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**Environment and Sustainable
Development Program**



Accumulation of Trace Metal Contaminants in Blue Swimming Crab in Bahrain

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of M.Sc. in Environment and Sustainable Development

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- ☐ Without making any changes to the thesis.
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Abstract

Blue swimming crab (*Portunus pelagicus*) is widely found in Indian and Pacific Oceans, and many other sea areas, including the Arabian Gulf. Like the other benthic crustaceans, *Portunus pelagicus* accumulates different trace metal ions among their tissues. Blue Swimming Crab (BSC) is popular seafood in Bahrain; therefore, it is important to study its trace metal ions content.

This study determined the levels of six trace metal ions in the tissues of male and female BSC collected from four marine sites within the territorial waters of Bahrain namely, Ra's Al M'ttalah (west), Marwada (north), Askar (east) and Tubli Bay. The study was divided into two parts; in the first part, the levels of Zn^{2+} , Cu^{2+} , Cr^{3+} , Ni^{2+} , Pb^{2+} and Cd^{2+} were determined in both white meat (muscles of the legs and claws) and brown meat (digestive system) of BSC on the dry weight basis. For all stations, male and female composite samples of white and brown meat were prepared from 15 crabs. Based on first part results, three metal ions (Zn^{2+} , Cu^{2+} and Cd^{2+}) were also determined in individual males and females BSC samples on wet weight basis. White and brown meat of ten male and ten female crabs from each site were analyzed.

The concentrations of Zn^{2+} , Cu^{2+} , Cr^{3+} , Ni^{2+} and Cd^{2+} were in the range of 174.6 – 300.4, 33.3 – 325.7, 0.025 – 0.25, 0.11 – 3.03 and <0.02 – 10.3 mg/kg dry weight, respectively. While Pb^{2+} was below method quantification limit (<0.2 mg/kg). Generally, the concentrations of trace metal ions were in the following order $\text{Zn}^{2+} > \text{Cu}^{2+} > \text{Cd}^{2+} > \text{Ni}^{2+} > \text{Cr}^{3+}$. The study revealed that BSC tends to accumulate Cu^{2+} and Cd^{2+} in brown meat more than in white meat, while no significant difference was observed between Zn^{2+} concentrations in the two tissues. Female crabs were found to accumulate Zn^{2+} more than male crabs in both white and brown meats, as well as Cu^{2+} in the white meat. Crabs from Ra's Al M'ttalah had higher concentrations of Cu^{2+} and Cd^{2+} in their brown meat compared to crabs from other sites while crabs from Marwada had higher concentration of Zn^{2+} in both white and brown meats compared to crabs from other sites.

In view of study results, Cd^{2+} and Pb^{2+} levels in white BSC meat are considered safe for human consumption according to both Bahrain's standards (GSO 1922/2009) and European standards (EU No 420/2011). However, it is recommended to refrain from eating BSC brown meat as it tends to have high level of Cd^{2+} .

Dedication

To..

the souls of my Mother and Father
for their loving memories

my beloved wife
for her patience and being with me all the way

my little kids, Hussain and Fatima
who made me smile

my family & friends
for supporting and believing in me

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List of Acronyms

As ²⁺	Arsenic (II) ion, metalloid
BAF	Bioaccumulation Factor
BSC	Blue Swimming Crab
Cd ²⁺	Cadmium (II) ion, trace metal
Cr ³⁺	Chromium (III) ion, trace metal
Cu ²⁺	Copper (II) ion, trace metal
CL	Carapace Length
CW	Carapace Width
EC	European Commission
GCC	Gulf Cooperation Council
GSO	GCC Standardization Organization
Hg ²⁺	Mercury (II) ion, trace metal
Pb ²⁺	Lead (II) ion, trace metal
ROPME	Regional Organization for the Protection of the Marine Environment
SCE	Supreme Council for Environment
Sb	Antimony, metalloid
Se	Selenium, non-metal
Zn ²⁺	Zinc (II) ion, trace metal

Chapter One

Introduction to the Study

Chapter One

Introduction to the Study

1.1 Introduction

Bahrain is located in the Arabian Gulf, which is a shallow, semi-closed sea that characterized by high salinity that exceeds 39‰ (Sheppard et al., 2010). Because of the human impacts, Arabian Gulf remarks as one of the highly anthropogenically impacted regions in the world (Halpern et al., 2008). One of the major sources of pollution in the Gulf is oil spills, where the region is the most active among the movement of oil tankers. In addition, discharging the treated water of industrial and domestic wastewater, and the coastal development projects on the coastlines (Naser, 2011; Chitrakar et al., 2019).

The study of pollution in the Arabian Gulf become highly important due to the slow flashing rate of water in this region (Sheppard et al., 2010), and because the marine environment of the Arabian Gulf provides the needs of the desalinated water (Freije, 2015). Around 45% of worldwide seawater desalination plants are connected to the Arabian Gulf with a production capacity of 11 million m³/day (Lattemann & Höpner, 2008). Currently, the production capacity of desalination plants connected to the Arabian Gulf is over 20 million m³/day, and expected to rise to 80 million m³/day after 30 years (Le Quesne et al., 2021). Furthermore, Arabian Gulf seawater is considered as a major source of food for these countries, and supports the activities of recreation and the industry (Naser, 2013).

Pollutants in the Arabian Gulf can be divided into two main groups, organic (petroleum hydrocarbon) and inorganic (potentially toxic trace metals) (Freije, 2015). Due to its bioaccumulation and toxicity, potentially

toxic trace metals represent a real threat to the marine environment and ecosystems (DeForest et al., 2007), especially Hg, Cd and Pb that are considered as highly toxic metals for the living organisms (Valavanidis & Vlachogianni, 2010), even at low concentrations (Jakimska et al., 2011). For example, bioaccumulation of methylmercury (CH_3Hg) in the high level feeders of the food chain (like piscivorous birds or mammals) can cause damage to their nervous and reproductive systems (Wolfe et al., 1998). Accumulation of large quantities of Cd found to affect the respiration system in the oyster, and in some cases leads to their mortality (Engel & Fowler, 1979). Lead bioaccumulation in marine organisms can cause mortality, and reduce the rate of growth and reproduction of these organisms (Eisler, 1988).

In the marine environment, the complexation of trace metal ions with the ligands (organic matter) available in seawater takes place according to the Hard and Soft Acids and Bases principle (HSAB principle). The majority of bioactive trace metals like Fe^{3+} , Zn^{2+} and Cu^{2+} preferring to forming complexes with the natural strong organic ligands that available in the seawater (Hirose, 2006).

Many studies conducted on the trace metals and petroleum hydrocarbons in both marine sediment and marine organisms of the Arabian Gulf (Naser, 2013; Freije, 2015). In Bahrain there are limited studies for trace metals in the marine environment, most of them were conducted on marine sediment and marine organisms, whereas scarce studies related to the trace metals in seawater of Bahrain. One of the studies conducted in Bahrain in the early eighties showed that the levels of Cu, Zn, Cd, Hg, and Pb metals in seawater and suspended particulates were similar to the metal levels in the open seas like the oceans or the Mediterranean Sea. This may gave an evidence that there was no metallic pollution in Bahrain at that time (Fowler et al., 1984). After that, in 1992,

trace metals measured in water samples collected from two stations in the period of March 1991–March 1992. The results showed that there were no significant difference in the concentrations of trace metals between the nearshore or offshore stations (Al-Sayed et al., 1994). In the first decade of this century, nine of trace metals were studied in 23 stations distributed in the territorial water of Bahrain. This study showed that trace metals were within the Quality Standards of the United Kingdom and the United States Environmental Protection Agency (USEPA) with an exception of Cu in all sites and Hg in one site which were found to be above those standards (Juma & Al-Madany, 2008).

Trace metals will not stay forever in the seawater, they rapidly undergo sorption (absorption or adsorption) with the suspended particulate matter, then they still fall down to the marine sediment (Hedge et al., 2009). Therefore, sediments can be considered as a reservoir of trace metals in the marine environment (Ruilian et al., 2008).

In Bahrain, the industrial activities are found to have an impact on the trace metals levels in the marine sediment. Akhter and Al-Jowder (1997) indicated that elevated levels of trace metals (Pb, Zn, Cd, Ni, Cu, and V) were found in sediments of 19 stations around the coastline of Bahrain, with remarkable higher levels of Zn and Pb. Similar results were found in 2002, when de Mora *et al.* (2004) found that the area close to a discharge point of the oil industry on the eastern coastline in Bahrain is suffering from high concentrations of Cu, Hg, Pb, and Zn. Furthermore, the same station was reported also in 2005 as the highest contaminated station by petroleum hydrocarbons (PHs) in the Arabian Gulf; therefore, the contamination in this station can be tagged as chronic (de Mora *et al.*, 2010).

Sediment is not the last stage for the trace metals in the marine environment, the bioavailable part of the trace metals can be transferred

and accumulated in the tissues of living organisms (Al-Madany et al., 1996). In Bahrain, the trace metals pollution was studied in algae, molluscs, fish, shellfish and recently in mangrove plants. Ali-Mohamed (2002) found that *Hormophysa triquetra* algae is a good indicator for trace metals pollution in Bahrain. That study indicated that the trace metals (Zn, Cu, Cd, Ni, Pb, and Cr) levels in Bahraini algae were found to be higher than the levels of these trace metal in the Atlantic Ocean and the Pacific Ocean. Pearl oyster (*Pinctada radiata*) flesh was studied for the trace metal contents and the result showed a high concentration of both Pb and Cd which were (1.8-14.0 µg/g wet weight) and (0.4-3.8 µg/g wet weight), respectively (Al-Sayed et al., 1994). Moreover, after 10 years de Mora *et al.* (2004) publish a study for the samples collected in 2000, they found high concentrations of trace metals in pearl oyster flesh samples that were collected from the eastern coast of Bahrain, close to the oil refinery in Bahrain. Tissues of marine snail (*Turbo coronatus* (Gmelin)) were studied for their contents of (Fe, Ni, Zn, Mn, Pb, Cu, Cd) in five nearshore stations. Results of that study showed that there were significant spatial and temporal variations in the levels of trace metals between the stations (Al-Sayed & Dairi, 2006). Recently, mangrove (*Avicennia marina*) leaves were analyzed for their contents of Cr, Cu, Fe, Mn, Mo, Ni, Pb, and Zn in three stations in Tubli Bay, and the results showed a high level of Cu (Abou Seedo et al., 2017). Al-Madany et al. (1996) studied the trace metals (Pb, Cd, Hg, and As) in fish and shellfish consumed in Bahrain, the results showed that the trace metal contents of shellfish were greater than fish. That study indicated that the contents of Pb and Cd in fish tissue found to be higher than their concentrations in the other areas within the Arabian Gulf, where the Hg and As were found to be less. Furthermore, the shellfish, shrimp, crabs and shovel lobsters were studied and the results showed the increasing order of Pb, Cd and Hg levels in these three species:

shrimp < crabs < shovel lobsters. However, the order of As was shrimp < shovel lobsters < crabs. Furthermore, Musaiger and Al-Rumaidh (2005) reported a low level of Pb, Hg, and Cd in the flesh of blue swimming crab (*Portunus pelagicus*) in Bahrain.

Studies conducted on the trace metal content in blue swimming crab (BSC) in Bahrain are limited. However, in other parts of the Arabian Gulf there were some studies, most of them in the northern part of the Arabian Gulf. This species found to have the ability to accumulate and magnify organic and inorganic pollutants in its body (Potter et al., 1983). This is because BSC feeds on a variety of bottom-dwelling animals even the debris (Monikh et al., 2014). Sarasiab and Hosseini (2014) studied the contamination of polychlorinated biphenyl, mercury and methyl mercury in different organs of BSC in Iran. Their results showed that hepatopancreas accumulated the largest amount of the contaminants, followed by the gills and the muscles. Also they reported a direct proportion between the size of the crabs and the concentration of these pollutants. Moreover, the crab's females were found to accumulate more pollutants than males. In contrast, Al-Mohanna and Subrahmanyam (2001) used the BSC to investigate the impact of oil spillage after the Gulf War 1990 in Kuwait's marine environment, the results showed that muscles accumulated the largest amounts of trace metals rather than the other organs, and both Zn and Cu were found to be the highest trace metals in the crabs tissue.

The BSC in the Arabian Gulf was found to have a high level of As in the Iranian coast of the Arabian Gulf (Heidarieh et al., 2013). The levels of trace metals were found to be affected by the habitat where the BSC live or feed (Abdolvand et al., 2014; Rahmanpour et al., 2014).

1.2 Research Problem

Trace metals are one of the pollutants in the marine environment associated with anthropogenic sources (Ruilian et al., 2008). In Bahrain, these anthropogenic sources are present in form of sewage discharges, industrial effluents, desalination plants, oil pollution; in addition to, reclamation and dredging activities (Khadija Zainal et al., 2008; Naser, 2013). However, it could be also resulted due to transboundary effect.

Trace metals in the marine environment eventually settle into marine sediments. The bioavailable part can be accumulated in different tissues of living organisms (Reynolds, 1993). Some trace metals may pose a threat to the life of marine organisms such as Hg, Cd, and Pb due to their toxicity (Valavanidis & Vlachogianni, 2010). Additionally, even though some metals are essential for many marine organisms such as Zn and Cu, their presence at high levels may pose toxicity and threat for many marine organisms (Goyer & Clarkson, 1996; Mason, 2013). Trace metals adverse effects are not limit to the environment, but also extend to the human through the bioaccumulation through food chain (Stankovic & Stankovic, 2013; Jaishankar et al., 2014).

Blue swimming crab is one of the marine organisms that commonly found on the whole coast of the Arabian Gulf (Monikh et al., 2014; Naderloo, 2017), and it is one of the foodstuff in Bahrain. This species has the ability to accumulate trace metals in its tissues and organs (Al-Mohanna & Subrahmanyam, 2001), so it can be used as a bioindicator for marine pollution (Monikh et al., 2014). Hence, it can be used to measure the trace metals levels in different Bahrain's coasts.

1.3 Research Questions

This study will try to find answers to the following questions regarding to marine pollution in Bahrain:

1. What is the level of the trace metal ions (Pb^{2+} , Cd^{2+} , Cu^{2+} , Cr^{3+} , Zn^{2+} , and Ni^{2+}) in the white and brown meats of BSC in different marine areas in Bahrain?
2. Is their significant difference in the trace metal ions (Cu^{2+} , Zn^{2+} , and Cd^{2+}) levels of BSC based on spatial variations in Bahrain?
3. Is there a significant difference in the concentrations of the trace metal ions (Cu^{2+} , Zn^{2+} , and Cd^{2+}) of white and brown meats of the BSC in Bahraini seawater?
4. Is there a significant difference in the concentrations of the trace metal ions (Cu^{2+} , Zn^{2+} , and Cd^{2+}) of males and females of the BSC in Bahraini seawater?
5. Are the levels of the selected trace metal ions (Pb^{2+} and Cd^{2+}) in the tissues of BSC, in Bahraini seawater, compatible with the national and international standards (GCC Standardization Organization "GSO" and European Commission "EU")?

1.4 Research Objectives

This study will attempt to achieve the following objectives in order to answer the above study questions:

1. Identify the levels of trace metal ions (Pb^{2+} , Cd^{2+} , Cu^{2+} , Cr^{3+} , Zn^{2+} , and Ni^{2+}) in the white and brown meats of BSC in different marine areas in Bahrain.
2. Investigate the spatial variation on the concentrations of the trace metal ions (Cu^{2+} , Zn^{2+} , and Cd^{2+}) in the BSC in Bahraini seawater.

3. Investigate the influence of tissue type on the concentrations of the trace metal ions (Cu^{2+} , Zn^{2+} , and Cd^{2+}) in the BSC in Bahraini seawater.
4. Investigate the influence of gender on the concentrations of the trace metal ions (Cu^{2+} , Zn^{2+} , and Cd^{2+}) in the BSC in Bahraini seawater.
5. Investigate the compatibility of the levels of the trace metal ions (Pb^{2+} and Cd^{2+}) in the BSC tissues in comparison with national and international standards (e.g. GSO and EU).

1.5 Research Hypotheses

The study will test the following hypotheses:

1. There are significant differences between the trace metal ions levels (Cu^{2+} , Zn^{2+} , and Cd^{2+}) of BSC based on spatial variations in Bahraini seawater.
2. There are significant differences between the concentrations of the trace metal ions (Cu^{2+} , Zn^{2+} , and Cd^{2+}) of white and brown meats of the BSC in Bahraini seawater.
3. There are significant differences between the concentrations of the trace metal ions (Cu^{2+} , Zn^{2+} , and Cd^{2+}) of males and females of the BSC in Bahraini seawater.

1.6 Research Significance

The marine environment in Bahrain, as part of the Arabian Gulf, is considered a fragile environment due to the high water temperature, high salinity, high evaporation rate, shallow water and poor water flushing (Sheppard et al., 2010). The slow water circulation process in the Arabian Gulf takes 3-5 years (Reynolds, 1993). This makes the dilution process of pollutants a slow process, which increases their residing time that leads to

an increase in their chances to accumulate at the bottom of the Gulf (Freije, 2015). These conditions keep the marine organisms living close to their tolerance limits, which means they will be very sensitive to any additional pressures related to the pollutions (Sheppard et al., 2010).

Pollutants like trace metals, especially non-essential metals, as well as high level of essential metals, can cause adverse effects on the marine environment, marine organisms, ecosystem health, and biodiversity (Al-Busaidi et al., 2011). Therefore, the additional pressures of trace metals in this fragile environment can be critical for both the environment and the humans' health (Sheppard et al., 2010). Consequently, it is very important to monitor continuously the trace metals levels in all components of the marine environment in Bahrain.

This study has significant implications on the environmental, human health and scientific research fields. Environmentally, this study can give an evidence about the trace metals level in the marine environment in Bahrain, which can identify whether these trace metals are present in normal levels in different marine areas in Bahrain or it is not. Also, it can detects the impact of industrial activities on the eastern coastline of Bahrain. On the other hand, there are limited studies conducted on pollutants in the crabs in Bahrain's marine environment. This study may contribute to the achievement of target number 14.1 of the SDGs by giving the decision-makers clear evidence of the level of trace metals in the marine environment. This can help in taking decisions to reduce or prevent the sources of these trace metal ions in the marine environment in Bahrain.

This study has implications on human health, because BSC is one of the seafood lists in Bahraini dishes. Finding out the trace metal ions levels in the tissues of BSC can help the decision-maker to pass legislation that regulates fishing or eating this kind of food to avoid the transmission of trace elements to the human body, especially toxic elements such as Cd^{2+} and Pb^{2+} .

The excessive consumption of foodstuff that contains a high level of trace metals may lead to adverse effects on humans' health. Therefore, monitoring the trace metals in marine organisms became more important because these organisms can accumulate trace metals in their different tissues (Lichtfouse et al., 2011).

For the scientific research field, this study could fill a knowledge gap in our understanding of trace metals concentration in BSC in Bahrain, as there are very limited old studies in this regard. The first study conducted by Al-Madany et al. (1996), were four non-essential elements studied (Cd, Pb, Hg and As) in the edible tissue only. The samples were collected randomly from all fishing locations around Bahrain. No differences were studied among the gender and the tissues of BSC. The second study conducted by Musaiger and Al-Rumaidh (2005), were six trace metals studied (Zn, Cu, Cd, Pb, Hg and Fe) in the edible tissue only for male and female crabs. That study did not investigate the differences among the tissues of BSC and the locations.

1.7 Definition of Terms

- **Bioaccumulation:** The net accumulation of the metal (or other organic or inorganic pollutants) in the tissues of the organisms (or part of it) after exposures to this metal (Mason, 2013).
- **Bioavailable Metals:** The metals that can be taken up by the organisms, mainly in the ionic form or the organometallic form. Accordingly, the metals that are in the unavailable form (not bioavailable) does not represent any toxicity for the organisms, even if it is toxic in nature. Therefore, the total level of metals in seawater or sediment does not represent metal's biological toxicity (Roane et al., 2015).

- **Bioindicators:** *"An organism used as an indicator of the quality of an ecosystem, especially in terms of pollution"* (Lexico, 2020). Organisms that are sensitive to their surrounding environment, and have the ability to reflect the situation of the ecosystem's health. Also, they have the ability to accumulate pollutants in their body for a long time (Stankovic & Stankovic, 2013).
- **Crab's Brown Meat:** The tissues of the crabs' carapace (the cephalothorax). It is everything in the carapace excluding the gills, which can be represented by crab's digestive organ (the hepatopancreas) (Bolam & Bersuder, 2013; Ariano et al., 2015). The muscles inside the carapace that hold the appendages are excluded from this study.
- **Crab's White Meat:** The edible tissues of the crabs' appendages (legs and claws) (Bolam & Bersuder, 2013; Ariano et al., 2015).
- **Essential Metals:** Metals that have at least one known essential function in the organisms, and presents in low and specific concentration range. They can become toxic at high levels. Further, declines in their concentration can leads to an adverse effect on the organism's functions (Mason, 2013). Organisms required these metals for many functions such as enzymes and protein synthesis, osmotic pressure control, and molecule transport (Roane et al., 2015).
- **Marine Pollution:** Change in the chemical, physical or biological characteristics of the marine environment due to introducing contaminants (compounds or energy) by humans. This change threatens marine organisms and affects the quality of the marine environment, which leads to reduction in the productivity of the ecosystems. Contaminants can be in form of direct effluent discharge (introducing toxic chemicals or changing the physical

properties of the sea, e.g. temperature), human activities (shipping), and solid wastes. (Noone et al., 2013).

- **Non-essential Metals:** Metals that have no known biological function in the organism's body and their presence leads to adverse effects on the organism's functions (Mason, 2013). Their toxicity takes place by substitution the essential metal's normal binding sites by another metal which disable the biological functions, e.g. displacement Zn^{2+} by Cd^{2+} leads to inhibit enzymatic functioning (Roane et al., 2015).
- **Trace Metals:** Metals present in the environment (water, soil, organism's tissue, plants, etc.) at very low concentration, mostly in ppm level or less (Sadiq, 1992; Duffus, 2002).

This study adopted the term *Trace Metal*, but many studies in the environmental field use the term *Heavy Metals*. Most environmentalists are usually using the term *heavy metals* to give a negative meaning to the elements (Hodson, 2004), even those are essential elements like Zn^{2+} or Cu^{2+} . Duffus (2002) considered that the term *heavy metals* is in fact a meaningless and misleading in an IUPAC Technical Report published in Pure and Applied Chemistry (the official IUPAC journal). Furthermore, other studies end up with the same conclusion which considered *heavy metal* as a poorly defined and loose term (Hodson, 2004; Pourret & Bollinger, 2018; Pourret & Hursthouse, 2019; Pourret et al., 2021). There are many definitions for *heavy metal* based on metal density, atomic weight, or atomic number. As an example of the poorly defined, Sc is considered a *heavy metal based* on the atomic number, but based on the definition that used the density, it is not *heavy metal* (Duffus, 2002). Moreover, As is considered a *heavy metal* but it is not metal, As is metalloid (Hodson, 2004).

Chapter Two

Theoretical Framework and Literature Review

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2.1 Trace Metals: Their Importance and Impacts on the Organisms

Essential trace metals are important for the life of the organisms due to their biological function in the organisms' body, but when their level exceeds the natural limit, they represent a threat to the organism's life (Chattopadhyaya et al., 2014). For instance, Cu and Zn represent the main part of numerous enzymes and proteins, also regulating different biochemical functions (Golovanova, 2008). For example, they are essential for photosynthesis in plants and phytoplankton (Mason, 2013). Nickel is present in the central sites of many enzymes such as urease, hydrogenase, methyl-coenzyme M reductase, superoxide dismutase, carbon monoxide dehydrogenase and acetyl-coenzyme A synthase (Ermler et al., 1998; Boer et al., 2014). Chromium (Cr^{3+}) is an essential for the controlling glucose, insulin and lipids in blood (Goyer & Clarkson, 1996).

Copper metal is an essential for all organisms, and its adverse effect to the human health can be caused by both the excess and the deficiency (Stern et al., 2007). Copper, as part of enzymes, play important roles in oxidation–reduction reactions in human body (WHO, 1996). There are more than 15 enzymes that have function based on Cu redox activity (Stern et al., 2007). For example, these enzymes involved formation of hemoglobin and cross-linking of hair keratin (Tchounwou et al., 2012). The deficiency of the Cu can cause Menkes disease. Excess intake of Cu can cause anemia when the Cu interfering Fe, and also it causes Wilson disease. However, there is no Cu toxicity for general population were recorded (Stern et al., 2007).

Zinc is essential for life and necessary for immune system (Mehri & Marjan, 2013). Zinc essentiality comes from the roles it plays in the formation of numerous enzymes in the human body, as well as involvement in the synthesis of proteins, carbohydrates, nucleic acids and lipids (WHO, 1996). Severe Zn deficiency can affect many organs in human body, for example, gastrointestinal, immune, epidermal, skeletal, central nervous and reproductive systems (Hambidge, 2000). Also, a severe deficiency of Zn leads to growth retardation (Mehri & Marjan, 2013). Toxicity due to an excess of Zn level in the body is uncommon, but when it happens, Zn interferences with the Cu metabolism or other trace metals in the body (WHO, 1996; Plum et al., 2010).

Nickel is an essential metal for many organisms and humans (Bencko, 1983), as other essential trace metals, nickel adverse effect can be caused by both the excess and the deficiency (Cempel & Nikel, 2006). Nickel cause dermatitis when reacting with the immune system (Sigel et al., 2013). The most toxic nickel compounds are nickel sulfide, nickel oxide and nickel carbonyl (Bencko, 1983). Nickel can enter the human body through two routes, inhalation or food chain. Inhalation of nickel compounds, especially in the occupational settings, can affect the respiratory tract, for example causing cancer for the nasal cavity, larynx and lungs, as well as affect the immune system (Bencko, 1983; Cempel & Nikel, 2006). Excessive exposure to nickel compounds can impairs the normal homeostasis of some essential metals in the body like Ca, Mg, Mn, Zn (Roussel et al., 2006). Whereas, chronic exposure to nickel's metal can lead to serious diseases in the kidney, cardiovascular and respiratory systems (Schiffer et al., 1991).

Chromium present in the marine environment mostly as Cr^{3+} and Cr^{6+} states (Monalisa & Kumar, 2013). It releases to the marine environment via fertilizers run off, sewage effluent or many industries effluent e.g.

electroplating, paint and pigment formation (Ghani, 2011). Chromium (III) is an essential metal for human which is important for glucose metabolism process (Scawarz & Mertz, 1959; Krejpcio, 2001; Lewicki et al., 2014). Whereas Cr^{6+} is highly toxic to human and known to be carcinogens. The respiratory system is affected by excessive exposure to Cr^{6+} , as it causes nose ulcers, shortness of breath, asthma and cough. Additionally, exposure to a high level of Cr^{6+} can cause damage to nerve tissues, liver, kidney, skin ulcers and skin irritation (Martin & Griswold, 2009).

Non-essential trace metals, such as Pb^{2+} , Cd^{2+} and Hg^{2+} , are toxic and cause harm even in low concentrations (Chattopadhyaya et al., 2014). No known biological function for Cd^{2+} (Golovanova, 2008). The toxicity of these minerals represented by deactivation of enzymes and distortion of proteins by competing or replacing the essential metals in the enzymes and proteins (Lichtfouse et al., 2011).

Trace metals have an adverse impact on marine ecosystems and different marine organisms (Jakimska et al., 2011), and it can affect the human health too (Dissanayake & Chandrajith, 2009; Tchounwou et al., 2012). For example, Cd is a toxic non-essential transition metal that poses a health risk for both human and animals (Genchi et al., 2020), and found to has negative effects on the kidney and the reproductive system of males. Other effects of Cd exposure are disturbances of calcium metabolism, hypercalciuria and formation of stones in the kidney. High exposure can lead to lung cancer and prostate cancer (Nordic Council of Ministers, 2003). Cadmium exposure occurs from the ingestion of contaminated food (e.g. crustaceans, organ meat, leafy vegetables) (Bernhoft, 2013). Whereas Pb has adverse effects on the nervous system (Shukla & Singhal, 1984).

Cadmium ion has a high toxicity due to its adverse effects on the enzymes in different organisms including human. (Irfan et al., 2013). Cadmium ion has the ability to replace the Zn ion in metallothionein

because they have the same oxidation state. This replacement can inhibit Zn metallothionein function, which is a free radical scavenger in the cells (Bhat et al., 2019), then the Cd^{2+} metallothionein accumulate in the kidney causing its damage (Siddiqui, 2010; Hambach et al., 2013; Tsutsumi et al., 2014). On the other hand, Cd^{2+} causes deficiency of Fe^{2+} by binding with cysteine, histidine and glutamate ligands (Castagnetto et al., 2002). In addition to kidney, Cd^{2+} accumulates also in other organs like lungs, liver, bones and nerve tissues and causes damage to these organs as well as malfunction (Tsutsumi et al., 2014).

Lead metal ion is highly toxic for living organisms and it can cause many adverse effects for human health (Jan et al., 2015). For example, it affects the nervous systems causing brain damage, intellectual disorders and disruption of the nervous system. Besides, hypertension, infertility, miscarriages, anemia and it causes damage to the kidney (ATSDR, 2020). Around 95% of the Pb in human body deposited in the bones as insoluble phosphate (Jan et al., 2015).

2.2 Characteristics of the Marine Environment in the Arabian Gulf

The Arabian Gulf is considered as a semi-closed sea, with one outlet, the Strait of Hormuz (which is 55 km wide at this strait). The volume of the water body of the Arabian Gulf is around $8,600 \text{ km}^3$ with an average depth around 35 m; therefore, it is considered as a shallow water (Fowler, 1993; Nadim et al., 2008; Sheppard et al., 2010). The Iranian side (Eastern) of the Arabian Gulf is deeper than the Arabian coast (Western) where the depth reach 110-160 m (Fowler, 1993; Swift, 2003).

The marine environment in the Arabian Gulf is subjected to harsh conditions represented by high temperature and salinity (Emery, 1956; Fowler, 1993; Sheppard, 1993; Coles, 2003). These extreme conditions are

the result of the arid climate in this region and the geographical nature of the Arabian Gulf (Swift, 2003; Sheppard et al., 2010). The temperature of surface seawater in the Arabian Gulf varies from 12°C in winter to 37°C in summer, but it elevates to around 40°C at some cases (Fowler, 1993; Sheppard & Loughland, 2002). The surface seawater temperature of the southern side of the Arabian Gulf is higher than the northern side by approximately 10°C. Generally, the temperature of the surface seawater became higher as one move towards the coastline, due to the impacts of land-base activities like desalination plants (Al-Rashidi et al., 2007; ROPME, 2013). There is an increase in the surface seawater temperature in the Arabian Gulf in the last 5 decades with a rate of 0.2°C/decade, but this increase became faster in the last 2 decades where the rate became 0.45°C/decade (Sheppard & Loughland, 2002).

In the same context, salinity in the Arabian Gulf is high compared with other regions (Sheppard, 1993). The salinity in the Arabian Gulf has a spatial and temporal variation, which is represented in Figure 1 (Piontkovski & Chiffings, 2014). For example, in case of temporal variation, the salinity of the seawater inflow at Strait of Hormuz that is coming from the Indian Ocean through the Arabian Sea and the Gulf of Oman is 36.5-37.0‰ (Chitrakar et al., 2019). However, the salinity of the outflow seawater from the Arabian Gulf at the Strait of Hormuz is 40.5‰ in summer and 38.5‰ in winter (Piontkovski & Chiffings, 2014). In addition, winter salinity in Arabian Gulf is greater than summer salinity, Figure 1, due to the increasing evaporation rate to the double in the winter (Swift, 2003). The reason for this is that winter possesses dry; northeast and northwesterly winds (Hunter, 1986; Johns et al., 2003).

