UNIVERSITY OF NAMIBIA

ASSESSMENT OF TRACE METALS POLLUTION ALONG THE CENTRAL NAMIBIAN MARINE COASTLINE: USING *CHOROMYTILUS MERIDIONALIS* (BLACK MUSSEL) AS INDICATOR ORGANISMS

A THESIS SUBMITTED IN FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF MASTER OF SCIENCE IN FISHERIES AND AQUATIC SCIENCES OF UNIVERSITY OF NAMIBIA

BY

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ABSTRACT

This study was carried out at four stations along the Central Namibian marine coastline towns (Walvis Bay, Swakopmund, Henties Bay and Cape Cross) to assess trace metals pollution using *Choromytilus meridionalis* as indicator organism. Samples were collected using randomized sampling techniques during winter and summer months of 2012. EPA 3050B and ICP-OES protocols were used to digest and assimilate the samples. Data were analysed using a 4x2x3 factorial model of a completely randomised design and results showed there was a significant interaction ($p < 0.05$) of only Pb and Fe levels between stations, seasons and substrates studied. However, there was no significant interaction ($p > 0.05$) between Cu and Zn levels between stations, substrates and seasons respectively. Metal levels in mussels and sediments ranked in the order: Fe > Zn > Cu > Pb. Walvis Bay recorded increased metal levels ($P < 0.05$) than other stations probably due to factors like port activities e.g. ship repairs, urban and residential settlements, tourism and recreation as well as run-off water from catchment areas. Sediments exhibited significantly higher mean trace metals than mussels and water ($P < 0.05$). In addition, clear-cut significant correlation ($p < 0.05$) of metals was observed between some stations and substrates. Maximum Fe, Zn, Cu, and Pb mean values in mussels were 483.7, 79.3, 6.50, and 2.30 ppm of sample dry weight respectively while maximum Fe, Zn, Pb; and Cu values in sediments were 44, 749.90, 100.0, 79.30 and 50.50 ppm of sample dry weight respectively. All metal values recorded in this study were lower or within those reported elsewhere. Overall, mussels grown in this part of the coastline could be regarded safe for human consumption; however, levels observed in samples from Walvis Bay are of health concerns. Findings from this study have stressed the need for continued monitoring of these metals along the Namibian marine shoreline for regulatory purposes.

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DEDICATION

I dedicate this work to my late aunt Jennifer Jahn Sandifolo who mysteriously died in 1981. You never lived to see my day but I have always been told about what happened at school on that fateful morning with mum Joyce and Susan. It is sad you didn't make it but we give thanks to God for everything. Blessed be His name. Many might have forgotten about your death but you will remain in my heart dear aunt. You are sincerely missed.

_____Rest In Peace Till We Meet Again_____

DECLARATION

I, Emmanuel Captain Vellemu, declare hereby that this study is a true reflection of my own research, and that this work, or part thereof has not been submitted for a degree in any other institution of higher education.

……. Date…...……..

Emmanuel Captain Vellemu

CHAPTER ONE

1. INTRODUCTION

Pollution of the aquatic environments is a worldwide challenge. Terrestrial anthropogenic activities contribute significantly to contaminating various marine ecosystems. Contaminants can be natural or synthetic compounds. However, when discharged into the sea they can stay in the water column in dissolved form or settle at the bottom in sediments. In addition, aquatic organisms can pick up these contaminants from the water column or in particulate organic matter and accumulate them in their bodies (Stewart, 1999). Thus, if organisms cannot excrete these substances from their bodies, they can pass and build up in the food chain level.

Organisms may react differently when exposed to contaminants as documented by Omoregie and Ufodike (2000). It has also been reported (Airas, 2003; Akueshi *et al.,* 2003) that certain animal groups may accumulate huge amounts of trace metals without any harmful effects while other groups which are more pollution sensitive might die in minute concentrations. This is the reason why biomonitors; organisms which accumulate trace metals in their tissues such as *Choromytilus meridionalis* (black mussels) and *Crassostrea gigas* (oysters), are used and analysed to monitor the bioavailability of such metals in an ecosystem (Viarengo and Canesi, 1991; Cardellicchio *et al.,* 2008).

The rich Benguela current yields a large number of *Choromytilus meridionalis* species in the Central Namibian marine coastline. However, it is quite evident and has been reported (World Bank, 2009) that the coastal ecosystems of Namibia are extremely fragile and are increasingly being disturbed by growing economic development including human activities that could increase the level of pollution. Additionally, the Namibian marine coastline has also received

discharges from industries including mining, private households and recreation during its history (World Bank, 2009).

Yet again, it is evident there are major rivers along the Central Namibian marine coastline that feed water into this part of the Atlantic Ocean on the south west of Africa. These include the Omaruru River in Henties Bay, Swakop River in Swakopmund and the Kuiseb River in Walvis Bay. Several land use practices take place in the catchment areas of these rivers such as agriculture (usually cattle farming practice), mining and tourism. During rainy season and flooding, some sediment originates from the topsoil lost through bad agricultural practices which in some cases probably carry nutrients and contaminants into these rivers. This is termed as 'sedimentation', a process in which there is a deposition of particles transported by flowing water which in turn and eventually increases the turbidity of the water in a reservoir such as lakes or the ocean. Thus, as too much sediment is transported into an aquatic water body, the sediment may be in suspension in the water column or settle at the bottom which might lead to pollution of that water body sooner or later (Cardellicchio *et al*., 2008).

In a recent study (Boateng *et al.*, 2011), it has been reported that the health risks associated with trace metal poisoning in humans and the environment are of concern to environmentalists and government agencies locally and around the world. This concern thus stresses a strong need for continuous research aimed at monitoring the levels of pollutants as well as setting measures to deal with the consequences of environmental pollution.

It was therefore the purpose of this study to provide the baseline data on the level of some selected trace metals (lead-Pb, copper-Cu, zinc-Zn and iron-Fe) in the Central Namibian marine coastline using *Choromytilus meridionalis*, seawater and sediment samples to determine the

environmental status of this part of the coastline.

1.1 Orientation of the study

Environmental contamination by various chemicals such as trace metals and persistent organochlorides has increased lately (Omoregie and Okunsebor, 2005; Ikemoto *et al.*, 2008). In addition, some recent studies have also reported that contaminants' levels in sediments are usually more than the equivalent metals in the water column (Giarratano and Amin, 2010). Hence, these concerns and knowledge gaps gave rise to this study in order to provide insights data and compare the relationship of metals between the sediments, water column and organisms in order to draw critical sound scientific and clear-cut conclusions about the environmental status of the Central Namibian marine coastline. The study also compared metal levels in substrates water, black mussels and sediments between winter (April – July) and summer (September - December) seasons respectively.

1.2 Statement of the problem

In recent years contaminant levels have been escalating in the marine environment as a result of human activities (Cardellicchio *et al*., 2008), and the environmental contamination by various chemicals such as trace metals and persistent organochlorides has increased (Omoregie and Okunsebor, 2005; Ikemoto *et al*., 2008). Furthermore, the rapid growth of urbanization, agriculture and industrial activities has resulted in increased production and use of +harmful elements such as trace elements; and there is likelihood for an eminent trace metal levels being transferred to higher trophic levels in the food web including human beings (Nguyen *et al.*, 2008). Thus, these pose major environmental and human health problems globally (Ensley, 2000).

Unlike many organic contaminants, most metals cannot be eliminated from the environment by chemical or biological transformation (Andres *et al.,* 2000). Lastly, there was no prevailing established and documented data in referenced scientific publications on the pollution status with regard to trace metals in the Central Namibian marine coastline prior to this research investigation.

1.3 Objectives of the study and/or research questions

1.3.1. Research question

This research investigation addressed the following question: What is the environmental pollution status along the Central Namibian marine coastline specifically between Walvis Bay and Cape Cross?

1.3.2. Objectives

1 To determine and compare trace metal levels in Namibian *Choromytilus meridionalis* samples collected from different Central Namibian marine coastline areas (Walvis Bay, Swakopmund, Henties Bay and Cape Cross stations).

2 To compare the metal levels in sediments, water column and the *Choromytilus meridionalis* samples collected from different Central Namibian marine coastline areas (Walvis Bay, Swakopmund, Henties Bay and Cape Cross stations).

3 To determine the trace metal levels between the two seasons (winter and summer).

4 To compare some metal levels in the Namibian *Choromytilus meridionalis* collected from Central Namibian marine coastline (Walvis Bay, Swakopmund, Henties Bay and Cape

Cross stations) with the permissible limit of 1 part per million (ppm) set by the European Community Commission Regulation (EC) No. 466/2001 for human consumption.

1.4 Hypotheses of the study

1. There are no significant difference in each mean trace metal levels (lead, copper, iron, zinc) in sediments, water column and *Choromytilus meridionalis* collected from Central Namibian marine coastline.

2. There are no significant differences in each mean trace metal levels between winter and summer seasons for water, sediments and *Choromytilus meridionalis* samples.

3. The mean trace metal levels in *Choromytilus meridionalis* recorded in the Central Namibian marine coastline are not within the permissible limit of 1 ppm set by the European Community Commission Regulation (EC) No. 466/2001 regarded safe for human consumption.

4. There is no significant interaction between stations, substrates and seasons with regard to trace metal level pollution in the Central Namibian marine coastline.

1.5 Significance of the study

While the core and fundamental intent of this research investigation was to asses and determine the environmental pollution status of the Central Namibian marine coastline, this study aimed at providing baseline data on the levels of Pb, Cu, Zn and Fe in *Choromytilus meridionalis,* water column and sediments for the Central Namibian marine coastline. This is quite substantial for further investigations in the field of ecotoxicology in as far as biomonitoring programs of the marine environment are concerned. The results also act as a benchmark in assisting government

agencies in setting up and mainstreaming various management policies and practices in different ministries responsible for not only marine but also environmental and education sectors to ensure the public at large consume non-hazardous fishery products both locally and internationally. Most importantly, the data provide a sound base for monitoring the Namibian coastline in terms of trace metal pollution. Thus, the data reveal, suggest and provide indications on areas that need critical attention to ensure that Namibia as a country does have a clean and pollution free marine environment.

Similarly, companies involved in the fisheries sector such as Kuiseb Fishing Enterprise, NAMSOV Fishing Enterprise, Hangana, Abroma Fishing Industries, Merlus Sea Food and even the Namibian Standards Institute (NSI) responsible for setting food and quality standards in Namibia will benefit from the findings of this study. Moreover, the consumers need to consume non-toxic food fishery products. Hence, this research investigation proved to be fundamentally imperative aimed at providing adequate and sound scientific information for decision and policy makers not only in Namibia but also around the world in as far as marine pollution is concerned.

1.6 Limitation of the study

Every scientific study investigation most probably has challenges to some extent and this study was not an exception. One notable limitation of this research is that sampling was not done on monthly basis due to time restriction of the Master's Degree study period; hence, sampling was only conical to one summer and winter season. The study did not also cover the entire Central Namibian marine shoreline due to resources but was rather restricted to only four stated stations. Lastly, there was lack of referenced historical data on black mussels.

CHAPTER TWO

2. LITERATURE REVIEW AND THEORETICAL FRAMEWORK

Trace metal contamination of the aquatic environment due to anthropogenic sources started to escalate when industrial revolution began (Nriagu, 1979). However, some previous studies (Falconer *et al.*, 1986 and Gonzalez *et al.*, 1998) reported that some of these trace metals have been introduced in marine environments through volcanic eruptions; although another study (Oehlenschläger, 2002) enlightens that the presence of trace metals in an aquatic environment is due to both natural and anthropogenic sources. For example, lead concentration has increased swiftly since it was added to gasoline fuel of vehicles. On the contrary, mercury levels have risen up as a result of degassing from the Earth's crust, industrial and urban activities. In addition, it has also been reported that these metals are not easy to quantify in an environment due to the very large natural inputs from the erosion or rocks, wind-blown dusts, volcanic activity and forest fires. The oceans therefore provide a vital sink for many trace metals and their compounds (Giarratano and Amin, 2010).

Marine species contain certain levels of metals since they live in aquatic ecosystems (Oehlenschläger, 2002). Yet, the levels of such metals usually depend on where the species resides in an ecosystem. For instance, it is further reported (Oehlenschläger, 2002) that in the open oceans fish would have the normal concentration of metals because there is no major pollution. However, some aquatic environments are located closer to industrial activities (Prudente *et al.*, 1997); such that organisms (e.g. *Choromytilus meridionalis* and *Crassotrea gigas*) found in these areas would normally contain metal levels surpassing the natural concentration (Kalay and Cahli, 1999; Dobson, 2000; Claisse *et al.*, 2001). In addition, some

studies (Cardellicchio *et al*., 2008; Boateng *et al*., 2011) have evidenced that seasons influence trace metal levels in bivalves with the belief that prior to spawning period, bivalves produce and store compounds that have an affinity to trace metals in the gonad tissue.

Various studies (Watling and Watling, 1976; Viarengo, 1985; Rainbow and Dallinger, 1993; Cardellicchio *et al*., 2008) have documented trace metal level accumulation by mussels such as *Mytilus edulis* and *Mytilus galloprovincialis* (with very little on *Choromytilus meridionalis*) and their potential as indicators of metal pollution. Unlike fishes which are migratory, *Choromytilus meridionalis* species are sessile filter-feeder organisms which makes these species prone to trace metal pollution in their habitants.

Some investigations (Viarengo and Canesi, 1991; Cardellicchio *et al*., 2008) have also reported that mussels are good bioaccumulators of some trace metals and as such they are used to indicate levels of those metals to monitor an aquatic body's status for any pollution. Thus, to determine trace metal accumulation in an ecosystem, certain marine species such as limpets and mussels are useful bio-indicators (Cardellicchio *et al*., 2008); and sometimes the metal accumulation levels in these organisms are directly proportional to the existing levels in the environment (Pe´rez-Lo´ pez *et al.*, 2003). In addition, transplanted or resident bivalves and other molluscs provide a good indicator of temporally and spatially average concentrations of bioavailable contaminants in aquatic ecosystems. Furthermore, different species of shellfish have been used for aquatic biomonitoring because they are widely distributed. In the Central Namibian marine shoreline, most of the rocky shores are inhabited by some type of mussels and they accumulate trace metals based on the physical factors taking place due to the Benguela upwelling, wave and wind action which most likely pose threats to these organisms.

However, some earlier findings (Watling and Watling, 1976) on this same species indicated that some metal levels in smaller individuals may be greater than those in larger individuals such that care must be taken when interpreting the differences in trace metals between populations or for populations sampled over a given period of time. As it is elsewhere, these species are commonly found and distributed in the Namibian coastline both in rocky and sandy beaches or at least suspended in the water column especially for farmed ones such as Walvis Bay mussels in this study. Again, in Namibia these species are available at low cost in terms of market price and availability. Their choice as study organisms was due to the ease of handling in laboratory tests, easy accessibility in terms of sampling and their ecological and economic importance for the country. Furthermore, (Cardellicchio *et al*., 2008) affirms that metal levels in *Choromytilus meridionalis* tissues readily respond to impinging levels in the environment.

Another study (Sulochanan *et al.,* 2007) documented that there is usually a limited water exchange in bays (e.g. Walvis Bay); and that shipping introduces contaminants such as oil, trace metals and organochlorides. The quality of water and sediments affect all living organisms in such ecosystems (Sulochanan *et al*., 2007); and trace metal toxicity in aquatic organisms in association with long residence time within food chains and the potential risk of human exposure, makes it necessary to monitor the levels of these contaminants in marine organisms (Giarratano and Amin, 2010). Sediments have been reported to form the major repository of trace metal in aquatic systems such that bioaccumulation and magnification are both capable of leading to toxic level of these metals in species such as mussels even at very low concentrations.

Further findings (Giarratano and Amin, 2010) noted that trace metal levels in sediments are usually more than the equivalent metals in the water column because the former can act as a scavenger for trace metals and an adsorptive sink in aquatic environment than the latter. It has

also been documented (Olowu *et al.,* 2010) that the distribution of trace metals in water bodies reveal that the levels of trace metals in the bottom sediment are usually higher than in the water columns which confirms that sediments act as sink for trace metals. In a related study however, (Obirikorang *et al.,* 2011), it was stressed that the analyses of sediments are a useful method of studying aquatic pollution with trace metals. This is apparently because the presence of increased trace metals in sediments found at the bottom of the water column can be a good indicator of man-induced contamination rather than natural enrichment of the sediment by geological weathering; although sediment metal levels in their study was found to be considerably low in variation per season.

Another study (Taylor and Kesterton, 2002) on the heavy metal contamination of the Gruben River in Namibia reported that the river area has a well-documented history of copper mining dating back to 1916. The river is one of the many tributaries of the Khan River which is a major tributary of the Swakop River and when in flood, the Swakop River drains to the southern Atlantic Ocean via the Swakopmund coastal town. They further stated that the area has metasediments associated with magmatic granites of the Damara Oregon rock such that the bedrock is highly weathered and fractured with materials being eroded in the valley and channeled into the Namibian marine shoreline via the Swakop River. In their findings, there was a high contamination of copper, nickel and zinc in the river. This holds true because in a separate study (Pecorari *et al.,* 2006) it is reported that rivers, lakes and other water bodies are frequently located in urbanized areas and such waters are not only used for recreational purposes but usually act as collectors of diverse types of effluents.

Water pollution occurs almost everywhere either direct or indirectly. Many countries have not sufficiently protected water quality. Both freshwater and seawater are prone to pollution. Major

sources of pollution have been reported to originate from various sources but usually due to anthropogenic practices and their externalities on the environment including aquatic bodies. Underground pollution such as landfill sites and rubbish dumps, buried harmful waste, industrial mining and human waste are just common examples of anthropogenic sources. These might severely affect aquatic water bodies in cases where there is flooding and runoffs as they carry along with them heavy and trace metals such as lead, mercury, copper, zinc and iron leading to contamination of such an ecosystem (The Ministry of Education, 2010).

Unpleasantly, polluted water supplies do not only limit water availability but also put most people at risk of water-related diseases if they use such water or consume fishery products originating from contaminated water (The Ministry of Education, 2010). In Namibia, some mining companies along the coastline depend on their freshwater supply from the Henties Bay AREVA Desalination Plant. Furthermore, aquaculture practice becomes a challenge if water is contaminated and can put social and economic problems on the country in many ways. Sulphur eruptions in the Namibian marine coastline also pose challenges not only to organisms such as fish and *Choromytilus meridionalis* but also the quality of water itself. It is believed that pollutants such as sulphur dioxide and oxides of nitrogen combine in the atmosphere to form acid rain which lowers the pH of fresh water if not buffered and can in turn kill fish that are acid sensitive. However, this acid rain can release heavy and trace metals in the top soil leading to contamination of oceans when flooding (The Ministry of Education, 2010).

Namibia has a very dry climate and does not have much fresh water. As mentioned, most mines along the coastline rely on freshwater supply from the desalination plants. The Ministry of Education, (2010) in Namibia has further reported that water pollution is a main concern to the country. Water pollution has become a key issue since the country became more economically

and industrially developed. Fisheries, mining and leather tanning industries are thought to be major contributors of trace metal pollution in coastal waters of Namibia due to lack of discharge treatment measures. Although special waste disposal sites exist at Walvis Bay town, reports indicate that these sites are not always used and that leachate from landfill sites is usually more polluted than sewage and more difficult to treat which eventually lead to seawater contamination (The Ministry of Education, 2010). Thus, pollution challenges in the Namibian coastal waters are associated with commercial and urban activities, especially in and around the Walvis Bay harbour area. Minor oil spills, discharge of waste containing traces of antifouling paints, sewage from ships and heavy metals from the export of semi-processed mine ore also contribute to the pollution of the seawater in the harbour and bay area. In addition, solid waste, sewerage and small amounts of oil products produced from the ships are occasionally dumped into the sea resulting in wastes being washed ashore on the beaches of the Namibian coastline (The Ministry of Education, 2010). Furthermore, the waste dumpsite at Walvis Bay is thought to pollute the ground water as the water table at the site coincides with the lower level of the dump. Underground pollution may also occur from the Rössing uranium mine despite the mine having an extensive programme for wastewater recovery (The Ministry of Education, 2010). Hence, conducting ecotoxicological studies in seawater ensures the monitoring of pollution status of such a water body.

The main reason therefore of biomonitoring trace metal levels has been to determine the toxicological threats posed to organisms and also for health risks to humans from the ingestion of those species that are edible. Thus, this study proved imperative based on the prevailing literature's arguments around the world so that it should act as a reference point for monitoring and further investigations in Namibia in the future.

CHAPTER THREE

3. METHODOLOGY

3.1 Research Design

This study was conducted using a quantitative research design. There were four variables analysed in each substrate (black mussels, water and sediments) namely iron (Fe), copper (Cu), lead (Pb) and zinc (Zn). In addition, seasonal variation (winter and summer) and location (Walvis Bay, Swakopmund, Henties Bay and Cape Cross) of the samples were also investigated to determine the trend and levels of these metals in the Central Namibian coastline.

The study was undertaken using a factorial model experiment of a 4x2x3 factor in a completely randomised design (CRD). Replication was done twelve times for all three substrates in each location per variable under investigation. The least significant difference (LSD) was used to compare the mean level of variables between the two seasons and stations where analysis of variance (ANOVA) showed significant differences at 0.05 significance level. All mean metal levels have been reported as a mean ± of standard error (SE).

3.1.1 Mathematical model for a factorial design (4x2x3 factor) in a completely randomised design

The following is the mathematical model for this study experiment/investigation;

$$Y_{ijk} = \mu + \alpha_i + \beta_j + Z_n + (\alpha\beta)_{ij} + (\alpha Z)_{in} + (\alpha\beta Z)_{ijn} + \varepsilon_{ijk}$$

Where:

Y_{ijzk} is the k^{th} observation /unit given the i^{th} level of factor A (station), the j^{th} level of factor B (season) and the z^{th} level of factor C (substrate)

μ is the overall mean for the study/experiment

α_i is the effect of the i^{th} level of factor A (station; i=1,2,3,4)

β_j is the effect of the jth level of factor B (season; j=1,2)

Z_k is the Z^{th} level of factor C (substrate; n = 1,2,3)

$(\alpha\beta)_{ij}$ is the interaction effect of the i^{th} level of factor A (station) and j^{th} level of factor B (season)

$(\alpha Z)_{in}$ is the interaction effect of the i^{th} level of factor A (station) and j^{th} level of factor C (substrate)

$(\alpha\beta Z)_{ijn}$ is the interaction effect of the i^{th} level of factor A (station), j^{th} level of factor B (season) and n^{th} level of factor C (substrate)

ε_{ijnk} is the random error of the k^{th} unit which has been given the i^{th} level of factor A (station), the j^{th} level of factor B (season) and z^{th} level of factor C

3.2 Population of Black mussel used for this study

Black mussel (Plate 1) sampled for this study were collected from the Central Namibian marine coastline (Figure 1).

Plate 1: The Black mussel **(***Choromytilus meridionalis*) collected from the Central Namibian

3.3. Study area

In the Central Namibian marine coastline - Walvis Bay (Latitude 22^{o} 56' 50.3"S, Longitude 014^{o} 30' 04.3"E), - Swakopmund (Latitude 22^{o} 42' 02.7"S, Longitude 014^{o} 31' 14.9"E), - Henties Bay (Latitude 22^{o} 24' 34.8"S, Longitude 014^{o} 26' 38.7"E) and Cape Cross (Latitude 21^{o} 45' 22.5"S, Longitude 013^{o} 58' 08.2"E), four sampling stations were selected (Figure 1). The latitude and longitude of the sampling sites were marked using a portable Global Positioning System (Garmin GPS, Model Etrex 30) device. Walvis Bay station was chosen based on the fact that there are a lot of activities taking place around the Kuiseb Area including the harbour that likely contribute to trace metal pollution. The high population of people dwelling in the area which also suggests the area activities could be a source of possible pollution based on prevailing literature. Swakopmund station was selected because of the Swakopmund River that discharges its effluents into the ocean and also considering the mining activities that take place in its catchment

area such as the Gruben and Khan areas in addition to the human settlements in this coastal town. As for Henties Bay station, the activities at the desalination plant in the area gave rise to this study while Cape Cross was partly chosen as a point of reference considering that there is not much activity taking place there compared to the other stations mentioned.

Prior to sample collection, an observational visit was undertaken to all four sampling stations (Figure 1) to roughly estimate and determine sample availability and get acquainted with these areas. The rocky shores are constantly exposed and experience strong wave action including the extensive Benguela upwelling. Mussels, sediments and water samples were collected from each of the above-mentioned stations.

Figure 1: Map of the Study Area (Source: Google Maps)

3.4. Sampling

3.4.1. Sampling Techniques

Probability sampling techniques were used in which the probability of any particular member of the population being chosen was unknown and arbitrary. An observation visit to the sampling locations was made prior to sample collection as stated earlier. Afterwards, the species, water and sediment samples were collected using transect random sampling technique. Hence, the selection of sampling units was entirely arbitrary. *In situ* observations of surface water temperature were measured. The pH and dissolved oxygen were estimated during each sampling period from April to December 2012. The pH was measured using the Voltcraft PH-100 ATC pH meter consisting of potentiometer, a glass electrode, a reference electrode and a temperature compensating device. Measurement of pH is one of the most important and frequently used tests in water chemistry. Usually, pH is used in alkalinity and carbon dioxide measurement and many other acid-base equilibria. The dissolved oxygen was determined by the YSI 550-12 model DO meter. All measurements for water parameters were done on site during each sampling period.

3.4.2. Sample size selection

All samples in this study were collected from the Central Namibian marine coastline between April and December of 2012. The sample size was chosen systematically to represent the at least the prevailing population in the ecosystem. There were four stations (Walvis Bay, Swakopmund, Henties Bay and Cape Cross stations) that were selected for this investigation. A transect was formed in each of the four stations of this study. From each station there were three (3) sub-stations selected for sampling using a hat method sampling technique to exclude any biasness. In principle, the substations were numbered 1, 2 and 3 on a piece of paper, folded and thrown into

the hat and shaken for sample selection. Randomly, 4 mussel samples were picked from each of the 3 substations per station for each of the 4 metals making a total of 12 mussels collected per station. For the 4 stations therefore, a total of 48 mussel samples were collected in winter season. Using the same sampling technique, sediment samples and water samples were also collected. Thus, the sample size of 48 from each station was reached and this gave a total of 144 samples for winter season for the three variables; and 144 samples for summer season making a complete sample total of 288 for this study for mussels, water and sediment samples. The samples were collected during the winter and summer seasons of Namibia.

3.4.3. Sampling procedure

The sampling date, location, station and number of mussels were recorded during sample collection. Water and sediment samples were collected following the standard sampling procedure as described in APHA (1999). A transect random sampling procedure was used for mussel collection. All samples were within a similar range of length and from within a specific range of intertidal depth. In both seasons, samples from all stations were collected during a low tide except those collected from Walvis Bay station where an engine boat was used to reach the Kuiseb Fishing Enterprise Mussel Farm.

For winter sampling, water and mussel samples were collected from the farm but sediments were collected from the Walvis Bay Harbour in the same vicinity (approximately 5 kilometres apart) because the bottom depth could not be reached due to lack of a Vaan Grab during the scheduled sampling period. Still more even if the instrument was there, the depth was more than 9 metres which was beyond reach. Only mussel samples were collected from the Kuiseb Mussel Farm in summer but water and sediments were both collected from Walvis Bay port. The rest of the

samples from the other stations came from same positions both in winter and summer with an aid of the GPS device. Thus, the APHA, (1999) modified with Olowu *et al*., (2010); Ayejuyo *et al.*, (2010) and Olowoyo *et al.*, (2012) sampling methods were used for sampling. All mussels and sediments were collected by hand. For water samples, the process essentially involved collection of surface water samples using acid clean plastic containers. Precisely, a 5L water sample was collected in an acid-cleaned plastic bottle from each station. The samples were spiked at the time of collection by adding 3 mL of nitric acid per litre of sample in order to stabilize the metal content in the sample matrix as analysis was done at a later stage. Thus, 5L water sample bottle was spiked with 15 mL of concentrated nitric acid before transporting the samples to the laboratory.

