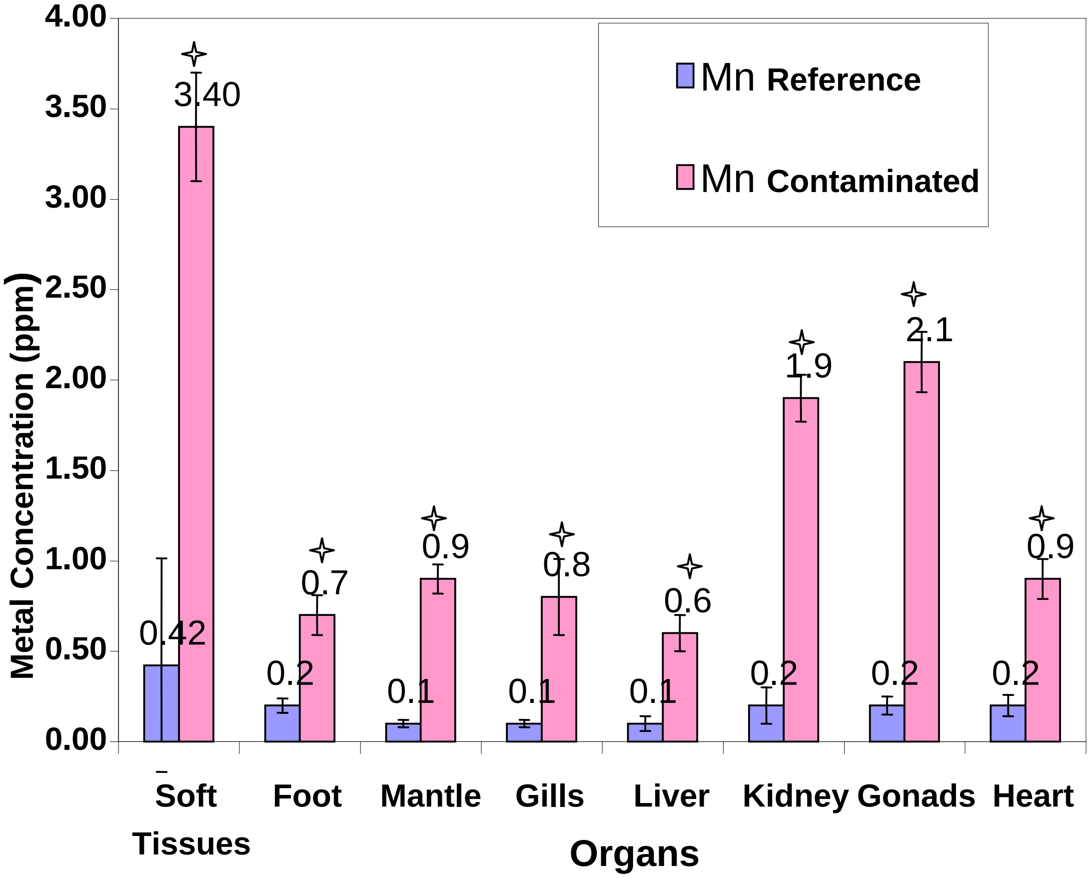

Figure 5. Mean Hg concentration in the various organs of clam *A. umbonella* between contaminated and the reference site.

Error bars show SD. ✧ Significantly different

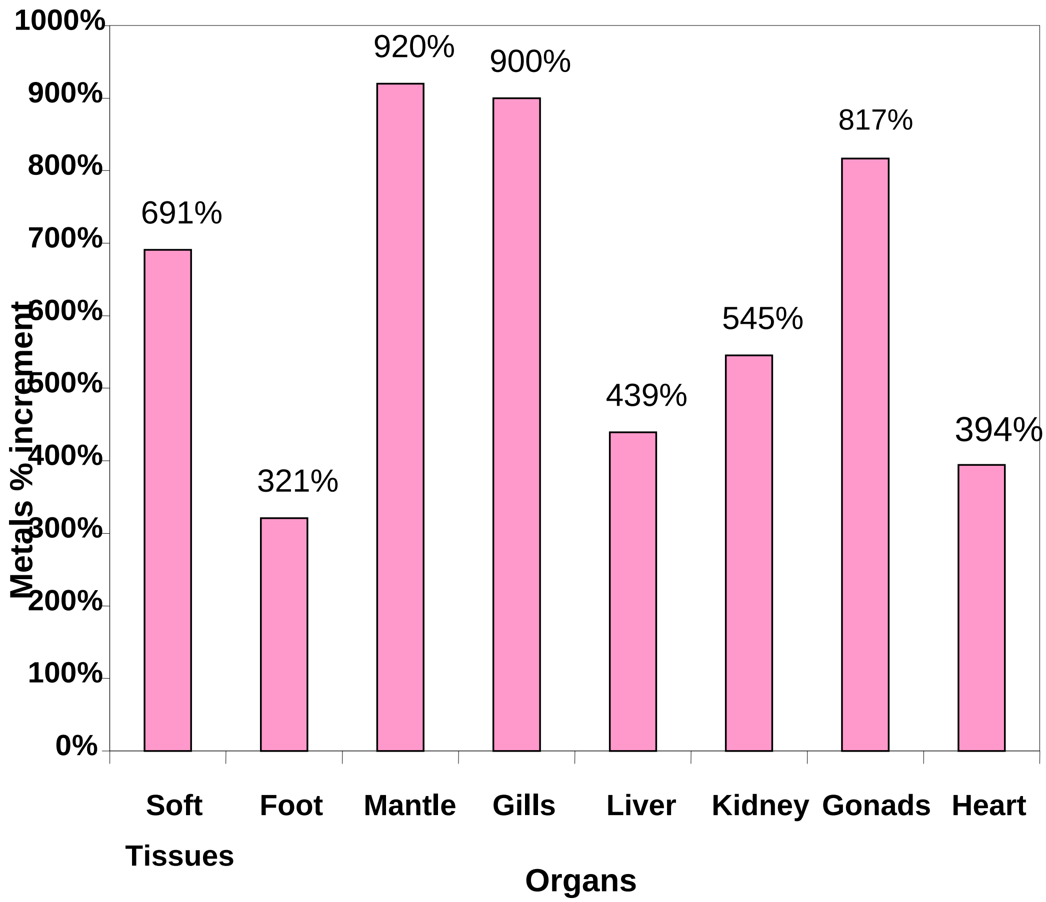

Figure 6. Incremental enrichment (%) of Hg in the various organs of clam *A. umbonella* at the contaminated sites with respect to the reference sites.

Table 2. Mean Cu concentration in various organs of clam

Mean Cu concentration in various organs of clam *A. umbonella* , SD (standard deviation),T-test, Contamination/Reference site, RSD (Relative standard deviation), A, B, C = Triplicate composite sample of 33 clams each.

Metals					Average	SD	T-test	Contami/ Referen	Relative SD	CI
	Sites	A	B	C						
Soft Tissues	Reference	3.2	3.4	3.4	3.3	0.1	6.01E-05	2.1	3%	13%
	Contaminated	6.7	7.5	6.9	7.0	0.4			6%	47%
Foot	Reference	1.7	1.7	1.5	1.6	0.1	1.21E-02	1.2	7%	13%
	Contaminated	1.9	1.9	2.1	2.0	0.1			6%	13%
Mantle	Reference	3.3	3.1	3.2	3.2	0.1	2.37E-02	4.0	3%	11%
	Contaminated	8.9	19.5	9.9	12.8	5.9			46%	662%
Gills	Reference	2.5	2.2	2.1	2.3	0.2	3.04E-04	2.0	9%	24%
	Contaminated	4.8	4.3	4.2	4.4	0.3			7%	36%
Liver	Reference	8.7	8.2	7.7	8.2	0.5	3.60E-05	2.0	6%	57%
	Contaminated	15.9	17.3	16.8	16.7	0.7			4%	80%
Kidney	Reference	3.8	2.8	2.7	3.1	0.6	9.54E-06	4.2	20%	69%
	Contaminated	13.3	12.6	12.7	12.9	0.4			3%	43%
Gonads	Reference	3.5	4.4	4	4.0	0.5	8.56E-06	3.8	11%	51%
	Contaminated	14.5	14.7	15.7	15.0	0.6			4%	73%
Heart	Reference	7.8	7.2	7.5	7.5	0.3	4.45E-04	1.3	4%	34%
	Contaminated	9.8	9.3	9.8	9.6	0.3			3%	33%

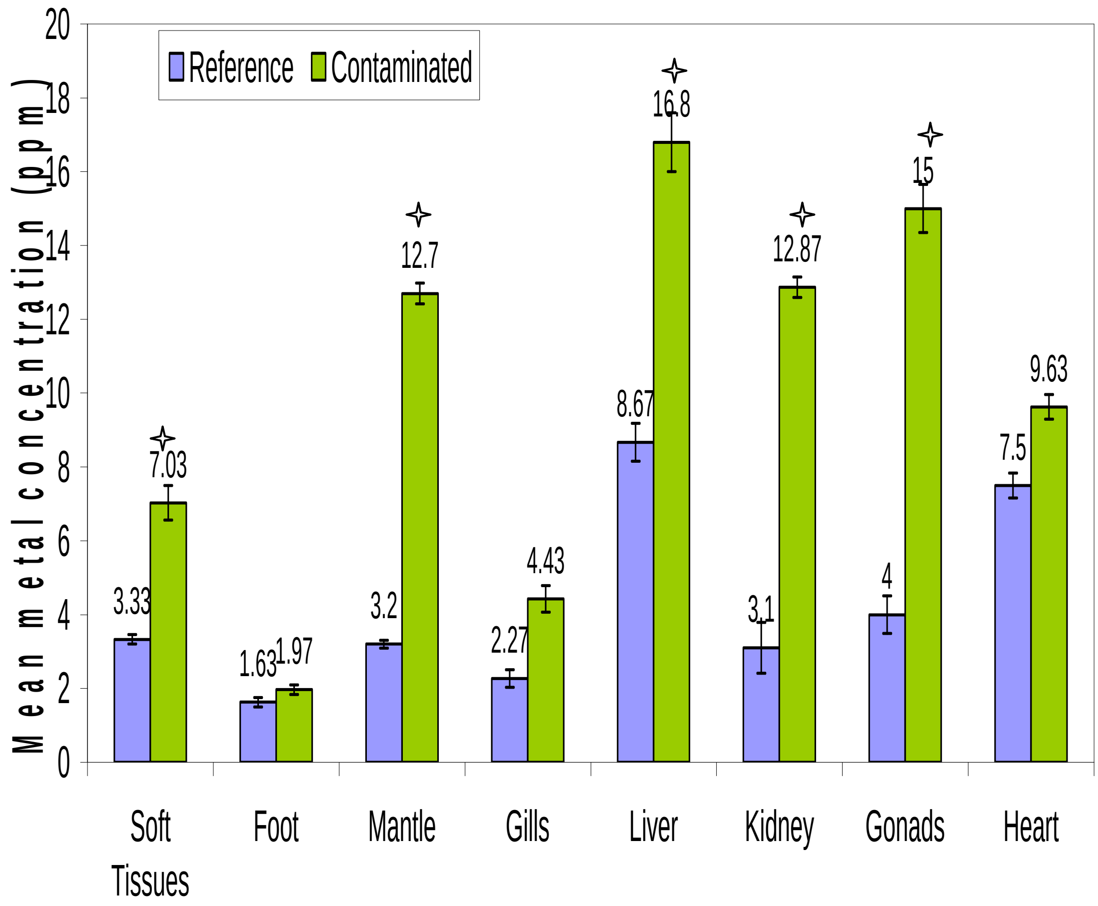

Figure 7. Mean Cu concentration in the various organs of clam *A. umbonella* between contaminated and the reference site.

Error bars show SD. ✧=Significantly different.

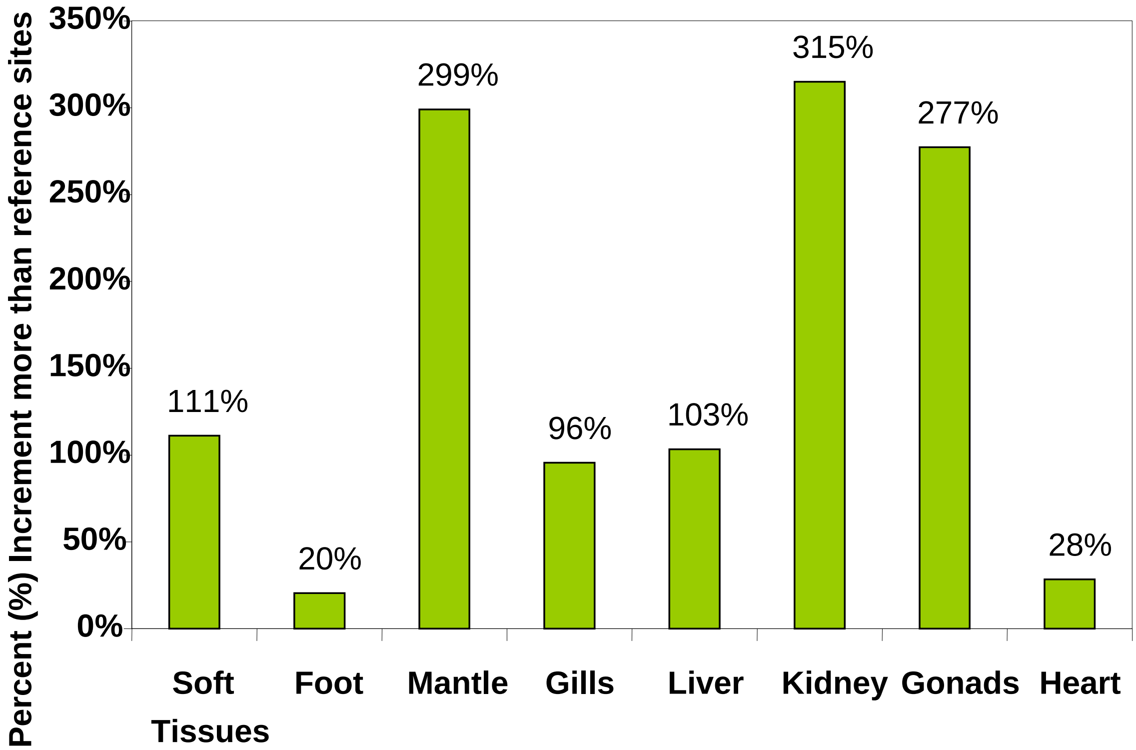

Figure 8. Incremental enrichment (%) of Cu in the various organs of clam *A. umbonella* at contaminated sites with respect to the reference site.

Table 3. Mean Cr concentration in various organs of clam

Mean Cr concentration in various organs of clam *A. umbonella* , SD (standard deviation),T-test, Contamination/Reference site, RSD (Relative standard deviation), A, B, C = Triplicate composite sample of 33 clams each.

Metals	Sites	A	B	C	Average	SD	T-test	Contam/ Reference	Relative SD	CI
Soft Tissues	Reference	3.1	2.9	3.1	3.0	0.1	1.80E-06	2.0	4%	13%
	Contaminated	6.1	6.2	6.3	6.2	0.1			2%	11%
Foot	Reference	10.5	9.8	9.3	9.9	0.6	1.61E-05	1.8	6%	68%
	Contaminated	17.7	17.9	17.4	17.7	0.3			1%	28%
Mantle	Reference	2.7	2.3	2.6	2.5	0.2	2.34E-07	4.9	8%	24%
	Contaminated	12.7	12.5	12.3	12.5	0.2			2%	23%
Gills	Reference	1.9	1.8	2.1	1.9	0.2	5.26E-06	3.9	8%	17%
	Contaminated	7.7	7.8	7.2	7.6	0.3			4%	36%
Liver	Reference	2.3	2.2	2.1	2.2	0.1	5.12E-06	3.4	5%	11%
	Contaminated	7.7	7.8	7.2	7.6	0.3			4%	36%
Kidney	Reference	4.5	4.8	4.4	4.6	0.2	2.15E-05	1.6	5%	24%
	Contaminated	7.6	7.3	7.4	7.4	0.2			2%	17%
Gonads	Reference	4.1	4.2	4.4	4.2	0.2	1.97E-04	1.2	4%	17%
	Contaminated	5.3	5.2	5.3	5.3	0.1			1%	7%
Heart	Reference	1.5	1.2	1.3	1.3	0.2	1.64E-04	2.1	11%	17%
	Contaminated	2.9	2.8	2.6	2.8	0.2			6%	17%

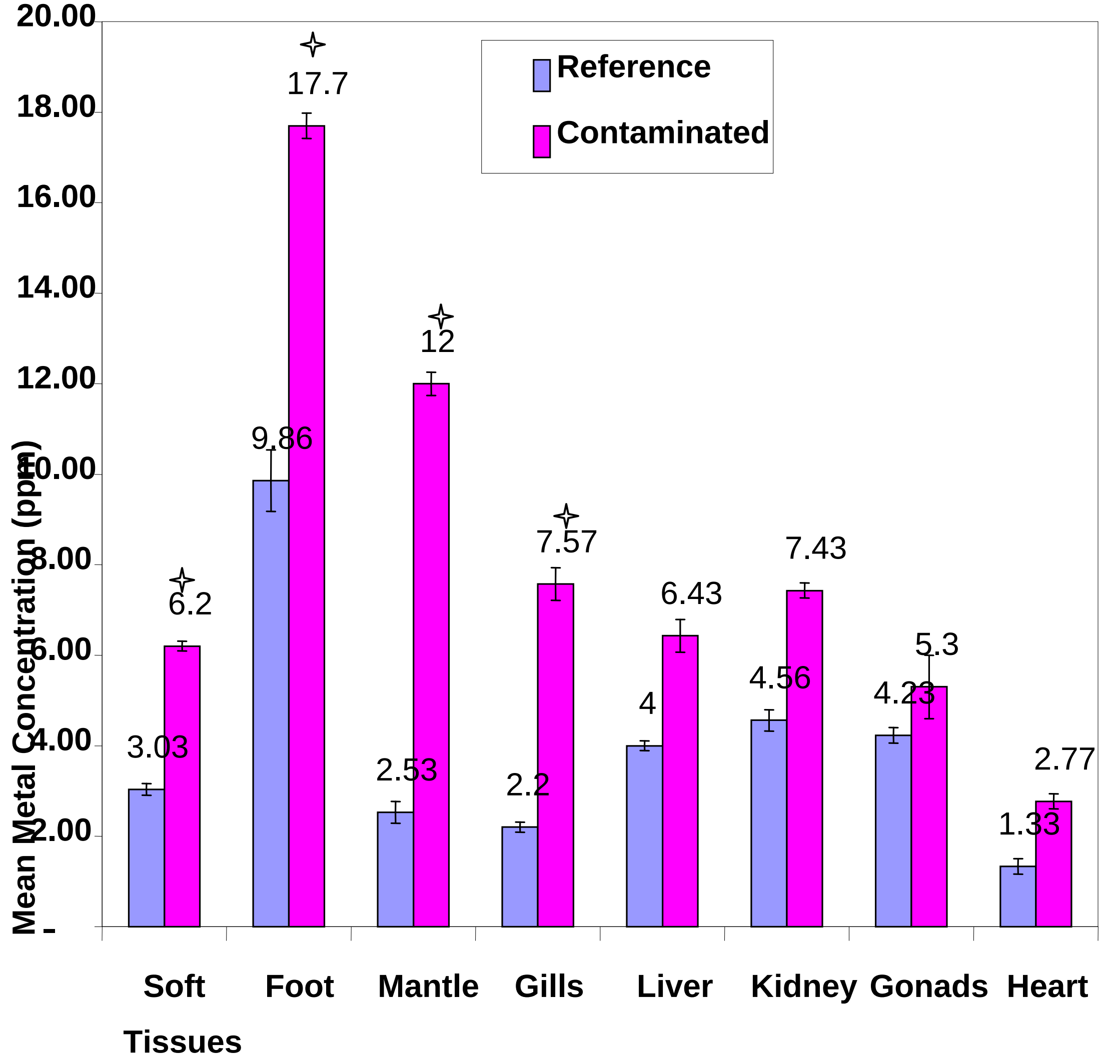

Figure 9. Mean Cr concentration in the various organs of clam *A. umbonella* between contaminated and reference site.

Error bars show SD. ✧= Significantly different.

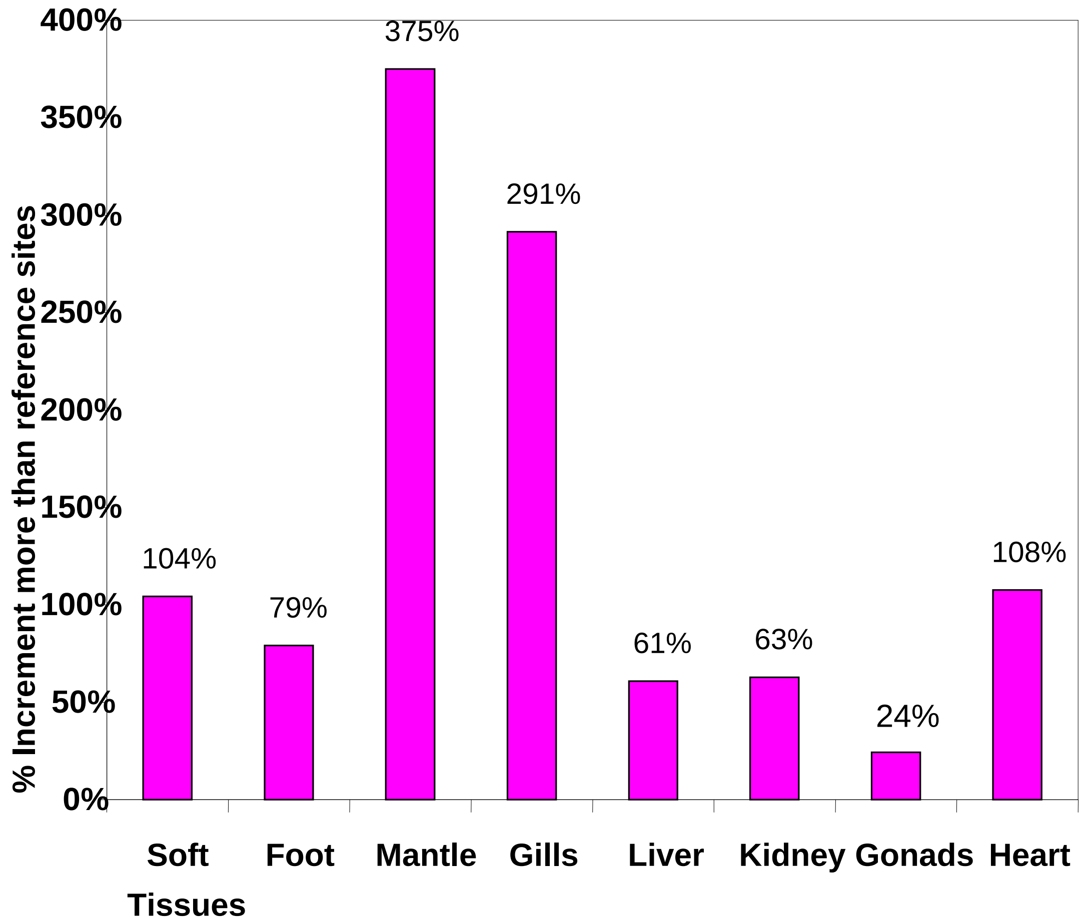

Figure 10. Incremental enrichment (%) of Cr in the various organs of clam *A. umbonella* at contaminated sites with respect to the reference site.

Table 4. Mean Ni concentration in various organs of clam *A. umbonella* , SD (standard deviation),T-test, Contamination/Reference site, RSD (Relative standard deviation), A, B, C = Triplicate composite sample of 33 clams each.

Metals	Sites	A	B	C	Average	SD	T-test	Contam/R efferenc	Relative SD	CI
Soft Tissues	Reference	2.5	2.4	2.4	2.4	0.1	3.34E-06	2.6	2%	7%
	Contaminated	6.2	6.5	6.1	6.3	0.2			3%	24%
Foot	Reference	0.8	0.81	0.8	0.8	0.0	1.15E-03	2.0	1%	1%
	Contaminated	1.7	1.4	1.8	1.6	0.2			13%	24%
Mantle	Reference	0.5	0.5	0.6	0.5	0.1	1.56E-03	1.6	11%	7%
	Contaminated	0.9	0.8	0.8	0.8	0.1			7%	7%
Gills	Reference	0.4	0.4	0.3	0.4	0.1	5.29E-04	2.1	16%	7%
	Contaminated	0.8	0.7	0.8	0.8	0.1			8%	7%
Liver	Reference	0.6	0.7	0.7	0.7	0.1	2.92E-04	1.7	9%	7%
	Contaminated	1.1	1.2	1.1	1.1	0.1			5%	7%
Kidney	Reference	1.8	1.4	1.4	1.5	0.2	3.35E-05	2.9	15%	26%
	Contaminated	4.3	4.6	4.3	4.4	0.2			4%	20%
Gonads	Reference	1.5	1.6	1.6	1.6	0.1	1.89E-03	2.8	4%	7%
	Contaminated	3.56	4.26	5.13	4.3	0.8			18%	89%
Heart	Reference	0.22	0.21	0.21	0.2	0.0	2.93E-04	7.2	3%	1%
	Contaminated	1.8	1.4	1.4	1.5	0.2			15%	26%

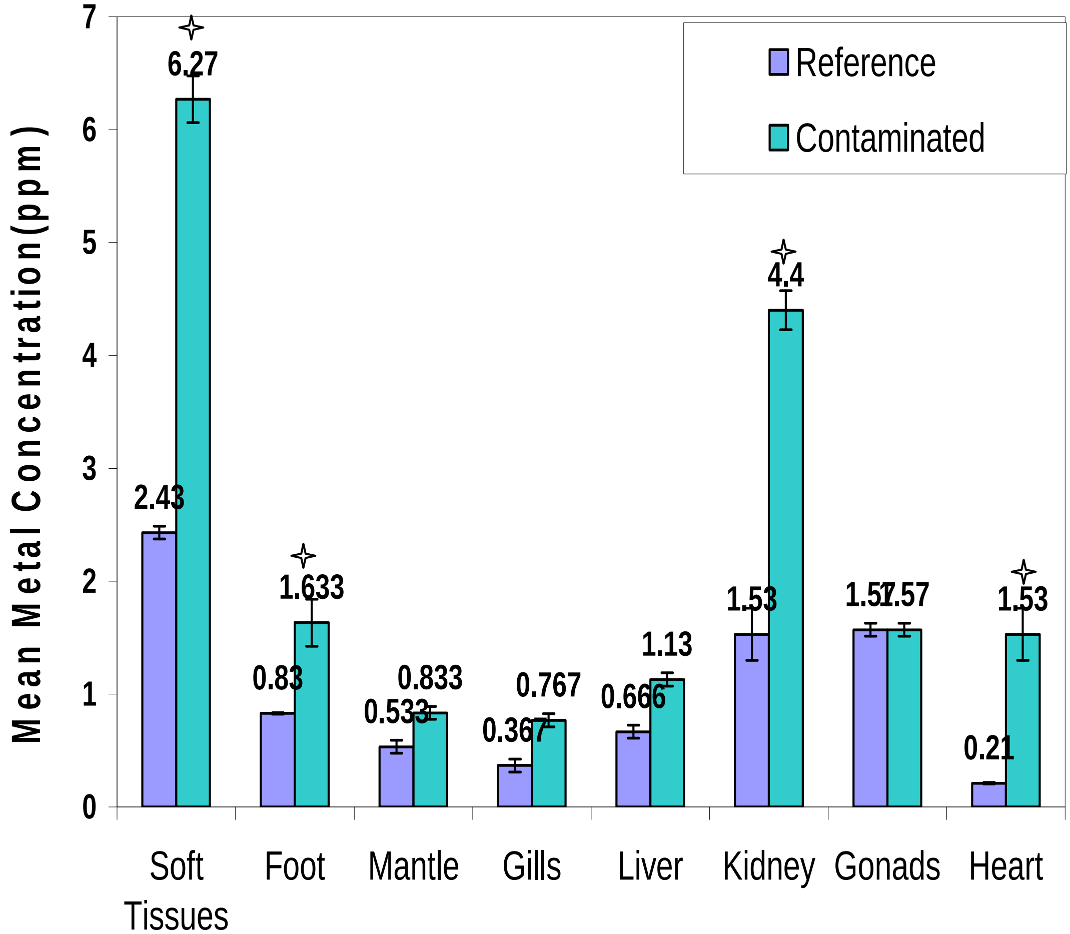

Figure 11. Mean Ni concentration in the various organs of clam *A. umbonella* between contaminated and the reference site.

Error bars show SD.✧ =Statistically different.

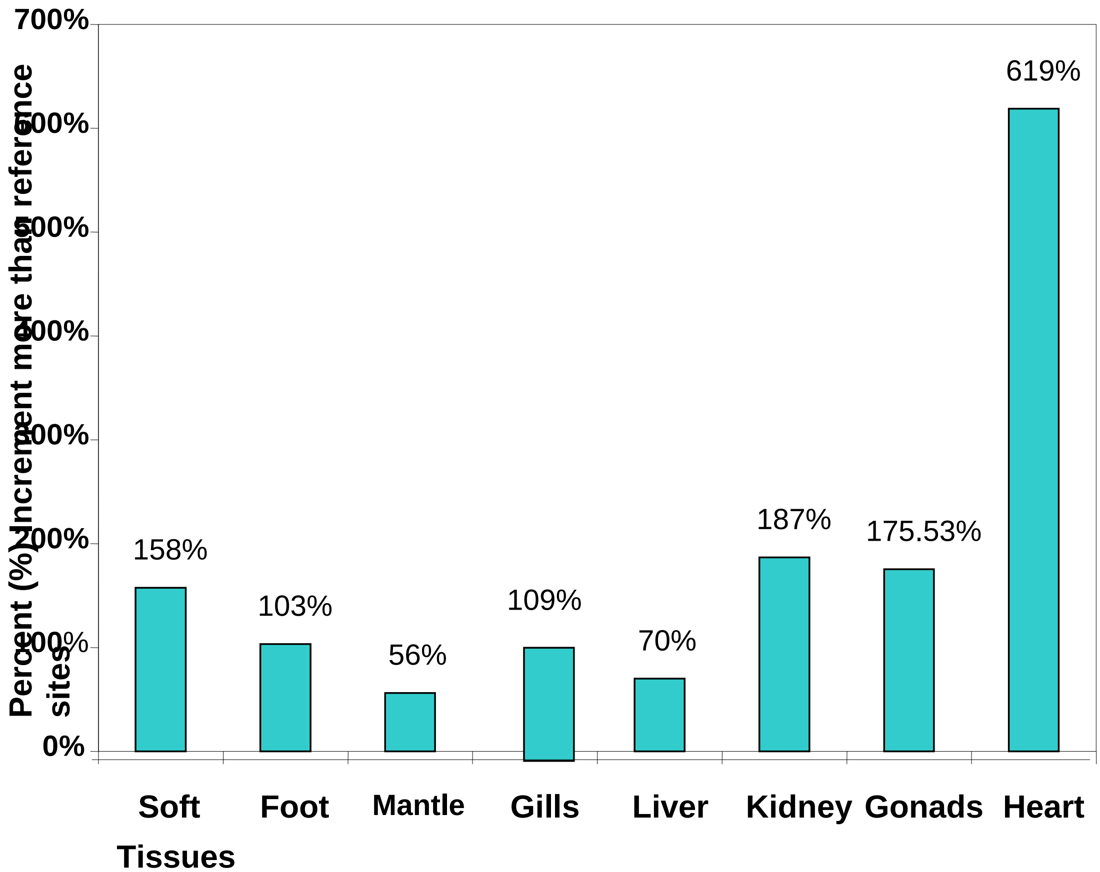

Figure 12. Relative enrichment (%) of Ni in the various organs of clam *A. umbonella* at contaminated sites with respect to the reference site.

Table 4. Mean Zn concentration in various organs of clam

Mean Zn concentration in various organs of clam *A. umbonella* , SD (standard deviation),T-test, Contamination/Reference site, RSD (Relative standard deviation), A, B, C = Triplicate composite sample of 33 clams each.