On the other hand, in the case of spatial variation within the Arabian Gulf, the salinity changes between 38-70‰ (Fowler, 1993; Al-Saad & Salman, 2012). Extreme levels of salinity in the Arabian Gulf are located

in the shallow regions, because seawater evaporates faster as, in the Gulf of Salwah (between Qatar, Bahrain and KSA) where the salinity reaches 70‰ (Sheppard et al., 1992; Kämpf & Sadrinasab, 2006; Al-Saad & Salman, 2012; ROPME, 2013). In contrast, lower salinity is observed in the Shatt Al-Arab (north-western Arabian Gulf), due to the discharging of freshwater from Tigris and Euphrates Rivers (Kämpf & Sadrinasab, 2006).

This high level of the salinity in the Arabian Gulf is caused by several reasons, firstly, the evaporation rate is higher than the freshwater's inflow into Arabian Gulf from both rivers runoff and precipitation (Sheppard, 1993; Al-Saad & Salman, 2012). Secondly, the Arabian Gulf is a shallow area, which means it evaporates faster by high temperature (Al-Saad & Salman, 2012). Thirdly, the dry winds in winter (Hunter, 1986). Fourthly, the slow circulation of the seawater in the Arabian Gulf (Sheppard et al., 1992; Barth & Khan, 2008; Nadim et al., 2008).

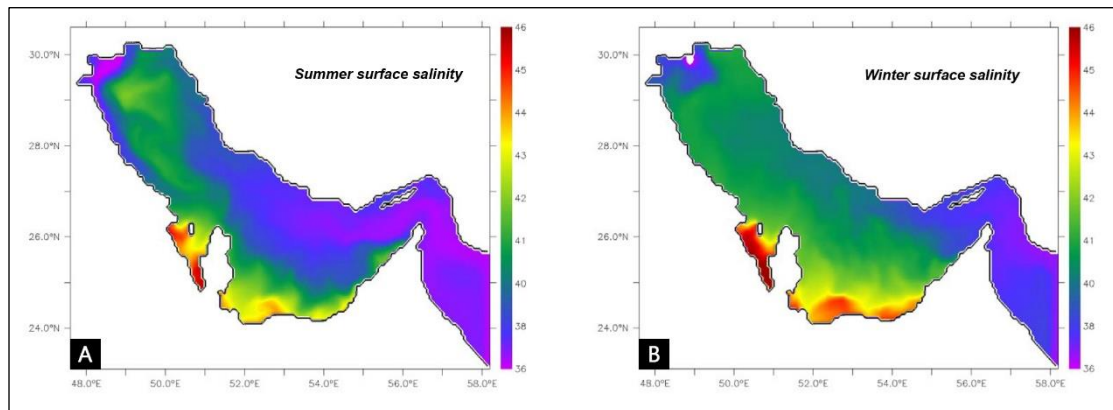


Figure 1 Spatial and temporal variation of the salinity within Arabian Gulf. (Kämpf & Sadrinasab, 2006)

The issue of water circulation in the Arabian Gulf plays an important role in increasing salinity as well as the concentration of pollutants in this semi-closed sea (Barth & Khan, 2008; Nadim et al., 2008). As explained by Sheppard et al. (2010) in Figure 2, the waters enter the Arabian Gulf from the Indian Ocean through the Strait of Hormuz, where the water entering is at the top and then moves along the Iranian coast in a counter-

clockwise movement. When the water moves to the southern coast of the Arabian Gulf, its density increases due to high salinity in the shallow areas. This saline water sinks to the bottom during its exit from the Strait of Hormuz, where Reynolds (1993) reported this process takes 3-5 years. This circulation is driven by the differences in the salinity within the Arabian Gulf seawater (Sheppard et al., 2010).

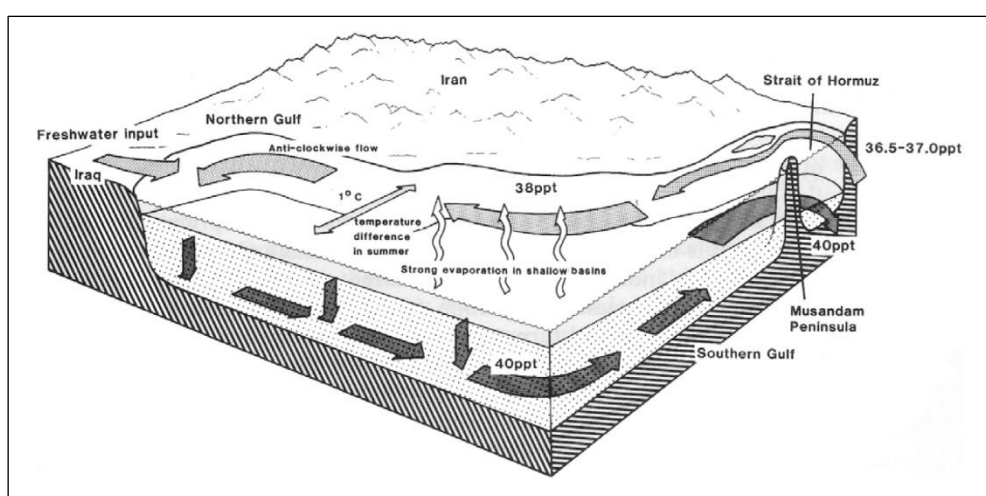


Figure 2 Circulation pattern of the Arabian Gulf. (Sheppard et al., 1992)

2.3 Marine Pollution in the Arabian Gulf

The natural characteristics of the Arabian Gulf (high salinity, high water temperature, and poor water flushing) are considered harsh conditions for many marine organisms in the region and force them to live close to their tolerance limits (Price et al., 1993; Coles, 2003; de Mora et al., 2003). The life of numerous marine organisms will be threatened if additional pressures are added due to the pollutants that enter the marine environment continuously (Fowler, 1993; de Mora et al., 2003). In addition to these natural pressures; on the other hand, the anthropogenic pressures also create an impact on the marine environment in the Arabian Gulf (Naser, 2014).

The environmental impacts of human activities within the Arabian Gulf are one of the highest around the world (Halpern et al., 2008). These activities are oil spill, reclamation and dredging projects, discharging of industrial wastewater, sewage treatment water, brine waters, ballast water of tankers (ballast), oil industries, increasing human population and the urbanization (Sale et al., 2011; Naser, 2014; Chitrakar et al., 2019).

Oil spill events are considered a main cause for marine pollution (Chitrakar et al., 2019), where the Arabian Gulf seawater received approximately 13.5 million barrels of oil during the last 50 years (Zhao et al., 2015). The Arabian Gulf is exposed to oil pollution from several sources, first, around 25,000 oil tankers cross the Arabian Gulf annually (Al-Saad & Salman, 2012), loaded with more than 50% of the world's demand for crude oil (UNEP, 1999; Literathy et al., 2002). This huge number of tankers within the Arabian Gulf makes oil spill accidents inevitable (Fowler, 1993) as presented in Table 1. Second, the discharging of tank washing waters and the ballast (around 100 tons of ballast water every year (Nasab & Makizadeh, 2016)) is another source of oil pollution, which estimated by 2 million barrels every year (Al-Saad & Salman, 2012). Third, three wars took place in the Arabian Gulf, Iraq-Iran War (1980-1988), Gulf War I (1991), and Gulf War II (2003), were accompanied by the dumping the oil tankers and releasing the oil wells into the seawater (Sheppard et al., 2010; Chitrakar et al., 2019). But the Gulf War I (1991) has the largest oil released not only in this region but in the history (Fowler, 1993; Literathy, 1993). The impact of the Gulf War I (1991) was not only the spilled oil directly into the Arabian Gulf, but there were approximately 500 million barrels of oil that were ignited during 250 days. This ignited oil emits gases, soot, aerosols and toxic combustion product, which may have sank into the Arabian Gulf in the end (Fowler, 1993). In addition to the above sources, discharges of refineries and industrial leakage are other

potential sources that lead to the oil spill in Arabian Gulf (Chitrakar et al., 2019).

In general, there is no evidence for elevated levels of different pollutants in the Arabian Gulf sea area. For example, petroleum hydrocarbons (PHs), aliphatic or polycyclic aromatic hydrocarbons (PAHs), chlorinated pesticides, and polychlorinated biphenyls (PCBs) in the Arabian Gulf are within the levels of other regions. But there are some exceptions in few sites close to the industrial effluents (de Mora *et al.*, 2010). Moreover, trace metal levels within the water, sediment, and marine organisms of the Arabian Gulf were found to be within the level of other regions or less. However, high level of trace metals were found to be in the zone area of some industrial discharges (de Mora et al., 2004; Naser, 2013; Dadar et al., 2014). Even after the Gulf War I (1991), Fowler *et al.* (1993) found that the impacts of the oil spilled were localized within 400 km from the source. Elevated levels of some trace metals were found nearshore of the Kuwait and Saudi Arabia (northern coast) coastline.

Table 1 Some oil spill incidents that occurred in the Arabian Gulf.

Year	Event	Quantity (in barrels)	Reference
1980	Oil spill in the north and west coasts of Bahrain	0.2 million	(Brown & James, 1985)
1980-1988	Iraq-Iran War, by the war of the dumping of the tankers.	6-8 million	(Sheppard et al., 2010)
1983	Tanker collision at Nowruz Oil Field	6 million	(Alam, 1993)
1991	During Gulf War I, deliberately discharged of Kuwait's oil into Arabian Gulf by Iraqi Forces	7-11 million	(Alam, 1993; Sale et al., 2011)
1994	"Baynunah" and "Seki" tanker collision loaded with Iranian crude.	0.10 million	(Chitrakar et al., 2019)

2.4 Trace Metals and their Sources in Bahrain's Marine Environment

In the marine environment, trace metals pollution is one of the major threats (Naser, 2013; Al Naggar et al., 2018). Naturally, trace metals represents a part of the marine environment, but their concentrations are

increasing due to the anthropogenic activities (Al Naggar et al., 2018). The problem associated with trace metals is their ability to accumulate in the living organisms in addition to their toxicity (Purves, 2012). Unlike organic pollutants, trace elements do not biodegrade in the living organisms (Nürnberg, 1984) nor decompose in the ecosystems (Förstner & Wittmann, 1981).

The trace metals in the marine environment can be divided into two main groups, *Essential Elements* such as Cu, Zn, Fe, Mn, Co, Ni, Mo and Cr (Mason, 2013; Roane et al., 2015), and *Non-Essentials Elements* like Cd, Pb, Hg, and As (Al-Busaidi et al., 2011; Stankovic & Stankovic, 2013). The importance of essential elements lies in the fact that they are an important part of the synthesis and functioning of enzymes and regulating different biochemical functions in the organisms (Javed & Usmani, 2015). Moreover, they are used in some organisms as protein structures stabilizer, and assist the electron transfer reactions (Stankovic et al., 2014). These essential elements have limitations of their importance in the organism, when their concentration exceeds certain levels, they constitute toxicity and danger to the organism life (Mason, 2013). For example, Cu is required at low concentration for the marine organisms, but elevated concentration can cause toxicity for numerous fish, invertebrates and aquatic plants (Moore & Ramamoorthy, 2012). In the same context, there are two effects of Zn concentration on the growth of marine diatom (*Thalassiosira weissflogii*). A low Zn concentration leads to a slow rate of growth, while a higher concentration converts Zn to toxin which poses a risk to lives and causes growth to stop (Mason, 2013).

Non-essentials (polluting) elements like Pb, Cd and Hg are toxic even in low concentration (Al-Busaidi et al., 2011), whereas no biological function is known for them (Singh et al., 2011; Roane et al., 2015; Herrera-Herrera et al., 2019). The toxicity caused by non-essential elements occurs

as a result of competing and replacing the sites of the essential elements in membrane protein and enzymes (Lichtfouse et al., 2011).

When trace metals released into the marine environment from their sources, they will rapidly undergo sorption (absorption or adsorption) with the suspended particulate matter, then they fall down to the marine sediment (Hedge et al., 2009). Therefore, sediment can be considered as a reservoir of trace metals in the marine environment (Ruilian et al., 2008). Then, the bioavailable part of the trace metals can be transferred and accumulated in the tissues of living organisms (Al-Madany et al., 1996). Firstly, trace metals accumulate in the phytoplankton and thus benthic marine organisms such as benthic algae and bacteria, then they distribute in the food web. Lower trophic level organisms accumulate the trace metals from food as well as directly from water, while the higher trophic level organisms accumulate most of the trace metals from food (Mason, 2013).

Trace metals enter the marine organisms through two main routes, water-borne and diet-borne. Water-borne, which is direct route, when the gills accumulate the trace metals that are in water, adsorbed on particulate matter in the water column and adsorbed on the sediment. Diet-borne, which is indirect route, the accumulating of the trace metals that enter the organisms bodies via food (Sauliutė & Svecevičius, 2015). The bioaccumulation of trace metals in the tissues of marine organisms is affected by internal and external factors. Internal factors like the chemistry of the trace metals and the biology of the organisms e.g. feeding behavior, body size and life cycle (Gerhardt, 1993). Where the external factors like the abundance of the metals in the surrounding environment, exposure duration to these metals and the availability of the food contaminated by these metals (Lee & Lee, 2005). In addition, physical and chemical characteristics of water such as salinity, temperature, dissolved oxygen, pH and sediment grain size (Luoma, 1983; Stankovic et al., 2014). The

resulting effects from these external factors are due to their effect on the bioavailability of the trace metals (Lichtfouse et al., 2011).

Trace metals to be bioavailable for uptaking by marine organisms should be present in one of the following two forms, free ion (cationic aqueous state), and part of either organometallic compound or hydroxyl complex. This bioavailability of the trace metals is affected by the environmental conditions that were mentioned above (the external factors of bioaccumulation). Furthermore, when a trace metal forms a complex with the clay or organic matter available in the aquatic system, it will lose its bioavailability for the marine organisms, which reduces its bioaccumulation chance and toxicity by 100 times (Javed & Usmani, 2015).

Trace metals in aquatic systems have a natural origin when they leachate from the rocks and soil (Stankovic & Stankovic, 2013; Viswanathan et al., 2013), as well as anthropogenic origin (Moore & Ramamoorthy, 2012). In Bahrain, most of the anthropogenic activities are located on the eastern coastline (Khadija Zainal et al., 2008). Table 2, shows the land-based activities in Bahrain and the potential trace metals associated with its discharge water. On the other hand, the Arabian Gulf receive $60\text{--}200 \times 10^6$ tons of the dust annually from the dust storms (Sheppard et al., 2010). These aerosols found to contain trace metals in their composition (Chester & Stoner, 1975; Ali-Mohamed & Matter, 1996), which may constitute a natural source of trace elements in the Bahrain's marine environment. Ali-Mohamed and Matter (1996) studied the composition of the aerosols fall down in Bahrain (during Gulf War (I) episode) and found Zn and Cu present in significant higher level than the other metals, followed by Ni and Cr. Very low level of toxic metals were recorded, As, Cd and Pb, whereas Sb was below the detection limit and V found in a negligible concentration.

Table 2 Trace metals pollution in the marine environment associated with different land-based activities.

Activity	Potential trace metal pollution
Desalination	Cu, Fe, Ni, Cr, Zn (Miri & Chouikhi, 2005) Al, Ba, Cr, Mn, Ni, Sr, V, Zn and B* (Mohamed et al., 2005) Cu (Hoepner & Lattemann, 2003) Fe, Cu, Pb, Cd, Zn and Ni (Cooley et al., 2006)
Petroleum Refinery	Cu, Cr, Pb and Zn (Nammanga, 1976) Cu and Pb (Bersuder et al., 2020) Mn, Fe, Ni, Zn, Pb and Cu (Relic et al., 2005) Cd, Pb and Hg (Croquer et al., 2016)
Petrochemical industries	As, Mo, Ni, S*, Se*, V, and Zn (Bosco et al., 2005) Cd, Pb, As*, Sn, Ag and Mo (Keshmiri et al., 2018) Cd, Cu, Zn and Pb (Cao et al., 2020)
Metallic Smellers	Pb, Zn, Cd and In (Boughriet et al., 2007) Cd and Pb (Schintu et al., 2010) Pb, Zn, Ni, Cu, Fe, and As* (Kabir et al., 2012)
Shipbuilding & Repair Yard Boaters and Boatyards	Cd, Cu, Pb, V and Zn (Anderlini et al., 1982) Cu and Pb (Choi & Jeon, 2018) Cd, Cr, Mn, and Zn (I. Alam et al., 2019) Cu, Zn, Ba, Cd, Cr, Ni, Pb and Sn (Turner, 2010) Cr, Cu, As* and Cd (Caballero-Gallardo et al., 2020)
Sewage Treatment Plant	Cu, Zn, Ni, Co, and Cd (Bradford et al., 1975) Cu and Zn (Bersuder et al., 2020) Cu, Zn and Ni (SCE, 2020) Zn and Mn (Álvarez et al., 2002)

* B and As are metalloid, S and Se are nonmetal

2.4.1 Trace Metals in Bahrain's Seawater

Trace metals in Bahrain's marine environment were studied by many researchers, regional and governmental organizations, as presented in Table 3. The first study of trace metals in Bahrain seawater showed that the levels of Cu, Zn, Cd, Hg, and Pb metals were similar to those trace metals levels in the open seas like the oceans or the Mediterranean Sea, which may gave evidence that there was no metallic pollution in Bahrain at that time (Fowler et al., 1984).

After 10 years, trace metals in seawater were studied by Al-Sayed *et al.* (1994). Two stations were chosen to study seven trace metals (Cu, Mn, Fe, Zn, Pb, Cd, and Ni) in seawater and pearl oyster (*Pinctada radiata*), the first is nearshore and the second offshore. The study conducted in the period March 1991–March 1992, were the sample collected every 6-8 weeks. The results showed that there was no significant difference in the concentration of trace metals between the nearshore and offshore stations.

In addition, the trace metals concentration in the summer months were higher.

A wide investigation of the trace metals in the Bahrain's seawater covering a large sea area was conducted by Juma and Al-Madany (2008). Ten trace elements (As, Cd, Cu, Fe, Mn, Ni, Pb, V, Zn, and Hg) were studied in the 23 fishing areas around the territorial water of Bahrain. This study can be considered as a baseline for these trace metals in Bahrain's seawater. The results indicated that Bahrain has a good marine water quality, because the levels of the trace metals were below the thresholds of United Kingdom Quality Standards and the United States Environmental Protection Agency (USEPA) criteria. However, there was an exception of Cu in all sites and Hg in one site, which were found to be above those thresholds. In general, the level of total trace metals in the studied stations were similar to the unpolluted sea areas worldwide.

2.4.2 Trace Metals in Bahrain's Marine Sediments

With regard to trace elements in Bahrain's marine sediments, in the regional level, Bahrain participated in the studies conducted by ROPME in 1980s and 2000s proposed to test the quality of sediment, seafood and bivalves with respect to trace metals content (Fowler et al., 1993; de Mora et al., 2004). During the period 1980-1991, Bahrain participated in ROPME-IAEA contaminant screening projects for the petroleum hydrocarbon and trace elements (Ag, As, Cd, Co, Cu, Cr, Fe, Mn, Ni, Pb, V, Zn and Se) in sediment and biota within the Arabian Gulf (Fowler et al., 1993). Five sites were selected in Bahrain for sediment study; Askar and Dar Island (east coast), Zallaq, Umm Na San, and Al-Malikiya (west coast). The results indicated remarkable level of As in June 1991 in Askar, Al-Malikiya, and Dar Island, with a concentration of 29.2 µg/g, 35.2 µg/g,

and 31.8 µg/g, respectively, where the last was the highest in the study. Besides, Askar elevated level of Pb was record in the study many times, 1983, 1986 and 1991 with a concentration of 13.5 µg/g, 15.9 µg/g, and 11.0 µg/g respectively. On the other coast, Zallaq record a high level of Pb also in 1983 and 1986 with a concentration of 24.0 µg/g and 9.36 µg/g, respectively.

In the beginning of this century, Bahrain was a part of another regional study conducted by ROPME that studied the trace metal in the Arabian Gulf (de Mora et al., 2004). Four sites in Bahrain were part of that study, Jasra (west), North of Meridien Hotel (north), Askar and close to the oil refinery (east). The results indicated high levels of Cu, Hg, Pb, and Zn in the site close to the oil refinery, which were 48.3, 0.22, 99.0, and 52.2 µg/g dry weight, respectively. Two sites within that study (from 21 sites) were found to have remarkable trace metals concentration; one site in UAE and the second in Bahrain close to the refinery facility.

Nationally, trace metals in the sediments studied by The Supreme Council for Environment (SCE) via ongoing monitoring program since 2007 (SCE, 2018). Sediments were collected once per year from up to 29 sites during this period, but the number of stations were reduced to 15 from 2017 according to the Ministerial Order (1) 2017. Eight trace metals (Cu, Cr, Cd, Ni, Pb, Zn, Al, and Fe) were studied. The results showed that Cd, Pb, Zn, Cu, and Fe metal ions are significantly higher in site close to the Refinery Area than the average of whole stations. In general, the comparison of the Canadian sediment quality guidelines and the trace metals (Cd, Cr, Cu, Pb, and Zn) of the stations, indicated the level of Cd and Pb (which is toxic metals) is less than Threshold Effect Level (TEL), except for site close to the Refinery Area that have elevated level of Pb that exceeded Probable Effect Level (PEL). Copper and Cr levels exceeded the TEL in 12 and 13 stations, respectively, but none of them exceeded the

PEL. Zinc level was also less than TEL except for stations that were expected to be affected by petroleum pollutions like the jetties, and the Refinery Area.

In the same way, SCE is conducting an ongoing monitoring program for water quality within Tubli Bay since 2009 (SCE, 2020). Trace metals in the sediments were studied once every year in 15 stations, but from 2017 the stations were reduced to 10 in total, 8 of them were studied for trace metals in sediments. The sites were divided into three main zones, Zone 1 for the sites within 0-700 m from discharge point of Tubli Sewage Treatment Plant, Zone 2 for the sites within 700-2000m and Zone 3 for the sites beyond 2000m. On comparison the results to the Canadian sediment quality guideline indicated Zn, Cu and Ni levels were higher than the Probable Effect Level (PEL) on the northern side of Tubli Bay. In addition, Cr, Cd and Pb levels exceeded the Threshold Effect Level (TEL), but not reached the PEL threshold. The levels of these trace metals decreased as one moved further towards the south side of Tubli Bay (Zone 3), away from the discharge point. This indicates that the northern side of Tubli Bay (Zone 1) is directly affected by the discharge of treated sewage water.

Similar conclusion reported by Abou Seedo *et al.* (2017), where the levels of Cr, Cu, Fe, Mn, Ni, Pb, and Zn in the sediments from the site close to the wastewater discharge were higher than other sites in the southern part of Tubli Bay. Furthermore, the second site (Sitra 1) it was found to have higher levels of Cr, Cu, Fe, Mn, and Ni than the third site (the farthest site from the discharge of Tubli Sewage Treatment Plant). The levels of tested trace metals were found to be within the international levels with an exception of Cu.

Akhter and Al-Jowder (1997) studied Pb, Zn, Cd, Ni, Cu, and V in 19 stations around Bahrain sea shores. The results showed higher levels of Pb and Zn compared to other elements studied. The decreasing order of the six

trace metal ions was $Pb > Zn > Ni > V > Cu > Cd$, with an average concentrations of 111, 104, 40.5, 21.9, 15.1, and 1.17 $\mu\text{g/g}$ respectively. Moreover, the levels of trace metals in the sites close to the industrial activities were found to be higher than the others.

Recently, Bersuder *et al.* (2020) studied the organic and inorganic contaminants in the sediments of 34 sites around Bahrain sea shores in 2017 and 2019. Eight trace elements ions were studied, the following sequence were found: $Zn > Cu > Cr > Ni > Pb > As > Cd > Hg$. The results indicated a clean environment in general, but there were three hotspots for some trace metals in Bahrain sea shores. For example, in Tubli Bay, elevated levels of Zn and Cu were recorded, especially on the Northern side of the bay. Also, around the Shipbuilding & Repair Yard, Zn and Cu were high, but Cu had more distribution area. The third hotspot was around the Refinery area, which had elevated level of Cu and Pb.

2.4.3 Trace Metals in Bahrain's Marine Organisms

Like in sediment, trace metals in marine organisms studied by researchers, regional, and national monitoring programs. Regionally, Bahrain participated in the studies conducted by ROPME in 1980s and 2000s proposed to test the quality of sediments, seafoods and bivalves with respect to trace metals content (Fowler *et al.*, 1993; de Mora *et al.*, 2004). During the period 1980-1991, Bahrain participated in ROPME-IAEA contaminant screening projects for the petroleum hydrocarbon and trace elements (Ag, As, Cd, Co, Cu, Cr, Fe, Mn, Ni, Pb, V, Zn and Se) in sediment and biota within the Arabian Gulf. Fowler *et al.* (1993) selected three sites in Bahrain for bivalves study; Askar (Rock Scallops, *Spondylus sp.*), Zallaq (pearl oyster, *P. radiata*), and Al-Malikiya (pearl oyster, *P. radiata*). The Pb concentration in the rock scallops was 1.2-7.2 $\mu\text{g/g}$,

during 1983-1986, but in 1991 (after the Gulf War I) its concentration elevated to 37.7 µg/g. On the other hand, high level of Zn were reported for the samples collected in June 1991, in three sites, Askar, Zallaq, and Al-Malikiya respectively: 461 µg/g, 898 µg/g, 1607 µg/g. During same study, trace elements in the fish tissues were examined in 4 sites in 1991, Fasht Al Jarim, Bandar Dar, Fasht Al Dibal, and Zallaq. Four fish species were studied greasy grouper, grouper, Two-banded Porgy, and Pigface Bream, which are locally known as Hamoor, Burtom, Fasker, and Sheiry, respectively. Very low levels of Cd and Pb were found in whole sites (which were not detected in the most of samples). The level of As (metalloid) was 53 µg/g in Hamoor fish from Fasht al Jarim, which was the highest between whole sites in that study.

Again, ROPME conducted a monitoring program for the trace elements contamination within the Arabian Gulf during the first decade of this century (de Mora et al., 2004). Trace metals content of the mussels and liver of orange spotted grouper fish (*Epinephelus coioides*, Hamoor) were examined in two stations in Bahrain during this study (Badaiya and Fasht Al-Adham). In addition, pearl oyster (*P. radiata*) as bivalves was collected from North of Meridien Hotel and off the refinery sites. The results showed elevated levels of Zn, V and Pb in pearl oyster tissues collected from off the refinery site where the concentrations were 4290 µg/g, 7.3 µg/g, and 3.92 µg/g, respectively. This is in the same context of elevated level of Zn and Pb in the sediment of same site that have been discussed above. On the other hand, As (metalloid) was reported as high concentration (related to the weight (size) of the fish) in one orange spotted grouper collected from Fasht al-Adham (east coast); 14.4 µg/g. However, according to the international standards, this level is considered low. Generally, the trace metals content of the livers found to be higher than muscles in case of V, Mn, Fe, Co, Ni, Cu, Zn, Se (nonmetal), Ag, Cd, and U. However, in case

of As (metalloid), Cr, Pb and Hg trace elements were found to be more in the muscles than the liver.

Researchers studied the trace metals pollution in many different organisms in Bahrain like algae, molluscs, fish, shellfish, and recently in the mangrove. Pearl oyster (*P. radiata*) was studied during March 1991 to March 1992 by Al-Sayed *et al.* (1994). Samples were collected every 6-8 weeks from two sites, first in nearshore in the north of Manama which received a discharge treated and untreated sewage water. The second site is offshore in the north east coast. The results showed site 1 had Ni, Mn, and Fe more than site 2. However, site 2 had Cd, Pb, and Zn more than site 1. The concentrations of Pb and Cd were found higher than the WHO level of some marine organisms. On the other hand, Cd had lower concentration than Cd level in the Arabian Gulf reported in Oman and UAE.

Seven trace metals were studied in the tissues of grouper fish (*E. coioides*) collected from four sites during the period 1990-1991 (Al-Sayed *et al.*, 1996). The results showed similar pattern of the trace metal ions in the four sites $Zn > Pb > Ni > Cu > Fe > Mn > Cd$ with a total mean concentration equal to 499, 10.8, 4.4, 3.9, 3.2, 2.2, and 1.2 $\mu\text{g/g}$ wet weight, respectively. Zinc found to have a significantly higher concentration than the other metals, and it exceeded the WHO the provisional tolerable weekly intake limit for fish in addition to Cd. There was no significant difference in the concentrations of Pb, Cu, Fe, and Mn within four sites. In contrast, site 2 found to have significantly higher Cd and Ni than the other sites. In addition, Zn in site 1 and 3 were found to have a significantly higher concentration than site 2 and 4, which may indicated the impact in site 1 and 2 by the dredging and reclamation activities that took place on the coastline. Furthermore, the results indicated the significant elevated level of Cu, Mn, Zn, and Cd in the summer compared with other seasons in some

sites. Besides that, strong correlations were found between Cu-Mn, Cu-Fe, and Mn-Fe within all sites.

Al-Madany et al. (1996) studied the content of Cd, Pb, Hg and As in 26 marine species; 23 fish and 3 shellfish species representing the common edible species in Bahrain. Total of 162 fish and shellfish were collected randomly from the fishing locations around Bahrain. The results showed that the overall average levels of As, Pb, Hg, and Cd in the shellfish were more than the content in the fish, but they had approximately same pattern. The concentrations were 1.7, 0.132, 0.084, and 0.032 in fish, while it was 3.61, 0.149, 0.042, and 0.045 µg/g wet weight in shellfish, respectively. The samples under this study were found to be suitable for human consumption from the perspective of the provisional tolerable weekly intake limits (PTWI) compared to WHO guidelines. The results did not indicate a clear significant differences according to the coasts around Bahrain.

Ali-Mohamed (2002) studied the level of Zn, Cu, Cd, Ni, Pb, and Cr in common marine algae (*Hormophysa triquetra*), samples were collected from different coastline around Bahrain (Duraz, Zallaq, Sitra, and Al-Jazair Beach). The decreasing order of the overall mean concentration was Zn > Pb > Ni > Cu > Cd > Cr with a mean concentration of 43.76, 31.49 8.79, 4.85, 1.89, and 0.66 µg/g dry weight, respectively. The concentrations of Zn and Pb were found to be close, but Zn found to be higher than Pb in all sites except Duraz coastline that had Pb more than Zn. Also, the levels of Cu and Ni were found within the same range with an exception of Al-Jazair Beach which had elevated level of Cu and Ni than other sites. The remarkable result in this study was recorded in Sitra site (where most the land-based activities located) which had not record any elevated level of any metal under this study. Al-Jazair Beach coastline recorded the highest level of Cu, Ni, and Cd, while Duraz coastline recorded highest level of Pb

and Cr, and the highest level of Zn was recorded in Zallaq coastline. The second part of that study was for investigating Cu, Pb, Cd, Zn, Mn and Ni in the 6 different algae (*sargassum*, *Fucus spiralis*, *Padina Pavonica*, *Ulva* (sea lettuce), *Enteromorpha intestinalis*) within another 4 sites (Zallaq, Sanabis, Manama, and Sitra). The remarkable point found in this part that, Cd and Ni were not detected in whole samples and in whole sites; in addition, Pb were detected only in two samples, *Padina Pavonica* (Sanabis coastline) and *Enteromorpha intestinalis* (Sitra coastline) with a concentration of 7.25 and 9.5 µg/g dry weight, respectively. There was no fixed pattern for the level of trace metals within the sites or species in this part. Zallaq coastline recorded the significantly highest level of Cu in *Fucus spiralis* with a concentration of 137.5 µg/g dry weight.

The content of Fe, Zn, Cu, Zn, Cd, Pb, and Hg were studied in the muscles of male and female of BSC in two sites in Bahrain, inshore and offshore (Musaiger & Al-Rumaidh, 2005). Results did not indicate differences between the levels of trace metals in male and female crabs. Zinc found to be significantly higher than the other trace metals and Cd was not detected in the study. The pattern of the trace metals in decreasing order was: Zn > Cu > Fe > Pb > Hg > Cd. There was no significant difference between the trace metals content of male and female reported, with an exception of Pb which was found to be in male more than female by 10 times. The female and male levels of Zn, Cu, Fe, Pb, and Hg were as follows: 2.6, 0.6, 0.4, 0.04, and 0.03 mg/100g wet weight, and 2.3, 0.6, 0.35, 0.4, 0.04 mg/100g wet weight, respectively.