In terms of moral values, research ethics were carefully followed and observed to make sure the whole investigation process, the environment and animal welfare were not negatively affected. In a scenario where too many live animals were collected, proper handling of the animal was followed such as returning them into the sea rather than just discarding them inhumanly.

This also included following proper laboratory procedures and taking care of the organism's welfare such as sacrificing them before analysis. Other than sacrificing the animals with a sharp knife or a stone, slow death technique was used by placing them under controlled freezer conditions. Then, all left over samples and/or shells after analysis were placed in the dust bin for proper disposal to avoid polluting the environment.

3.5. Procedure and quality control

All the four sampling stations are located and well distributed on the Central Namibian marine coastline between Walvis Bay and Cape Cross. Except for mussel and water samples from

Walvis Bay station which were collected from the Kuiseb Fishing Enterprise Mussel Farm and area (at an approximately 9 metres depth) during winter season, samples were collected from rocky shores in the inter-tidal zones across all stations with a depth of not exceeding 6 metres. Collected live specimens were transported right away from the sampling stations in small sea water tanks to the Sam Nujoma Marine Research and Coastal Resources Centre (SANUMARC) Laboratory of the University of Namibia at Henties Bay campus. In the laboratory, morphometric (fresh weight and length) measurements of the mussel specimens were taken before sorting them into similar length and weight accordingly. For convenience sake, only large sized mussels (6.0 – 9.50 cm) were selected and measured with an exception of those mussels from Henties Bay mussels in which over half of the samples were smaller in size and on average below 6.0 cm. Weight of the mussel samples was recorded for each sample under investigation. Afterwards, the mussel samples were placed in clean polythene bags and kept in the refrigerator for 24 hours to sacrifice them and drain the excess water from the shells before preserving them in a deep freezer at - 30°C in the SANUMARC Laboratory waiting for digestion process.

For sediments, the samples were first prepared by removing stones and other debris. Then, both the mussel and sediment samples were air dried thoroughly and separately in the Incubator 2000 Series model at 70°C for 48 hours and 110°C for 48 hours respectively. The *Choromytilus meridionalis* samples were crushed into fine particles using the metal free mortar and pestle while the sediments were also pulverised in the same way but had to pass through a 250 micro metre mesh sieve to obtain fine and uniform particles. Thus, each sample powder was then shaken meticulously to ensure homogeneity before a known weight of sample was taken for analysis. Twelve replicate samples were then prepared for each variable per treatment.

Afterwards, samples were taken to Windhoek Analytical Laboratory Services for analysis. Samples were further digested using the Environmental Protection Agency **(EPA) 3050B** acid digestion method. Sediment and mussel samples were further air-dried and manually grinded once again and passed through a 0.17 cm sieve. All samples passed through this mesh size sieve to ensure very fine particles are obtained. For water samples, no digestion was done as samples had no visual turbidity (< 1 NTU) and were analysed on 'as is' basis. Precisely, during the digestion of the samples, 1g sample was digested with repeated additions of nitric acid and hydrogen peroxide. The hydrochloric acid was added to the initial digestate and the sample refluxed. The digestate was then diluted to a final volume of 25 ml. Copper, zinc, lead and iron levels were determined by the inductively coupled plasma optical emission spectrometry (**ICP-OES),** Perkin Elmer Optima 7, 000 DV model on the following wavelengths; Cu: 324.752; Zn: 213.860; Fe: 259.943 and Pb: 220.353.

In addition, all samples in this study were analysed for Near Total Concentration (NTC) and because of that there was no separation of the tissue and the shells. Again, it is a very strong acid that will dissolve almost all elements that could become "environmentally available" in a sample.

Additionally, a multi-element standard solution was prepared from certified single element standards (10, 000 ppm). The calibration values ranged from 0.01 ppm up to 5.0 ppm whereby a five point calibration was determined (0.01, 0.25, 1.0, 2.5 and 5.0 ppm). For water samples, the matrix adjusted standards were prepared in artificial seawater whilst for sediment and mussels the standards were prepared in nitric acid.

All calibration curves were verified utilizing a calibration verification sample and blank. As for water samples, it included one artificial seawater blank per sampling site while for sediment and

mussels, one method blank per sampling site was used. All samples involved the inclusion of two laboratory duplicates per sampling site. For sediments, however, the iron concentrations exceeded the upper standard and thus the samples were re-analysed in a 1:100 dilution. Duplicate samples were prepared to assess the precision of the method used for the analysis. Finally, all glassware used was of analytical grade and each instrument from sampling phase to analysis of trace metals was calibrated first before use.

3.6. Data analysis

All metal levels in this study have been expressed as NTC in ppm for water samples and ppm for *Choromytilus meridionalis* and sediment samples respectively. GENSTAT Discovery Edition 4 software (VSN International, Hertfordshire HP1 1ES, UK) for statistical analysis was used to analyse the data of this research investigation. Data were tested for normality at 0.05 level of significance prior to analysis to determine which analysis method to use either parametric or nonparametric. Data were analysed using a mathematical model for a factorial design (4x2x3 factor) in a completely randomised design. Mean metal levels that had shown significant differences were assessed using the least significant difference technique. The mean levels of each variable in *Choromytilus meridionalis*, water and sediment samples were measured as the mean of the standard error (± SE) as can be observed in summary tables in the results section. In addition, Pearson's correlation coefficient was used to determine correlation between trace metals in *Choromytilus meridionalis*, sediment and water samples in both winter and summer season and between these variables. All descriptive statistics and graphs were executed using GENSTAT Discovery Edition 4 software and redrawn by Microsoft Office Excel application for easy formatting.

3.7 Bioaccumulation factor calculations (BAF)

Bioaccumulation Factor is defined as the ratio of the contaminant in an organism to the concentration in the ambient environment at a steady state, where the organism can take in the contaminant through ingestion with its food as well as through direct content (U.S. Environmental Protection Agency, 2010).

The following formulas were used to calculate the Bioaccumulation Factor (BAF) by black mussels for each metal across the four stations along the Central Namibian marine coastline.

1. **BAF between black mussel tissue and water**

$$BAF_{water} = \frac{\text{Pollutant concentration in mussel tissue (ppm)}}{\text{Pollutant concentration in water (ppm)}}$$

2. **Between black mussel and sediment**

$$BAF_{sediment} = \frac{\text{Pollutant concentration in mussel tissue (ppm)}}{\text{Pollutant concentration in sediment (ppm)}}$$

CHAPTER FOUR

4. RESULTS

This part of the report presents observations and findings of this study from sampling phase to data analysis. However, for ease of referencing the section has been arranged into subsections. The first part outlines observations and findings on water parameters while the second part reports observations and results of trace metal levels accumulated by mussel samples followed by trace metal levels in seawater samples. The final subsections reveal the observations and findings of trace metals accumulated in sediment samples and also outline some notable correlations of the metals of this study. Both winter and summer seasons' metal level findings and correlations have thoroughly been presented in various tables and graphs with the means and standard errors.

4.1 Variations in water parameters

This study investigation showed some variations in temperature mean values, dissolved oxygen (DO) and the potential of hydrogen ions (pH). Maximum surface seawater temperatures of 17.40^{o}C and 18.90^{o}C in surface seawater temperatures were recorded during winter and summer seasons respectively. This investigation recorded the highest temperature value in Swakopmund station during winter season compared to summer season while the maximum temperature value (18.90^{o}C) was recorded in Walvis Bay station. Again, while the minimum temperature value of 14.40^{o}C in seawater was recorded in winter season and in Henties Bay station, the minimum value of temperature recorded in summer season was 14.10^{o}C. However, mean temperature values of 15.83 ± 0.09^{o}C and 16.38 ± 0.27^{o}C were both recorded for winter and summer months

respectively. This shows the temperature degree change between winter and summer season was an average of 0.55^{o}C.

The pH in water samples for all stations ranged from 7.94 to 8.67 during winter season compared to the pH values in the range between 4.67 and 8.59 in summer season (Appendix 1).

Additionally, this study showed that both minimum and maximum mean dissolved oxygen values (7.69 ppm and 9.26 ppm) were recorded at Swakopmund station during winter and summer seasons respectively. The study noted that both Walvis Bay and Swakopmund stations had registered increased DO in summer compared to winter while there was a drop in DO levels at Henties Bay and Cape Cross stations. The highest mean DO value in summer was observed in Walvis Bay station.

While the lowest Do value recorded was 3.95 ppm during winter and in Walvis Bay station, in summer period DO value recorded was 4.53 ppm slightly higher than that recorded during winter season. Overall, the mean DO value of 6.37 ± 0.44 ppm for winter was recorded while for summer DO value was 6.83 ± 0.72 ppm in all stations as seen in Table 1. Figures 2, 3 and 4 indicate the values for all parameters in much detailed illustration.

The following Figures 2 - 4 indicate the values of winter and summer for dissolved oxygen, pH and temperatures of the surface seawater in the study locations.

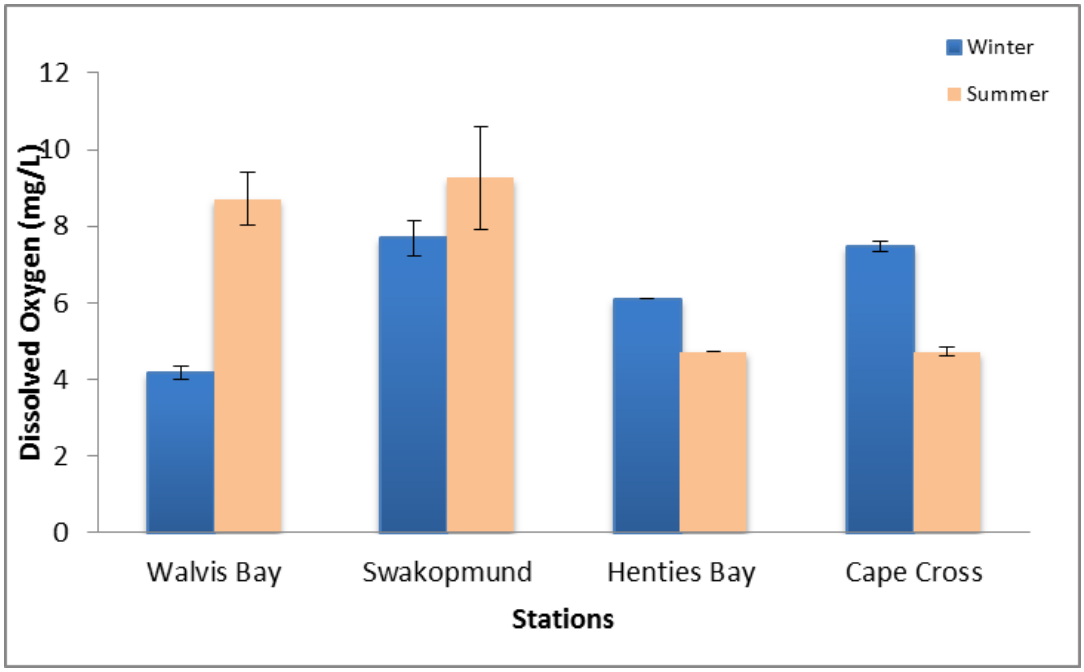

Figure 2: Mean dissolved oxygen values of seawater

The standard errors of the mean for dissolved oxygen (Figure 2) in winter and summer were 0.44 and 0.72 respectively. Statistically, using a two sample independent *t* - test, there were no significant differences in mean dissolved oxygen values between winter and summer seasons in seawater across the stations 95% confidence interval ($p > 0.05$).

Table 1: Mean dissolved oxygen values of seawater and probability value

Season	**Mean + S.E.**	**P value**
Winter	6.37 ± 0.44	0.59
Summer	6.83 ± 0.72	

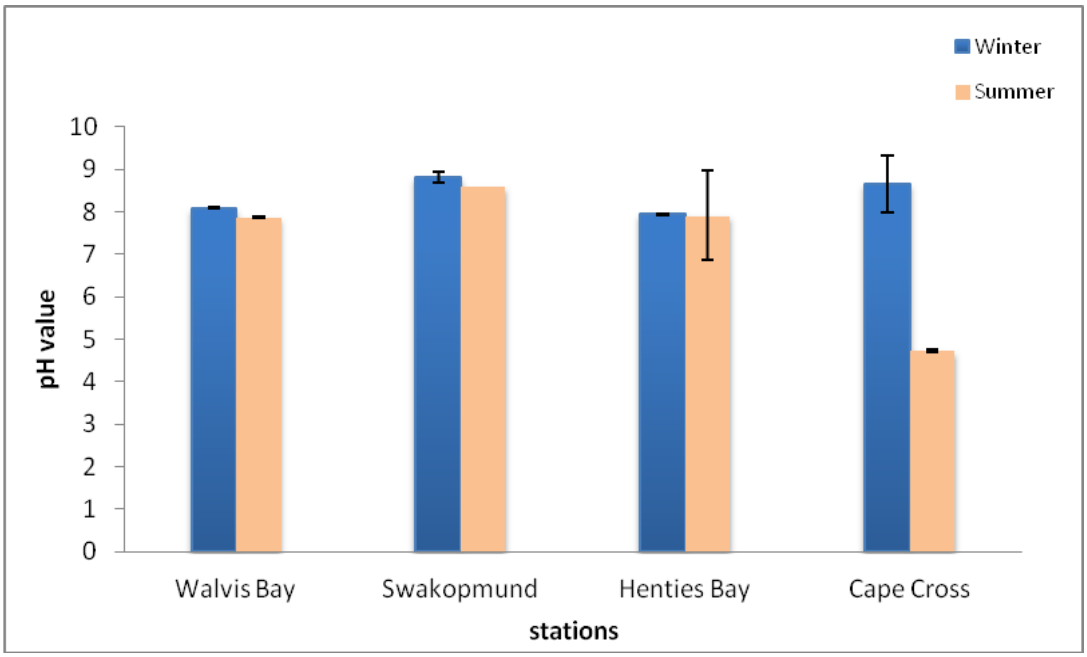

Figure 3: pH levels of seawater per station

The graph above shows the pH values across the stations during this study.

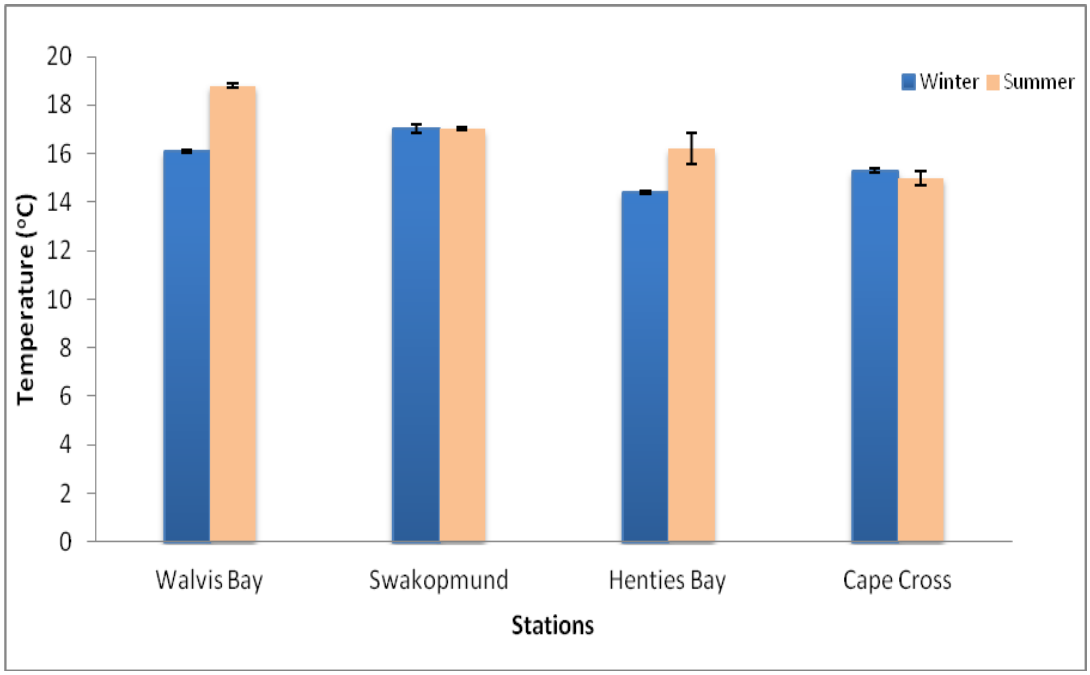

Figure 4: Mean temperature values of seawater

The standard errors of the mean for temperature (Figure 4) in winter and summer seasons were 0.30 and 0.46 respectively. Statistically, using a two sample independent *t* - test, the analysis showed that there were no significant differences in mean temperature value between winter and summer seasons in seawater across the stations at 95% confidence interval ($p > 0.05$) as can be seen in Table 2.

Table 2: Mean temperature of seawater and probability value

Season	**Mean + S.E.**	**P value**
Winter	15.82 ± 0.30	0.32
Summer	16.38 ± 0.46	

4.2. Trace metal levels in *Choromytilus meridionalis*, water column and sediment samples

Mussels are usually attached to substrates e.g. rocks; like in this investigation, except for mussels collected from Walvis Bay station that were suspended in the water column in the Kuseib Fishing Enterprise Mussel Farm. Thus, these mussels were not in direct contact with the sediment nor attached to the rocks but rather only exposed to fine sediments that are resuspended in the water column by wave action or the Benguela upwelling currents. In both seasons, it was observed that mussels from Walvis Bay station were of relatively larger sizes in length and weight while Henties Bay mussels were smaller in size and those from Swakopmund station had very hard shells than the rest.

Results of this investigation showed that during winter season there were no metals recorded in all water samples as the levels were way below detection limit while during summer season metals were recorded at Walvis Bay station alone. It is important to note that water samples obtained from Walvis Bay Station were blackish in appearance and had a slight odour indicating some turbidity as compared to the other stations.

In both seasons sediment samples from Walvis Bay had very fine particles and dark in colour with some silt-clay but gravel while samples from the rest of the stations mainly constituted of gravel and sand; and had brownish colour. Sediment samples from Henties Bay and Cape Cross stations mainly constituted of large sized particles. All sediment samples contained some amount of debris. Walvis Bay's station sediment had the most odour followed by Swakopmund's samples and the remaining two station's sediment were odourless, perhaps this could suggest that there was enough oxygen available for aerobic degradation of sedimentary organic matter.

Lead

No lead levels were recorded in *Choromytilus meridionalis* samples in all stations of this study during winter months. However, it is intriguing to note that this research investigation did record some values of lead in *Choromytilus meridionalis* species during summer season in the same stations. Precisely, the highest value level of lead was recorded at Swakopmund station (1.40 ppm) while the maximum value of Pb for Walvis Bay was 0.70 ppm in which 50% of the mussels registered some Pb values. On average, Walvis Bay station recorded maximum levels of Pb within the Central Namibian marine coastline. This station is located in the vicinity where there are high numbers of industrial companies and increased human settlement. Walvis Bay station also houses Namibia's most important harbour and is the major export and import area. Nevertheless, Walvis Bay and Swakopmund stations are the only ones that recorded some Pb values in mussel samples during summer. This study also revealed that Walvis Bay Station had the most Pb content accumulated by mussels.

Unlike in winter season where no single Pb was recorded in all stations in water samples, some traces of Pb were recorded in Walvis Bay station alone during summer season with a mean Pb value of 0.01 ppm as seen in Table 3. Remarkably, this study did not record any single value of Pb in Cape Cross' sediments while Swakopmund and Henties Bay stations recorded mean Pb values of 1.07 ppm and 1.83 ppm during winter season respectively. However, these values had doubled during summer months in Henties Bay station while Cape Cross station recorded a mean value of 0.30 ppm.

Lead interaction between stations, substrates and seasons

Using the 4x2x3 factorial model analysis of a completely randomised design, there was a significant interaction ($p = 0.019$) between stations, substrates and seasons with regard to lead pollution along the Central Namibian marine coastline (Appendix 2). The factorial model also showed significant differences ($p < 0.05$). As can be seen in the summary Table 3, sediment samples had accumulated more Pb in both winter and summer seasons at Walvis Bay than any of the stations under investigation.

Table 3: Interaction table for lead between stations, substrates and seasons

	Mussels (ppm)		**Sediments (ppm)**		**Water (ppm)**	
Station	Winter	Summer	Winter	Summer	Winter	Summer
Walvis Bay	0	0.25 ± 0.08	47.31 ± 2.63	39.06 ± 4.89	0	0.01 ± 0.01
Swakopmund	0	0.12 ± 0.12	1.07 ± 0.26	1.39 ± 0.13	0	0
Henties Bay	0	0	1.83 ± 0.50	3.02 ± 0.15	0	0
Cape Cross	0	0	0	0.28 ± 0.08	0	0

Lead levels across the stations

Lead levels were highly significantly different ($p < 0.05$) across the four different stations of Central Namibian marine coastline (Appendix 2). Walvis Bay station had accumulated more lead than any other station as can be seen in Figure 5.

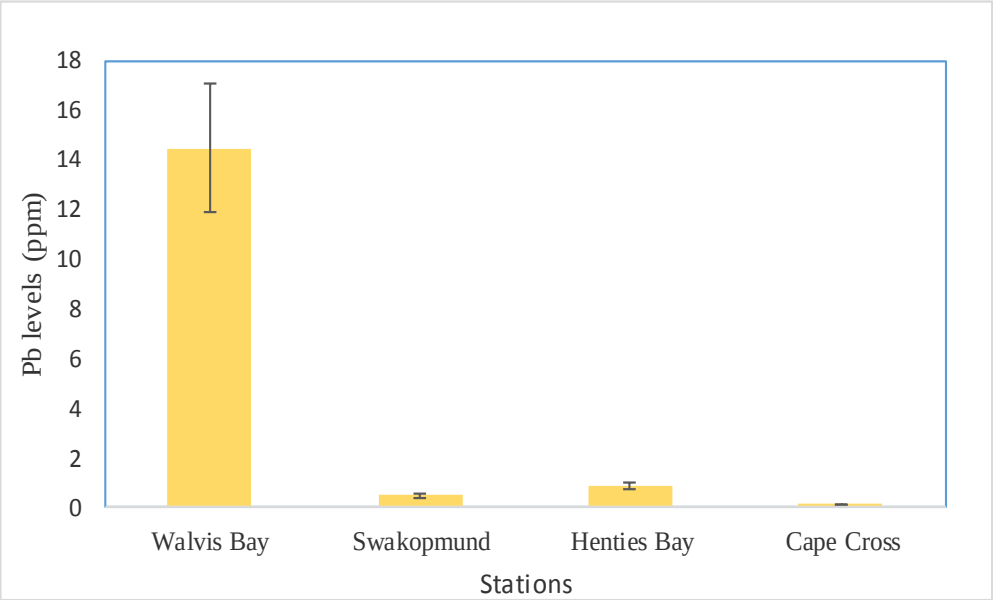

Figure 5: Overall lead levels across the stations

Lead levels in substrates

Statistically, there was a highly significant difference (p = 0.01) across the substrates (water, mussels and sediments) in the Central Namibian marine coastline. As can be seen in Figure 6 below, sediments had accumulated more Pb followed by mussels and then water.

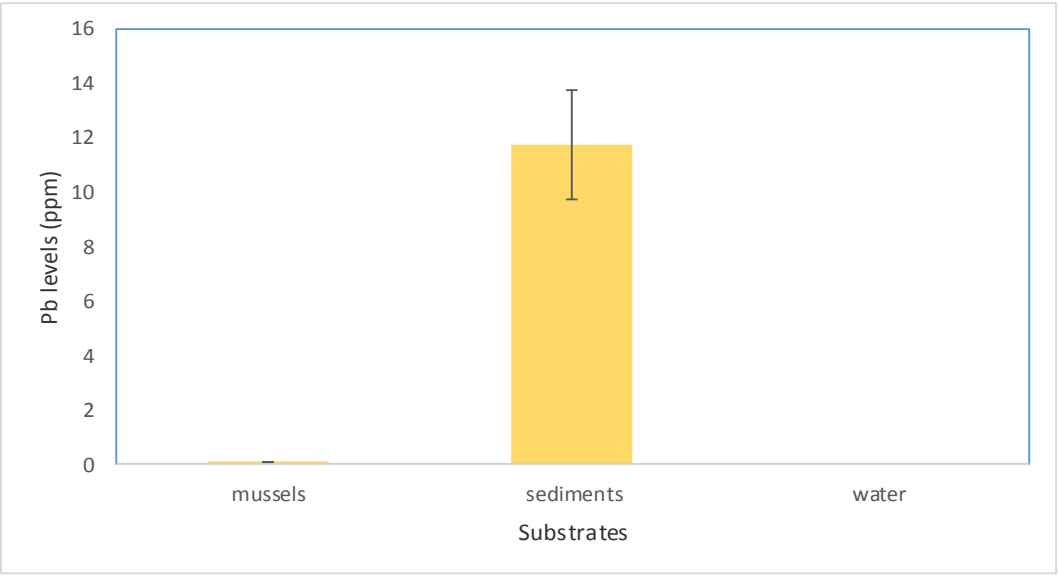

Figure 6: Lead levels accumulated by substrates

Lead levels between the seasons

Statistically (p = 0.286), there was no significant difference between lead levels accumulated between the two seasons despite winter accumulating more Pb than summer season. However, there was more lead accumulated by substrates during winter along the Namibian marine coastline as can be seen in Figure 7. This was despite not being significant between the seasons.

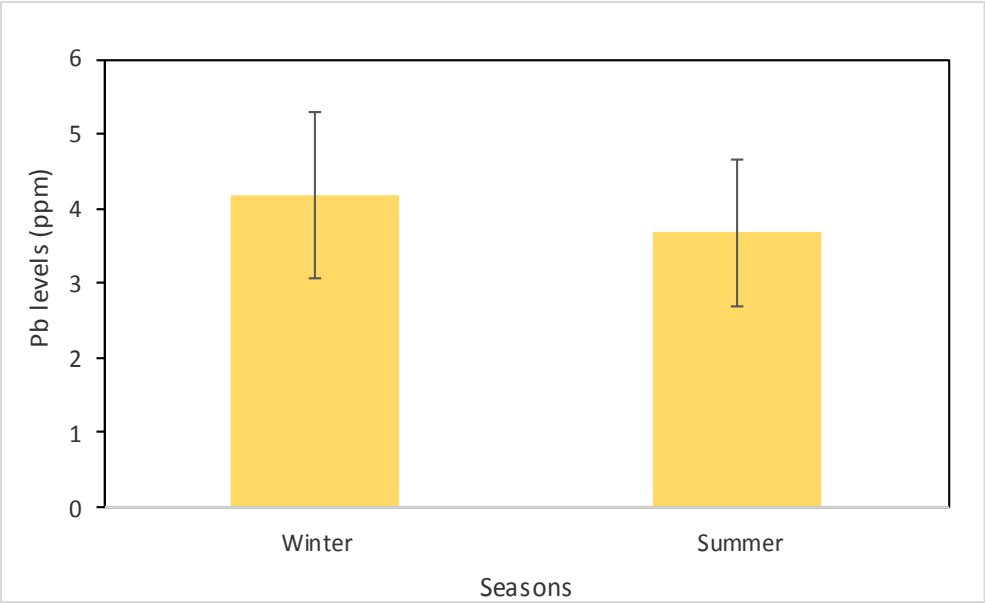

Figure 7: Lead accumulation between winter and summer seasons

Interaction between stations and seasons in terms of Pb accumulation

Walvis Bay had the most lead levels accumulated than any other station in both seasons as can be seen in Figure 8. Statistically, there was no significant interaction (p = 0.068) between lead levels accumulated between the stations and seasons.