Metals					Average	SD	T-test	Contam/ Referen	Relative SD	CI
	Sites	A	B	C						
Soft Tissues	Reference	81.1	82.1	87.2	83.5	3.3	5.35E-04	1.2	4%	370%
	Contaminated	103.2	99.8	103.1	102.0	1.9			2%	219%
Foot	Reference	49.4	53.3	47.8	50.2	2.8	3.35E-03	1.2	6%	320%
	Contaminated	58.2	59.8	58.8	58.9	0.8			1%	91%
Mantle	Reference	41	38.7	39	39.6	1.3	1.65E-02	1.1	3%	141%
	Contaminated	41.6	44.1	42.8	42.8	1.3			3%	141%
Gills	Reference	49.7	47.5	50.1	49.1	1.4	5.91E-05	1.3	3%	158%
	Contaminated	64.7	64.7	62.9	64.1	1.0			2%	118%
Liver	Reference	91.8	98.4	92.1	94.1	3.7	1.64E-01	1.0	4%	422%
	Contaminated	94.4	97.5	98.8	96.9	2.3			2%	256%
Kidney	Reference	178	189.5	172	179.8	8.9	1.57E-02	1.1	5%	1006%
	Contaminated	195.7	201.9	195.8	197.8	3.6			2%	402%
Gonads	Reference	56.5	56.5	55.5	56.2	0.6	3.75E-05	1.2	1%	65%
	Contaminated	67.6	68.5	69.9	68.7	1.2			2%	131%
Heart	Reference	131.3	134.4	132.97	132.9	1.6	1.52E-03	1.1	1%	176%
	Contaminated	143.1	147.1	142.2	144.1	2.6			2%	295%

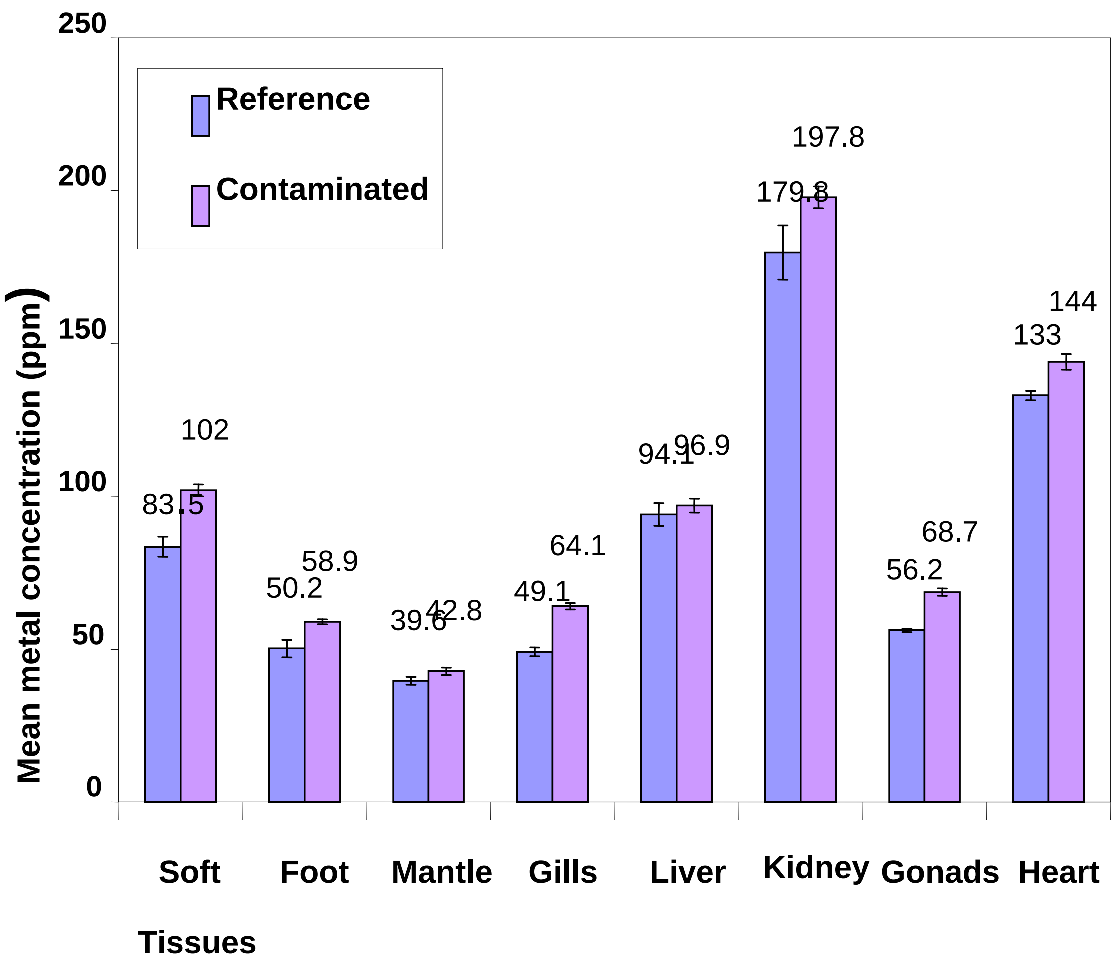

Figure 13. Mean Zn concentration in the various organs of clam *A. umbonella* at contaminated and the reference site.

Error bars show SD.

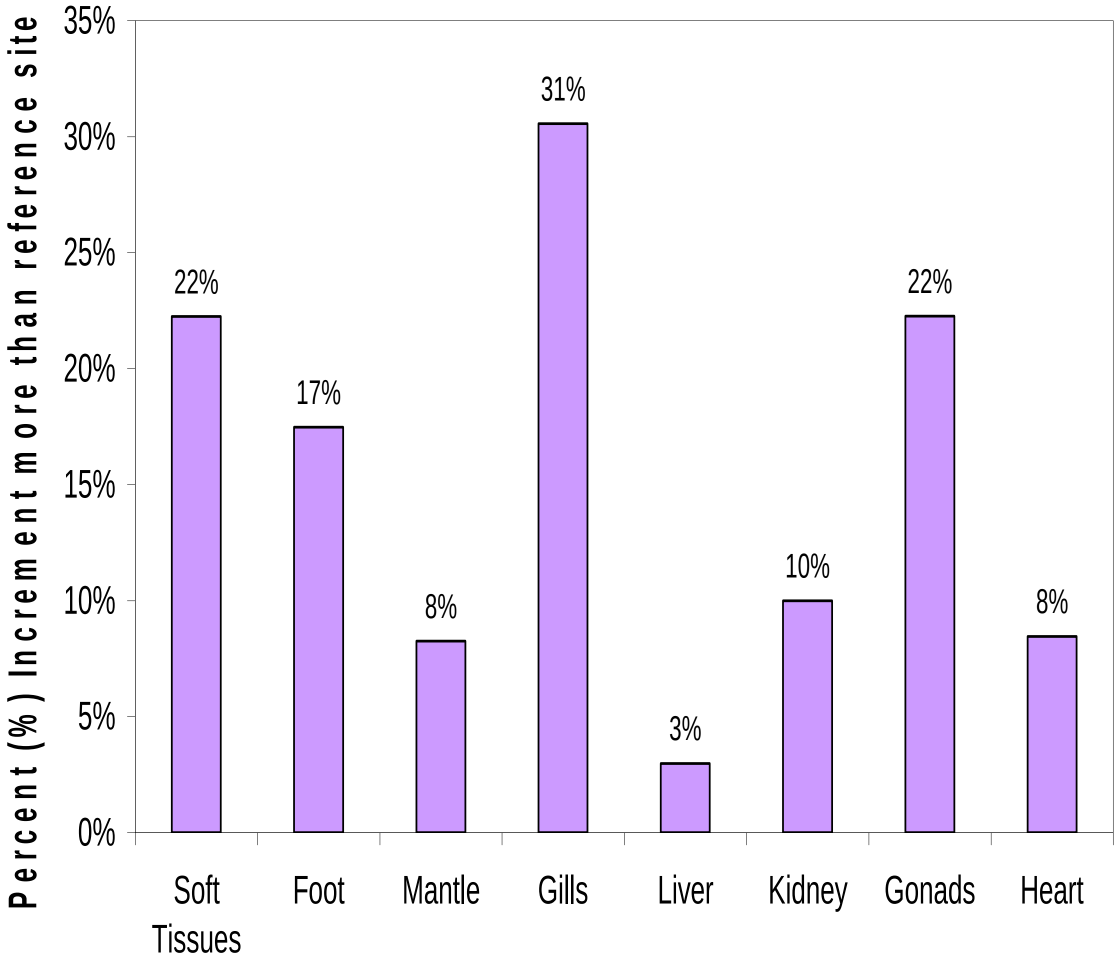

Figure 14. Incremental enrichment (%) of Zn in the various organs of clam *A. umbonella* at contaminated sites with respect to the reference site.

Table 5. Mean Fe concentration in various organs of clam

Mean Fe **concentration in various organs of clam *A. umbonella* , SD (standard deviation),T-test, Contamination/Reference site, RSD (Relative standard deviation), A, B, C = Triplicate composite sample of 33 clams each.**

Metals					Average	SD	T-test	Contam/ Referen	Relative SD	CI
	Sites	A	B	C						
Soft Tissues	Reference	0.51	0.51	0.53	0.5	0.0	2.41E-02	1.2	2%	1%
	Contaminated	0.65	0.55	0.6	0.6	0.0			8%	6%
Foot	Reference	0.3	0.3	0.29	0.3	0.0	1.12E-04	1.4	2%	1%
	Contaminated	0.42	0.42	0.45	0.4	0.0			4%	2%
Mantle	Reference	0.27	0.25	0.26	0.3	0.0	2.90E-06	2.0	4%	1%
	Contaminated	0.52	0.53	0.51	0.5	0.0			2%	1%
Gills	Reference	0.82	0.82	0.83	0.8	0.0	1.40E-06	1.2	1%	1%
	Contaminated	1.01	1	1	1.0	0.0			1%	1%
Liver	Reference	0.45	0.38	0.54	0.5	0.1	7.70E-05	2.4	18%	9%
	Contaminated	1.1	1.11	1.1	1.1	0.0			1%	1%
Kidney	Reference	3.5	3.7	3.2	3.5	0.3	2.06E-05	1.8	7%	28%
	Contaminated	6.4	6.4	6.3	6.4	0.1			1%	7%
Gonads	Reference	1.01	1.02	1.06	1.0	0.0	3.38E-03	4.5	3%	3%
	Contaminated	3.33	4.76	5.72	4.6	1.2			26%	136%
Heart	Reference	1.5	1.4	1.1	1.3	0.2	3.70E-02	1.2	16%	24%
	Contaminated	1.6	1.6	1.7	1.6	0.1			4%	7%

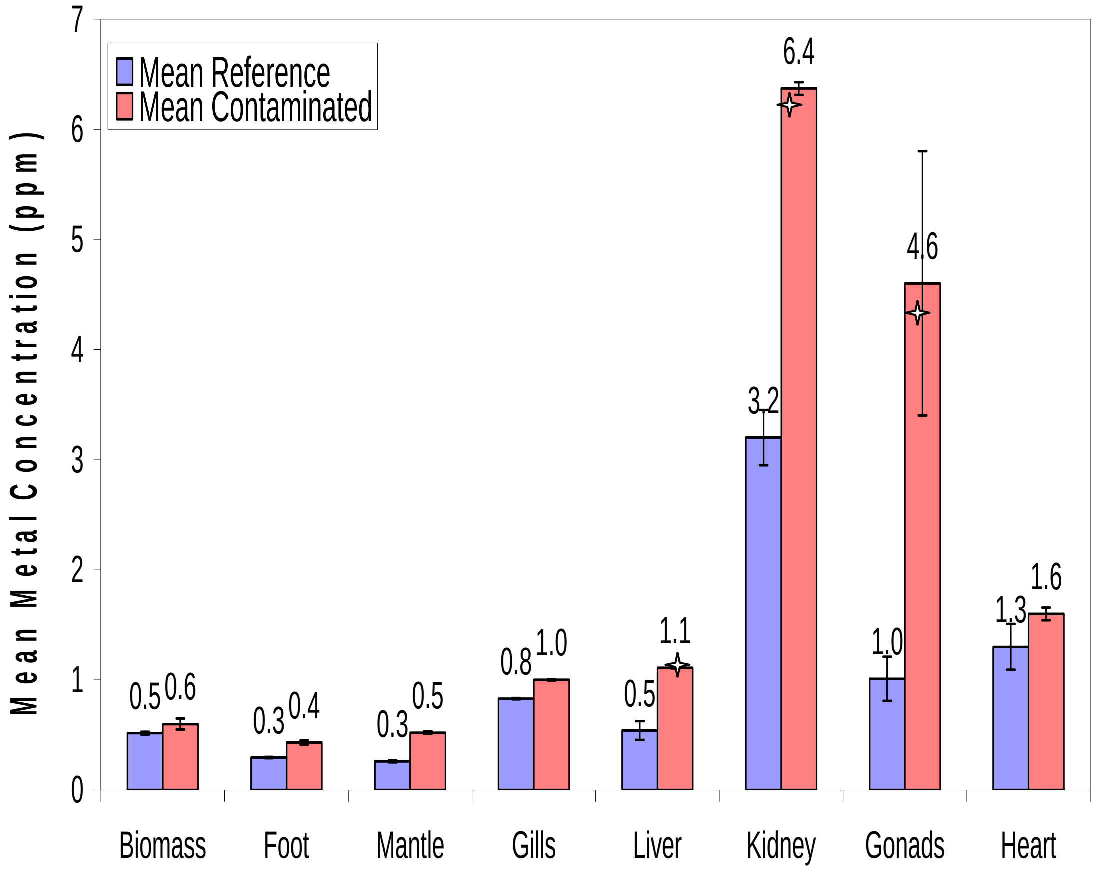

Figure 15. Mean Fe concentration in the various organs of clam *A. umbonella* at contaminated and the reference site.

Error bars show SD. ✧=Significantly different.

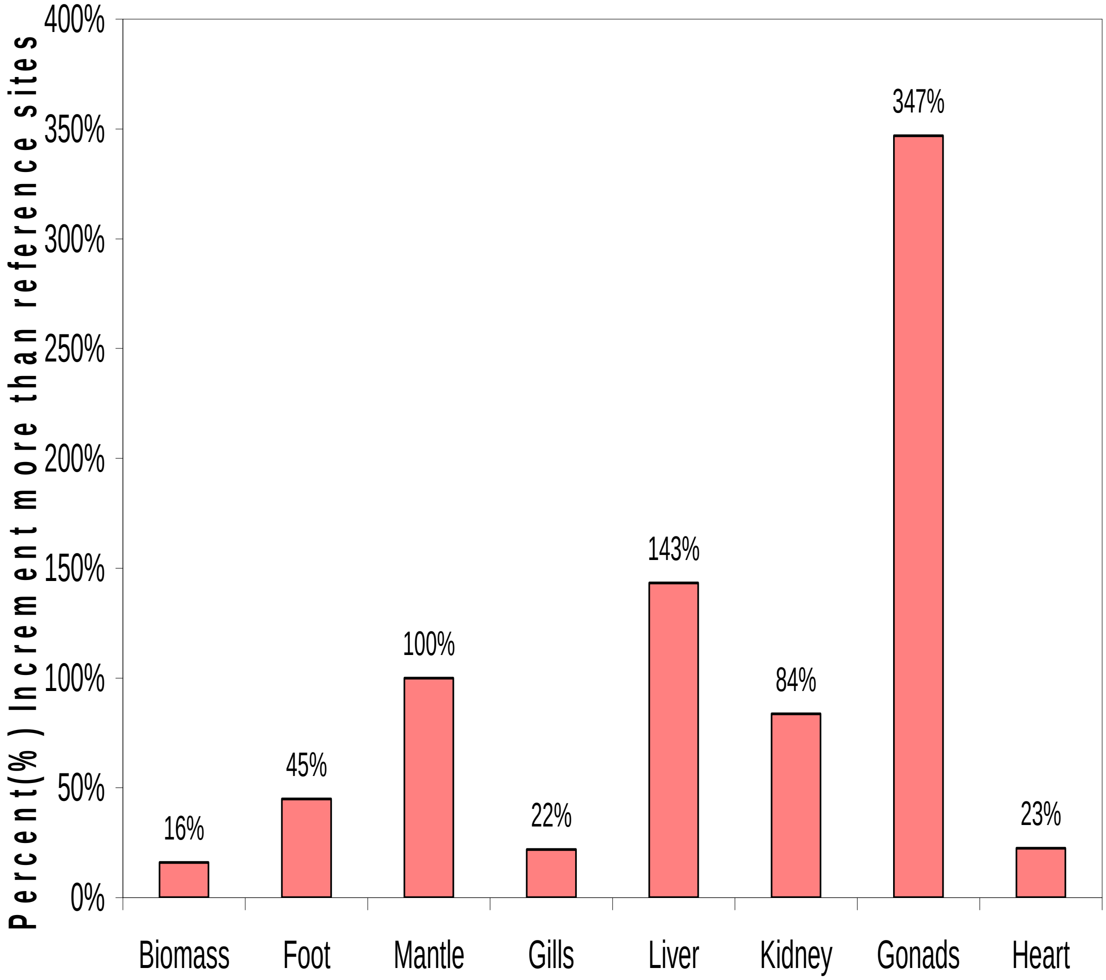

Figure 16. Incremental enrichment (%) of Fe in the various organs of clam *A. umbonella* at the contaminated sites with respect to the reference site.

Table 6. Mean Pb concentration in various organs of clam

Mean Pb concentration in various organs of clam *A. umbonella* , SD (standard deviation),T-test, Contamination/Reference site, RSD (Relative standard deviation), A, B, C = Triplicate composite sample of 33 clams each.

				Average	SD	T-test	Contam/ Referen	Relative SD	CI
Sites	A	B	C						
Reference	0.21	0.23	0.24	0.2	0.0	1.57E-06	2.5	7%	2%
Contaminated	0.57	0.58	0.58	0.6	0.0			1%	1%
Reference	0.11	0.11	0.12	0.1	0.0	7.37E-06	2.0	5%	1%
Contaminated	0.227	0.22	0.23	0.2	0.0			2%	1%
Reference	0.4	0.4	0.3	0.4	0.1	1.56E-03	1.8	16%	7%
Contaminated	0.7	0.7	0.6	0.7	0.1			9%	7%
Reference	0.8	0.7	0.8	0.8	0.1	7.26E-05	1.9	8%	7%
Contaminated	1.4	1.4	1.5	1.4	0.1			4%	7%
Reference	1.1	1.1	0.9	1.0	0.1	1.00E-06	4.5	11%	13%
Contaminated	4.8	4.7	4.6	4.7	0.1			2%	11%
Reference	31.4	32.7	29.8	31.3	1.5	2.14E-06	3.4	5%	164%
Contaminated	110	107	103	106.7	3.5			3%	397%
Reference	2.1	2.4	2.1	2.2	0.2	1.60E-07	4.5	8%	20%
Contaminated	9.9	9.8	9.7	9.8	0.1			1%	11%
Reference	9.8	9.2	8.8	9.3	0.5	3.27E-07	2.9	5%	57%
Contaminated	27.1	26.9	27.4	27.1	0.3			1%	28%

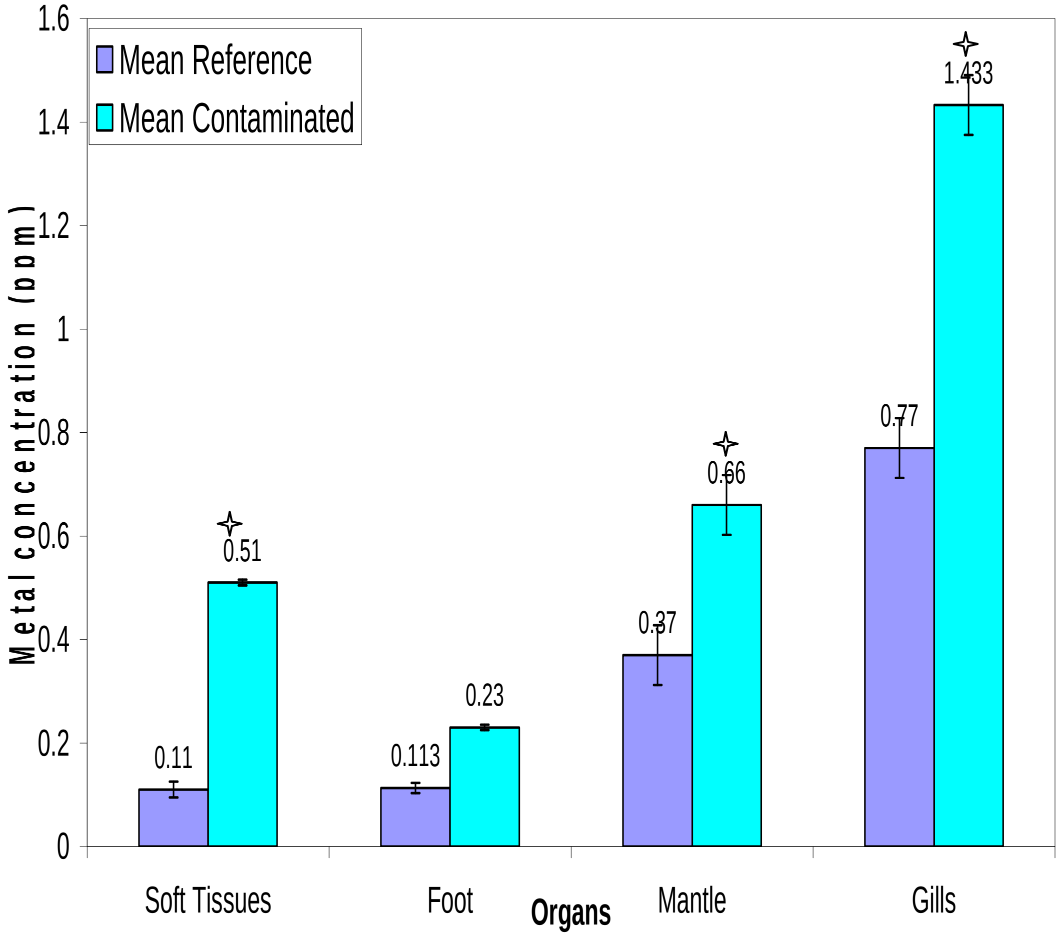

Figure 17. Mean Pb concentration in the various organs of clam *A. umbonella* at contaminated and the reference site.

Error bars show SD. ✧ = Significantly different

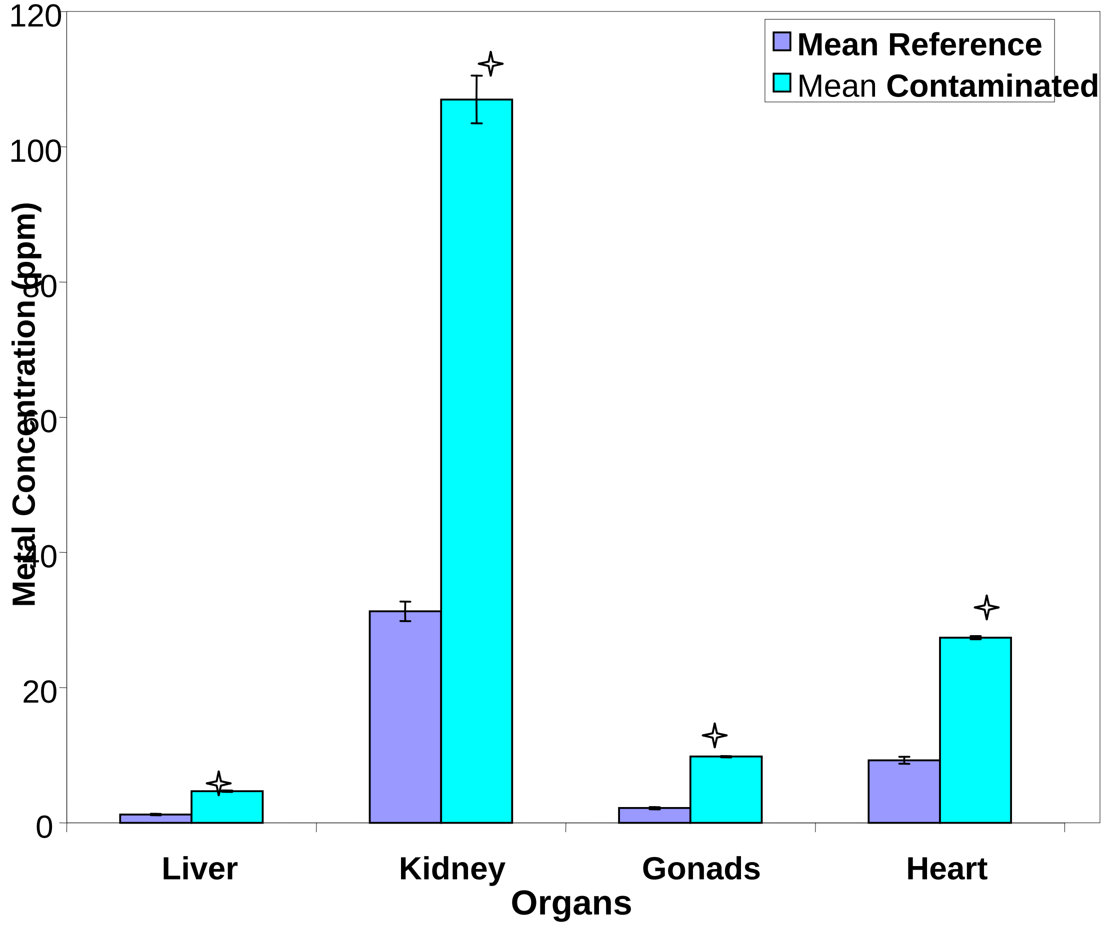

Figure 18. Mean Pb concentration in the various organs of clam *A. umbonella* at the contaminated and the reference site.

Error bars show SD. ✧ = Significantly different.

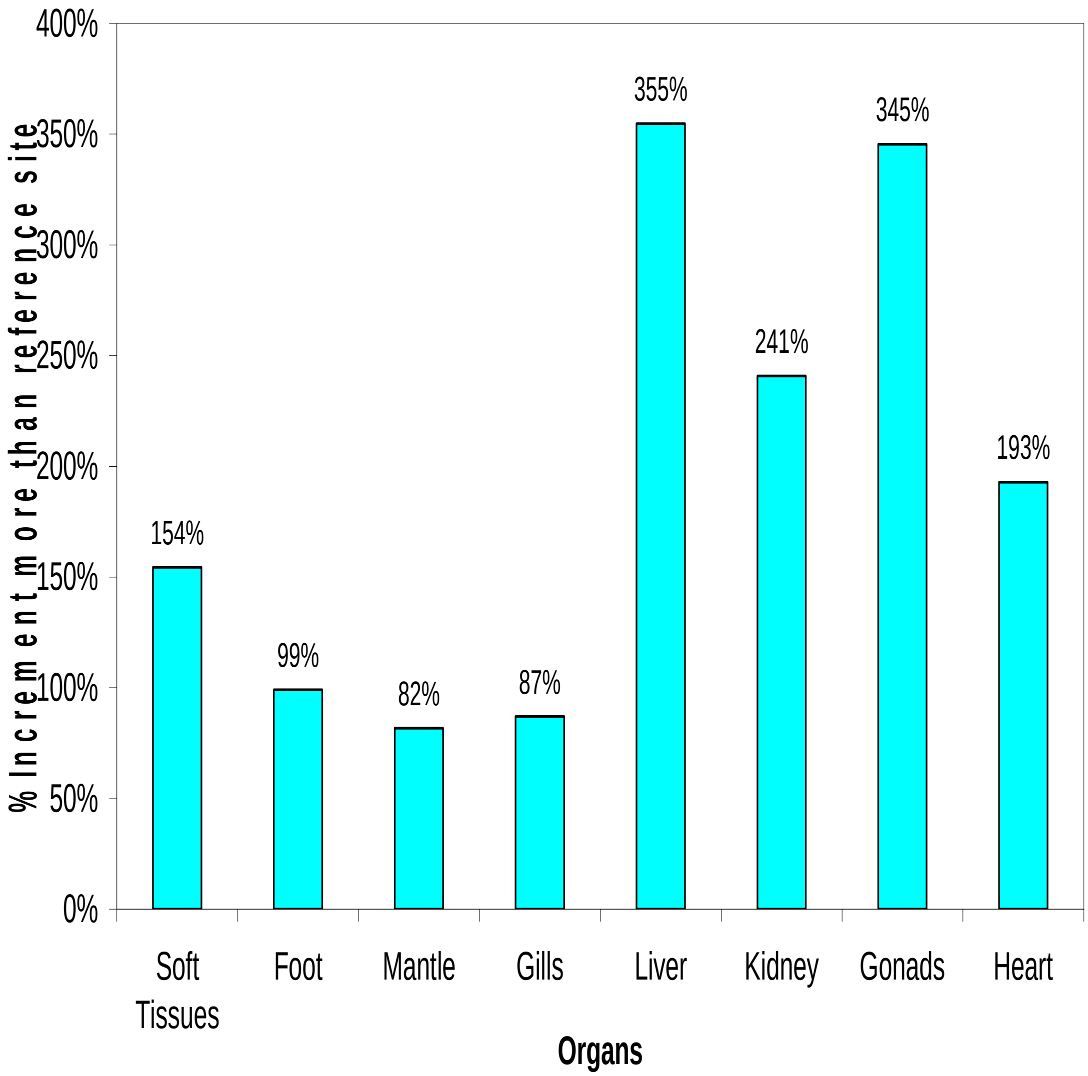

Figure 19. Incremental enrichment (%) of Pb in the various organs of clam *A. umbonella* at the contaminated sites with respect to the reference site.

Table 7. Mean Cd concentration in various organs of clam

Mean Cd concentration in various organs of clam *A. umbonella* , SD (standard deviation),T-test, Contamination/Reference site, RSD (Relative standard deviation), A, B, C = Triplicate composite sample of 33 clams each.

Metals					Average	SD	T-test	Contam/ Reference	Relative SD	CI
	Sites	A	B	C						
Soft Tissues	Reference	0.28	0.25	0.23	0.3	0.0	3.90E-05	7.2	10%	3%
	Contaminated	1.67	1.79	1.99	1.8	0.2			9%	18%
Foot	Reference	0.65	0.54	0.53	0.6	0.1	1.17E-03	1.6	12%	8%
	Contaminated	0.98	0.88	0.89	0.9	0.1			6%	6%
Mantle	Reference	0.21	0.22	0.27	0.2	0.0	3.32E-04	3.8	14%	4%
	Contaminated	0.98	0.92	0.76	0.9	0.1			13%	13%
Gills	Reference	0.21	0.23	0.24	0.2	0.0	1.45E-05	3.9	7%	2%
	Contaminated	0.94	0.84	0.87	0.9	0.1			6%	6%
Liver	Reference	0.23	0.25	0.24	0.2	0.0	3.73E-04	1.5	4%	1%
	Contaminated	0.34	0.36	0.38	0.4	0.0			6%	2%
Kidney	Reference	31.4	32.7	29.8	31.3	1.5	1.24E-07	3.1	5%	164%
	Contaminated	97.5	98.2	96.7	97.5	0.8			1%	85%
Gonads	Reference	0.21	0.22	0.23	0.2	0.0	6.00E-04	4.7	5%	1%
	Contaminated	0.98	0.89	1.22	1.0	0.2			17%	19%
Heart	Reference	1.8	1.7	1.7	1.7	0.1	2.70E-04	2.1	3%	7%
	Contaminated	3.33	3.67	3.98	3.7	0.3			9%	37%

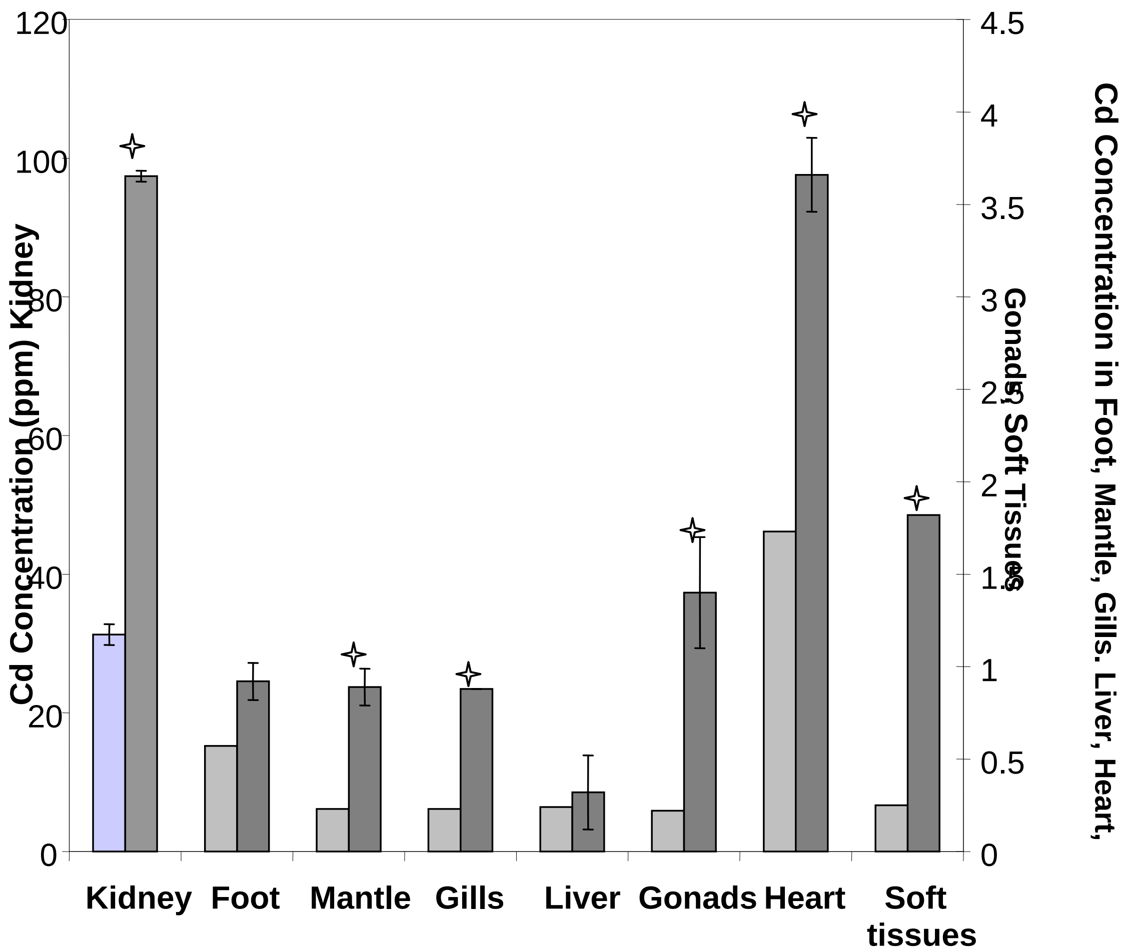

Figure 20. Mean Cd concentration in the various organs of clam *A. umbonella* at the contaminated and the reference site.
Error bars show SD. ✧ = Significantly different.
Right axis: Kidney
Left axis: Foot, Mantle, Gills, Liver, Gonads, Heart, Soft tissues.