Tissues of marine snail (*Turbo coronatus* (Gmelin)) were studied for the contents of (Fe, Ni, Zn, Mn, Pb, Cu, and Cd) in 5 stations in Bahrain by Al-Sayed and Dairi (2006). The results showed that there were significant spatial and temporal variations in the levels of trace metals between the stations. The elevated level of Zn was in September and Cd

elevated in March, which might be related with the spawning season. The highest level of Cd was reported in Fasht Al-Jarim site while the Jazirat Al-Shaikh (Askar) site recorded the highest level of Zn.

Nine of the most common consumed seafood stuff in Bahrain were tested for the proximate, mineral and trace metals content by Musaiger and D'Souza (2008). Eight fish species (*Seriolina nigrofasciata*, *Siganus canaliculatus*, *Rhabdosargus haffara*, *Scomberomorus commerson*, *Liza alata*, *Epinephelus areolatus*, *Plectorhinchus sordidus*, and *Lethrinus nebulosus*) and one shrimp species (*Penaeus semisilcatus*) were collected randomly from central market in Manama. Results showed very low content of toxic trace metals Pb, Hg and Cd, where the Cd detected only in *Rhabdosargus haffara* fish at 0.03 µg/g wet weight. Moreover, Pb and Hg were below the limit of detection in many species, *Liza alata* found to contain highest level of Pb and *E. areolatus* recorded the highest level of Hg, which were 0.50 and 0.20 µg/g wet weight, respectively. In contrast, Pb, Hg, and Cd were not detected in shrimp (*P. semisilcatus*) species. But Cu was found to be highest in shrimp with a concentration of 2.8 mg/100g wet weight. *R. haffara* found to have the highest level of Zn and Fe, where the concentrations were 2.2 and 1.1 mg/100g wet weight, respectively.

Total mercury (T-Hg) and methyl mercury (MeHg) were tested in five fish species in Bahrain, *S. commerson*, *S. analiculatus*, *E. coioides*, *L. nedulosus* and *Sardinella albella* (Freije & Awadh, 2009). *S. commerson* found to contain the highest level of T-Hg and MeHg with a concentration of 0.126 and 0.123 mg/kg wet weight, respectively, which were less than the international limits (0.5 and 0.3 mg/kg wet weight, respectively). Methyl mercury found to represent 94.6% of the total mercury in the tissues of the fish in that study.

Trace metals content of the mangrove trees were studied within Tubli Bay by Abou Seedo *et al.* (2017) and Almahasheer (2019). Abou Seedo *et*

al. (2017) studied the impact of discharging treated sewage water on the levels of trace metals (Cr, Cu, Fe, Mn, Mo, Ni, Pb, and Zn) in Tubli Bay. Three sites were selected, one close to the discharge point of Tubli Sewage Treatment Plant, and the other two sites far in the southern side of Tubli bay. Unlike the sediment, gray mangrove (*A. marina*) leaves did not show a significant difference in most of the trace metals tested, exception Zn, which found to be highest in the site close to the discharge point. Moreover, Mo found to be the highest in the second site (Sitra (1) site). The overall mean order of the trace metals was found to be as $Fe > Zn > Mn > Mo > Cu > Ni > Cr$. After one year, Almahasheer (2019) studied the content of 13 metals in the leaves and branches of gray mangrove (*Avicennia marina*) within Tubli Bay. Three stations were selected, one close to the discharge point of sewage treatment plant and two in the southern side of the bay, each the samples were collected from 10 sites. The results showed that the leaves accumulated trace metals more than the branches, with the exception of Ag, Al, and Cu which accumulated more in branches. No significant differences were observed in the trace metals content of leaves and branches within the stations in Tubli Bay. The decreasing order of the elements in leaves and branches was as following $As > Pb > Mo > Fe > Al > Cd > Ni > Ag > Cr > Cu > Zn > V > Mn$ and $As > Al > Pb > Mo > Fe > Ag > Cd > Cu > Cr > Zn > V > Mn$, respectively. The levels of the studied trace metals in Tubli Bay were found to be within the comparable level of Tarut Bay in KSA, with exception of Cd and V which were higher in Tubli Bay. In general, the trace metals reported in this study found to be higher than other regions.

Table 3 Studies conducted on trace metal ions in Bahrain's marine environment.

#	Year	Metals	Type of sample	Reference
1	1983-1991	Ag, As*, Cd, Co, Cu, Cr, Fe, Mn, Ni, Pb, V, Zn and Se*	Sediments Rock Scallops (<i>Spondylus sp.</i>), pearl oyster (<i>P. radiata</i>), Greasy grouper (Hamoor), Grouper (Burtom), Two-banded Porgy (Fasker), Pigface Bream (Sheiry)	(Fowler et al., 1993)
2	1984	Cu, Zn, Cd, Hg, and Pb	Seawater Suspended particulates	(Fowler et al., 1984)
3	1990-1991	Zn, Pb, Ni, Cu, Fe, Mn, and Cd	Grouper fish (<i>E. coioides</i> , Hamoor)	(Al-Sayed et al., 1996)
4	1991-1992	Cu, Mn, Fe, Zn, Pb, Cd, and Ni	Seawater Pearl oyster (<i>P. radiata</i>)	(Al-Sayed et al., 1994)
5	1993	Cd, Pb, Hg, and As*	162 different sample of edible fish and shellfish (23 species of fish and 3 species of shellfish)	(Al-Madany et al., 1996)
6	1997	Pb, Zn, Cd, Ni, Cu, and V	Sediments	(Akhter & Al-Jowder, 1997)
7	2002	Cu, Pb, Cd, Zn, Cr, Mn and Ni	Marine algae: <i>H. triquetra</i> , <i>sargassum</i> , <i>F. spiralis</i> , <i>P. Pavonica</i> , <i>Ulva</i> , <i>E. Intestinalis</i>	(Ali-Mohamed, 2002)
8	2000-2001	Ag, Hg, As*, Cd, Co, Cu, Cr, Fe, Mn, Ni, Pb, V, Zn, Se*, Al, Sb*, and U	Sediments Pearl oyster (<i>P. radiata</i>) Orange spotted grouper (<i>E. coioides</i> , Hamoor)	(de Mora et al., 2004)
9	2005	Fe, Cu, Zn, Pb, Hg, and Cd	Blue swimming crab (<i>P. pelagicus</i>)	(Musaiger & Al-Rumaidh, 2005)
10	2006	Fe, Ni, Zn, Mn, Pb, Cu, and Cd	Tissue of marine snail (<i>T. coronatus</i>) (Gmelin)	(Al-Sayed & Dairi, 2006)
11	2007	As*, Cd, Cu, Fe, Mn, Ni, Pb, V, Zn, and Hg	Seawater	(Juma & Al-Madany, 2008)
12	2007-2017	Cu, Cr, Cd, Ni, Pb, Zn, Al, and Fe	Sediments	(SCE, 2018)
13	2008	Fe, Cu, Zn, Pb, Hg, and Cd	8 fish species and one shrimp species.	(Musaiger & D'Souza, 2008)
14	2009	Total-Hg and Methyl-Hg	Five fish species: <i>S. commerson</i> , <i>S. canaliculatus</i> , <i>E. coioides</i> , <i>L. nedulosus</i> and <i>S. albella</i>	(Freije & Awadh, 2009)
15	2009-2020	Cu, Cr, Cd, Ni, Pb and Zn	Sediments	(SCE, 2020)
18	2016	Cr, Cu, Fe, Mn, Mo, Ni, Pb, and Zn	Sediments and leaves of gray mangrove (<i>Avicennia marina</i>)	(Abou Seedo et al., 2017)
19	2017	Ag, Al, As*, Cd, Cr, Cu, Fe, Mn, Mo, Ni, Pb, V and Zn	Sediments, branches and leaves of gray mangrove (<i>Avicennia marina</i>)	(Almahasheer, 2019)
20	2020	Cr, Ni, Cu, Zn, As*, Cd, Pb and Hg	Sediments	(Bersuder et al., 2020)

* As and Sb are metalloid, Se is a nonmetal

2.5 Trace Metals in BSC in the Arabian Gulf

There are limited studies that had investigated the trace metals content of the BSC in Bahrain and the Arabian side (west coast) of the Arabian Gulf, southern coastlines. On the other hand, many studies were conducted in the Iranian side (east coast) of the Arabian Gulf, as well as in the northern coastlines, Table 4.

The trace metals content of BSC was investigated only in two studies in Bahrain (Al-Madany et al., 1996; Musaiger & Al-Rumaidh, 2005), which studied the edible tissues of the crabs. Al-Madany *et al.* (1996) did not report a significant differences in the levels of Cd, As, Pb, and Hg in the tissues of BSC between the coasts of Bahrain. Arsenic found to be significantly higher than the other metals which have the following order $As > Pb > Cd > Hg$ with a mean concentration of 3.62, 0.163, 0.054, and 0.04 mg/kg wet weight, respectively. This pattern was different in the next decade, where Musaiger and Al-Rumaidh (2005) reported Hg more than Cd where the latter has not been detected in the study, whereas Pb and Hg almost equal in concentration 0.04-0.03 mg/100g wet weight.

In Kuwait, Al-Mohanna & Subrahmanyam (2001) used the BSC to investigate the impact of oil spillage after the Gulf War I in Kuwait marine environment. Nine trace metals (As (metalloid), Cr, Cu, Pb, Me, Mn, Se (metalloid), V and Zn) were studied in different crab organs (gills, hepatopancreas, gonad, gastric stomach and muscle). Their results showed that muscles and hepatopancreas accumulated more trace metals than the other organs. Between the tested trace metals, Zn and Cu were found to be significantly higher than the other trace metals in the crabs tissue, whereas Zn found to be higher than Cu. These results indicated the ability of crabs to accumulate metals to a detectable levels, especially Zn and Cu. On the other hand, the elevated concentrations of Zn and Cu might indicated that

these sites were still affected by the Gulf War I oil spill in 1991, or other land-based activities.

In Saudi Arabia, BSC was studied in three estuarine ecosystems along the eastern coastline (Ibrahim et al., 2020). The results showed elevated levels of trace metals (Cu, Zn, Cd, Pb, Cr, Al, Fe, Mn and Ni) measured in the gills of the crabs. Zinc found to have significantly higher level than the other trace metals which found to have the following distribution pattern $Zn > Al > Cu > Pb > Mn > Fe > Ni > Cd > Cr$. On the other hand, Pb, Cu, Cr and Al found to be higher in winter compare to other seasons, whereas Zn and Ni found to be higher in autumn season. Southern Khobar site found to have the highest content of trace metals, and the percentage of trace metals found to be higher than Saudi Arabian Standards.

On the eastern coast of the Arabian Gulf, several studies were conducted on the Iranian coastline to investigate the trace metals in the BSC's tissues. Three studies investigated the levels of Fe, Hg, Ni and Pb in the hepatopancreas, exoskeleton and muscle of BSC in 11 sites along the Iranian coastline (Hosseini et al., 2012a; Hosseini et al., 2012b; Hosseini et al., 2015). The results showed the decreasing level of the trace metals as followed hepatopancreas > muscle > exoskeleton and the distribution pattern of the trace metals as followed $Fe > Ni > Hg > Pb$. The concentration of the trace metals found to be higher in summer than in winter. However, no significant differences between the levels of the tested trace metals were recorded. On the other hand, the accumulation of Hg within different tissues of BSC were studied by Elahi *et al.* (2012). The accumulation of Hg in the viscera and muscles of the crab was significantly increased with an increase of the size and the weight of the crabs. But the accumulation of Hg in the skeleton was not affected by the size and weight of the crabs. However, the concentration of Hg was not affected by the gender of the crabs.

Mercury also were studied within the hepatopancreas, muscle and exoskeleton of BSC in 6 stations eastern coast of the Arabian Gulf in Iran (Hosseini et al., 2012c). The accumulation pattern of Hg was found to be in the order of: hepatopancreas > muscle > exoskeleton, but there was no significant difference between the Hg concentration within these three organs. The Hg levels were between 4.70 - 0.11 µg/g dry weight, and no significant differences were observed between the male and female crabs.

Heidarieh *et al.* (2013) studied the levels of Fe, Zn, As, Co and Mn in the whole body of BSC near Bushehr coastline. Arsenic contamination was recorded in the BSC's body. The distribution pattern of the studied trace metals was found in order: Zn > Fe > As > Mn > Co with a concentration of 66.64, 62.87, 21.38, 1.91 and 0.15 µg/g dry weight, respectively.

The concentration of both mercury (Hg) and methyl mercury (MeHg) were investigated in the tissues of hepatopancreas, muscle and gill of BSC by Sarasiab and Hosseini (2014). The accumulation pattern found to be in decreasing order hepatopancreas > gill > muscle for both Hg and MeHg. The results showed positive significant relationships between the concentration of Hg and MeHg with the crab size. Furthermore, the level of Hg and MeHg in the male crab's organs (hepatopancreas, muscle and gill) were found significantly higher than female crab's organs, except in muscle which were found to accumulate more Hg in female crabs.

Rahmanpour *et al.* (2014) used BSC as a bioindicator for the trace element loads in intertidal ecosystems along the eastern coast of Arabian Gulf. Six trace elements (Ni, Pb, V, Cd, As and Hg) were measured in the hepatopancreas of BSC collected from four intertidal ecosystems, coral reef, rocky shore, mangrove forest, and muddy habitat. The results indicated that the highest trace metals content in the crabs was found in the coral reef, followed by muddy habitat and mangrove forest, and the lowest content was in the rocky habitat. The trace metals accumulation in the

crab's hepatopancreas was different from one habitat to another, but in general, Ni was significantly higher than the other metals, and V was the lowest. The overall mean sequence of the trace metals was in the following decreasing order $\text{Ni} > \text{Cd} > \text{Pb} > \text{As} > \text{Hg} > \text{V}$ with a concentration of 393.23, 6.00, 5.21, 4.58, 3.15 and 0.89 $\mu\text{g/g}$ wet weight, respectively.

Blue swimming crab was used as a bioindicator for studying As level in coastline of Asalouyeh city in Iran during the winter months (Khoramnejadian & Fatemi, 2015). The level of As was found to be in the range of 0.13-0.23 mg/kg dry weight. There was a significant difference between the As concentrations during the January, February and March month, where the concentrations were 0.08, 0.21 and 0.22 mg/kg dry weight, respectively.

In Iran, BSC has succeeded to be a bioindicator for Cd and As contamination in the Asalouyeh coast, which has been affected by many petrochemical, petroleum, and natural gas industries (Fatemi & Khoramnejadian, 2016). The levels of Cd and As were found to be higher in impacted regions than the sample collected from the a protected area (in Bushehr Province) as control area. Cadmium found to be significantly higher than As in the soft tissues of male crabs, where the level of Cd and As recorded were 0.011-3.503 and 0.075-0.616 $\mu\text{g/g}$ dry weight, respectively. On the other hand, temporal variation in the contents of Cd and As was observed in this study, where the decreasing order of their contents were as in the following order: March > February > January. The reported levels of Cd and As found to be not safe for human consumption.

Table 4 Studies conducted on trace metals in BSC in the Arabian Gulf.

#	Year	Country	Trace Metal ions	Parts of Crabs	Reference
1	1993	Bahrain	Cd, Ph, Hg, As*	Edible tissues	(Al-Sayed et al., 1996)
2	1998-1999	Kuwait	As*, Cr, Cu, Pb, Mn, Mg, Se*, V	Hepatopancreas, gills, gonad, gastric stomach, and muscle	(Al-Mohanna & Subrahmanyam, 2001)
3	2005	Bahrain	Fe, Cu, Zn, Pb, Hg, Cd	Edible muscles in the carapace	(Musaiger & Al-Rumaidh, 2005)
4	2011	Iran	Total Hg	Hepatopancreas, muscle and exoskeleton	(Hosseini et al., 2012c)
5	2012	Iran	Fe, Hg, Ni, Pb	Hepatopancreas, muscle and exoskeleton	(Hosseini et al., 2012a; Hosseini et al., 2012b; Hosseini et al., 2015)
6	2012	Iran	Total Hg	Edible tissue, exoskeleton and viscera	(Elahi et al., 2012)
7	2013	Iran	Zn, Fe, As*, Mn, Co	Whole body tissue	(Heidarieh et al., 2013)
8	2014	Iran	Ni, Pb, V, Cd, As*, Hg	Hepatopancreas	(Rahmanpour et al., 2014)
9	2014	Iran	PCB, PAH, Hg, CH ₃ Hg	Gills, hepatopancreas, and muscle tissue	(Sarasiab & Hosseini, 2014)
10	2014	Iran	As*	Soft tissues	(Khoramnejadian & Fatemi, 2015)
11	2014	Iran	Cd, As*	Soft tissues	(Fatemi & Khoramnejadian, 2016)
12	2017-2018	Saudi Arabia	Cu, Zn, Cd, Pb, Cr, Al, Fe, Mn, Ni	Gill	(Ibrahim et al., 2020)

* As is metalloid, Se is nonmetal

2.6 Trace Metals in BSC: Other Regions

Globally, there were many studies conducted on the accumulation of trace metal contaminants in the tissues of BSC. Not far from the Arabian Gulf, in the Indian Ocean, the levels of Cd, Cu, Cr, Pb and Zn were studied in the hepatopancreas, gill and muscle of BSC collected from Ennore estuary (Viswanathan et al., 2013). The results showed Zn and Cu were significantly higher than the other trace metals in all organs under study; moreover, Zn were higher than Cu in the gills and muscles but in hepatopancreas Cu was higher. Gills found to accumulate the highest

overall mean of the tested trace metals in the BSC, then muscles and the hepatopancreas found to accumulate the least amount.

In the sites along the coast of Mannar, Negombo and down south in Sir Lanka, Hg, Cd, As, Pb, Cu, Fe and Zn were investigated in the muscles of BSC (Jinadasa et al., 2018). The results showed a similar distribution pattern of the trace metals in the tissues of male and female crabs that had the following sequence $Zn > Fe > Cu > As > Pb > Cd > Hg$. The overall mean concentrations of Zn, Fe, Cu, As, Pb, Cd and Hg were found as following: 22.04, 13.41, 7.97, 4.45, 0.22, 0.15 and 0.08 mg/kg wet weight, respectively. Females found to accumulate Zn and Fe, in their tissues, significantly higher than the male crabs, whereas no significant difference found between the other metals between the male and female crabs. According to the European Commission, for trace metal levels in crustaceans (EU/EC-1881, 2006), based on the level of Hg, Cd and Pb, the crab sample in this study found be safe for human consumption.

Nine trace metals (Cu, Mn, Cd, Ni, Pb, Co, Hg, Cr and U) content of the gill, muscle and hepatopancreas of BSC were studied along the southeast coast of India (Kumar et al., 2019). The distribution pattern of the metals in the different organs of the crabs was found in decreasing order: gill > hepatopancreas > muscle. The distribution patterns of the metals within each crab organs were not identical, but Cu found to be the highest and U was the least in all organs.

In Bangladesh, the northern Bay of Bengal, Karar *et al.* (2019) measured the level of Cd, Cu, Cr and Pb in the gill, muscle and hepatopancreas of BSC during 12 months. Results showed that Cu present in the concentration significantly higher than the other trace metals. Gill was found to accumulate more Cu than other organs in that study, but hepatopancreas was accumulating more Cd, Cr and Pb than other organs. Instead, muscles found to have the lowest level of the trace metals studied.

The total trace metals concentrations in all organs were in the following distribution order: Cu > Pb > Cd > Cr with a total concentration of 39.51, 4.82, 4.54 and 2.84 µg/g wet weight, respectively. These levels were found to be safe when they were compared with the levels recommended by WHO and US FAD. On the other hand, this study reported that salinity affect the accumulation of trace metals in the tissues of crabs. The level of trace metals was found higher during the monsoon season than then non-monsoon seasons where the salinity is higher.

Also in Indian Ocean, Anandkumar *et al.* (2019) studied the concentration of Zn, Cu, Rb, Mn, Cr, Pb, Co, Ni and Cd in the carapace and waking leg muscle tissues of BSC in the coastal Borneo, Malaysia. The results showed Cu, Mn, Rb, Cr and Co were accumulated in the waking leg macules higher than carapace tissues, whereas Zn was significantly highly accumulated in the carapace tissues. Zinc trace metal found to be significantly the highest concentration between the studied trace metals, then Mn, Cu, Co, Cr and Pb found to be higher than the rest of metals. The concentration of Zn, Cu, Rb, Mn, Cr, Pb, Co, Ni and Cd found to be 394, 33.5, 11.2, 7.2, 5.1, 4.1, 3.8, 2.4 and 0.6 µg/g dry weight in carapace tissues, and 319, 85.7, 10.5, 15.9, 7.2, 4.0, 6.8, 3.1 and 0.6 µg/g dry weight in waking leg tissues, respectively. These levels found to be safe for the human consumption.

In the Red Sea, along the Suez Canal, Hg was studied in the gonads, muscles, hepatopancreas and gills of BSC in Bitter lakes and Lake Timsah, Egypt (El-Moselhy, 2006). Blue swimming crab found to accumulate higher level of Hg compared with other marine organisms tested in this study, which had a decreasing order: crab > bivalve > shrimp > fish. The distribution of Hg in the organs of male and female BSC was not similar in the gender or in the location. However, in general, gills found to accumulate the lowest level of Hg compared to other organs. Female found

to accumulate more Hg than male crabs in all organs tested except in gonad in case of Bitter Lake that had lower level than the gonad of the male crabs. The accumulation of Hg found to be affected by the gender, growth rate, size and variation of the species; in addition, the habitat and the source of the pollutants surrounding those marine organisms.

In the Red Sea also, on Saudi Arabia's Jeddah coastline, the edible muscle tissues of male and female BSC were investigated for the content of Fe, Cu, Zn, Co, Se, Hg, Pb and Cd (Abdel-Salam & Hamdi, 2014). Results showed significant differences in the content of the trace metals in male and female crabs. The male crabs found to accumulate Fe, Zn, Co, Hg and Pb more than female, whereas female found to have significantly higher level of Cu, Se and Cd than male. Iron (Fe) and zinc (Zn) found to have a significantly higher concentration than the other metals which have the following sequence: $Fe > Zn > Pb > Cu > Hg > Co > Cd > Se$.

In the Mediterranean, at the Northeastern coast in Mersin Bay (Turkey), Cd, Cr, Pb, Cu, Zn and Fe levels were measured in the carapace meat of male and female BSC (Ayas & Özogul, 2011). Results showed elevated level of Zn and Cu compared to the other trace metals, which had the following pattern: $Zn > Cu > Fe > Cd > Cr > Pb$. The levels of Zn, Cu, Fe, Cd, Cr and Pb in male and female crabs were found to be: 102.51, 29.98, 22.64, 0.88, 0.55 and 0.21 $\mu\text{g/g}$ dry weight and 107.27, 26.92, 15.21, 1.11, 0.40 and 0.33 $\mu\text{g/g}$ dry weight, respectively. Male BSC found to have accumulated Cr, Cu and Fe significantly higher than females, whereas females had higher levels of Cd, Pb and Zn.

In the Mersin Bay, Northeastern Mediterranean Sea, Ayas (2013) studied the levels of Fe, Cu, Zn, Cr, Pb and Cd in the lump crab meat (LCM) and chela crab meat (CCM), of male and female adult BSC over the four seasons. Results showed that the accumulation of those trace metals was not affected by the gender of the BSC, but it was affected by

the season. Chromium was highest in winter, Pb was highest in summer, Cd, Zn and Fe were highest in spring and Cu was higher in spring and summer. CCM was found to accumulate more Zn and Cu than CLM, whereas CLM found to accumulate more Fe. Zinc, Cu and Fe were found to be significantly higher than the other metals in that study. The BSC tissues were found to be not contaminated by Cr and Pb, but the levels of Zn, Cu and Cd were higher than the accepted limit for human consumption in Turkey.

Later in 2016, Zn, Fe, Cu, Al, Mn, Cr and Pb studied in the carapace meat of male and female BSC in Mersin Bay also (Olgunoğlu & Olgunoğlu, 2016). Results were not different from those reported earlier before Zn, Fe, and Cu were found significantly higher in concentration than the other trace metals. Besides, the distribution pattern in both male and female was in the following order: $Zn > Fe > Cu > Al > Mn > Cr > Pb$, with no significant difference between the trace metals in male and female crabs. The elevated level of Zn, Fe, and Cu may indicated that Mersin Bay affected by anthropogenic activities. The contents of Zn (334.50 and 345.94 mg/kg wet weight), Fe (146.28 and 149.70 mg/kg wet weight), and Cu (145.00 and 143.02 mg/kg wet weight) in male and female, respectively, were found to be not safe for human consumption.

The potential of trace elements bioaccumulation in the tissues of BSC were studied by McPherson and Brown (2001). Blue swimming crabs were feed on tissues of *Trychomya hirsute* contaminated by Cd, Cu, Zn, As, Fe and Al for 8 weeks. Results showed, only Cd was accumulated in the hepatopancreas tissues, which started to be detected within 4 weeks of the study. Cadmium and mercury, found to have an effect on the hemolymph glucose of BSC (Saravanan et al., 2018). Elevated level of glucose observed after exposing the BSC to Cd and Hg for 2 and 1 day(s), respectively. Thus this may indicate the effect of Cd and Hg on the

performance of both crustacean hyperglycemic hormone and hepatopancreas glycogen in the BSC.

Chapter Three

Methods and Procedures

Chapter Three

Methods and Procedures

To achieve the objectives mentioned in Chapter Two, the current study was conducted in two parts as shown in Figure 3. The first part was conducted on the dry weight of the crab samples, where the white meat of 15 crabs was collected into one composite sample (the same was done for brown meat). Then, these composite samples were dried using a freeze drier followed by digestion and analyzed by an atomic absorption spectrometer (AAS). In the second part of the current study, the white and brown meat of 10 crabs were digested individually, and then analyzed by AAS.

The objective of the first part was to identify the levels of Zn, Cu, Cr, Ni, Pb and Cd in different tissues and genders of BSC. Moreover, this part contributed to identify which metal ions can be detected and have to be included in the second part of the study. This is because the metals are concentrated in the dry tissues more than wet tissues.

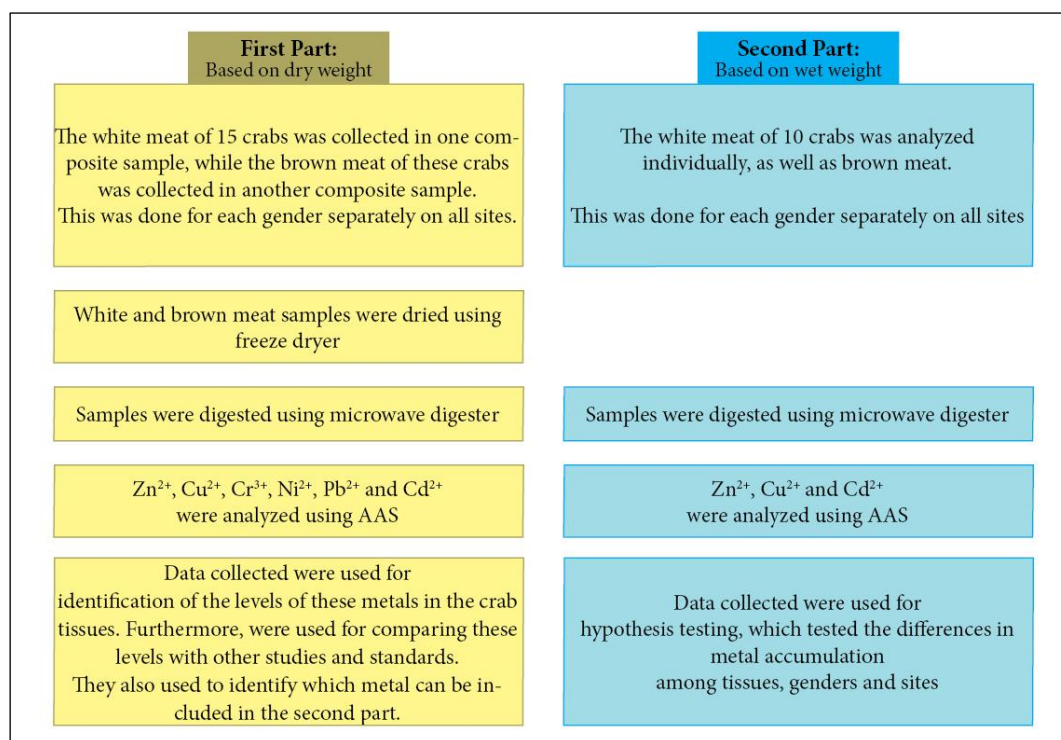


Figure 3 A flowchart for the processes carried out in this study.

3.1 Study Area

According to Ali and Abahussain (2013), the General Directorate for the Protection of Marine Resources in Bahrain divided the fishing grounds into three main areas, namely Western, Northern and Eastern, Figure 4. The decreasing order of the total commercial fisheries collected from those areas were in the following order Northern > Eastern > Western, which represented 51.3%, 38.7% and 10.0%, respectively. The finfish fisheries were the largest percentage in both Northern and Western areas, whereas BSC was the largest percentage in the Eastern area. The BSC represent 12.2%, 44.6% and 29.1% of the total fisheries collected from Northern, Eastern and Western fishing grounds, respectively. In general, Eastern fishing ground is the largest source of BSC in Bahrain, which represents 65% (more than 3000 tons/year), followed by the northern coast and the west coast (less than 1000 tons/year for each).

The current study was conducted in the Bahrain's territorial water. Four sites were selected in the different fishing grounds, Figure 4 and Table 5. The first site (Askar) was selected in the eastern coast of Bahrain where the most land-based activities are located (Khadija Zainal et al., 2008), and 65% of BSC fisheries are collected from this coast (Ali & Abahussain, 2013). The second site (Ra's M'ttalah) was selected in the western coast which has limited land-based activities. The third site (Marwada) is located in the northern coast of Bahrain which represents the largest commercial fisheries stock, and characterized by limited land-based activities. Whereas the last site is located in Tubli Bay, which is affected by the discharge water of the sewage treatment plant (Abou Seedo et al., 2017).

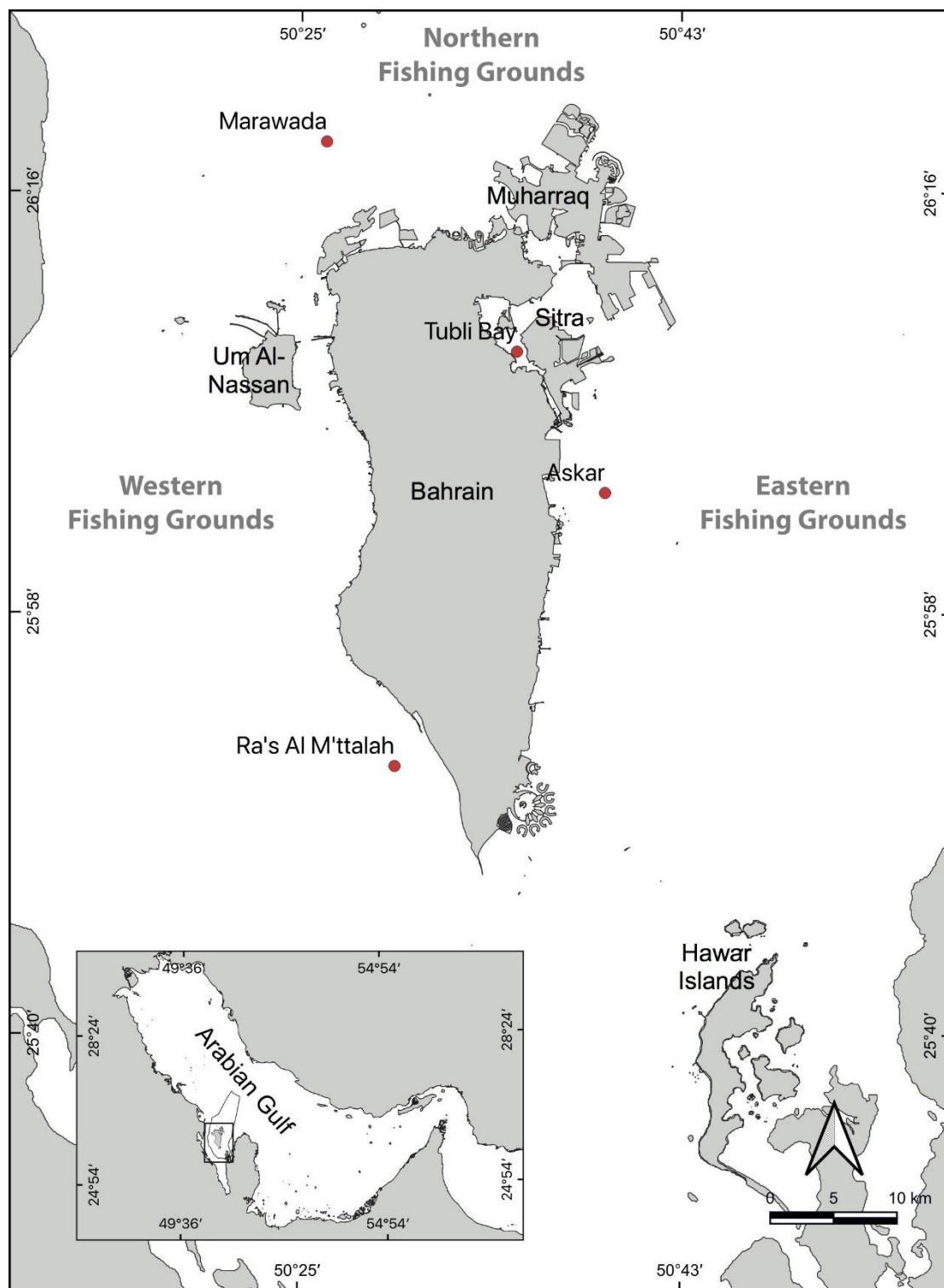


Figure 4 The fishing grounds in Bahrain and the locations of the sampling sites.