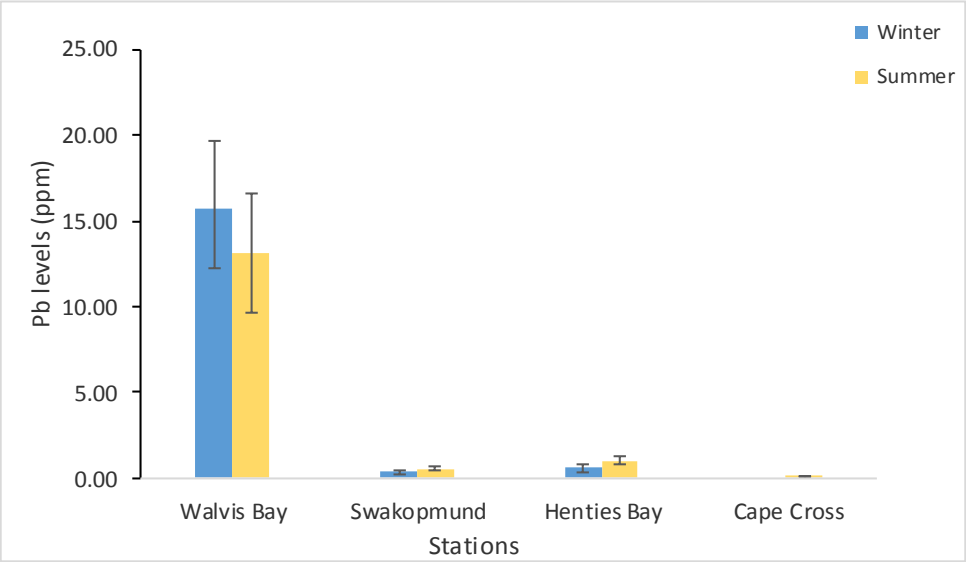

Figure 8: Lead levels across the stations between winter and summer seasons

Interaction of lead between stations and substrates

There was a high significant interaction ($p < 0.05$) in terms of lead accumulation between the stations and substrates as can be seen in Table 4 below. Sediments had accumulated more Pb than any other substrate with a mean of 43.19 ppm.

Table 4: Interaction of lead between stations and substrates

Station	Substrate		
	Mussels (ppm)	**Sediments (ppm)**	**Water (ppm)**
Walvis Bay	0.12 ± 0.004	43.19 ± 3.45	0.01
Swakopmund	0.06	1.23 ± 0.11	0
Henties Bay	0	2.42 ± 0.30	0
Cape Cross	0	0.14	0

Interaction between substrates and seasons in terms of lead accumulation

Statistically, there was no significant interaction ($p = 0.244$) between substrate and season in this study as seen in Appendix 2.

Multiple comparisons of means for Pb using the least significant difference (LSD) between stations

Using the LSD technique to separate the means for Pb across the stations, only Swakopmund – Henties Bay, Swakopmund – Cape Cross and Henties Bay – Cape Cross did not show any significant differences at 0.05 significance level (Appendix 2).

Multiple comparisons of means for Pb using LSD technique between substrates

Using the LSD technique to separate the means for Pb across the substrates, water – black mussels did not show any significant differences at 0.05 significance level (Appendix 2).

Copper:

One remarkable observation of this study is that only copper metal had recorded a direct relationship in terms of accumulation by *Choromytilus meridionalis* in all stations and both seasons. Thus, the value levels of copper recorded in winter had all increased during summer as compared to winter season. Additionally, Walvis Bay station recorded the highest mean values of copper during both winter and summer seasons than any station with values of 2.0 ppm and 3.4 ppm respectively. Again, while Swakopmund station (Walvis Bay's adjacent station) recorded the lowest values of copper on average during both seasons than the rest, it was at Cape Cross station where mussels had accumulated second highest mean values of Cu traces after Walvis Bay station. Henties Bay was third highest in terms of copper mean values accumulated by *Choromytilus meridionalis.*

For water samples, Walvis Bay station registered minimum and maximum values of copper (0.07 ppm, 0.14 ppm) in summer season than the 0 ppm recorded in winter season. The minimum mean value of copper in sediment samples was recorded in Cape Cross station (2.08 ppm) in winter season which had slightly increased in summer season to a value of 2.25 ppm. The maximum mean value of Cu was recorded in Walvis Bay station's sediments (37.16 ppm) in winter season but had declined in summer months (35.16 ppm). Again, both Swakopmund and Henties Bay stations had recorded mean copper values in the range of 5.20 ppm and 6.90 ppm. These values indicate that the levels of Cu had actually increased from winter to summer season. It should also be noted that mean values of copper in sediments were higher than those recorded in mussels and water samples.

Copper interaction between stations, substrates and seasons

Using the 4x2x3 factorial model analysis of a completely randomised design, there was a no significant interaction ($p = 0.716$) between stations, substrates and seasons (Table 5) with regard to copper pollution along the Central Namibian marine coastline (Appendix 2). As can be seen in the summary table below, sediment samples had accumulated more copper in both winter and summer seasons at Walvis Bay than any of the stations under investigation.

Table 5: Copper interaction between stations, substrates and seasons

	Mussels (ppm)		Sediments (ppm)		Water (ppm)	
Station	Winter	Summer	Winter	Summer	Winter	Summer
Walvis Bay	2.16 ± 0.19	3.36 ± 0.33	37.16 ± 2.03	35.16 ± 2.64	0	0.09 ± 0.01
Swakopmund	0.73 ± 0.14	1.06 ± 0.26	5.2 ± 0.46	6.73 ± 2.28	0	0
Henties Bay	0.61 ± 0.12	1.48 ± 0.12	6.76 ± 1.25	6.90 ± 0.24	0	0
Cape Cross	1.46 ± 0.16	2.01 ± 0.20	2.08 ± 0.21	2.25 ± 0.15	0	0

Copper levels across the stations

Copper levels were highly significantly different ($p < 0.05$) across the four different stations of Central Namibian marine coastline (Appendix 2). Walvis Bay station had accumulated more copper than any other station as can be seen in Figure 9 below.

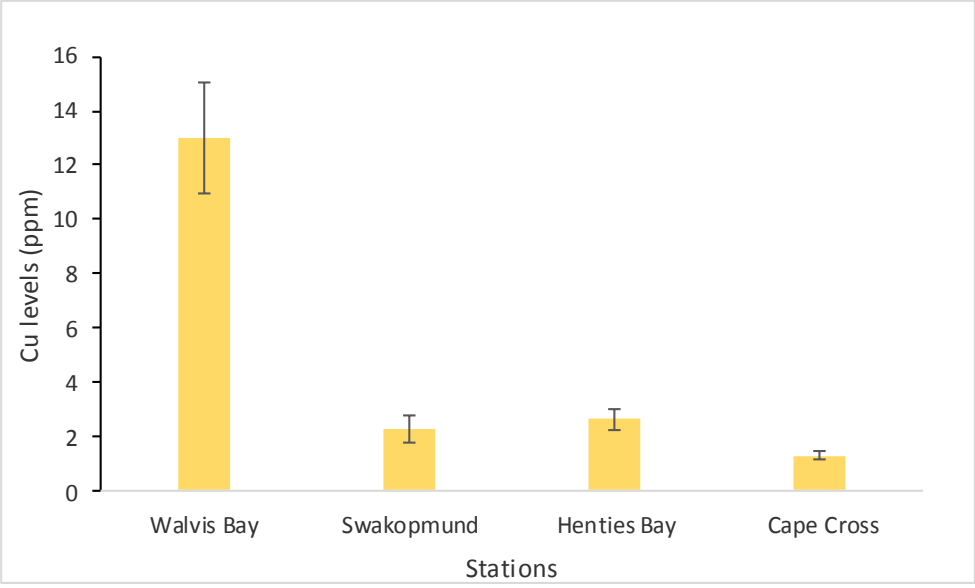

Figure 9: Overall copper levels across the stations

Copper levels in substrates

Statistically, there was a highly significant difference (p = 0.01) across the substrates (water, mussels and sediments) in the Central Namibian marine coastline. As can be seen in Figure 10 below, sediments had accumulated more Cu followed by mussels then water.

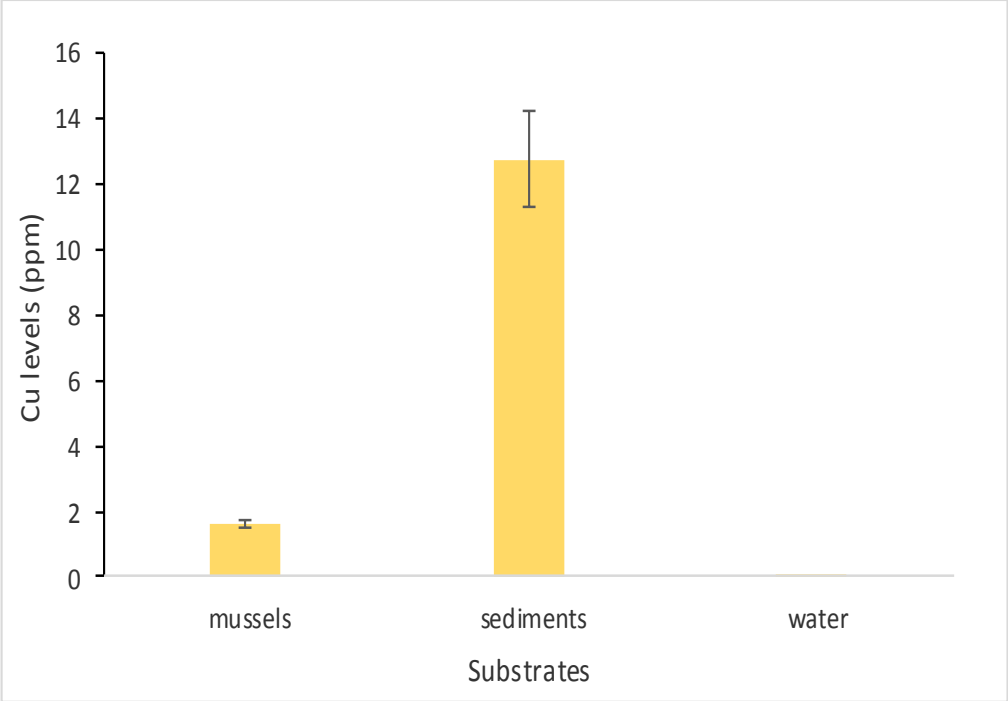

Figure 10: Copper levels accumulated by substrates

Copper levels between the seasons

Statistically (p = 0.505), there was no significant difference in copper levels accumulated between the two seasons despite summer accumulating more Cu than during winter season.

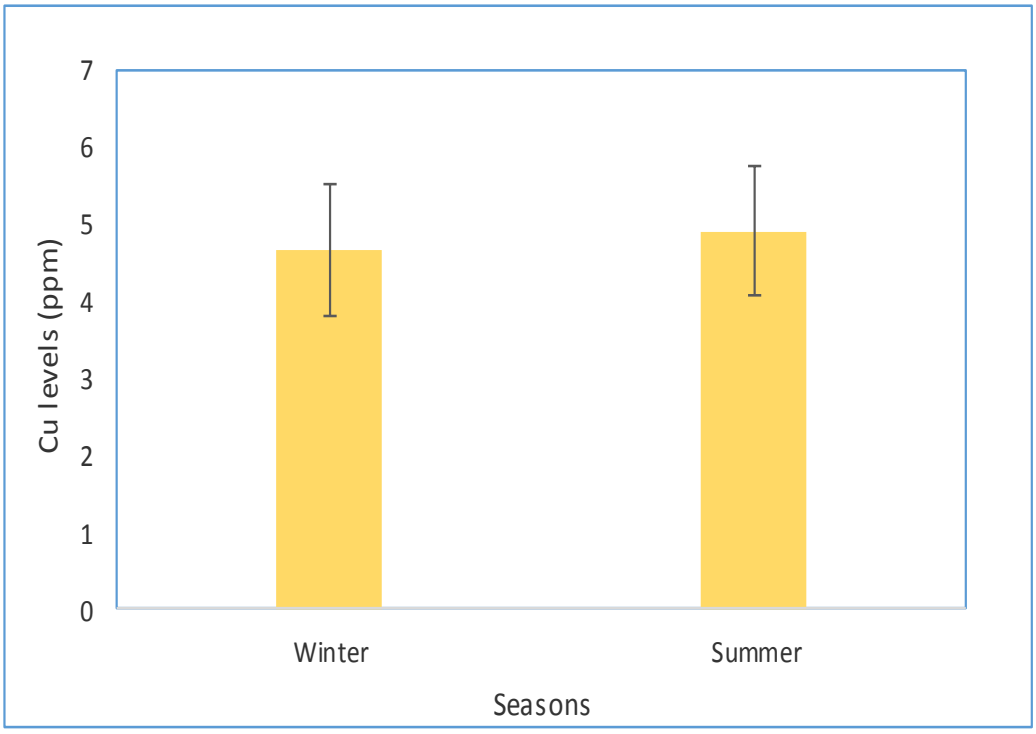

Figure 11: Copper accumulation between winter and summer seasons

Interaction between stations and seasons in terms of Cu accumulation

Walvis Bay had the most Cu levels accumulated than any other station in both seasons as can be seen in Figure 12. Statistically, there was no significant interaction ($p = 0.865$) between lead levels accumulated between the stations and seasons. Except at Walvis Bay station, more copper had been accumulated in summer season as compared to winter season as can be seen in Figure 12.

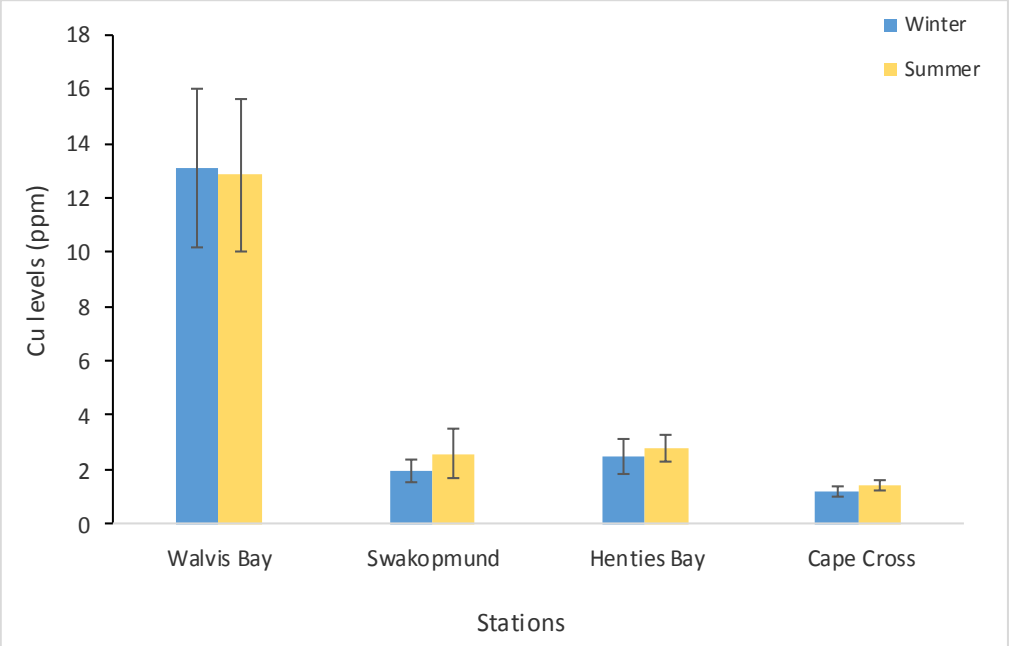

Figure 12: Copper levels across the stations between winter and summer seasons

Interaction of copper between stations and substrates

There was a high significant interaction ($p < 0.05$) in terms of copper accumulation between the stations and substrates as can be seen in Table 6. Sediments had accumulated more Cu than any other substrate with a mean of 36.16 ppm (Table 6).

Table 6: Interaction of copper between stations and substrates

Station	Substrates		
	Mussels (ppm)	**Sediments (ppm)**	**Water (ppm)**
Walvis Bay	2.76 ± 0.23	36.16 ± 5.34	0.05
Swakopmund	0.9 ± 0.01	5.96 ± 1.02	0
Henties Bay	1.04 ± 0.02	6.83 ± 1.23	0
Cape Cross	0	0.14 ± 0.08	0

Interaction between substrate and seasons in terms of copper accumulation

Statistically, there was no significant interaction ($p = 0.616$) between substrate and season in this study (Appendix 2).

Multiple comparisons of means for Cu using the least significant difference (LSD) between stations

Using the LSD technique to separate the means for Cu across the stations, only Swakopmund – Henties Bay and Cape Cross and Swakopmund stations did not show any significant differences at 0.05 significance level as seen in Appendix 2.

Multiple comparisons of means for Cu using the least significant difference (LSD) between substrates

Using the LSD technique to separate the means for Cu between substrates, water and black mussels, all means were significantly different 0.05 significance level (Appendix 2).

Zinc

Interestingly, except for Walvis Bay station, *Choromytilus meridionalis* species from all stations recorded higher values of zinc in winter months but registered a decline in summer season. While Walvis Bay station recorded positive zinc values accumulated by mussels from winter to summer season, Henties Bay alone registered the highest mean zinc values (27.40 ppm) in winter months yet saw a decline in summer (14.10 ppm). Thus, Walvis Bay mean zinc values had actually increased from 14 ppm to 18.30 ppm in winter and summer months respectively. Cape Cross had recorded lowest mean levels of zinc than any other station.

While the minimum and maximum levels of zinc recorded were 0.02 ppm and 0.05 ppm during summer season in Walvis Bay station, the mean value of zinc registered in Walvis was Zn 0.04 ppm in water samples. However, the grand mean value of zinc in all stations was 0 and 0.01 ppm for both winter and summer seasons respectively.

For sediment samples, Swakopmund station had the least mean value of zinc (26.20 ppm) in winter season followed by Cape Cross station (44.20 ppm) while Walvis Bay station registered the maximum value of zinc (75.50 ppm). It is important to note that all stations recorded a decline in Zn values in summer season as compared to winter season. Additionally and in both seasons, Walvis Bay station's sediments topped in accumulating Zn levels than any other station. The grand mean values of Zn recorded across the stations were 44.20 ppm in winter and 19.68 ppm in summer season. This study investigation also observed that zinc values recorded were actually higher in sediments as compared to mussels and water samples.

Interaction of zinc between stations, substrates and seasons

Using the 4x2x3 factorial model analysis of a completely randomised design, there was no significant interaction ($p = 0.137$) between stations, substrates and seasons with regard to Zn pollution along the Central Namibian marine coastline (Appendix 2). The factorial model showed significant differences ($p < 0.05$). As can be seen in Table 7, sediment samples had accumulated more Zn in both winter and summer seasons at Walvis Bay than any of the stations under investigation.

Table 7: Interaction of zinc between stations, substrates and seasons

	Mussels (ppm)		Sediments (ppm)		Water (ppm)	
Station	Winter	Summer	Winter	Summer	Winter	Summer
Walvis Bay	13.57 ± 1.59	18.33 ± 1.54	75.53 ± 3.66	47.27 ± 3.36	0	0.04 ± 0.003
Swakopmund	16.77 ± 2.51	15.05 ± 2.00	26.22 ± 5.73	9.67 ± 0.63	0	0
Henties Bay	27.44 ± 6.31	14.13 ± 0.94	42.86 ± 6.97	18.07 ± 0.76	0	0
Cape Cross	13.42 ± 1.95	8.48 ± 1.07	32.14 ± 7.19	3.72 ± 0.59	0	0

Zinc levels across the stations

Zinc levels were highly significantly different ($p < 0.05$) across the four different stations of Central Namibian marine coastline (Appendix 2). Walvis Bay station had accumulated more lead than any other station as can be seen in Figure 13.

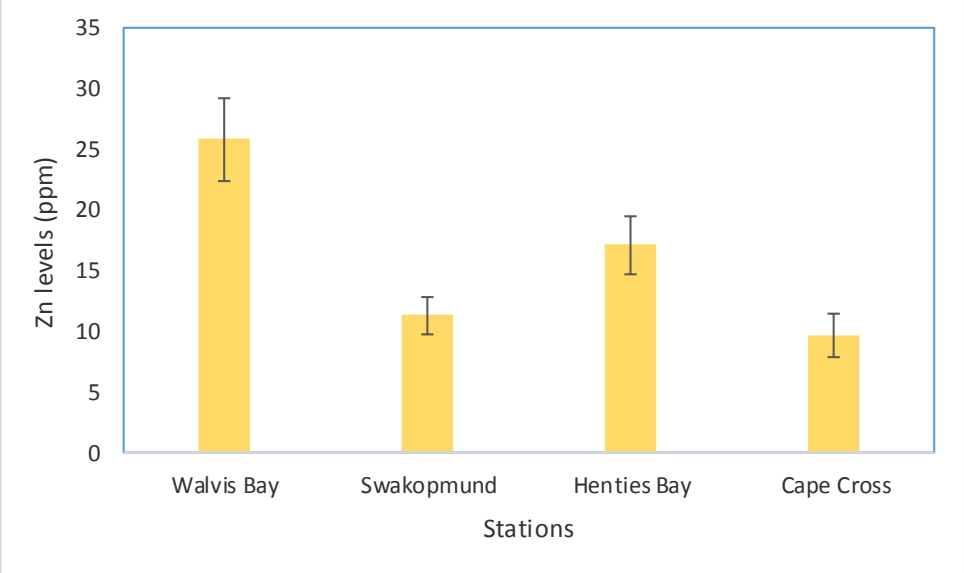

Figure 13: Overall zinc levels across the stations

Zinc levels in substrates

Statistically, there was a highly significant difference ($p < 0.01$) across the substrates (water, mussels and sediments) in the Central Namibian marine coastline. As can be seen in Figure 14 below, sediments had accumulated more Zn followed by mussels then water.

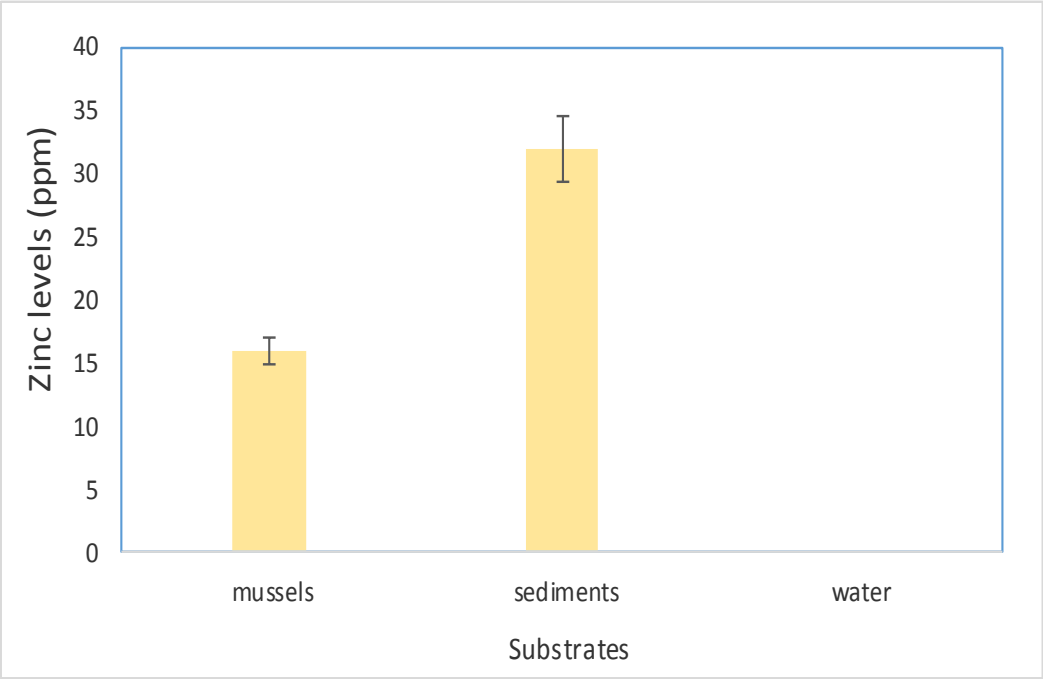

Figure 14: Zinc levels accumulated by substrates

Zinc levels between the seasons

Statistically ($p < 0.05$), there was a high significant difference between Zn levels accumulated between the two seasons despite winter accumulating more Zn than in summer season (Figure 15).

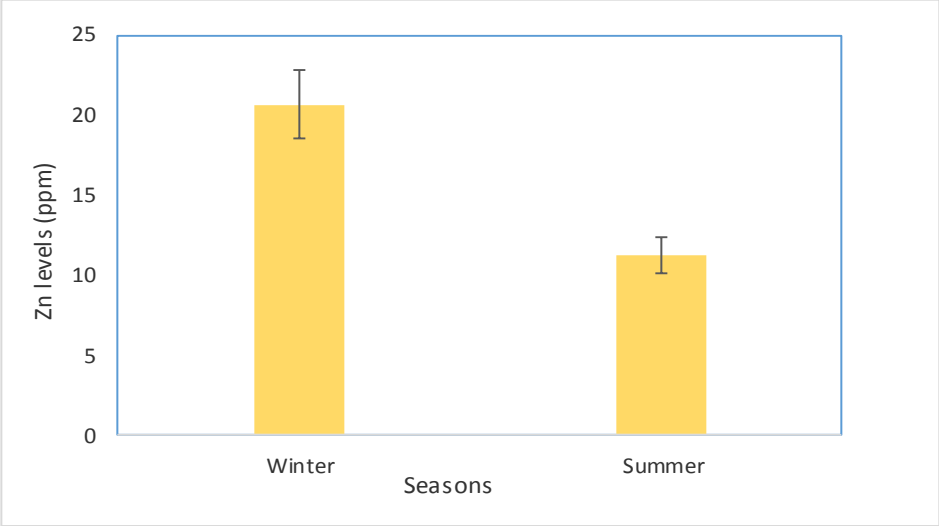

Figure 15: Zinc accumulation between winter and summer seasons

Interaction between stations and seasons in terms of Zn accumulation

Walvis Bay had the most Zn levels accumulated than any other station in both seasons as can be seen in Figure 16. Statistically, there was no significant interaction ($p = 0.219$) between lead levels accumulated between the stations and seasons

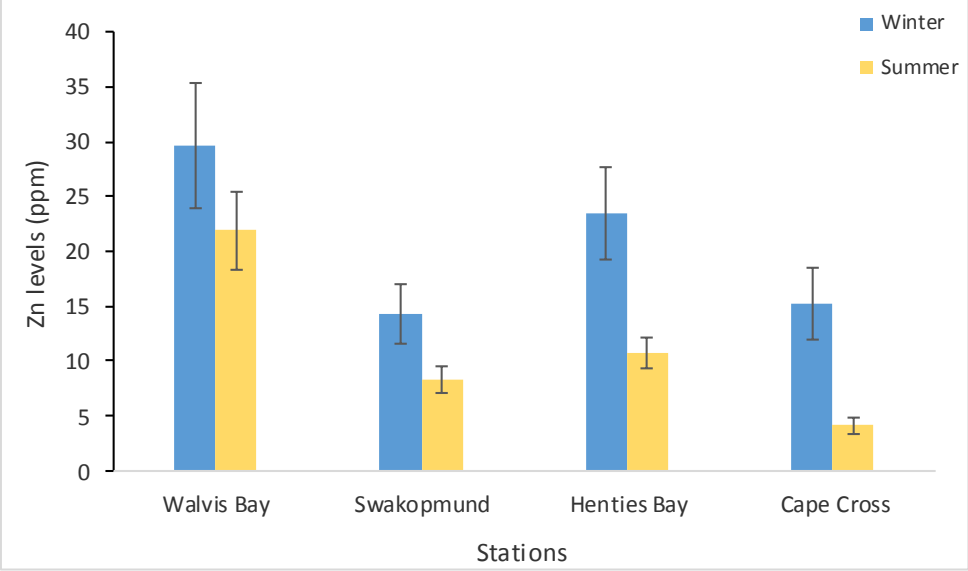

Figure 16: Zinc levels across the stations between winter and summer seasons

Interaction of Zn between stations and substrates

There was a high significant interaction ($p < 0.05$) in terms of Zn levels between the stations and substrates as can be seen in Table 8. As the case with other metals, sediment samples had accumulated more Zn than any other substrate with a mean value of 61.43 ppm at Walvis Bay alone

Table 8: Interaction of Zn between stations and substrates

Station	Substrates		
	Mussels (ppm)	**Sediments (ppm)**	**Water (ppm)**
Walvis Bay	15.95 ± 1.60	61.48 ± 6.95	0.02
Swakopmund	15.91 ± 1.57	17.95 ± 2.34	0
Henties Bay	20.78 ± 2.90	30.47 ± 3.30	0
Cape Cross	10.95 ± 0.91	17.93 ± 3.01	0

Interaction between substrate and seasons in terms of Zn levels

Statistically, there was a high significant interaction ($p < 0.05$) between substrate and season in this study (Appendix 2). Figure 17 illustrates Zn levels accumulated by each of the substrates in both seasons.