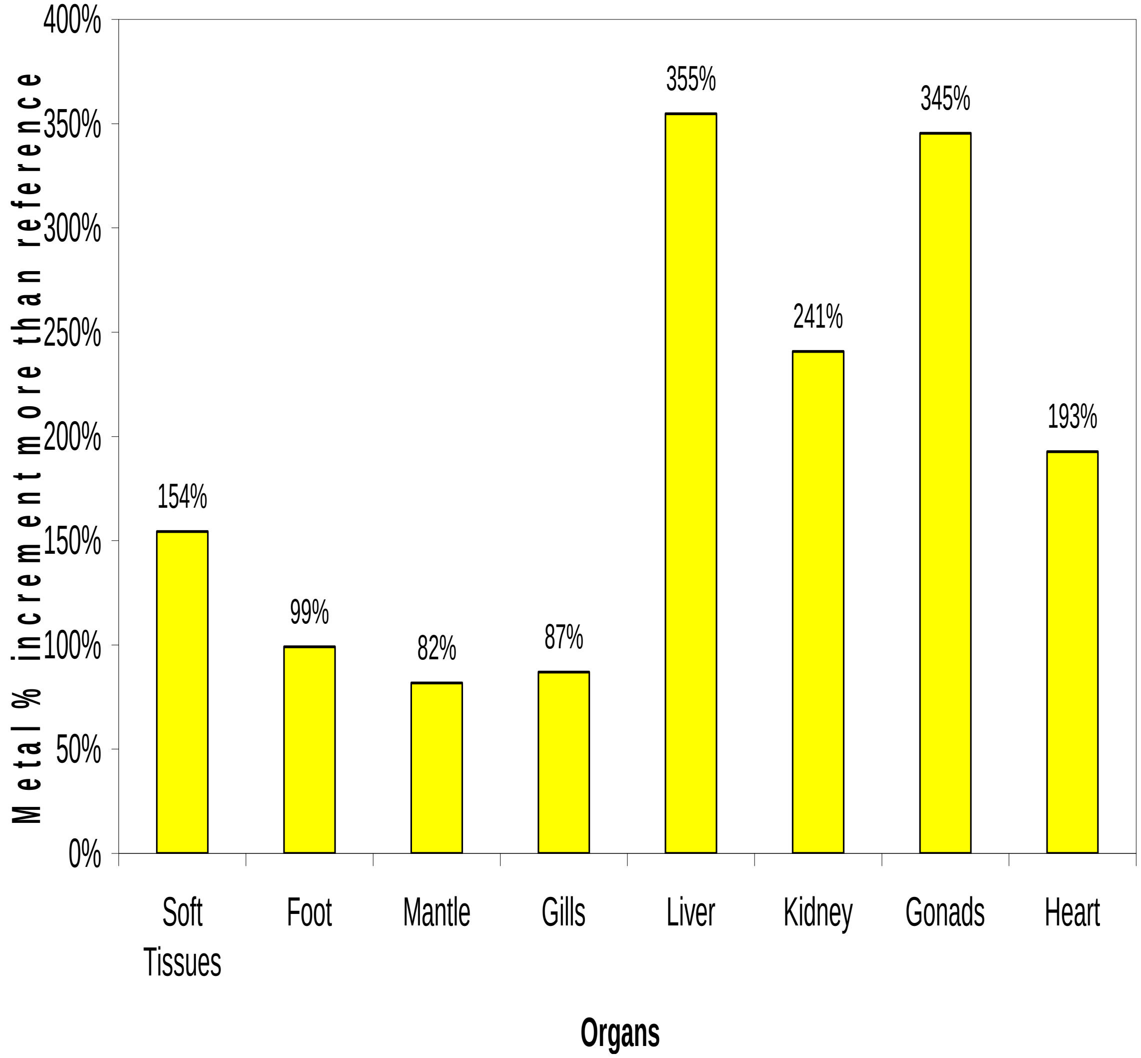

Figure 21. Incremental enrichment (%) of Cd in the various organs of clam *A. umbonella* at the contaminated sites with respect to the reference site.

Table 8. Mean V concentration in various organs of clam

Mean V concentration in various organs of clam *A. umbonella* , SD (standard deviation),T-test, Contamination/Reference site, RSD (Relative standard deviation) A, B, C = Triplicate composite sample of 33 clams each.

Metals	Sites	A	B	C	Average	SD	T-test	Contam/ Reference	Relative SD	CI
Soft Tissues	Reference	0.4	0.5	0.4	0.4	0.1	4.69E-02	1.2	13%	7%
	Contaminated	0.51	0.51	0.5	0.5	0.0			1%	1%
Foot	Reference	0.2	0.2	0.3	0.2	0.1	1.74E-04	3.3	25%	7%
	Contaminated	0.7	0.8	0.8	0.8	0.1			8%	7%
Mantle	Reference	0.3	0.2	0.2	0.2	0.1	2.66E-03	2.6	25%	7%
	Contaminated	0.5	0.7	0.6	0.6	0.1			17%	11%
Gills	Reference	0.4	0.4	0.3	0.4	0.1	2.15E-03	4.0	16%	7%
	Contaminated	1.1	1.6	1.7	1.5	0.3			22%	36%
Liver	Reference	0.31	0.31	0.32	0.3	0.0	3.05E-06	4.4	2%	1%
	Contaminated	1.4	1.4	1.3	1.4	0.1			4%	7%
Kidney	Reference	7.9	7.8	7.9	7.9	0.1	9.52E-07	3.4	1%	7%
	Contaminated	27.8	26.5	26.4	26.9	0.8			3%	88%
Gonads	Reference	0.1	0.2	0.2	0.2	0.1	5.99E-05	5.2	35%	7%
	Contaminated	0.9	0.8	0.9	0.9	0.1			7%	7%
Heart	Reference	0.35	0.3	0.3	0.3	0.0	3.71E-03	1.3	9%	3%
	Contaminated	0.42	0.4	0.4	0.4	0.0			3%	1%

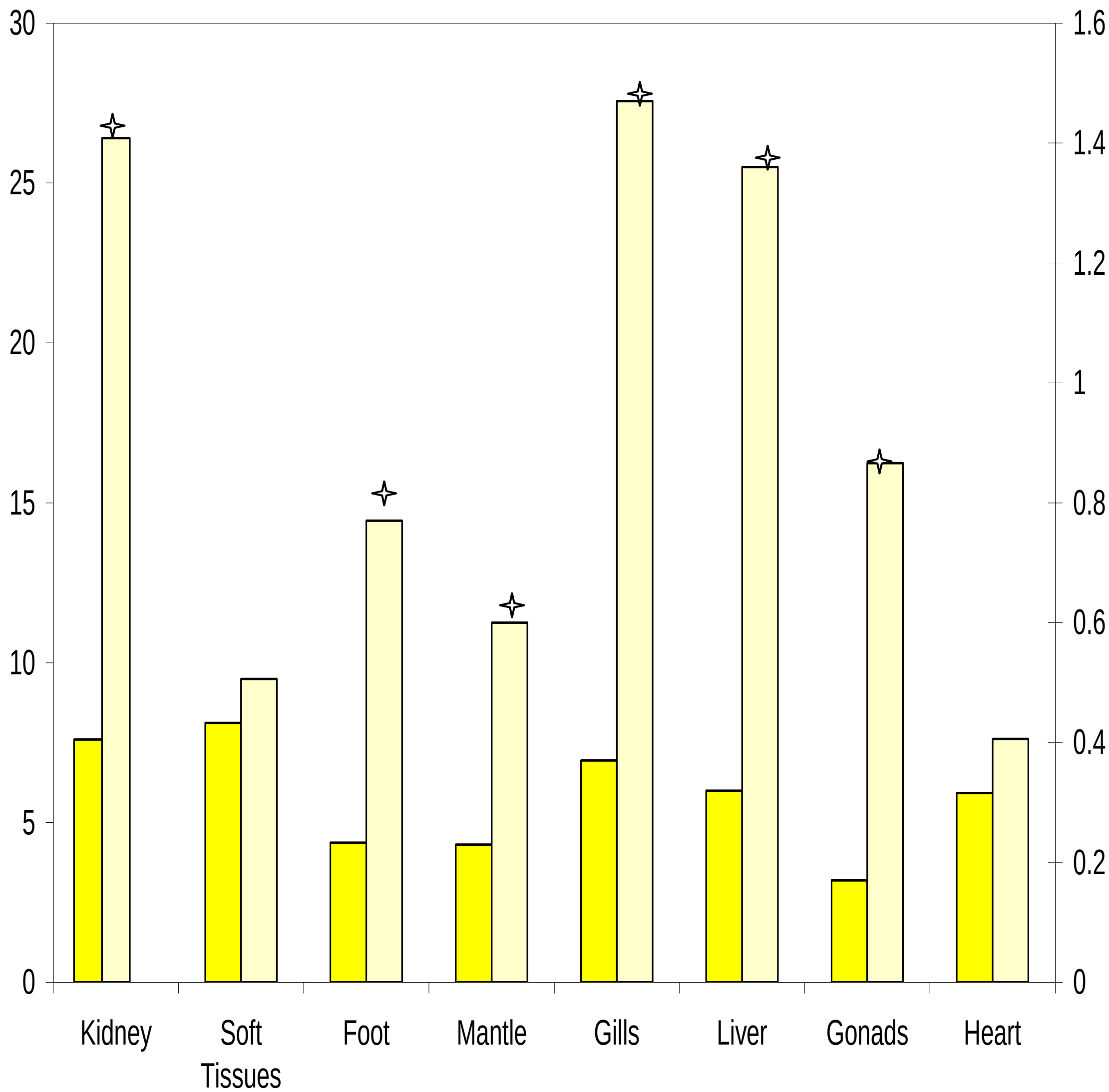

Figure 22. Mean V concentration in the various organs of clam *A. umbonella.*

Right axis: Kidney
Left axis: Soft tissues, Foot, Mantle, Gills, Liver, Gonads, Heart at contaminated and the reference site.
Error bars show SD. ✧ = Significantly different

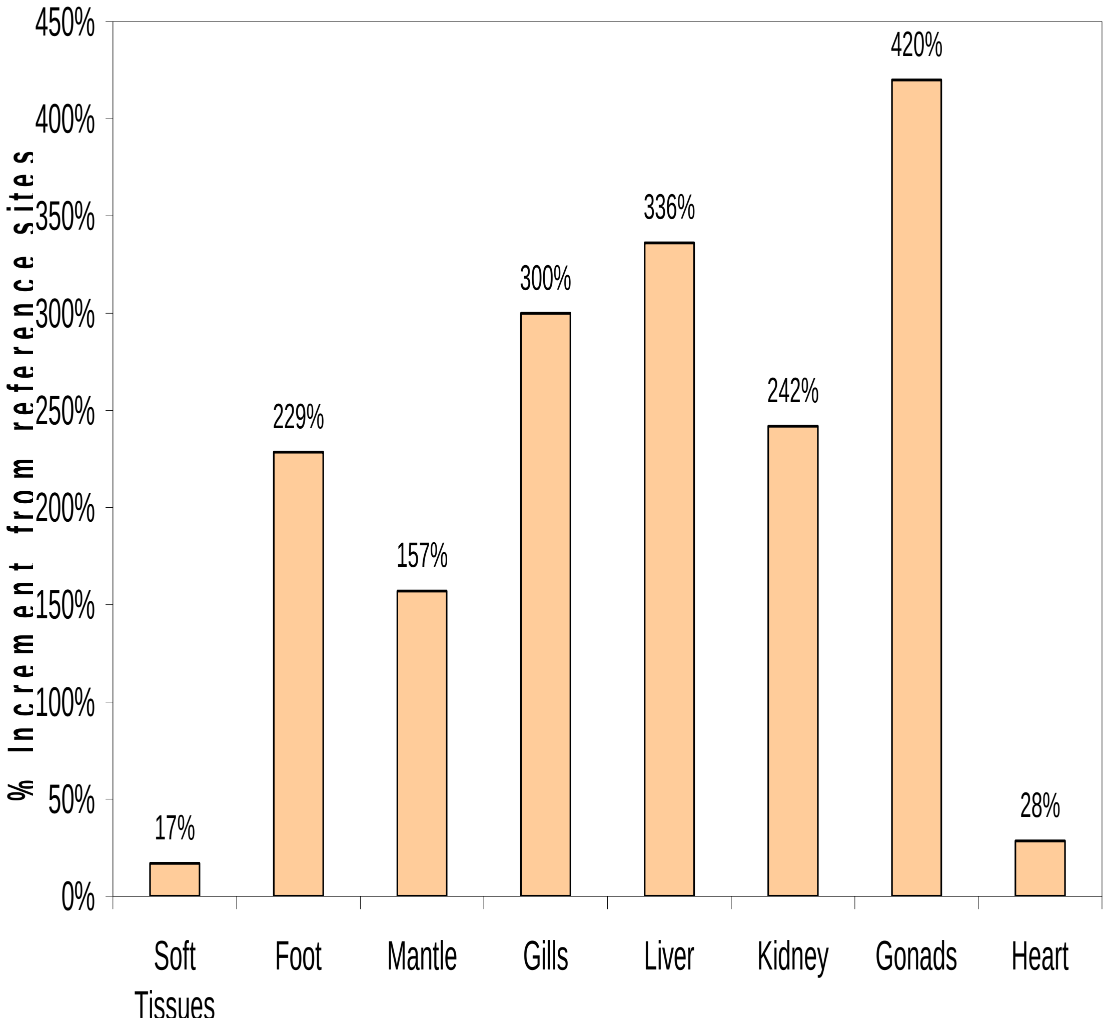

Figure 23. Incremental enrichment (%) of V in the various organs of clam *A. umbonella* at the contaminated sites with respect to the reference sites.

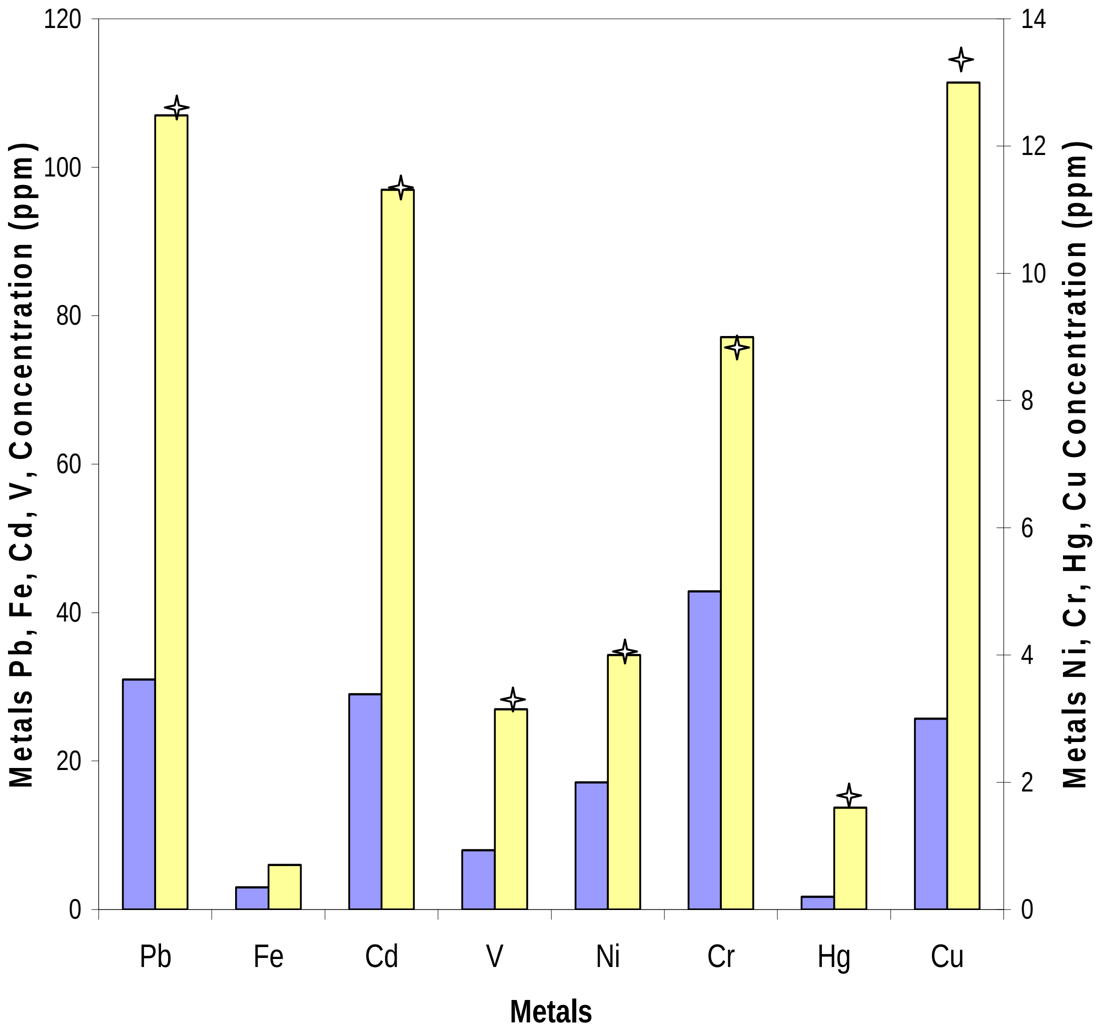

Figure 24. Mean metal concentration (ppm) in clam *A. umbonella* kidney with respect to the reference site.

✦ = Significantly different.

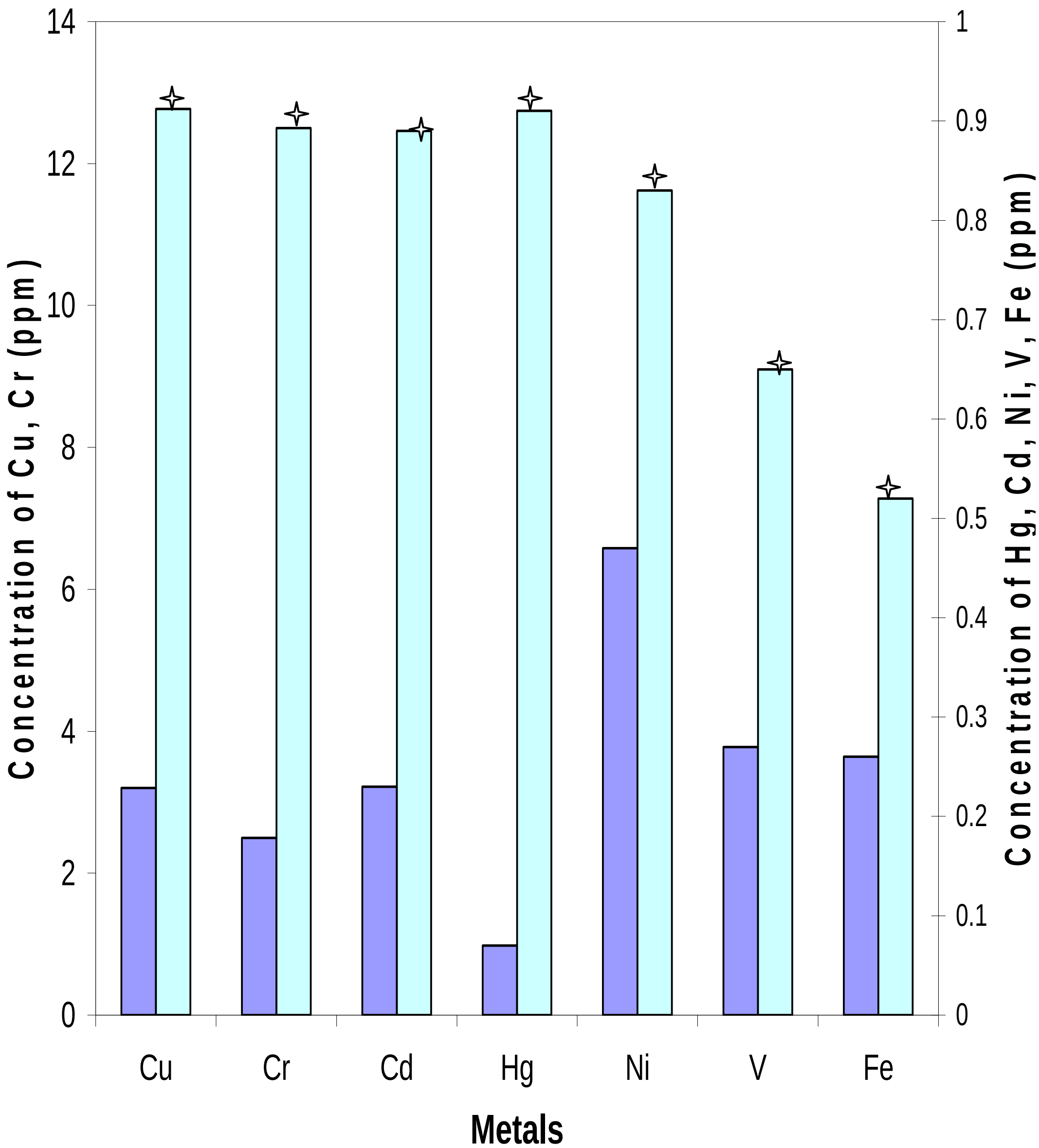

Figure 25. Mean metal concentrations (ppm) in clam's mantles at the contaminated and the reference site.
Left axis: Cu, Cr
Right axis: Cd, Hg, Ni, V, Fe.
✧ = Significantly different.

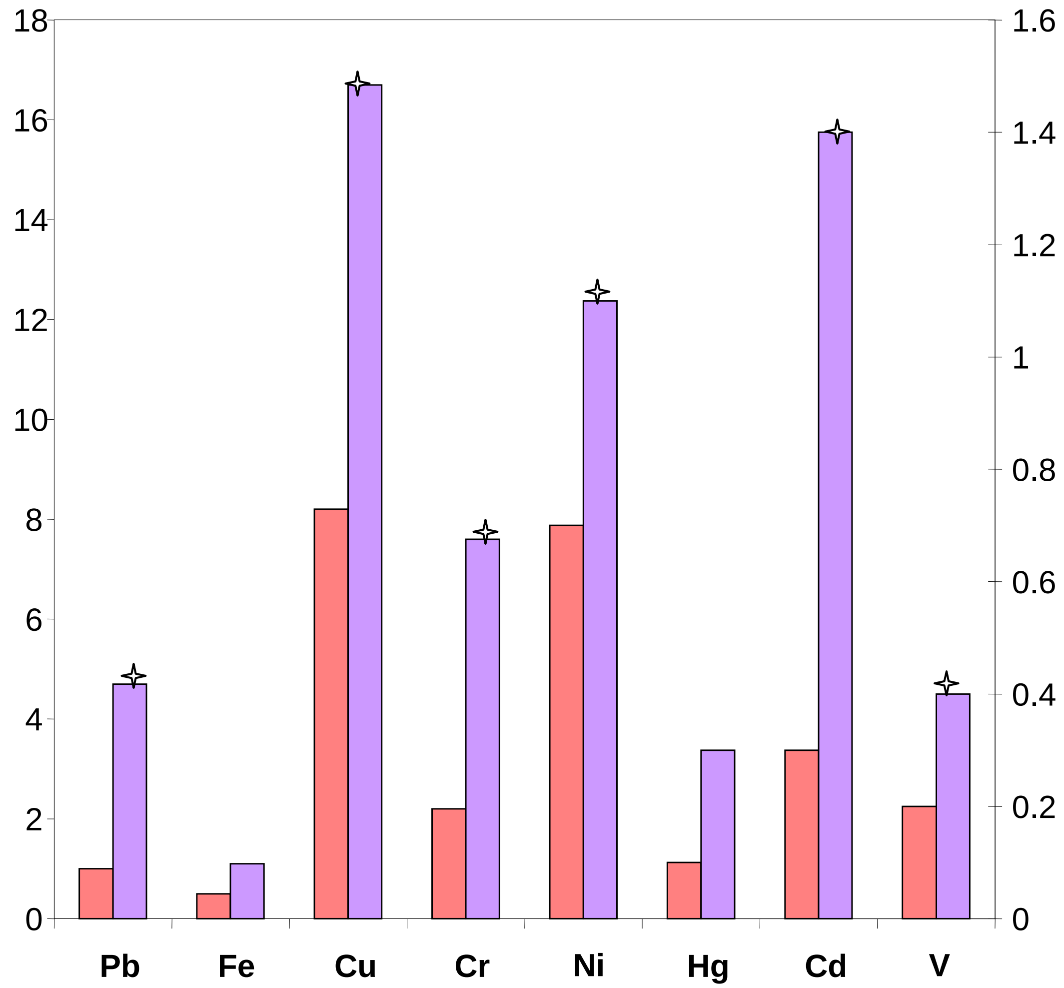

Figure 26. Mean metal concentration (ppm) in clam's livers at the contaminated and the reference site.

Right axis: Pb, Fe, Cu, Cr; Left axis: Ni, Hg, Cd, And V.
✧ = Significantly different.

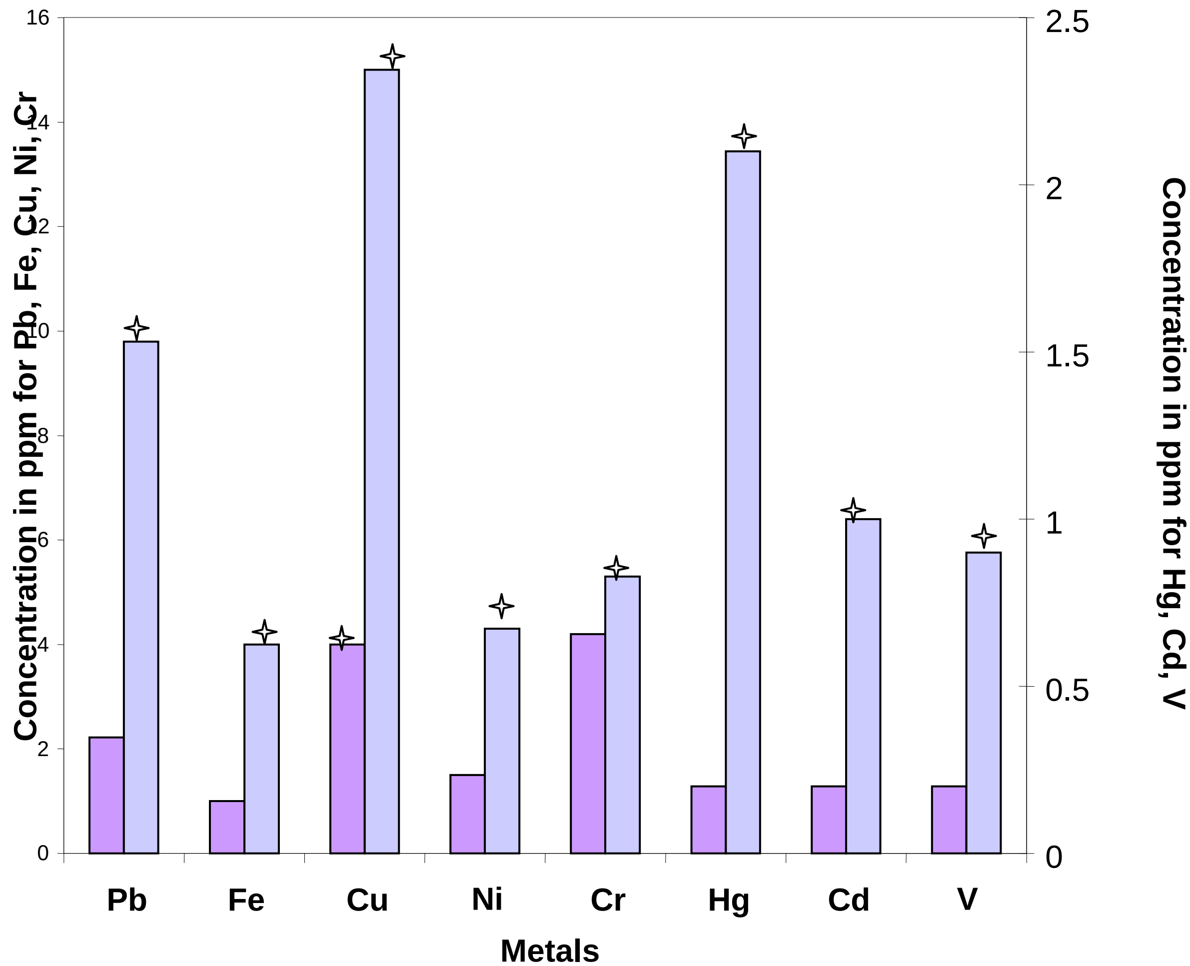

Figure 27. Mean metal concentration (ppm) in clam's gonads at the contaminated and the reference site.
Left axis: Pb, Fe, Cu, Ni, Cr
Right axis: Hg, Cd, V.
✧ = Significantly different.

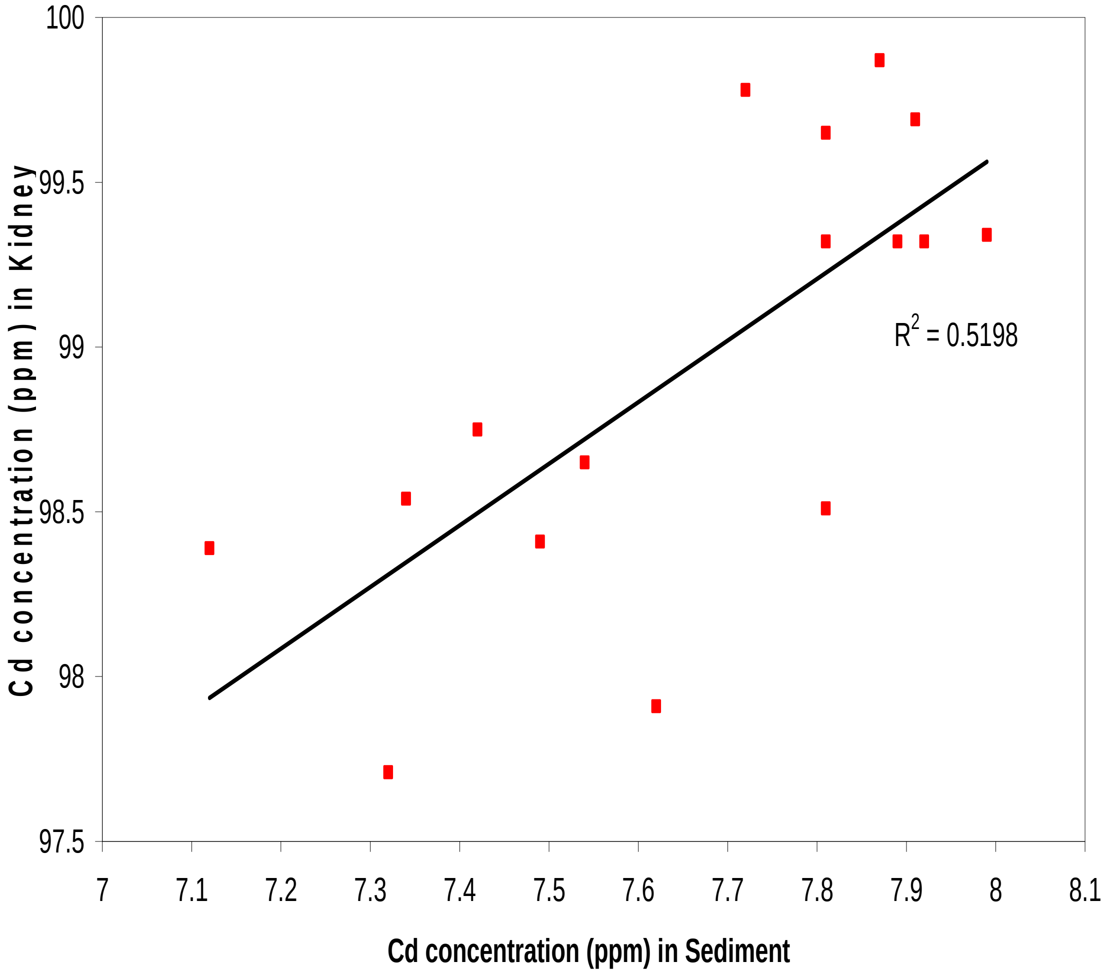

Figure 28. Correlation of Cd concentration (ppm) between sediment and kidney at the contaminated site.