Table 5 Description of the sites investigated.

Site	Name	Description	Latitude	Longitude
1	Ra's Al M'ttalah	Western, limited land based activities.	25°52.017	50°30.002
2	Marawada	Northern, limited land-based activities	26°18.673	50°26.694
3	Tubli Bay	Tubli Bay, affected by a sewage treatment plant	26°09.716	50°35.730
4	Askar	Eastern, affected by land-based activities	26°03.684	50°39.911

3.2 Selecting the Bioindicator

Bioindicators are considered as an important tool for studying the state of the environment and detecting the environmental pressures and their impacts on the ecosystems (Stankovic & Stankovic, 2013). The first use of bioindicators dates back to the 1960s (Holt & Miller, 2011). At that time the bioindicators were used as indicators for the pollutants in the aquatic system (Sinicropi et al., 2010). Approximately 40% of studies that use bioindicators are associated with a study of trace metals pollution (Burger, 2006).

Bioindicator selection is a complex issue. Therefore, it is not an easy process to find a bioindicator that has the ability to achieve research objectives (Han et al., 2015). In order to select a good bioindicator in the marine environment, the aquatic organism should have the following characteristics, easily identified and collected, long-lived and sedentary, available throughout the year and reasonably abundant (Cantillo, 1998). In addition, tolerant to pollution and different environmental conditions (Lichtfouse et al., 2011). For the study of trace metals in the aquatic system, in addition to the criteria mentioned above, bioindicators must have the ability to accumulate trace metals constantly and tolerate a high level of non-essential metals (toxic metals), as well as they must have sufficient tissues to do the analysis (Stankovic et al., 2014).

This study used the blue BSC as a bioindicator for trace metals in the marine environment of Bahrain.

Several factors influence the choice of BSC as a bioindicator; firstly, BSC has the ability to accumulate the trace metals in different organs tissues (Potter et al., 1983; Al-Mohanna & Subrahmanyam, 2001; McPherson & Brown, 2001). Because BSC is a benthic organism, it accumulates the trace metals from the sediment and it feeds on benthic preys (Zhao et al., 2012). Furthermore, BSC is a scavenger organism (Albelali et al., 2010), which leads to accumulation of trace metals in its different tissues. Secondly, this species is abundant in the territorial waters of Bahrain and the Arabian Gulf (Albelali et al., 2010; Monikh et al., 2014; Naderloo, 2017). Additionally, it has a long life-cycle due to the slow growing rate (Williams, 1981). Besides, its limited mobility (Sarasiab & Hosseini, 2014), BSC is suitable for the comparisons of pollution in different regions or habitats. Thirdly, there are many other marine organisms that feed on this crab e.g. fish and an octopus (Castro & Huber, 2002), which means its contents of trace metals can accumulate and magnify in marine organisms that come at higher levels in the food chain. Fourthly, BSC is eaten by some of the Bahraini people (Musaiger & Al-Rumaidh, 2005), so it has a direct impact on human health. Lastly, this kind of crab has an economic value and it is a commercially important species on the international level (Romano & Zeng, 2007; Albelali et al., 2010; Viswanathan et al., 2013), whereas locally in Bahrain, BSC has a low local consumption (Khadija Zainal, 2013).

Currently, the price of BSC in Bahrain local market is about 1.5 BD/kg (equivalent to US \$3.970). The traditional recipe for BSC in Bahrain is by boiling the crabs with spices for around 2 hours, then eating the tissues of the appendages and the tissues that are in carapace without gills. The gills are not eaten "because they are filters that collect urine"

according to some people's beliefs in Bahrain. Female's crabs are more preferred to be eaten than males.

During the sampling period, it was observed that more than 90% of the catchments were male. The fishermen clarified that in summer season the male ratio is high, while in the winter season (November) the female ratio is the highest.

BSC's fishery was decreased (SCE, 2015) due to the overfishing, which leads to issuing several Ministerial Orders to control BSC's fishing in Bahrain. The first Ministerial Order controlling the fishing of the BSC was issued in 2007 by the Public Commission for the Protection of Marine Resources, Environment and Wildlife. Ministerial Order No. 2 2007 with respect to Preventing Crab Fishing, prevents the fishing of crabs with carapace width less than 7 cm as well as females with eggs. This Order prevents fishing the mentioned cases at any time of year.

Then, from 2012 to 2014, Bahrain issued annually Ministerial Order to prevent the BSC's fishery from March 15th to May 15th. Ministerial Order No. 12 of 2015 expanded the period of banning to be four months starting from March 15th until July 15th. Finally, in 2016 General Directorate for the Protection of Marine Resources in Bahrain issued the Ministerial Order No. 52 of 2016, which controls the fishing of this species by annual ban within the reproduction period that starts from March 15th to May 15th.

3.3 Samples Collection

The crab's samples were collected by traditional fishing using large-wire traps (called "Gargor" in Bahrain) by the fishermen. For the first part of this study, the samples were collected during 25th May and 22nd June 2020 for the Ra's Al M'ttalah, Marwada and Askar sites, whereas for Tubli Bay site collection was carried out on 6th of November 2020. This

difference in the dates of sampling was related to obtaining the appropriate size of crab for this study, which is discussed below. Fifteen crabs of each gender were collected from each site and washed with tap water followed by double distilled water to get rid of external contaminants attached to the body of the crab.

For the second part of this study, 2x4 two-way ANOVA design was used to test for difference among different genders and locations (2 gender types and 4 stations). According to Bausell and Li (2002), the required group size for 2x4 two-way ANOVA design is 10 at $\alpha=0.05$, statistical power=0.80, and effect size=0.80. Thus, 10 male crabs and 10 female crabs were collected from each of the 4 stations (40 males and 40 females in total). It is worth mentioning that this sample size was more than sufficient as well for testing the differences among the two tissues using paired t-test as the test requires a total number of samples of 18 only at $\alpha=0.05$, statistical power=0.80, effect size=0.80, and $r=0.4$.

The samples were collected from 13th August to 24th September 2021 for the four sites. Ten crabs of each gender were collected from each site. Then they were washed with tap water followed by double distilled water to get rid of external contaminants attached to the body of the crab.

In the laboratory the morphometric measurements such as carapace width (CW), carapace length (CL) and the weight of the samples, were measured using digital caliper and balance, Figure 5 and Figure 6. Finally, samples were stored individually in polyethylene bags at -20°C for the time of dissection.

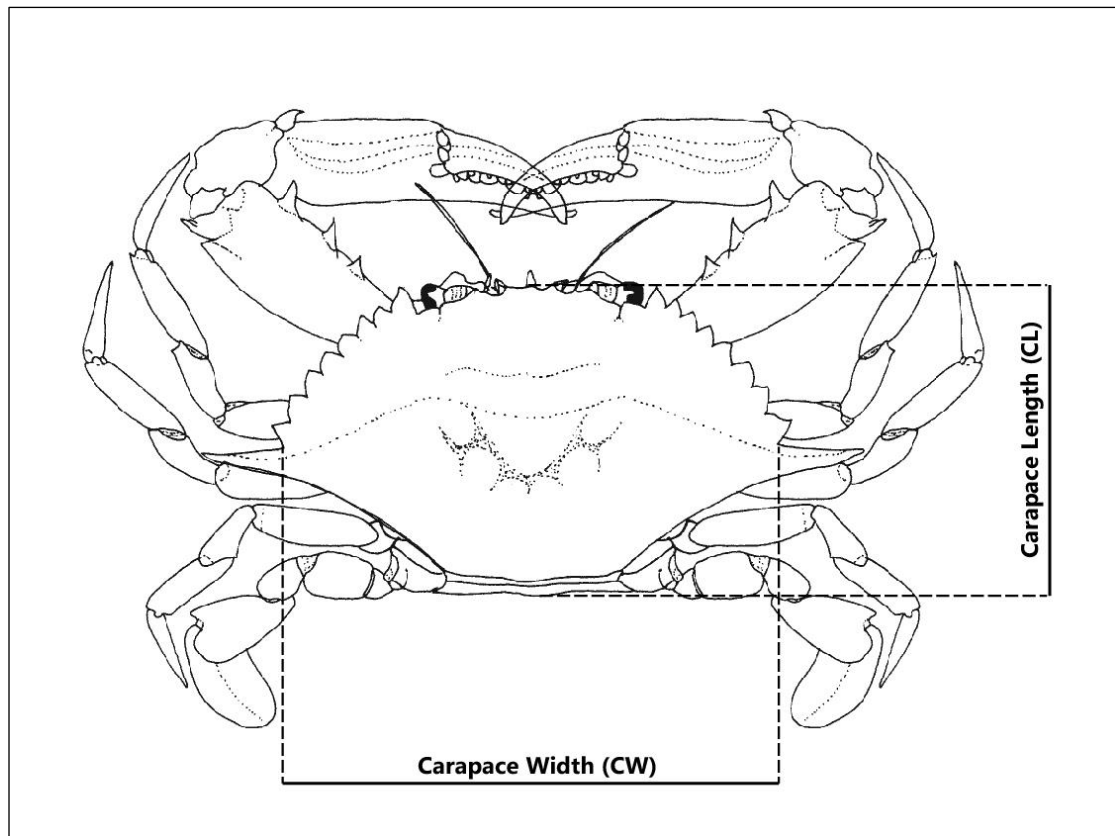


Figure 5 Schematic drawings of a BSC, with carapace width (CW) and carapace length (CL).

The longevity of BCS found to be up to 3 years in Queensland, Australia (Smith & Sumpton, 1987), and up to 2.5 years in India (Sukumaran & Neelakantan, 1997), whereas Al-Rumaidh (2002) reported that Bahrain's BSC live up to 3 years. The maximum size (carapace width) of BCS in Bahrain was reported to be 16 cm for Male and 17 cm for female. The average growth rate of BSC in Bahrain was found to be 1.3 cm/month, while the growth rate in summer is the highest. In Bahrain, the male of BSC reaches the sexual maturity before the females. Males start to reach sexual maturity from 4 cm CW, but at 8 cm CW 50% of males reach sexual maturity. In contrast, females reach their sexual maturity from 7 cm CW while at the size of 9 cm 50% reach sexual maturity (Al-Rumaidh, 2002).



Figure 6 Measuring the carapace width (CW) for a female BSC.

There were some studies that showed effective size of the crab's on the trace metals accumulation in its tissues (Zhao et al., 2012). To eliminate the crab's size factor, a specific size of the crabs in all sites was selected. The size of BSC studied in this research was 11 ± 1 cm for both males and females. At this size, crabs had reached adulthood and had lived long enough to accumulate pollutants in their bodies. Moreover, this size is expected to be available in all sampling sites.

Differences in color were used to distinguish between the genders of crabs. Males' appendages are typically light blue in color, while females are green-brown in color, Figure 7. In addition, they differ in the genital organ area, the males have a narrow genital organ and its shape like the spear, while the females genital organ is round abdomens (Potter & De Lestang, 2000), Figure 8. Moreover, males have larger body dimensions than females, especially chela or manus of chelipeds (Giraldes et al., 2016; Khadija Zainal, 2017). The current research studied each gender separately

because some of the previous studies have shown that there is a significant difference between the bioaccumulation of pollutants in the tissues of BSC males and females (El-Moselhy, 2006; Elahi et al., 2012; Hosseini & Parsa, 2013; Abdel-Salam & Hamdi, 2014; Monikh et al., 2014; Sarasiab & Hosseini, 2014; Ghaeni et al., 2015).



Figure 7 Dorsal view of the BSC, the difference in color between the male (right) and female (left).

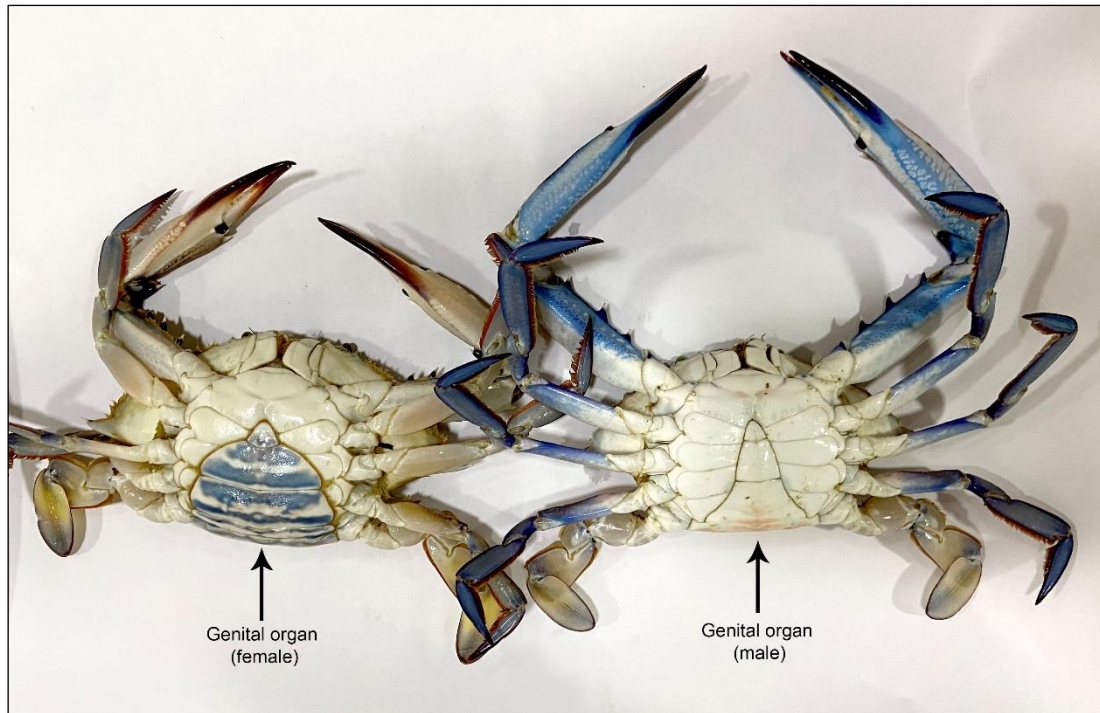


Figure 8 Ventral view of the BSC, the difference in genital organs between the female (left) and male (right).

3.4 Sample Preparation

White and Brown meats in the current study were collected like what was mentioned by Bolam and Bersuder (2013) and Ariano et al. (2015).

For the first part of this study, crabs were taken out from the freezer and kept to reach room temperature. The appendages tissue of 15 male crabs was extracted and collected together in a single jar. This tissue represents the muscles, which is called white meat in current study. Crabs have five pair of legs, the muscles (white meat) was collected from the whole the first pair that bearing claws (pincers), and the first segment of the rest legs (four pairs) as shown in Figure 9. The muscles which are inside the carapace (that hold the appendages) were excluded from the white meat sample because in most of the cases it was mixed with the digestive system, Figure 10.

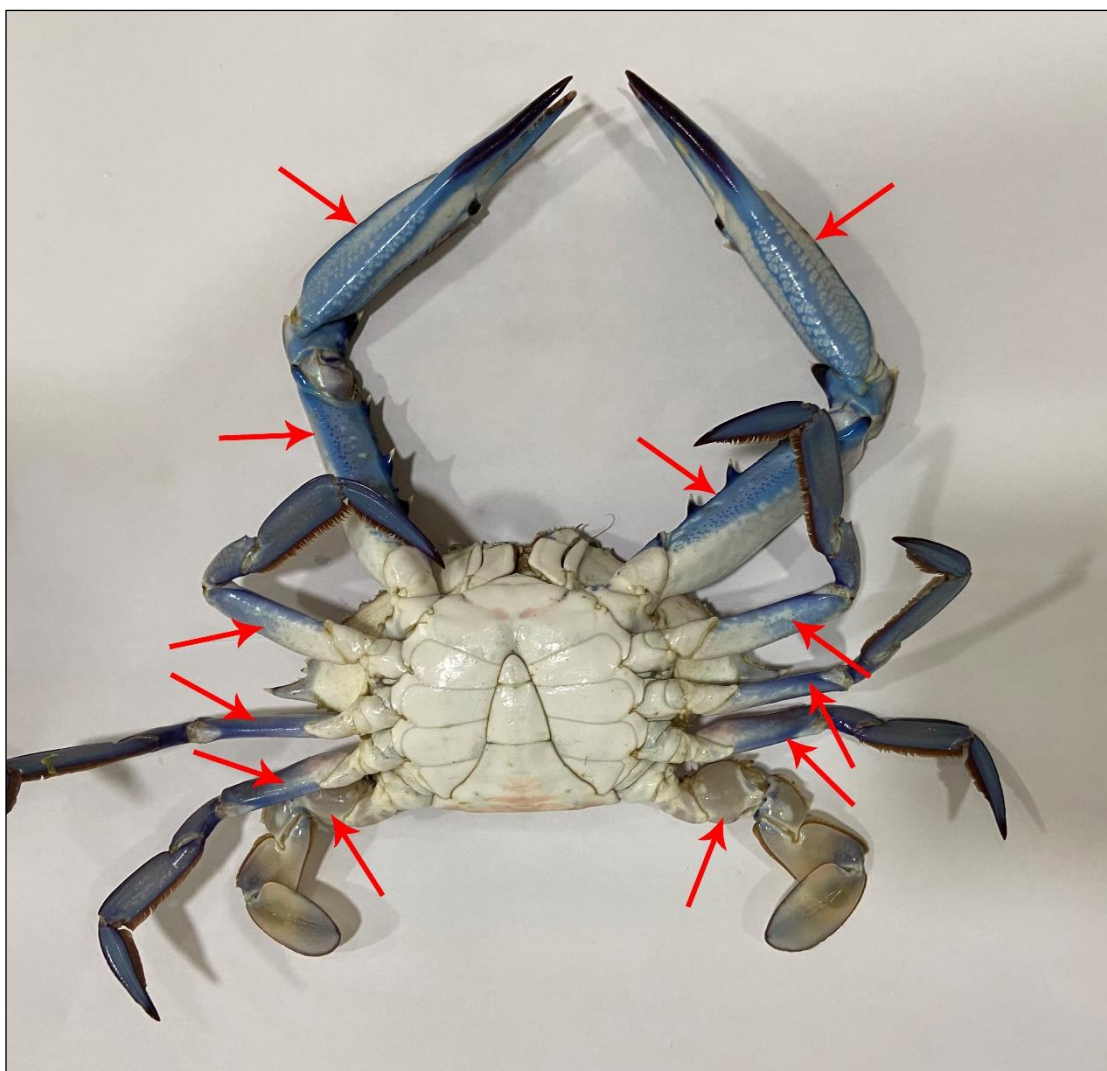


Figure 9 The places from where the white meat (muscle) samples were collected.

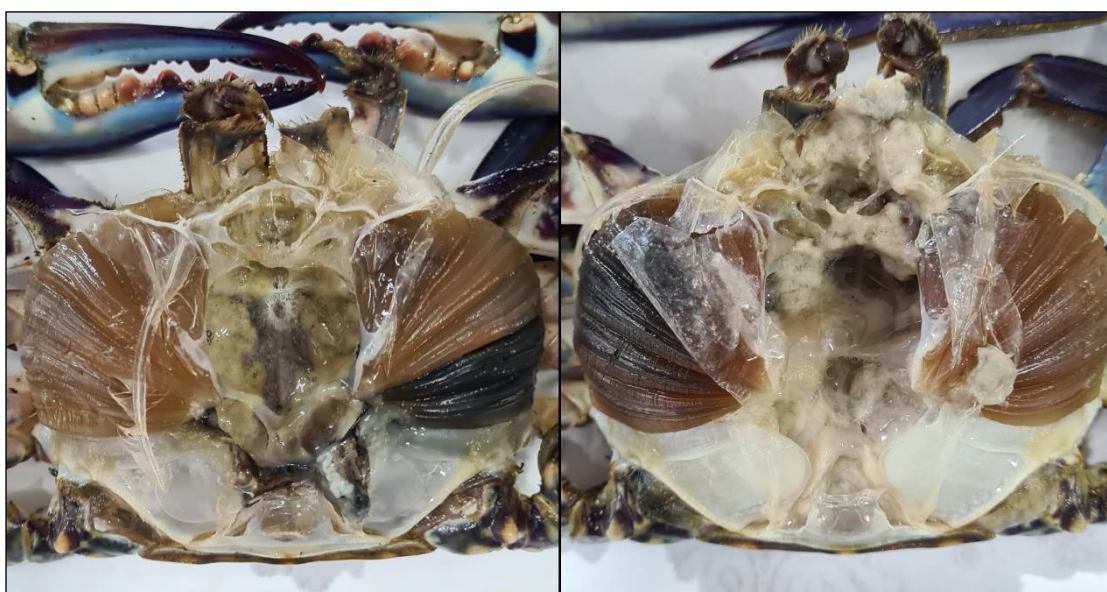


Figure 10 The digestive system tissues attached to the muscles inside the carapace.

The carapace tissues (without gills and muscles) of the 15 male crabs were collected in another jar. This tissue called "brown meat" in current study due to its brownish color after drying, which represent the digestive system and everything within the carapace excluding gills and muscles, Figure 11 and Figure 12. The same procedure was used to collect, the tissues of white and brown meat of 15 female crabs and stored in separate jars. These tissues were dried for 10 – 15 days using freeze dryer, LABCONCO FreeZone 4.5, Figure 13. The differences between brown and white meat is clearly presented in Figure 14.

All glassware used for freeze drying and storing the dried tissues were cleaned by immersing in 10% nitric acid (v/v) for one day and then washed with deionized water. After drying, glassware were then cleaned by hexane and dried in oven for one day at 105°C.



Figure 11 The parts from which the brown meat (digestive system) sample were collected.



Figure 12 The composition of the brown meat sample in the freezing flask.



Figure 13 Drying the crab tissues using LABCONCO FreeZone 4.5 freeze dryer.



Figure 14 Dried white (left) and brown (right) crab meat showed clear differences in colors.

The dried tissues were grinded and stored in amber glass container, ready for digestion. The moisture content of the dried tissues was estimated using the method in the Manual of Oceanographic Observations and Pollutant Analyses Methods (MOOPAM) method (ROPME, 2010). Around 1.0 gram of the sample was dried in the oven at 105°C for 24 hours. Then the weight of the dry sample was determined to calculate the moisture content by the following equation.

$$\text{Moisture (\%)} = \frac{\text{mass of the moisture}}{\text{Wet weight}} \times 100$$

The concentration obtained from the Atomic Absorption Spectrometer was corrected by subtracting the moisture content value from the sample weight, as showing in the following equation.

$$M_c = M_{AAS} \times \frac{100}{100 - \text{Mois}}$$

Where M_c is the corrected concentration of the metal, M_{AAS} is the concentration of the metal obtained from AAS, and Mois is the moisture percentage.

3.5 Acid Digestion of Crab's Tissues

In the first part of this study, the method used to digest the crab's tissues is the MOOPAM method published by ROPME (2010). The sample vials were shaken for 2 min to homogenize the sample. Around 0.3 g of the dried tissue was placed in the Teflon bombs, and then 5 ml of concentrated nitric acid were added. The flask was kept at room temperature for 1 hour. Then the Teflon bombs were closed tightly and placed in the Microwave Digestion System, (MILSTONE model, ETHOS PLUS). The digestion routine in Table 6 was applied to solubilize all trace metals in the sample. When the sample cooled to the room temperature, the pressure was released and the bombs were opened. The sample was then transferred into a 50 ml polypropylene graduated tube and the bomb was washed with deionized water at least 3 times (to washout the traces of the sample) and the wash water was added to the sample tube. The sample was diluted to the mark of 50 ml with deionized water and the sample was stored in the refrigerator (4°C) until the time of analysis.

Table 6 The digestion routine of biota samples

Step	Power (W)	%Power	Ramp time (min)	Pressure (PSI)	°C	Hold Time (min)
1	1200	100	5	600	50	5
2	1200	100	5	600	100	5
3	1200	100	10	600	200	12

In each run, there was one blank which was prepared in the same way as the samples. For this part, two Certified Reference Materials (CRM) were used for quality purposes, IAEA-436 (Tuna fish flesh) and IAEA-452 (Scallop, *Pecten maximus*). The values of the selected trace metals were listed in the Table 7.

Table 7 The certified values in mg/kg for the metal in the CRM

Trace Metals	IAEA-436		IAEA-452		NIST 2976	
	Mean	Std. Dev	Mean	Std. Dev	Mean	Std. Dev
Zn	19.0	1.3	167	10	137	13
Cu	1.73	0.19	11.0	0.8	4.02	0.33
Ni	0.069	0.041	-	-	-	-
Cr	0.194	0.058	5.2	0.7	-	-
Pb	-	-	2.4	0.3	-	-
Cd	0.052	0.007	30.5	1.6	0.82	0.16

For the second part of this study, the wet tissue of each crab sample was digested individually. For white meat, around 3 g of muscle in the claws (pincers) were homogenized using mortar and pestle, then around 0.5 g were balanced in the Teflon bombs. For brown meat, around 3 g of digestive system were homogenized using mortar and pestle, and then around 0.5 g was balanced directly in the Teflon bombs. After that, 7 ml of concentrated nitric acid (HNO_3) were added followed by 1 ml of hydrogen peroxide (H_2O_2). The flasks were kept at room temperature for 1 hour. Then the Teflon bombs were closed tightly and placed in the Microwave Digestion System, (MILSTONE model, ETHOS PLUS). The digestion routine in Table 6 (above) was applied to solubilize all trace metals in the sample. When the sample was cooled to room temperature, the pressure was released and the bombs were opened. The sample was then transferred into a 50 ml polypropylene graduated tube and the bomb was washed with deionized water at least 3 times (to washout the traces of the sample) and the wash water was added to the sample tube. The sample was diluted to the mark of 50 ml with deionized water and the sample was stored in the refrigerator (4°C) until the time of analysis.

In each run, there was one blank, which was prepared in the same way as the samples. For this part, three Certified Reference Materials (CRM) were used for quality purposes, IAEA-436 (Tuna fish flesh), IAEA-452

(Scallop, *Pecten maximus*) and NIST 2976 (Mediterranean mussel, *Mytilus galloprovincialis*). The values of the selected trace metals were listed in the Table 7 (above).

3.6 Determination of Trace Metal Ions

The trace metals ions content of the digested samples were measured using Atomic Absorption Spectroscopy (AAS), (Thermo model, *iCE 3000 SERIES*). The flame technique was used for Zn^{2+} and Cu^{2+} but when the level of the trace metal was low, the graphite furnace technique was used, e.g. for Ni^{2+} , Cr^{3+} , Pb^{2+} and Cd^{2+} . The calibration curves used for estimating the concentration of the trace metals are constructed by three to five points in addition to the zero point for the blank. The fit of these curves was at least 0.995.

3.7 Risk to Human Health

During 2004-2009, the BSC fishing status in Bahrain was 3000-4000 tons per year (Ali & Abahussain, 2013), the majority of this quantity was for exportation, whereas the consumption of BSC meat inside Bahrain is very low (Khadija Zainal, 2013). BSC meat was found to be rich in protein but it has low lipids level (Musaiger & Al-Rumaidh, 2005). Besides, some trace metals were detected in the BSC meat (Al-Madany et al., 1996; Musaiger & Al-Rumaidh, 2005).

To evaluate the potential risks that may be involved by consuming the crabs inhabiting Bahrain's marine environment, the trace metal contents were compared with different standards. These standards and their metal levels are listed in Table 8.

GSO 1922:2009 issued by GCC Standardization Organization (<https://www.gso.org.sa>), and it is the standard that is used in Bahrain. The

scope of this standard is concerned with the chilled and frozen crabs that are used for human consumption. The trace elements included in this standard are Pb, Cd, As and Hg only, which should not exceed 1.0, 0.1, 0.5 and 0.5 mg/kg, respectively, based on wet weight of the crab meat. The type of the meat is not specified in this standard. However, since it used the term "edible tissues", this means the standard is limited to muscles, which represents white meat in this study.

Since February 2018, the Cd threshold in the crab's edible tissues (GSO 1922:2009) was updated in Bahrain to be 0.5 mg/kg according to the Ministerial Order No. 10 of 2018. This Ministerial Order is concerned with the amendment of Ministerial Order No. 108 of 2010 with respect to the adoption of the unified Gulf standard specifications (Shellfish - Part 2: Chilled and frozen sea crab), which was issued by the Minister of Industry, Commerce, and Tourism.

EU No. 420/2011 issued by the European Commission Regulation concerning with amending Regulation (EC) No 1881/2006 (<https://www.fao.org>), which set the limits for certain contaminants in foodstuffs. The trace metal ions covered in this standard were Cd, Pb and Hg only, which should not exceed 0.5 mg/kg, each, based on the wet weight of the crustacean's meat. This limit is applied to the muscle tissues in the crab legs and claws only, but the tissues in the crab's (crustaceans) cephalothorax are excluded. Thus EU Standard applies to white crab meat only, whereas brown crab meat is not cover.

European Commission studied the Cd level in brown crab meat foodstuff in the Member States. Centre for Environment, Fisheries, and Aquaculture Science (Cefas) and the Food Standards Agency in the United Kingdom conducted a survey for analysis of Cd concentration in 399 samples of brown crab meat and brown crab meat products in the UK. The

concentration of cadmium ranged from 0.1 to 26 mg/kg wet weight with an average of 3.4 mg/kg wet weight (Bolam & Bersuder, 2013).

Table 8 Thresholds for trace metal ions in crabs white meat.

Standard	Unit	Pb	Cd
GSO 1922/2009	mg/kg	1.00	0.10*
EU No 420/2011	mg/kg	0.50	0.50

* The Cd standard in Bahrain was updated to be 0.5 mg/kg wet weight.

As mentioned above, the standards using the thresholds are based on the wet weight of the samples, whereas in the first part of the current study the trace metals content were estimated based on the dry weight. So for comparison purposes, the water content of the crab meats was estimated for normalization of the results.

Water content of the crab meat sample was estimated by Hardy *et al.* (2000), where around two grams of wet crab meat were dried in an oven for 48 hours at 96°C and the difference in weight was estimated, as shown in the following equation.

$$\text{Water (\%)} = \frac{(\text{Wet weight} - \text{Dry weight})}{\text{Wet weight}} \times 100$$

The metal concentration estimated based on dry weight was normalized by adding the water content value from the sample weight, as shown in the following equation.

$$M_{WW} = M_{DW} \times \frac{100 - WC}{100}$$

Where M_{WW} is the approximately corrected concentration of the trace metal ions based on wet weight, M_{DW} is the concentration of the trace metal ions based on dry weight, and WC is the percentage (mass/mass) water content in the crab meat.