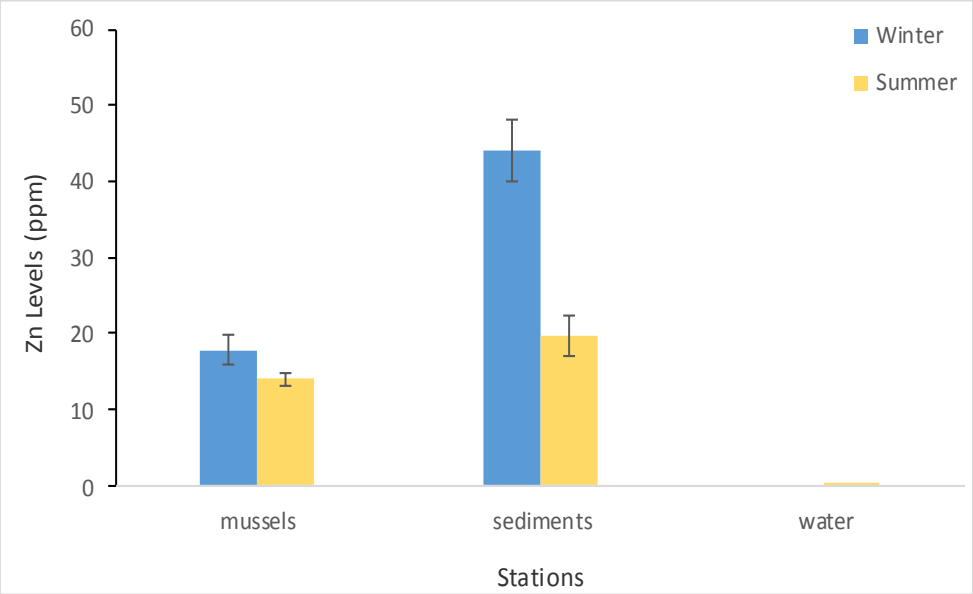

Figure 17: Substrate and season interaction in Zinc levels

Multiple comparisons of means for Zn using the least significant difference (LSD) between stations

Using the LSD technique to separate the means for Zn across the stations, all stations were significantly different ($p < 0.05$) except between station Swakopmund and Cape Cross (Appendix 2).

Multiple comparisons of means for Zn using the least significant difference (LSD) between substrates

Using the LSD technique to separate the means for Zn across the substrates (sediments, water and black mussels), all substrates were significantly different at 0.05 significance level (Appendix 2).

Iron

This study revealed that Fe was the most abundant trace metal in mussel samples compared to the other trace metals in both seasons. The grand mean Fe value during winter for Walvis Bay station was much lower (10.40 ppm) than that recorded in summer in the same station (115 ppm). It is interesting to note that mussel samples collected at Henties Bay station had accumulated higher levels of Fe on average during winter and summer season (139.37 ppm and 103 ppm) respectively; and on average both of these values were still higher than those recorded in the other stations. While Cape Cross registered mean Fe values of 33.10 ppm during winter season, which had increased in summer months (56.18 ppm), Swakopmund was the station that had the least Fe accumulated by mussels on average with mean values of 23.20 ppm in winter and 29.40 ppm in summer.

For water samples, the minimum and maximum values of Fe recorded in summer season were 0.05 ppm and 0.57 while the mean value of Fe recorded in this study was 0.16 ppm for Walvis Bay. The grand mean value of Fe recorded in water samples across the stations was 0.04 ppm.

Henties Bay station had the highest mean value of Fe (20, 152 ppm) in sediment samples during winter which had also increased in summer season and was still the highest and most accumulated trace metal (31, 459 ppm). Both Cape Cross and Swakopmund stations recorded increased iron levels in summer than winter season. However, Walvis Bay station where there are a lot of harbour activities taking place and industries including human settlements recorded a decline of Fe levels in summer than during winter months.

Interaction of Fe between stations, substrates and seasons

Using the 4x2x3 factorial model analysis of a completely randomised design, there was very high significant interaction ($p = 0.001$) between stations, substrates and seasons with regard to iron pollution along the Central Namibian marine coastline (Appendix 2). The factorial model also showed significant differences ($p < 0.05$). As can be seen in Table 9 below, sediment samples had accumulated more Fe in both winter and summer seasons at Henties Bay than any of the stations under investigation.

Table 9: Interaction of Fe between stations, substrates and seasons

	Mussels		**sediments**		**Water**	
Station	Winter	Summer	Winter	Summer	Winter	Summer
Walvis Bay	10 ± 1.21	115 ± 22.73	11055 ± 308.08	7513 ± 379.22	0	0.17 ± 0.05
Swakopmund	23 ± 4.42	29 ± 5.62	6711 ± 564.88	14304 ± 728.29	0	0
Henties Bay	139 ± 46.90	103 34.59	20152 ± 3843.30	31459 ± 1099.70	0	0
Cape Cross	33 ± 4.22	56 ± 21.68	3088 ± 316.50	4408 ± 464.21	0	0

Iron levels across the stations

Fe levels were highly significantly different ($p < 0.05$) across the four different stations of Central Namibian marine coastline (Appendix 2). Henties Bay station had accumulated more iron than any other station as can be seen in Figure 18.

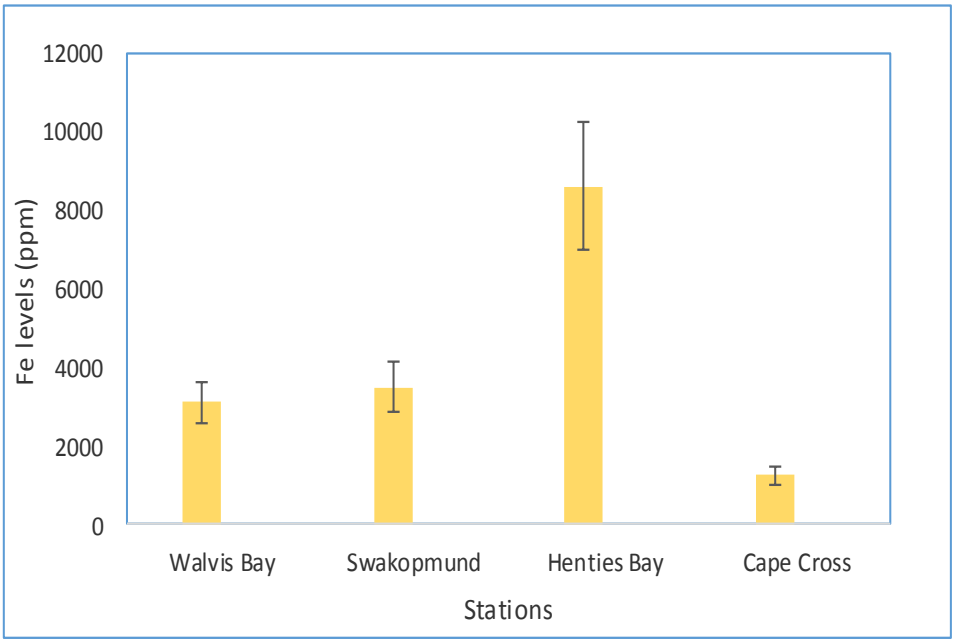

Figure 18: Overall Fe levels across the stations

Iron levels in substrates

Statistically, there was a highly significant difference (p = 0.01) across the substrates (water, mussels and sediments) in the Central Namibian marine coastline. As can be seen in Figure 19, sediments had accumulated more Fe followed by mussels then water.

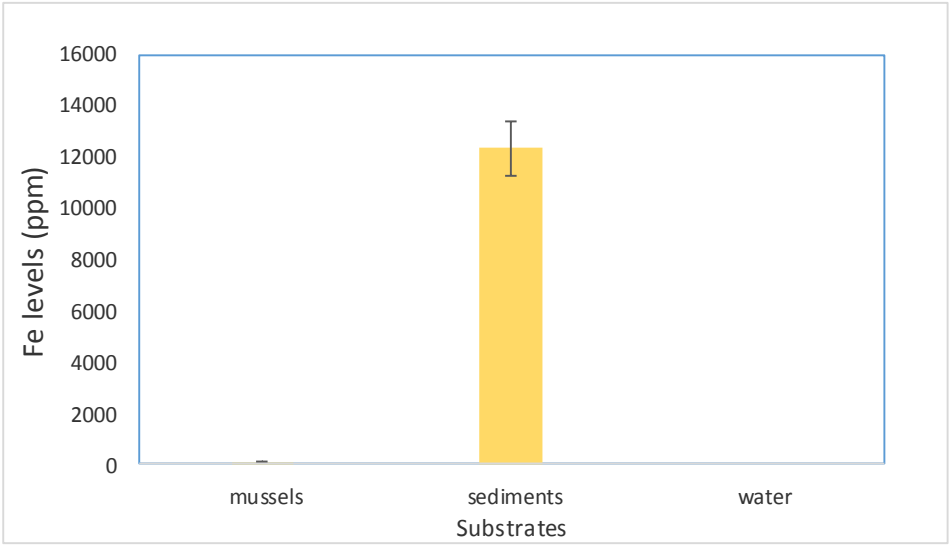

Figure 19: Fe levels accumulated by substrates

Iron levels between the seasons

Statistically ($p < 0.05$), there is a high significant difference between Fe levels accumulated between the two seasons. There was more Fe accumulated in summer season than during winter season as can be seen in Figure 20.

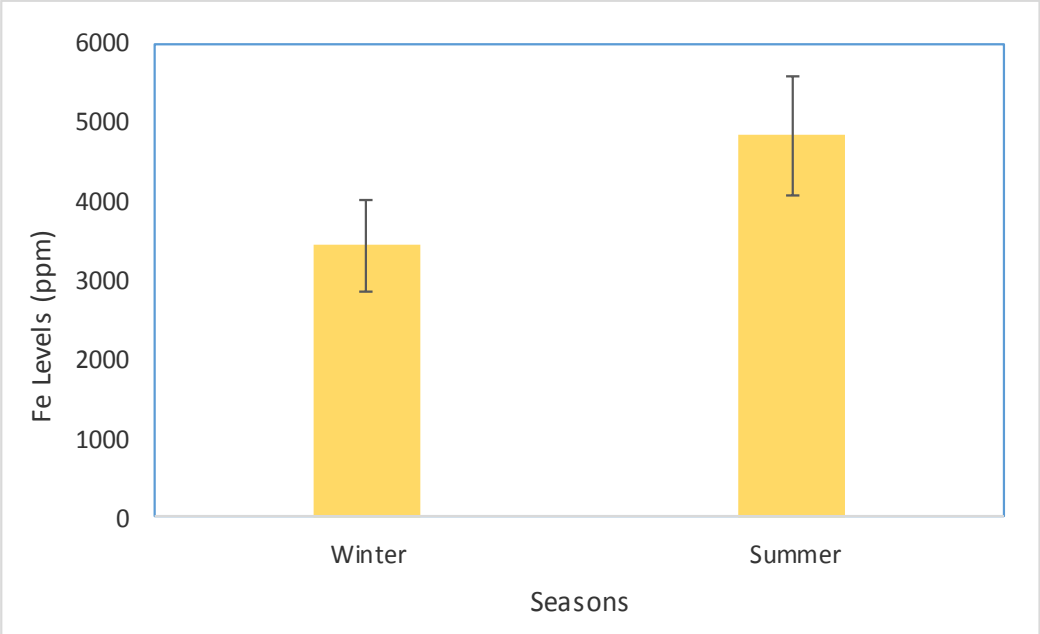

Figure 20: Fe accumulation between winter and summer seasons

Interaction between stations and seasons in terms of Fe accumulation

Walvis Bay had the most lead levels accumulated than any other station in both seasons as can be seen in Figure 21. Statistically, there was no significant interaction ($p > 0.05$) between lead levels accumulated between the stations and seasons.

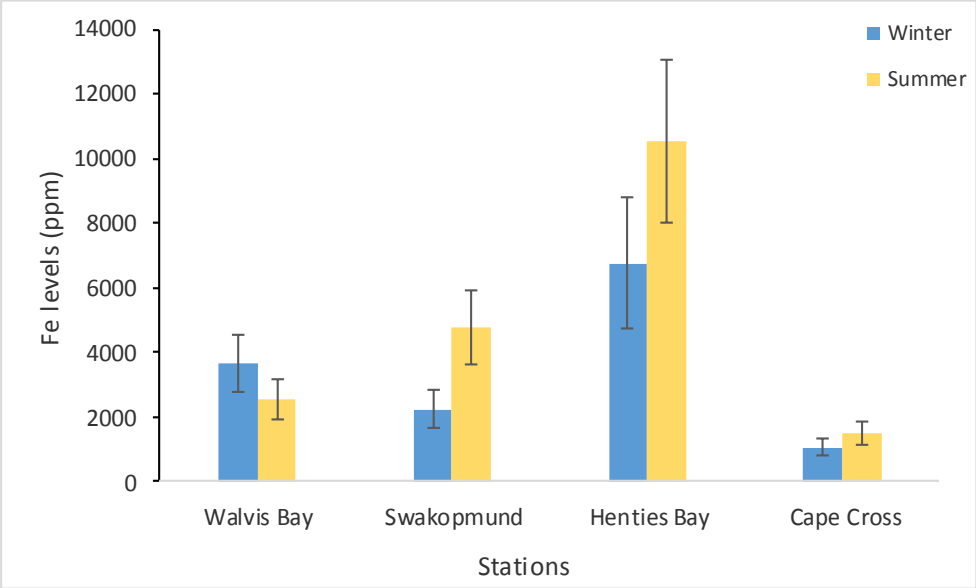

Figure 21: Iron levels across the stations between winter and summer seasons

Interaction of Fe between stations and substrates

There was a high significant interaction ($p < 0.05$) in terms of Fe accumulation between the stations and substrates as can be seen in Table 20. Sediments had accumulated more Fe than any other substrate with a mean of 25, 806 ppm for Henties Bay station while the least was Cape Cross (3, 748 ppm) still higher than those reported in mussels and water as seen in Table 10.

Table 10: Interaction of Fe between stations and substrates

Station	Substrates		
	Mussels (ppm)	**Sediments (ppm)**	**Water (ppm)**
Walvis Bay	63 ± 12.34	9284 ± 805.98	0
Swakopmund	26 ± 3.47	10507 ± 1045.67	0
Henties Bay	121 ± 13.45	25806 ± 4356.72	0
Cape Cross	45 ± 4.41	3748 ± 234.0	0

Interaction between substrate and seasons in terms of Fe accumulation

Statistically, there was a high significant interaction ($p < 0.05$) between substrate and season in this study as seen in Appendix 2. Sediment samples had accumulated more Fe than mussels and water as seen in Figure 22 below.

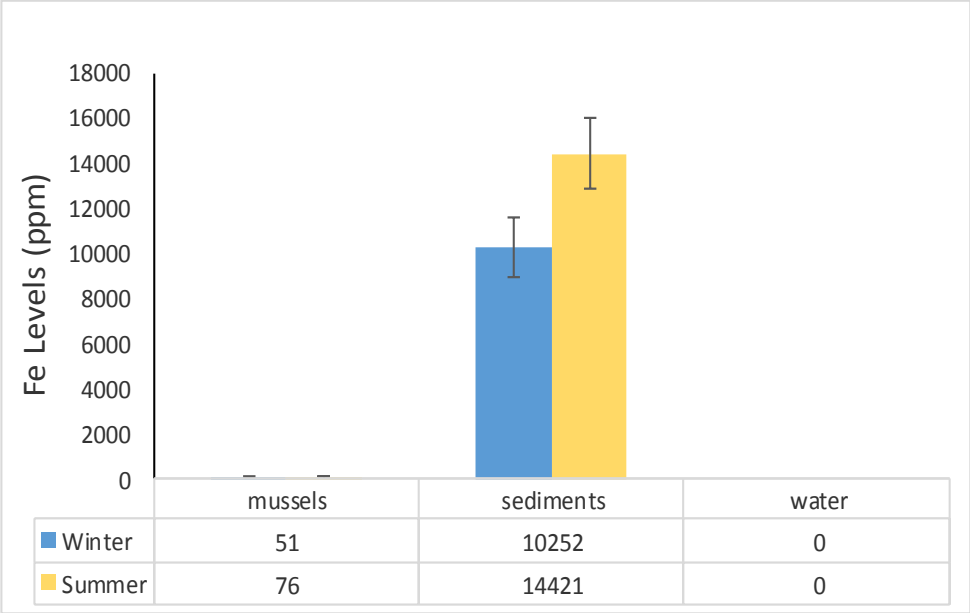

Figure 22: Iron interaction between substrates and seasons

Multiple comparison of means using the least significant difference (LSD) between stations

Using the LSD technique to separate the means for Fe across the stations' Fe levels were significantly different at 5% significance level except between Walvis Bay and Swakopmund stations.

Multiple comparisons of means for Fe using the least significant difference (LSD) between substrates

Using the LSD technique to separate the means for Fe across the substrates, only water-black mussels did not show any significant differences at 0.05 significance level (Appendix 2).

4.2.1 Bioaccumulation factors for each trace metal in *Choromytilus meridionalis*

Lead:

In terms Pb concentration along the Central Namibian marine coastline, it is only at Walvis Bay station during summer season where the BAF was more than 1 between *Choromytilus meridionalis* tissue and water (Table 11). Hence, in this case the BAF can be deduced to be hyperaccumulator.

Table 11: Bioaccumulation factors of Pb in black mussels

Pb	Season	BAF_{water}	$BAF_{sediment}$
Walvis Bay	Winter	0	0.00
	Summer	25	0.01
Swakopmund	Winter	0	0.00
	Summer	0	0.09
Henties Bay	Winter	0	0.00
	Summer	0	0.00
Cape Cross	Winter	0	0.00
	Summer	0	0.00

Copper:

In terms of Cu concentration along the Central Namibian marine coastline, it is also only at Walvis Bay station during summer season (Table 12) where the BAF was more than 1 between *Choromytilus meridionalis* tissue and water. Hence, in this case the BAF can be deduced to be hyperaccumulator.

Table 12: Bioaccumulation factors for Cu in black mussels

Cu	Season	BAF_{water}	$BAF_{sediment}$
Walvis Bay	Winter	0	0.06
	Summer	37.33	0.10
Swakopmund	Winter	0	0.14
	Summer	0	0.16
Henties Bay	Winter	0	0.09
	Summer	0	0.21
Cape Cross	Winter	0	0.70
	Summer	0	0.89

Zinc:

As with the case for Pb and Cu at Walvis Bay station, the BAF between mussel tissues and water can be said to be hyperaccumulator since it is greater than 1. In addition, BAF was also greater than 1 for Zn at Cape Cross station during summer season (Table 13).

Table 13: Bioaccumulation factors for Zn in black mussels

Zn	Season	BAF_{water}	$BAF_{sediment}$
Walvis Bay	Winter	0	0.18
	Summer	458.25	0.39
Swakopmund	Winter	0	0.64
	Summer	0	0.98
Henties Bay	Winter	0	0.64
	Summer	0	0.78
Cape Cross	Winter	0	0.42
	Summer	0	2.28

Iron:

The BAF was less than 1 in all mussel samples in relation to sediments and water and the metal content.

Table 14: Bioaccumulation factors for Fe in black mussels

Fe		BAF_{water}	$BAF_{sediment}$
Walvis Bay	Winter	0	0.00
	Summer	0	0.02
Swakopmund	Winter	0	0.00
	Summer	0	0.00
Henties Bay	Winter	0	0.01
	Summer	0	0.00
Cape Cross	Winter	0	0.01
	Summer	0	0.01

CHAPTER FIVE

5. DISCUSSION

For ease of referencing, this section has been arranged based on the variable studied with detailed discussion on each metal per substrate across the stations including any interaction. The first part discusses the findings of this study on water parameters followed by trace metal levels in mussel samples and metal levels in water samples then in sediment samples. Both winter and summer seasons' metal level findings and correlations have thoroughly been explained followed by a comparison analysis of these trace metals in general across the stations. In addition, the discussion also compares the findings of this study with various studies from different areas around the world.

5.1. Water parameters

Natural waters become contaminated when the contaminant material disturbs the natural balance of living organisms near or in the water or when it makes the water unsafe for human consumption or recreation. Lately, it is evident that human activities are considered to be the most driving factors of water pollution in addition to the natural factors. Anthropogenic activities originated from different sources such as industries and farming practices affect water quality and usually such activities occur in the catchment areas (Ministry of Education, 2010). The analysis of dissolved oxygen in water samples is a key test when it comes to water pollution. Dissolved oxygen is an important component to the survival of most aquatic organisms. Generally, dissolved oxygen plays a vital role in the process of cellular respiration, and if insufficient, aquatic life would not thrive or survive. Aquatic plant populations, precipitation,

rocks in the riverbed, time of the day, water velocity and temperature of the water are all contributing factors that influence total dissolved oxygen in an aquatic body.

It has been reported and commonly known that natural waters usually have pH values in the range of 4 to 9; and most are slightly basic because of the presence of bicarbonates and carbonates of the alkali and alkaline earth metals. However, the pH values of seawater across the stations in the areas of this research investigation were within the value of 8.4 which has been specified by WHO (1987) for coastal waters. It is also widely accepted that pH values between 7 and 8 are optimal for supporting a diverse aquatic ecosystem. Hence, the pH values recorded in this study were slightly higher than that but not significantly different ($p > 0.05$) in winter season to cause alarm or concern despite seasonal variations between the two seasons. Additionally, Swakopmund station registered higher pH, DO and temperature values in in comparison with the other stations although Walvis Bay station recorded the lowest dissolved oxygen levels. In terms of pH for water, however, there were significant differences in pH levels between winter and summer seasons across the stations at 95% confidence interval ($p < 0.05$).

Usually, the water pH is affected mainly by the chemicals discharged by communities and industries because most water bodies become more acidic with time due to the build-up of organic materials. And, taking a glance at the areas of this study, it vindicates and supports this claim as there are almost major tributary rivers along the coastline which discharge effluents into the sea such as the Swakopmund River which is connected to Khan and Gruben Rivers that are well known for mining activities such as copper. In Henties Bay there is the Omaruru River while in Walvis Bay there is Kuiseb River combined with a lot of recreation activities in the area. In all these areas there are also human settlements and these people discharge some amount of

waste into this part of the coastline which might eventually affect the water quality in the long run.

As for temperature, this parameter also has many vital effects on water chemistry and all living organisms in a particular water body. For instance, prevailing literature indicates that water with lower temperatures can hold more dissolved oxygen than warmer waters. This was the expected scenario of this research investigation with the study area being in the cold Benguela current ecosystem. To the contrary, the dissolved oxygen levels were not significantly different. It has also been reported that many biological processes such as spawning and egg hatching in most species such as *Choromytilus meridionalis* are geared to temperature variations. This study recorded temperature levels of seawater above 14.0^{o}C and less than 19^{o}C in both winter and summer seasons. Statistically however, there were no significant differences in mean temperature values of seawater between winter and summer seasons in the stations at 95% confidence interval ($p > 0.05$).

Thus, only pH values between winter and summer seasons showed that there were significant mean differences ($p < 0.05$). Both dissolved oxygen and temperature levels between the two seasons did not record significant mean differences ($p > 0.05$).

5.2. Trace metal levels in *Choromytilus meridionalis*.

Several studies have assessed the bioaccumulation of trace metals in various species of mussels and bivalves in general reaching varied findings in different organs and species themselves. It is not easy however to compare results of this kind of study since responses vary among species and there are also complex environmental factors including threats to organisms (Luoma and Rainbow, 2005). Limited literature on *Choromytilus meridionalis* species also renders

comparison difficult for the findings of this current study. Nevertheless, organisms such as mussels when they are exposed to toxic stress have been reported (Moore and Halton, 1977; Moore *et al.*, 1978; Moore, 1985) to encounter effects at different levels of biological organisation e.g. molecular, cellular and physiological levels. Though this current study did not look at the stress effects of the toxic levels on these study species, *Choromytilus meridionalis* species do accumulate trace metals when exposed to toxic stress (Reineeke and Reineeke, 1999).

This section presents detailed discussion on each of the trace metals accumulated by *Choromytilus meridionalis* species along the Central Namibian marine coastline.

Interaction of lead between stations, substrates and seasons

Using the 4x2x3 factorial model analysis of a completely randomised design, there was a significant interaction ($p = 0.019$) between stations, substrates and seasons with regard to lead pollution along the Central Namibian marine coastline. This most probably indicates that the Pb levels along the coastline were partly influenced by factors such as season. Thus, among the substrat**es** of this study, sediment samples had accumulated more lead followed by mussels and the least being water samples.

Lead in black mussels across the stations

The results of this study showed significant differences ($p < 0.05$) in terms of Pb levels across the four different stations of the Central Namibian marine coastline. Pb was not accumulated by *Choromytilus meridionalis* during winter season despite being recorded in sediment samples across the stations. Some studies (Giarratano and Amin, 2010) have reported that Pb is mainly associated with the residual fraction of the sediment, which could explain why the mussels could

not accumulate this metal during this period. However, it should be noted here once more that the black mussel samples collected from Walvis Bay station came from the Kuiseb Fishing Enterprise Mussel Farm. This farm is located at about 5 kilometres offshore Walvis Bay port and is a protected area from vessels and the species grown in this farm are suspended in the water column. The water depth in this area is about 9 metres. Perhaps the movement of the water cleans up any contaminant traces for these black mussels to accumulate in their bodies or shells as compared to those mussels found right at the harbour area where it has been reported that there is usually a limited water exchange than in open seas. However, Pb value levels were recorded in summer season but only at Walvis Bay and Swakopmund stations. The mean Pb values were actually higher in Walvis Bay Station than in Swakopmund which most probably reflect the port activities taking place in the area than in Swakopmund station. It has been reported (Besada *et al.,* 2011) that close relationship between the atmospheric deposition and/or urban inputs and distribution of Pb in the oceans explains the reason why mussels collected in harbours/bays tend to accumulate high levels of Pb. This study noted that Pb levels had actually been accumulated by *Choromytilus meridionalis* species in summer months and were higher than those values reported during winter season.

Choromytilus meridionalis species collected at Cape Cross and Henties Bay stations did not pick up any Pb amount and most probably indicate that the areas are Pb free zones than the other two stations mentioned earlier since this trace metal was also not recorded in water samples and sediments in these latter stations. Furthermore, it should be noted here that the values recorded in Henties Bay sediments could be due to the ability of the sediments to act as an adsorptive sink for most trace metals. Perhaps the high metal levels could have also been attributed due to sampling and machine errors. Based on the factorial model, the mean Pb value findings of this

current study were significantly different across the stations ($p < 0.05$) but still within the permissible limit of 1 ppm set by the European Community Commission Regulation (EC) No. 466/2001 regarded safe for human consumption (European Community, 2001). Additionally, black mussel samples found along the Central Namibian marine coastline had lower levels of Pb compared to those mussels reported from various sources in Taiwan (Lin *et al.,* 2004) and in the Spanish North-Atlantic coast of 1.3 ppm of dried weight limit for *Mytilus galloprovincialis* (Besada *et al.,* 2011). This therefore should not cause concern for consumers although the Pb level increase or accumulation along the Central Namibian coastline should actually cause an alarm and concern to all since the areas continue to expand for tourism, industrial activities and human settlements including demand for more fishery products in the country and the world as a whole. Finally, there was no significant correlation between Pb levels and rest of the other trace metals of this study in both winter and summer seasons.

Copper across the stations:

Copper is an important element for mussels and it has been widely reported that these organisms do have a biological mechanism to regulate it within their tissues. This study has found out that Cu levels had actually increased from winter to summer season in all stations. This phenomenon was also observed in Galicia and Catabria (Besada *et al.,* 2011) which in fact also agrees that there are high levels of Cu produced in the catchment areas of this study since 1916 especially in Swakopmund station (Taylor and Kesterton, 2002). However, while it is evident that Cu was the only trace metal that showed a positive relationship in mean values accumulated by mussels from winter to summer season in all stations of this study, these value levels were also statistically significant ($p < 0.05$).