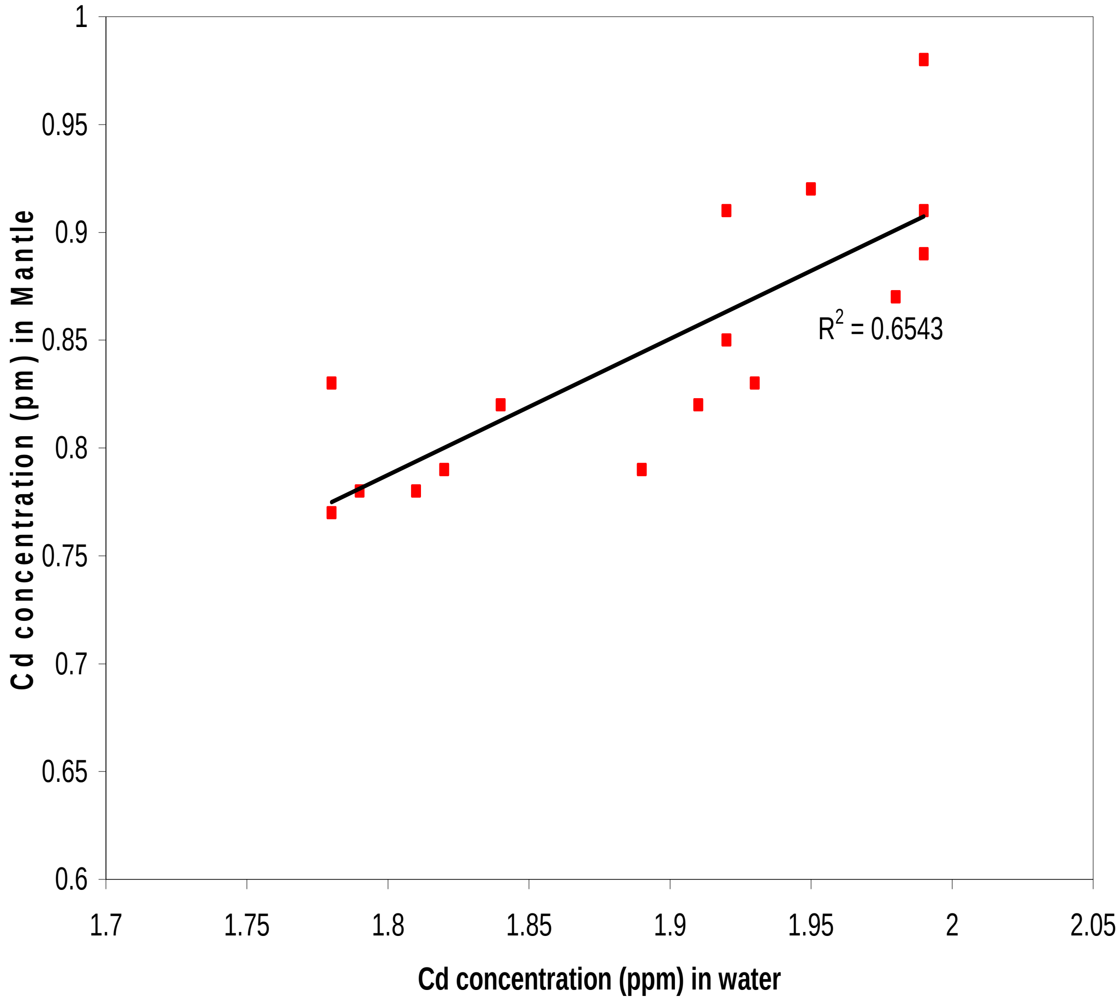

Figure 29. Correlation of Cd concentration between water (ppb) and mantle at the contaminated site.

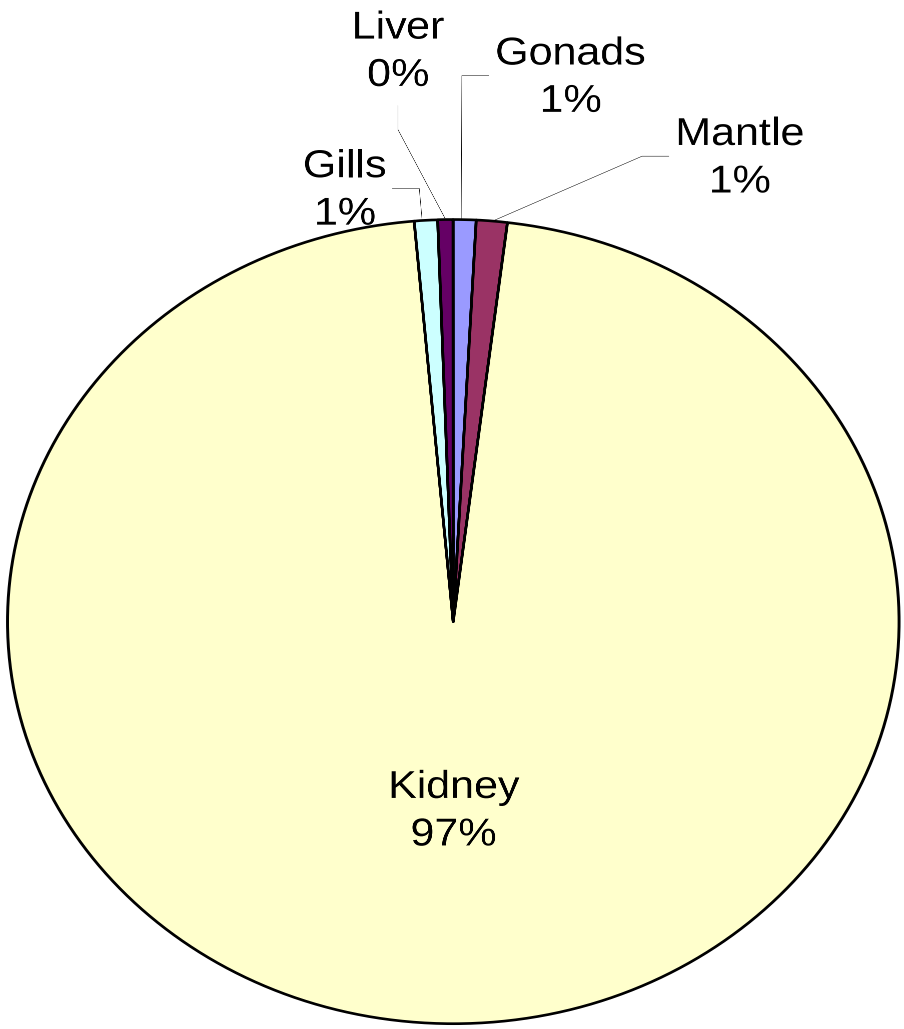

Figure 30. The percent (%) of Cd Concentration in various organs; gonads, mantles, kidneys, gills and livers.

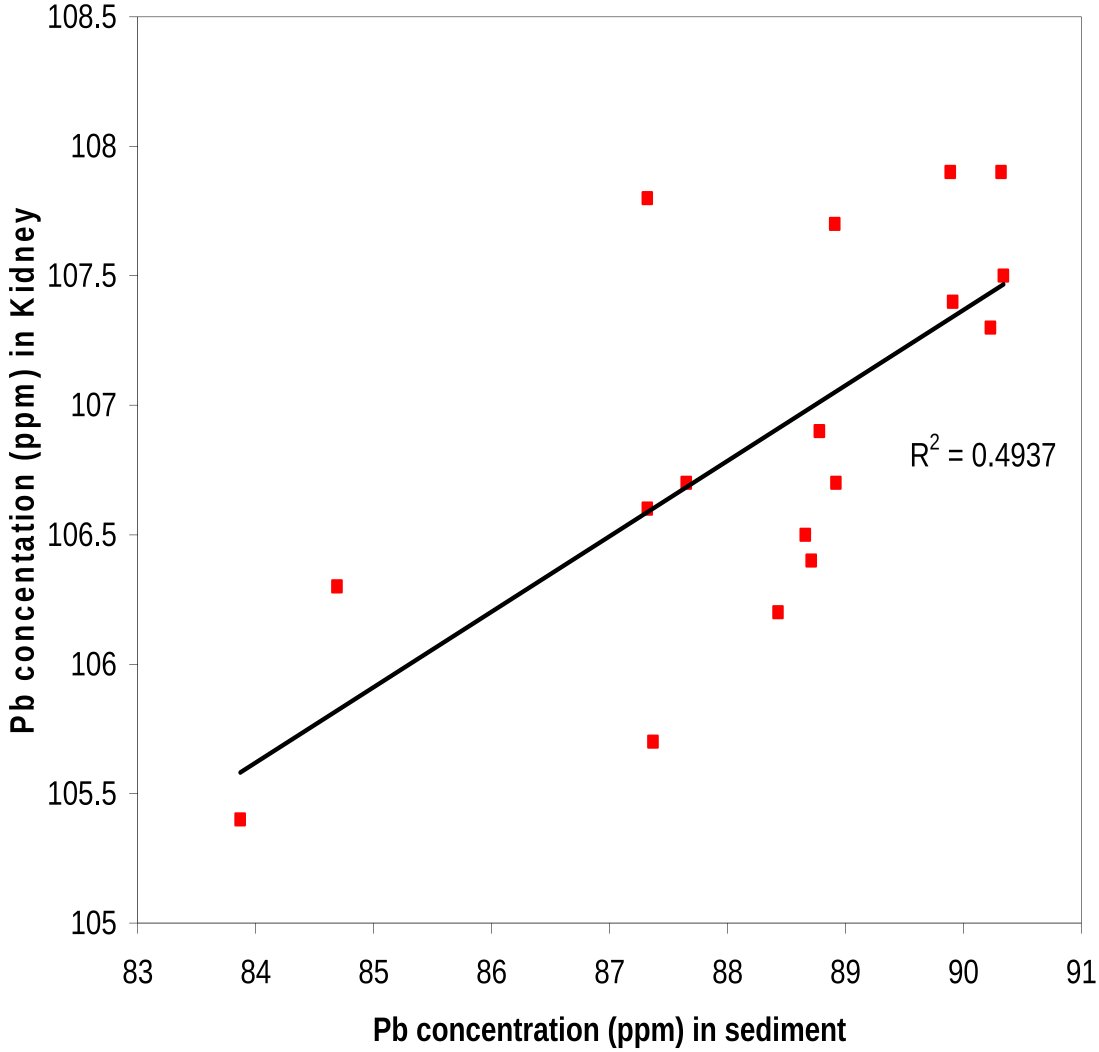

Figure 31. Correlation of Pb concentration (ppm) between sediment and kidneys at the contaminated site.

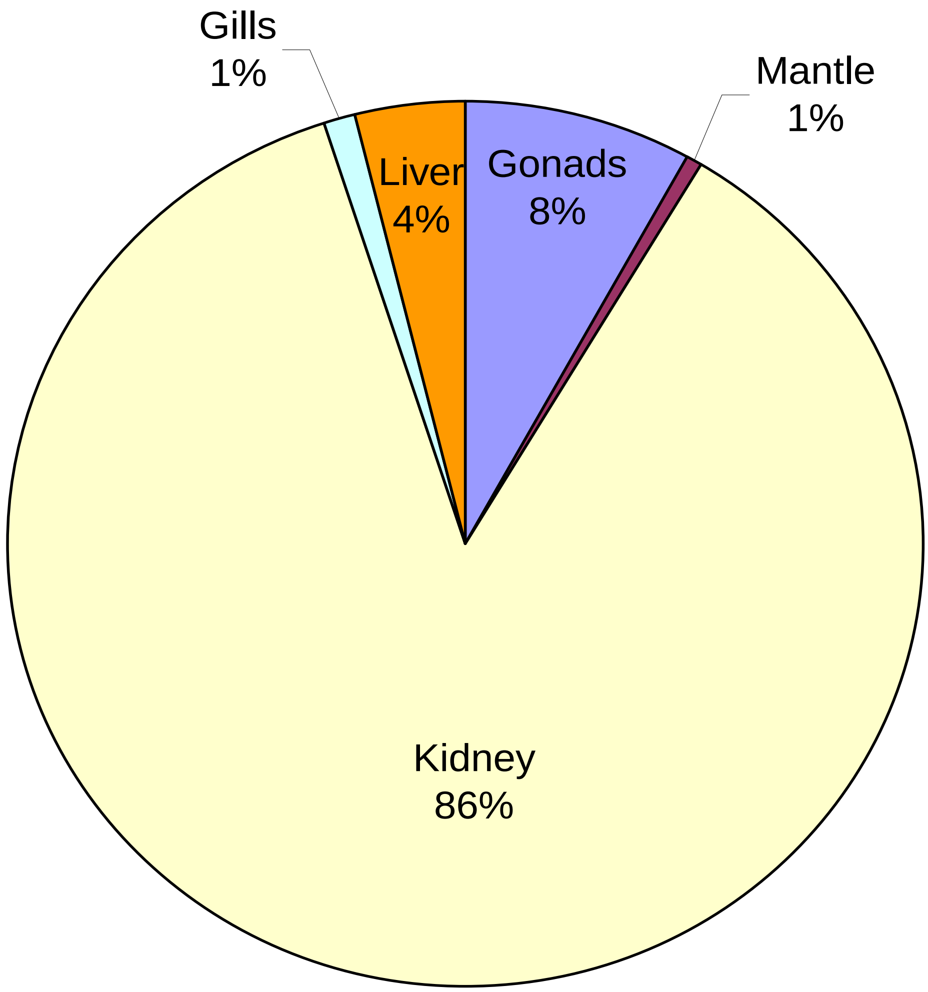

Figure 32. The percent (%) Pb concentration in various organs; mantles, kidneys, gills, livers and gonads.

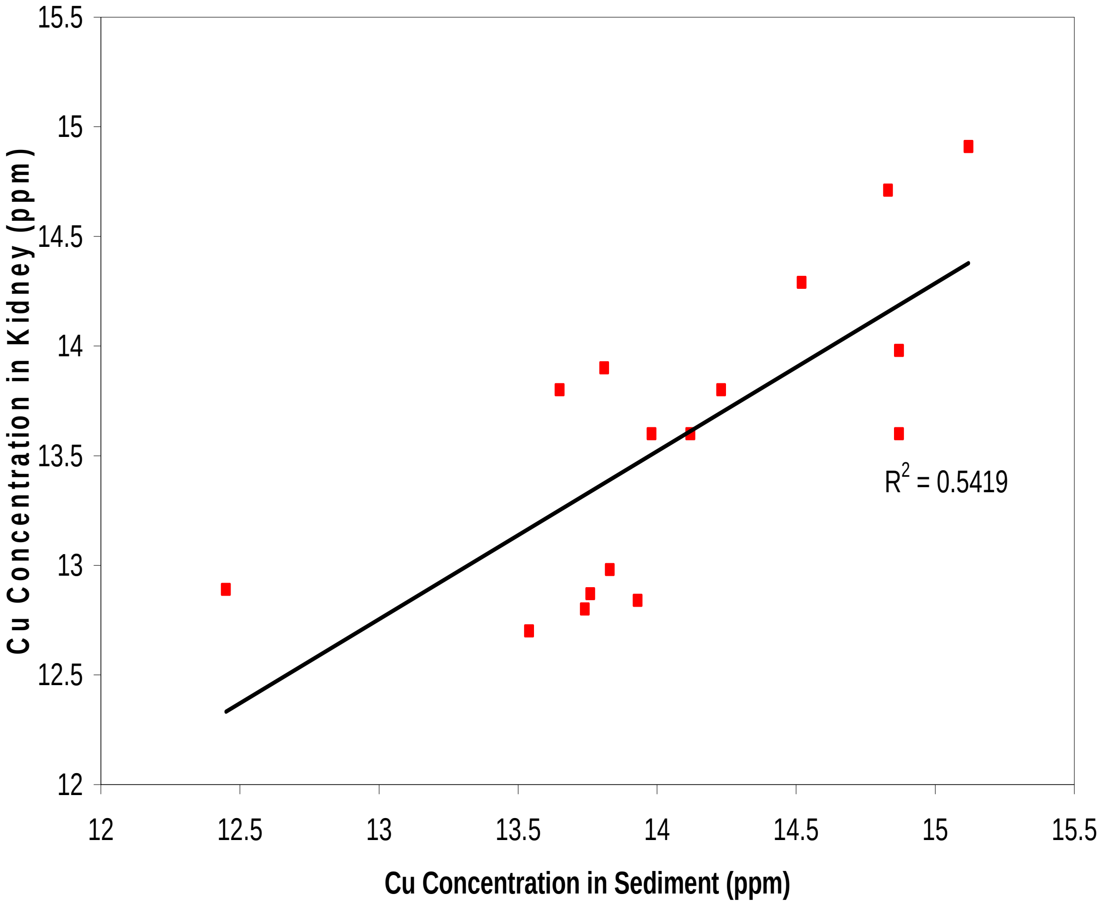

Figure 33. Correlation of Cu concentration (ppm) between sediment and kidneys at the contaminated site.

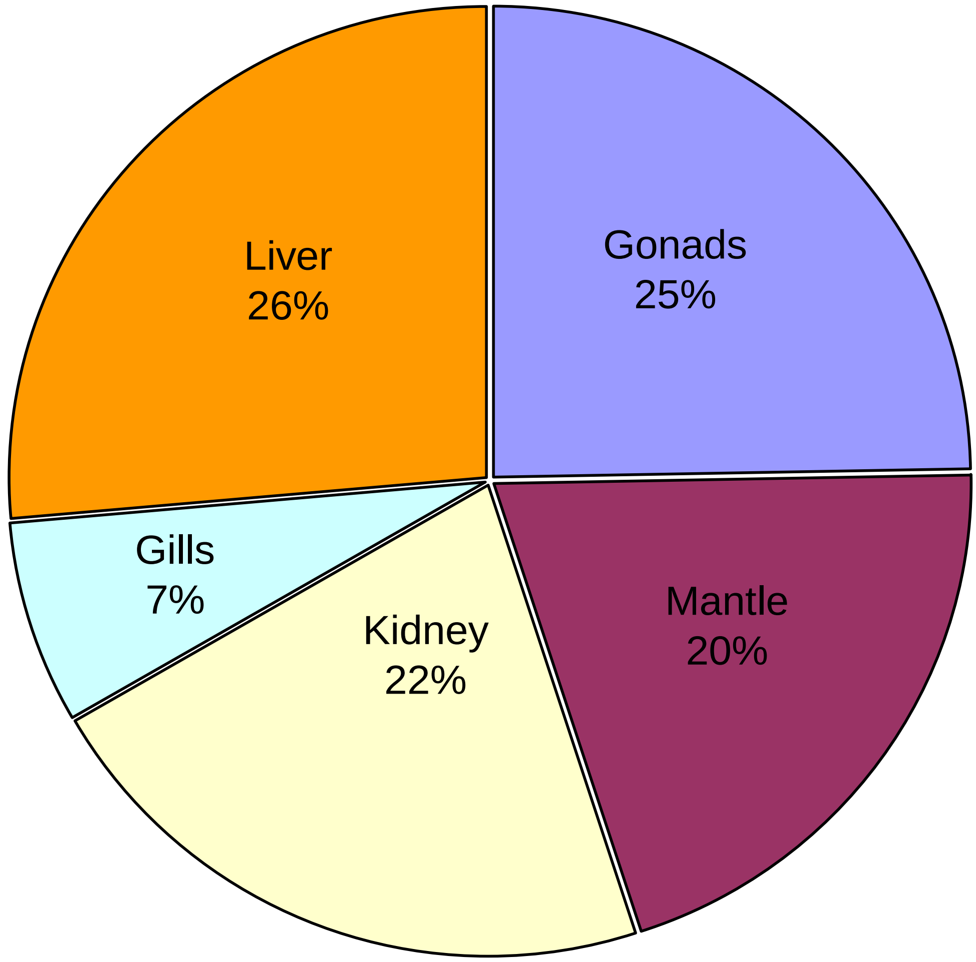

Figure 34. The percent (%) Cu Concentration in various organs; gonads, mantles, kidneys, gills and livers.

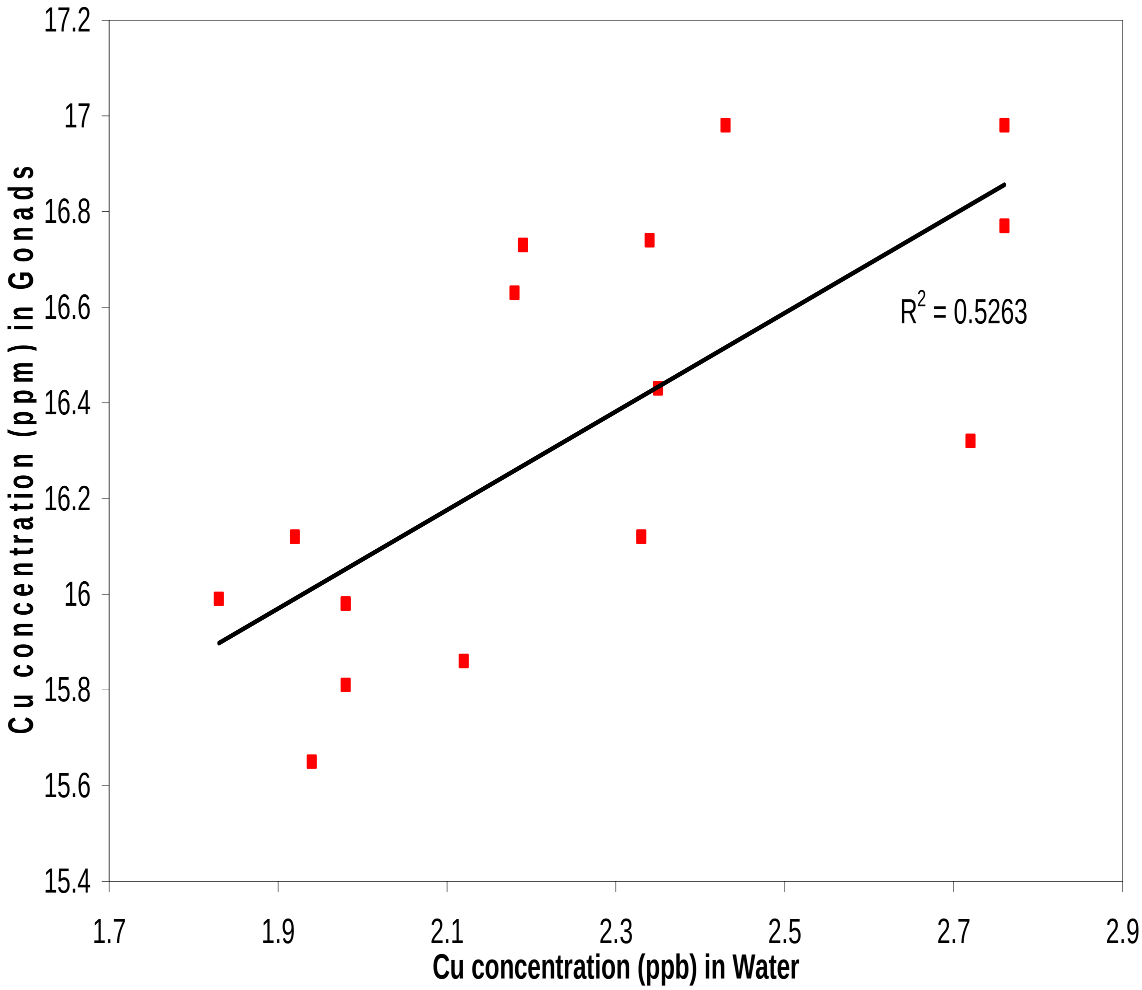

Figure 35. Correlation of Cu concentration between water (ppb) and gonads (ppm) at the contaminated site.

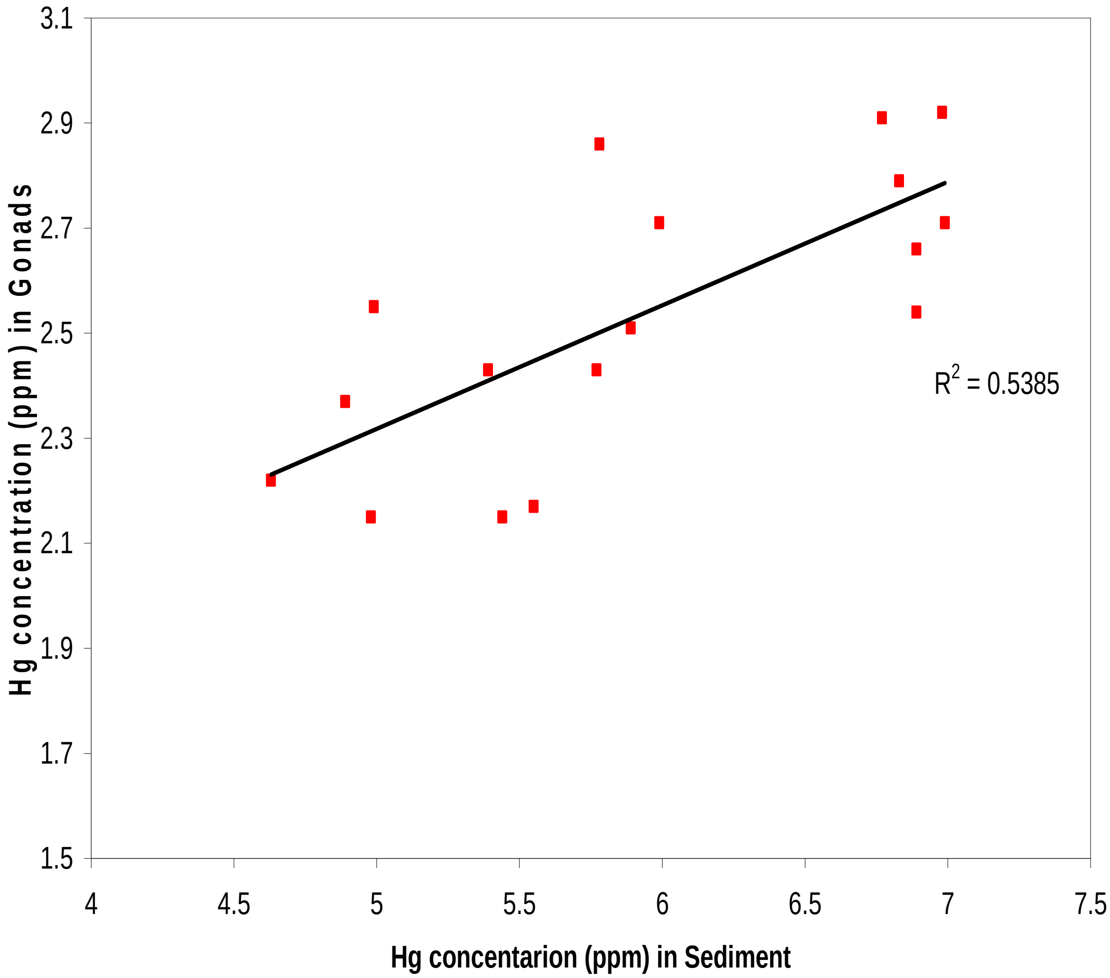

Figure 36. Correlation of Hg concentration (ppm) between sediment and gonads.

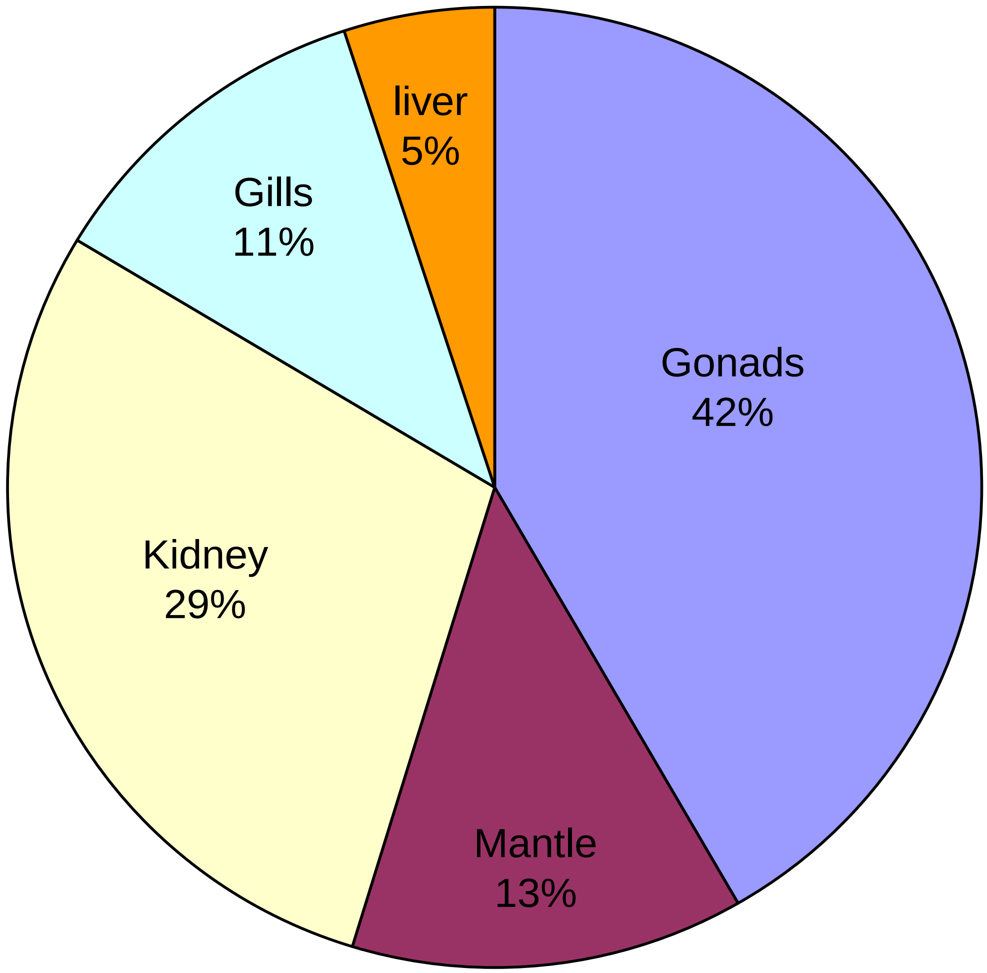

Figure 37. The percent (%) of Hg concentration in various organs; gonads. mantles, kidneys, gills and livers.

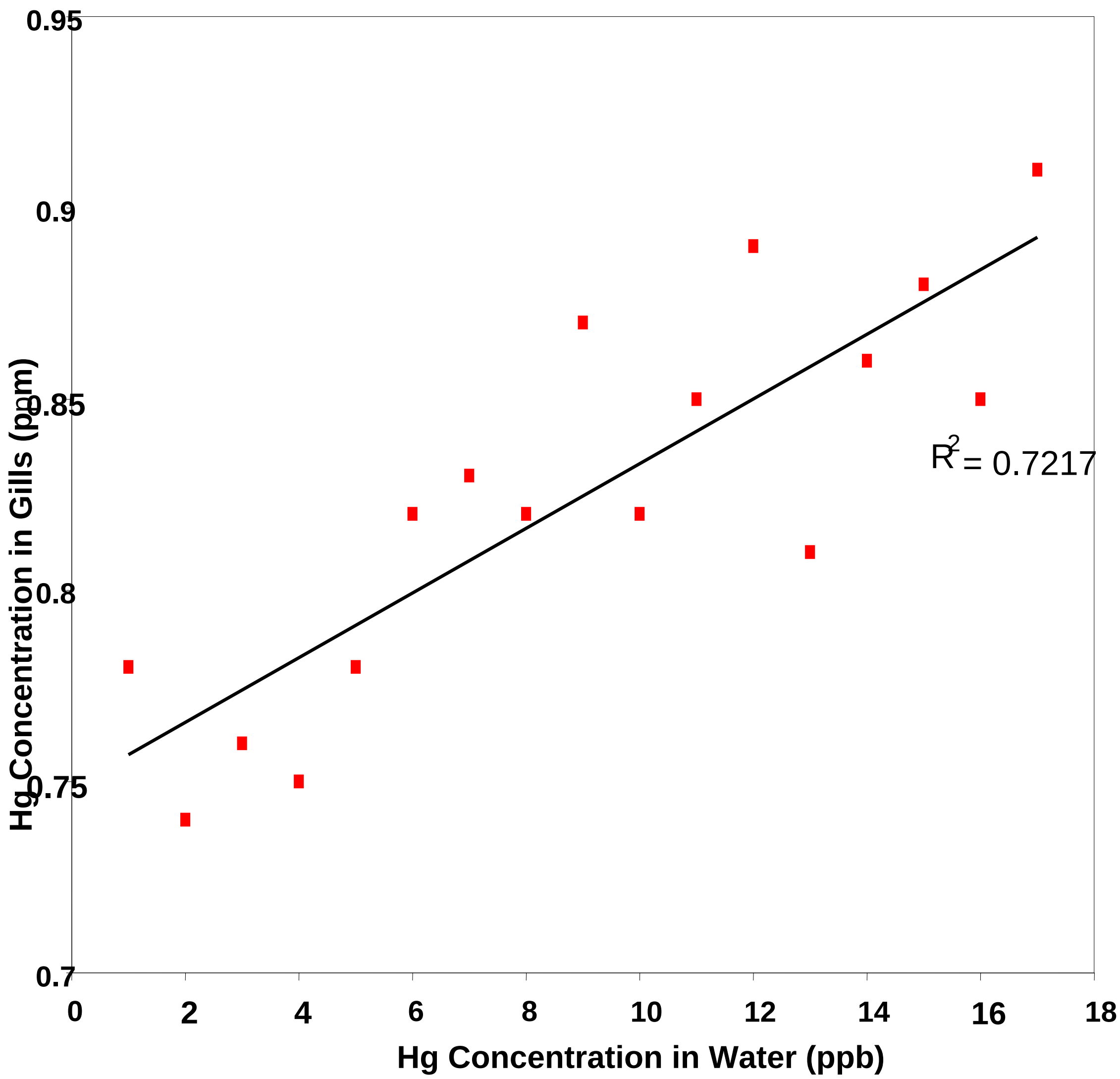

Figure 38. Correlation of Hg concentration (ppm) between water and Gills at the contaminated site.