3.8 Statistical Analysis

To achieve the objectives of this study, and to test its hypotheses, Minitab 19 application was used to work out the data analysis. Both

associations and differences of trace metal ion concentrations were investigated statistically based on the site, gender and tissue type.

In the first part of this study, the levels of the Zn^{2+} , Cu^{2+} , Ni^{2+} , Cr^{3+} and Cd^{2+} in the dried crab meat samples were reviewed and the outliers reading were excluded using the box plot test in Minitab 19. Then the mean and standard deviation were calculated.

In the second part of the current study, the hypothesized were tested for significant differences in trace metal ions concentrations (Cd^{2+} , Zn^{2+} and Cu^{2+}) among different locations, genders, and tissues.

Two-way ANOVA test is used to study the effect of two factors (independent variables) on a single response variable (dependent variable) (Bausell & Li, 2002). In the current study, two-way ANOVA was used to study the effect of location (site) and gender on metal concentration in white and brown crab meat. Therefore, the test was performed twice, once using the white meat data and second using the brown meat data.

For the cases that showed significant differences based on location, Tukey's post hoc test was used in order to understand how these sites are different from each other.

To compare metal concentrations in different crab tissues, paired t-test was performed. This test is usually used to compare two sets of associated measurements e.g., before and after measurements (Bausell & Li, 2002; McCrum-Gardner, 2008). In this study, white and brown meat were separated from each crab individually. Metal concentrations in these two tissues were dependent on each other (associated). Thus, paired t-test was used.

Chapter Four

Results, Discussion, Conclusion and Recommendations

Chapter Four

Results, Discussion, Conclusion and Recommendations

4.1 Results

In the first part, the average carapace width (CW) of overall male crabs in the current study is 10.81 ± 0.41 cm with a range of 10.13 – 11.83 cm, whereas the average CW of overall female crabs is 10.82 ± 0.55 cm, with a range of 10.04 – 12.28 cm. The weight of the male crabs was found to be higher than that of female crabs. More description data for the crab samples collected from the sites under this study, such as carapace width (CW), carapace length (CL) and weight are shown in Table 9.

Table 9 Description data of the crab's samples

Site	Gender		CW (cm)	CL (cm)	Weight (g)
Ra's Al M'ttalah	Male	Average	10.92	6.92	207
		SD	0.42	0.31	22
	Female	Average	10.51	6.75	155
		SD	0.32	0.24	16
Marwada	Male	Average	10.71	6.85	205
		SD	0.42	0.30	22
	Female	Average	10.50	6.79	166
		SD	0.43	0.26	21
Tubli Bay	Male	Average	10.54	6.58	186
		SD	0.25	0.18	15
	Female	Average	11.04	7.00	183
		SD	0.33	0.23	16
Askar	Male	Average	11.08	7.06	204
		SD	0.34	0.24	22
	Female	Average	11.21	7.19	181
		SD	0.67	0.49	26
Overall	Male	Average	10.81	6.85	201
		SD	0.41	0.31	22
	Female	Average	10.82	6.93	171
		SD	0.55	0.36	23

The N value for each gender in all sites is 15

Results have shown that Zn^{2+} ion was accumulated in both white and brown meats in almost a close concentration with a slight difference. Female crabs accumulated Zn^{2+} ion in brown meat slightly more than in white meat, but it was the opposite in male crabs. The range of Zn^{2+} ion in white meat of overall male crabs was 209.25 – 240.25 mg/kg dry weight, while it was 174.56 – 256.81 mg/kg dry weight in brown meat with an average equal to 229.94 mg/kg and 202.09 mg/kg, respectively. In overall female crabs, Zn^{2+} ion concentration range was 208.27 – 255.32 mg/kg in white meat and 232.53 – 300.45 mg/kg in brown meat with an average equal to 242.41 and 268.56 mg/kg, respectively. The mean concentration, standard deviation and the ratio of Zn^{2+} ion in white and brown meats of crabs from different sites in Bahrain are listed in Table 10.

Table 10 Mean concentration of Zn^{2+} ion (mg/kg dry weight) in tissues of crabs from different sites.

Site	Gender	Tissue	N	Mean	St. Div	Ratio (W:B)
Ra's Al M'ttalah	Male	White	12	231.16	3.48	1 : 0.8
		Brown	12	176.59	2.83	
	Female	White	12	254.35	4.00	1 : 1
		Brown	11	248.21	3.04	
Marwada	Male	White	12	239.08	4.00	1 : 1.1
		Brown	12	256.81	2.84	
	Female	White	12	255.32	4.12	1 : 1.2
		Brown	12	300.45	6.54	
Tubli Bay	Male	White	12	209.25	2.92	1 : 0.8
		Brown	11	174.56	2.81	
	Female	White	11	208.27	2.68	1 : 1.1
		Brown	12	232.53	2.83	
Askar	Male	White	12	240.25	3.26	1 : 0.8
		Brown	12	198.11	3.36	
	Female	White	12	248.84	3.32	1 : 1.2
		Brown	10	295.92	4.00	
Overall	Male	White	48	229.94	13.01	1 : 0.9
		Brown	47	202.09	33.83	
	Female	White	47	242.41	19.55	1 : 1.1
		Brown	45	268.56	30.38	

Copper (II) ion was accumulated in brown meat more than white meat by around three times in male crabs while it was more than double in female crabs. The range of Cu^{2+} ion in white meat of overall male crabs was 33.28 – 87.42 mg/kg dry weight, while it was 40.81 – 325.71 mg/kg dry weight in brown meat with an average equal to 67.99 mg/kg and 213.88 mg/kg, respectively. In the overall female crabs, Cu^{2+} ion range was 36.92 – 95.25 mg/kg in white meat and 97.38 – 312.90 mg/kg in brown meat with an average equal to 70.99 and 184.36 mg/kg, respectively. The mean concentration, standard deviation and the ratio of Cu^{2+} ions in white and brown meats of crabs from different sites in Bahrain are listed in Table 11.

Table 11 Mean concentration of Cu^{2+} ions (mg/kg dry weight) in tissues of crabs from different sites.

Site	Gender	Tissue	N	Mean	SD	Ratio (W:B)
Ra's Al M'ttalah	Male	White	12	87.42	1.19	1 : 3.4
		Brown	12	298.70	3.31	
	Female	White	12	95.25	1.36	1 : 3.3
		Brown	12	312.90	4.88	
Marwada	Male	White	12	77.04	0.85	1 : 2.4
		Brown	12	185.18	1.76	
	Female	White	12	72.35	0.98	1 : 2.7
		Brown	11	195.09	1.52	
Tubli Bay	Male	White	12	33.28	0.31	1 : 1.2
		Brown	11	40.81	0.37	
	Female	White	11	36.92	0.40	1 : 2.6
		Brown	12	97.38	0.96	
Askar	Male	White	12	74.23	1.21	1 : 4.4
		Brown	11	325.71	2.78	
	Female	White	12	76.60	0.72	1 : 1.7
		Brown	12	132.96	2.18	
Overall	Male	White	48	67.99	20.87	1 : 3.1
		Brown	46	213.88	111.90	
	Female	White	47	70.99	20.99	1 : 2.6
		Brown	47	184.36	83.72	

Chromium (III) ion was accumulated in both brown meat and white meat, in general, female accumulated Cr^{3+} ions in brown meat more than male crabs. In some cases Cr^{3+} ion was found in brown meat more, like female and male crabs in Tubli Bay and female crabs in Marwada. However, there were some cases where Cr^{3+} ion was accumulated more in white meat like female crabs in Askar and male crabs in both Ra's Al M'ttalah and Marwada sites. The range of Cr^{3+} ions in white meat of overall male crabs was 0.062 – 0.227 mg/kg dry weight, while it was 0.032 – 0.202 mg/kg dry weight in brown meat with an average equal to 0.135 mg/kg and 0.130 mg/kg, respectively. In the overall female crabs, Cr^{3+} ions range was 0.025 – 0.124 mg/kg in white meat and 0.040 – 0.254 mg/kg in brown meat with an average equal to 0.083 and 0.135 mg/kg, respectively. The mean concentration, standard deviation and the ratio of Cr^{3+} ion in white and brown meats of crabs from different sites in Bahrain are listed in Table 12.

Table 12 Mean concentration of Cr^{3+} ion (mg/kg dry weight) in tissues of crabs from different sites.

Site	Gender	Tissue	N	Mean	SD	Ratio (W:B)
Ra's Al M'ttalah	Male	White	11	0.157	0.055	1 : 0.7
		Brown	11	0.114	0.036	
	Female	White	12	0.124	0.060	1 : 1.1
		Brown	11	0.141	0.018	
Marwada	Male	White	12	0.062	0.023	1 : 0.5
		Brown	12	0.032	0.018	
	Female	White	11	0.025	0.017	1 : 2.6
		Brown	12	0.066	0.018	
Tubli Bay	Male	White	11	0.095	0.013	1 : 2.1
		Brown	11	0.202	0.012	
	Female	White	11	0.070	0.013	1 : 3.6
		Brown	12	0.254	0.018	
Askar	Male	White	12	0.227	0.136	1 : 0.8
		Brown	12	0.178	0.020	
	Female	White	12	0.107	0.036	1 : 0.4
		Brown	7	0.040	0.007	
Overall	Male	White	46	0.135	0.098	1 : 1
		Brown	46	0.130	0.071	
	Female	White	46	0.083	0.053	1 : 1.6
		Brown	42	0.135	0.086	

Results show that the Ni^{2+} ion was accumulated in brown meat more than white meat by more than seven times. The brown meat of male and females crabs at Marwada site accumulated Ni^{2+} ions more than the other sites. In Tubli Bay and Askar sites, males accumulated Ni^{2+} ions in their brown meat more than female crabs. The range of Ni^{2+} ions in white meat of overall male crabs was 0.111 – 0.192 mg/kg dry weight, while it was 0.831 – 1.771 mg/kg dry weight in brown meat with an average equal to 0.148 mg/kg and 1.152 mg/kg, respectively. In the overall female crabs, Ni^{2+} ions range was 0.145 – 0.202 mg/kg in white meat and 0.421 – 3.028 mg/kg in brown meat with an average equal to 0.170 and 1.348 mg/kg, respectively. The mean concentration, standard deviation and the ratio of Ni^{2+} ions in white and brown meats of crabs from different sites in Bahrain are listed in Table 13.

Table 13 Mean concentration of Ni^{2+} ions (mg/kg dry weight) in tissues of crabs from different sites.

Site	Gender	Tissue	N	Mean	SD	Ratio (W:B)
Ra's Al M'ttalah	Male	White	11	0.1700	0.0613	1 : 6
		Brown	10	0.9755	0.0129	
	Female	White	12	0.1470	0.0413	1 : 6
		Brown	11	0.9449	0.0126	
Marwada	Male	White	12	0.1198	0.0211	1 : 15
		Brown	12	1.7711	0.0185	
	Female	White	12	0.1446	0.0153	1 : 21
		Brown	11	3.0281	0.0926	
Tubli Bay	Male	White	11	0.1114	0.0299	1 : 7
		Brown	11	0.8311	0.0154	
	Female	White	12	0.2018	0.0953	1 : 4
		Brown	10	0.7784	0.0144	
Askar	Male	White	11	0.1918	0.0426	1 : 5
		Brown	11	0.9591	0.0369	
	Female	White	12	0.1882	0.0114	1 : 2
		Brown	9	0.4207	0.0093	
Overall	Male	White	45	0.148	0.052	1 : 7.8
		Brown	44	1.152	0.388	
	Female	White	48	0.170	0.057	1 : 7.9
		Brown	41	1.348	1.048	

Cadmium (II) ion was accumulated mostly in brown meat, which was more than white meat by at least thirteen times. Males found to accumulate more Cd^{2+} ions in their brown meat than female crabs. Tubli bay site found the lowest level of Cd^{2+} ions compared with the other sites. The range of Cd^{2+} ion in white meat of overall male crabs was 0.004 – 0.636 mg/kg dry weight, while it was 0.402 – 10.309 mg/kg dry weight in brown meat with an average equal to 0.265 and 5.826 mg/kg, respectively. In the overall female crabs, Cd range was 0.001 – 0.665 mg/kg in white meat and 0.266 – 8.350 mg/kg in brown meat with an average equal to 0.281 and 3.917 mg/kg, respectively. The mean concentration, standard deviation and the ratio of Cd^{2+} ions in white and brown meats of crabs from different sites in Bahrain are listed in Table 14.

Table 14 Mean concentration of Cd^{2+} ions (mg/kg dry weight) in tissues of crabs from different sites.

Site	Gender	Tissue	N	Mean	SD	Ratio (W:B)
Ra's Al M'ttalah	Male	White	11	0.636	0.0326	1 : 13
		Brown	12	8.054	0.3126	
	Female	White	11	0.665	0.0370	1 : 13
		Brown	11	8.350	0.3648	
Marwada	Male	White	11	0.229	0.0241	1 : 45
		Brown	12	10.309	0.4781	
	Female	White	12	0.166	0.0227	1 : 30
		Brown	12	5.032	0.3010	
Tubli Bay	Male	White	12	<0.020	-	1 : >20
		Brown	10	0.402	0.0227	
	Female	White	8	<0.020	-	1 : >20
		Brown	12	0.266	0.0331	
Askar	Male	White	10	0.211	0.0153	1 : 17
		Brown	12	3.635	0.1629	
	Female	White	12	0.232	0.0174	1 : 10
		Brown	11	2.251	0.0968	
Overall	Male	White	44	0.265	0.236	1 : 22
		Brown	46	5.826	3.819	
	Female	White	43	0.281	0.242	1 : 14
		Brown	46	3.917	3.070	

Lead (II) ion was below the quantification limit of the AAS, which was <0.20 mg/kg in all sites.

In order to compare the level of the trace metal ions in the current study with other studies that recorded their results based on wet weight, the water content of the crab's meats was estimated, Table 15. The water content in white meat of overall male and female crabs was 79.7% and 80.2%, respectively, whereas in brown meat it was 80.4% and 79.3% for overall males and females, respectively.

The concentrations of trace metal ions were normalized with the percentage of the water content of tissues to estimate their concentration based on wet weight, the normalized results are listed in Table 16. Also, the results should be based on wet weight to be comparable with different standards for the suitability of the crab meat to human consumption.

Table 15 The mean percentage of water content in different tissues of crabs.

Sample	Gender	Tissue	Water Content (%)	Standard Deviation
Ra's Al M'ttalah	Male	White	79.76	1.66
		Brown	78.47	1.07
	Female	White	75.70	0.97
		Brown	80.15	0.41
Marwada	Male	White	80.61	2.49
		Brown	84.35	0.99
	Female	White	78.55	0.07
		Brown	78.59	0.24
Tubli Bay	Male	White	76.39	9.03
		Brown	80.74	0.33
	Female	White	83.60	0.21
		Brown	79.80	0.77
Askar	Male	White	81.85	1.23
		Brown	81.93	0.72
	Female	White	82.90	0.75
		Brown	78.80	0.42

The N value for each tissue in every gender in all sites was 3

Table 16 Normalized concentrations (mg/kg wet weight) of the trace metal ions in different gender and tissues of BSC in Bahrain.

Site	Gender	Tissue	Zn ²⁺	Cu ²⁺	Cr ³⁺	Ni ²⁺	Cd ²⁺
Ra's Al M'ttalah	Male	White	46.79	17.69	0.032	0.034	0.129
		Brown	38.02	64.31	0.025	0.210	1.734
	Female	White	61.81	23.14	0.030	0.036	0.162
		Brown	49.27	62.11	0.028	0.188	1.658
Marwada	Male	White	51.28	16.52	0.013	0.026	0.049
		Brown	40.19	28.98	0.005	0.277	1.613
	Female	White	54.77	15.52	0.005	0.031	0.036
		Brown	64.33	41.77	0.014	0.648	1.077
Tubli Bay	Male	White	49.40	7.86	0.022	0.026	<0.02
		Brown	33.62	7.86	0.039	0.160	0.077
	Female	White	34.16	6.05	0.011	0.033	<0.02
		Brown	46.97	19.67	0.051	0.157	0.054
Askar	Male	White	43.60	13.47	0.041	0.035	0.038
		Brown	35.80	58.86	0.032	0.173	0.657
	Female	White	42.55	13.10	0.018	0.032	0.040
		Brown	62.73	28.19	0.008	0.089	0.477
Average	Male	White	46.68	13.80	0.027	0.030	0.054
		Brown	37.59	39.78	0.024	0.214	1.084
	Female	White	48.00	14.06	0.016	0.034	0.056
		Brown	53.18	36.50	0.027	0.267	0.776
Overall		White	48.05	14.17	0.022	0.032	0.057
		Brown	46.37	38.97	0.025	0.238	0.918

In the second part, the average carapace width (CW) of overall male crabs was 10.60 ± 0.47 cm with a range of 10.01 – 11.73 cm, whereas the average CW of overall female crabs was 10.40 ± 0.38 cm, with a range of 10.02 – 11.39 cm. The weight of the male crabs found to be higher than the female crabs. More description data for the crab samples collected from the sites under this study, such as carapace width (CW), carapace length (CL) and weight are shown in Table 17.

Table 17 Description data of the crab's samples used in second part.

Site	Gender		CW (cm)	CL (cm)	Weight (g)
Ra's Al M'ttalah	Male	Average	10.54	6.73	182
		SD	0.33	0.26	26
	Female	Average	10.14	6.58	138
		SD	0.23	0.15	11
Marwada	Male	Average	10.43	6.56	170
		SD	0.38	0.22	19
	Female	Average	10.38	6.72	149
		SD	0.26	0.23	17
Tubli Bay	Male	Average	11.12	7.01	212
		SD	0.43	0.46	37
	Female	Average	10.58	6.63	150
		SD	0.46	0.37	26
Askar	Male	Average	10.29	6.51	168
		SD	0.26	0.19	18
	Female	Average	10.51	6.75	159
		SD	0.41	0.29	26
Overall	Male	Average	10.60	6.70	183
		SD	0.47	0.35	31
	Female	Average	10.40	6.67	149
		SD	0.38	0.27	22

The N value for each gender in all sites was 10

Results have shown that Zn^{2+} ion was accumulated in both white and brown meats in almost a close concentration with a slight difference. Female crabs accumulated Zn^{2+} ion in brown and white meats more than male crabs. The range of Zn^{2+} ion in white meat of overall male crabs was 19.5-97.1 mg/kg wet weight, while it was 25.0-115.0 mg/kg wet weight in brown meat with an average equal to 47.1 mg/kg and 48.2 mg/kg, respectively. In overall female crabs, Zn^{2+} ion range was 29.8-97.2 mg/kg wet weight in white meat and 28.1-136.9 mg/kg wet weight in brown meat with an average equal to 60.1 and 70.6 mg/kg, respectively. The mean concentration, standard deviation and, minimum and maximum levels of Zn^{2+} ion in white and brown meats of crabs from different sites in Bahrain are listed in Table 18.

Table 18 Mean concentration of Zn^{2+} ion (mg/kg wet weight) in tissues of crabs from different sites.

Sample	Gender	Tissue	Mean	SD	Minimum	Maximum
Ra's Al M'ttalah	Male	White	45.14	7.66	37.09	59.07
		Brown	46.80	28.68	25.02	115.03
	Female	White	55.41	8.10	42.00	66.66
		Brown	38.11	5.53	28.12	49.62
Marwada	Male	White	55.47	15.42	37.28	79.23
		Brown	45.63	15.49	26.91	76.99
	Female	White	75.23	11.25	52.68	92.25
		Brown	84.01	31.00	41.20	128.37
Tubli Bay	Male	White	35.71	6.75	19.52	43.65
		Brown	52.08	27.81	27.59	111.21
	Female	White	47.32	20.53	29.77	97.21
		Brown	94.60	32.89	42.91	136.87
Askar	Male	White	52.11	19.62	28.21	97.09
		Brown	48.14	16.50	34.11	88.90
	Female	White	62.56	12.58	45.76	88.05
		Brown	65.67	20.23	42.45	92.40
Overall	Male	White	47.1	15.0	19.5	97.1
		Brown	48.2	22.2	25.0	115.0
	Female	White	60.1	16.9	29.8	97.2
		Brown	70.6	32.3	28.1	136.9

The N value for each case was 10

The results of Cu^{2+} ion concentrations showed that Cu^{2+} ion was accumulated in brown meat more than white meat. The range of Cu^{2+} ion in white meat of overall male crabs was 5.2-30.3 mg/kg wet weight, while it was 5.9-220.2 mg/kg wet weight in brown meat with an average equal to 12.8 mg/kg and 48.6 mg/kg, respectively. In overall female crabs, Cu^{2+} ion range was 7.8-28.3 mg/kg wet weight in white meat and 10.2-143.6 mg/kg wet weight in brown meat with an average equal to 15.7 and 49.7 mg/kg, respectively. The mean concentration, standard deviation and, minimum and maximum levels of Cu^{2+} ion in white and brown meats of crabs from different sites in Bahrain are listed in Table 19.

Table 19 Mean concentration of Cu^{2+} ion (mg/kg wet weight) in tissues of crabs from different sites.

Sample	Gender	Tissue	Mean	SD	Minimum	Maximum
Ra's Al M'ttalah	Male	White	12.32	3.73	5.94	17.34
		Brown	99.88	61.65	12.26	220.19
	Female	White	16.92	3.13	12.79	23.71
		Brown	88.05	30.87	32.15	143.57
Marwada	Male	White	13.17	5.39	7.20	22.32
		Brown	22.32	14.00	10.34	58.54
	Female	White	15.91	4.31	11.78	26.40
		Brown	44.73	18.20	26.39	77.02
Tubli Bay	Male	White	12.77	8.19	5.24	30.30
		Brown	37.65	23.89	5.91	81.16
	Female	White	14.84	6.93	7.80	28.31
		Brown	33.02	17.55	10.18	64.03
Askar	Male	White	12.87	3.53	7.37	16.39
		Brown	34.74	32.95	6.70	123.41
	Female	White	15.31	3.59	10.37	20.94
		Brown	33.01	18.54	11.04	63.93
Overall	Male	White	12.8	5.3	5.2	30.3
		Brown	48.6	47.3	5.9	220.2
	Female	White	15.7	4.6	7.8	28.3
		Brown	49.7	31.2	10.2	143.6

The N value for each case was 10

Cadmium (II) ion was accumulated in brown meat more than white meat. The range of Cd^{2+} ion in white meat of overall male crabs was 0.001-0.306 mg/kg wet weight, while it was 0.02-37.57 mg/kg wet weight in brown meat with an average equal to 0.02 mg/kg and 3.05 mg/kg, respectively. In overall female crabs, Cd^{2+} ion range was 0.001-0.106 mg/kg wet weight in white meat and 0.02-13.52 mg/kg wet weight in brown meat with an average equal to 0.014 and 2.91 mg/kg, respectively. The mean concentration, standard deviation and, minimum and maximum levels of Cd^{2+} ion in white and brown meats of crabs from different sites in Bahrain are listed in Table 20.

Table 20 Mean concentration of Cd^{2+} ion (mg/kg wet weight) in tissues of crabs from different sites.

Sample	Gender	Tissue	Mean	SD	Minimum	Maximum
Ra's Al M'ttalah	Male	White	0.0250	0.0201	0.0068	0.0628
		Brown	8.34	10.86	0.85	37.57
	Female	White	0.0138	0.0067	0.0061	0.0231
		Brown	6.20	3.62	1.86	13.00
Marwada	Male	White	0.0119	0.0128	0.0022	0.0469
		Brown	1.65	1.44	0.23	4.40
	Female	White	0.0299	0.0314	0.0032	0.1060
		Brown	2.92	3.89	0.18	13.52
Tubli Bay	Male	White	0.0011	0.0005	0.0005	0.0020
		Brown	0.10	0.06	0.02	0.20
	Female	White	0.0011	0.0005	0.0006	0.0021
		Brown	0.14	0.14	0.02	0.49
Askar	Male	White	0.0412	0.0937	0.0023	0.3063
		Brown	2.10	1.59	0.81	6.27
	Female	White	0.0111	0.0122	0.0042	0.0444
		Brown	2.37	1.47	0.52	5.44
Overall	Male	White	0.020	0.049	0.001	0.306
		Brown	3.05	6.20	0.02	37.57
	Female	White	0.014	0.020	0.001	0.106
		Brown	2.91	3.44	0.02	13.52

The N value for each case was 10

To investigate the differences of Zn^{2+} , Cu^{2+} and Cd^{2+} accumulation based on the tissue type, paired t-test was applied and the results are shown in Table 21. Results showed that both Cu^{2+} and Cd^{2+} were accumulated in brown meat significantly more than in white meat in all sites. Two exceptions were observed, in which the male crabs in Askar and Marwada sites showed no significant differences. In the case of Zn^{2+} , no significant differences were reported between its levels in white and brown meats in general. Two exceptions were reported, female crabs in the Tubli Bay site accumulated Zn^{2+} in brown meat significantly more than white meat. The second case was in Ra's Al M'ttalah where female crabs accumulated Zn^{2+} in white meat significantly more than brown meat. Generally, the overall result of this test showed no significant differences were observed among Zn^{2+} , but Cu^{2+} and Cd^{2+} found to be accumulated in the brown meat significantly higher than the white meat.

Table 21 The p-value of the *Paired T-Test* for the difference of trace metal ions accumulation based on the tissue type.

Site	Gender	Zn^{2+}	Cu^{2+}	Cd^{2+}
Ra's Al M'ttalah	Male	0.846	0.002	0.039
	Female	<0.001	<0.001	<0.001
	Overall	0.120	<0.001	0.001
Marwada	Male	0.286	0.111	0.006
	Female	0.401	<0.001	0.042
	Overall	0.939	<0.001	0.003
Tubli Bay	Male	0.112	0.004	<0.001
	Female	0.002	0.016	0.009
	Overall	0.001	<0.001	<0.001
Askar	Male	0.649	0.072	0.003
	Female	0.699	0.020	0.001
	Overall	0.940	0.004	<0.001
All	Male	0.814	<0.001	0.004
	Female	0.064	<0.001	<0.001
Overall Sample	Both male and female	0.109	<0.001	<0.001

There is a significant difference of accumulation if p-value < 0.05

No significant difference observed
Accumulation in white meat is significantly more than brown meat
Accumulation in brown meat is significantly more than white meat

A Two-way ANOVA test was applied to investigate the differences in the Zn^{2+} , Cu^{2+} and Cd^{2+} levels based on genders and sites, and the results are shown in Table 22. Results showed that the Cu^{2+} in white meat and Zn^{2+} in both white and brown meats found to be accumulated by female crabs significantly more than male crabs. On the other hand, no significant differences were observed in the accumulation of Cu^{2+} in brown meat and Cd^{2+} in both white and brown meats between male and female crabs.

Moreover, the results showed significant differences among the sites and the accumulation of Zn^{2+} , Cu^{2+} and Cd^{2+} in brown meat. In the case of white meat, only Zn^{2+} showed significant differences based on site, whereas there were no significant differences among Cu^{2+} and Cd^{2+} .

Table 22 The p-value of the Two-way ANOVA test for the difference of metal ions accumulation based on the site and gender.

Tissue Type	Comparison	Zn^{2+}	Cu^{2+}	Cd^{2+}
White	Genders	<0.001	0.011	0.481
	Sites	<0.001	0.951	0.161
Brown	Genders	<0.001	0.879	0.886
	Sites	0.003	<0.001	<0.001

There is a significant difference of accumulation if p-value < 0.05

Accumulation in female crabs is significantly more than male crabs.
No significant difference observed
There is a significant difference in the metal ions accumulation based on site.

The Tukey Pairwise Comparisons were used to investigate the differences within the sites and the results are shown in Table 23. In the white meat, the decreasing order of the Zn^{2+} levels were as following Marwada > Askar > Ra's Al M'ttalah > Tubli Bay. In brown meat, the level of Cu^{2+} and Cd^{2+} were found to be in Ra's Al M'ttalah significantly higher than the other sites while no significant differences were observed between the other sites in white meat. In Zn concentration, the decreasing order was found to be as the following Marwada and Tubli Bay > Askar > Ra's Al M'ttalah.

Table 23 Grouping information using the Tukey method for the difference of metal ions accumulation based on the site.

Tissue	Site	Zn ²⁺	Cu ²⁺	Cd ²⁺
White	Ra's Al M'ttalah	BC		
	Marwada	A		
	Tubli Bay	C		
	Askar	AB		
Brown	Ra's Al M'ttalah	B	A	A
	Marwada	A	B	B
	Tubli Bay	A	B	B
	Askar	AB	B	B

A, B, C and D are the grouping according to the Tukey Method

No significant difference observed
Highest concentration than the others
The second highest concentration than the others
The second lowest concentration
Lowest concentration

The Pearson correlation test indicated a significance moderate positive correlation between Cd²⁺-Cu²⁺ in the brown meat of BSC, where the r-value was 0.587 and the p-value was <0.001. No more correlations indicated. Details about the Pearson correlation of the metal ions and the level of significance of this correlation is found in the matrix in Figure 15. The correlation coefficient (r-value) is high if its value in the range of 0.7-1.0 and it is moderate if the range is 0.5-0.69, whereas p-value represents the Level of Significance of this correlation.

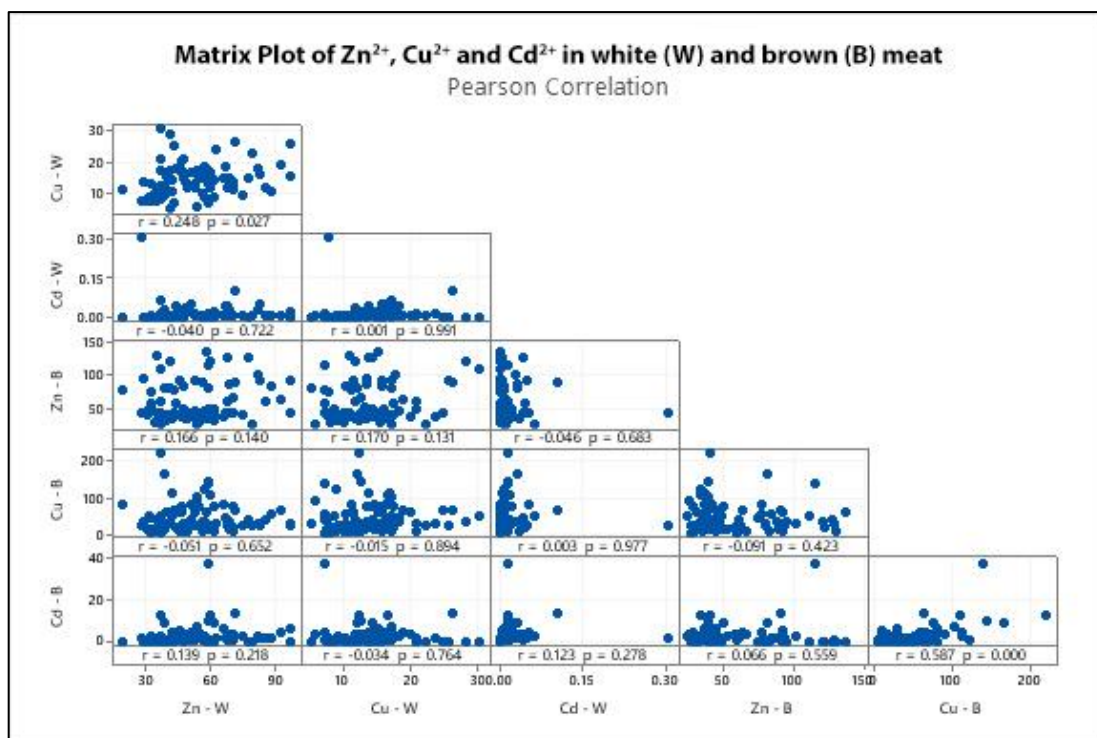


Figure 15 Pearson correlation of the trace metal ions within BSC tissues.

The results of the recovery percentage of the Certified Reference Materials (CRM) that were used in the first and second parts of the current study are shown in Table 24 and Table 25, respectively. The overall recovery percentage of the CRMs used ranged from 92 to 108% for all trace metal ions.