A previous study (Lloyd, 1992) stated that copper has entered the aquatic environment by leaching from paints on the hulls of ships. In addition, copper has been reported as used fungicides and algicides. This present study noted that *Choromytilus meridionalis* species collected from Walvis Bay station had accumulated the highest value levels of Cu in both seasons. These values accumulated in Walvis Bay station were actually about six times more than that accumulated by the same species in the rest of the stations. This could be due to the fact that there are activities taking place in this Walvis Bay station ranging from recreation and shipping to all port activities associated which eventually introduce contaminants such as copper in bays (Sulochanan *et al*., 2007).

On the contrary, although black mussel samples from Swakopmund were collected from the Swakopmund river mouth area; and close to the sand dunes where recreation activities are frequent, the mussel samples in this area had accumulated mean Cu value levels lower than those that were collected at Cape Cross station where there are no major recreational activities. There is no major river inflow around Cape Cross area but rather little fishing and accommodation businesses such as the famous Cape Cross Lodge which might suggest that there might be some release of effluents into the ocean that later on lead to an increase in mean copper levels accumulated by these mussels. This lodge also generates its own electricity and desalinates its own water. However, this present study could not verify if the effluents from this lodge were contributing to the high levels of copper accumulated by mussel species in this area. Nevertheless, copper is commonly reported as a naturally occurring element and perhaps the area does not have any geological activities taking place to contribute to copper levels.

The traces of copper accumulated by mussels in Swakopmund and Walvis Bay stations could partly be due to the highly contaminated river tributaries such as the Khan and Gruben rivers in

the catchment. Despite the desalination plant activity at Henties Bay station, the mean value levels of Cu were lower than those reported at Cape Cross but slightly higher than those recorded at Swakopmund station. An explanation for this could be the smaller size of mussels found in this area as compared to the other stations such that body surface area of the animal might be related to increased metal accumulation. This is evident in some earlier research study which claimed that the level of metals in mussel species depends on the metal itself, size of the animal, its age as well as gender which has been claimed to be vital (Pip, 1995). However, this present study did not look at age and size as factors to determine on whether the levels accumulated by *Choromytilus meridionalis* species were due to such reported factors or not. Hence, in terms of copper accumulation by *Choromytilus meridionalis* species in the Central Namibian coastline, this study evidenced that Walvis Bay station recorded the highest levels of Cu than the other stations. This plainly reflects that the kind of activities taking place in the area may have a significant direct relationship with the metal accumulation by black mussel species.

Thus, Walvis Bay station would be considered to be polluted than the rest of the stations in terms of copper content in the species. Compared to other trace metals in this study it is seen that mean Cu values were higher than those reported for Pb but lower than those recorded for Zn and Fe in the same species. Copper is an important part of several enzymes and is necessary for the synthesis of haemoglobin in the body (Sivaperumal *et al.,* 2007). It should be noted however that a high intake of this trace metal has been documented to cause adverse health problems (Gorell *et al.,* 1997). Nevertheless, although the differences in the means of Cu value levels in mussels were extremely significant across the stations and in both seasons ($p < 0.05$) as mentioned earlier; the values of copper recorded were lower than the toxic limit of 30 ppm (FAO, 1983). And, despite the fact that there is no set limit for intake level of copper in Namibia, the

prevailing Cu levels in these species do not pose health problems to the Namibian society.

Furthermore, compared to other studies, the mean value levels of copper accumulated by *Choromytilus meridionalis* species in the Central Namibian marine coastline were found to be generally very low than those reported for the same species sampled along the west coast of South Africa in Bloubergstrand (Orren *et al.,* 1980); the *Mytilus galloprovincialis* (blue mussels) collected along the Albanian coast (Cullaj *et al.,* 2006) and also lower than those values that had been reported in the Montenegrin coastal area of the South-eastern Adriatic Sea (Joksimovic *et al.,* 2011). However, it should be noted that there are various factors to be considered when making these comparisons such as species, age and environmental factors before conclusions are drawn.

Zinc across the stations:

Statistically, there was a highly significant difference ($p < 0.01$) between the substrates (water, mussels and sediments) in the Central Namibian marine coastline. Based on the LSD technique, the differences were observed between water and sediment samples. In addition, the differences Zinc mean values accumulated by mussel samples across the stations in this study were slightly higher than those recorded for copper and lead. The values recorded ranged between 4.1 ppm and 79.3 ppm in both seasons and these were actually higher during winter season but had declined in summer season except for Walvis Bay station which registered an increase. A possible reason for an increased level of zinc in Walvis Bay station could still be that of port activities taking place in the area such as ship repairs as the metals are scrapped off from the hulls into the sea which eventually tend to be picked up by *Choromytilus meridionalis* species.

Additionally, the presence of higher trace metals such as zinc in species tissues indicate that the movement of metals is not localised but spread out and depends on various other factors such as wind velocity and current pattern of the area (Sulochanan *et al*., 2007). Again, the environments (e.g. oceans) receiving the runoff from urban areas have been reported to experience an increase in their concentrations of suspended sediments, nutrients and metals (Pecorari *et al*., 2006). In terms of zinc values, Cape Cross station can be concluded to be less polluted while Henties Bay station would be regarded zinc polluted followed by Walvis Bay and Swakopmund stations respectively.

In comparison with other similar studies, Zn value levels accumulated by *Choromytilus meridionalis* species along the Central Namibian marine coastline were on average four times lower than those reported in the south east Adriatic sea (Joksimovic *et al*., 2011). This would mean that the Central Namibian marine coastline is very much less polluted in terms of Zn. It has been reported that Zn is an essential trace metal for both animals and humans. In addition, the recommended daily intake of Zn has been reported to be 10 mg/day for children and 15 mg/day for adults (Sivaperumal *et al*., 2007). Also, Zn has been reported to have a defensive effect against the toxicities of both cadmium and lead (Malik *et al.,* 2010). However, there is no documented and regulated limit for intake of Zn in Namibia; and findings of this study might probably just aid in setting a baseline in this case as the value levels accumulated by *Choromytilus meridionalis* species in Central Namibian marine coastline are higher than the recommended limit.

Interaction of Fe between stations, substrates and seasons

Using the 4x2x3 factorial model analysis of a completely randomised design, there was very high significant interaction ($p < 0.001$) between stations, substrates and seasons with regard to iron pollution along the Central Namibian marine coastline. Sediment samples had accumulated more Fe than water and mussels.

Iron across the stations

Iron is an important element in an organism's life (Cardellicchio *et al*., 2008). This trace metal was the most accumulated trace metal element by mussels than any other metal in this study. The mean value levels of iron picked up by *Choromytilus meridionalis* were significantly different in the Central Namibian marine coastline in both seasons. Precisely, the value levels of Fe were higher in Henties Bay station than any other station. An explanation of this phenomenon could be the weathering of the rocks in close proximity and in the shoreline, which are most likely Fe-rich such that these mussels tend to accumulate them when they are broken down and transported into the sea. This weathering could also be intensified or attributed to the wave action, the Benguela Upwelling and the easterly wind action originating from the Namib Desert inland into the South West Atlantic Ocean. It is also widely known that Fe is a naturally occurring and commonly available element in an ecosystem because of the rocks which could also explain why the mussels accumulated more Fe than the other metals.

Swakopmund station could be regarded as less Fe polluted followed by Cape Cross station despite both stations indicating a positive Fe increment in accumulation from winter to summer season. However, Walvis Bay stations' Fe levels are a cause of concern as the mean values had drastically increased from winter to summer despite the high levels recorded in Henties Bay

station. In addition, considering that Walvis Bay stations' mussels are farmed and used for human consumption, the Fe levels accumulated by these species were higher than the acceptable trace metal intake limit of 48 mg per day (Joint FAO/WHO, 1999). Compared to Fe levels accumulated by *Mytilus galloprovincialis* samples from different locations along the Ionian Sea in Southern Italy (Cardellicchio *et al*., 2008), Fe levels accumulated by *Choromytilus meridionalis* samples along the Central Namibian marine coastline were in fact lower than those reported in Italy meaning the Namibian coastline is less polluted in terms of Fe content.

No significant correlation was found between Fe and Cu in this study during winter season. This pattern was also observed along the west coast of South Africa (Orren *et al*., 1980).

5.3. Trace metal levels in sediments

Contaminants can persist for many years in sediments and have the potential to negatively affect human health and the environment (Obirikorang *et al*., 2011). When contaminated sedimentary material is released into an ecosystem such as an aquatic water body, it equilibrates with the water, silt and living food materials which results in ongoing contamination of all environmental compartments. It has been claimed (Luoma *et al.,* 2008) that organisms such as *Choromytilus meridionalis* get exposed to this situation throughout their lifetime to the point that this type of sediment metal accumulation is thought to reduce or eliminate populations of fish or mussels without killing them (e.g. by inhibiting reproduction) or without leaving visually clear evidence of effects. In addition, particle size of the sediment renders it hard to interpret the metal levels in sediments because highest level of metals are usually associated with fine grained sediments; and those dominated by sand typically have lower metal levels because of smaller ratio of surface area to mass.

Furthermore, separation of the fine fraction of sediments for trace metal analysis reduces the physical variability among sediment samples which in turn reduces the impact of grain size variability on concentration (Luoma *et al.,* 2008). It is also a well-known fact that historically sediments do contain higher levels of metals than the mussels and water. These are just some of the many reasons why this study had to assess the levels of trace metals in sediment samples along the Central Namibian marine coastline. Hence, based on the findings of this study, the levels of trace metals accumulated in sediment samples along the coastline were found to be extremely higher than those recorded in *Choromytilus meridionalis* and water samples in the same stations. In addition, all trace metals showed intra and inter seasonal variations in terms of sediment accumulation across the study stations. Thus, this section discusses the findings in detail for this phenomenon and in comparison with various studies as the case for trace metals in *Choromytilus meridionalis* species.

Lead:

Pb was the least accumulated trace metal in both sediments and mussels including water samples. This metal has a tendency to bioaccumulate but at low levels in terms of distribution in the marine environment (Cardellicchio *et al*., 2008). Sediment samples collected from Cape Cross station did not accumulate any values of Pb in winter months while in summer season, this station recorded some levels indicating the area experienced an increase in Pb levels in sediments for some reason. However, these values were lower than those recorded in Swakopmund and Henties Bay stations though Walvis Bay station, which is active in terms of port activities recorded Pb values of about 40 times higher than any station in both seasons despite a slight drop in levels of this metal in summer season. Additionally, mean Pb values across the stations showed that they were highly different and significantly ($p < 0.05$). Again, Walvis Bay station

could be regarded more Pb contaminated than the rest of the stations with regard to mean value levels of Pb on comparison basis.

Several studies have documented that sediments act as a scavenger and an adsorptive sink for trace metals (Giarratano and Amin, 2010; Olowu *et. al*., 2010); and that paint scrapping off from vessels and boats eventually contribute to the accumulation of Pb in a water body (Lloyd, 1992). In addition, it has also been reported that in bays there is a limited water exchange to clean up the system (Sulochanan *et al*., 2007) which probably explains why there is an increase in metal content in Walvis Bay than the other stations which are not bays. Also, Walvis Bay town is an industrial town coupled with a high number of people residing in this area such that factories and industries discharge their effluents into this part of the ocean. Several recreation activities in this area including the port works all act as anthropogenic sources of these contaminants and this could possibly explain the high level of Pb recorded in sediment samples for this station. Swakopmund station's Pb levels could probably be attributed due to its adjacent location with Walvis Bay station in which there is no clear boundary to demarcate the water flow. In addition, the area has some major rivers in the catchments which tribute their waters into the Swakopmund River and town alongside its well-known recreation activities. As for Cape Cross station, no major activities take place in this area and no significant human settlement for one to assume that the activities could be sources of lead contamination although there are fishing and accommodation businesses in the area. Perhaps these activities do not significantly contribute to Pb levels and hence no traces of Pb were recorded in winter and little in summer. Mean values of Pb in Walvis Bay station's sediments show that the area is far more polluted as they were twice as high as those reported in Beagle Channel of Argentina (Giarratano and Amin, 2010) while the

rest of the stations recorded way less Pb content in sediments and could be regarded as less polluted.

Copper:

One way analysis of variance of a completely randomised design showed highly significant differences in mean values of Cu ($p < 0.05$) in sediment samples across the stations of this study both in winter and summer season. As expected, Walvis Bay station recorded the highest levels of copper in sediment samples (37 ppm) compared to the other stations in which the mean value levels of Cu were within the range of 2 ppm and 7 ppm. The grand mean value of Cu accumulated in sediment samples were higher than those recorded for Pb in same stations. However, worth noting is that the mean value levels of Cu in sediments showed a very good correlation with Pb levels between winter and summer seasons ($r = 0.969$; $r = 0.909$) respectively. This might explain why this station recorded higher values of this trace metal as the presence of one had an influence on another. Additionally, Cu values recorded in the Central Namibian marine coastline's sediments had slightly increased from winter to summer season in three stations except at Walvis Bay station which had recorded a slight drop. The accumulation of Cu is explained by Lloyd (1992) findings that Cu originates from the hulls of ships. Of course it is also true that fishing, transport and most cargo vessels pass through this area of the sea en-route to Walvis Bay Port and other parts of the world such that high volume of traffic in this regard could lead to trace metal accumulation such as Cu.

Furthermore, some previous studies (Taylor and Kesterton, 2002) on the heavy metal contamination of the Gruben River in Namibia, which is a tributary of the Swakopmund River, reported that the river area has a well-documented history of copper mining dating back to 1916.

Findings of this study could prove that these claims are actually true because the levels of copper in Swakopmund station had actually increased between the two seasons. Additionally, the results of this study could infer that the Central Namibian marine coastline is far more polluted in terms of Cu content accumulated in sediment samples than those reported in Palk Bay and Gulf coastline of Mannar of the Indian Ocean (Sulochanan *et al*., 2007) though this study established that there were no significant differences in mean value levels of Cu between the two seasons.

Zinc:

In this present study Zn was the second most abundant trace metal accumulated in both sediments and mussel samples along the Central Namibian marine coastline. Statistically, the differences in mean values of Zn accumulated in sediment samples were highly significant across the stations during winter and summer season. Like the accumulation pattern with other trace metal levels in Walvis Bay station, Zn mean values recorded in this station were above 70 ppm in sediment samples. The other stations recorded Zn value levels of less than 45 ppm in sediments with Swakopmund and Cape Cross stations being the least in Zn content on average despite the latter recording a sharp decline from winter to summer season. Nevertheless, the grand Zn mean levels across stations was recorded to be 44.27 ppm during winter months, about three times higher than that recorded for Cu and Pb but lower than Fe. Though the mean values of this metal had dropped significantly in summer months from winter, the levels were still above those recorded for Pb and Cu. With mean Zn levels ranging between 3.72 ppm and 75.50 ppm in this present study, it is also important to note that Zn was the only trace metal that had declined (negative relationship) in terms of accumulation in sediment samples along the Central Namibian marine coastline from winter to summer season across the stations. This finding is

contrary to Cu which had displayed a positive relationship in *Choromytilus meridionalis* samples.

Prevailing literature (Otchere, 2003) reports that most metals such as Zn are found in agricultural products e.g. fertilisers used in agricultural practices that eventually may accumulate in soils and become exposed to water bodies through runoffs in rainy season. However, Zn values recorded in sediment samples along the Central Namibian marine shoreline were lower than those reported in Brown Bay (Giarratano and Amin, 2010) and in Chilean Andes (Lamy *et al.,* 2001; Dezileau *et al.,* 2007) but could be regarded as more polluted compared to Palk Bay and Gulf of Mannar (Sulochanan *et al*., 2007). Unlike Zn levels reported in the Spanish North-Atlantic coast (Besada *et al*., 2010), this study found the levels of Zn in the Central Namibian marine coastline to be heterogeneous in all stations and both seasons yet still lower. Furthermore, winter Zn levels recorded in sediments along the Central Namibian marine shoreline were actually higher than those reported in the four Spanish North Atlantic coastline locations (Zanjice, Sv. Stasija, H. Novi and Bar) but less than Rt Deran location. Even Zn levels recorded in sediments collected from Walvis Bay station in both seasons were in fact lower than this Rt Deran location.

Iron:

Using a multivariate analysis (MANOVA), this study showed that there were significant differences in mean Fe value levels across the stations in winter season but highly significant mean differences in summer season. As observed in Fe values recorded in the South East of Adriatic sea (Joksimovic *et al.*, 2011), Fe was by far the most abundant trace metal accumulated in sediment samples across the stations of this investigation. This metal had increased in accumulation in all stations from winter to summer season except in Walvis Bay Station as the

case observed for Pb and Cu metals in the same station which had both declined in levels. The mean value levels accumulated in sediment samples along the Central Namibian marine coastline during both winter and summer season across the stations were surprisingly about 1, 000 times higher than those recorded for Pb and Cu. This simply indicates how abundant Fe is in the Central Namibian marine coastline. Additionally, Henties Bay station had the most Fe content in sediment samples than the rest of the stations ($p < 0.05$); and could be regarded as more polluted than Cape Cross, Swakopmund and Walvis Bay stations respectively. It is also important to note that this metal was the most distributed along the Namibian coastline.

One of the reasons for the presence, abundance and distribution of Fe could be the physical action brought by the Benguela Upwelling in the Namibian waters which most probably bring along sedentary rocks. The wind also contributes to the accumulation of sediments due to weathering of the available rocks along the shoreline.

Another possible explanation of this accumulation increase of Fe in sediment samples in the shoreline is reported by Obirikorang *et al*., (2011). Their study has stated that higher levels of Fe and Zn might as well be due to import from surrounding settlements as most roofing are made of galvanized iron sheets, most of which are presently rusty. This study observed that there are more settlements along the Central Namibian marine coastline (scattered and stretched apart from each other) with old buildings which could support this rust idea as a contributing factor to trace metal pollution in the shoreline. However, this study could not verify this claim but it could prove to be true based on the levels of Fe recorded in this study. Even Henties Bay station sediments which recorded quite significant amounts of Fe than the rest of the stations does not really justify this suggestion as there are few people living in this area as compared to

Swakopmund or Walvis Bay towns which also have industrial activities and mines in close proximity yet they recorded way lower amounts of Fe in sediment samples.

Perhaps the various stones and rocks observed in these areas are Fe rich and the weathering due to the Easterly strong winds blowing from the Namib Desert and inland into the Namibian coastline, combined with the wave action and the physical forces brought by the Benguela currents, are directly related to the high levels of Fe accumulated in sediments. On the other hand, it is interesting to note that Walvis Bay station had experienced a drop in Fe content from winter to summer season as the scenario that had been observed in Pb, Cu and Zn content. This study could not answer this phenomena and perhaps another investigation needs to be done and on a long term basis to monitor if the situation remains constant or not and why.

Furthermore, a different study (Olowu *et al*., 2010) reported that high levels of Fe in sediment samples may be because of the clayey material that forms the river beds in catchments as the case for this study. This is complemented by another study (Heine and Völkel, 2010) that further suggests and claims that clay minerals are the most common components of all sediments and soils which are found in the soils and sediments from polar to equatorial regions, from sea level to high mountains, from deserts to rain forests and in marine sediments from coasts to deep seas. Thus, based on the Namibian geographical physical features (land forms and drainages such as rivers and a lot of clay ant hills in the country), it probably also gives an explanation as to why Fe was the most abundant metal than any other in the study stations. Nonetheless, high levels recorded may also be attributed to the ability of the sediments to act as a sink (Giarratano and Amin, 2010; Olowu *et al*., 2010) and anthropogenic practices leading to its accumulation.

Except for sediment samples collected at Cape Cross station, mean Fe values were much higher than those reported in Ghana's Volta Estuary sediments (Madkour *et al.,* 2011). These values were also higher than those reported in the South East of Adriatic sea (Joksimovic *et al*., 2011) and the four stations (Zanjice, Sv. Stasija, H. Novi and Bar) on average during both winter and summer seasons and years despite both studies reporting variations in sediment Fe accumulation. However, when compared to one single location (Rt Deran) in their study, the Central Namibian marine coastline could be regarded as less polluted in Fe content in sediments. Again, it is true that compared to Brown Bay in Argentina (Giarratano and Amin, 2010), the Central Namibian coastline is extremely polluted in terms of Fe levels in sediments. Thus, all values of Fe recorded in sediment samples along the Central Namibian marine coastline have been reported elsewhere.

5.4. Trace metal levels in the water column

Trace metal pollution of seawater most probably has long term effects many of which are still unknown and pose increasing risks for many species such as humans, fish and shellfish or bivalves. The state of freshwater resources, acid rain and various activities in harbour areas all contribute to the deterioration of coastal waters and oceans in general (Ministry of Education, 2010). It is therefore critical to conduct various toxicological tests to monitor the ocean pollution state and the environment broadly. Hence, this section presents and outlines detailed findings and discussion of this study with regard to the accumulation of trace metals in the water column in relation to sediments and mussels metal levels of the Central Namibian marine coastline. However, it should be noted that this study did not record any trace metals in seawater samples during winter season in all stations. Again, the findings of this study recorded significant mean trace metal values during summer season ($p < 0.05$). The least but not surprising finding of this

study was that all stations did not record any trace metal values except for Walvis Bay station as was the case for most metals in sediment and mussel samples.

The quality of water affects all living organisms in the Namibian coastline. This study recorded trace metal levels in Walvis Bay station's seawater during summer. These values were significantly different at 95% confidence interval. The reason why there were no trace metals recorded during winter season could be that of sampling location difference of the water samples. It should be noted once more that during winter season, seawater samples were collected from the Kuiseb Fishing Enterprise Mussel Farm which is situated at approximately 5 km from the harbour. This area is calm in terms of physical disturbances other than the upwelling, such that water exchange is seen to be good which allows free flow and movement of water that in turn clears up the suspended particles rendering no single trace metal recorded for this study during winter season. This could also prove to be true for the other stations to some degree.

On the contrary and for comparison purposes, summer seawater samples were collected right on the Walvis Bay harbour to determine if there would be any significant differences between the metal levels. This was bearing in mind that Walvis Bay harbour is a very active port hence prone to trace metal contamination from various kind of sources such as sewerage discharges, mining and fisheries discharges including human settlement (The Ministry of Education, 2010). Overall, this study proved true as the findings found significant trace metal levels ($p < 0.05$) in this vicinity during summer. Again, the pH of seawater between the two seasons had changed significantly and could perhaps also indicate why water samples had accumulated some trace metal levels in this period as compared to winter season. Finally, these trace metals had also evidenced very good correlation in terms of accumulation with an exception of Pb and Fe.

Lead:

Maximum recorded values of Pb including those in Walvis Bay station's seawater collected from the harbour were higher than those reported in Palk Bay and Gulf of Mannar (Sulochanan *et al*., 2007). However, this study has revealed that seawater in the Central Namibian marine coastline has not exceeded the permissible limit set by the European Community Commission of 0.02 ppm regarded safe for human consumption. The value levels of this metal recorded in Walvis Bay station could partly be due to the Benguela upwelling system (Bruland, 1983) and various physical factors that occur in the area including the wave and wind action that bring along particulates of sand from the dunes and the settlements.

Copper:

Maximum value levels of Cu including those recorded in Walvis Bay station's seawater collected from the harbour were extremely higher than those reported in Palk Bay and Gulf of Mannar (Sulochanan *et al*., 2007). However, the mean value of Cu along the Central Namibian marine coastline exceeds the permissible limit specified by European Community Commission of 0.01 ppm regarded safe for human consumption. This should cause an alarm to all parties. The traces of Cu element recorded in this study most probably are contributed by the mining activities taking place around Swakopmund and Walvis Bay including shipping and all harbour activities in this area.

Zinc:

The value levels of Zn in seawater vary largely (Besada *et al*., 2010). The maximum value levels of Zn including those recorded in Walvis Bay station's seawater collected from the harbour were

extremely lower than those reported in Palk Bay and Gulf of Mannar (Sulochanan *et al*., 2007). Still, Zn value levels recorded in the central coastal waters of the Namibian marine coastline do not exceed the permissible limit set by the European Community Commission of 0.04 ppm and could be regarded as safe and not polluted. Compared to Zn value levels reported in Besada *et al*., (2011), the Central Namibian coastline's seawater could be regarded as very clean and pollution free. However, climatic conditions (Cantillo *et al.,* 1997) in the Namibian coast could explain why some traces of this metal were accumulated in seawater

Iron:

As seen with the tendency of this metal accumulation by mussel and sediment samples, mean values of Fe recorded in seawater were higher than any other trace metal under investigation. The maximum values of Fe were observed in Walvis Bay station's seawater which reflects the various kinds of activities that occur in this part of the coastline from human settlements to port activities. The reasons for Fe accumulation in seawater have already been outlined for *Choromytilus meridionalis* and sediment samples. Thus, the abundance and distribution of Fe in the Central Namibian coastal waters could be due to the physical actions brought by the Benguela upwelling that mixes the water column including most sedimentary material at the bottom. This turning of nutrients in turn leads to trace metal accumulation as some rock particles break up in the process. In addition, the wind also contributes to the accumulation of sediments due to weathering of the available rocks along the shoreline. Ship repairs and industrial activities that involve breaking and casting of metals also do contribute to Fe content in the long run as most metal bars do contain Fe amounts in their compositions.

To sum it all and as a general observation with regard to water column, this study observed that seawater samples obtained from the harbour at Walvis Bay station, an urban and industrial area, had recorded higher values of trace metals than water samples collected from Swakopmund, Henties Bay and Cape Cross stations respectively. These areas do not have many activities as compared to Walvis Bay station such as the port. Nevertheless, it has been reported (Sulochanan *et al*., 2007) that these trace metals can be removed from the water column only by accumulation in the biota or by sedimentation though the values recorded in Namibia generally do not pose health risk at the moment to take that action. The increasing levels of these metals should however be a cause of concern to all. There is also a need for continued monitoring of these trace metals along the Namibian marine coastline to ensure the levels do not exceed the permissible limits. It will also ensure that the Namibian coastline in general is environmentally pollution.

5.5. Seasonal variations of trace metals in water, sediment and mussel samples

Various studies (Cardellicchio *et al*., 2008; Giarratano and Amin, 2010; Boateng *et al.*, 2011) have documented seasonal changes in trace metal levels in mussels as has been reported in this present study between winter and summer season. In this study, the two seasons had both recorded significant mean trace metal differences in all three substrates across the stations. One notable observation is that the percentage metal accumulation change by mussels was higher than in sediments which indeed substantiate some claims that metal levels in mussels do accumulate at the end of winter season. Thus, a very possible explanation of this development could partly be due to the reproductive activity of mussels (Cardellicchio *et al*., 2008) as gonadal development might be attributed to the accumulation of proteins and carbohydrates during spawning; and for gonad tissue production and energy consumption (Boateng *et al.*, 2011). Conversely, despite all trace metals of this study yielding variations between the two seasons,

only copper had showed a 100% positive relationship in terms of accumulation in mussel species with an increase in summer from winter. This observation is similar to the findings of Boateng *et al.*, (2011) who studied *Galatia paradoxa* (Born 1778) in the Volta Estuary and observed a peak accumulation of different but similar metal manganese (Mn) within the same study months. However, this study did not look at reproduction of *Choromytilus meridionalis* species and there is no referenced and documented data about the exact reproduction period/months for these species in the Central Namibian marine coastline to conclude that reproduction occurs during the same period as those reported elsewhere.

Additionally, the other trace metals recorded higher mean values during winter than summer season which was similar to some previous studies (Giarratano and Amin, 2010) that suggested it could be due to factors such as food supply for *Choromytilus meridionalis* population, fresh water runoff of particulate metal to the coastal water, weight changes caused by gonadal development and release of sexual products. For this study though, suggesting that a large difference in seawater temperature might affect metal variations (Szefer *et al.*, 2004) could not be true as the temperature changes were not significant ($p > 0.05$) between the two seasons. The findings of this study could also not be of the same suggestion that temperature may be the cause of these variations as reported by Orren *et al.*, (1980). Only pH changes had recorded and showed significant mean variations in seawater ($p < 0.05$) between the two seasons but this study did not evidence this finding as the reason for the seasonal variations in metal accumulation by *Choromytilus meridionalis* species along the Central Namibian marine coastline. Thus, trace metal levels of the mussel *Choromytilus meridionalis* showed significant variations between the seasons and stations overall ($p < 0.05$). Based on this study therefore, perhaps these variations may be related to pH of the water (though not evidenced) during sampling period which was

significantly different between the two seasons as mentioned earlier. In addition, the presence of higher trace metals Pb and Zn in mussel samples simply indicate that the movement of these metals is not localised but rather spread out and depends on various other factors such as wind action and currents (Sulochanan *et al*., 2007) like the Benguela upwelling in the Namibian coastline.