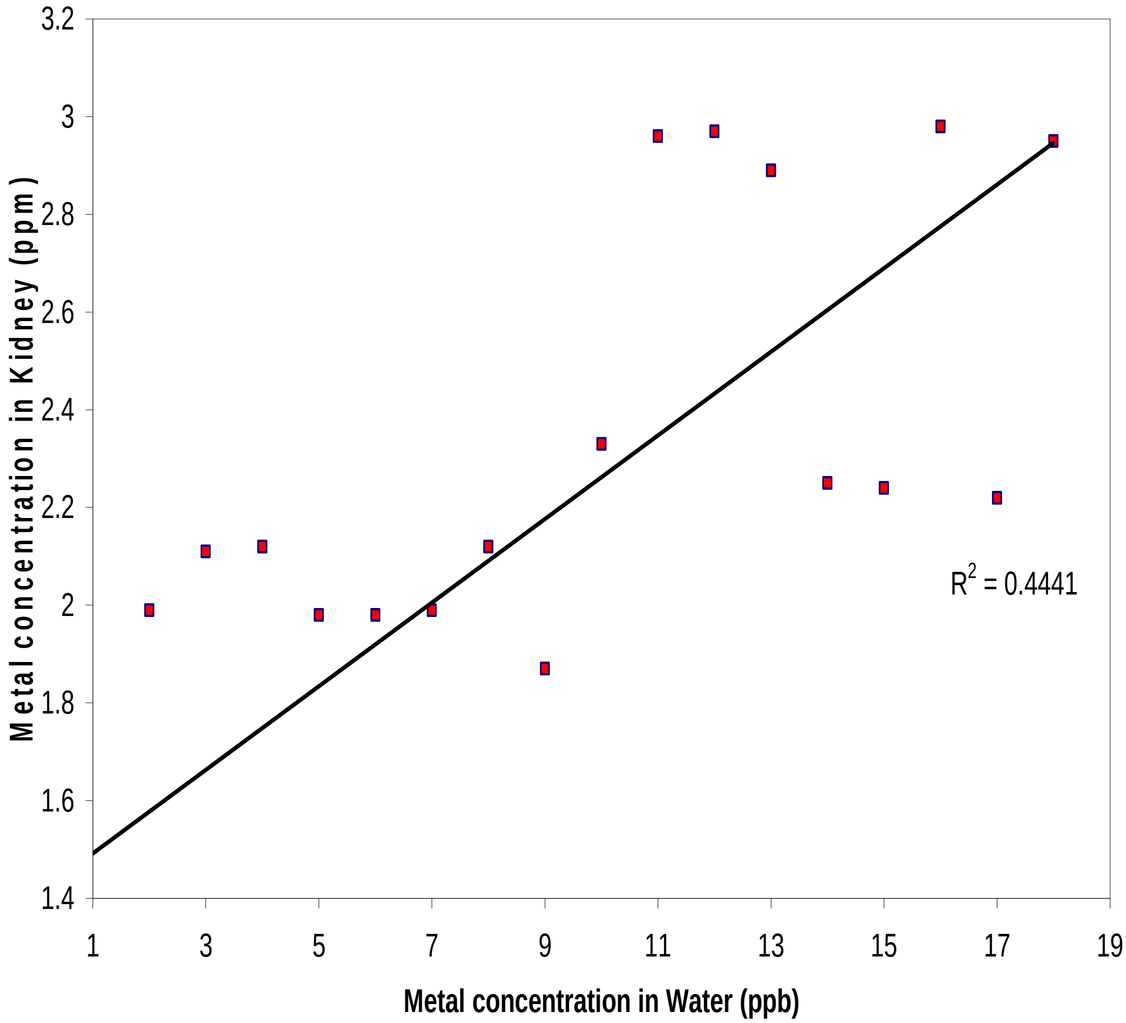

Figure 39. Correlation of Hg concentration (ppb) between water and kidneys (ppm) at the contaminated sites.

3.6. Discussion

The main findings in this study were: 1) the mean wet weight of gonads, mantles and gills in contaminated clams were significantly reduced (up to a factor of 2, 2.1, 2.2 respectively) at the contaminated site clams compared to the reference site clams; 2) The highest inter-location variability of metal concentrations was in the kidney of the contaminated site clams compared to all other organs. However, the clam *A. umbonella* did not show any differences in kidney size or weight in contaminated site clams compared to the reference site clams; 3) The concentration of Zn in various organs did not differ between contaminated sites and the reference sites; 4) The liver (digestive gland) was the main depository of Cu at the contaminated site clams compared to the reference site clams; 5) Strong inter metal correlations among organs and water as well as organs and sediments were observed.

3.6.1. Organ weights at the contaminated and the reference site clams.

Low gonad weights observed at the contaminated site clams may be due to higher metal levels (Hg, Cu, Cr, Ni, Pb and Cd) in the clams of the contaminated sites than at the reference site clams. In particular, reduced gonad weight (poor gonad development) at the contaminated sites could be due to the elevated levels of Hg (2.3 ppm) and Cu (15.0 ppm). Previous studies indicated that the elevated levels of Cu and Hg have a strongly inhibitory effect on gamete production and maturation in scallop (E Gould et al., 1988). Total gamete weight per scallop doubled in reference sites individuals but dropped by 60% in both high metal exposure groups. The soft shell clam (*Mya arenaria)* exhibited delay in gonad maturation when exposed to the heavy metals such as Hg, Pb, Zn and Cu (Gauthier-Clerc et al., 2002). Furthermore histological observations have shown that the gonads of soft shell clams were devoid of mature germ cells (Siah et al., 2003). The same histopathological observations were reported in the fresh water zebra mussels (*D. polymorpha*), (Regoli et al., 2001). Though not definitive, the results of the present study were consistent with the hypothesis that certain metals affect the development and growth of the excretory (kidneys, mantles, gills) and reproductive organs.

Reduced mantles and gills wet weights in clams from the contaminated sites compared to the reference sites may also be the consequences of higher accumulation of metals in these organs. However since all metals were higher in clams from the contaminated sites, gills and mantles weight may reflect a general toxic effect.

3.7. Kidney weights and metal accumulation

Interestingly, there were no signs of visible stress in kidney size and weight in the present study. It is possible the relatively high metal concentrations observed in the kidney could be due to accumulation of Pb, Cd and V in intracellular membrane-bound cytoplasmic granules or metallothioneins (Walsh, 1990). As indicated by Kotsonis et al ., (1978), Cd accumulated in liver, kidney rather than in muscle and may be replacing the Zn in some enzymes and has a long half-life (10-30 yr) Metallothioneins are associated with a detoxifying role and can account for 20% of the kidney volume in bivalves (Langston et al., 1998) and allow the kidney to function as a reservoir for excess trace metals. The kidney has an extra capacity to accumulate almost all metals and demonstrate tolerance without harming itself. This may reflect specific regulatory activities for the extremely high accumulation of metals in the kidney particularly Cd and

Pb (Ahn in-Young et al., 2001). The ability of the kidney to concentrate a diversity of metals infers an abundance of non-specific metal binding sites in its tissue, which immobilize and retain metals for relatively long period of time (Denton.Gary and Leroy Heitz, 1988). The mechanism of immobilization and internal detoxification of Cd, Cu, and Pb by bivalves has been also described by Schulz-Baldes, 1977. These metals are taken up in the gills and distributed by the blood and finally stored as a phosphorus sulfur-rich complex in membrane-bound vesicles within the excretory cells of the kidney.

3.8. Comparison among tissues

The highest Cr concentration was found in mantles compared to the kidneys, gonads and livers in clams from the contaminated sites compared to clam from the reference sites. There were also significantly higher concentrations of Cd, Hg, Ni, V, and Fe in mantles and gills of contaminated clams compared to the reference site clams. Previous studies have reported that, generally, tissues where absorption takes place (like gills and mantles), were enriched in a wider range of different metals than other tissues (Bebianno et al., 1985, Siha et al., 2006). However, the gills and mantles are in contact with the external medium and considered responsible for metal transfer to the organisms. Thus, based on the results derived, the preceding evidence supports the first hypotheses that organs rich in protein (livers, kidneys and gonads) accumulate more Hg, Cu, and Cd than other organs.

The concentration of Cu was significantly higher (16.7 ppm) in the liver (digestive gland) than in any other organ, and could be associated with metal binding protein (metallothioneins). Langston et al., (1998) observed that 70% of Cu within the digestive gland in bivalves may be associated with metallothioneins, which may infer protection from cytoxicity. The digestive gland plays also an important role in heavy metal metabolism and contributes to their detoxification (Viarengo, 1992). Irato et al., (2005) also reported that Japanese little neck clam accumulates Cu in the digestive gland more readily than do other bivalves, maybe due to increased induction of metallothioneins like proteins. Viarengo et al., (1985) demonstrated that Cu^{2+}, when present in sea water at the sub-lethal concentration of 0.08 mg L^{-1}, is able to induce within 48 hours the synthesis of Cu-binding proteins in the gills, mantles and livers (digestive gland) in bivalves.

In all tissues, Zn concentrations were not significantly varies in clams from the reference and the contaminated sites. Similar results of field studies on *Mytilus edulis*, have shown that the levels of Zn do not vary considerably between polluted and unpolluted areas, and confirm these observations that Zn is regulated by all bivalves (Phillips & Yim, 1981; Klumpp & Burdon-Jones, 1982; Lobel et al; 1982, Philips, 1985). Among mollusks, bivalves are able to regulate Zn and their Zn accumulation does not reflect environmental exposure (Bryan, 1979; George & Pirie, 1980, Triquet et al,. 1986. A similar Zn regulating ability has been recorded for decapods crustaceans (Bryan, 1976; White & Rainbow, 1982; Rainbow, 1985, 1988).

3.9. Inter-correlation among metals

Several inter-metal correlations among organs, water and sediment were found in the present study. This was may be the consequences of specific mechanisms as uptake, transport, storage and excretion of metals (Simkiss and Mason, 1983; Phillips and Rainbow, 1989). There were positive correlations of Cd and Cu concentrations between water and mantles, water and gonads (R^2 =0.65, R^2 =0.52 respectively). These correlations may be attributable to the fact that clam *A. umbonella* required Cu and Cd in the same proportion during the life cycle for metabolic processes. Paez-Osuna and Marmolejo-Rivas (1990a) observed the same correlations in *Crassostrea corteziensis* from the port of Mexico. The Cd is found in marine waters mostly in dissolved form (Balls, 1985). This was expected since the clam *A. umbonella* is a filter feeder, there would be a stronger correlation between metal concentration in various organs and metal concentration in water. However, these results also indicate a statistically strong correlation of Cu, Cd and Hg concentration between sediment and kidneys and between sediment and gonads (R^2 = 0.51, R2= 0.54, R^2 =0.53 respectively). The ability of many in- faunal benthic species to switch from deposit feeding to suspension feeding depend on the flow regime (Taghon et al, 1980). Dauer et al., 1981 used the term “interface” feeders to refer species that were not obligatory deposit or suspension feeders; it could be due to stress or poor quantity of suspended particulate matter that caused the switch from filter feeding to deposit feeding.

3.10. Suitability of individual organs as a bioindicators

The distribution of metals in the isolated tissues of clam *A. umbonella* is useful to identify specific organs that may be particularly selective and sensitive to the accumulation of heavy metals. In this study (see Chapter 1), clams show differences in metal concentrations between the contaminated and the reference sites, making them useful bio-indicators. However, all organs do not show such a difference. Several inter-metal correlations between organs and water as well as organs and sediment suggest the use of these organs as bio-indicators for assessing changes in the marine coastal environment. In particular, gills accumulate all seven metals (Cu, Cr, Cd, Hg, Ni, V and Fe), indicating that gills may be the most suitable bio-indicator for changes in metal levels (solute) in the surrounding sea water. Metal concentrations in the gills of other mollusks have already been shown to increase proportionally to the concentration of the ambient environment (Langston & Zhou, 1987; Roesijadi et al., 1992,1993; Odzak et al., 1994). The kidneys of clam *A. umbonella* strongly accumulate most of the metals and therefore serve as a useful bio-indicator. For Cu and other metals associated with food and particulate matter the digestive gland would serve as a better bioindicator (Ahn In-Young et al. 2001). The Table 9 summarizes the strong metal accumulation by organs in this study.

Table 9. Metal bioaccumulation in clams' organs.
"**W**" indicates that the metal accumulated in the organ in proportion to the concentration of the metal in water.
"**S**" indicates that the metal accumulated in the organ in proportion to the concentration of the metal in sediment.

Organs	*Hg*	*Pb*	*Cd*	*Cu*	*Cr*	*V*	*Fe*
Gonads	*S*	*W, S*		*W*		*W, S*	*W*
Mantle					*W*		*S*
Liver		*W*			*W*	*S*	*W*
Gill	*W*	*W*	*W*		*S*	*W*	*S*
Kidney	*W*	*S*	*S*	*S*	*W*	*S*	

4. Accumulation of Pb, Hg, Cu and Cd as a function of size in the clam *A. umbonella* living near a desalination / power plant in Kuwait.

4.1. Abstract

This study was designed to compare the metal concentrations and metal burdens of Pb, Hg, Cu, and Cd in the soft tissues of clam *Amiantis umbonella* as a function of clam size. Clams were collected near a desalination/power plant and a reference site 5 km away in Kuwaiti Bay. The objectives of this study were to: 1) examine the correlation between the weight of the soft tissues and shell length in clams from the contaminated and the reference sites: 2) determine the metal concentration and body burden variations of four metals (Cu, Cd, Pb and Hg) in relation to clam shell length and wet weight of contaminated site clams: 3) develop an allometric relationship between the shell length and width and the weight of the soft tissues that provides a base for predicting metal concentration in various organs as a function of clam size. Clam length and soft tissue wet weight were significantly correlated (R^2 = 0.40, $p < 0.005$) in clams from the reference site, but were not correlated (R^2 =0.030) in contaminated site clams. Log transformed regression relationships between body burden and size/wet weight were used to establish the correlations. Among the metals, Cd body burden was positively related to clam wet weight and length, and similarly Cd concentration was also positively correlated with clam length. The body burdens of the other metals (Hg, Pb and Cu) were positively correlated to clam wet weight. However, the concentrations of these metals were not correlated with clam length or weight. This indicates that the concentrations of Hg, Pb, and Cu are biologically regulated in *A. umbonella*.

4.2. Introduction

Monitoring programs and research for metals in the environmental samples have become widely established because of concerns over accumulation and toxic effects, particularly in aquatic organisms and to humans consuming these organisms (Lauenstein et al., 1990). The criteria by which organisms are accepted as biological indicator for the assessment of environmental contamination were proposed more than twenty five years ago and remain unchanged (Phillips, 1976; Bryan et al., 1980, 1985).

A number of programs have operated throughout the world, including the United States (Goldberg, 1986; O'Connor, 1998), France (Claisse, 1989; Belieff et al., 1998), the Mediterranean, Caribean, (Kistner, 1984), and throughout Asia (Tanabe, 2000). As time passes trend detection improves and it is expected that trends that are currently obscured by natural factors will eventually emerge (O'Connor et al., 1994)

Bivalves are widely used as bio-indicators of heavy metals pollution in coastal areas because they are known to accumulate these elements, providing a time integrated measure and indication of environmental pollution. In comparison to fish and crustacean, bivalves have a very low level of activity of enzyme systems capable of metabolizing persistent organic pollutants, such as aromatic hydrocarbons and polychlorinated biphenyls (Phillips, 1977). Therefore contaminants concentrations in the tissues of bivalves more accurately reflect the magnitude of environmental contamination (Phillip, 1980, 1990). Bivalve mollusks are routinely employed to define the temporal and spatial distributions of biologically available environmental pollutants, including trace metals, in coastal areas and estuaries (Goldberg et al; 1978, Farrington 1983; Bryan et al., 1985).

Mussels, in particular species of the genus *Mytilus*, have been used most extensively as they fulfill all the requirements of an ideal bio-indicator. Hence, as a group, they have world wide distribution and where found, are usually abundant (Farrington et al., 1983). Interpretation when and where contaminant concentrations in the bodies of organisms reflect environmental exposure, however, can be complicated by biological processes such as reproductive cycles, and age (Phillips 1976a; Borchardt et al., 1988). One of the criteria when an organism is proposed as a bio-monitoring agent is a simple correlation between pollutant levels present in the organism and those in its environment (Goldberg\, 1975; Phillip and Rainbow, 1997). Factors known to influence metal concentrations and accumulation in these organisms include metal bioavailability, season of sampling, hydrodynamics of the environment, size, sex, changes in tissue composition and reproductive cycle (Boyden and Phillips, 1981). Different animals in the same community at the same trophic level could accumulate pollutants differently due to differences in habitat/niche's physical and chemical properties (Reinfelder et al., 1997). Bioaccumulation of pollutants can occur from sea water, suspended particles, and sediments and through food chains (Otchere et al., 2003). The rate at which accumulation occurs in an organism depends not only on the availability of the pollutant but also on a whole range of biological, chemical and environmental factors. The ultimate level which is reached is governed by the ability of the organism to excrete the pollutant or, alternatively store it or pollutants along food chains and the development of tolerance which sometimes occurs (Bryan, 1979).

Studying bivalve growth and establishing allometric relationship are essential for generating useful information for managing resources and understanding changing environmental conditions due to environmental metal contamination (Palmer 1990; Paez-Osuna et al., 1993a).

Recently baseline metal concentrations were determined in the principal body organs of *Laternula elliptica* with a strong metal accumulating tendency near shore site at King George Island (Ahn et al, 1996).

This study was designed to compare metal concentration and metal burden of Pb, Hg, Cu, and Cd in soft tissues of clam as a function of clam size. Clams *A. umbonella* were collected near a desalination/power plants and a reference site 5 km away in Kuwaiti Bay. The objectives of this study are to: 1) examine the correlation between the weight of the soft tissues and shell length in clams from contaminated sites compared to the reference sites clams: 2) determine the metal concentration and body burden variation of four metals (Cu, Cd, Pb, Hg) in relation to clams shell length and total wet weight of contaminated site clams: 3) develop an allometric relationships between the shell length and width and the weight of the soft tissue that provides a base for predicting metal concentrations in various organs as a function of clam size.

It is hypothesized that:

1. There will be a strong positive relationship between clam total wet weight and shell length in clams from contaminated sites compared to the reference sites clams.

2. Total metal burden in clam will increase with the weight gain, whereas the metal concentration will diminish with increase size and weight of the clam.

4.3. Materials and Methods

4.3.1. Study Sites

The sampling sites of clams *A. umbonella* is shown in Figure/Map 2. Description about sampling sites can be found in Chapter 2.

4.3.2. Clams Collection and Preparation

Clams *A. umbonella* were collected randomly from an inter-tidal flat (29.30' North, 48.00 'East) within Kuwait Bay on the west coast of Kuwait (Persian Gulf). Sea water temperature in the Bay varies seasonally from a maximum 28 C^o to a minimum $4C^{0,}$ and salinity ranges from 40.8 psu to 18.3 psu.

For organs analysis, thirty three clams were randomly collected from four contaminated Sites 2A, 2B, 3A and 3B and from reference sites during December 1999 and January 2000 with similar shell length to minimize the effect of body weight (Marina and Enzo, 1983). These sampling sites are shown in Figure/Map 2 (Chapter 2). After collection clams were transported to the laboratory in self-supporting tanks (capacity of 150 liters, aerated sea water continuously flowing). Clams were depurated in seawater for approximately 24 hours. Seawater salinity and temperature were measured during each sampling.

Each clam opened carefully using stainless steel scalpel blades. Soft organs such as gonads, stomach, digestive glands, gills, mantles, foot, hearts, and kidneys were removed. The fresh weight of each organ was recorded (mg). The following weights were recorded:

10. FW: flesh weight

11. GW: gonads weight
12. DG: digestive glands weight
13. SW: both shells weight
14. MW: mantle weight
15. St W: stomach weight
16. Kd W: kidney weight
17. Ft W: foot weight
18. HW: heart weight

All these organs were transferred to different vials and stored at -20 ^{o}C for further investigation of metal depositions using atomic absorption.

Prior to analysis, clam tissues were freeze dried to a constant weight (±0.5%). Than the sample was ground to homogenize it and sieved in order to normalize the variation in the grain size distributed. Samples were extracted by placing 0.5 gm of the homogenized sample in a Teflon beaker and heating (120 °C) with concentrated HNO3 until dry. 5 ml acid (HNO3+HCLO4+HF) was added to sample and heated up to 120 °C until dry. Then the sample was cooled to room temperature and passed through filter paper and poured into the 50 ml volumetric flask, diluting with HCl 0.1% to make up the volume.

4.4. Sediment and Water collection

Water and sediment samples were also collected during sampling from each of the two contaminated sites and reference site (see chapter 1)

4.5. Results

4.5.1. Length and weight of clam A. umbonella

Mean length, and weight of clam (*A. umbonella*) varied between 2 cm-4.2 cm for length, and 2.4g-4.48g for weight at the reference study sites (Figure/Map 2, Chapter 2). Regression between the clams length and wet weight were tested and it showed a positive correlation between shell length and wet weight at the reference sites ($p<0.05$). Mean length, and weight of clam *(A. umbonella*) were between 2.5 cm - 4.6 cm for length, and 1.2 g-1.8 g wet weight at the contaminated study sites (Figure/Map 2, Chapter 2). The length of the clams had no correlation with the wet weight ($p<0.5$) at the contaminated sites. The length of the clams increased with minimally gain wet weight and weight increased without gaining length.

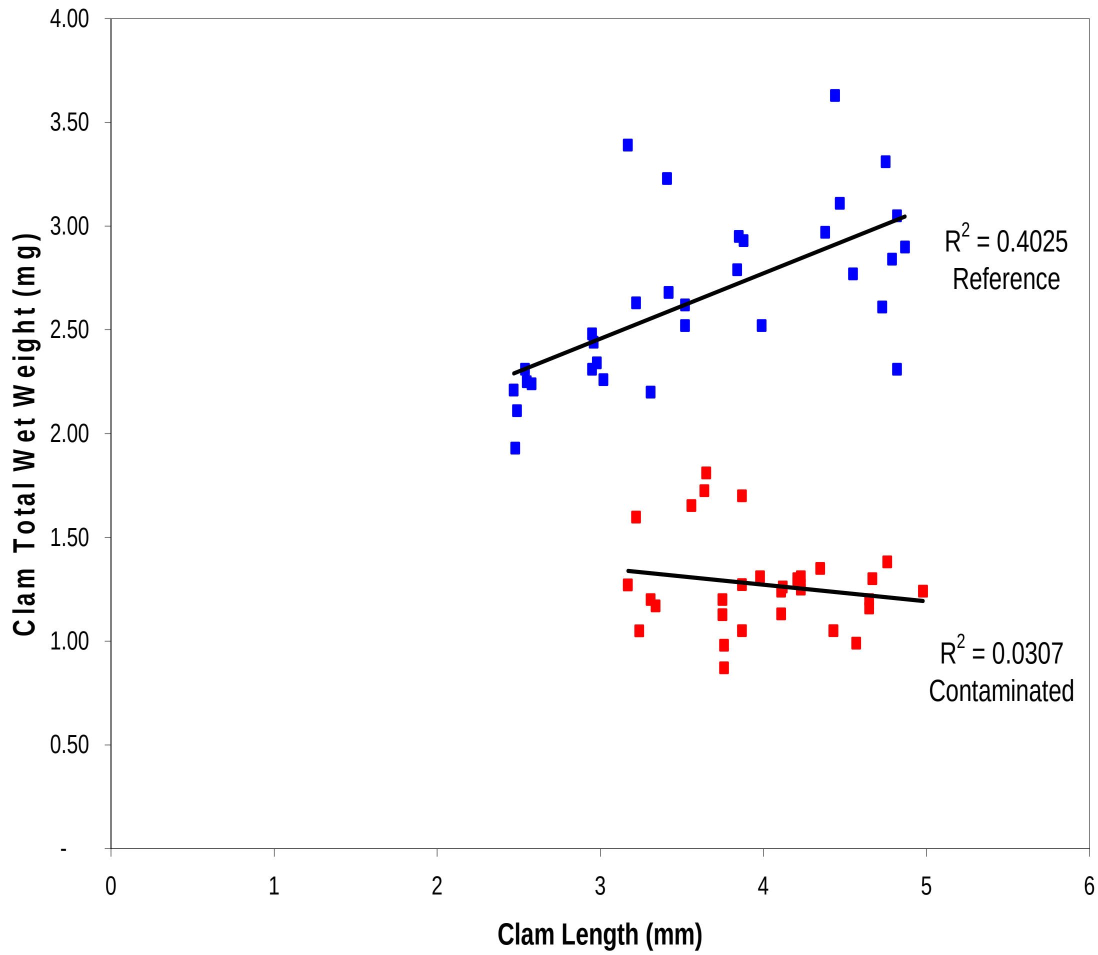

Figure 1. Correlation between mean length (cm) and mean wet weight of clam *A. umbonella* at the contaminated and the reference site.

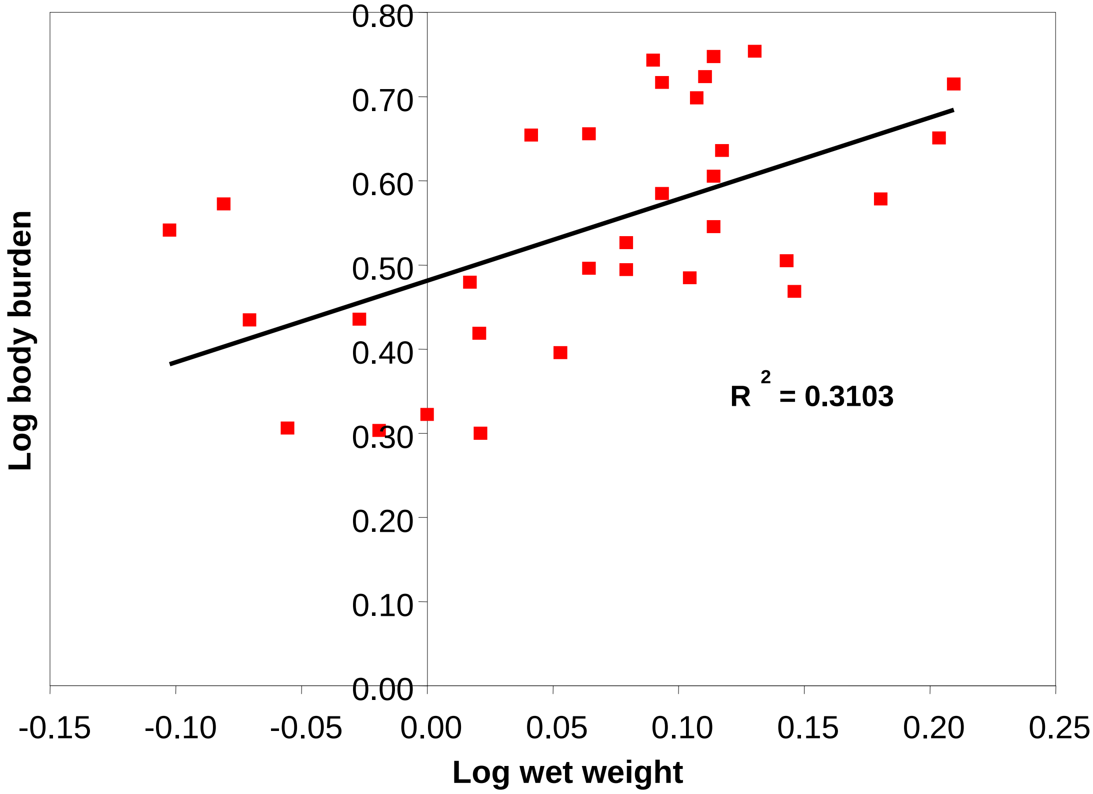

Figure 2. Correlation of Log Cd body burden (mg) between clam soft tissues and clam length (mm).

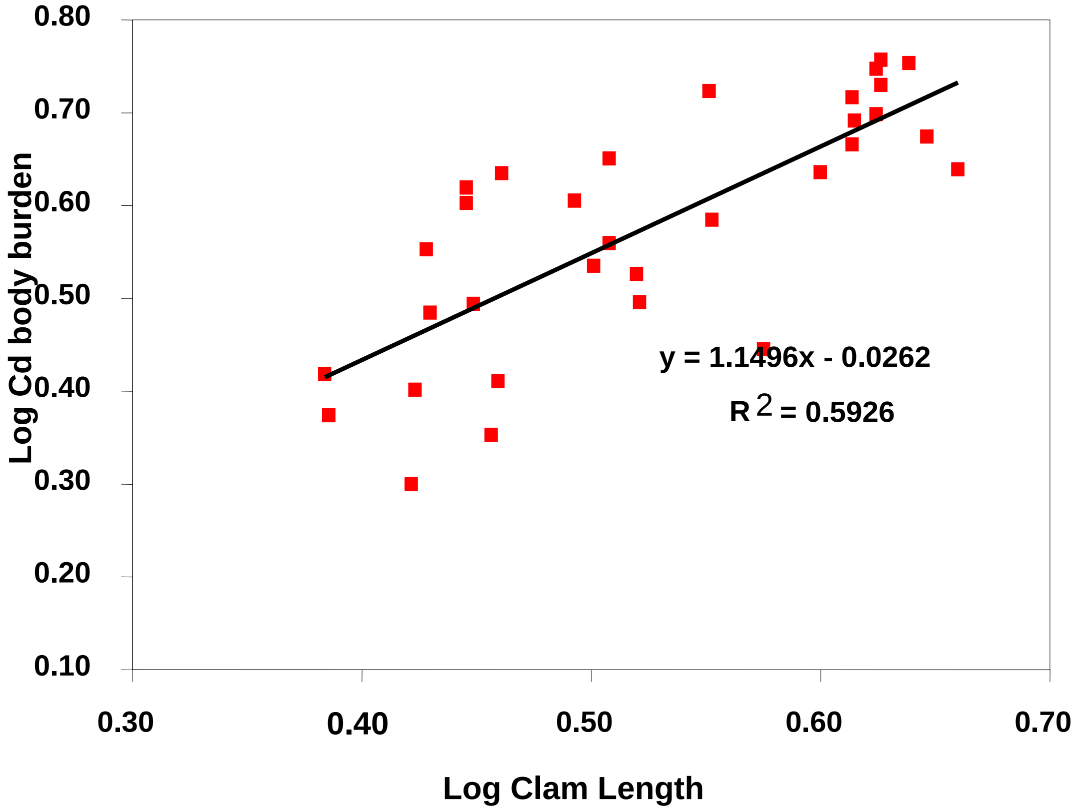

Figure 3. Correlation of log Cd body burden between clam soft tissues and clam total wet weight.

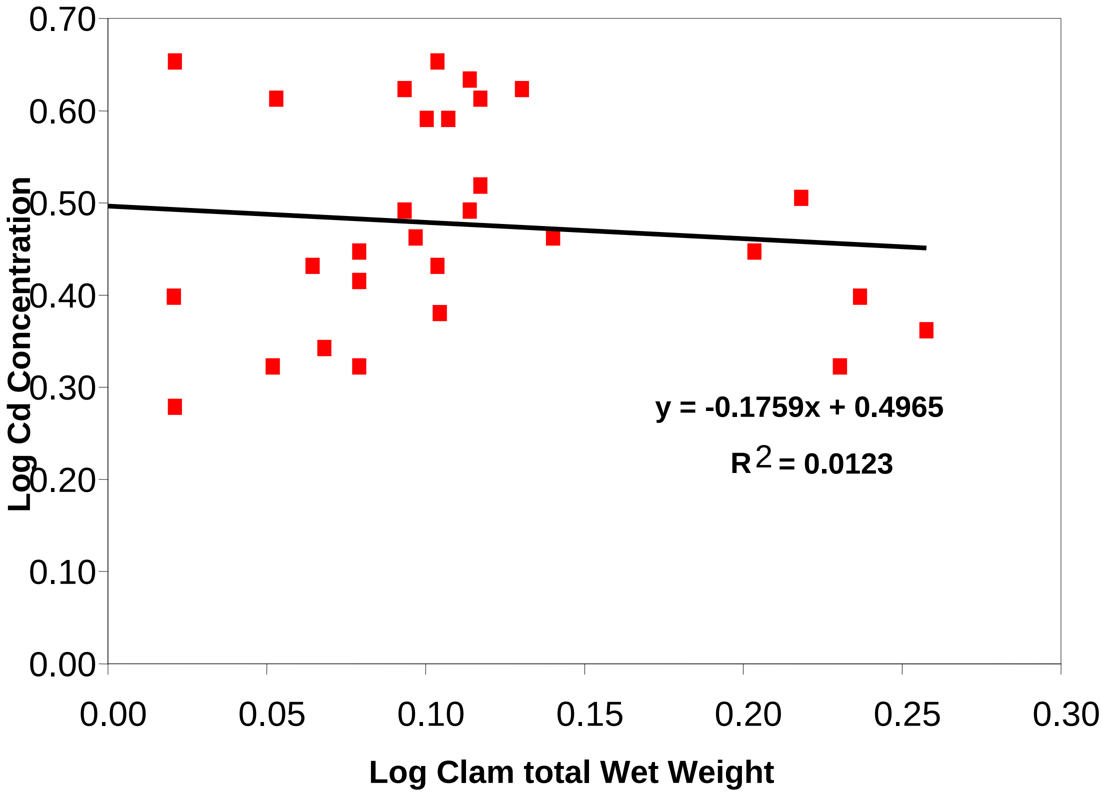

Figure 4. Correlation between Log clam wet weight and Log Cd concentration at the contaminated site.