Table 24 Recovery values of CRM trace metal ions within the first part

Metal ions	IAEA-436			IAEA-452		
	Certified Value	Measured	Recovery	Certified Value	Measured	Recovery
Zn ²⁺	19.0	18.03	95%	167	168.67	101%
Cu ²⁺	-	-	-	11.0	10.67	97%
Ni ²⁺	0.069	0.064	93%	-	-	-
Cr ³⁺	0.194	0.182	94%	5.2	4.78	92%
Pb ²⁺	-	-	-	2.4	2.26	94%
Cd ²⁺	0.052	0.048	92%	30.5	30.19	99%

Table 25 Recovery values of CRM trace metal ions within the second part

Meta l ions	IAEA-436			IAEA-452			NIST 2976		
	Certified Value	Measured	Recovery	Certified Value	Measured	Recovery	Certified Value	Measured	Recovery
Zn ²⁺	19.0	18.17	96%	167	160.91	96%	137	130.0	95%
Cu ²⁺	-	-	-	11.0	10.76	98%	4.02	4.18	104%
Cd ²⁺	0.052	0.052	100%	30.5	29.18	96%	0.82	0.88	108%

4.2 Discussion

The first part of the current study aimed to determine the concentration of Zn^{2+} , Cu^{2+} , Ni^{2+} , Cr^{3+} , Pb^{2+} and Cd^{2+} in different tissues of male and female BSC. The results obtained were compared with different marine organisms studied in Bahrain, and then compared to the levels of these trace metal ions in the BSC's tissues in the Arabian Gulf and other sea areas. As well as, the suitability of these crabs for human consumption was investigated. In the second part of the current study, the differences based on the type of tissues and genders were studied, in addition, the differences based on the site location.

4.2.1 The Levels of Trace Metal Ions in BSC

The results showed higher levels of both Zn^{2+} and Cu^{2+} compared to the rest of trace metal ions; Cr^{3+} , Ni^{2+} , Pb^{2+} and Cd^{2+} . Lead (Pb^{2+}) was found to be below the quantification limit, so it was excluded from the comparison and the trace metal ions pattern.

The dominant pattern of the trace metals under current study is $\text{Zn}^{2+} > \text{Cu}^{2+} > \text{Cd}^{2+} > \text{Ni}^{2+} > \text{Cr}^{3+}$, which is, also, the pattern of overall white and brown meats as shown in Table 26. Tubli Bay site was found to have a different pattern of both genders and tissues than the main pattern. In Tubli Bay the white meat in both male and female found to be in the order of $\text{Zn}^{2+} > \text{Cu}^{2+} > \text{Ni}^{2+} > \text{Cr}^{3+} > \text{Cd}^{2+}$ whereas brown meat of both male and female showed the following order $\text{Zn}^{2+} > \text{Cu}^{2+} > \text{Ni}^{2+} > \text{Cd}^{2+} > \text{Cr}^{3+}$. Male crabs in the Askar site was showed a different order of trace metal ions than the other sites, which was in the order of $\text{Zn}^{2+} > \text{Cu}^{2+} > \text{Cr}^{3+} > \text{Cd}^{2+} > \text{Ni}^{2+}$ and $\text{Zn}^{2+} > \text{Cu}^{2+} > \text{Cd}^{2+} > \text{Ni}^{2+} > \text{Cr}^{3+}$ for white and brown meats, respectively.

The main pattern of trace metals in the current study is similar to that reported in BSC in Turkey (Ayas & Özogul, 2011; Ayas, 2013), However, differences in patterns were reported in different areas, Table 27.

Table 26 The order of the trace metal ions in different tissues and genders of BSC in Bahrain.

Site	Gender	Tissue	Pattern
Ra's Al M'ttalah	Male	White	$Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$
		Brown	$Cu^{2+} > Zn^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$
	Female	White	$Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$
		Brown	$Cu^{2+} > Zn^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$
Marwada	Male	White	$Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$
		Brown	$Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$
	Female	White	$Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$
		Brown	$Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$
Tubli Bay	Male	White	$Zn^{2+} > Cu^{2+} > Ni^{2+} > Cr^{3+} > Cd^{2+}$
		Brown	$Zn^{2+} > Cu^{2+} > Ni^{2+} > Cd^{2+} > Cr^{3+}$
	Female	White	$Zn^{2+} > Cu^{2+} > Ni^{2+} > Cr^{3+} > Cd^{2+}$
		Brown	$Zn^{2+} > Cu^{2+} > Ni^{2+} > Cd^{2+} > Cr^{3+}$
Askar	Male	White	$Zn^{2+} > Cu^{2+} > Cr^{3+} > Cd^{2+} > Ni^{2+}$
		Brown	$Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$
	Female	White	$Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$
		Brown	$Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$
Average	Male	White	$Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$
		Brown	$Cu^{2+} > Zn^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$
	Female	White	$Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$
		Brown	$Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$
Overall		White	$Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$
		Brown	$Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$

Similar colors represent similar order

Table 27 The comparison of the order of trace metal ions in BSC tissues from Bahrain and the other regions.

Area	Gender	Tissue	Pattern	Reference
Bahrain	Male	White	$Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{2+}$	Current study
		Brown	$Cu^{2+} > Zn^{2+} > Cd^{2+} > Ni^{2+} > Cr^{2+}$	
	Female	White	$Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{2+}$	
		Brown	$Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{2+}$	
Bahrain	-	-	$As^* > Pb > Cd > Hg$	(Al-Madany et al., 1996)
Bahrain	Male	-	$Zn > Cu > Pb > Hg > Cd$	(Musaiger & Al-Rumaidh, 2005)
	Female	-	$Zn > Cu > Pb = Hg > Cd$	
Kuwait	Mixed	White	$Zn > Cu > Pb > Cr$	(Al-Mohanna & Subrahmanyam, 2001)
		Brown	$Zn > Cu > Pb > Cr$	
Turkey	Male	Carapace meat	$Zn > Cu > Cd > Cr$	(Ayas & Özogul, 2011)
	Female	Carapace meat	$Zn > Cu > Cd > Cr$	
Turkey	Male	Chela crab meat	$Zn > Cu > Cd > Cr$	(Ayas, 2013)
		Lamp crab meat	$Zn > Cu > Cd > Cr$	
	Female	Chela crab meat	$Zn > Cu > Cd > Cr$	
		Lamp crab meat	$Zn > Cu > Cd > Cr$	
India	-	Muscle	$Cu > Zn > Cr > Cd$	(Viswanathan et al., 2013)
		Hepatopancreas	$Zn > Cu > Cr > Cd$	
India	-	Muscle	$Cu > Cr > Ni > Cd$	(Kumar et al., 2019)
		Hepatopancreas	$Cu > Cd > Ni > Cr$	
Malaysia	-	Muscle - Carapace	$Zn > Cu > Cr > Pb > Ni > Cd$	(Anandkumar et al., 2019)
		Muscle - Walking leg	$Zn > Cu > Cr > Pb > Ni > Cd$	

* As is metalloid

Zinc metal ion was found to be the highest in almost all of the cases, exceptions to this were reported in the white meat of male crabs in Askar site and brown meat of both male and female of Ra's Al M'ttalah site. In these cases, Cu^{2+} concentration was found to be more than Zn^{2+} ; Table 10 and Table 11. The average concentration of Zn^{2+} in whole white meat was 235.8 mg/kg where female crabs in Tubli Bay have the lowest concentration (208.2 mg/kg) and female crabs in Marwada have the highest content (255.3 mg/kg). However, in brown meat which was close to white meat, the average concentration 235.4 mg/kg where male crabs in Tubli Bay site have the lowest while female crabs in Marwada have the highest content of Zn^{2+} , with a concentration of 174.6 and 300.4 mg/kg, respectively.

These levels of Zn^{2+} are higher than the other studies conducted in the Arabian Gulf and other regions as shown in Table 28 and Table 29. For example, in Iran Zn^{2+} concentration in the muscles of BSC was found at 48.34 mg/kg dry weight (Koosej et al., 2016), while it was 66.6 mg/kg in the whole body of crabs (Heidarieh et al., 2013). However, in Kuwait, Al-Mohanna and Subrahmanyam (2001) reported Zn^{2+} level in muscles and hepatopancreas at 206.2 mg/kg dry weight (188.7-208.3 mg/kg) and 160.6 mg/kg (155.0-165.6 mg/kg), respectively, which is the closest level within the Arabian Gulf to the current study. Outside the Arabian Gulf, in Mersin Bay (Turkey), Ayas and Ozogul (2011) reported Zn^{2+} at 102.5 and 107 mg/kg dry weight in carapace meat of male and female crabs, respectively, whereas it was reported at 117 and 130 mg/kg in chela tissue of male and female crabs (Ayas, 2013). On the other hand, Zn^{2+} was reported higher than the current study. For example, in Malaysia, Anandkumar et al. (2019) reported Zn^{2+} at 394 and 319 mg/kg dry weight in muscles of the carapace and walking legs, respectively. Moreover, Olgunoğlu and Olgunoğlu

(2016) reported an elevated levels of Zn^{2+} in *Portunus segnis* species in Turkey, which was found to have a mean of 340.2 mg/kg wet weight.

Copper trace metal ions were found to be high in all sites, but lower than Zn^{2+} with exception of some cases that were found to have Cu^{2+} more than Zn^{2+} ; Table 11. Overall Cu^{2+} concentration in white meat was 69.1 mg/kg, where male crabs in Tubli Bay site have the lowest concentration (33.3 mg/kg) and female crabs in Ra's Al M'ttalah the highest content (95.2 mg/kg). However, Cu^{2+} concentration in brown meat was found to be more than 2-fold of white meat content.

These levels of Cu^{2+} are higher than the other reported studies conducted in other regions as shown in Table 28 and Table 29. For example, in Turkey (Mersin Bay) muscles tissue content of Cu^{2+} was 29.98 (29.3-31.1) and 26.9 (25.4-28.2) mg/kg dry weight for male and female of BSC (Ayas & Özogul, 2011). Besides, in muscles of chela in male and female of crabs Cu^{2+} was 44.5 (26.7-72.5) and 43.7 (25.1-70.6) mg/kg, respectively, in Mersin Bay (Ayas, 2013). However, higher level than the level in white meat of the current study was reported in the other areas. For example, in Kuwait it was around 123.99 (110.2-142.8) mg/kg in the muscles of the same species (Al-Mohanna & Subrahmanyam, 2001). In Malaysia, Cu^{2+} content of walking leg muscles found to be at 85.7 mg/kg (Anandkumar et al., 2019), which are greater than the current study. In addition, Kumar et al. (2019) reported that Cu in the range of <0.01-156.8 mg/kg in muscles tissues.

On the other hand, in the case of brown meat, the current study reported the highest level of Cu^{2+} in the Arabian Gulf. For example, in Kuwait Cu^{2+} was reported at 50 mg/kg with a range of 46.2-52.5 mg/kg (Al-Mohanna & Subrahmanyam, 2001). In the other sea areas, Kumar et al. (2019) reported that Cu^{2+} in the range up to 222.92 mg/kg in hepatopancreas tissues of BSC in India. Moreover, in Mersin Bay (Turkey)

elevated level of Cu^{2+} in *Portunus segnis* species was reported by Olgunoğlu and Olgunoğlu (2016), which was found to have a mean of 144.0 mg/kg wet weight.

These elevations of Zn^{2+} and Cu^{2+} concentrations in the crabs' tissues are proportionate to the concentration in the surrounding environment. For example, Zn^{2+} and Cu^{2+} were found in the range of 4.06-118.0 and 4.53-119.0 $\mu\text{g/L}$ in seawater, respectively (Juma & Al-Madany, 2008). Besides, they were in the range of 25.0-239 and 10.7-213 mg/kg in the marine sediment, respectively (Bersuder et al., 2020). Blue swimming crab found to have a very high Bioaccumulation Factor (BAF) for Zn^{2+} and Cu^{2+} ; 6825 and 7059, respectively (Anandkumar et al., 2019). The BAF estimated as the ratio of the metal concentration in the organisms tissues and its surrounding environment e.g. seawater or marine sediment (Gobas et al., 2009). In addition, Zn^{2+} is an essential element for the crabs that involves in protein synthesis, which increases at the time of molting (Anandkumar et al., 2019). On the other hand, Cu^{2+} is essential element for the respiratory process in the crabs because it is a component of the respiratory pigment of the crabs' gills; hemocyanin that is participating in the oxygen transportation (Engel & Brouwer, 1987). Hemocyanin synthesis takes place in the hepatopancreas of the crab (Rainer & Brouwer, 1993).

The decapod crustaceans like crabs, shrimps and lobsters found to have a kind of regulatory mechanism that maintains the concentration of Zn^{2+} and Cu^{2+} within their different tissues at certain levels. Even if they were exposed to a higher level from the surrounding environment until a specific threshold. This regulatory mechanism falls down when this threshold crossed and then these metal ions will start accumulating in different tissues of these organisms (White & Rainbow, 1982; Rainbow, 1985; Waiwood et al., 1987; Hilmy & Ghazaly, 1988; Nugegoda &

Rainbow, 1989; Uma Devi & Prabhakara Rao, 1989; Canli & Furness, 1993).

The uptake rate of Zn^{2+} and Cu^{2+} in decapod crustaceans found to be affected by the increase of salinity and temperature of the seawater (Chan et al., 1992). However, different studies found evidence for trace metal ions uptake (or toxicity) by marine and estuarine invertebrates increases at high temperature and low salinity. Whereas minimum (amount of trace metal ion) takes place at low temperature and high salinity (McLusky & Hagerman, 1987; Niyogi et al., 2016). Furthermore, the trace metal ions uptake rate in crustaceans increases in the period of molting (Rainbow, 1990). The content of the trace metal ions of muscles and hepatopancreas of crabs was found to be proportional directly to the conditions of the foods (Reichmuth et al., 2010). Zinc ions uptake by crabs found to be similar to that of Ca^{2+} uptake pathway (Niyogi et al., 2016), which increases during the postmoult period (Nørum et al., 2005).

The elevation of Zn^{2+} and Cu^{2+} in current study is not unique, elevated values in different marine organisms were reported in previous studies, Table 30. For example, Al-Sayed et al. (1996) reported Zn^{2+} in grouper fish (*E. coioides*) were in the range of 223 – 1253 mg/kg wet weight. Whereas de Mora et al. (2004) reported Zn^{2+} in the muscles and livers of the *E. coioides* in the range of 15.9 – 27.1 mg/kg dry weight and 183 – 421 mg/kg dry weight, respectively. Moreover, Pearl Oyster (*P. radiata*) was reported to accumulate high Zn^{2+} content; 333.8 mg/kg dry weight (Al-Sayed et al., 1994), 1825 mg/kg dry weight (de Mora et al., 2004) and 898 – 4607 mg/kg dry weight (Fowler et al., 1993). Furthermore, Zn^{2+} level in the Rock scallop (*Spondylus sp.*) was reported at 461 mg/kg dry weight by Fowler et al. (1993). Elevated level of Cu^{2+} was reported by de Mora et al. (2004) in the liver of orange grouper fish (*E. coioides*), which were found in the range of 75.5 – 276 mg/kg dry weight. Ali-Mohamed (2002) also

reported an elevated level of Cd^{2+} in brown algae (Spiral wrack, *Fucus spiralis*); 137.5 mg/kg dry weight in Zallaq site (west coastline of Bahrain).

Crabs accumulated Ni^{2+} mostly in the brown meat, which were around 8-fold its concentration in the white meat, Table 13. Overall Ni^{2+} concentration in white meat was 0.159 mg/kg were male crabs in Tubli Bay have the lowest concentration (0.111 mg/kg) and female crabs in Tubli Bay have the highest content (0.202 mg/kg). However, in brown meat, the average concentration of Ni^{2+} was 1.25 mg/kg where female crabs in Askar site have the lowest while female crabs in Marwada have the highest content of Ni^{2+} , with a concentration of 0.421 and 3.028 mg/kg, respectively.

Nickel metal ions level in the seawater of Bahrain were in the range of 0.71 and 20.1 $\mu\text{g/l}$ (Juma & Al-Madany, 2008) and in the range of 16.6 – 42.6 in the sediment (Bersuder et al., 2020). Anandkumar et al. (2019) report the BSC's bioaccumulation factor for Ni^{2+} around 35 in Malaysia. The level of Ni^{2+} reported in current study is lower than Ni^{2+} level in same species studies in the Arabian Gulf and other regions, Table 28 and Table 29. Hosseini et al. (2012b) reported Ni^{2+} in the north east coast of the Gulf in the muscles and hepatopancreas of BSC were in the range of 7.5 – 20.8 mg/kg dry weight and 6.3 – 13.8 mg/kg dry weight, respectively. Similarly, Ni^{2+} recorded at 393.2 mg/kg wet weight in the hepatopancreas of BSC (Rahmanpour et al., 2014). However, Koosej et al. (2016) reported Ni^{2+} in muscles at level slightly higher than what have been found in the current study (0.62 mg/kg dry weight). Outside the Arabian Gulf, Anandkumar et al. (2019) reported Ni^{2+} in the walking leg muscles to be around 3.1 mg/kg dry weight in Malaysia. Whereas in India, Ni^{2+} was reported in the range of <0.1 – 32.8 mg/kg and <0.01 – 223 mg/kg dry weight in the muscles and hepatopancreas of BSC (Kumar et al., 2019), respectively.

The level of Ni^{2+} in the current study was within the same range as most of the marine organisms studied in Bahrain, Table 30. Pearl oyster (*Pinctada radiata*) content of Ni^{2+} was in the range of 0.2 – 8.95 mg/kg dry weight (Fowler et al., 1993; Al-Sayed et al., 1994; de Mora et al., 2004), while rock scallop (*Spondylus sp.*) has higher level; 9.3 mg/kg (Fowler et al., 1993). Moreover, grouper fish found to have Ni^{2+} level in the range of <0.01 – 0.085 mg/kg dry weight (de Mora et al., 2004), and in the range of 0.7 – 9.0 mg/kg wet weight (Al-Sayed et al., 1996). On the other hand, the highest level of Ni^{2+} was reported by Ali-Mohamed (2002) in marine algae (*Hormophysa triquetra*), which was in the range of 4.17 – 22.36 mg/kg dry weight.

The accumulation of Cr^{3+} was not the same in all sites. There was a fluctuation in the Cr^{3+} levels among the genders and tissue types; Table 12. Overall Cr^{3+} concentrations in white and brown meats were 0.110 and 0.133 mg/kg dry weight, respectively. Chromium (III) ion concentration in the white meat was in the range of 0.025 – 0.227 mg/kg, whereas in brown meat was in the range of 0.032 – 0.254 mg/kg. Female crabs in Marwada and male crabs in Askar found to have the lowest and highest level of Cr^{3+} in the white meat, respectively. However, male crabs in Marwada and female crabs in Tubli Bay were found to have the lowest and highest level of Cr^{3+} in the brown meat, respectively.

Chromium's (III) ion bioaccumulation factor found to be 197 in Malaysia (Anandkumar et al., 2019). The level of Cr^{3+} reported in the current study is lower than the Cr^{3+} level in the same species studies in Arabian Gulf and other regions, Table 28 and Table 29. Within the north west of the Arabian Gulf sea area, Al-Mohanna and Subrahmanyam (2001) reported Cr^{3+} in the range of 0.15 to 0.66 mg/kg dry weight with an average of 0.48 mg/kg dry weight in the muscles. In the hepatopancreas tissues, it was in the range of 0.13 to 0.52 mg/kg with an average of 0.38 mg/kg dry

weight. Outside the Arabian Gulf, the Cr^{3+} level of the current study was also lower than values recorded in Turkey; 0.3-0.61 mg/kg dry weight (Ayas & Özogul, 2011), 0.31 – 0.53 mg/kg dry weight (Ayas, 2013). In Malaysia, Anandkumar et al. (2019) reported Cr^{3+} concentration at 5.1 and 7.2 mg/kg dry weight in carapace and walking leg muscles, respectively. Moreover, in India Cr^{3+} levels were in the range of 0.2 – 2.39 mg/kg wet weight (Karar et al., 2019). On the other hand, Viswanathan et al. (2013) reported lower Cr^{3+} content than the current study in India; 0.03 and 0.04 mg/kg dry weight in the muscle and hepatopancreas, respectively.

In Bahrain marine environment, Cr^{3+} level in the sediment were in the range of 25.0 – 71.1 mg/kg dry weight (Bersuder et al., 2020). Orange spotted grouper (*Epinephelus coioides*) was found to have lower level of Cr^{3+} than current study; 0.013 – 0.075 mg/kg dry weight (de Mora et al., 2004). But pearl oyster (*Pinctada radiata*) found to have higher level Cr^{3+} than the current study; 0.79 and 0.67 mg/kg dry weight (de Mora et al., 2004). Fowler et al. (1993) reported Cr^{3+} in the *Pinctada radiata* with an average of 0.34 and 0.43 mg/kg dry weight, and in rock scallop (*Spondylus* sp.) with an average of 2.8 mg/kg dry weight. Ali-Mohamed (2002) reported Cr^{3+} in marine algae (*Hormophysa triquetra*) in the range of 0.03 – 1.2 mg/kg dry weight.

Lead trace metal ion was not detected (<0.20 mg/kg) in the tissues of crabs in the current study. Lead trace metal ion was found to have a low bioaccumulation factor in BSC muscles; which was equal to 34 in Malaysia (Anandkumar et al., 2019). However, Pb reported in higher level than 0.20 mg/kg dry weight, which is the limit of quantification of current study; Table 28 and Table 29. For example, in Kuwait coast; Pb^{2+} concentration in BSC was ranged from 0.71 – 1.72 mg/kg dry weight (Al-Mohanna & Subrahmanyam, 2001), and in the north east coast of the Arabian Gulf it

was ranged from 0.10 – 2.85 mg/kg dry weight (Hosseini et al., 2012b). Abdel-Salam and Hamdi (2014) reported Pb^{2+} in BSC collected from the marine coast of the KSA in the mean of 5.8 mg/kg dry weight within the Arabian Gulf area, while in the Red Sea area it was around 5.95 mg/kg dry weight.

In Bahrain marine environment, Pb^{2+} concentration in seawater ranged from 1.13 to 2.01 $\mu\text{g/L}$ (Juma & Al-Madany, 2008), and in sediment from 3.69 to 277 mg/kg dry weight (Bersuder et al., 2020). In the marine organisms, Pb^{2+} concentrations in the grouper fish (*Epinephelus coioides*) were in the range of 4.3 to 15.2 mg/kg with an average of 10.8 (Al-Sayed et al., 1996). Furthermore, elevated level of Pb^{2+} were observed in pearl oyster; 3.9 and 6.4 mg/kg dry weight (de Mora et al., 2004), rock scallop; 37.7 mg/kg dry weight (Fowler et al., 1993), and marine algae; 21.76 – 45.96 mg/kg dry weight (Ali-Mohamed, 2002). On the other hand, very low levels of Pb^{2+} were observed in marine organism in Bahrain (Al-Madany et al., 1996; de Mora et al., 2004).

Cadmium trace metal ion accumulated mostly in the brown meat; hepatopancreas, which was around 15-fold its concentration in the white meat; Table 14. Overall Cd^{2+} concentration in white meat was in the range of <0.02 to 0.66 mg/kg with an average of 0.27 mg/kg, where Tubli Bay crabs found to have the minimum content of Cd^{2+} and males crabs in Ra's Al M'ttalah found to have the highest content. However, in the brown meat the average concentration of Cd^{2+} was 4.87 mg/kg with a range of 0.27 to 10.3 mg/kg. Female crabs in Tubli Bay found to have the lowest content of Cd^{2+} , while male crabs in Marwada found to have the highest Cd^{2+} .

Cadmium trace metal ions in Bahrain marine environment were at very low levels; in seawater, it ranged from 0.06 to 5.20 $\mu\text{g/L}$ (Juma & Al-Madany, 2008), and in the marine sediment it was ranged from <0.08 to 0.62 mg/kg (Bersuder et al., 2020). However, in the current study it ranges

from <0.02 to 10.3 mg/kg dry weight. This is due to the fact that decapods did not have a regulated mechanism that regulates the body concentration of Cd^{2+} at a certain level, which leads to net bioaccumulation of the Cd^{2+} from the surrounding environment (Jennings & Rainbow, 1979; White & Rainbow, 1982; Rainbow, 1985). Most of Cd^{2+} accumulate in the hepatopancreas (90%), then in the gills and other organs (Canli & Furness, 1993).

Crabs uptake cadmium trace metal ions from the surrounding environment through the calcium channel (Verbost et al., 1989; Wright, 1995; Bondgaard & Bjerregaard, 2005; Nørum et al., 2005), due to the similarity of the ionic radii between Cd^{2+} and Ca^{2+} ; 0.97 Å and 0.99Å, respectively (Bondgaard & Bjerregaard, 2005; Rainbow & Black, 2005). This accumulation increases especially in the postmoult stage, where the crab has to increase the Ca^{2+} uptake to substitute the large losses of Ca^{2+} in the old exoskeleton (Neufeld & Cameron, 1993; Nørum et al., 2005).

The level of Cd^{2+} reported in the current study is lower than the Cd^{2+} level in the same species studied in the Arabian Gulf and other regions; Table 28 and Table 29. Within white meat (muscles), BSC was found to have a low bioaccumulation factor in Malaysia; which was equal to 37 (Anandkumar et al., 2019). In Bahrain, Al-Madany et al. (1996) reported Cd^{2+} in edible tissues of BSC were at range of 0.001 to 0.20 mg/kg wet weight with a mean of 0.054 mg/kg wet weight. This is almost in the same level as the current study that reported in the white meat, which was at 0.054 with a range of <0.02 to 0.162 mg/kg wet weight. Within the Arabian Gulf area, higher level of Cd^{2+} than the current study was observed in the north east coast of the Arabian Gulf; 0.32 – 1.54 mg/kg dry weight (Fatemi & Khoramnejadian, 2016), 0.54 – 9.4 mg/kg wet weight (Rahmanpour et al., 2014). However, in marine coast of KSA, Cd^{2+} concentration was close to the current study which was reported at 0.26 and 0.25 mg/kg dry weight

for male and female BSC (Abdel-Salam & Hamdi, 2014). Outside the Arabian Gulf, in Mersin Bay (Turkey) Cd^{2+} concentration was ranged from 0.50 to 1.47 mg/kg dry weight (Ayas, 2013). Furthermore, in Malaysia Anandkumar et al. (2019) reported Cd^{2+} at 0.6 mg/kg dry weight in the muscles of walking legs. In India, Kumar et al. (2019) reported Cd^{2+} within muscle at the range of 0.01 to 2.74 mg/kg dry weight, and in the range of 0.01 to 30.23 mg/kg dry weight within the hepatopancreas tissues, which is higher than what have been observed in the current study. European Commission conducted a monitoring exercise in Europe in 2009-2010 for the Cd^{2+} level in crabs, the results showed that Cd^{2+} concentration was 0.08 mg/kg in white meat but it was 8 mg/kg in brown meat (European Commission, 2011).

In Bahrain, the Cd^{2+} levels in different marine organisms were higher than what have been found in the white meat of the current study; Table 30. Al-Sayed et al. (1996) reported Cd^{2+} in grouper fish in the range of 0.2 to 3.1 mg/kg wet weight. Moreover, Pearl oyster (*Pinctada radiata*) content of Cd^{2+} was in the range of 0.25 – 17.9 mg/kg dry weight (Fowler et al., 1993; Al-Sayed et al., 1994; de Mora et al., 2004), while rock scallop (*Spondylus sp.*) has higher level also; 2.0 mg/kg dry weight (Fowler et al., 1993). Ali-Mohamed (2002) reported Cd^{2+} in marine algae (*Hormophysa triquetra*) in the range of 1.17 – 3.30 mg/kg dry weight. On the other hand, de Mora et al. (2004) reported Cd^{2+} within *Eponephelus coioides* muscles in the range of <0.001 to 0.002 mg/kg dry weight, which is lower than the current study. Also in the liver of *Eponephelus coioides* accumulate Cd^{2+} less than brown meat of BSC, which ranged from 0.37 to 2.14 mg/kg dry weight (de Mora et al., 2004).

In general, the results of metal ion concentrations indicate a difference in the bioaccumulation of trace metal ions at white and brown meats of crabs. This might be attributed to the bioavailability of the trace metal ions

in the surrounding environment, biological activities in the crab's body and ecological factors like temperature and salinity.

Table 28 Comparison of trace metal ions content (mg/kg dry weight) in the BSC tissues from Bahrain with the other regions.

Area	Gender	Tissue	Zn ²⁺	Cu ²⁺	Ni ²⁺	Cr ³⁺	Pb ²⁺	Cd ²⁺	Reference
Bahrain	Male	White	229.94	67.99	0.148	0.135	<0.20	0.265	Current Study, Part 1
		Brown	202.09	213.88	1.152	0.130	<0.20	5.826	
	Female	White	242.41	70.99	0.170	0.083	<0.20	0.281	
		Brown	268.56	184.36	1.348	0.135	<0.20	3.917	
Kuwait	Mixed	Muscle	206.22 (188.69-208.28)	123.99 (110.15-142.8)	-	0.48 (0.15-0.66)	1.90 (1.72-2.08)	-	(Al-Mohanna & Subrahmanyam, 2001)
		Hepatopancreas	160.59 (155.04-165.63)	50.00 (46.23-52.45)	-	0.38 (0.13-0.52)	1.18 (0.71-1.40)	-	
Iran	-	Muscle	-	-	13.29 (7.54-20.8)	-	0.73 (0.10-2.40)	-	(Hosseini et al., 2012b)
		Hepatopancreas	-	-	13.75 (6.34-13.75)	-	0.77 (0.14-2.85)	-	
Iran	-	Whole Body	66.64	-	-	-	-	-	(Heidarieh et al., 2013)
Iran	Male	Soft tissue	-	-	-	-	-	0.96 (0.32-1.54)	(Fatemi & Khoramnejadian, 2016)
Iran	-	Muscle	48.34 (1.11-88.30)	0.71 (0.62-0.80)	0.62 (0.37-0.84)	-	0.021 (0.014-0.031)	-	(Koosej et al., 2016)
Turkey	Male	Carapace meat	102.51 (101.40-104.13)	29.98 (101.40-104.13)	-	0.55 (0.5-0.61)	0.21 (0.19-0.23)	0.88 (0.84-0.91)	(Ayas & Özogul, 2011)
	Female	Carapace meat	107 (106.13-108.64)	26.92 (25.39-28.20)	-	0.40 (0.30-0.52)	0.33 (0.22-0.43)	1.11 (1.05-1.17)	
Turkey	Male	Lump crab meat	45.66 (37.98-66.77)	22.2 (9.64-50.32)	-	0.41 (0.38-0.44)	0.34 (0.30-0.40)	0.77 (0.50-1.01)	(Ayas, 2013)
		Chela crab meat	117 (89.85-150.13)	44.51 (26.74-72.50)	-	0.36 (0.31-0.38)	0.38 (0.29-0.46)	0.99 (0.63-1.47)	
	Female	Lump crab meat	58.39 (37.80-78.16)	22.25 (11.74-33.85)	-	0.41 (0.37-0.45)	0.25 (0.10-0.30)	0.85 (0.71-1.01)	
		Chela crab meat	130 (93.99-178.55)	43.72 (25.13-70.62)	-	0.43 (0.32-0.53)	0.30 (0.12-0.52)	1.12 (1.02-1.23)	
India	-	Muscle	2.34	2.63	-	0.03	0.03	0.02	(Viswanathan et al., 2013)
		Hepatopancreas	2.82	0.89	-	0.04	0.03	0.02	

Continue

Area	Gender	Tissue	Zn ²⁺	Cu ²⁺	Ni ²⁺	Cr ³⁺	Pb ²⁺	Cd ²⁺	Reference
KSA - Arabian Gulf	Male	Muscle	45	6	-	-	6	0.26	(Abdel-Salam & Hamdi, 2014)
	Female	Muscle	43	5	-	-	5.6	0.25	
KSA - Red Sea	Male	Muscle	47	4	-	-	6.2	0.24	
	Female	Muscle	44	6	-	-	5.7	0.28	
Malaysia	-	Muscle - Carapace	394	33.5	2.4	5.1	4.1	0.6	(Anandkumar et al., 2019)
		Muscle - Walking leg	319	85.7	3.1	7.2	4	0.6	
India		Muscle	-	<0.01-156.84	<0.01-32.77	<0.01-81.69	<0.01-1.37	0.01-2.74	(Kumar et al., 2019)
		Hepatopancreas	-	<0.01-222.92	<0.01-14.86	<0.01	<0.01-4.32	0.01-30.23	

Table 29 Comparison of trace metal ions contents (mg/kg wet weight) in the BSC tissues from Bahrain with the other regions.