In comparison with *Choromytilus meridionalis* and water samples across the stations, this study could therefore report that mean values of trace metals Pb, Cu, Zn and Fe were higher in sediment samples followed by mussels with the least values recorded in water samples. This substantiates some earlier findings (Giarratano and Amin, 2010) that trace metals in sediment samples are many times more than the same metals in the water column because sediments act as scavenger agents for trace metals and an adsorptive sink in aquatic ecosystems. Furthermore, no significant correlations of trace metals in black mussels were recorded between winter and summer seasons during this investigation as compared to other studies that reported some relationships such as Pb-Cu and Pb-Fe (Cardellicchio *et al*., 2008). Perhaps some further studies would prove vital to establish why significant trace metal correlations were observed in sediments but not ($p > 0.05$) in black mussels. However, correlation observations were noted between trace metal levels accumulated in *Choromytilus meridionalis* samples, water and sediments samples overall. This proves some claims reported elsewhere that some trace elements of these metals either stay in the water column or absorbed by mussels before settling and accumulating at the bottom in sediments.

Overall, this research study has established that the Central Namibian marine coastline is relatively less polluted compared to other coastlines and coastal waters around the world. The study has also noted some significant increases in metal accumulation in the water column,

mussel and sediment samples across the stations within the two seasons. This finding should raise some concerns to all so that precaution measures can be put in place to control the situation, monitor and prevent it from reaching a critical point as the clean up is usually costly. Once measures are put into place then it will also monitor the levels of these metals so that they do not exceed their permissible limits for human consumption and ensure the Namibian coastline is environmentally pollution free. Further, this study has also found out that Walvis Bay station is far more polluted mainly due to the activities that take place in the area. The least polluted station in this study is Cape Cross mainly because this part of the coastline does not have much activities taking place as compared to Henties Bay, Swakopmund and Walvis Bay stations. In addition, this investigation has found that Fe is the most abundant trace metal in the Central Namibian marine coastline followed by Zn, Cu and the least abundant trace metal being Pb. In comparisons with the three variables under investigation along the Central Namibian marine coastline, this study has established that trace metal accumulations were higher in sediments, *Choromytilus meridionalis* and seawater respectively.

Furthermore, findings of this research investigation have proved variations between winter and summer seasons in terms of trace metal accumulations by *Choromytilus meridionalis* species, sediments and seawater. Not all metals had shown significant positive trace metal accumulation in summer season from winter as reported by some literature because this study has evidenced some negative relationships for some trace metals such as Zn in sediment samples. However, it could be true that metal accumulation increases from winter to summer period given the claims about reproduction in these mussels though this study could not establish this claim. This study could also not reveal why Zn had shown a negative correlation in terms of accumulation by sediments along the Central Namibian coastline.

Mean trace metal values recorded in the Namibian *Choromytilus meridionalis*, water and sediment samples have all been analysed and compared thoroughly. Thus, it can be conclusively stated that this study has ultimately provided very significant data which will act as a benchmark for further investigations in this field to monitor the Namibian marine coastline pollution status. The data and information in this report will most probably also help the country to set possible measures that will regulate some foodstuff limits in terms of trace metal compositions where needed for Namibia. Finally, Pb levels have been compared with the permissible limit of 1 ppm set by the European Community Commission Regulation (EC) No. 466/2001 and regarded safe for human consumption; and the values were found to be slightly lower than this standard though this metal increase should set an alert for monitoring.

CHAPTER SIX

6. RECOMMENDATIONS

Trace metals are not only limited to the marine ecosystem but rivers, lakes, estuaries and in foodstuffs as well. There is therefore a strong need to conduct and have continued intense ecotoxicological studies in Namibian waters either freshwater or marine and foodstuffs in order to monitor the levels of these metals accumulated by organisms, water and sediments as well as foodstuffs. This will ensure that citizens consume foodstuffs that have metal levels within the permissible limits without affecting their health. It will also simplify and aid in the determination of the pollution status of various ecosystems so that prevention and mitigation measures are setup beforehand. This study being the baseline and first of its kind in the Central Namibian marine coastline, it is evident that data set obtained once off and in only a one year period is not sufficient enough to construct a reliable trend of trace metal levels especially where comparisons and monitoring of an ecosystem metal levels is concerned. Besides, the Namibian coastline is vast and there is a need to intensify this study such as at Luderitz area, another harbour and industrial town for Namibia. In addition, some studies should be undertaken (such as isotope studies) that will involve observing and identification of point sources of pollution. Thus, for monitoring and comparison purposes in which mussel samples are used as bio-indicator organisms, there should be a unique and precise pattern adopted during the species sampling period.

Specifically, mussel samples should at least be of the same size limits such as age and length; and be collected at the same depth where possible. The samples should also be collected in the same period or at the end of the reproductive cycle (winter end), when the metal levels have been

reported to be the highest. Perhaps by following these recommendations it might be possible to obtain, in different locations, uniform data which can easily be compared to each other.

At the minimum however, the following general recommendations have been drawn based on experiences and challenges encountered during this study investigation;

There is need for continued research on trace metals along the Namibian marine coastline which will act as a monitoring aspect for country and will also ensure that data is readily available for any related projects. Perhaps the Government of Namibia should develop this monitoring programme that will aid in monitoring of trace metals in various ecosystems of ecological and economical importance. This should as well include assessment of trace metal levels found in foodstuffs within and those brought into the country for human consumption.

There is also a need to propose policy reforms including re-organising policy institutions and structures to enable the creation of a policy environment that is adaptive and responsive to the ever changing developmental challenges in Namibia. This is in a case whereby the findings of this research study are to be used elsewhere as is the purpose of many research studies.

7. CONCLUSION

Metal values recorded in *Choromytilus meridionalis* species and sediments from sampling stations in this study ranked in the following order of accumulation from highest to lowest: Fe > Zn > Cu > Pb. The levels of all metals recorded in all three different samples across the stations were well below or within those reported elsewhere in literature in general; indicating that the Namibian coastline is not polluted. However, compared to each of the stations studied, Walvis Bay station recorded more metal accumulation in all samples ($P < 0.05$). The findings of this

study thus infer that Walvis Bay station is more polluted compared to Swakopmund, Henties Bay and Cape Cross stations. The least polluted station is Cape Cross while Henties Bay holds the most Fe content while Cu levels in seawater in Walvis Bay station exceed permissible levels.

In terms of mariculture, the mussels grown in Walvis Bay station experience diverse threats and stress from physical and biochemical factors as evidenced in this study which lead to the accumulation of the metals in their body and shells. Thus, though applicable to all stations, pollution status in Walvis Bay station is quite alarming and increased metal accumulation by mussels and sediments in this locality could pose health dangers to consumers and the public at large eventually. Additionally, since these mussels are usually exported to generate income for the companies involved in this business and the country in general, necessary steps should be put in place and implemented to avoid losing the economic value of these species in cases where the limits reach unacceptable and permissible limits or standards as this might also cause serious diseases. It is evident though that this present study has found higher levels of trace metals in sediments than in mussels and water across the stations.

The findings are indicative of trace metal pollution from various sources but most probably from activities in Walvis Bay harbour such as ship repairs, industrial, urban and residential sources, tourism and recreation including run-off water from various catchments around the sampling stations. Thus, sediments exhibited significantly higher mean trace metal levels than mussels and water ($P < 0.05$). In addition, mean trace metal values in mussel samples showed no correlation while a clear-cut significant correlation was exhibited in sediment samples. For water samples, trace metals had also evidenced very good correlation in terms of accumulation with an exception of Pb and Fe.

Broadly, trace metal values between winter and summer season were highly variable across the stations in this study. Only Cu and Pb mean values in mussels showed significant differences ($P < 0.05$) while Zn and Fe mean values were significant ($P < 0.05$) in sediments between the seasons than water samples in which all were significant ($P < 0.05$). The mean value level of Pb in sampled mussels is within permissible limit of 1 ppm set by the European Community Commission Regulation (EC) No. 466/2001 for human consumption though an increased metal accumulation in the farmed stations could pose a threat to consumers and the public as stated earlier. Thus, based on the fact these mussels are safe for consumption presently and that relatively high levels of metals in sediment might vary their availability in cases where physical factors change; there is a need for continued monitoring of these metals to avoid human risks in consuming contaminated mussel products as stated in the recommendations above.

Finally, *Choromytilus meridionalis*, sediments and seawater samples collected from the Central Namibian marine coastline are within the range of the mean values that have been reported in the prevailing literature elsewhere. The study has also supplied the most valuable data and information pertaining to the trace metal levels in the mussel *Choromytilus meridionalis*, sediments and seawater along the Central Namibian marine coastline. These results will most definitely be used to estimate and monitor the quality of mussels farmed in the area as a whole; and to evaluate possible risks associated with their consumption. In addition, the data has also provided a benchmark for further investigations in the field including the monitoring programme to assess the state of pollution in this part of the ocean if this work is going to continue. Hence, this study has served as baseline assessment of the pollution status of the Central Namibian coastline waters.

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APPENDIX 1

The following section shows the table for mean values of water parameters and ANOVA tables of mean trace metal values in the species under investigation.

Appendix 1.1: Mean values and standard errors (± SE) for temperature, DO and pH of seawater in stations

Sample ID	Station	Temp (^{O}C)		DO (ppm)		pH readings	
		Winter	**Summer**	**Winter**	**Summer**	**Winter**	**Summer**
1	Walvis Bay	16.10	18.90	3.95	8.37	8.09	7.87
2	Walvis Bay	16.20	18.70	4.50	9.91	8.04	7.96
3	Walvis Bay	16.00	19.00	4.10	7.49	8.05	7.93
	Mean	**16.10**	**18.87**	**4.18**	**8.59**		
4	Swakopmund	16.8	16.8	7.83	8.22	8.77	8.59
5	Swakopmund	17.2	16.9	8.4	11.93	8.4	8.61
6	Swakopmund	17.4	17	6.85	7.63	8.84	8.57
	Mean	**17.13**	**16.90**	**7.69**	**9.26**		
7	Henties Bay	14.4	14.7	6.11	4.73	7.94	7.91
8	Henties Bay	14.4	16.2	6.11	4.74	7.95	4.73
9	Henties Bay	14.5	14.1	6.12	4.71	7.94	4.76
	Mean	**14.43**	**15.00**	**6.11**	**4.73**		
10	Cape Cross	15.80	15.70	7.71	4.72	7.96	4.70
11	Cape Cross	15.60	14.70	7.22	4.53	8	4.70
12	Cape Cross	15.50	15	7.52	4.93	10	4.60
	Mean	**15.7**	**15.1**	**7.48**	**4.73**	**-**	**-**
	SE ($\bar{x}$)	**0.30**	**0.46**	**0.44**	**0.72**	**-**	**-**
	S.e.d.	**0.57**		**0.59**			

Note: S.e.d is the standard error of the difference between the two means for winter and summer except for pH which has been presented as a range in the report.

The following section includes all observations and raw data for this study investigation.

Appendix 1.2: Water parameters

A. Temperature Readings of Seawater (^{o}C)

Sample ID	Station	Winter	Summer
1	Walvis Bay	16.1	18.9
2	Walvis Bay	16.2	18.7
3	Walvis Bay	16	19
4	Swakopmund	16.8	16.8
5	Swakopmund	17.2	16.9
6	Swakopmund	17.4	17
7	Henties Bay	14.4	14.7
8	Henties Bay	14.4	16.2
9	Henties Bay	14.5	14.1
10	Cape Cross	15.8	15.8
11	Cape Cross	15.60	15.60
12	Cape Cross	15.50	15.50

B. Dissolved Oxygen Readings (ppm)

Sample ID	Station	Winter	Summer
1	Walvis Bay	3.95	8.37
2	Walvis Bay	4.5	9.91
3	Walvis Bay	4.1	7.49
4	Swakopmund	7.83	8.22
5	Swakopmund	8.4	11.93
6	Swakopmund	6.85	7.63
7	Henties Bay	6.11	4.73
8	Henties Bay	6.11	4.74
9	Henties Bay	6.12	4.71
10	Cape Cross	7.7	4.72
11	Cape Cross	7.22	4.53
12	Cape Cross	7.52	4.93

C. pH reading levels in Seawater

Sample ID	Station	Winter	Summer
1	Walvis Bay	8.09	7.87
2	Walvis Bay	8.04	7.96
3	Walvis Bay	8.05	7.93
4	Swakopmund	8.77	8.59
5	Swakopmund	8.4	8.61
6	Swakopmund	8.84	8.57
7	Henties Bay	7.94	7.91
8	Henties Bay	7.95	4.73
9	Henties Bay	7.94	4.76
10	Cape Cross	7.96	4.7
11	Cape Cross	8	4.7
12	Cape Cross	10	4.6

APPENDIX 2

Appendix 2.1: ANOVA table for Pb across the stations using a 4X2X3 factorial design

Source of variation	**d.f.**	**s.s.**	**m.s.**	**v.r.**	**F pr.**
Station	3	10622.11	3540.7	226.49	<.001
Substrate	2	8793.65	4396.82	281.25	<.001
Season	1	18.54	18.54	1.19	0.277
Station.Season	3	112.53	37.51	2.4	0.068
Station.Substrate	6	21077.25	3512.88	224.71	<.001
Substrate.Season	2	44.36	22.18	1.42	0.244
Station.Substrate.Season	6	243.04	40.51	2.59	0.019
Residual	264	4127.18	15.63		
Total	287	45038.65			

Appendix 2.2: ANOVA table for Cu across the stations using a 4X2X3 factorial design

Source of variation	**d.f.**	**s.s.**	**m.s.**	**v.r.**	**F pr.**
Station	3	6506.346	2168.782	234.88	<.001
Substrate	2	9292.885	4646.442	503.21	<.001
Season	1	4.122	4.122	0.45	0.505
Station.Season	3	6.78	2.26	0.24	0.865
Station.Substrate	6	11336.66	1889.443	204.63	<.001
Substrate.Season	2	8.974	4.487	0.49	0.616
Station.Substrate.Season	6	34.202	5.7	0.62	0.716
Residual	264	2437.662	9.234		
Total	287	29627.63			

Appendix 2.3: ANOVA table for Zn across the stations using a 4X2X3 factorial design

Source of variation	**d.f.**	**s.s.**	**m.s.**	**v.r.**	**F pr.**
Station	3	11508.8	3836.3	34.84	<.001
Substrate	2	48940.4	24470.2	222.26	<.001
Season	1	6406.4	6406.4	58.19	<.001
Station.Season	3	490.9	163.6	1.49	0.219
Station.Substrate	6	19942.4	3323.7	30.19	<.001
Substrate.Season	2	8351.4	4175.7	37.93	<.001
Station.Substrate.Season	6	1080.7	180.1	1.64	0.137
Residual	264	29066.1	110.1		
Total	287	125787.1			

Appendix 2.4: ANOVA table for Fe across the stations using a 4X2X3 factorial design

Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.
Station	3	2.16E+09	7.20E+08	82.77	<.001
Substrate	2	9.69E+09	4.85E+09	557.25	<.001
Season	1	1.41E+08	1.41E+08	16.18	<.001
Station.Season	3	2.56E+08	8.54E+07	9.82	<.001
Station.Substrate	6	4.27E+09	7.12E+08	81.84	<.001
Substrate.Season	2	2.77E+08	1.38E+08	15.9	<.001
Station.Substrate.Season	6	5.26E+08	8.76E+07	10.08	<.001
Residual	264	2.30E+09	8.69E+06		
Total	287	1.96E+10			

Appendix 2.5: Descriptive statistics for lead (Pb)

Variable	Station	Substrate	Season	Mean	Std. Error	Std. Deviation	*n* (sample number)
Pb	Walvis Bay	Water	Winter	0	0	0	12
			Summer	0.01	0.01	0.02	12
			Total	0.01	0.01	0.01	24
		Mussels	Winter	0	0	0	12
			Summer	0.25	0.08	0.27	12
			Total	0.12	0.08	0.23	24
		Sediment	Winter	47.31	2.63	9.13	12
			Summer	39.06	4.89	16.95	12
			Total	43.19	7.52	13.96	24
		Total	Winter	15.77		23.19	36
			Summer	13.11		20.94	36
			Total	14.44		21.96	72
	Swakopmund	Water	Winter	0	0	0	12
			Summer	0	0	0	12
			Total	0	0	0	24
		Mussels	Winter	0	0	0	12
			Summer	0.12	0.12	0.40	12
			Total	0.06	0.12	0.29	24
		Sediment	Winter	1.07	0.26	0.91	12
			Summer	1.39	0.13	0.45	12
			Total	1.23	0.39	0.72	24
		Total	Winter	0.36		0.72	36
			Summer	0.5		0.72	36

Variable	Station	Substrate	Season	Mean	Std. Error	Std. Deviation	*n* (sample number)
			Total	0.43		0.72	72
	Henties Bay	Water	Winter	0	0	0	12
			Summer	0	0	0	12
			Total	0		0	24
		Mussels	Winter	0	0	0	12
			Summer	0	0	0	12
			Total	0		0	24
		Sediment	Winter	1.83	0.5	1.73	12
			Summer	3.02	0.15	0.51	12
			Total	2.42		1.39	24
		Total	Winter	0.61		1.31	36
			Summer	1.01		1.47	36
			Total	0.81		1.40	72
	Cape Cross	Water	Winter	0	0	0	12
			Summer	0	0	0	12
			Total	0		0	24
		Mussels	Winter	0	0	0	12
			Summer	0	0	0	12
			Total	0		0	24
		Sediment	Winter	0	0	0	12
			Summer	0.28	0.08	0.28	12
			Total	0.14		0.24	24
		Total	Winter	0		0	36
			Summer	0.09		0.21	36
			Total	0.05		0.15	72
	Total	Water	Winter	0	0	0	48
			Summer	0	0.01	0.01	48
			Total	0		0.01	96
		Mussels	Winter	0	0	0	48
			Summer	0.09	0.24	0.26	48
			Total	0.05		0.19	96
		Sediment	Winter	12.55	4.67	20.79	48
			Summer	10.94	8.48	18.38	48
			Total	11.75		19.53	96
		Total	Winter	4.18		13.32	144
			Summer	3.68		11.73	144
			Total	3.93		12.53	288

Appendix 2.5.1: Multiple comparisons of means for Pb using the least significant difference (LSD) between stations

Dependent Variable			Mean Difference	Sig.
Pb	Walvis Bay	Swakopmund	14.01*	.000
		Henties Bay	13.63*	.000
		Cape Cross	14.39*	.000
	Swakopmund	Walvis Bay	14.01*	.000
		Henties Bay	.38	.568
		Cape Cross	.38	.560
	Henties Bay	Walvis Bay	13.63*	.000
		Swakopmund	.38	.568
		Cape Cross	.76	.249
	Cape Cross	Walvis Bay	14.39*	.000
		Swakopmund	.38	.560
		Henties Bay	.76	.249

* Significant difference at 0.05 probability level

Appendix 2.5.2: Multiple comparisons of means for Pb using the least significant difference (LSD) between substrates

Dependent Variable			Mean Difference	Sig.
Pb	Water	Black Mussels	4	.94
		Sediments	11.74*	.00
	Black Mussels	Water	0.04	.94
		Sediments	11.70*	.000
	Sediments	Water	11.74*	.000
		Black Mussels	11.70*	.000

* Significant difference at 0.05 probability level

Appendix 2.6: Descriptive statistics for copper (Cu)

Variable	Station	Substrate	Season	Mean	Std. Error	Std. Deviation	*n* (sample number)
Cu	Walvis Bay	Water	Winter	0	0	0.00	12
			Summer	0.09	0.006	0.02	12
			Total	0.05		0.05	24
		Mussels	Winter	2.16	0.19	0.65	12
			Summer	3.36	0.33	1.15	12
			Total	2.76		1.10	24
		Sediment	Winter	37.16	2.03	7.03	12
			Summer	35.16	2.64	9.13	12
			Total	36.16		8.03	24
		Total	Winter	13.11		17.72	36
			Summer	12.87		16.85	36
			Total	12.99		17.17	72
	Swakopmund	Water	Winter	0	0	0.00	12
			Summer	0	0	0.00	12
			Total	0		0.00	24
		Mussels	Winter	0.73	0.14	0.50	12
			Summer	1.06	0.26	0.89	12
			Total	0.9		0.73	24
		Sediment	Winter	5.2	0.46	1.58	12
			Summer	6.73	2.28	7.89	12
			Total	5.96		5.62	24
		Total	Winter	1.98		2.51	36
			Summer	2.59		5.36	36
			Total	2.29		4.17	72
	Henties Bay	Water	Winter	0	0	0.00	12
			Summer	0	0	0.00	12
			Total	0		0.00	24
		Mussels	Winter	0.61	0.12	0.42	12
			Summer	1.48	0.12	0.42	12
			Total	1.04		0.60	24
		Sediment	Winter	6.76	1.25	4.33	12
			Summer	6.9	0.24	0.83	12
			Total	6.83		3.05	24
		Total	Winter	2.46		3.94	36
			Summer	2.79		3.05	36
			Total	2.63		3.51	72

Variable	Station	Substrate	Season	Mean	Std. Error	Std. Deviation	*n* (sample number)
	Cape Cross	Water	Winter	0	0	0.00	12
			Summer	0	0	0.00	12
			Total	0		0.00	24
		Mussels	Winter	1.46	0.16	0.54	12
			Summer	2.01	0.2	0.68	12
			Total	1.73		0.67	24
		Sediment	Winter	2.08	0.21	0.73	12
			Summer	2.25	0.15	0.51	12
			Total	2.16		0.62	24
		Total	Winter	1.18		1.02	36
			Summer	1.42		1.13	36
			Total	1.3		1.08	72
	Total	Water	Winter	0	0	0.00	48
			Summer	0.02	0.01	0.04	48
			Total	0.01		0.03	96
		Mussels	Winter	1.24		0.81	48
			Summer	1.98		1.19	48
			Total	1.61		1.08	96
		Sediment	Winter	12.8	4.22	14.89	48
			Summer	12.76	6.05	14.45	48
			Total	12.78		14.59	96
		Total	Winter	4.68		10.32	144
			Summer	4.92		10.03	144
			Total	4.8		10.16	288

Appendix 2.6.1: Multiple comparisons of means for Cu using LSD between stations

Dependent Variable			Mean Difference	Sig.
Cu	Walvis Bay	Swakopmund	10.70*	.000
		Henties Bay	10.36*	.000
		Cape Cross	11.69*	.000
	Swakopmund	Walvis Bay	10.70*	.000
		Henties Bay	.34	.503
		Cape Cross	.99	.052
	Henties Bay	Walvis Bay	10.36*	.000
		Swakopmund	.34	.503

Dependent Variable			Mean Difference	Sig.
		Cape Cross	1.33*	.009
	Cape Cross	Walvis Bay	11.69*	.000
		Swakopmund	.99	.052
		Henties Bay	1.33*	.009

* Significant difference at 0.05 probability level

Appendix 2.6.2: Multiple comparisons of means for Cu using LSD between substrates

Dependent Variable			Mean Difference	Sig.
Cu	Water	Black Mussels	1.60*	.000
		Sediments	12.77*	.000
	Black Mussels	Water	1.60*	.000
		Sediments	11.17*	.000
	Sediments	Water	12.77*	.000
		Black Mussels	11.17*	.000

* Significant difference at 0.05 probability level

Appendix 2.7: Descriptive statistics for zinc

Variable	Station	Substrate	Season	Mean	Std. Error	Std. Deviation	*n* (sample number)
Zn	Walvis Bay	Water	Winter	0	0	0.00	12
			Summer	0.04	0.003	0.01	12
			Total	0.02		0.02	24
		Mussels	Winter	13.57	1.59	5.49	12
			Summer	18.33	1.54	5.35	12
			Total	15.95		5.83	24
		Sediment	Winter	75.53		12.67	12
			Summer	47.27		11.64	12
			Total	61.4		18.71	24
		Total	Winter	29.7		34.23	36
			Summer	21.88		20.99	36
			Total	25.79		28.46	72

Variable	Station	Substrate	Season	Mean	Std. Error	Std. Deviation	*n* (sample number)
	Swakopmund	Water	Winter	0	0	0.00	12
			Summer	0	0	0.00	12
			Total	0		0.00	24
		Mussels	Winter	16.77	2.51	8.71	12
			Summer	15.05	2	6.93	12
			Total	15.91		7.75	24
		Sediment	Winter	26.22		19.86	12
			Summer	9.67		2.17	12
			Total	17.95		16.20	24
		Total	Winter	14.33		16.40	36
			Summer	8.24		7.51	36
			Total	11.29		13.03	72
	Henties Bay	Water	Winter	0	0	0.00	12
			Summer	0	0	0.00	12
			Total	0		0.00	24
		Mussels	Winter	27.44	6.31	21.87	12
			Summer	14.13	0.94	3.25	12
			Total	20.78		16.73	24
		Sediment	Winter	42.86		24.16	12
			Summer	18.07		2.64	12
			Total	30.47		21.04	24
		Total	Winter	23.43		25.63	36
			Summer	10.73		8.21	36
			Total	17.08		19.95	72
	Cape Cross	Water	Winter	0	0	0.00	12
			Summer	0	0	0.00	12
			Total	0		0.00	24
		Mussels	Winter	13.42	1.95	6.77	12
			Summer	8.48	1.07	3.71	12
			Total	10.95		5.90	24
		Sediment	Winter	32.14		24.92	12
			Summer	3.72		2.03	12
			Total	17.93		22.58	24
		Total	Winter	15.19		19.71	36
			Summer	4.07		4.24	36
			Total	9.63		15.22	72
	Total	Water	Winter	0	0	0.00	48

Variable	Station	Substrate	Season	Mean	Std. Error	Std. Deviation	*n* (sample number)
			Summer	0.01	0.004	0.02	48
			Total	0		0.01	96
		Mussels	Winter	17.8	12.55	13.45	48
			Summer	14	5.02	6.04	48
			Total	15.9		10.55	96
		Sediment	Winter	44.19		27.97	48
			Summer	19.68		17.92	48
			Total	31.94		26.41	96
		Total	Winter	20.66		25.47	144
			Summer	11.23		13.65	144
			Total	15.95		20.94	288

Appendix 2.7.1: Multiple comparisons of means for Zn using LSD between stations

Dependent Variable			Mean Difference	Sig.
Zn	Walvis Bay	Swakopmund	14.50*	.00
		Henties Bay	8.71*	.00
		Cape Cross	16.16*	.00
	Swakopmund	Walvis Bay	14.50*	.00
		Henties Bay	5.80*	.00
		Cape Cross	1.66	.34
	Henties Bay	Walvis Bay	8.71*	.00
		Swakopmund	5.80*	.00
		Cape Cross	7.46*	.00
	Cape Cross	Walvis Bay	16.16*	.00
		Swakopmund	1.66	.34

* Significant difference at 0.05 probability level

Appendix 2.7.2: Multiple comparisons of means for Zn using LSD between substrates

Dependent Variable			Mean Difference	Sig.
	Water	Black Mussels	15.90*	.00
		Sediments	31.93*	.00

Dependent Variable			Mean Difference	Sig.
Zn	Black Mussels	Water	15.90*	.00
		Sediments	16.04*	.000
	Sediments	Water	31.93*	.00
		Black Mussels	16.04*	.00

* Significant difference at 0.05 probability level

Appendix 2.8: Descriptive statistics for iron (Fe)

Variable	Station	Substrate	Season	Mean	Std. Error	Std. Deviation	*n* (sample number)
Fe	Walvis Bay	Water	Winter	0	0.00	0.00	12
			Summer	0.16	0.05	0.17	12
			Total	0.08		0.14	24
		Mussels	Winter	10.41	1.21	4.18	12
			Summer	114.9	22.73	78.76	12
			Total	62.66		76.31	24
		Sediment	Winter	11055.5	308.08	1067.21	12
			Summer	7512.68	379.22	1313.66	12
			Total	9284.09		2155.08	24
		Total	Winter	3688.64		5316.82	36
			Summer	2542.58		3640.11	36
			Total	3115.61		4560.71	72
	Swakopmund	Water	Winter	0	0.00	0.00	12
			Summer	0	0.00	0.00	12
			Total	0		0.00	24
		Mussels	Winter	23.19	4.42	15.30	12
			Summer	29.39	5.62	19.48	12
			Total	26.29		17.42	24
		Sediment	Winter	6710.68	564.88	1956.80	12
			Summer	14304.29	728.29	2522.86	12
			Total	10507.48		4462.94	24
		Total	Winter	2244.62		3385.46	36
			Summer	4777.89		6976.62	36
			Total	3511.26		5592.03	72

Variable	Station	Substrate	Season	Mean	Std. Error	Std. Deviation	*n* (sample number)
	Henties Bay	Water	Winter	0	0.00	0.00	12
			Summer	0	0.00	0.00	12
			Total	0		0.00	24
		Mussels	Winter	139.28	46.90	162.46	12
			Summer	103.02	34.59	119.84	12
			Total	121.15		140.84	24
		Sediment	Winter	20152.03	3843.30	13313.58	12
			Summer	31459.42	1099.70	3809.48	12
			Total	25805.73		11183.34	24
		Total	Winter	6763.77		12161.52	36
			Summer	10520.81		15167.18	36
			Total	8642.29		13780.05	72
	Cape Cross	Water	Winter	0	0.00	0.00	12
			Summer	0	0.00	0.00	12
			Total	0		0.00	24
		Mussels	Winter	33.07	4.22	14.63	12
			Summer	56.06	21.68	75.11	12
			Total	44.56		54.21	24
		Sediment	Winter	3088.08	316.50	1096.38	12
			Summer	4408.07	464.21	1608.08	12
			Total	3748.07		1505.38	24
		Total	Winter	1040.38		1592.00	36
			Summer	1488.04		2280.38	36
			Total	1264.21		1965.61	72
	Total	Water	Winter	0	0.00	0.00	48
			Summer	0.04	0.08	0.11	48
			Total	0.02		0.08	96
		Mussels	Winter	51.49		94.74	48
			Summer	75.84		86.32	48
			Total	63.67		90.98	96
		Sediment	Winter	10251.57	6771.70	9188.18	48
			Summer	14421.12	2509.40	10853.71	48
			Total	12336.34		10219.61	96
		Total	Winter	3434.35		7151.95	144
			Summer	4832.33		9220.40	144
			Total	4133.34		8266.57	288

Appendix 2.8.1: Multiple comparisons of means for Fe using LSD between stations

Dependent Variable			**Mean Difference**	**Sig.**
Fe	Walvis Bay	Swakopmund	395.65	.42
		Henties Bay	5526.68*	.00
		Cape Cross	1851.40*	.00
	Swakopmund	Walvis Bay	395.65	.42
		Henties Bay	5131.03*	.00
		Cape Cross	2247.05*	.00
	Henties Bay	Walvis Bay	5526.68*	.00
		Swakopmund	5131.03*	.00
		Cape Cross	7378.08*	.00
	Cape Cross	Walvis Bay	1851.40*	.00
		Swakopmund	2247.05*	.00
		Henties Bay	7378.08*	.00

* Significant difference at 0.05 probability level

Appendix 2.8.2: Multiple comparisons of means for Fe using LSD between substrates

Dependent variable			**Mean Difference**	**Sig.**
	Water	Black Mussels	63.64	0.88
		Sediments	12336.32*	0
Fe	Black Mussels	Water	63.64	0.88
		Sediments	12272.68*	0
	Sediments	Water	12336.32*	0
		Black Mussels	12272.68*	0

APPENDIX 3

Appendix 3.1: Trace metal readings (ppm) in winter and summer for water samples in stations.