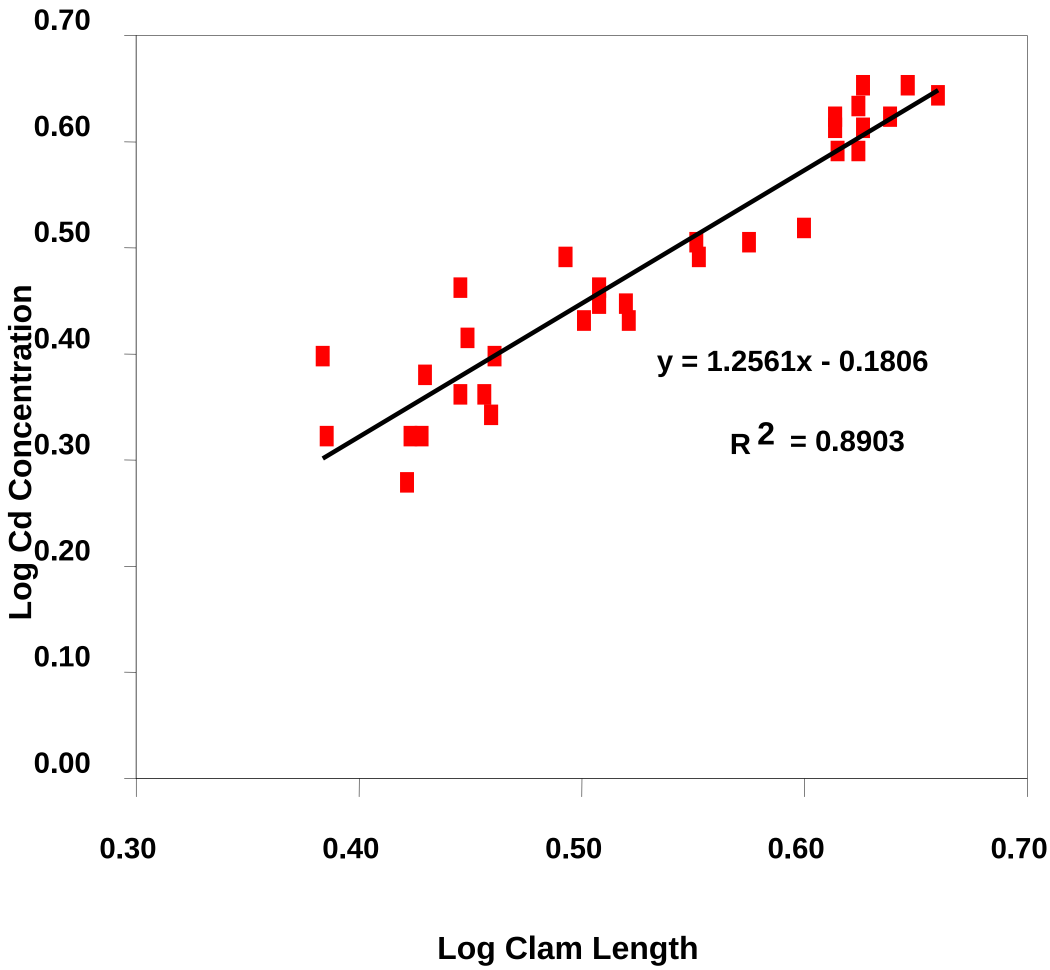

Figure 5. Correlation between Log lengths of clam *A. umbonella* and Log Cd concentration at the contaminated site.

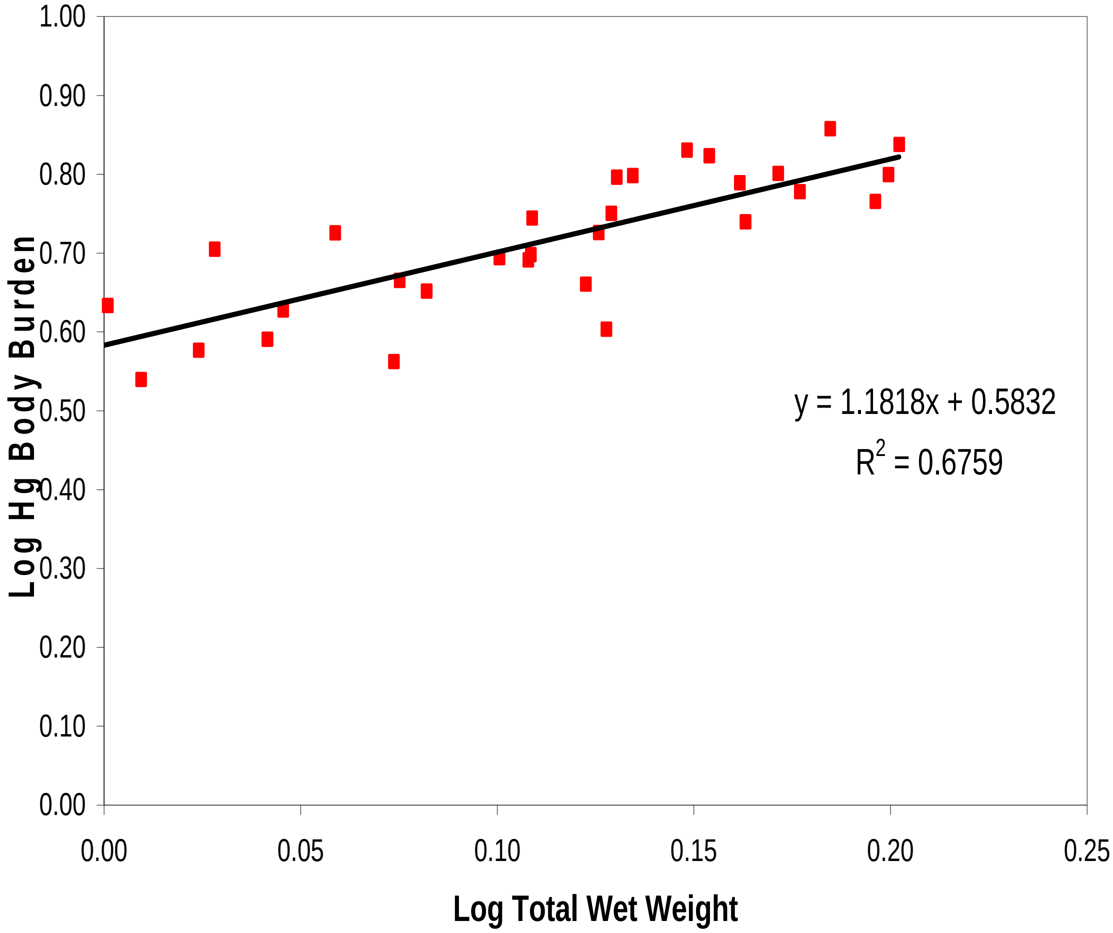

Figure 6. Correlation between Log clam total wet weight and Log Hg body burden at the contaminated sites.

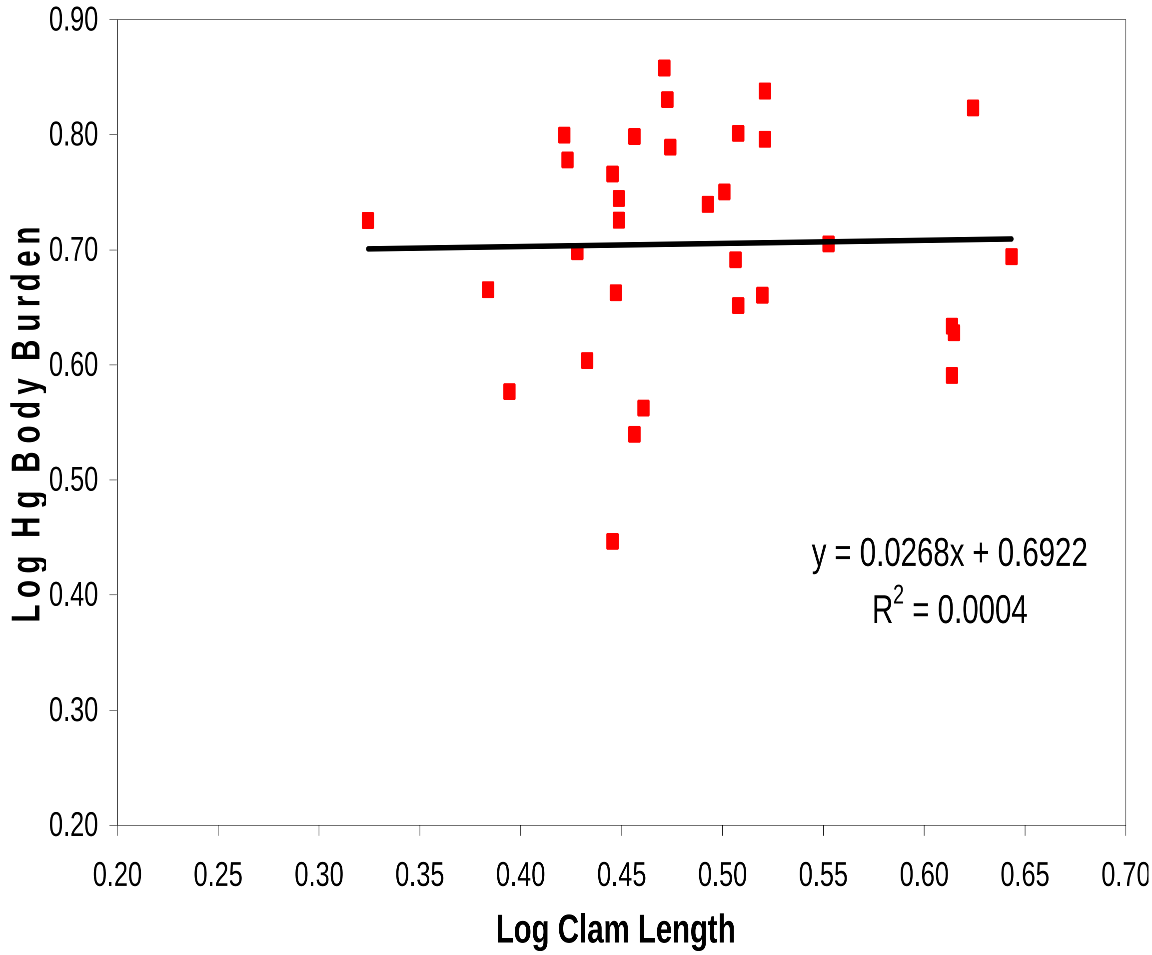

Figure 7. Correlation of Hg concentration (ppm) between clam length (mm) and clam soft tissues at the contaminated sites.

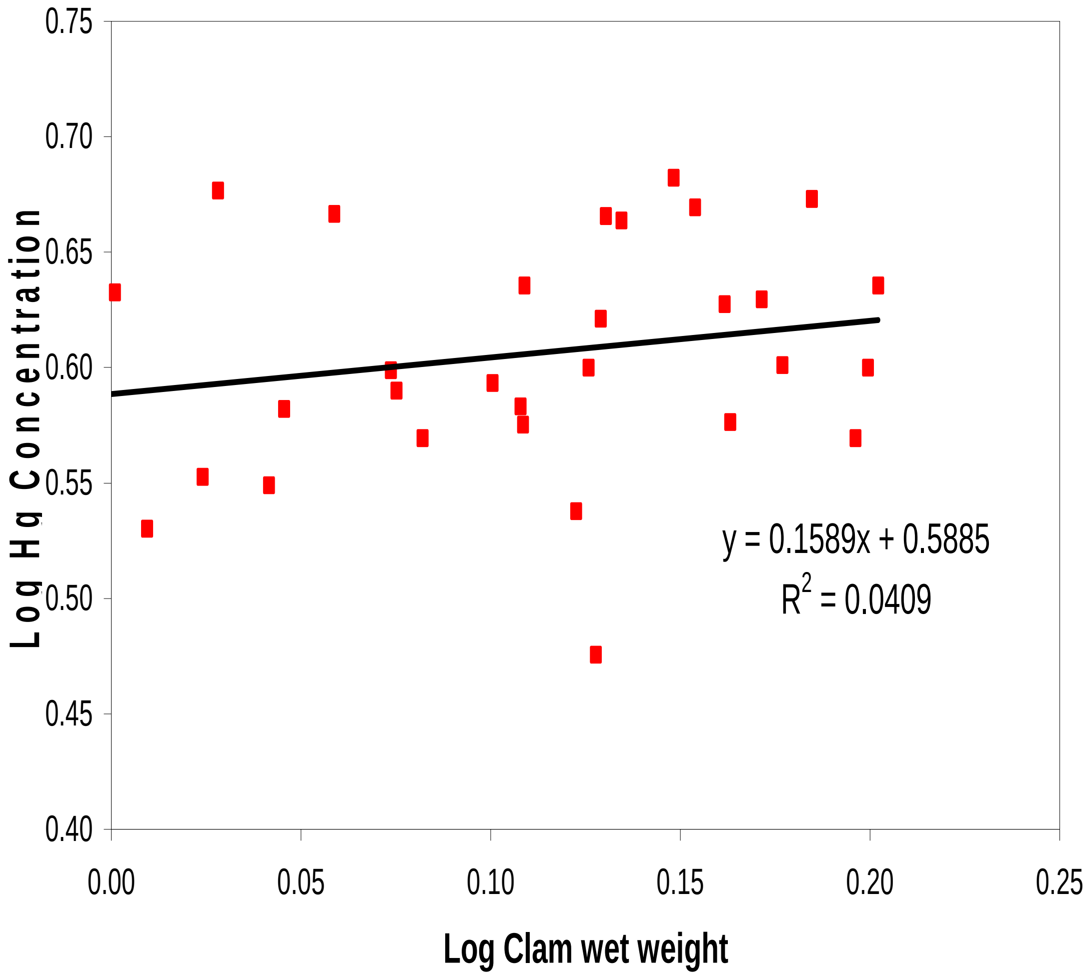

Figure 8. Correlation between Log clam wet weight and Log Hg concentration at the contaminated sites.

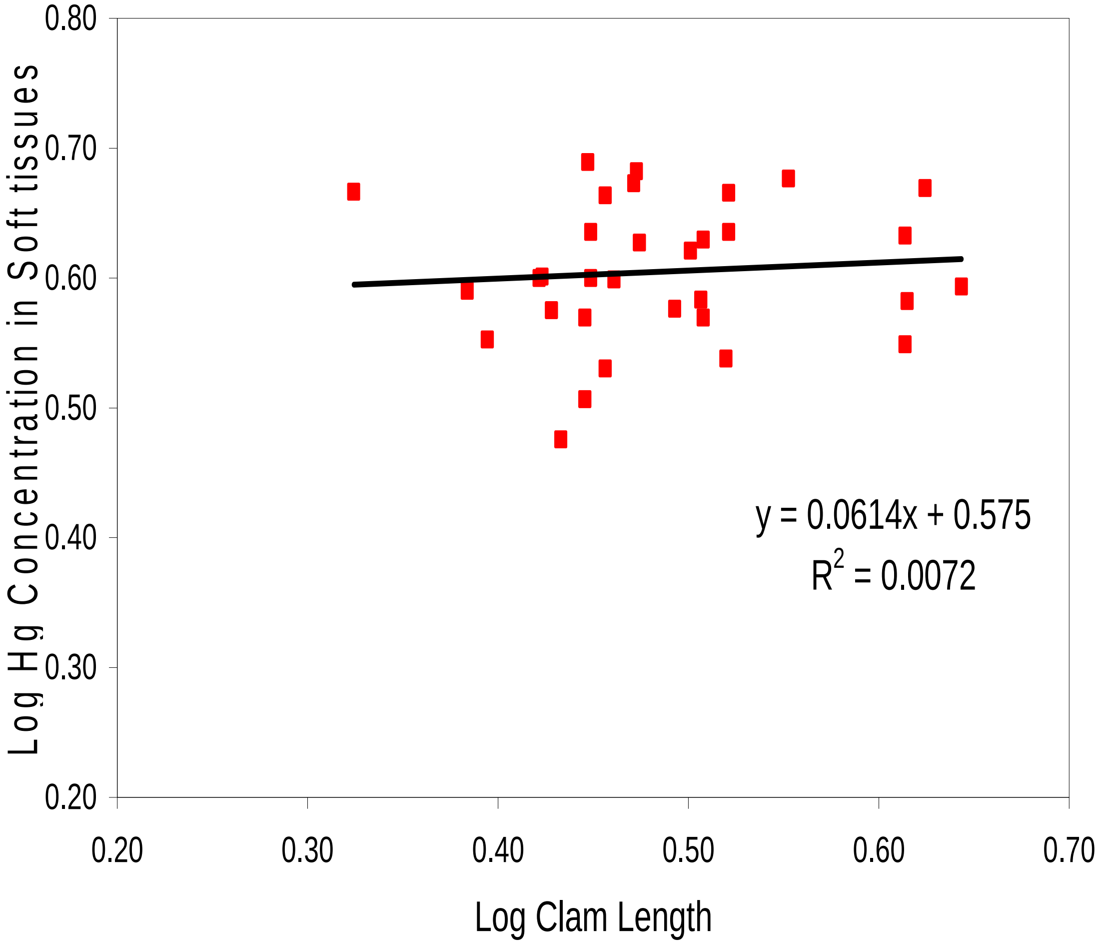

Figure 9. Correlation between Log clam length and Log Hg concentration at the contaminated sites.

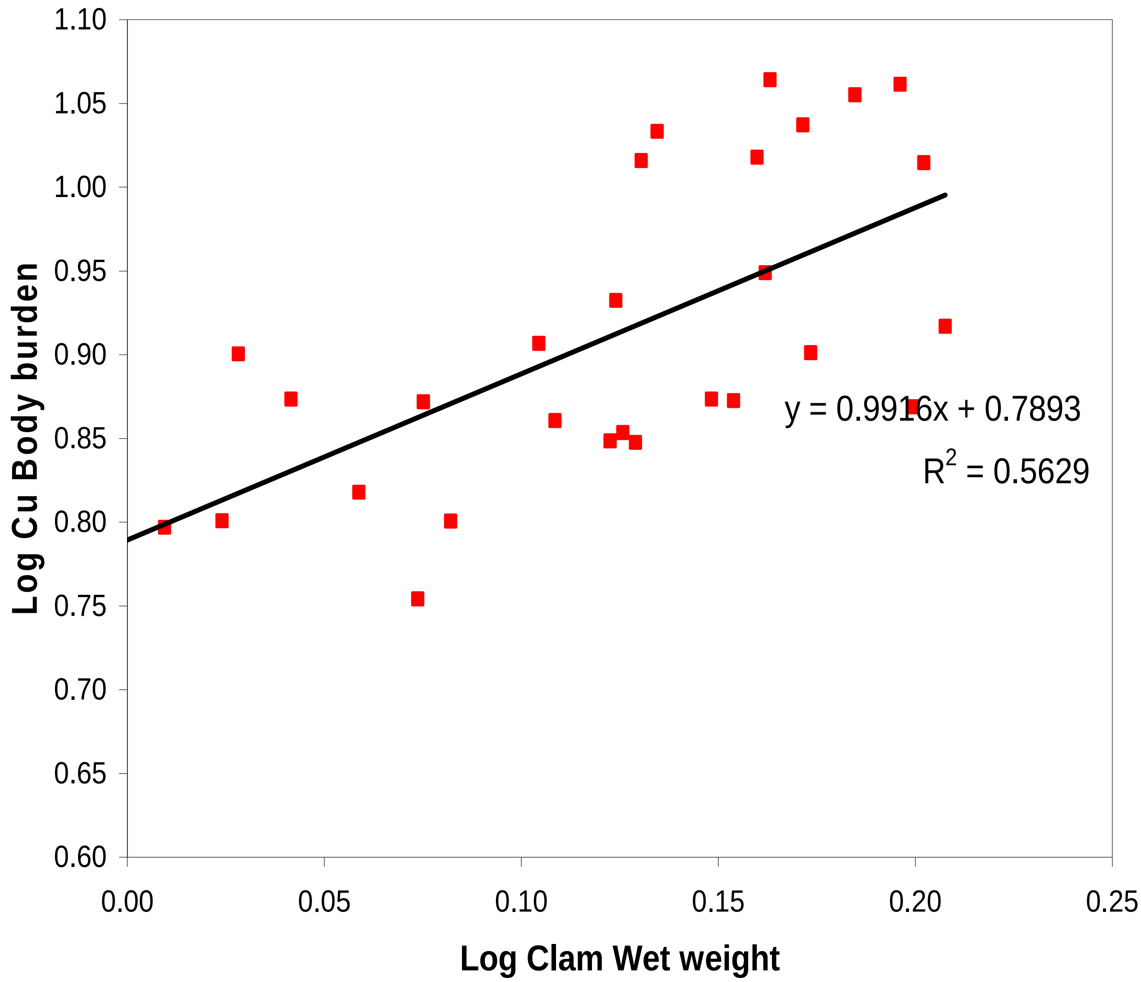

Figure 10. Correlation between Log wet weight and Log Cu body burden at the contaminated sites.

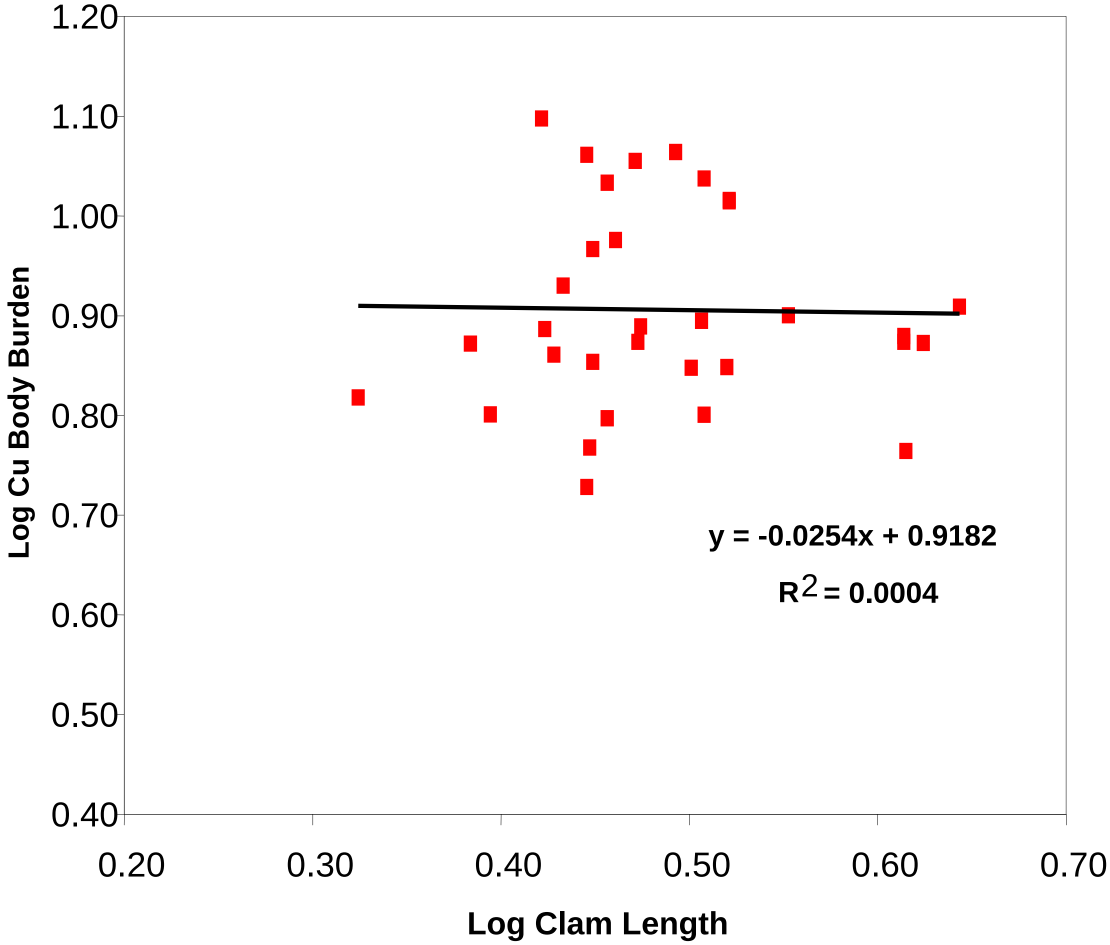

Figure 11. Correlation between Log clam length and Log Cu body burden at the contaminated sites.

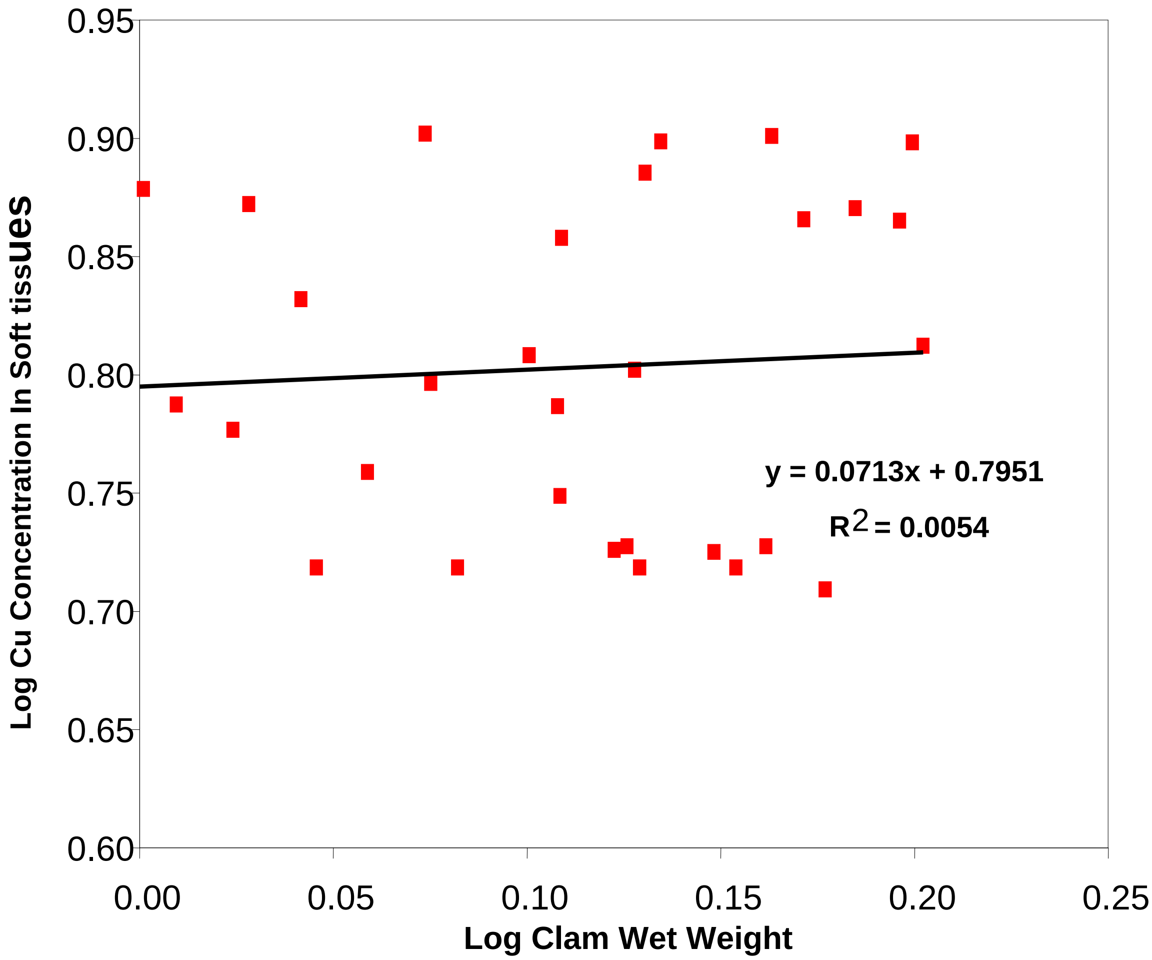

Figure 12. Correlation between Log clam total wet weight and Log Cu body burden at the contaminated sites.

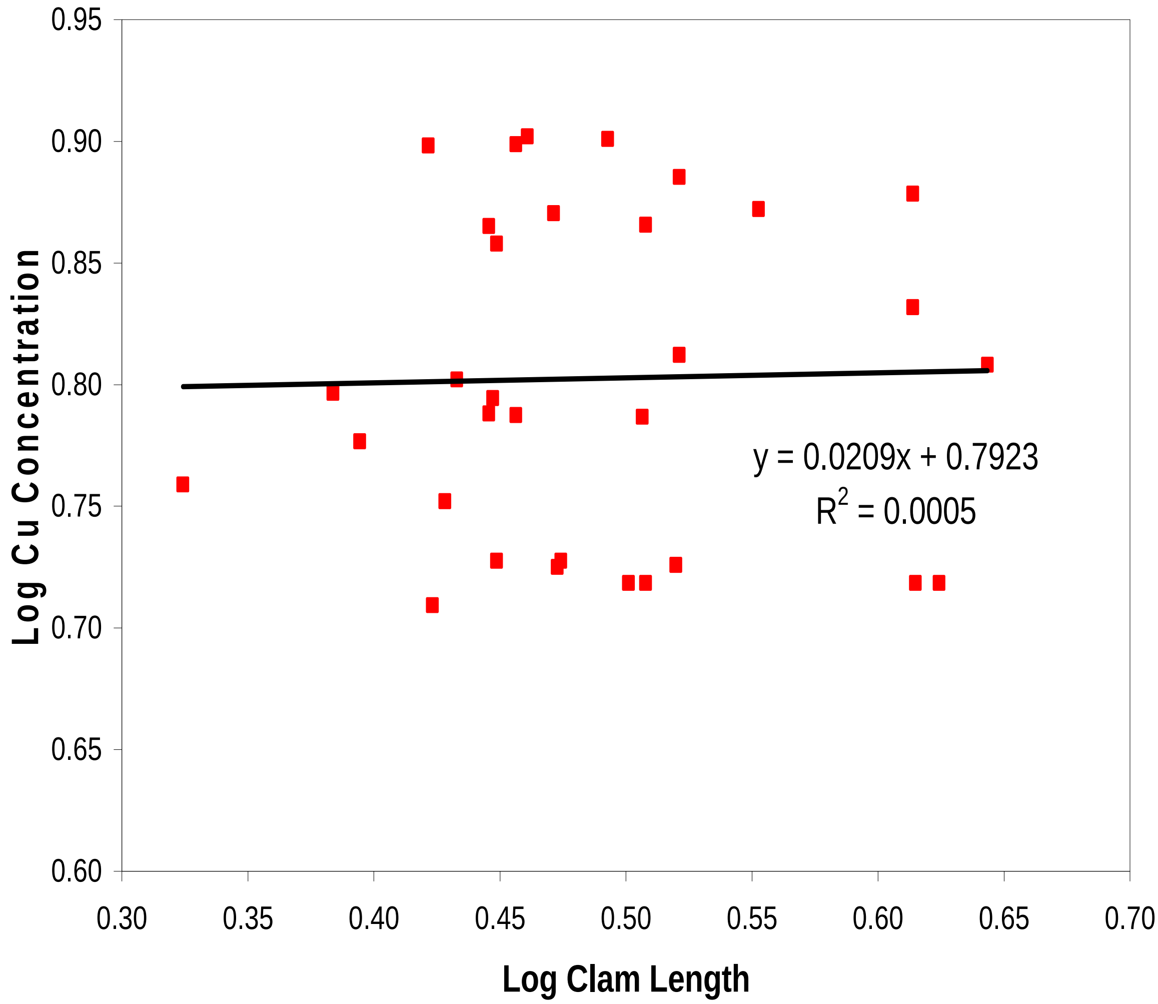

Figure 13. Correlation between Log clam length and Log Cu concentration in soft tissues at the contaminated sites.