Area	Gender	Tissue	Zn ²⁺	Cu ²⁺	Ni ²⁺	Cr ³⁺	Pb ²⁺	Cd ²⁺	Reference
Bahrain	Male	White	46.68	13.80	0.030	0.027	<0.2	0.054	Current Study, Part 1
		Brown	37.59	39.78	0.214	0.024	<0.2	1.084	
	Female	White	48.00	14.06	0.034	0.016	<0.2	0.056	
		Brown	53.18	36.50	0.267	0.027	<0.2	0.776	
Bahrain	-	-	-	-	-	-	0.163 (0.008-0.820)	0.054 (0.001-0.200)	(Al-Madany et al., 1996)
Bahrain	Male	Composition meat	23	6.0	-	-	4.0	ND	(Musaiger & Al-Rumaidh, 2005)
	Female	Composition meat	26	6.0	-	-	0.4	ND	
Iran	-	Hepatopancreas	-	-	393.23 (214.0-674.28)	-	5.21 (1.78-8.94)	6.00 (0.54-9.40)	(Rahmanpour et al., 2014)
India	Male	Muscle	-	5.99 (3.86-8.54)	-	0.68 (0.2-1.47)	1.08 (0.28-1.75)	0.71 (0.41-1.31)	(Karar et al., 2019)
		Hepatopancreas	-	12.46 (7.21-17.03)	-	1.33 (0.66-2.39)	2.07 (0.53-4.72)	2.82 (0.95-5.86)	
Sri Lanka	Male	Carapace meat	21.82	6.44	-	-	0.25	0.16	(Jinadasa et al., 2018)
	Female	Carapace meat	22.25	9.49	-	-	0.2	0.14	

Table 30 Comparison of trace metal ions contents (mg/kg) in the BSC tissues and other marine organisms studied in Bahrain.

Species	Tissue	Zn ²⁺	Cu ²⁺	Ni ²⁺	Cr ³⁺	Pb ²⁺	Cd ²⁺	Reference
Blue swimming crab (<i>P. pelagicus</i>)	White	229.94	67.99	0.148	0.135	<0.20	0.265	Current Study, based on dry weight
	Brown	202.09	213.88	1.152	0.130	<0.20	5.826	
	White	242.41	70.99	0.170	0.083	<0.20	0.281	
	Brown	268.56	184.36	1.348	0.135	<0.20	3.917	
Grouper fish (<i>E. coioides</i>)	Tissues	499 (223-1253)	3.9 (0.6-9.3)	4.4 (0.7-9.0)	-	10.8 (4.3-15.2)	1.2 (0.2-3.1)	(Al-Sayed et al., 1996)
BSC (<i>P. pelagicus</i>)	Crab meat	24.5	6	-	-	2.2	-	(Musaiger & Al-Rumaidh, 2005)
Shrimp (<i>P. semisulcatus</i>)	Edible tissue	-	-	-	-	0.008 – 0.15	0.001 – 0.06	(Al-Madany et al., 1996)
BSC (<i>P. pelagicus</i>)	Edible tissue	-	-	-	-	0.008 – 0.84	0.001 – 0.20	
Shovel lobsters (<i>Thennus sp.</i>)	Edible tissue	-	-	-	-	0.01 – 0.97	0.025 – 0.10	
Pearl Oyster (<i>P. radiata</i>)		333.8 (159-895)	1.52 (0.4-3.4)	3.25 (0.2-8.95)	-	-	1.65 (0.25-3.8)	(Al-Sayed et al., 1994)
Orange spotted grouper (<i>E. coioides</i>)	Muscles	15.9 – 27.1	0.235 – 0.593	<0.01 – 0.05	0.013 – 0.075	0.005 – 0.028	<0.001 – 0.002	(de Mora et al., 2004)
	Livers	183 – 421	75.5 – 276	0.039 – 0.085	0.019 – 0.04	0.001 – 0.017	0.369 – 2.14	
Pearl Oyster (<i>P. radiata</i>)	Tissues	4290 & 1825	4.46 & 3.13	0.884 & 0.709	0.794 & 0.672	3.92 & 6.396	3.68 & 3.79	
Rock Scallop (<i>Spondylus sp.</i>)	Tissues	461	15.3	9.3	2.8	37.7	2.0	(Fowler et al., 1993)
Pearl Oyster (<i>P. radiata</i>)	Tissues	898 & 4607	1.8 – 6.9	0.44 & 1.24	0.34 & 0.43	0.32 – 3.9	3.3 – 17.9	
Marine algae (<i>H.triquetra</i>)		39.4 – 48.0	4.17 – 5.99	4.17 – 22.36	0.03 – 1.22	21.76 – 45.96	1.17 – 3.30	(Ali-Mohamed, 2002)

Based on wet weight

Based on dry weight

4.2.2 The Differences among the Sites

In the second part of the current study, the differences in accumulations of Zn^{2+} , Cu^{2+} and Cd^{2+} among gender, tissues and sites were tested in a wet weight of 10 individual crabs in each gender in every site.

Two-way ANOVA test showed differences in the crab's Zn^{2+} , Cu^{2+} and Cd^{2+} content among the sites in the brown meat. However, only Zn^{2+} showed significant differences in white meat of BSC, Table 22. Tukey ad-hoc were used to identify the differences in sites, Table 23. Marwada site was found to have higher than the other sites in the Zn^{2+} level among white meat. Whereas both Marwada and Tubli Bay were found to have higher than the other sites in the Zn^{2+} level among brown meat. Ra's Al M'ttallah found to have higher level of Cu^{2+} and Cd^{2+} than the other sites in Brown mean. This is in agreement with what was reported by Juma and Al-Madany (2008) that sites on the west and north-west coasts of Bahrain showed higher levels of Zn^{2+} , Cu^{2+} and Cd^{2+} than other sites around Bahrain, in seawater.

On the other hand, there are limited anthropogenic activities close to the Marwada and Ra's Al M'ttallah sites, which eliminate the possibility of point source pollution hypothesis in these sites. The remarkable thing for these sites is the elevated salinity level that may exceed 57 ppt in Ra's Al M'ttallah site (Zainal et al., 2007), because this site is affected directly by the high salinity waters coming from Salwa Bay. Salinity in the Marwada site (around 43.95 – 49.31 ppt) (<https://www.sce.gov.bh>) is lower than in Ra's Al M'ttallah, but the water is shallow providing the opportunity for high temperature.

The proper salinity range for BSC that increases both the growth rate (size and weight) and the survival rate was reported in Australia from 20 to 35 ppt. When the salinity rises to around 45 ppt, significant decreases in both growth rate and the survival rate were recorded. Furthermore,

increasing the salinity up to 45 ppt causes an increase in the metabolic rate and generating "hyper-osmoregulation" condition that increases the stress on crabs (Romano & Zeng, 2006). It can be suggested that, crabs that are in Marwada and Ra's Al M'ttallah sites are facing harsh conditions, which increase their food demands. Crabs have to feed more in order to meet their requirements for salinity tolerance. In that case, crabs are increasing their possibility of accumulating more trace metal ions through the food route.

The findings in the current study is not in agreement with what was concluded by Al-Madany et al. (1996), where no significant differences were indicated for the mean levels of Pb^{2+} , As^{2+} , Hg^{2+} and Cd^{2+} in the edible tissues of crustaceans collected from the north, south, east and west coast of Bahrain.

Contrary to what was expected, crabs in the Tubli Bay did not show significantly higher metal content than those in the other sites, except in the case of Zn^{2+} in brown meat. Tubli Bay was reported by Bersuder et al. (2020) as a hot spot in Bahrain that suffers from high levels of Zn^{2+} , Cu^{2+} and Cd^{2+} . Due to the organic matter discharged with incomplete treated sewage water as shown in Figure 16, Tubli Bay samples did not show a higher level of metals compared to those taken from other sites. Because the organic matter combines with the trace metals and reduces their bioavailability for marine organisms (Javed & Usmani, 2015). So the trace metal ions are present in high concentrations in the sediment of Tubli Bay as reported by Bersuder et al. (2020), but they are not in the available form that can be accumulated by crabs.



Figure 16 Sediment from the northern side of Tubli Bay.

Askar site was found to have no significant elevation of Zn^{2+} , Cu^{2+} and Cd^{2+} metal ions content compared to other sites. Askar site is located on the eastern coast of Bahrain, where most land-based activities are located. The same site is located around 5.8 km from the effluent of the Refinery facility. The results found in this current study corresponds with what was found by Bersuder et al. (2020), which indicated that the trace metal ions pollution in Bahrain is located and limited in close area to the discharge point of some of the heavy industrial facilities.

Some studies indicated the presence of some trace metal ions in the southern coast of Bahrain significantly higher than in the other coasts. For example, Al-Madany et al. (1996) reported that Cd^{2+} in the shellfish collected from the south coast is more than in others. In addition, Ali-

Mohamed (2002) reported Cd^{2+} and Ni^{2+} in marine algae (*Hormophysa triquetra*) at Al-Jazair Beach were higher than the other sites. Where Al-Jazair Beach is on the west coast of Bahrain and it is the closest site to the Ra's Al M'ttalah site in the current study. On the other hand, the southern site was found to have Cr^{3+} significantly lower than the other sites in *Hormophysa triquetra* (Ali-Mohamed, 2002).

4.2.3 The Differences among the Tissue Type

The differences of the trace metal ions accumulation based on the tissue types were investigated by Paired t-test; Table 21. Both Cu^{2+} and Cd^{2+} were significantly accumulated in brown meat more than in white meat. However, no significant differences were observed for Zn^{2+} in general. In the Tubli Bay site, Zn^{2+} was found to be accumulate in brown meat significantly more than in white meat.

Other studies showed similar findings. For instance, Kumar et al. (2019) reported that Cu^{2+} and Cd^{2+} more in hepatopancreas, but Ni^{2+} and Cr^{3+} were accumulated more in muscles. In India also, Karar et al. (2019) reported that Cu^{2+} , Cr^{3+} , Pb^{2+} and Cd^{2+} accumulations were more in hepatopancreas than in muscles. However, Al-Mohanna and Subrahmanyam (2001) reported Zn^{2+} and Cu^{2+} accumulated in muscles more than the hepatopancreas. On the other hand, Viswanathan et al. (2013) observed no differences between the concentration of Zn^{2+} , Cr^{3+} , Pb^{2+} and Cd^{2+} in the muscles and hepatopancreas, while Cu^{2+} was found to have accumulate more in muscles.

The accumulation of trace metal ions in the brown meat (digestive system; hepatopancreas) is expected, especially for those trace metal ions that are toxic or not essential in high levels like Cd^{2+} , Pb^{2+} , Cr^{3+} and Ni^{2+} . This is because the functional role of the hepatopancreas (which is a metabolically active organ) is detoxification, for such trace metal ions

(Çoğun et al., 2017). Hepatopancreas uses metallothioneins in order to detoxicate the trace metal ions (Truchet et al., 2020). Copper metal ion is an essential element for the crab (and other organisms) and some of Cu^{2+} present in the hepatopancreas because it is the site where the hemocyanin synthesis takes place (Rainer & Brouwer, 1993).

4.2.4 The Differences among the Gender

The differences in the accumulation of Zn^{2+} , Cu^{2+} and Cd^{2+} based on the crab gender were investigated by two-way ANOVA test, Table 22. Female crabs were found to accumulate Zn^{2+} and Cu^{2+} significantly more than male crabs in their white meat. Moreover, only Zn^{2+} was found to be accumulated significantly by females more than male crabs in their brown meat. No significant differences were observed in the accumulation of Cd^{2+} in both white and brown meats. In Bahrain, no attempt was made to investigate the accumulation of Zn^{2+} , Cu^{2+} and Cd^{2+} based on gender of BSC.

In Sri Lanka, Jinadasa et al. (2018) reported a significantly higher content of Cu^{2+} and Fe^{2+} in female BSC than male. However, no differences were observed in Zn^{2+} , Pb^{2+} , Hg^{2+} and Cd^{2+} . Furthermore, No differences were observed by Hosseini et al. (2014) between the accumulation of Hg^{2+} in tissues of male and female of the BSC in Iran. In Turkey, Ayas (2013) did not report a significance different between the accumulation of Cd^{2+} , Cr^{3+} , Pb^{2+} , Cu^{2+} , Zn^{2+} and Fe^{2+} in the tissues of BSC male and female. This is in agreement with result reported previously by Ayas and Ozogul (2011) that no significant differences were observed between male and female crabs of BSC in accumulation of Zn^{2+} , Cu^{2+} , Cr^{3+} , Pb^{2+} and Cd^{2+} , but a significant differences were indicated for Fe^{2+} , which was accumulated more in male crabs.

The reason that allows female crabs to accumulate trace metal ions more than males may be referred to the physiological needs of female crabs (Beltrame et al., 2011; Simonetti et al., 2013). Beltrame et al. (2010) suggested that during the reproduction stage, female increase their demand for food, which increase the chance of bioaccumulation of trace metals that enter the body through the food uptake route. Female crabs have to migrate to the shallow water to spawn, and then they have to move back to deep water to release the fertilized eggs for hatching (Kumar et al., 2003). The females' food demands increase during this stage because they have to carry the fertilized eggs for more than one month, which needs more energy (Chen et al., 2005). Al-Syaed (2009) reported that BSC has three spawning seasons in Bahrain, the main in spring (May). The rest is during autumn and winter, during autumn (November) and winter (December), respectively. This increases their potential for accumulation of more trace metal ions within their tissues more than male crabs.

4.2.5 The Suitability of the BSC Meats for human Consumption

The suitability of these crabs for human consumption was tested with the thresholds permitted in the GSO 1922/2009 and EU No 20/2011, which reported that the level of Pb^{2+} and Cd^{2+} should not exceed 0.5 mg/kg wet weight. It was based on levels of both Pb^{2+} and Cd^{2+} in the white meat of crabs only. The Pb^{2+} was below the quantification limit; <0.20 mg/kg dry weight. Overall Cd^{2+} concentrations in the white meat ranged from <0.02 to 0.16 mg/kg with an average of 0.06 mg/kg wet weight, whereas in brown meat Cd^{2+} ranged from 0.05 to 1.73 mg/kg with an average of 0.92 mg/kg wet weight. These results do not show any violations in the mentioned thresholds, which indicates that the white meat of the BSC that lives in Bahrain's marine environment can be consumed safely and does not pose a

threat to human health according to the Pb^{2+} and Cd^{2+} levels. This result is in close agreement with what is reported by Al-Madany et al. (1996).

On the other hand, high levels of Cd^{2+} were reported in the brown meat that exceeded the 0.5 mg/kg threshold. People who eat brown meat of the BSC will be exposed to elevated levels of Cd^{2+} , so it is recommended to avoid eating much of brown meat (European Commission, 2011).

4.3 Conclusion

This study aimed to determine the concentration of six trace metal ions in BSC meat collected from four sites in Bahrain's marine environment, to investigate how gender, and geographical location influence trace metal concentrations, and to explore the tendency of BSC to accumulate trace metals in its white and brown tissues.

This study revealed that BSC tends to accumulate high amounts of Zn^{2+} and Cu^{2+} compared to Cd^{2+} , Ni^{2+} , Cr^{3+} , and Pb^{2+} . Overall, concentration of trace metal ions in BSC meat was in the following order: $\text{Zn}^{2+} > \text{Cu}^{2+} > \text{Cd}^{2+} > \text{Ni}^{2+} > \text{Cr}^{3+}$. Zinc (II) ion concentrations reported in this study in both white and brown crabmeat as well as Cu^{2+} concentrations in brown crabmeat are the highest in the region.

It was found that females tend to accumulate more Zn^{2+} and Cu^{2+} in their white meat and more Cu^{2+} in their brown meat compared to males.

Trace metals concentrations in crabmeat was found to vary among different locations. Crabs collected from Ra's Al M'ttallah were found to have higher concentrations of Cu^{2+} and Cd^{2+} in brown crabmeat compared to other sites. Crabs collected from Marwada showed higher concentrations of Zn^{2+} in white crabmeat compared to other locations, whereas crabs

collected from Marwada and Tubli Bay sites were found to have a higher concentration of Zn^{2+} in brown crabmeat.

In terms of crabs tissues, this study revealed that BSC accumulates Cu^{2+} and Cd^{2+} in its brown meat.

Concentrations of Pb^{2+} and Cd^{2+} in BSC white meat were found to comply with thresholds recommended by GSC and European Commission standards for edible crab meat for Cd^{2+} and Pb^{2+} . This indicates that the consumption of BSC white meat in Bahrain does not pose any health risks in terms of Pb^{2+} and Cd^{2+} . On the other hand, BSC was found to accumulate high amounts of Cd^{2+} in its brown meat, which suggests that BSC should be avoided.

4.4 Recommendations and Future Work

In the end, the results of this study lead to recommend a set of recommendations related to levels of Zn^{2+} , Cu^{2+} , Cr^{3+} , Ni^{2+} , Pb^{2+} and Cd^{2+} in crabs' tissues, as follows:

- Increase the awareness of consumers to the elevated level of Cd^{2+} in the brown crab meat and advise them to change their dietary habits by refraining from eating brown meat. In order to reduce the exposure of the consumer to the higher levels of Cd^{2+} , they can be advised to exclude the crab's digestive system then washing the crabs thoroughly before cooking.
- Bahrain has to formulate a threshold for the essential trace metal ions (e.g. Zn^{2+} , Cu^{2+} , Ni^{2+} and Cr^{3+}) in crabs' tissues.

Moreover, a prospect for future work and research concerning the trace metal ions content in BSC to be suggested on:

- Conducting a continues program for monitoring the trace metals in BSC tissues collected from different habitat in Bahrain's sea area.
- Assess other trace metal ions in the tissues of BSC, especially the toxic metals like Hg^{2+} , $(\text{CH}_3\text{Hg})^+$, and metalloid ions such as As^{2+} .
- Investigate the organic pollutants in the tissues of BSC including the crab's gills and other organs.
- Identify the effect of temperature and salinity on the accumulation of trace metal ions among different tissues of BSC. Taking into account the wide range of both salinity and temperature in Bahrain's seawater.
- Evaluate the variation of trace metal ions level in the BSC tissues based on different sizes and different seasons.
- Study the trace metals contents in different marine organisms to investigate the elevation of Cu^{2+} and Zn^{2+} whither it is unique for crabs or may include other shellfish.
- Adopt monitoring locations associated with the potential pollution sources for the monitoring program implemented by SCE.

References

- Abdel-Salam, H. A., & Hamdi, S. A. H. (2014). Heavy metals monitoring using commercially important crustaceans and mollusks collected from Egyptian and Saudi Arabia coasts. *Animal and Veterinary Sciences*, 2(3), 49–61.
- Abdolvand, S., Esfahani, S. K., & Dmirchi, S. (2014). Mercury (Hg) and methyl mercury (MMHg) bioaccumulation in three fish species (sea food) from Persian Gulf. *Toxicology and Environmental Health Sciences*, 6(3), 192–198.
- Abou Seedo, K., Abido, M. S., Salih, A. A., & Abahussain, A. (2017). Assessing heavy metals accumulation in the leaves and sediments of urban mangroves (*Avicennia marina* (Forsk.) Vierh.) in Bahrain. *International Journal of Ecology*, 2017.
- Akhter, M. S., & Al-Jowder, O. (1997). Heavy metal concentrations in sediments from the coast of Bahrain. *International Journal of Environmental Health Research*, 7(1), 85–93.
- Al-Busaidi, M., Yesudhason, P., Al-Mughairi, S., Al-Rahbi, W. A. K., Al-Harthy, K. S., Al-Mazrooei, N. A., & Al-Habsi, S. H. (2011). Toxic metals in commercial marine fish in Oman with reference to national and international standards. *Chemosphere*, 85(1), 67–73.
- Al-Madany, I. M., Wahab, A. A. A., & Al-Alawi, Z. (1996). Trace metals concentrations in marine organisms from the coastal areas of Bahrain, Arabian Gulf. *Water, Air, and Soil Pollution*, 91(3–4), 233–248.
- Al-Mohanna, S. Y., & Subrahmanyam, M. N. V. (2001). Flux of heavy metal accumulation in various organs of the intertidal marine blue crab, *Portunus pelagicus* (L.) from the Kuwait coast after the Gulf War. *Environment International*, 27(4), 321–326.
- Al-Rashidi, T. B., Amos, C. L., & El-Gamily, H. I. (2007). Utilization of remotely sensed data to detect anthropogenic impacts on sea surface temperature of Kuwait Bay, Kuwait. *OCEANS 2007-Europe*, 94–98.
- Al-Rumaidh, M. J. (2002). *The Biology, Population Dynamics and Fishery Management of the Blue Swimming Crab, Portunuspelagicus (Linnaeus, 1758), in Bahraini Waters*. University of Wales, Bangor.
- Al-Saad, H. T., & Salman, N. A. (2012). Status of oil pollution in the Arabian Gulf and Shatt Al-Arab estuary: a review. In *Coastal Environments: Focus on Asian Regions* (pp. 36–46). Springer.

- Al-Sayed, H. (2009). Review of Spawning Studies of Some Commercially Important Marine Organisms in the Coastal Waters of Bahrain. *Journal of the Association of Arab Universities for Basic & Applied Sciences*, 8, 75–92.
- Al-Sayed, H., & Dairi, M. (2006). Metal accumulation in the edible marine snail *Turbo coronatus* (Gmelin) from different locations in Bahrain. *Arab Gulf Journal of Scientific Research*, 24(2), 48–57.
- Al-Sayed, H., Mahasneh, A. M., & Al-Saad, J. (1994). Variations of trace metal concentrations in seawater and pearl oyster *Pinctada radiata* from Bahrain (Arabian Gulf). *Marine Pollution Bulletin*, 28(6), 370–374.
- Al-Sayed, H. A., Al-Saad, J., Madany, I. M., & Al-Hooti, D. (1996). Heavy metals in the grouper fish *Epinephelus coioides* from the coast of Bahrain: an assessment of monthly and spatial trends. *International Journal of Environmental Studies*, 50(3–4), 237–246.
- Al Naggar, Y., Khalil, M. S., & Ghorab, M. A. (2018). Environmental pollution by heavy metals in the aquatic ecosystems of Egypt. *Open Acc J of Toxicol*, 3(1), 555603.
- Alam, I. A. H. (1993). The 1991 Gulf War Oil Spill—Lessons from the past and a warning for the future. *Marine Pollution Bulletin*, 27, 357–360.
- Alam, I., Barua, S., Ishii, K., Mizutani, S., Hossain, M. M., Rahman, I. M. M., & Hasegawa, H. (2019). Assessment of health risks associated with potentially toxic element contamination of soil by end-of-life ship dismantling in Bangladesh. *Environmental Science and Pollution Research*, 26(23), 24162–24175.
- Albelali, A. S., Alsaqabi, S. M., & Eshky, A. A. (2010). Sexual maturation and fertility in blue swimming crab *Portunus pelagicus* in the Arabian Gulf, (Saudi Arabia). *Alexandria Journal of Agricultural Research*, 55(3), 1–10.
- Ali-Mohamed, A. Y. (2002). Trace metal concentrations in Marine Algae *Hormophysa triquetra*, Bahrain coastline (Arabian Gulf). *Pollution Research*, 21(4), 397–402.
- Ali-Mohamed, A. Y., & Matter, H. A. (1996). Determination of inorganic particulates: (Cationic, anionic and heavy metals) in the atmosphere of some areas in Bahrain during the Gulf crisis in 1991. *Atmospheric Environment*, 30(20), 3497–3503.

- Ali, T. S., & Abahussain, A. (2013). Status of commercial fisheries in the Kingdom of Bahrain. *Basrah Journal of Agricultural Sciences*, 1, 220–238.
- Almahasheer, H. (2019). High levels of heavy metals in Western Arabian Gulf mangrove soils. *Molecular Biology Reports*, 46(2), 1585–1592.
- Álvarez, E. A., Mochón, M. C., Sánchez, J. C. J., & Rodríguez, M. T. (2002). Heavy metal extractable forms in sludge from wastewater treatment plants. *Chemosphere*, 47(7), 765–775.
- Anandkumar, A., Nagarajan, R., Prabakaran, K., Bing, C. H., Rajaram, R., Li, J., & Du, D. (2019). Bioaccumulation of trace metals in the coastal Borneo (Malaysia) and health risk assessment. *Marine Pollution Bulletin*, 145, 56–66.
- Anderlini, V. C., Mohammad, O. S., Zarba, M. A., Fowler, S. W., & Miramand, P. (1982). Trace metals in marine sediments of Kuwait. *Bulletin of Environmental Contamination and Toxicology*, 28(1), 75–80.
- Ariano, A., Lo Voi, A., D'Ambola, M., Marrone, R., Cacace, D., & Severino, L. (2015). Levels of Cadmium in White and Brown Meat of Warty Crab (*Eriphia verrucosa*). *Journal of Food Protection*, 78(12), 2253–2256.
- ATSDR. (2020). *Toxicological profile for lead*. ATSDR.
- Ayas, D. (2013). Effects of gender and season on potentially toxic metal levels in muscles of adult blue swimmer crabs (*Portunus pelagicus*) from the northeastern Mediterranean Sea. *J Mar Biol Oceanogr*, 2, 2:2. <http://dx.doi.org/10.4172/2324-8661.1000110>
- Ayas, D., & Özogul, Y. (2011). The chemical composition of sexually mature blue swimmer crab (*Portunus pelagicus*, Linnaeus 1758) in the Mersin Bay. *Journal of Fisheries Sciences. Com*, 5(4), 308–316.
- Barth, H.-J., & Khan, N. Y. (2008). Biogeophysical setting of the Gulf. In *Protecting the Gulf's marine ecosystems from pollution* (pp. 1–21). Springer. https://link.springer.com/chapter/10.1007/978-3-7643-7947-6_1
- Bausell, R. B., & Li, Y.-F. (2002). *Power analysis for experimental research: a practical guide for the biological, medical and social sciences*. Cambridge University Press.
- Beltrame, M. O., De Marco, S. G., & Marcovecchio, J. E. (2010). Influences of sex, habitat, and seasonality on heavy-metal

- concentrations in the burrowing crab (*Neohelice granulata*) from a coastal lagoon in Argentina. *Archives of Environmental Contamination and Toxicology*, 58(3), 746–756.
- Beltrame, M. O., De Marco, S. G., & Marcovecchio, J. E. (2011). The burrowing crab *Neohelice granulata* as potential bioindicator of heavy metals in estuarine systems of the Atlantic coast of Argentina. *Environmental Monitoring and Assessment*, 172(1–4), 379–389.
- Bencko, V. (1983). Nickel: a review of its occupational and environmental toxicology. *Journal of Hygiene, Epidemiology, Microbiology, and Immunology*, 27(2), 237–247.
- Bernhoft, R. A. (2013). Cadmium Toxicity and Treatment. *The Scientific World Journal*, 2013, 1–7.
- Bersuder, P., Smith, A. J., Hynes, C., Warford, L., Barber, J. L., Losada, S., Limpenny, C., Khamis, A. S., Abdulla, K. H., Le Quesne, W. J. F., & Lyons, B. P. (2020). Baseline survey of marine sediments collected from the Kingdom of Bahrain: PAHs, PCBs, organochlorine pesticides, perfluoroalkyl substances, dioxins, brominated flame retardants and metal contamination. *Marine Pollution Bulletin*, 161, 111734.
- Bhat, S. A., Hassan, T., & Majid, S. (2019). Heavy metal toxicity and their harmful effects on living organisms—a review. *International Journal of Medical Science And Diagnosis Research*, 3(1), 106–122.
- Boer, J. L., Mulrooney, S. B., & Hausinger, R. P. (2014). Nickel-dependent metalloenzymes. *Archives of Biochemistry and Biophysics*, 544, 142–152.
- Bolam, T., & Bersuder, P. (2013). A survey of cadmium in brown crabmeat and brown crabmeat products (FS102010). *Cefas Project Report C*, 5700, 55.
- Bondgaard, M., & Bjerregaard, P. (2005). Association between cadmium and calcium uptake and distribution during the moult cycle of female shore crabs, *Carcinus maenas*: an in vivo study. *Aquatic Toxicology*, 72(1–2), 17–28.
- Bosco, M. L., Varrica, D., & Dongarrà, G. (2005). Case study: Inorganic pollutants associated with particulate matter from an area near a petrochemical plant. *Environmental Research*, 99(1), 18–30.
- Boughriet, A., Proix, N., Billon, G., Recourt, P., & Ouddane, B. (2007). Environmental impacts of heavy metal discharges from a smelter in

- Deûle-canal sediments (Northern France): concentration levels and chemical fractionation. *Water, Air, and Soil Pollution*, 180(1–4), 83–95.
- Bradford, G. R., Page, A. L., Lund, L. J., & Olmstead, W. (1975). Trace Element Concentrations of Sewage Treatment Plant Effluents and Sludges; Their Interactions with Soils and Uptake by Plants. *Journal of Environmental Quality*, 4(1), 123–127.
- Brown, D. J. S., & James, J. D. (1985). Major oil spill response coordination in the combat of spills in Bahrain waters. *Journal of Petroleum Technology*, 37(01), 131–136.
- Burger, J. (2006). Bioindicators: a review of their use in the environmental literature 1970–2005. *Environmental Bioindicators*, 1(2), 136–144.
- Caballero-Gallardo, K., Alcala-Orozco, M., Barraza-Quiroz, D., De la Rosa, J., & Olivero-Verbel, J. (2020). Environmental risks associated with trace elements in sediments from Cartagena Bay, an industrialized site at the Caribbean. *Chemosphere*, 242, 125173.
- Canli, M., & Furness, R. W. (1993). Toxicity of heavy metals dissolved in sea water and influences of sex and size on metal accumulation and tissue distribution in the norway lobster *Nephrops norvegicus*. *Marine Environmental Research*, 36(4), 217–236.
- Cantillo, A. . (1998). Comparison of results of Mussel Watch Programs of the United States and France with Worldwide Mussel Watch Studies. *Marine Pollution Bulletin*, 36(9), 712–717.
- Cao, L., Lin, C., Gao, Y., Sun, C., Xu, L., Zheng, L., & Zhang, Z. (2020). Health risk assessment of trace elements exposure through the soil-plant (maize)-human contamination pathway near a petrochemical industry complex, Northeast China. *Environmental Pollution*, 263, 114414.
- Castagnetto, J. M., Hennessy, S. W., Roberts, V. A., Getzoff, E. D., Tainer, J. A., & Pique, M. E. (2002). MDB: the metalloprotein database and browser at the Scripps Research Institute. *Nucleic Acids Research*, 30(1), 379–382.
- Castro, P., & Huber, M. (2002). *Marine Biology* (4th ed.). McGraw-Hill.
- Cempel, M., & Nikel, G. (2006). Nickel: A review of its sources and environmental toxicology. *Polish Journal of Environmental Studies*, 15(3), 375–382.
- Chan, H. M., Bjerregaard, P., Rainbow, P. S., & Depledge, M. H. (1992).