Sample ID	Station	Winter Cu	Summer Cu	Winter Zn	Summer Zn	Winter Fe	Summer Fe	Winter Pb	Summer Pb
1	Walvis Bay	0	0.08	0	0.05	0	0.06	0	0
2	Walvis Bay	0	0.07	0	0.02	0	0.05	0	0
3	Walvis Bay	0	0.10	0	0.04	0	0.57	0	0.01
4	Walvis Bay	0	0.12	0	0.05	0	0.46	0	0.01
5	Walvis Bay	0	0.09	0	0.03	0	0.05	0	0
6	Walvis Bay	0	0.08	0	0.04	0	0.10	0	0
7	Walvis Bay	0	0.09	0	0.04	0	0.15	0	0.01
8	Walvis Bay	0	0.09	0	0.04	0	0.12	0	0.01
9	Walvis Bay	0	0.08	0	0.03	0	0.08	0	0.01
10	Walvis Bay	0	0.09	0	0.05	0	0.10	0	0.06
11	Walvis Bay	0	0.14	0	0.04	0	0.14	0	0.01
12	Walvis Bay	0	0.08	0	0.03	0	0.09	0	0.01
13	Swakopmund	0	0	0	0	0	0	0	0
14	Swakopmund	0	0	0	0	0	0	0	0
15	Swakopmund	0	0	0	0	0	0	0	0
16	Swakopmund	0	0	0	0	0	0	0	0
17	Swakopmund	0	0	0	0	0	0	0	0
18	Swakopmund	0	0	0	0	0	0	0	0
19	Swakopmund	0	0	0	0	0	0	0	0
20	Swakopmund	0	0	0	0	0	0	0	0
21	Swakopmund	0	0	0	0	0	0	0	0
22	Swakopmund	0	0	0	0	0	0	0	0
23	Swakopmund	0	0	0	0	0	0	0	0
24	Swakopmund	0	0	0	0	0	0	0	0

Sample ID	Station	Winter Cu	Summer Cu	Winter Zn	Summer Zn	Winter Fe	Summer Fe	Winter Pb	Summer Pb
25	Henties Bay	0	0	0	0	0	0	0	0
26	Henties Bay	0	0	0	0	0	0	0	0
27	Henties Bay	0	0	0	0	0	0	0	0
28	Henties Bay	0	0	0	0	0	0	0	0
29	Henties Bay	0	0	0	0	0	0	0	0
30	Henties Bay	0	0	0	0	0	0	0	0
31	Henties Bay	0	0	0	0	0	0	0	0
32	Henties Bay	0	0	0	0	0	0	0	0
33	Henties Bay	0	0	0	0	0	0	0	0
34	Henties Bay	0	0	0	0	0	0	0	0
35	Henties Bay	0	0	0	0	0	0	0	0
36	Henties Bay	0	0	0	0	0	0	0	0
37	Cape Cross	0	0	0	0	0	0	0	0
38	Cape Cross	0	0	0	0	0	0	0	0
39	Cape Cross	0	0	0	0	0	0	0	0
40	Cape Cross	0	0	0	0	0	0	0	0
41	Cape Cross	0	0	0	0	0	0	0	0
42	Cape Cross	0	0	0	0	0	0	0	0
43	Cape Cross	0	0	0	0	0	0	0	0
44	Cape Cross	0	0	0	0	0	0	0	0
45	Cape Cross	0	0	0	0	0	0	0	0
46	Cape Cross	0	0	0	0	0	0	0	0
47	Cape Cross	0	0	0	0	0	0	0	0
48	Cape Cross	0	0	0	0	0	0	0	0

Appendix 3.2: Trace metal readings (ppm) in winter and summer for *Choromytilus meridionalis* samples in stations**.**

Sample ID	Station	Pb Winter	Pb Summer	Fe Winter	Fe Summer	Cu Winter	Cu Summer	Zn Winter	Zn Summer
1	Walvis Bay	0	0.3	16	141	2.7	3.4	13	15
2	Walvis Bay	0	0	7.3	59	1.5	3.1	8.7	14
3	Walvis Bay	0	0	12	74	1.7	2.7	11	19
4	Walvis Bay	0	0.3	12	57	1.7	3.7	28	16
5	Walvis Bay	0	0.7	7.6	201	1.7	2.3	11	13
6	Walvis Bay	0	0	7.8	55	2.0	3.0	12	14
7	Walvis Bay	0	0.4	4.8	179	2.2	2.7	11	21
8	Walvis Bay	0	0	7.5	47	1.8	2.3	11	20
9	Walvis Bay	0	0	20	30	3.9	2.7	13	14
10	Walvis Bay	0	0.3	12	234	2.5	6.5	11	20
11	Walvis Bay	0	0.7	9.3	239	1.9	3.9	11	21
12	Walvis Bay	0	0.2	8.7	63	2.3	3.9	21	33
13	Swakopmund	0	1.4	24	50	1.3	0.5	16	7.7
14	Swakopmund	0	0.0	24	23	0.4	1.3	25	16
15	Swakopmund	0	0.0	26	24	0.9	0.8	8.7	14
16	Swakopmund	0	0.0	8.7	82	0.0	0.2	6.5	14
17	Swakopmund	0	0.0	16	21	0.6	0.3	15	12
18	Swakopmund	0	0.0	15	26	0.4	0.3	25	10
19	Swakopmund	0	0.0	19	26	1.1	2.3	12	27
20	Swakopmund	0	0.0	52	12	0.0	0.2	11	10
21	Swakopmund	0	0.0	55	16	0.9	0.7	34	11
22	Swakopmund	0	0.0	12	29	0.5	3.0	26	26
23	Swakopmund	0	0.0	19	32	1.0	1.1	14	25
24	Swakopmund	0	0.0	8.1	12	1.6	1.7	8.4	9.5

Sample ID	Station	Pb Winter	Pb Summer	Fe Winter	Fe Summer	Cu Winter	Cu Summer	Zn Winter	Zn Summer
25	Henties Bay	0	0.0	90	40	0.5	1.3	19	11
26	Henties Bay	0	0.0	45	36	0.5	1.7	33	15
27	Henties Bay	0	0.0	103	205	1.2	1.6	79	14
28	Henties Bay	0	0.0	484	23	0.9	0.9	11	20
29	Henties Bay	0	0.0	56	23	0.3	1.7	57	16
30	Henties Bay	0	0.0	483	20	0.2	1.9	15	17
31	Henties Bay	0	0.0	35	388	0.2	2.0	16	14
32	Henties Bay	0	0.0	54	269	0.4	1.0	12	12
33	Henties Bay	0	0.0	106	41	0.5	1.6	17	11
34	Henties Bay	0	0.0	48	101	0.8	1.8	4.1	9.4
35	Henties Bay	0	0.0	76	49	0.1	1.4	39	12
36	Henties Bay	0	0.0	91	42	1.5	0.7	25	18
37	Cape Cross	0	0.0	35	69	1.7	1.3	7.4	2.8
38	Cape Cross	0	0.0	49	20	1.3	1.4	26	5.8
39	Cape Cross	0	0.0	18	18	0.6	2.9	6.6	13
40	Cape Cross	0	0.0	55	21	2.1	2.0	11	5.7
41	Cape Cross	0	0.0	26	21	0.6	2.9	6.9	14
42	Cape Cross	0	0.0	22	32	1.1	2.4	7.2	12
43	Cape Cross	0	0.0	39	29	1.6	1.4	13	13
44	Cape Cross	0	0.0	27	25	2.3	2.3	13	7.7
45	Cape Cross	0	0.0	59	75	2.1	2.4	20	7.7
46	Cape Cross	0	0.0	22	49	1.6	1.7	20	4.6
47	Cape Cross	0	0.0	29	287	1.4	2.5	8.7	8.7
48	Cape Cross	0	0.0	16	27	1.2	0.8	21	6.6

Appendix 3.3: Trace metal readings (ppm) in winter and summer for sediment samples in stations.

Sample ID	Station	Winter Cu	Summer Cu	Winter Zn	Summer Zn	Winter Fe	Summer Fe	Winter Pb	Summer Pb
1	Walvis Bay	36	22	64	33	11502	6696	45	30
2	Walvis Bay	44	43	72	42	11561	6603	46	39
3	Walvis Bay	37	39	83	49	11872	7170	43	27
4	Walvis Bay	46	34	84	50	10553	8392	50	41
5	Walvis Bay	39	32	93	40	11712	6977	60	20
6	Walvis Bay	31	31	57	44	8642	7489	42	45
7	Walvis Bay	36	25	72	41	11450	6402	50	28
8	Walvis Bay	46	23	67	44	10984	6011	44	26
9	Walvis Bay	21	35	69	39	11563	7606	25	27
10	Walvis Bay	33	46	66	48	9481	6823	51	47
11	Walvis Bay	39	50	81	59	12459	10142	59	79
12	Walvis Bay	38	40	100	77	10887	9841	52	60
13	Swakopmund	5.6	4.2	17	10	7895	14582	0.7	2.3
14	Swakopmund	6.1	5.2	11	10	8002	14411	0.6	0.9
15	Swakopmund	6.0	5.6	12	11	7960	17717	0.8	1.8
16	Swakopmund	8.7	5.3	72	10	10584	16594	2.4	1.4
17	Swakopmund	3.9	5.0	39	12	6981	15911	3.4	1.8
18	Swakopmund	6.3	4.6	14	9.2	8484	16082	1.1	1.1
19	Swakopmund	4.8	4.2	13	7.0	6322	11701	0.7	1.0
20	Swakopmund	4.6	2.7	18	4.3	5044	8408	0.8	0.7
21	Swakopmund	4.8	4.6	52	10	4820	15034	0.6	1.6
22	Swakopmund	4.5	31.6	36	11	5404	13463	1.1	1.4
23	Swakopmund	5.2	4.1	26	9.3	5359	12435	0.7	1.2
24	Swakopmund	2.1	3.6	4.7	12	3673	15312	0.1	1.6

Sample ID	Station	Winter Cu	Summer Cu	Winter Zn	Summer Zn	Winter Fe	Summer Fe	Winter Pb	Summer Pb
25	Henties Bay	11	6.8	41	13	44750	23346	5.0	2.2
26	Henties Bay	12	5.7	45	16	39595	30216	3.3	3.0
27	Henties Bay	17	6.8	80	20	41109	29718	4.9	2.8
28	Henties Bay	3.9	6.7	52	19	11298	31515	0.4	3.0
29	Henties Bay	4.2	8.5	80	21	15388	36411	0.8	4.1
30	Henties Bay	6.0	7.8	28	22	17332	34026	2.9	3.5
31	Henties Bay	4.3	6.4	28	21	14476	38002	0.9	3.5
32	Henties Bay	4.8	6.2	77	19	13607	33588	1.3	2.7
33	Henties Bay	3.7	7.3	17	16	8376	30448	0.1	2.6
34	Henties Bay	4.2	7.6	27	18	10438	31179	0.5	3.0
35	Henties Bay	4.3	5.8	18	15	12971	28472	0.9	2.7
36	Henties Bay	5.4	7.1	23	18	12484	30592	1.0	3.2
37	Cape Cross	2.6	1.6	18	2.8	5134	3432	0	0.0
38	Cape Cross	3.2	2.8	11	3.1	5654	3945	0	0.5
39	Cape Cross	1.4	2.3	18	2.4	2876	3079	0	0.0
40	Cape Cross	3.5	2.2	50	3.8	2745	4499	0	0.2
41	Cape Cross	1.9	2.9	48	2.8	2574	3311	0	0.0
42	Cape Cross	2.4	2.6	39	4.3	2559	5261	0	0.2
43	Cape Cross	1.3	2.5	32	9.8	2582	8546	0	0.7
44	Cape Cross	1.6	2.1	26	2.4	2823	3595	0	0.2
45	Cape Cross	1.3	2.6	3.6	3.7	2729	4910	0	0.8
46	Cape Cross	1.8	1.4	92	2.2	2497	2234	0	0.0
47	Cape Cross	2.5	2.5	45	3.8	2209	5103	0	0.3
48	Cape Cross	1.6	1.5	3.5	3.6	2676	4985	0	0.3

APPENDIX 4

Appendix 4.1: Data collection sheets for winter season

The following tables present the data recording sheets for *Choromytilus meridionalis* species, sediments and water during winter season of this investigation. The size and weight are for readings for mussels, temperature, dissolved oxygen and pH for seawater and the rest apply to all samples respectively.

Walvis Bay Station

Sampling Date	Sample ID	Site	Season	GPS coordinates	Temp (°C)	DO	pH	Fresh weight (g) live	Length (cm)
19/04/12	1	Walvis Bay	Winter	S:22°56'50.3" E: 014°30'04.3"	16.1	4.18	8.09	24.019	5.5
19/04/12	2	Walvis Bay	Winter	S:22°56'50.3" E: 014°30'04.3"	16.1	4.18	8.09	26.9001	6.66
19/04/12	3	Walvis Bay	Winter	S:22°56'50.3" E: 014°30'04.3"	16.1	4.18	8.09	25.7892	6.52
19/04/12	4	Walvis Bay	Winter	S:22°56'50.3" E: 014°30'04.3"	16.1	4.18	8.09	27.6387	6.48
19/04/12	5	Walvis Bay	Winter	S:22°56'50.3" E: 014°30'04.3"	16.1	4.18	8.09	27.4002	6.42
19/04/12	6	Walvis Bay	Winter	S:22°56'50.3" E: 014°30'04.3"	16.1	4.18	8.09	28.2535	6.4
19/04/12	7	Walvis Bay	Winter	S:22°56'50.3" E: 014°30'04.3"	16.1	4.18	8.09	26.3987	6.42
19/04/12	8	Walvis Bay	Winter	S:22°56'50.3" E: 014°30'04.3"	16.1	4.18	8.09	26.8014	6.54
19/04/12	9	Walvis Bay	Winter	S:22°56'50.3" E: 014°30'04.3"	16.1	4.18	8.09	25.1932	6.6
19/04/12	10	Walvis Bay	Winter	S:22°56'50.3" E: 014°30'04.3"	16.1	4.18	8.09	24.5186	5.38
19/04/12	11	Walvis Bay	Winter	S:22°56'50.3" E: 014°30'04.3"	16.1	4.18	8.09	19.3502	5.32
19/04/12	12	Walvis Bay	Winter	S:22°56'50.3"E: 014°30'04.3"	16.1	4.18	8.09	24.969	6.4

Swakopmund station

Date	Sample ID	Sampling Site	Season	GPS coordinates (South)	GPS coordinates (East)	Temp (^{O}C)	Mean DO	pH	Fresh weight (g) live	Length (cm)
05/07/12	13	Swakopmund	Winter	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	17.03	7.69	8.81	43.83	7.38
05/07/12	14	Swakopmund	Winter	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	17.03	7.69	8.81	62.8509	7.94
05/07/12	15	Swakopmund	Winter	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	17.03	7.69	8.81	41.53	8.33
05/07/12	16	Swakopmund	Winter	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	17.03	7.69	8.81	50.767	7.98
05/07/12	17	Swakopmund	Winter	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	17.03	7.69	8.81	45.8122	7.2
05/07/12	18	Swakopmund	Winter	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	17.03	7.69	8.81	44.685	7.94
05/07/12	19	Swakopmund	Winter	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	17.03	7.69	8.81	46.8677	6.56
05/07/12	20	Swakopmund	Winter	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	17.03	7.69	8.81	55.595	7.94
05/07/12	21	Swakopmund	Winter	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	17.03	7.69	8.81	39.7194	6.72
05/07/12	22	Swakopmund	Winter	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	17.03	7.69	8.81	49.0593	7.98
05/07/12	23	Swakopmund	Winter	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	17.03	7.69	8.81	45.4082	8.3
05/07/12	24	Swakopmund	Winter	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	17.03	7.69	8.81	44.7092	8.2

Henties Bay Station

Sampling Date	Sample ID	Sampling Site	Season	GPS coordinates (South)	GPS coordinates (East)	Temp (^{O}C)	Salinity	Mean DO	pH	Fresh weight (g) live	Length (cm)
15/05/12	25	Henties Bay	Winter	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	14.4	94.40%	6.11	7.94	7.9887	2.14
15/05/12	26	Henties Bay	Winter	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	14.4	94.40%	6.11	7.94	4.5345	3.09
15/05/12	27	Henties Bay	Winter	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	14.4	94.40%	6.11	7.94	6.5467	2.90
15/05/12	28	Henties Bay	Winter	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	14.4	94.40%	6.11	7.94	5.7892	1.99
15/05/12	29	Henties Bay	Winter	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	14.4	94.40%	6.11	7.94	5.9976	4.17
15/05/12	30	Henties Bay	Winter	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	14.4	94.40%	6.11	7.94	3.4423	3.75
15/05/12	31	Henties Bay	Winter	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	14.4	94.40%	6.11	7.94	5.3451	3.05
15/05/12	32	Henties Bay	Winter	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	14.4	94.40%	6.11	7.94	6.8767	2.76
15/05/12	33	Henties Bay	Winter	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	14.4	94.40%	6.11	7.94	5.6353	2.63
15/05/12	34	Henties Bay	Winter	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	14.4	94.40%	6.11	7.94	4.6747	3.01
15/05/12	35	Henties Bay	Winter	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	14.4	94.40%	6.11	7.94	7.3590	2.87
15/05/12	36	Henties Bay	Winter	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	14.4	94.40%	6.11	7.94	5.4244	3.47

Cape Cross Station

Sampling Date	Sample ID	Sampling Site	Season	GPS coordinates (South)	GPS coordinates (East)	Temp (^{O}C)	Dissolved oxygen (DO) – average	pH	Fresh weight (g) live	Length (cm)
06/06/12	37	Cape Cross	Winter	S:21°45'22.5"	E: 013°58'08.2"	15.3	7.48	8.65	64.5391	9
06/06/12	38	Cape Cross	Winter	S:21°45'22.5"	E: 013°58'08.2"	15.3	7.48	8.65	60.364	9.2
06/06/12	39	Cape Cross	Winter	S:21°45'22.5"	E: 013°58'08.2"	15.3	7.48	8.65	37.425	7.52
06/06/12	40	Cape Cross	Winter	S:21°45'22.5"	E: 013°58'08.2"	15.3	7.48	8.65	32.402	6.9
06/06/12	41	Cape Cross	Winter	S:21°45'22.5"	E: 013°58'08.2"	15.3	7.48	8.65	31.196	6.3
06/06/12	42	Cape Cross	Winter	S:21°45'22.5"	E: 013°58'08.2"	15.3	7.48	8.65	29.539	6.6
06/06/12	43	Cape Cross	Winter	S:21°45'22.5"	E: 013°58'08.2"	15.3	7.48	8.65	29.1667	6.57
06/06/12	44	Cape Cross	Winter	S:21°45'22.5"	E: 013°58'08.2"	15.3	7.48	8.65	30.229	6.57
06/06/12	45	Cape Cross	Winter	S:21°45'22.5"	E: 013°58'08.2"	15.3	7.48	8.65	26.1009	6.6
06/06/12	46	Cape Cross	Winter	S:21°45'22.5"	E: 013°58'08.2"	15.3	7.48	8.65	24.5045	6.61
06/06/12	47	Cape Cross	Winter	S:21°45'22.5"	E: 013°58'08.2"	15.3	7.48	8.65	23.7481	5.8
06/06/12	48	Cape Cross	Winter	S:21°45'22.5"	E: 013°58'08.2"	15.3	7.48	8.65	22.7289	6.3

Appendix 4.2: Data collection sheets for summer season

The following tables present the data recording sheets for *Choromytilus meridionalis* species, sediments and water during summer season. The size and weight are for readings for mussels, temperature, dissolved oxygen and pH for seawater and the rest apply to all samples respectively.

Walvis Bay Station

Sampling Date	Sample ID	Site	Season	GPS coordinates	Temp (°C)	DO	pH	Fresh weight (g) live	Length (cm)
14/11/12	1	Walvis Bay	Summer	S:22°56'50.3" E: 014°30'04.3"	18.80	8.59	7.92	24.1752	6.40
14/11/12	2	Walvis Bay	Summer	S:22°56'50.3" E: 014°30'04.3"	18.80	8.59	7.92	43.1462	8.02
14/11/12	3	Walvis Bay	Summer	S:22°56'50.3" E: 014°30'04.3"	18.80	8.59	7.92	32.6986	7.30
14/11/12	4	Walvis Bay	Summer	S:22°56'50.3" E: 014°30'04.3"	18.80	8.59	7.92	16.8036	6.80
14/11/12	5	Walvis Bay	Summer	S:22°56'50.3" E: 014°30'04.3"	18.80 18.80	8.59	7.92	38.5373	7.40
14/11/12	6	Walvis Bay	Summer	S:22°56'50.3" E: 014°30'04.3"	18.80	8.59	7.92	29.1017	5.67
14/11/12	7	Walvis Bay	Summer	S:22°56'50.3" E: 014°30'04.3"	18.80	8.59	7.92	31.9508	6.02
14/11/12	8	Walvis Bay	Summer	S:22°56'50.3" E: 014°30'04.3"	18.80	8.59	7.92	22.2358	5.90
14/11/12	9	Walvis Bay	Summer	S:22°56'50.3" E: 014°30'04.3"	18.80	8.59	7.92	15.0701	5.56
14/11/12	10	Walvis Bay	Summer	S:22°56'50.3" E: 014°30'04.3"	18.80	8.59	7.92	25.5497	6.0
14/11/12	11	Walvis Bay	Summer	S:22°56'50.3" E: 014°30'04.3"	18.80 18.80	8.59	7.92	26.4930	6.10
14/11/12	12	Walvis Bay	Summer	S:22°56'50.3"E: 014°30'04.3"	18.80	8.59	7.92	24.5310	5.90

Swakopmund station

Sampling Date	Sample ID	Sampling Site	Season	GPS coordinates (South)	GPS coordinates (East)	Temp (^{O}C)	Mean DO	pH	Fresh weight (g) live	Length (cm)
02/11/12	13	Swakopmund	Summer	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	16.9	9.26	8.59	35.3112	6.67
02/11/12	14	Swakopmund	Summer	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	16.9	9.26	8.59	21.3625	7.89
02/11/12	15	Swakopmund	Summer	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	16.9	9.26	8.59	21.0213	8.0
02/11/12	16	Swakopmund	Summer	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	16.9	9.26	8.59	28.4461	9.07
02/11/12	17	Swakopmund	Summer	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	16.9	9.26	8.59	22.9576	7.42
02/11/12	18	Swakopmund	Summer	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	16.9	9.26	8.59	15.7124	7.87
02/11/12	19	Swakopmund	Summer	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	16.9	9.26	8.59	23.4676	5.47
02/11/12	20	Swakopmund	Summer	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	16.9	9.26	8.59	23.1335	7.82
02/11/12	21	Swakopmund	Summer	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	16.9	9.26	8.59	12.0702	6.77
02/11/12	22	Swakopmund	Summer	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	16.9	9.26	8.59	19.4118	7.06
02/11/12	23	Swakopmund	Summer	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	16.9	9.26	8.59	17.8240	8.38
02/11/12	24	Swakopmund	Summer	S:22^{o}42’02.7”	E: 014^{o}31’14.9”	16.9	9.26	8.59	21.6868	8.68

Henties Bay station

Sampling Date	Sample ID	Sampling Site	Season	GPS coordinates (South)	GPS coordinates (East)	Temp (^{O}C)	Mean DO	pH	Fresh weight (g) live	Length (cm)
31/10/12	25	Henties Bay	Summer	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	15.0	4.73	5.80	11.4072	2.2
31/10/12	26	Henties Bay	Summer	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	15.0	4.73	5.80	22.0818	3.08
31/10/12	27	Henties Bay	Summer	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	15.0	4.73	5.80	13.7389	2.98
31/10/12	28	Henties Bay	Summer	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	15.0	4.73	5.80	16.9001	5.20
31/10/12	29	Henties Bay	Summer	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	15.0	4.73	5.80	18.7469	4.11
31/10/12	30	Henties Bay	Summer	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	15.0	4.73	5.80	14.0261	3.95
31/10/12	31	Henties Bay	Summer	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	15.0	4.73	5.80	11.6716	5.56
31/10/12	32	Henties Bay	Summer	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	15.0	4.73	5.80	15.8589	2.76
31/10/12	33	Henties Bay	Summer	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	15.0	4.73	5.80	13.7480	2.64
31/10/12	34	Henties Bay	Summer	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	15.0	4.73	5.80	11.6757	3.03
31/10/12	35	Henties Bay	Summer	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	15.0	4.73	5.80	8.4043	2.70
31/10/12	36	Henties Bay	Summer	S:22^{o}24’34.8”	E: 014^{o}26’38.7”	15.0	4.73	5.80	9.5698	3.21