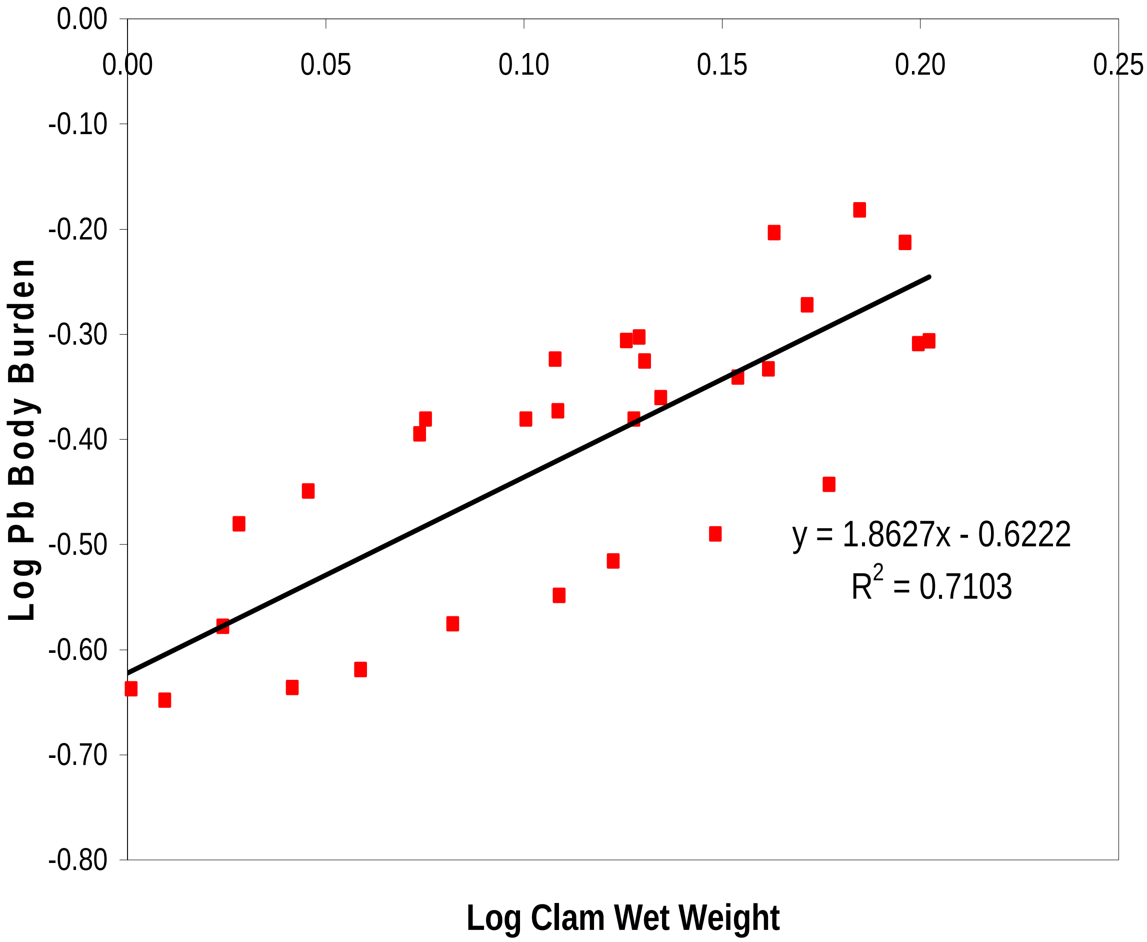

Figure 14. Correlation between Log total clam wet weight and Log Pb body burden at the contaminated site.

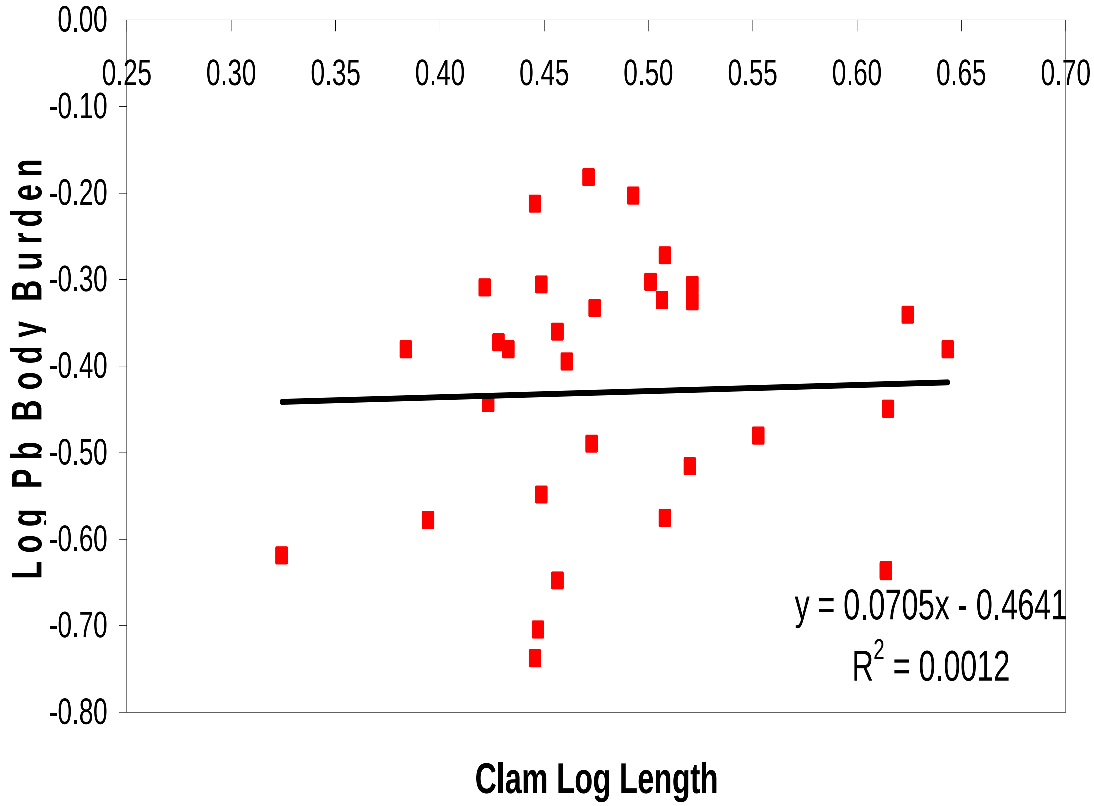

Figure 15. Correlation between Log clam length and Log Pb body burden at the contaminate sites.

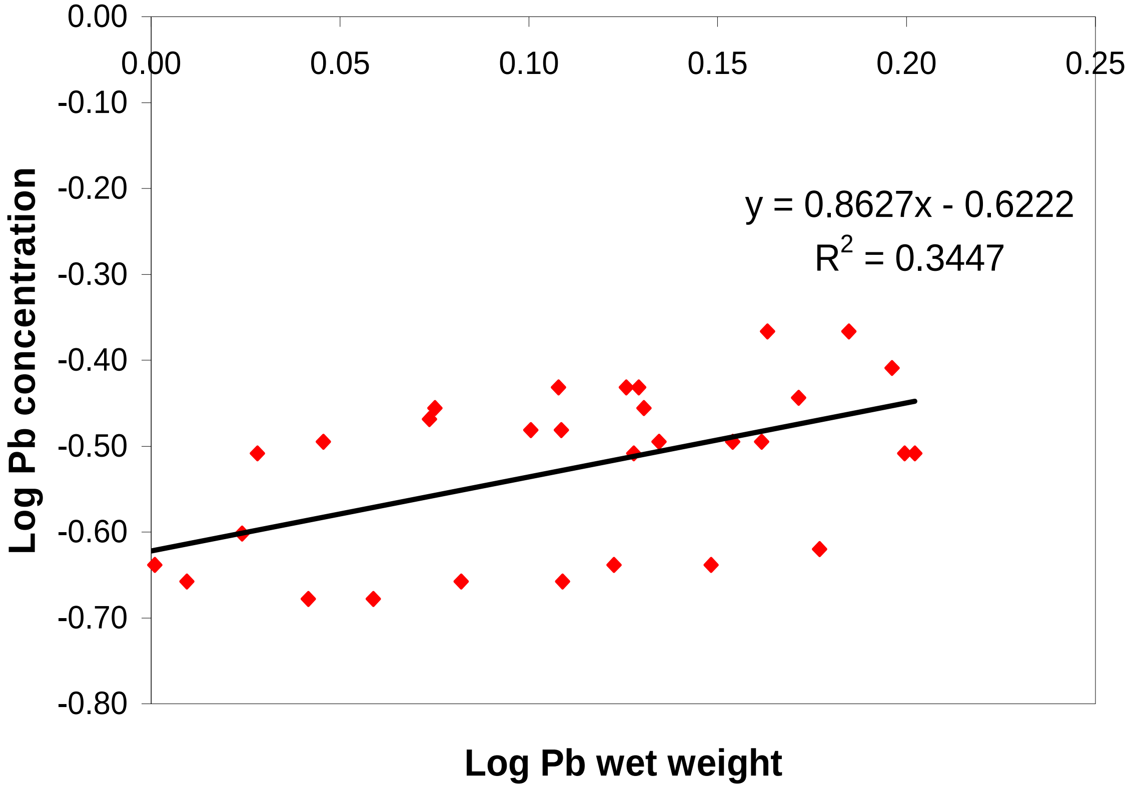

Figure 16. Correlation between Log clam wet weight and Log Pb soft tissue concentration at the contaminated sites.

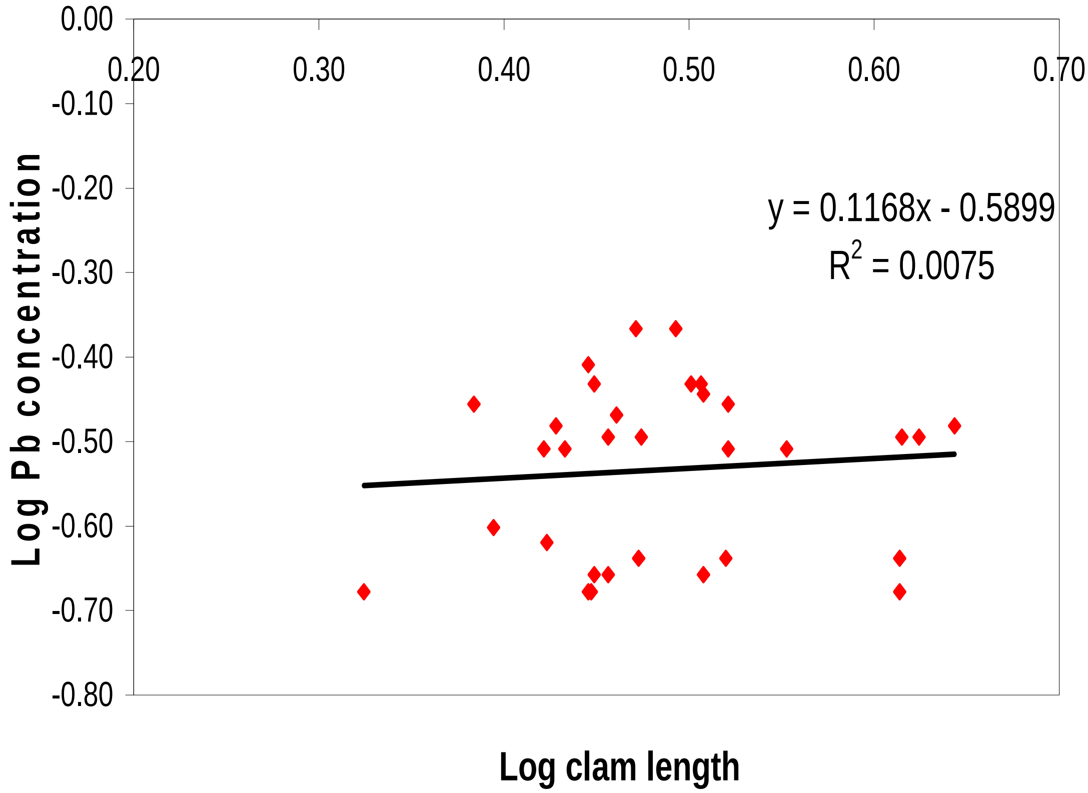

Figure 17. Correlation between Log clam length and Log Pb concentration at the contaminated sites.

4.6. Results

4.6.1. Correlation between length and wet weight of clam *A. umbonella*

A total of 33 clams were analyzed. Mean length and wet weight varied between 2.5cm - 5 cm for length (Mean=3.75 cm), and 2.6 mg- 3.48 mg for weight (Mean = 3.4) for the reference site clams. Regression between the clam length and wet weight showed a positive correlation (R^2=0.40) between shell length and wet weight at the reference sites.

Mean length and weight were between 3 cm - 5 cm for length (Mean = 4cm) and 0.8mg - 1.7mg wet weight (Mean = 85 mg) of the contaminated site clams. The length of the clam had no correlation (R^2=0.03) with wet weight at the contaminated sites. The length of the clam increased with minimal gain wet weight but weight increased without gaining length (Figure 1).

4.6.2. Correlation between length and wet weight of clam *A. umbonella*

The positive (P<.05) correlation found between log clam wet weight (Figure 2) and Log Cd body burden (R^2=21) and also between log length and log Cd body burden (R2=59). Figure. 5 showed the strong correlation (R^2=89, P< 0.05) between log clam length and log Cd concentration. There was no correlation between log clam wet weight and log Cd concentration (Figure 4).

There was a strong positive correlation (Figure 6) between log clam total wet weight and log Hg body burden (R^2=67, P< 0.05)). There was no correlation (R2=0004) between log clam length and log Hg body burden (Figure 7) and between log clam length and log Hg concentration (Figure. 5, 6).

The strong correlation ($p<0.05$, Figure 10) were found between log clam wet weight and log Cu body burden (R^2=54). There was no correlation between log clam length and log Cu concentration and also between log wet weight and log Cu concentration (Figures 7, 8, 9).

There was a strong positive correlation (R^2=71, $P<0.05$)) between log wet weight and log Pb body burden (Figure 14). There was no correlation between log length and log Pb body burden and also no correlation between log length and log Pb concentration (Figures 13, 14, 15).

4.7. Discussion

The main findings in this study were: 1) the insignificant correlations between the length and wet weight of clams from the contaminated sites; 2) Cd body burden increased with clam wet weight/length and Cd concentration also increased with clam's length at the contaminated site clams; 3) body burdens of Hg, Pb and Cu were positively correlated to clam wet weight, however, the concentrations of these metals were not correlated with clam length or weight at contaminated sites.

4.7.1. Correlation between length and wet weight of clam A. umbonella

The mean soft tissue wet weight was lower by a factor of 2.2 in the contaminated sites clams compared to the reference site clams (chapter 2), moreover, clam length and tissue wet weight were strongly correlated at the reference site clams, but were not correlated at the contaminated clams. The lack of growth as indicated by no soft tissue weight increase as length increased was indicative of the toxic effects in the clams from the contaminated sites. Although some clams increased in shell length, they did not gain weight. Thus, although the elevated levels of metals and perhaps other contaminants suppress tissue growth, they were not lethal. An apparent effect of heavy metal contamination on wet weight is the possibility that these clams were more tolerant of a wide range of metal contamination. This variation may explain the insignificant correlations between the length and total wet weight of the contaminated site clams. Bivalve exhibited the ability to exist under stressful condition and bio-concentrate contaminants without harming themselves (Goldberg, et al,. 1978; Jenkin, 1981).

4.7.2. Correlation between Cd body burden and log wet weight of clam A. umbonella

Cd body burden increased with clam wet weight/length and Cd concentration also increased with clam's length. The positive correlation between clam length and Cd concentration perhaps could be due to the biomagnifications which is a well known process in the aquatic food web (D' Itri 1991), or may be explained by the extremely slow rates of elimination from the body. Cd has no known biological use in animals (although it may substitute for zinc in certain enzymes in phytoplankton; Lane and Morel, 2000. Cd might biomagnify if consumers efficiently assimilate and slowly lose it (Reinfelder et al., 1998; Wang 2002). Langston (1987) also observed a positive relationship between metal concentration and length in some bivalve species due to the extremely slow rates of elimination of a metal from the body of an organism, coupled with non regulatory uptake. Since soft tissue weight did not increase with shell length, the increase in Cd concentration with shell length occurred in the near absence of an increase in soft tissue weight. Therefore, the increase in Cd concentration with shell length indicates increase with clam age. It is well known that cadmium readily accumulates through food webs (Fisher & Reinfelder, 1995; Nott, 1998). However, the size of the animal could be mainly responsible for the Cd concentration variability. Clams are part of a complex food web and the benthic invertebrates are an important link between phytoplankton and fish (Berkman and Nigro, (1992). Benthic organisms often lengthen the food chains and causes further biomagnifications (Bargagli et al., 1996).

4.7.3. Correlation between Hg, Cu and Pb body burden and wet weight/length of clam A. umbonella.

In general body burden is directly related to body weight in several species, for a variety of element (Boyden, 1974). Indeed, body burdens of Cd, Hg, Cu, and Pb in *A. umbonella* did increase with wet weight. The amount of metal, however, may not depend on the amount of binding compounds within the tissues, but on the amount of metal available in that environment. Thus both biotic and a-biotic factors probably change their relative importance in these bivalves and it is unlikely that a steady state could ever be reached (Otchere Fred, 2003).

The changes in metabolic rates of bivalve with length, storage mechanism as well as the variation in bioavailability of metals in the surrounding environment with time might be responsible for these correlations in these mollusks (Boyden, 1974; Cossa and Rondeau, 1985).

The tendency to decrease the metal concentrations with an increase in body weight/length was not significant for Hg, Cu and Pb. Perhaps growth was not more rapid than the accumulation rate. It is likely that when tissues grow more quickly than the metal can be absorbed, there will be reduction in metal concentration in soft tissues (Ahn, et al., 2001). Cu concentration in cockles in dry season decreases with size/length as a "growth dilution" effect, but is stable in the wet season. Thus alterations in the concentration of metals reciprocated with those of the whole soft tissues weight (Otchere Fred, 2003).

4.8. Summary

Among metals, only Cd increased with wet weight and length. One of the reasons for the strong positive correlation of Cd concentration and body burden (consistent with wet weight/length) is very likely due to bio-magnification, which is a well- known process in the aquatic food web (D' Itri 1991). Cadmium, one of the most toxic heavy metals (US EPA 1999; Langston 1990), is of particular importance in marine ecosystems (Fowler, 1990). Further investigation will be needed to verify the ecological factors involved in this process. The body burdens of the other metals (Hg, Pb and Cu) were positively correlated to clam wet weight. However, the concentrations of these metals were not correlated with clam length or weight. This indicates that the concentrations of Hg, Pb, and Cu are biologically regulated in *A. umbonella*.

5. Conclusions of the Dissertation

There is a need to critically assess the present quality of the marine ecosystem, especially the connection between ecosystem change and threats to human health. In this study an indigenous clam *Amiantis umbonella* was used as an effective bio-indicator species for desalination plant and steam power plant's waste water discharge in Kuwait. In addition to its use as a bio-indicator, the high levels of some metals are a concern as these bivalves are consumed by human and wildlife. Three metals had high enough levels in clams from Kuwait to suggest that they pose a risk to people who eat them. Bivalves are capable of accumulating extremely high levels of Cd in edible portions and therefore represent a greater hazard to human consumers than Cd does to other marine organisms (EPA, 1978; Langston et al., 1998; Roesijiadi, 1993). Among metals, only Cd increased with wet weight and length of the clam *A. umbonella*. One of the possible reasons for the significant correlations of Cd concentration and body burden (consistent with wet weight/length) is bio-magnification, which is a well known process in aquatic food webs (D' Itri 1991; Luoma et al., 1996; Chong and Wen-Xiong Wang, 2000). Cd might bio-magnify if consumers efficiently assimilate and slowly lose it (Rienfelder et al., 1998; Wang, 2000). Cd, one of the most toxic heavy metals, is of particular importance in marine ecosystems (Fowler, 199 ;US EPA 1999; Langston, 1990). Pb level in clams from Kuwait was 1.9 to 2.00 ppm that could pose a hazard to human consumers.

WHO (1976) sets a limit of 0.5 ppm for mercury in shell fish for human consumption, the U.S. level is 1 ppm (FAO, 2005). The mean levels of Hg in clams from Kuwait ranged from 0.6 - 6.2 ppm, which also exceeded the limit levels by WHO and

U.S. FDA. Metal levels in isolated tissues of clam *A. umbonella* are useful for identifying specific organs that may be particularly selective and sensitive to accumulation of heavy metals. Several inter- metal correlations between organs and water, as well as organs and sediment, suggest the use of these organs as bio-indicators for assessing changes in the marine coastal environment. In particular, gill accumulated all seven metals (Cu, Cr, Cd, Hg, Ni, V, Fe), indicating that the gill may be the most suitable bio-indicator for changes in metal levels (solute) in the surrounding sea water. The Kidney of clam *A. umbonella* also strongly accumulated most of the metals and therefore serves as a useful bio-indicator. For Cu and other metals associated with food and particulate matter, the digestive gland would serve as a better bio-indicator (Ahn, In-Young et al).

This study indicates that clams can serve as an early detection of potential marine-based contaminants, thus allowing for the protection of marine ecosystems and the prevention of associated human exposure. It also provides baseline data for future sustainable monitoring programs of the coastal environment, to reveal especially for Hg, Pb and Cd.

6. Future Direction

Further investigations will be needed to verify the ecological factors (age, trophic transfer) involved in Cd bio-magnification processes.

7. References

Abufayed, A. A. (2003). Impacts of desalination on coastal environment Middle East and North Africa region. INOC-INBA International Workshop on Desalination & Marine Pollution,

Ahn, L-Y., Chung, H., Choi, K.S. (2001a). Some ecological features of the Antarctic clam, *Laternula elliptica* (King and Broderip) in a near shore habitat on King George Island. Ocean Polar Res. **23:** 419-424.

Ahn, L-Y., Kang, J., Kim, K-W. (2001). The effect of body size on metal accumulations in the bivalve *Laternula elliptica*. Antarctica. Sci. B; **13:**355-362.

Ahn, L-Y., Lee, S.H., Kim, K.T.,Shim, J.H., Kim, D.Y. (1996). Baseline heavy metal concentrations in the Antarctic clam., *Laternula elliptica* (King and Broderip) in Maxwell Bay, King George Island. Antarctica. Mar. Pollut. Bull.; **32:**592-598.

Al Bakri Dhia & Kittaneh Wajeh. (1998). Physicochemical Characteristic and Pollution Indicators in the Intertidal Zone of Kuwait : mplication for Benthic Ecology: Environ. Management ; **22 (3)**415-424.

Al Bakri. D., A.Khuraibet, & M. Behbehani. (1997b). Quantitative assessment of the intertidal environment of Kuwait—11: Controlling factors. Journal of Environmental Management .

Al-Mafda, H., M.A.R. Abdel-Moati & F.H. Al-Gimaly. (1998). *Pinctada radiata* (Pearl Oyster): A Bio-indicator for Metal Pollution Monitoring in the Qatari Waters (Arabian Gulf). Bull. Environm. Contam Toxicol. **60:** 245-251.

Al-Majeed N.B & RaJab, W.A. (1998). Levels of mercury in the marine environment of the ROPME sea area. In: Otsaki, Abdulrahim, M.Y. and Reynolds, R.M., Editors, Offshore Environment of the ROPME Sea Area after the War-Related oil Spills, Terra Scientific, Tokyo, pp. 124-147.

Al-Mohanna and Subrehmanyam, M. N. V. (2001). Flux of heavy metal accumulation in various organs of the intertidal marine blue crab, Portunus pelagicus (L) from the Kuwait coast after the Gulf War. Environ. Int. **27,** pp. 321-326.

Aly Mohammed Aly Abdallah. (2003). Impact of desalination on the marine environment. INOC-INBA International Workshop on Desalination & Marine Pollution.

Al-Yamani. F., JBishop et al . (2001). A strategic plan for sustainable utilization of Kuwait;s Marine Environment. Kuwait Foundation for the advancement of Sciences (Publisher). pp. **40**

Amiard-Triquet C, Rainglet F, Larroux C, Regoli F, Hummel H. (1998). Metallothioneins in Arctic bivalves. Ecotoxicol Environ Saf;41: 96-102.

Amiard-Triquet, C., Berthet, B. and Metayer, C. and Amiard. (1986). Contribution to the ecotoxicological study of cadmium, copper and zinc in the mussel *Mytilus edulis*. 11. Experimental study. Mar. Biol; **92**: 7-13.

Augusto-R. Carlos. E Silva, Brian D. Smith and Phillip S. Rainbow. (2006). Comparative bio-monitors of coastal trace metal contamination in tropical South America (N. Brazil), Marine. Environ. Research; **61**, issue **4**:439-455.

Awadhi, F.(1999). The year of the ocean and its crucial importance to the Gulf. Desalination, 123:127-133. Bayne, B, L., Thompson, R. J., and Widdos, (1976a), Physiology: 1.In “Marine Mussels: Their Ecology and Physiology” (B. L. Bayne, Ed.) International Biological Programme Synthesis, pp. 121-206. Cambridge Univ. Press, London and New York.

Ball. PW. (1985). Copper, Lead, and Cadmiumin in coastal waters of the western North Sea. Marine Chemistry. **15**: 363-378.

Bargagli, R., L. Nelli, S. Ancora, and S. Focardi. (1996). Elevated cadmium accumulation in marine organisms from Terra Nova Bay (Antarctica). Polar Bio; **l16**:513-520.

Bebianno MJ, Nott JA, Langston WJ. (1993). Cadmium metabolismin the clam *Ruditapes decussata:* the role of metallothioneins. Aquat Toxicol; **27**:315-334.

Belanger, S. E., L.J. Farris, D.S. Cherry & J. Cairns. (1990). Validation of Corbicula fluminea growth reductions induced by copper in artificial streams and river systems. Can. J. Fish. Aquat. Sci. **47:**904-914.

Beliaeff, B., O'Connor, T. P., Daskalakif, D. K. and Smith, P. J. (1997). US mussel watch data from 1986±1994. Temporal trend detection at large spatial scales. American Journal of Chemistry and Society **31** (5), 1411±1415.

Beliaeff B, O'connor TP, Claisse, D. (1998). Comparison of chemical concentrations in mussels and oysters from the United States and France. Environ Monit and Assess; **49**:87-95..

Bellos D & Sawidis T, 2005, Chemical pollution monitoring of the river Pinios. Journal of Environ. Management; V:**76,4**, pp. 282-292.

Berkman, P.A. and M. & Nigro (1992). Trace metal concentrations in Scallops around Antarctica: extending the Mussel Watch Programme to the Southern Ocean. Mar. Poll. Bull; **24**:322-

Bodek, I., L. W. Lyman, W.F. Rechi & D. Rosenblat. (1995). Environmental Inorganic Chemistry. 1988. Pergamon Press, NewYork.

Borchardt, T. (1983). Influence of food quantity on the kinetics of cadmium uptake and loss via food and sea water in Mytilus edulis. Mar. Biol. **76.** pp. 67-76. Abstract-EMBASE.

Boyden CR, Phillip DJH. (1981). Seasonal variation and inherent variability of trace elements in oysters and their implications for indicator studies. Mar. Ecol. Prog. Ser. **5**:29-40.

Boyden, C.R. (1974). Trace metals content and body size in molluscs. Nature **251**: 311-314.

Boyden, C.R. (1977). Effect of size upon metal contents of shellfish. J. Mar. Biol. Ass. UK, **57** : 675-714.

Brown, C.L. and Luoma, S.n. (1995). Use of the euryhaline bivalve *Potamocorbula amurensis* as bio-sentinel species to assess trace metal contamination in San Francisco Bay, Marine Ecology Progress Series;**124**:129-142.

Bryan, GW, Langston, W.J. Hummerstone, L.G, Burt GR. (1985). A guide to the assessment of heavy metal contamination in estuaries using biological indicators. Occasional Publication Number 4, Marine Biological Association of the United Kingdom. Plymouth.

Bryan, et.al. (1985). Heavy metals in the burrowing bivalve *Scrobicularia plana* from the Tamer estuary in relation to environmental levels. J. Mar. Biol. Ass. U.K ; **58**: 89-108.

Bryan, G. W. and P. E. Gibbs. (1983). Heavy metals in the Fal Estuary, Cornwall: A study of long-term contamination by mining waste and its effects on estuarine organisms. Occusional Publication, Marine Biological Association of the United Kingdom

Bryan, G. W. (1980). Recent trends in research on heavy metal contamination in the sea. Helgol. Meeresunters., **33**:6-25

Bryan, G.W. (1973). The occurrence and seasonal variation of trace metals in the scallops *Pecten maximus* (L) and *Chlamys opercularis* (L). J. Mar. Biol. Ass. U. K. **53**: 145-166.

Bryan, G.W., Langston, W. J. & Hummerstone, L.G. (1980). The use of biological indicators of heavy metal contamination in estuaries. Occasional Publ. Mar. Biol. Ass. U.K., **1,** 1-73.

Bryan, G.W., Langston, W.J. hummerstone, L.G. and Burt, G.R. (1985).

A guide to the assessment of heavy metal contamination in estuaries using biological indicators.

Bryan, G.W. (1977). Indicators of heavy metal contamination in the loose estuary (Cornwall) with particular regard to silver and lead . J. Mar. Biol. Ass. UK, **57**:75-92.

Bu-Olayan and BV Thomas. (2006). Assessment on biocides bioaccumulation in *MulletLiza klunzingeri* in Kuwaiti waters, off the Arabian Gulf. American Journal of environmental science ; **2** (**3**):109-113.

Bu-Olayan. A. H., Subrahmanyam, M.N.V. (1996). Heavy metals in marine algae of the Kuwait coast. Bull. Environ. Toxicol.,; **57**: 816-825.

Bu-Olayan. A. H., Subrahmanyam, M.N.V. (1997). Accumulation of copper, nickel, lead and zinc by snail, Lunella coronatus and pearl oyster, Pictada from the Kuwait coast before and after the Gulf War oil spill. Sci. Total Environ; **197**: 161-165.

Burdon-Jones, C and Denton, G, R, W. (1984). Metals in Marine Organisms from the Great Barrier Reef Province. Final Report, Part 2. Department of Marine Biology, School of Biological Sciences, James Cook University, Townsville, QId.

Burger J. (2001). Gochfeld M. On developing Bio-indicators for Human and Ecological Health. Environ Monit Assess ; **66**:23-46.

Burger J. (2002). Food Chain Differences Affect Heavy metals in Bird Eggs in Barnegat Bay, New Jersey. Environmental Research Section A; **90**, 33-39

Burger J. (2006). Bio-indicators: Types, Development, and use in Ecological Assessment and Research. Environmental Bioindicators, **1**:22-39.

Burger J., Gochfeld, M. (2004). Metal Levels in eggs of common terns (Sterna hirundo) in New Jersey: Temporal trends from 1971 to 2002) Environmental Research **94**, 336-343

Burger, J. (1997). Heavy metals and selenium in herring gulls (Larusution: functional argentatus). Nesting in colonies from eastern Long Island to Virginia. Environmental Monitoring and assessment.

Calabrese, A., J. R. Maclnnes, D.A. Greig, and P.P. Yevich. (1984). Effects of long-term exposure to silver or copper on growth, bioaccumulation and histopathology in the blue mussel Mytilus edulis. Mar. Environ.; **11,** 253-274..

Campanella. L. Conti M E., Cubadda. F. & Sucpane, C. (2001). Trace metals in seagrass, algae and mollusks from an uncontaminated area in the Mediterranean. Environ. Poll. :**111,** 117-126.

Campbell, P. G. C. Campbell. (2003). Interaction between trace metals and aquatic organisms: a critique of the free- ion activity model, In:A. Tessier and D.R. Turner, Editors, Metal Speciation and Aquatic Systems, Wiley, NewYork (1995), pp. 45-10

Campbell, P.G.C., 1995. A critique of the free ion activity model. In: Tessier, a. and Turner. D. R., Editors. (1995). Metal Speciation and Bioavailability in aquatic systems, John Wiley, Chchester, pp. 45-102.

Cantillo, A.Y. and O;Connor, T.P. (1992). Trace element contaminants in sediments from the NOAA, National Status and Trends programme compared to data from throughout the world. Chemistry and Ecology **7**, pp. 31-50

Cheggour. M., Langston, W.J., Chafik, A., Texier, H. and Idrissi, H. (1999). Phosphate Industry discharges and their impact on metal contamination and intertidal macrobenthos: Jorf Lasfar and Safi coastline (Morocco). Toxocology and Environmental Chemistry **70**, pp. 159-179.

Chong, I. and Wang, W-X. (2000). Assimilation of Cd, Cr, and Zn by the green mussel *Perna Virid* and the clam *Ruditapes philippinarum*. Environ. Toxicol. Chem. **19,** pp. 1660-1667.

Chong, I. and Wang, W-X. (2001). Comparative studies on the biokinetics of Cd, Cr, Zn in the green mussel *Perna viridis* and the Manila clam *Ruditapes philippinarum*. Environ. Pollut. V, **115**, Issue 1, Nov 2001, pp.107-121.

Chouikhi. A (2003). Desalination Technology and impact on the coastal environment. INOC-INBA International Workshop on Desalination & Marine Pollution,

Claisse D, Cossa D, Bretaudeau-sanjuan J, Youchard G, Bombled B. (2001). Methylmercury in mollusks Marine. Pollut. Bull;**42**: 329-332.

Claisse D, (1989). Chemical contamination of French coasts. The results of a ten years mussel watch. Marine Pollut Bulle **20 (10**): 52-528.

Cossa D, Coquery M, Nakhle K, Claisse D. (2002). Determination of mercury and monmethylmercury in marine biota and sediments. Methods Anal Milea Mar.; **1-** 16.

Cossa, D., Bourget, E. and Piuze, J. (1979). Sexual maturation as a source of variation in the relationship between cadmium concentration and the body weight of the *Mytilus edulis*. Mar. Pollut. Bull. **10,**pp. 174-178.

Cosson, R.P. (2000). Bivalve metallothionein as a biomarker of aquatic ecosystem pollution by trace metals: limits and perspectives. Cell. Mol. Biol. 46, 295-309.

Cunningham, P A. (1979). The use of bivalve mollusks in heavy metal pollution research. In “Marine Poll responses” (W. B. Vernberg, F. P. Thurberg, A. Calabrese and F. J. Vernberg, eds), pp. 183-221. Acadmic Press, New York.

D. J. H. (1980a). Quantitative aquatic biological indicators: their use to monitor trace metal and organochlorinepollution. Applied Science Publishers Ltd.,

D’ v Itri. (1991) DiToro, D. M., J. D. Mahony, D. J. Hanse Scott, M. B. Hicks, S. M. Mayr, and Redmond, M. S., 1990, Toxicity of cadmium in sediments: The role of acid volatile sulfide: Environmental Toxicology and Chemistry,.**9,** p. 1487-1502.

Dallinger, R. and Rainbow, P. (1993). Ecotoxicology of metals in invertebrates. Boca Raton, Florida. CRC Lewis Publishers/SETAC. USA

Dauer, D. M., C. A. Maybury and R.M. Ewing. (1981). Feeding behaviour and general ecology of several spionid polychaetes from the Chesapeake Bay, J. Exp. Mar. Biol. Ecol., **54**, 21-38.

Denton, G, R, W, and Burdon-Jones, C. (1986). Trace metals in fish from the Great Barrier Reef. Marine Pollution Bulletin **17**, 201-209.

Depledge, M.H. & P.S. Rainbow. (1990). Cited by Rainbow (1993). The significance of trace metal concentrations in marine invertebrates p. 3-24. In R. Dallinger and P.S. Rainbow (eds.). Ecotoxicology of metals in invertebrates. Lewis, Boca Raton, Florida.

Diaz, O., Ortega, O.,Del Valle, S., Encina, F., Munoz, G, F., Figueroa, A.(2001). Journal of Architecture and culture **1,** 122-131.

Eisler, (1981). Trace metal concentration in marine organisms. Pergamon, New York, 687p.

Emara, I. Hossssny, 2003.

EPA. (1978). Reviews of the environmental effects of pollutants; 1V. Cadmium. U.S. Environmental Protection Agency, ORNL/EIS-106, 600/1-78-026. 251 p.

Evtushenko ZS, Belcheva NN, Lukyaanova ON. (1986). Cadmium accumulation in organs of the scallop Mizuhopecten yessoensis. 1.Activities of phosphatases and composition and amount of lipids. Comp Biochem Physiol; **83** C:371-376.

Farrington, J.W., Davis, AC., Tripp, B.W., Phelps, D.K. and Galloway, W.B. (1987). Mussel Watch Program Measurement of chemical pollutants in bivalves as one indicator of coastal environmental quality. New Approaches to Monitoring Aquatic Ecosystems

FDA. (1993). Guidance Document for nickel in Shellfish. U.S. Food and Drug Administration. Center for food Safety and Applied Nutrition. Washington, DC.

FDA, (1993). FDA, Guidance document for cadmium in shellfish, Centre food and safety and applied nutrition, Food and Drug Administration, Washingtion (DC) (1993).

FDA, (1993). Guidance document for cadmium in shellfish, Centre for food and safety and applied nutrition, Food and Drug Administration,Washington (DC,). Fialkowski, W. And New man, W. A.(1998). A pilot study of heavy metals accumulations in a barnacle from the Salton Sea, Southern California. Marine Pollution Bulletin **36**, 138-143.

Fisher, N.S. and Reinfelder, J.R.(1995). The trophic transfer of metals in marine systems. In:Tessier, A. Turner, D.R., Editors, 1995. Metal Speciation and Bioavailability in Aquatic Systems, John WILEY, NEW, PP. 363-406.

Fowler , S. W. (1990). Critical review of selected heavy metal and chlorinated hydrocarbon concentrations in the marine environment. Mar. Environ.; **29**: 1-64.& Oregioni 1976.

Fowler, S. W. (1990). Critical review of selected heavy metal and chlorinated hydrocarbon concentrations in the marine environment. Marine Environmental research**; 29.1.**

Fowler, S.W. & B. Oregioni. (1976). Trace metals in mussels from the NW Mediterranean. Mar. Pollut. Bull**.** **7:** 26-29.

Gagne. F, Blaise, C, Pellerin, J, Strand, J. (2006). Health Status of *Mya arenaria* bivalves collected from contaminated sites in Canada (Saguenay Fjord) and Denmark (Odense Fjord) during their reproductive period. Ecotoxicol & Environment Safety; **64**:**3**, 348-361.

Galtsoff, P.S. (1964). The American Oyster Crassostrea virginica Gmelin. Fish. Bull. Fish and Wild life Some. Government Printing Office, Washington, D.C. 480p.

Genest, P. E. & Hatch, W.I. (1981). "Heavy metals in Mercenaria and sediments from the New Bedford harbor region of Buzzard's Bay, Massachusetts,' Bull. Environ. Contamin. Toxicol. **26,** 124-130.

George, S.G., B.J.S. Pirie, A.R. Cheyne, T.L. Coombs & P.T. Grant. (1978). Detoxification of metals by marine bivalves: an ultrastructural study of the compartmentation of copper and zinc in the oyster Ostrea edulis. Mar. Biol. **45**: 147-156.

Goldberg, E. D. (1986). The Mussel Watch concept. Environmental Monitoring and Assessment **7:**91-103.

Goldberg, E.D., Bowen. V.T, Farrington, J.H., Harvey, G., Martin, J.H., Parker, P.L., Riseborough, R.W., Robertson, W., Schneider, E. and Gamble, E. (1978). The Mussel Watch. Environment. Conver., 5:1-25. N.R.C. (1980). The International Mussel Watch: Report of a workshop. Washington DC, U.S. National Academy of sciences , National Research Council, Publications Office, 248p

Goldberg, E.D., M. Koide, V. Holdge, A.R. Flegal & J. Martin. (1983). U.S. mussel watch: 1977-1978 results on trace metals and radionuclides. Estuar. Coast. Shelf. Sci. **16**: 69-93.

Gunther, AJ, Davis JA, Hardin DD, Gold , Bell D, Scelfo GM, et al. (1999). Long- term bioaccumulation monitoring with transplanted bivalves in the San Francisco Estuary. Mar pollut Bull **38:**21-34. Heavy metals in the rock oyster Crassostrea iridescens

Hosch G. (1996). Prospective mapping of total cadmium, copper, zinc and lead contamination in water, sediment and clams (Chione gnidia) of Guaymas Bay, Sonora, Mexico. MSc. Thesis. University of Hull, England.

Hung T, Meng P, Han B, Chuang A, Huang C. (2001). Trace metals in different species of mollusca, water and sediments from Taiwan coastal area. Chemosphere; **44**:833-41.

Islam and Tanaka, M.D. Islam and M. Tanaka (2004). Impact of pollution on coastal and marine ecosystems includind coastal and marine fisheries and approach for management :a review and synthesis,. Marin. Pollut. Bull.; **48**:624-649.

Jassim A. Al-Khayat and Mohammed. S.Al-Mohannadi. (2006). Egyptian Journal of Aquatic Research Vol. **32:** 1, 419-430.

Jensen, K.,Randlov, A.. &Rissgard, H.U. (1981). Heavy metal pollution from a point source demonstrated by means of mussels, *Mytilus edulis*. Chemosphere, 10, 761-5. Journal of the Marine Biological Association of the United Kingdom **4**, p. 92.

Keen, A.M. (1971). Sea Shells of Tropical West America: Marine mollusks from Baja California to Peru. Stanford University, Stanford, California, 356p.

Kistner, W. (1984). International Mussels Watch. Concerned scientists gather data world wide. Oceans **17** (November); 64-67.

Langston & Spence, (1995). W.J, Langston and S.K. Spence, Biological factors involved in metal concentrations observed in aquatic organisms. In: A. Tessier and D.R. Turne, Editors, Metal Speciation and Bioavailability in Aquatic System, Wiley, Chichester, UK (1995), pp.407-478

Langston, W. j. & Zhou, M. (1987). Cadmium accumulation, distribution and elimination in the bivalve *Macoma Balthica*; neither metallothionein-like proteins are involved. Mar. Environ. Res. ;**21**: 225-237.

Langston, W. J. (1990). Toxic effects of metals and the incidence of metal pollution in marine ecosystems. P. 101-122. In: Heavy metals in the marine environment. Eds. By R.W. Furness and P.S. Rainbow. CRC Press. Boca Raton.

Langston, W. J., Bebianno, M.J., Burt, G. R. (1998). Metal handling strategies in mlluscks. In: Langston, W.J., Bebianno, W.J. (Eds), Metal metabolism in aquatic environments. Chapman and Hall, London,; pp. 219-283.

Latouche, Y.D & M.C. Mix. (1981). Seasonal variation in soft tissue weights and trace metal burdens in the Bay Mussel, Mytilus edulis. Bull. Environ. Contam. Toxicol. **27**: 821-828.

Lauenstein, G. G., Robertson, O’Connor, T. P. (1990). Comparison trace metal data in mussels and oysters from a mussels watch programme of the 1970s with those from a 1980sprogramme. Mar. Pollut. Bull. **21**: 440-447.

Liang, N. L.He, B. Jiang, G.B. Chen, D.Y. Yao, Z.W. (2004). Evaluation of mollusks as biomonitors to investigate heavy metal contaminations along the Chinese Bohai Sea. Science of the Total Environ; **324** :105-113.

Lobel, PB, Mogie P, wright DA, Wu Bl. (1982). Metal accumulation in four mollusks. Mar Pollut Bull 13:170-174.

Luoma, S,N., D.J. Cain, K. Ho and A. Hutchinson. (1983). Variable tolerance to copper in two species from San Francisco Bay. Mar. Environ. Res., Vol., 10, pp. 209-222.

Luoma, S. N. (1993). Bioavailability of trace metals to aquatic organisms---a review, Sci. Tot. Environ. **28:**1-22.

Luoma, S. N. Cain, D. & Johansson, C. (1985). Temporal fluctuations of silver, copper and zinc in the bivalve Macoma balthica at five stations in south San Francisco Bay. Hydrobiologia;**129**:109-120.

Luoma, S. N. (1989). Can we determine the biological availability of sediment-bound trace elements? Hydrobiologia;: 176-177, 379-396.

Luoma, S. N. (1990). Processes affecting metal concentrations in estuarine and coastal marine sediments. In Heavy Metals in the Marine Environment (R. W. Furness & P. S. Rainbow, eds), 51-66.CRC Press.

Luoma, S.N. and E. A. Jenne. (1977). The availability of sediment bound cobalt , silver and Zinc to a deposit feeding clam. In: Biological implications of metals in the environment, edited by R. W. Wildung and h. Drucker, ERDA Conf. 750920, NTIS, Springfield, VA, pp. 213-230.

Luoma, S.N. and Fisher, N.S. (1997). Uncertainties in assessing contaminant exposure from sediment. In: Ingersoll, C. G., Dillon, T. and Biddinger, G.R., Editor, 1997. Ecological Risk Assessment of Contaminated Sediments, SETAC Spec. PubI. Series, Pensacda, FL. pp. 211-237. Abstract + Reference in Scopus.

Luoma, S.N. Fisher,N.(1997). Uncertainties in assessing contaminant exposure from sediment. In “Ecological Risk Assessment of Contaminated Sediments”. (Eds T. Dillon and G. Biddinger) pp. 211-237.

Marcus, J. M. and Scott, G.I. (1990). The Use of Sediment Heavy Metal Bioavailability as a Method of Environmental Hazard Assessment, Aquatic Toxicology and Risk Assessment;: **13**, pp. 110-122.

Marcus, J. M. and Scott, G.I. (1990). The Use of Sediment Heavy Metal Bioavailability as a Method of Environmental Hazard Assessment, Aquatic Toxicology and Risk Assessment; **13**, pp. 110-122. BRYAN, G. W. 1979. Bioaccumulation of marine pollutants.

Marina, M. & O. Enzo. (1983). Variability of zinc and manganese concentrations in relation to sex and season in the bivalve Donax trunculus. Mar. Poll. Bull. **4**: 342-346.

Mendze, L, L. M. Salas,-Flores, A. Arreola-Lizarraga, S.T. (2002). Alvarez-Castaneda, B. Acosta, Heavy metals in clams from Guaymas Bay, Mexico. Bull. Environ. Contam. Toxicol; **68**: 217-223.

Miller, J.C. & J.N. Miller. (1987). Statistics for analytical chemistry. Wiley; New York.

Moody, J.R. & R.N. Lindstrom. (1977). Selection and cleaning of plastic containers for storage trace element samples. Anal. Chem**. 49:** 2264-2267.

Mora De Stephen, Fowler. W Scott, Wyse Eric, and Sabine Azemard, (2004). Ditribution of heavy metals in marine bivalves, fish and coastal sediment in the Gulf and Gulf of Oman. Marin. Pollu. Bull**; 49**: Issues 5-6, pp. 410-424.

N.R.C.(1980). The International Mussel Watch: Report of a workshop. Washington DC, U.S. National Academy of sciences, National Research Council, Publications Office, 248 pp

National Research Council (NRC). (1986). Ecological knowledge and environmental problem solving. Washington, DC: National Academy Press; National Research Council (NRC). Issues in risk assessment. Washington, DC: National Academy Press; 1993.

National Research Council (NRC). (1983). Risk assessment in the federal government. Washington, DC: National Academy Press;

Negri Andrew, Burns Kathryn, Boyle Steve, Brinkman Diane and Webster Nicole. (2006). Contamination in sediment, bivalves and sponges of McMurdo Sound, Antarctica. Environmental Pollution; **143**, V,**3**, pp 456-467.

Nott, J. A. Metals and marine food chains. (1998). In: Metal metabolisms in aquatic environments. Eds. By W. J. Langston and M.J. Bebianno. Chapman & Hall, London. pp. 387-414

O'Connor TP. (1992). Recent trends in coastal environmental quality :results from the first five years of the NOAA, National Mussel Watch Project. US Dept Commerce, NOAA, National Ocean Service, Washington, DC, p 46.

O'Connor, T, P. (1992) Recent trends in Coastal Environmental Quality: Results from the first five years of the NOAA Mussel Watch Project. P. 46. National Ocean

Service, National Oceanic and Atmospheric Administration, United States Department of Commerce, Rockville.

O'Connor, T. P. (2004) Trends in chemical concentrations in mussels and oysters collected along the US coast from 1986 to 1993. Mar. Environ. Res., **41**, 183-201.

O'Connor, T. P. (1898) Mussels watch results from 1986 to 1996. Marine Pollution Bull.1998**, 37,** (**1-2**), 14-19.

O'Connor, T.P. and Beliaeff, B. (1995). Recent Trends in Coastal Environmental Quality: Results from the Mussel Watch Project. NOAA/NOS, NOAA, Silver Springs, MD.

Odzak, N., Martinic, D., Zvonaric, T. and Branica, M. (1994). Bioaccumulation rate of Cd and Pb in M. galloprovincialis foot and gills. Marine Chemistry 46, 119±131.

Otchere FA, Joiris C, Holsbeek L, Ali IB, Vanderpuye CJ. (2000). Heavy metals concentration and burden in bivalves Anadara (Senilia), Perna perna and Cressostrea tulipa from Ghana; In: 11th annual International Conference of Heavy Metals in the Environment (J. Nriague, ed.) Contribution number 10161. University of Michigan, School of Public Health, Ann Arbor.

Otchere Fred A. (2003). Heavy metals concentrations and burden in the bivalves (Anadara (Senilia) senilis, Crassostrea tulipa and Perna perna) from Lagoons in Ghana: Model to describe mechanisms of accumulation/ excretion Afr. J. Biotechnol Vol. 2 No. **9:** 280-287.

Podgurskaya, O.V..Ya. Kavun and O.N. Lukyanova, (2004). Heavy metals accumulation and distribution in organs of the mussel *Crenomytilus grayanus* from upwelling areas of the sea of Okhotsk and the sea of Japan..Russian journal of Marine Biology,. V;**30**:188-195.

P. Irato, G. Santovito, A. Cassini, E. Piccinni, V. Albergoni. (2003). Metal Accumulation and Binding Protein Induction in Mytilus galloprovincialis, Scapharca inaequivalvis, and tapes philippinarum from the lagoon of Venice; Arch. Environ. Contam. Toxicol; **44:**476-484.

Páez-Osuna, F. & C. Marmolejo-Rivas. (1990 a). Trace Metal in Tropical Coastal Lagoon Bivalves; Crassostrea corteziensis. Bull. Environ. Contam. Toxicol. **45**: 538-544.

Páez-Osuna, F. & C. Marmolejo-Rivas. (1990 b). Occurrence and seasonal variation of heavy metals in the oysters *Saccrostrea iridescens* Bull. Environ. Contam. Toxicol. **44**: 129-134.

Páez-Osuna, F., J.I. Osuna-López, G. Izaguirre-Fierro & H.M. Zazueta-Padilla. (1993a). Heavy metals in oysters from a subtropical coastal lagoon associated with an agricultural drainage basin. Bull. Environ. Contam. Toxicol. **50**: 696-702.

Páez-Osuna, F., J.I. Osuna-López, G. Izaguirre-Fierro & H.M. Zazueta-Padilla. (1993b). Heavy metals in clams from a subtropical coastal lagoon associated with an agricultural drainage basin. Bull. Environ. Contam. Toxicol. **50**: 915-921.

Páez-Osuna, F., M.G. Frías-Espericueta & J.I. Osuna-López. (1995). Trace metals concentrations in relation to season and gonadal maturation in the oyster Crassostrea iridescens. Mar. Environ. Res. **40:** 19-32.

Paulson AJ, Sharack B, Zdanowicz. (2003). Trace metals in ribbed mussel from Arthur kill, New York/New Jersey, USA. Mar Pollut Bull **46:**139-52.

Peakall D. (1992). Animal biomarkers as pollution indicators. London: Chapman and Hall; Philips, 1976-The common mussel *Mytilus edulis* as an indicator of pollution by zinc, cadmium, lead and copper. Effects of the environmental variables on uptake of metals. Mar Biol; **38**, pp. 71-80.

Phillip DJH. (1980). Quantitative Aquatic Biological Indicators: Their use to monitor trace metal and organochlorine pollution. Lond. Appl, Sci. Ltd. U.K.

Phillip DJH (1990). In: Furness, R.W. and Rainbow, P.S. (eds) Heavy metals in the marine environment. CRC Press, Florida:pp. 81-99.

Phillip, D. J. H. and Rainbow, P.S. (1981). Bio-monitoring of Aquatic Trace Contaminants., Chapman and Hall, London.

Phillip, D.J.H. (1977). Effect of salinity on the net uptake of zinc by the common mussel *Mytilus edulis*, Mar. Biol., **41**, 79-88.

Phillip, D.J.H. (1976). The common mussel Mytilus edulis as an indicator of pollution by Zinc, Cadmium, Lead, and Copper. Effects of environmental variables on uptake of metals. Mar. Biol. **38,** 59-69.

Phillips, D. J. H. (1979). The rock oyster Saccostrea glomerata as an indicator of trace metals in Hong Kong. Mar. Biol. **53**: 353-360 13, Food. 26-29

Phillips, D. J. H., Yim, W. W.-S. (1981). A comparative evaluation of oysters, mussels and sediments as indicators trace metals in Hong Kong waters. Mar. Ecol. Prog. Ser. 285-293

Phillips, D.J.H. & P.S. Rainbow. (1989). Strategies of trace metal sequestration in aquatic organisms. Mar. Environ. Res. **28**: 207-210.

Phillips, D.J.H. & P.S. Rainbow. (1993). Bio-monitoring of trace aquatic contaminants. Elsevier, New York. 371p.

Phillips. D. J. H. (1980b). Proposal for monitoring studies on he contamination of the East Asian Seas by trace metals and organochlorines. Report UNEP/WG.41/INF.

Phillips. Rainbow, P. S. (1995). Bio-monitoring of heavy metal availability in the marine environment. Marine Pollut., **31:** 183-192. Philosophical Transactions of the Royal Society of London Series B, Biological Sciences **286**~483-505.

Picer, M. and Picer, N. (1991). Levels and long- term trends of some molecular chlorinated hydrocarbons in mussels collected from the western Istrian coastal waters-Northern Adriatic. Chemosphere; **23** (6), 747-759.

Popham, J.D., Johnson, D.C. & D'Auria, J.M. (1980). Mussel (*Mytilus edulis*) as 'point source' indicators of trace metal pollution. Mar. Pollut. Bull., **11:2**

Rainbow ,PS. (1993). The significance of trace metal concentrations in marine invertebrates. In; Dallinger R, Rainbow PS (Eds) Ecotoxicology of metals in invertebrates. Lewis publishers, Boca Raton, FL, p 4-23.

Rainbow, P. S. (1995). Biomonitoring of heavy metal availability in the marine environment. Mar. Pollut. Bull. **31,** 4-12, :183-192.

Rainbow, P. S. (1993). The significant of trace metals concentrations in marine invertebrates, pp. 3-24. In R. Dallinger & P.S. Rainbow (eds.) Ecotoxicology of metals in invertebrates. Lewis, Boca Raton, Florida.

Rainbow, P.S. (1990). Heavy metal levels in marine invertebrates. In Heavy Metals in the Marine Environment. R.W. Furness & P. S. Phillip, D.J.H.,1990. Arsenic in aquatic organisms: a review emphasizing chemical speciation. Aquatic Toxicol. 16, pp. 151-186.

Rainbow, P.S. (1988). The significant of trace metals concentrations in decapods. Symp. Zool. Soc. London, **59:** 291-299.

Rainbow, P.S. (1993). The significance of trace metal concentrations in marine invertebrates. In: Dallinger, R., Rainbow, P.S. (Eds.), Ecotoxicology of metals in invertebrates. Lewis Publishers, Chelsea, USA, pp. 3-23.

Ray, S. (1984). Bioaccumulation of cadmium in marine organisms. Experientia **40:** 14-23. Received 18-VIII-1998. Corrected 18-III-1999. Accepted 10-V-1999.

Regoli. L, H. M. Chan, Y. De Lafontaine and I. Mikaelian. (2001). Organotins in zebra mussel (*Dreissena polymorpha*)) and sediments of the quebec City Harbour area of the St. Lawrence River. Aquat Toxicol. **53**, pp.115-126.

Reinfelder, J.R., N.S. Fisher,S.N. Luoma and Wang. (1997). Assimilation efficiencies and turnover rates of trace element in marine bivalves: Marine Biol.,129 :443-452

Reinfelder, J.R., N.S. Fisher,S.N. Nichols. J.W. Luoma and Wang. (1998). Trace element trophic transfer in aquatic organisms:A critique of the kinetic model approach. The Science of the Total Environment**; 219**,pp. 117-135.

Riget, F, Johansen P, Asmund G, (1996). Influence of length on element concentrations in blue mussel (Mytilus edulis). Mar Pollut Bull **32:**745-751.

Roesijadi G, Robinson, WE. (1994). Metal regulation in aquatic animals: mechanisms of uptake, accumulation and release, In: Malins DC, Ostrander GK (eds) Aquatic toxicology; molecular, biochemical and cellular percpectives. Lewis Publishers, Boca Raton, p 387-420.

Sabine Lattemann. (2003). Chemical Constituents of Desalination Plant Reject Streams and their Potential Impacts on the Marine Ecosystems. INOC-INBA International Workshop on Desalination & Marine Pollution,

Sericano J. W.,Wade, T.L.,Jackson, T. J.,Brooks, J. M., Tripp, B. T., Farrington, J. W., Mee, L. DReadman. J.,Villeneuve J. P.,and Golberg, E. D. (1995). Trace organic contamination in the Americas; An overview of the U.S. National Status and Trends and the International 'Mussel Watch"programs. Marine Pollution bulletin; **31**:214-225.

Shulkin, VM, Kavun VYa, Tkalin AV, Presley BJ. (2002). The influence of metal concentrations in bottom sediments on metal accumulation of *mytilids Crenomytilus* grayanus and *Modiolus kurilensis*. Biol Morya/Mar Biol; **28**:53-60.

Shulkin, VM, Presley BJ, Kavun Via. (2003). Metal concentrations in mussel *Crenomytilus grayanus* and oyster *Cressostrea gigas i*nrelation to contamination of ambient sediments. Environ Int; **29**:493-502.

Siah.A, J. Pellerin, J. C. Amiard, E.Pelletier and L.Viglino, (2003). Delayed gametogenesis and progesterone levels in soft- shell clams (*Mya arenaria*) in relation to in situ contamination to organotins and heavy metals in the St. Lawrence River (Canada), Toxicology and Pharmacology:**135**,:145-156.

Silva, R. Augusto Carlos & Phillip, S. Rainbow. (2006). Comparative biomonitors of coastal trace metal contamination in tropical South America (N. Brazil). In Marin. Environ. Research; **61**: 430-455.

Simkiss, K & Mason A.Z (1984). Cellular responses of molluscan tissues to environmental metals. Mar Environ Res ;**14**:103-118.

Simkiss, K. & A.Z. Mason. (1983). Metal ions: metabolic and toxic effects, p. 102-164. In P.W. Hochachka (ed.). The mollusca. Academic, New York. 285-293

Subba Rao, D. and Al-Yamani, F. (2000). The Arabian Gulf. In:Seas at the millennium; An environment al evaluation. chapter **53**, 2: 1-16.

Szefer P,Frelek K, Szefer K, Lee CB, Warzocha J, Zdrojewska I, et al; (2002). Distribution and relationship of trace metals in soft tissues, byssus and shells of *Mytilus edulis trossulus f*rom the southern Baltic. Environ Pollut **120:**423-44.

Taghon, G.L, A.R.M. Nowell and P.A. Jumars, (1980). Induction of suspension feeding in spionid polychaet by high particulate fluxes, Science;**210:** 562-564.

Thomson, E. A.., Luoma, S. N. Johansson, C. E. , and Cain, D. J., (1984) Water Research;**18**: 755-765.

Tkalin, A. V., Lishavavskaya, T.S., and Shulkin, V. M. (1998). Radionuclides and Trace Metals in mussels and bottom sediments around Viadivostok, Russia, Mar. Pollut. Bull ;**36**: 551-554.

Tanabe, S. (2000). Asia-Pacific Mussels Watch Progress report. Marine Pollution Bull **40 (8):**651

Townsend, D, W, (1992). An overview of the oceanography and biological production. In The Gulf of Maine, Regional synthesis Series, No. 1, pp.5-25. USNOAA.

Tripp, B. W. & Farrington J. W. (1985). Using sentinel organisms to monitor chemical changes in the coastal zone: Progress or paralysis. In proceedings of the ninth annual conference of the Coastal Society. Atlantic City, NJ.

UNEP. (1999). Overview on land-based sources and activities affecting the marine environment in the ROPME sea area, UNEP regional seas reports and studies. ;168 : 127 p.

USFDA,US Food and Drug Administration. (1993). Guidance documents for cadmium, chromium, lead and nickel. Center for Food Safety and Applied Nutrition US Food and Drug Administration , Washington , DC.

Vercauteren K. (1999). Uptake of cadmium and zinc by the mussel *Mytilus edulis* and inhibition by calcium channel and metabolic blockers. Mar Biol; **135:** 615-26.

Viarengo A, Palmero S, Zanicchi G, Capelli R, Vaissiere R, Orunesu M. (1985). Role of metallothioneins in cu and cd accumulation and elimination in the gill and digestive gland cells of *Mytilus galoprovincialis* lam. Mar. Environ Res; **16**:23-36.

Viarengo, A. and Nott, J. A. (1992). Mechanisms of heavy metal cation homeostasis in marine invertebrates. Camp. Biochem. Physiol. 00c **(0),** 1-14.

Walsh, PM. (1990), The use of sea birds as monitors of heavy metals in the marine environment. In: Furness RW, Rainbow PS (ed) Heavy metals in the marine environment. CRC Press, Boca Raton,pp. 183-204.

WHO. (1976). Environmental health criteria. I. Mercury. Geneva: World Health Organization; Who. Toxicological evaluation of certain additives and contaminants. Joint FAO/WHO Expert Committee on Food Additives, WHO Food additives Series no 17, World Health Organization, Geneva; 1982 pp, 28-35.

Widdow, J., B.L. Bayne, D.A.Brown, K. Burns, D.R. Dixon, A. Ivanovici, D.R. Livingston, D.M. Lowe, M.N. Moore, A.R.D. Stebbing and J. Widdow. (1985).

The effect of stress and pollution on Marine Animals. Praege Press, New York, pp. 161-178.

Widdow, J., Donkin, P., Brinsley, M. D., Evans, S. V., Salkeld, P. N., Franklin, A., Law, R. J. and Waldock, M. J. (1995). Scope for growth and contaminant levels in North sea Mussels, *Mytilus edulis*. Marin Ecology Progress Series **127**, 131-148.

Widdow, J., Nasci, C. and Foossato, V.U. (1997). Effects of pollution on the scope for growth of mussels (*Mytilus galloprovincialis*) from the Venice Lagoon, Italy. Marine environmental Research **43,** 69-79.

Wong JW, Yang CL. (1995). The effect of pH and redox potential on the release of nutrients and heavy metals from a contaminated marine sediment. Toxicol Environ Chem ; **62.** 1-10.

Zorba, M. A. Jacob, P.G., al-Bloushi, A. and al-Nafisi, R., R. (1992). 'Clams as pollution bio-indicators and depuration, Sci. Total Environ. **120**, 195-204

8. Curriculum Vita

Education: **Ph.D.** in Ecology, Evolution, and Natural Resources - 2008
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Advisor: Dr. Joanna Burger, Co-Advisor: Dr John Reinfelder
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Summary of Work Experience:

Jul `07 – Aug `07 **Adjunct Professor**
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Sep `97 – May '07 **Full-time Gradate Student/ Teaching Assistant**
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Aug `01 – Jan `01 **Instructor**
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Sep `86 – May `96 **Instructor/Teaching Assistant**
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Sep `86 – May `96 **Research Assistant**
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Research Experience:

Sep `97 – Present DEPARTMENT OF ECOLOGY, EVOLUTION AND NATURAL RESOURCES

RUTGERS UNIVERSITY, NEW BRUNSWICK, NEW JERSEY

Projects:

- Monitoring Benthic Invertebrates Fauna (Supervisor: Dr Gary Taghon, Marine Coastal Sciences, Rutgers Cook College)
- Metal contamination in birds and fish (Supervisor: Dr Joanna Burger. Rutgers Busch Campus)
- Metal Contamination through Atomic Absorption in Bivalves (Supervisor: Dr Joanna Burger, Eoshi lab, Busch Campus)

Sep `95 – May `96 DEPARTMENT OF ZOOLOGY

KUWAIT UNIVERSITY, KHALDIYA, KUWAIT

Projects:

- Systematic Studies of Amphipods (Supervisor: Dr Manaf Behbehani, Marine Science Department)
- Systematic Studies of Marine Fauna of Coastal Habitats in Kuwait (Marine Science Department).
- Identification of Gastropods and Bivalves and Polychaetes (Zoology Department).
- Identification of Zooplankton and Phytoplankton in waters of Kuwait (Supervisor: Dr Faiza Al-Yamani, Kuwait Scientific Institute of Research)
- Shrimp Larvae of Kuwaiti Waters (Supervisor: Dr Faiza Al-Yaman, Kuwait Scientific Institute of Research)

Publications:

- F.Y. Al-Yamani, Tarique, Q. 1995. Description of Larval, Postlarval and Juvenile stages of commercially important species of Metapenaeus affinis In Kuwaiti waters. Kuwait Institutes of Scientific Research. Technical Report. KISR 4693, Kuwait. PP 49-78.

- F.Y. Al-Yamani, Tarique, Q., I, Wafa. 1995. Studies on the larval stages of commercially important species of penaeid shrimp, Penaeus semisulcatus. In Kuwaiti waters. Kuwait Institutes of Scientific Research. Technical Report. KISR 4693, Kuwait PP 1-45

- F.Y. Al-Yamani, Tarique, Q. 1995. Studies on the larval development of the commercially important species Parapenaeopsis stylifera in Kuwaiti waters. Kuwait Institutes of Scientific Research. Technical Report. KISR 4693, Kuwait. PP 82-109.

- Tarique, Q., A.S.D Farmer; T. Watanabe; and A.B. Al-Hajj. 1990. Studies on the larval stages of commercially important species of penaeid shrimp. Annual Research Report, KISR PP50-52

- F.Y. Al-Yamani, Tarique, Q. 1995. Early developmental stages of Metapenaeus stebbingi reared in Laboratory in Kuwait. KISR 4693, Kuwait. PP 111-135.

Work in Progress:

- Tarique, Q., Burger, J., Reinfelder, J.R. 2007. An evaluation of trace metals in the clam *Amiantis umbonella* near a Desalination/Power Plant compared to a Reference Site. Submitted to Science of the Total Environment, September 2007.
- Tarique, Q. An Evaluation of the clam *Amiantis umbonella* (Bivalve) as a Bioindicator of Metal Pollution in the Kuwait Marine Environment. Ph.D Dissertation, Rutgers University, New Brunswick, NJ

Other Skills:

- Extensive international travel. Resided in five countries
- Good understanding of the geography and the cultures of Middle East and South-East Asia
- Computers: Internet savvy, Microsoft Word, Microsoft Excel, and Microsoft PowerPoint