Cape Cross station

Sampling Date	Sample ID	Sampling Site	Season	GPS coordinates (South)	GPS coordinates (East)	Temp (^{O}C)	Mean DO	pH	Fresh weight (g) live	Length (cm)
10/09/12	37	Cape Cross	Summer	S:21^{o}45’22.5”	E: 013^{o}58’08.2”	15.7	4.73	4.67	30.4474	8.90
10/09/12	38	Cape Cross	Summer	S:21^{o}45’22.5”	E: 013^{o}58’08.2”	15.7	4.73	4.67	25.9166	8.34
10/09/12	39	Cape Cross	Summer	S:21^{o}45’22.5”	E: 013^{o}58’08.2”	15.7	4.73	4.67	26.7622	7.68
10/09/12	40	Cape Cross	Summer	S:21^{o}45’22.5”	E: 013^{o}58’08.2”	15.7	4.73	4.67	18.5614	6.95
10/09/12	41	Cape Cross	Summer	S:21^{o}45’22.5”	E: 013^{o}58’08.2”	15.7	4.73	4.67	17.7697	6.0
10/09/12	42	Cape Cross	Summer	S:21^{o}45’22.5”	E: 013^{o}58’08.2”	15.7	4.73	4.67	11.2580	8.86
10/09/12	43	Cape Cross	Summer	S:21^{o}45’22.5”	E: 013^{o}58’08.2”	15.7	4.73	4.67	14.1216	6.9
10/09/12	44	Cape Cross	Summer	S:21^{o}45’22.5”	E: 013^{o}58’08.2”	15.7	4.73	4.67	15.3027	6.39
10/09/12	45	Cape Cross	Summer	S:21^{o}45’22.5”	E: 013^{o}58’08.2”	15.7	4.73	4.67	12.8631	5.97
10/09/12	46	Cape Cross	Summer	S:21^{o}45’22.5”	E: 013^{o}58’08.2”	15.7	4.73	4.67	16.9075	6.71
10/09/12	47	Cape Cross	Summer	S:21^{o}45’22.5”	E: 013^{o}58’08.2”	15.7	4.73	4.67	14.7788	6.43
10/09/12	48	Cape Cross	Summer	S:21^{o}45’22.5”	E: 013^{o}58’08.2”	15.7	4.73	4.67	13.4940	8.33

APPENDIX 5

ACID DIGESTION OF SEDIMENTS, SLUDGES, AND SOILS (EPA METHOD 3050B)

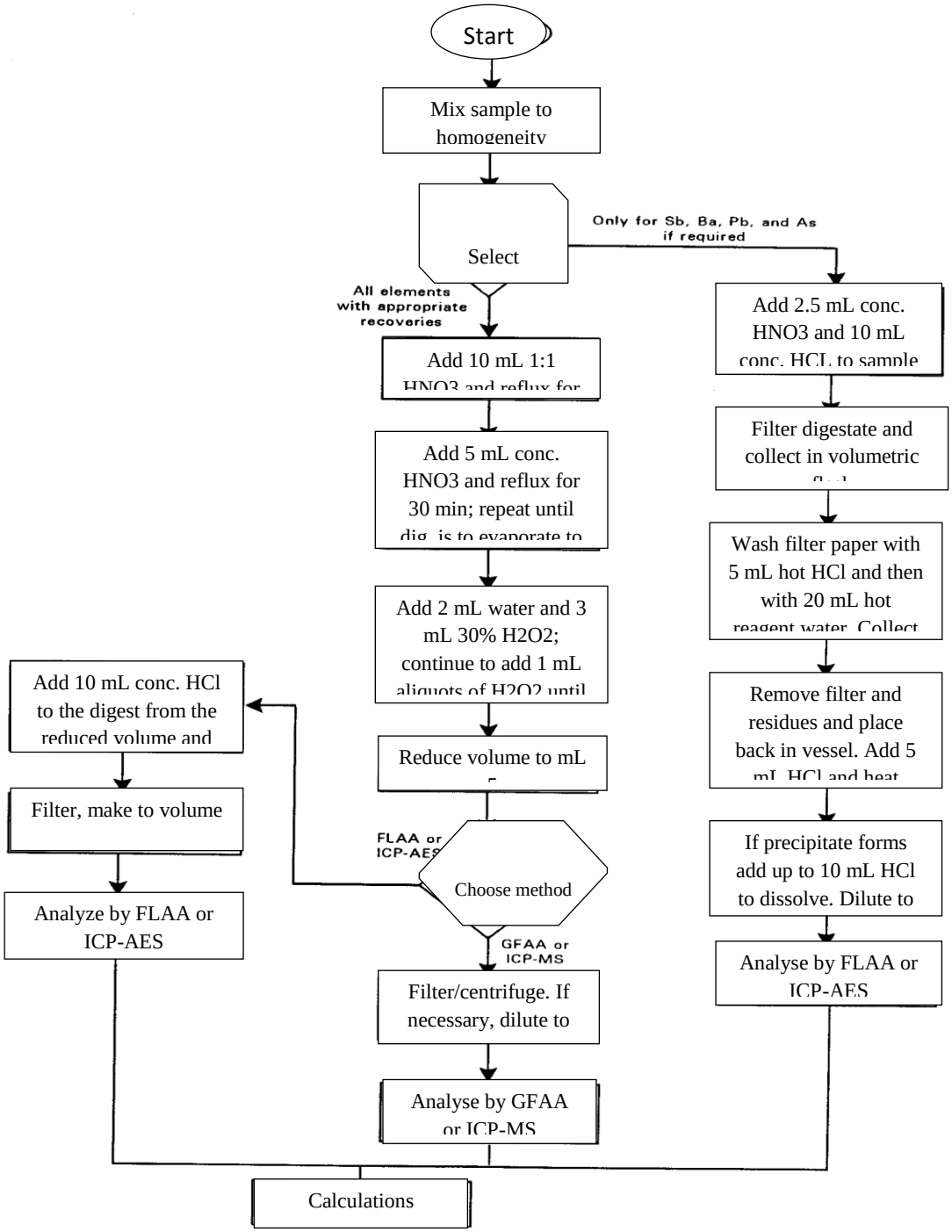

Figure 23: Digestion protocol (EPA method)

APPENDIX 6

The following tables represent calibration values and information for the machine (Perkin Elmer Optima 7, 000 DV) used in this study to analyse samples.

Water samples calibration data and information for the machine (Perkin Elmer Optima 7, 000 DV) used in this study.

Sample ID	Date	Analyte Name	Concentration (Sample)	RSD (Conc)	Correlation Coefficient
BLK N	07/01/2013	Cu 324.752		0	
BLK N	07/01/2013	Zn 213.857		0	
BLK N	07/01/2013	Fe 259.939		0	
BLK N	07/01/2013	Pb 220.353		0	
UN-0.01	07/01/2013	Cu 324.752		1.65E-06	
UN-0.01	07/01/2013	Zn 213.857		1.65E-06	
UN-0.01	07/01/2013	Pb 220.353		1.65E-06	
UN-0.25	07/01/2013	Cu 324.752		0	
UN-0.25	07/01/2013	Zn 213.857		0	
UN-0.25	07/01/2013	Fe 259.939		0	
UN-0.25	07/01/2013	Pb 220.353		0	
UN-1.0	07/01/2013	Cu 324.752		0	
UN-1.0	07/01/2013	Zn 213.857		0	
UN-1.0	07/01/2013	Fe 259.939		0	
UN-1.0	07/01/2013	Pb 220.353		0	
UN-2.5	07/01/2013	Cu 324.752		0	
UN-2.5	07/01/2013	Zn 213.857		0	
UN-2.5	07/01/2013	Fe 259.939		0	
UN-2.5	07/01/2013	Pb 220.353		0	
UN-5.0	07/01/2013	Cu 324.752		0	
UN-5.0	07/01/2013	Zn 213.857		0	
UN-5.0	07/01/2013	Fe 259.939		0	
UN-5.0	07/01/2013	Pb 220.353		0	
UN-0.25	07/01/2013	Cu 324.752	0.249355326	1.811014717	0.999980973
UN-0.25	07/01/2013	Zn 213.857	0.251214205	0.299692381	0.999995116
UN-0.25	07/01/2013	Fe 259.939	0.184873828	0.370749492	0.999719256
UN-0.25	07/01/2013	Pb 220.353	0.247018835	0.766850422	0.999994718
BLK	07/01/2013	Cu 324.752	0.001060149	21.84848064	0.999980973

Sample ID	Date	Analyte Name	Concentration (Sample)	RSD (Conc)	Correlation Coefficient
BLK	07/01/2013	Zn 213.857	-0.00112523	11.2380591	0.999995116
BLK	07/01/2013	Fe 259.939	0.001547781	14.89306602	0.999719256
BLK	07/01/2013	Pb 220.353	-0.000351804	270.1237371	0.999994718

New calibration curve

Sample ID	Date	Analyte Name	Concentration (Sample)	RSD (Conc)	Correlation Coefficient
BLK N	07/01/2013	Cu 324.752		0	
BLK N	07/01/2013	Zn 213.857		0	
BLK N	07/01/2013	Fe 259.939		0	
BLK N	07/01/2013	Pb 220.353		0	
UN-0.01	07/01/2013	Cu 324.752		1.65E-06	
UN-0.01	07/01/2013	Zn 213.857		1.65E-06	
UN-0.01	07/01/2013	Pb 220.353		1.65E-06	
UN-0.25	07/01/2013	Cu 324.752		0	
UN-0.25	07/01/2013	Zn 213.857		0	
UN-0.25	07/01/2013	Fe 259.939		0	
UN-0.25	07/01/2013	Pb 220.353		0	
UN-1.0	07/01/2013	Cu 324.752		0	
UN-1.0	07/01/2013	Zn 213.857		0	
UN-1.0	07/01/2013	Fe 259.939		0	
UN-1.0	07/01/2013	Pb 220.353		0	
UN-2.5	07/01/2013	Cu 324.752		0	
UN-2.5	07/01/2013	Zn 213.857		0	
UN-2.5	07/01/2013	Fe 259.939		0	
UN-2.5	07/01/2013	Pb 220.353		0	
UN-5.0	07/01/2013	Cu 324.752		0	
UN-5.0	07/01/2013	Zn 213.857		0	
UN-5.0	07/01/2013	Fe 259.939		0	
UN-5.0	07/01/2013	Pb 220.353		0	
UN-0.25	07/01/2013	Cu 324.752	0.265469957	3.26875994	0.99994782
UN-0.25	07/01/2013	Zn 213.857	0.265728666	0.971712175	0.999763708
UN-0.25	07/01/2013	Fe 259.939	0.183054783	1.76084843	0.999801513
UN-0.25	07/01/2013	Pb 220.353	0.257285259	3.102810451	0.999895231
BLK	07/01/2013	Cu 324.752	-0.000360271	57.39202977	0.99994782
BLK	07/01/2013	Zn 213.857	0.000177332	5.477380631	0.999763708

Sample ID	Date	Analyte Name	Concentration (Sample)	RSD (Conc)	Correlation Coefficient
BLK	07/01/2013	Fe 259.939	0.001414816	202.7339908	0.999801513
BLK	07/01/2013	Pb 220.353	0.02597087	8.473463745	0.999895231

New calibration curve

Sample ID	Date	Analyte Name	Concentration (Sample)	RSD (Conc)	Correlation Coefficient
BLK N	07/01/2013	Cu 324.752		0	
BLK N	07/01/2013	Zn 213.857		0	
BLK N	07/01/2013	Fe 259.939		0	
BLK N	07/01/2013	Pb 220.353		0	
UN-0.01	07/01/2013	Cu 324.752		1.65E-06	
UN-0.01	07/01/2013	Zn 213.857		1.65E-06	
UN-0.01	07/01/2013	Pb 220.353		1.65E-06	
UN-0.25	07/01/2013	Cu 324.752		0	
UN-0.25	07/01/2013	Zn 213.857		0	
UN-0.25	07/01/2013	Fe 259.939		0	
UN-0.25	07/01/2013	Pb 220.353		0	
UN-1.0	07/01/2013	Cu 324.752		0	
UN-1.0	07/01/2013	Zn 213.857		0	
UN-1.0	07/01/2013	Fe 259.939		0	
UN-1.0	07/01/2013	Pb 220.353		0	
UN-2.5	07/01/2013	Cu 324.752		0	
UN-2.5	07/01/2013	Zn 213.857		0	
UN-2.5	07/01/2013	Fe 259.939		0	
UN-2.5	07/01/2013	Pb 220.353		0	
UN-5.0	07/01/2013	Cu 324.752		0	
UN-5.0	07/01/2013	Zn 213.857		0	
UN-5.0	07/01/2013	Fe 259.939		0	
UN-5.0	07/01/2013	Pb 220.353		0	
UN-0.25	07/01/2013	Cu 324.752	0.306658511	0.372729248	0.999875408
UN-0.25	07/01/2013	Zn 213.857	0.271351868	0.897242913	0.999940238
UN-0.25	07/01/2013	Fe 259.939	0.308717263	1.26668825	0.999934733
UN-0.25	07/01/2013	Pb 220.353	0.253003395	1.83925577	0.999958292
BLK	07/01/2013	Cu 324.752	0.001238356	27.83668129	0.999875408
BLK	07/01/2013	Zn 213.857	-0.000310419	95.08397597	0.999940238
BLK	07/01/2013	Fe 259.939	5.45E-05	299.1623317	0.999934733
BLK	07/01/2013	Pb 220.353	-0.002045229	173.4213117	0.999958292
BLK N	07/01/2013	Cu 324.752		0	

Sample ID	Date	Analyte Name	Concentration (Sample)	RSD (Conc)	Correlation Coefficient
BLK N	07/01/2013	Zn 213.857		0	
BLK N	07/01/2013	Fe 259.939		0	
BLK N	07/01/2013	Pb 220.353		0	
UN-0.01	07/01/2013	Cu 324.752		1.65E-06	
UN-0.01	07/01/2013	Zn 213.857		1.65E-06	
UN-0.01	07/01/2013	Pb 220.353		1.65E-06	
UN-0.25	07/01/2013	Cu 324.752		0	
UN-0.25	07/01/2013	Zn 213.857		0	
UN-0.25	07/01/2013	Fe 259.939		0	
UN-0.25	07/01/2013	Pb 220.353		0	
UN-1.0	07/01/2013	Cu 324.752		0	
UN-1.0	07/01/2013	Zn 213.857		0	
UN-1.0	07/01/2013	Fe 259.939		0	
UN-1.0	07/01/2013	Pb 220.353		0	
UN-2.5	07/01/2013	Cu 324.752		0	
UN-2.5	07/01/2013	Zn 213.857		0	
UN-2.5	07/01/2013	Fe 259.939		0	
UN-2.5	07/01/2013	Pb 220.353		0	
UN-5.0	07/01/2013	Cu 324.752		0	
UN-5.0	07/01/2013	Zn 213.857		0	
UN-5.0	07/01/2013	Fe 259.939		0	
UN-5.0	07/01/2013	Pb 220.353		0	
UN-0.25	07/01/2013	Cu 324.752	0.261605073	2.279876557	0.999996
UN-0.25	07/01/2013	Zn 213.857	0.263086789	0.73023703	0.999972
UN-0.25	07/01/2013	Fe 259.939	0.205724567	0.392785967	0.999886
UN-0.25	07/01/2013	Pb 220.353	0.253639763	1.31821776	0.999998
BLK	07/01/2013	Cu 324.752	0.001725924	33.66022909	0.999996
BLK	07/01/2013	Zn 213.857	-0.000572038	29.52424428	0.999972
BLK	07/01/2013	Fe 259.939	0.001221528	61.16750083	0.999886
BLK	07/01/2013	Pb 220.353	-0.002425486	21.55743053	0.999998

Mussel calibration data and information for the machine (Perkin Elmer Optima 7, 000 DV) used in this study

Sample ID	Date	Analyte Name	Concentration (Sample)	RSD (Conc)	Correlation Coefficient
BLK N	30/01/2013	Cu 324.752		0	
BLK N	30/01/2013	Zn 213.857		0	
BLK N	30/01/2013	Fe 259.939		0	
BLK N	30/01/2013	Pb 220.353		0	
UN-0.01	30/01/2013	Cu 324.752		1.65E-06	
UN-0.01	30/01/2013	Zn 213.857		1.65E-06	
UN-0.01	30/01/2013	Pb 220.353		1.65E-06	
UN-0.25	30/01/2013	Cu 324.752		0	
UN-0.25	30/01/2013	Zn 213.857		0	
UN-0.25	30/01/2013	Fe 259.939		0	
UN-0.25	30/01/2013	Pb 220.353		0	
UN-1.0	30/01/2013	Cu 324.752		0	
UN-1.0	30/01/2013	Zn 213.857		0	
UN-1.0	30/01/2013	Fe 259.939		0	
UN-1.0	30/01/2013	Pb 220.353		0	
UN-2.5	30/01/2013	Cu 324.752		0	
UN-2.5	30/01/2013	Zn 213.857		0	
UN-2.5	30/01/2013	Fe 259.939		0	
UN-2.5	30/01/2013	Pb 220.353		0	
UN-5.0	30/01/2013	Cu 324.752		0	

Sample ID	Date	Analyte Name	Concentration (Sample)	RSD (Conc)	Correlation Coefficient
UN-5.0	30/01/2013	Zn 213.857		0	
UN-5.0	30/01/2013	Fe 259.939		0	
UN-5.0	30/01/2013	Pb 220.353		0	
UN-1.0	30/01/2013	Cu 324.752	1.012393588	1.032975183	0.999989408
UN-1.0	30/01/2013	Zn 213.857	1.002241513	0.599270819	0.999975561
UN-1.0	30/01/2013	Fe 259.939	1.00556531	1.745131818	0.999772088
UN-1.0	30/01/2013	Pb 220.353	0.990703173	0.999534905	0.999992675
BLK	30/01/2013	Cu 324.752	-0.001032072	13.25097481	0.999989408
BLK	30/01/2013	Zn 213.857	0.000373844	46.77725538	0.999975561
BLK	30/01/2013	Fe 259.939	0.000745548	21.11704272	0.999772088
BLK	30/01/2013	Pb 220.353	-0.001530189	101.4817653	0.999992675
UN-1.0	30/01/2013	Cu 324.752	1.032969332	0.553294083	0.999989408
UN-1.0	30/01/2013	Zn 213.857	1.021440265	0.663594478	0.999975561
UN-1.0	30/01/2013	Fe 259.939	1.005666296	0.384316562	0.999772088
UN-1.0	30/01/2013	Pb 220.353	1.003000902	0.312753293	0.999992675
UN-1.0	30/01/2013	Cu 324.752	1.062469307	1.288604225	0.999989408
UN-1.0	30/01/2013	Zn 213.857	1.007775358	0.867171544	0.999975561
UN-1.0	30/01/2013	Fe 259.939	1.036532581	0.45458894	0.999772088
UN-1.0	30/01/2013	Pb 220.353	1.021482312	0.487119489	0.999992675
UN-1.0	30/01/2013	Cu 324.752	1.054814794	2.163223507	0.999989408
UN-1.0	30/01/2013	Zn 213.857	0.982530448	0.691469548	0.999975561
UN-1.0	30/01/2013	Fe 259.939	1.027813478	1.206142698	0.999772088

Sample ID	Date	Analyte Name	Concentration (Sample)	RSD (Conc)	Correlation Coefficient
UN-1.0	30/01/2013	Pb 220.353	0.986858374	0.420876892	0.999992675
UN-1.0	30/01/2013	Cu 324.752	-0.021496003	21.42912046	0.999989408
UN-1.0	30/01/2013	Zn 213.857	-0.002998075	9.415681387	0.999975561
UN-1.0	30/01/2013	Fe 259.939	-0.004733927	2.573878762	0.999772088
UN-1.0	30/01/2013	Pb 220.353	-0.02446688	10.15595827	0.999992675
UN-1.0	30/01/2013	Cu 324.752	1.055564599	0.851921254	0.999989408
UN-1.0	30/01/2013	Zn 213.857	0.947424509	1.203182537	0.999975561
UN-1.0	30/01/2013	Fe 259.939	0.992660661	0.450710304	0.999772088
UN-1.0	30/01/2013	Pb 220.353	0.958052432	0.588357456	0.999992675
BLK N	30/01/2013	Cu 324.752		0	
BLK N	30/01/2013	Zn 213.857		0	
BLK N	30/01/2013	Fe 259.939		0	
BLK N	30/01/2013	Pb 220.353		0	
UN-0.01	30/01/2013	Cu 324.752		1.65E-06	
UN-0.01	30/01/2013	Zn 213.857		1.65E-06	
UN-0.01	30/01/2013	Pb 220.353		1.65E-06	
UN-0.25	30/01/2013	Cu 324.752		0	
UN-0.25	30/01/2013	Zn 213.857		0	
UN-0.25	30/01/2013	Fe 259.939		0	
UN-0.25	30/01/2013	Pb 220.353		0	
UN-1.0	30/01/2013	Cu 324.752		0	

Sample ID	Date	Analyte Name	Concentration (Sample)	RSD (Conc)	Correlation Coefficient
UN-1.0	30/01/2013	Zn 213.857		0	
UN-1.0	30/01/2013	Fe 259.939		0	
UN-1.0	30/01/2013	Pb 220.353		0	
UN-2.5	30/01/2013	Cu 324.752		0	
UN-2.5	30/01/2013	Zn 213.857		0	
UN-2.5	30/01/2013	Fe 259.939		0	
UN-2.5	30/01/2013	Pb 220.353		0	
UN-5.0	30/01/2013	Cu 324.752		0	
UN-5.0	30/01/2013	Zn 213.857		0	
UN-5.0	30/01/2013	Fe 259.939		0	
UN-5.0	30/01/2013	Pb 220.353		0	
UN-1.0	30/01/2013	Cu 324.752	1.001236566	2.719286709	0.999978875
UN-1.0	30/01/2013	Zn 213.857	1.019344756	0.950944364	0.99997877
UN-1.0	30/01/2013	Fe 259.939	0.991931202	1.199603127	0.999724671
UN-1.0	30/01/2013	Pb 220.353	1.007016236	1.026303376	0.999993661
UN-1.0	30/01/2013	Cu 324.752	1.032126202	2.054584473	0.999978875
UN-1.0	30/01/2013	Zn 213.857	1.022678458	0.17602045	0.99997877
UN-1.0	30/01/2013	Fe 259.939	1.021106957	1.096661807	0.999724671
UN-1.0	30/01/2013	Pb 220.353	0.995677935	0.339210317	0.999993661
UN-1.0	30/01/2013	Cu 324.752	1.003326976	1.472544552	0.999978875
UN-1.0	30/01/2013	Zn 213.857	0.964738576	0.529315002	0.99997877
UN-1.0	30/01/2013	Fe 259.939	0.978840411	1.009232118	0.999724671

Sample ID	Date	Analyte Name	Concentration (Sample)	RSD (Conc)	Correlation Coefficient
UN-1.0	30/01/2013	Pb 220.353	0.950514741	0.931608726	0.999993661
UN-1.0	30/01/2013	Cu 324.752	1.013813095	1.891364799	0.999978875
UN-1.0	30/01/2013	Zn 213.857	0.951793344	1.125337806	0.99997877
UN-1.0	30/01/2013	Fe 259.939	0.988339829	0.763881966	0.999724671
UN-1.0	30/01/2013	Pb 220.353	0.939222982	0.487114376	0.999993661

Sediment calibration data and information for the machine (Perkin Elmer Optima 7, 000 DV) used in this study

Sample ID	Date	Analyte Name	Concentration (Sample)	RSD (Conc)	Correlation Coefficient
BLK N	07/02/2013	Cu 324.752		0	
BLK N	07/02/2013	Zn 213.857		0	
BLK N	07/02/2013	Fe 259.939		0	
BLK N	07/02/2013	Pb 220.353		0	
UN-0.01	07/02/2013	Cu 324.752		1.65E-06	
UN-0.01	07/02/2013	Zn 213.857		1.65E-06	
UN-0.01	07/02/2013	Pb 220.353		1.65E-06	
UN-0.25	07/02/2013	Cu 324.752		0	
UN-0.25	07/02/2013	Zn 213.857		0	
UN-0.25	07/02/2013	Fe 259.939		0	
UN-0.25	07/02/2013	Pb 220.353		0	
UN-1.0	07/02/2013	Cu 324.752		0	
UN-1.0	07/02/2013	Zn 213.857		0	
UN-1.0	07/02/2013	Fe 259.939		0	
UN-1.0	07/02/2013	Pb 220.353		0	
UN-2.5	07/02/2013	Cu 324.752		0	
UN-2.5	07/02/2013	Zn 213.857		0	
UN-2.5	07/02/2013	Fe 259.939		0	
UN-2.5	07/02/2013	Pb 220.353		0	
UN-5.0	07/02/2013	Cu 324.752		0	
UN-5.0	07/02/2013	Zn 213.857		0	
UN-5.0	07/02/2013	Fe 259.939		0	
UN-5.0	07/02/2013	Pb 220.353		0	
UN-1.0	07/02/2013	Cu 324.752	1.026738511	2.694318942	0.999965986

Sample ID	Date	Analyte Name	Concentration (Sample)	RSD (Conc)	Correlation Coefficient
UN-1.0	07/02/2013	Zn 213.857	1.01961359	1.063986574	0.999945886
UN-1.0	07/02/2013	Fe 259.939	1.005717509	0.931656483	0.999752165
UN-1.0	07/02/2013	Pb 220.353	1.001719095	0.764054243	0.999995731
BLK	07/02/2013	Cu 324.752	0.001330658	10.08288157	0.999965986
BLK	07/02/2013	Zn 213.857	-5.76E-05	113.5506624	0.999945886
BLK	07/02/2013	Fe 259.939	0.000285208	15.23237053	0.999752165
BLK	07/02/2013	Pb 220.353	-0.001686511	63.29482618	0.999995731
UN-1.0	07/02/2013	Cu 324.752	1.034490733	1.516535494	0.999965986
UN-1.0	07/02/2013	Zn 213.857	1.023147665	1.106848578	0.999945886
UN-1.0	07/02/2013	Pb 220.353	1.013601319	0.889648769	0.999995731
UN-1.0	07/02/2013	Cu 324.752	1.024640289	1.166343877	0.999965986
UN-1.0	07/02/2013	Zn 213.857	1.011285065	0.405413123	0.999945886
UN-1.0	07/02/2013	Pb 220.353	1.004105981	0.394886589	0.999995731
UN-1.0	07/02/2013	Cu 324.752	1.027193504	0.573390296	0.999965986
UN-1.0	07/02/2013	Zn 213.857	1.022486145	0.89859744	0.999945886
UN-1.0	07/02/2013	Pb 220.353	1.008986841	0.507955467	0.999995731
UN-1.0	07/02/2013	Cu 324.752	1.044938138	2.261652029	0.999965986
UN-1.0	07/02/2013	Zn 213.857	1.022041409	0.400826617	0.999945886
UN-1.0	07/02/2013	Pb 220.353	1.015616437	0.22510482	0.999995731
UN-1.0	07/02/2013	Cu 324.752	1.037621133	0.401621698	0.999965986
UN-1.0	07/02/2013	Zn 213.857	1.025185581	0.748859054	0.999945886
UN-1.0	07/02/2013	Pb 220.353	1.01240252	1.043490512	0.999995731
UN-1.0	07/02/2013	Cu 324.752	1.040731487	1.420562803	0.999965986
UN-1.0	07/02/2013	Zn 213.857	1.032577108	0.496321247	0.999945886
UN-1.0	07/02/2013	Pb 220.353	1.018977201	0.259723817	0.999995731
UN-1.0	07/02/2013	Cu 324.752	1.027617507	2.051601297	0.999965986
UN-1.0	07/02/2013	Zn 213.857	1.021381247	0.474280609	0.999945886
UN-1.0	07/02/2013	Pb 220.353	1.023703183	0.900795526	0.999995731