- Uptake of zinc and cadmium by two populations of shore crabs *Carcinus maenas* at different salinities. *Marine Ecology Progress Series*, 86, 91–97.
- Chattopadhyaya, M. C., Soares, M. G., Campos, M. T. de J. S., Ismadji, S., Lofrano, G., Barakat, M. A., Ikehata, K., Yin, X.-B., Chowdhury, P., & Sharma, S. K. (2014). *Heavy metals in water: presence, removal and safety*. Royal Society of Chemistry.
- Chen, M. H., Chen, C. Y., Chou, H. Y., & Wen, T. C. (2005). Gender and size effects of metal bioaccumulation on the rock crab, *Thalamita crenata*, in Dapeng Bay, southwestern Taiwan. *Marine Pollution Bulletin*, 50(4), 463–469.
- Chester, R., & Stoner, J. H. (1975). Trace elements in total particulate material from surface sea water. *Nature*, 255(5503), 50–51.
- Chitrakar, P., Baawain, M. S., Sana, A., & Al-Mamun, A. (2019). Current Status of Marine Pollution and Mitigation Strategies in Arid Region: A Detailed Review. *Ocean Science Journal*, 54(3), 317–348.
- Choi, J.-S., & Jeon, S. kyung. (2018). A Geo-statistical Assessment of Heavy Metal Pollution in the Soil Around a Ship Building Yard in Busan, Korea. *Journal of the Korean Society of Marine Environment & Safety*, 24(7), 907–915.
<https://doi.org/10.7837/kosomes.2018.24.7.907>
- Çoğun, H. Y., Firat, Ö., Aytekin, T., Firidin, G., Firat, Ö., Varkal, H., Temiz, Ö., & Kargin, F. (2017). Heavy Metals in the Blue Crab (*Callinectes sapidus*) in Mersin Bay, Turkey. *Bulletin of Environmental Contamination and Toxicology*, 98(6), 824–829.
- Coles, S. L. (2003). Coral species diversity and environmental factors in the Arabian Gulf and the Gulf of Oman: a comparison to the Indo-Pacific region. *Atoll Research Bulletin*.
- Cooley, H., Gleick, P. H., & Wolff, G. (2006). *Desalination, with a grain of salt: A California Perspective*.
- Croquer, A., Bone, D., Bastidas, C., Ramos, R., & García, E. (2016). Monitoring coastal pollution associated with the largest oil refinery complex of Venezuela. *PeerJ*, 4, e2171.
- Dadar, M., Peyghan, R., & Memari, H. R. (2014). Evaluation of the bioaccumulation of heavy metals in white shrimp (*Litopenaeus vannamei*) along the Persian Gulf coast. *Bulletin of Environmental Contamination and Toxicology*, 93(3), 339–343.

- de Mora, S., Fowler, S. W., Cassi, R., & Tolosa, I. (2003). Assessment of organotin contamination in marine sediments and biota from the Gulf and adjacent region. *Marine Pollution Bulletin*, 46(4), 401–409.
- de Mora, S., Fowler, S. W., Wyse, E., & Azemard, S. (2004). Distribution of heavy metals in marine bivalves, fish and coastal sediments in the Gulf and Gulf of Oman. *Marine Pollution Bulletin*, 49(5–6), 410–424.
- de Mora, S., Tolosa, I., Fowler, S. W., Villeneuve, J.-P., Cassi, R., & Cattini, C. (2010). Distribution of petroleum hydrocarbons and organochlorinated contaminants in marine biota and coastal sediments from the ROPME Sea Area during 2005. *Marine Pollution Bulletin*, 60(12), 2323–2349.
- DeForest, D. K., Brix, K. V., & Adams, W. J. (2007). Assessing metal bioaccumulation in aquatic environments: the inverse relationship between bioaccumulation factors, trophic transfer factors and exposure concentration. *Aquatic Toxicology*, 84(2), 236–246.
- Dissanayake, C. B., & Chandrajith, R. (2009). Phosphate mineral fertilizers, trace metals and human health. *Journal of the National Science Foundation of Sri Lanka*, 37(3), 153–165.
- Duffus, J. H. (2002). “Heavy metals” a meaningless term?(IUPAC Technical Report). *Pure and Applied Chemistry*, 74(5), 793–807.
- Eisler, R. (1988). *Lead hazards to fish, wildlife, and invertebrates: a synoptic review* (Issue 14). US Fish and Wildlife Service, Patuxent Wildlife Research Center.
- El-Moselhy, K. M. (2006). Bioaccumulation of mercury in some marine organisms from Lake Timsah and Bitter Lakes (Suez Canal, Egypt). *Egyptian Journal of Aquatic Research*, 32(1), 124–134.
- Elahi, M., Esmaili-Sari, A., & Bahramifar, N. (2012). Total mercury levels in selected tissues of some marine crustaceans from Persian Gulf, Iran: variations related to length, weight and sex. *Bulletin of Environmental Contamination and Toxicology*, 88(1), 60–64.
- Emery, K. O. (1956). Sediments and water of Persian Gulf. *AAPG Bulletin*, 40(10), 2354–2383. <https://doi.org/10.1306/5CEAE595-16BB-11D7-8645000102C1865D>
- Engel, D. W., & Brouwer, M. (1987). Metal regulation and molting in the blue crab, *Callinectes sapidus*: metallothionein function in metal metabolism. *The Biological Bulletin*, 173(1), 239–251.
- Engel, D. W., & Fowler, B. A. (1979). Factors influencing cadmium

- accumulation and its toxicity to marine organisms. *Environmental Health Perspectives*, 28, 81–88.
- Ermiler, U., Grabarse, W., Shima, S., Goubeaud, M., & Thauer, R. K. (1998). Active sites of transition-metal enzymes with a focus on nickel. *Current Opinion in Structural Biology*, 8(6), 749–758.
- European Commission. (2011). *Information Note: Consumption of Brown Crab Meat*.
- Fatemi, F., & Khoramnejadian, S. (2016). Investigation of Cadmium and Arsenic Accumulation in *Portunus pelagicus* along the Asalouyeh Coast, Iran. *Journal of Earth, Environment and Health Sciences*, 2(1), 34–38.
- Förstner, U., & Wittmann, G. (1981). *Metal pollution in the aquatic environment*. Springer, Berlin, Heidelberg. <https://doi-org.libweb.uob.edu.bh/10.1007/978-3-642-69385-4>
- Fowler, S. W. (1993). Pollution in the Gulf: Monitoring the marine environment. *IAEA Bulletin*, 35(2), 9–13.
- Fowler, S. W., Huynh-Ngoc, L., & Fukai, R. (1984). Dissolved and particulate trace metals in coastal waters of the Gulf and Western Arabian Sea. *Deep Sea Research Part A. Oceanographic Research Papers*, 31(6–8), 719–729.
- Fowler, S. W., Readman, J. W., Oregioni, B., Villeneuve, J.-P., & McKay, K. (1993). Petroleum hydrocarbons and trace metals in nearshore Gulf sediments and biota before and after the 1991 war: an assessment of temporal and spatial trends. *Marine Pollution Bulletin*, 27, 171–182.
- Freije, A. (2015). Heavy metal, trace element and petroleum hydrocarbon pollution in the Arabian Gulf. *Journal of the Association of Arab Universities for Basic and Applied Sciences*, 17(1), 90–100.
- Freije, A., & Awadh, M. (2009). Total and methylmercury intake associated with fish consumption in Bahrain. *Water and Environment Journal*, 23(2), 155–164. <https://doi.org/10.1111/j.1747-6593.2008.00129.x>
- Genchi, G., Sinicropi, M. S., Lauria, G., Carocci, A., & Catalano, A. (2020). The Effects of Cadmium Toxicity. *International Journal of Environmental Research and Public Health*, 17(11), 3782.
- Gerhardt, A. (1993). Review of impact of heavy metals on stream invertebrates with special emphasis on acid conditions. *Water, Air, and Soil Pollution*, 66(3–4), 289–314.

- Ghaeni, M., Pour, N. A., & Hosseini, M. (2015). Bioaccumulation of polychlorinated biphenyl (PCB), polycyclic aromatic hydrocarbon (PAH), mercury, methyl mercury, and arsenic in blue crab *Portunus segnis* from Persian Gulf. *Environmental Monitoring and Assessment*, 187(5), 253. <https://doi.org/10.1007/s10661-015-4459-9>
- Ghani, A. (2011). Effect of chromium toxicity on growth, chlorophyll and some mineral nutrients of brassica juncea L. *Egyptian Academic Journal of Biological Sciences, H. Botany*, 2(1), 9–15.
- Giraldes, B. W., Al-Maslamani, I., Al-Ashwel, A., Chatting, M., & Smyth, D. (2016). Basic assessment of *Portunus segnis* (Forskål, 1775) – A baseline for stock management in the Western Arabian Gulf. *The Egyptian Journal of Aquatic Research*, 42(1), 111–119.
- Gobas, F. A. P. C., de Wolf, W., Burkhard, L. P., Verbruggen, E., & Plotzke, K. (2009). Revisiting Bioaccumulation Criteria for POPs and PBT Assessments. *Integrated Environmental Assessment and Management*, 5(4), 624.
- Golovanova, I. L. (2008). Effects of heavy metals on the physiological and biochemical status of fishes and aquatic invertebrates. *Inland Water Biology*, 1(1), 93.
- Goyer, R. A., & Clarkson, T. W. (1996). Toxic effects of metals. In C. D. Klaassen (Ed.), *Casarett and Doull's toxicology: the basic science of poisons* (5th ed., Vol. 5, pp. 813–867). McGraw-Hill New York.
- Halpern, B. S., Walbridge, S., Selkoe, K. A., Kappel, C. V., Micheli, F., D'Agrosa, C., Bruno, J. F., Casey, K. S., Ebert, C., Fox, H. E., Fujita, R., Heinemann, D., Lenihan, H. S., Madin, E. M. P., Perry, M. T., Selig, E. R., Spalding, M., Steneck, R., & Watson, R. (2008). A Global Map of Human Impact on Marine Ecosystems. *Science*, 319(5865), 948–952.
- Hambach, R., Lison, D., D'Haese, P. C., Weyler, J., De Graef, E., De Schryver, A., Lamberts, L. V., & van Sprundel, M. (2013). Co-exposure to lead increases the renal response to low levels of cadmium in metallurgy workers. *Toxicology Letters*, 222(2), 233–238.
- Hambidge, M. (2000). Human zinc deficiency. *The Journal of Nutrition*, 130(5), 1344S–1349S.
- Han, Y.-G., Kwon, O., & Cho, Y. (2015). A study of bioindicator selection for long-term ecological monitoring. *Journal of Ecology and Environment*, 38(1), 119–122.

- Hardy, D., Dutil, J.-D., Godbout, G., & Munro, J. (2000). Survival and condition of hard shell male adult snow crabs (*Chionoecetes opilio*) during fasting at different temperatures. *Aquaculture*, 189(3–4), 259–275.
- Hedge, L. H., Knott, N. A., & Johnston, E. L. (2009). Dredging related metal bioaccumulation in oysters. *Marine Pollution Bulletin*, 58(6), 832–840.
- Heidarieh, M., Maragheh, M. G., Shamami, M. A., Behgar, M., Ziaei, F., & Akbari, Z. (2013). Evaluate of heavy metal concentration in shrimp (*Penaeus semisulcatus*) and crab (*Portunus pelagicus*) with INAA method. *SpringerPlus*, 2(1), 72.
- Herrera-Herrera, C., Fuentes-Gandara, F., Zambrano-Arévalo, A., Higueta, F. B., Hernández, J. P., & Marrugo-Negrete, J. (2019). Health risks associated with heavy metals in imported fish in a coastal city in Colombia. *Biological Trace Element Research*, 190(2), 526–534.
- Hilmy, A. M., & Ghazaly, K. S. (1988). Variation of zinc and copper levels with sex and size in different tissues of the crab *Portunus pelagicus* (Linnaeus). *Folia Morphologica*, 36(2), 160–166.
- Hirose, K. (2006). Chemical Speciation of Trace Metals in Seawater: a Review. *Analytical Sciences*, 22(8), 1055–1063.
- Hodson, M. E. (2004). Heavy metals—geochemical bogey men? *Environmental Pollution*, 129(3), 341–343.
- Hoepner, T., & Lattemann, S. (2003). Chemical impacts from seawater desalination plants — a case study of the northern Red Sea. *Desalination*, 152(1–3), 133–140.
- Holt, E., & Miller, S. (2011). Bioindicators: Using Organisms to Measure Environmental Impacts. *Nature Education Knowledge*, 2(2), 8.
- Hosseini, M., Abdi Bastami, A., Karami, O., & Parsa, Y. (2012). Seasonal and sexuality variation of mercury concentrations in selected tissues of blue swimming crab, *Portunus pelagicus* (Linnaeus, 1758) and sediments from South Iran Persian Gulf. *American-Eurasian Journal of Agricultural and Environmental Sciences*, 12, 827–834.
- Hosseini, M., Amiri, A. M. D., Sahafi, H. H., Baniamam, M., & Lakzaei, F. (2015). Distribution of heavy metals in (Fe, Hg, Ni and Pb) sediment and blue crab, *Portunus pelagicus* from four estuaries, Persian Gulf. *Indian Journal of Geo-Marine Sciences*, 44(7), 1035–1042.
- Hosseini, M., Bastami, A. A., Khoei, J. K., Esmailian, M., Songhori, E. J.,

- & Najafzadeh, M. (2012). Concentrations of heavy metals in selected tissues of blue swimming crab, *Portunus pelagicus* (Linnaeus, 1758) and sediments from Persian Gulf. *World Applied Sciences Journal*, 19(10), 1398–1405.
- Hosseini, M., Nabavi, S. M. B., Bastami, A. A., & Parsa, Y. (2012). Mercury concentration in tissues of blue swimming crab, *Portunus pelagicus* (Linnaeus, 1758) and sediments from Persian Gulf coasts, Iran. *World Applied Sciences Journal*, 18(3), 322–327.
- Hosseini, M., Nabavi, S. M. B., Monikh, F. A., & Peery, S. (2014). Blue swimming crab, *Portunus pelagicus* (Linnaeus, 1758) as monitors of mercury contamination from Persian Gulf, South Iran. *Indian Journal of Geo-Marine Sciences*, 43(3), 377–383.
- Hosseini, M., & Parsa, Y. (2013). The Levels of Toxic Metals in Blue Crab *Portunus segnis* from Persian Gulf. *Journal of Marine Science: Research & Development*, 04(01), 1–5.
- Hunter, J. R. (1986). The physical oceanography of the Arabian Gulf: a review and theoretical interpretation of previous observations. *The First Arabian Gulf Conference on Environment and Pollution*. Kuwait University, Faculty of Science, Kuwait, 1–23.
- Ibrahim, G. A., Atallah, A., Anbtawi, N. M. G., & Ben Ahmed, A. (2020). Interaction of physicochemical parameters and the blue crab *Portunus pelagicus* (Linnaeus, 1758) in the Arabian Gulf. *International Journal of Oceans and Oceanography*, 14(1), 33–76.
- Irfan, M., Hayat, S., Ahmad, A., & Alyemeni, M. N. (2013). Soil cadmium enrichment: Allocation and plant physiological manifestations. *Saudi Journal of Biological Sciences*, 20(1), 1–10.
- Jaishankar, M., Tseten, T., Anbalagan, N., Mathew, B. B., & Beeregowda, K. N. (2014). Toxicity, mechanism and health effects of some heavy metals. *Interdisciplinary Toxicology*, 7(2), 60–72.
- Jakimska, A., Konieczka, P., Skóra, K., & Namieśnik, J. (2011). Bioaccumulation of Metals in Tissues of Marine Animals, Part I: the Role and Impact of Heavy Metals on Organisms. *Polish Journal of Environmental Studies*, 20(5), 1117–1125.
- Jan, A. T., Azam, M., Siddiqui, K., Ali, A., Choi, I., & Haq, Q. M. (2015). Heavy metals and human health: mechanistic insight into toxicity and counter defense system of antioxidants. *International Journal of Molecular Sciences*, 16(12), 29592–29630.

- Javed, M., & Usmani, N. (2015). Impact of heavy metal toxicity on hematology and glycogen status of fish: a review. *Proceedings of the National Academy of Sciences, India Section B: Biological Sciences*, 85(4), 889–900.
- Jennings, J. R., & Rainbow, P. S. (1979). Studies on the uptake of cadmium by the crab *Carcinus maenas* in the laboratory. I. Accumulation from seawater and a food source. *Marine Biology*, 50(2), 131–139.
- Jinadasa, B., Jayasinghe, G., Nethmina, N. A. N., & Abeysinghe, S. S. (2018). Bio-accumulation of trace metals in Blue Swimmer Crab (*Portunus pelagicus*) in Sri Lanka. *Journal of the National Aquatic Resources Research and Development Agency of Sri Lanka*, 45–47, 17–25.
- Johns, W. E., Yao, F., Olson, D. B., Josey, S. A., Grist, J. P., & Smeed, D. A. (2003). Observations of seasonal exchange through the Straits of Hormuz and the inferred heat and freshwater budgets of the Persian Gulf. *Journal of Geophysical Research: Oceans*, 108(C12).
- Juma, H. A., & Al-Madany, I. (2008). Concentration of heavy metals in the territorial sea water of the Kingdom of Bahrain, Arabian Gulf. *Arab Gulf Journal of Scientific Research* (1989), 26(1–2), 19–32.
- Kabir, E., Ray, S., Kim, K.-H., Yoon, H.-O., Jeon, E.-C., Kim, Y. S., Cho, Y.-S., Yun, S.-T., & Brown, R. J. C. (2012). Current Status of Trace Metal Pollution in Soils Affected by Industrial Activities. *The Scientific World Journal*, 2012, 1–18.
- Kämpf, J., & Sadrinasab, M. (2006). The circulation of the Persian Gulf: a numerical study. *Ocean Science, European Geosciences Union*, 2(1), 27–41. <https://doi.org/10.5194/os-2-27-2006>
- Karar, S., Hazra, S., & Das, S. (2019). Assessment of the heavy metal accumulation in the Blue Swimmer Crab (*Portunus pelagicus*), northern Bay of Bengal: Role of salinity. *Marine Pollution Bulletin*, 143, 101–108. <https://doi.org/10.1016/j.marpolbul.2019.04.033>
- Keshmiri, S., Pordel, S., Raeesi, A., Nabipour, I., Darabi, H., Jamali, S., Dobaradaran, S., Heidari, G., Ostovar, A., Ramavandi, B., Tahmasebi, R., Marzban, M., Khajeian, A., Sanati, A., & Farrokhi, S. (2018). Environmental Pollution Caused by Gas and Petrochemical Industries and Its Effects on the Health of Residents of Assaluyeh Region, Iranian Energy Capital: A Review Study. *Iranian South Medical Journal*, 21(2), 162–185.
- Khoramnejadian, S., & Fatemi, F. (2015). Bioaccumulation of arsenic in

- blue swimmer crab. *International Proceedings of Chemical, Biological and Environmental Engineering*, 88, 59–64.
- Koosej, N., Jafariyan, H., Rahmani, A., Patimar, A., & Gholipoor, H. (2016). The Study of Some Heavy Metals (Lead, Nickel, Zinc, Copper and Iron) in Muscle Tissue of Blue Swimming Crab *Portunus pelagicus* in Some Areas of the Hormozgan Province, Iran. *American Journal of Life Science Researches*, 4(4), 140–144.
- Krejpcio, Z. (2001). Essentiality of chromium for human nutrition and health. *Polish Journal of Environmental Studies*, 10(6), 399–404.
- Kumar, M. S., Xiao, Y., Venema, S., & Hooper, G. (2003). Reproductive cycle of the blue swimmer crab, *Portunus pelagicus*, off southern Australia. *Journal of the Marine Biological Association of the United Kingdom*, 83(5), 983–994.
- Kumar, S. B., Padhi, R. K., & Satpathy, K. K. (2019). Trace metal distribution in crab organs and human health risk assessment on consumption of crabs collected from coastal water of South East coast of India. *Marine Pollution Bulletin*, 141, 273–282.
- Lattemann, S., & Höpner, T. (2008). Environmental impact and impact assessment of seawater desalination. *Desalination*, 220(1–3), 1–15.
- Le Quesne, W. J. F., Fernand, L., Ali, T. S., Andres, O., Antonpoulou, M., Burt, J. A., Dougherty, W. W., Edson, P. J., El Kharraz, J., Glavan, J., Mamiit, R. J., Reid, K. D., Sajwani, A., & Sheahan, D. (2021). Is the development of desalination compatible with sustainable development of the Arabian Gulf? *Marine Pollution Bulletin*, 173, 112940.
- Lee, J.-S., & Lee, B.-G. (2005). Effects of salinity, temperature and food type on the uptake and elimination rates of Cd, Cr, and Zn in the asiatic Clam *Corbicula fluminea*. *Ocean Science Journal*, 40(2), 79–89.
- Lewicki, S., Zdanowski, R., Krzyzowska, M., Lewicka, A., Debski, B., Niemcewicz, M., & Goniewicz, M. (2014). The role of Chromium III in the organism and its possible use in diabetes and obesity treatment. *Annals of Agricultural and Environmental Medicine*, 21(2), 331–335.
- Lexico. (2020). *Algorithm | Definition of Algorithm by Oxford Dictionary on Lexico.com also meaning of Algorithm*. Lexico.Com. <https://www.lexico.com/definition/bioindicator>
- Lichtfouse, E., Schwarzbauer, J., & Robert, D. (2011). *Environmental Chemistry for a Sustainable World: Volume 1: Nanotechnology and Health Risk* (Vol. 1). Springer Science & Business Media.

- Literathy, P. (1993). Considerations for the assesement of environmental consequences of the 1991 Gulf War. *Marine Pollution Bulletin*, 27, 349–356.
- Literathy, P., Khan, N. Y., & Linden, O. (2002). Oil and petroleum industry. In *The Gulf Ecosystem Health and Sustainability*, Backhuys (pp. 127–156). <https://www.jstor.org/stable/10.14321/j.ctt1tm7jkg>
- Luoma, S. N. (1983). Bioavailability of trace metals to aquatic organisms—a review. *Science of the Total Environment*, 28(1–3), 1–22.
- Martin, S., & Griswold, W. (2009). Human health effects of heavy metals. *Environmental Science and Technology Briefs for Citizens*, 15, 1–6.
- Mason, R. P. (2013). *Trace metals in aquatic systems* (1st ed.). John Wiley & Sons.
- McCrum-Gardner, E. (2008). Which is the correct statistical test to use? *British Journal of Oral and Maxillofacial Surgery*, 46(1), 38–41.
- McLusky, D. S., & Hagerman, L. (1987). The toxicity of chromium, nickel and zinc: effects of salinity and temperature, and the osmoregulatory consequences in the mysid *Praunus flexuosus*. *Aquatic Toxicology*, 10(4), 225–238.
- McPherson, R., & Brown, K. (2001). The bioaccumulation of cadmium by the Blue Swimmer Crab *Portunus pelagicus* L. *Science of the Total Environment*, 279(1–3), 223–230.
- Mehri, A., & Marjan, R. F. (2013). Trace elements in human nutrition: A review. *International Journal of Medical Investigation*, 2(3), 115–128.
- Miri, R., & Chouikhi, A. (2005). Ecotoxicological marine impacts from seawater desalination plants. *Desalination*, 182(1), 403–410.
- Mohamed, A. M. O., Maraqa, M., & Al Handhaly, J. (2005). Impact of land disposal of reject brine from desalination plants on soil and groundwater. *Desalination*, 182(1), 411–433.
- Monalisa, M., & Kumar, P. H. (2013). Effect of ionic and chelate assisted hexavalent chromium on mung bean seedlings (*Vigna radiata* L. wilczek. var k-851) during seedling growth. *Journal of Stress Physiology & Biochemistry*, 9(2), 232–241.
- Monikh, F. A., Hosseini, M., & Rahmanpour, S. (2014). The effect of size and sex on PCB and PAH concentrations in crab *Portunus pelagicus*. *Environmental Monitoring and Assessment*, 186(3), 1575–1582.

- Moore, J. W., & Ramamoorthy, S. (2012). *Heavy metals in natural waters: applied monitoring and impact assessment*. Springer Science & Business Media.
- Musaiger, A. O., & Al-Rumaidh, M. J. (2005). Proximate and mineral composition of crab meat consumed in Bahrain. *International Journal of Food Sciences and Nutrition*, 56(4), 231–235.
- Musaiger, A. O., & D'Souza, R. (2008). Chemical composition of raw fish consumed in Bahrain. *Pak. J. Biol. Sci*, 11, 55–61.
- Naderloo, R. (2017). *Atlas of crabs of the Persian Gulf*. Springer, Cham.
- Nadim, F., Bagtzoglou, A. C., & Iranmahboob, J. (2008). Coastal management in the Persian Gulf region within the framework of the ROPME programme of action. *Ocean & Coastal Management*, 51(7), 556–565.
- Namminga, H. E. (1976). Effects of high discharge and an oil refinery cleanup operation on heavy metals in water and sediments in Skeleton Creek. *Proceedings of the Oklahoma Academy of Science*, 56, 133–138.
- Nasab, G. A., & Makizadeh, I. (2016). How can reduce the transfer of harmful aquatic organism and pathogens by ballast water in the ROPME Sea area. *International Journal of Biology, Pharmacy and Allied Sciences*, 5(8), 1923–1935.
- Naser, H. (2011). Human impacts on marine biodiversity: macrobenthos in Bahrain, Arabian Gulf. *The Importance of Biological Interactions in the Study of Biodiversity*; Lopez-Pujol, J., Ed, 109–126.
- Naser, H. (2013). Assessment and management of heavy metal pollution in the marine environment of the Arabian Gulf: a review. *Marine Pollution Bulletin*, 72(1), 6–13.
- Naser, H. (2014). Marine ecosystem diversity in the Arabian Gulf: threats and conservation. In *Biodiversity–The Dynamic Balance of the Planet* (pp. 297–328).
- Neufeld, D. S., & Cameron, J. N. (1993). Transepithelial movement of calcium in crustaceans. *Journal of Experimental Biology*, 184(1), 1–16.
- Niyogi, S., Blewett, T. A., Gallagher, T., Fehsenfeld, S., & Wood, C. M. (2016). Effects of salinity on short-term waterborne zinc uptake, accumulation and sub-lethal toxicity in the green shore crab (*Carcinus maenas*). *Aquatic Toxicology*, 178, 132–140.

- Noone, K. J., Sumaila, U. R., & Diaz, R. J. (2013). *Managing ocean environments in a changing climate: sustainability and economic perspectives*. Newnes.
- Nordic Council of Ministers. (2003). *Cadmium Review. Report No: 1, Issue No: 04, CRL. EHN*.
- Nørum, U., Bondgaard, M., Pedersen, T. V., & Bjerregaard, P. (2005). In vivo and in vitro cadmium accumulation during the moult cycle of the male shore crab *Carcinus maenas*—interaction with calcium metabolism. *Aquatic Toxicology*, 72(1–2), 29–44.
- Nugegoda, D., & Rainbow, P. S. (1989). Zinc uptake rate and regulation breakdown in the decapod crustacean *Palaemon elegans* rathke. *Ophelia*, 30(3), 199–212.
- Nürnberg, H. W. (1984). The voltammetric approach in trace metal chemistry of natural waters and atmospheric precipitation. *Analytica Chimica Acta*, 164, 1–21. [https://doi.org/10.1016/S0003-2670\(00\)85613-5](https://doi.org/10.1016/S0003-2670(00)85613-5)
- Olgunoğlu, M. P., & Olgunoğlu, İ. A. (2016). Heavy Metal Contents in Blue Swimming Crab from the Northeastern Mediterranean Sea, Mersin Bay, Turkey. *Polish Journal of Environmental Studies*, 25(5), 2233–2237.
- Piontkovski, S. A., & Chiffings, T. (2014). Long-term changes of temperature in the Sea of Oman and the western Arabian Sea. *International Journal of Oceans and Oceanography*, 8(1), 53–72.
- Plum, L. M., Rink, L., & Haase, H. (2010). The essential toxin: impact of zinc on human health. *International Journal of Environmental Research and Public Health*, 7, 1342–1365.
- Potter, I. C., Chrystal, P. J., & Loneragan, N. R. (1983). The biology of the blue manna crab *Portunus pelagicus* in an Australian estuary. *Marine Biology*, 78(1), 75–85.
- Potter, I. C., & De Lestang, S. (2000). Biology of the blue swimmer crab *Portunus pelagicus* in Leschenault Estuary and Koombana Bay, southwestern Australia. *Journal of the Royal Society of Western Australia*, 83(4), 443–458.
- Pourret, O., & Bollinger, J.-C. (2018). “Heavy metal” - What to do now: To use or not to use? *Science of The Total Environment*, 610–611, 419–420.
- Pourret, O., Bollinger, J.-C., & Hursthouse, A. (2021). Heavy metal: a

- misused term? *Acta Geochimica*, 40(3), 466–471.
- Pourret, O., & Hursthouse, A. (2019). It's Time to Replace the Term "Heavy Metals" with "Potentially Toxic Elements" When Reporting Environmental Research. *International Journal of Environmental Research and Public Health*, 16(22), 4446.
- Price, A. R. G., Sheppard, C. R. C., & Roberts, C. M. (1993). The Gulf: its biological setting. *Marine Pollution Bulletin*, 27, 9–15.
- Purves, D. (2012). *Trace-element Contamination of the Environment*. Elsevier.
- Rahmanpour, S., Ghorghani, N. F., & Ashtiyani, S. M. L. (2014). Heavy metal in water and aquatic organisms from different intertidal ecosystems, Persian Gulf. *Environmental Monitoring and Assessment*, 186(9), 5401–5409.
- Rainbow, P. S. (1985). Accumulation of Zn, Cu and Cd by crabs and barnacles. *Estuarine, Coastal and Shelf Science*, 21(5), 669–686.
- Rainbow, P. S. (1990). Heavy metal levels in marine invertebrates. In *Heavy metals in the marine environment* (pp. 67–79). CRC Press Boca Raton, FL.
- Rainbow, P. S., & Black, W. H. (2005). Cadmium, zinc and the uptake of calcium by two crabs, *Carcinus maenas* and *Eriocheir sinensis*. *Aquatic Toxicology*, 72(1–2), 45–65.
- Rainer, J., & Brouwer, M. (1993). Hemocyanin synthesis in the blue crab *Callinectes sapidus*. *Comparative Biochemistry and Physiology Part B: Comparative Biochemistry*, 104(1), 69–73.
- Reichmuth, J. M., Weis, P., & Weis, J. S. (2010). Bioaccumulation and depuration of metals in blue crabs (*Callinectes sapidus* Rathbun) from a contaminated and clean estuary. *Environmental Pollution*, 158(2), 361–368.
- Relic, D., Dordevic, D., Popovic, A., & Blagojevic, T. (2005). Speciations of trace metals in the Danube alluvial sediments within an oil refinery. *Environment International*, 31(5), 661–669.
- Reynolds, R. M. (1993). Physical oceanography of the Gulf, Strait of Hormuz, and the Gulf of Oman—Results from the Mt Mitchell expedition. *Marine Pollution Bulletin*, 27, 35–59.
- Roane, T. M., Peer, I. L., & Gentry, T. J. (2015). Chapter 18 À Microorganisms and Metal Pollutants. In *Environmental*

Microbiology. 3rd ed, 415–439. Amsterdam, The Netherlands: Academic Press.

- Romano, N., & Zeng, C. (2006). The effects of salinity on the survival, growth and haemolymph osmolality of early juvenile blue swimmer crabs, *Portunus pelagicus*. *Aquaculture*, 260(1–4), 151–162.
- Romano, N., & Zeng, C. (2007). Ontogenetic changes in tolerance to acute ammonia exposure and associated gill histological alterations during early juvenile development of the blue swimmer crab, *Portunus pelagicus*. *Aquaculture*, 266(1), 246–254.
- ROPME. (2010). Manual of oceanographic observations and pollutant analyses methods (MOOPAM). In *Moopam 1999* (Fourth). ROPME.
- ROPME. (2013). *State of the Marine Environment Report- 2013*. <http://ropme.org/>
- Roussel, A.-M., Favier, A. E., & Anderson, R. A. (2006). *Trace elements in man and animals 10*. Springer Science & Business Media.
- Ruilian, Y. U., Xing, Y., Yuanhui, Z., Gongren, H. U., & Xianglin, T. U. (2008). Heavy metal pollution in intertidal sediments from Quanzhou Bay, China. *Journal of Environmental Sciences*, 20(6), 664–669.
- Sadiq, M. (1992). *Toxic metal chemistry in marine environments*. Marcel Dekker, Incorporated.
- Sale, P. F., Feary, D. A., Burt, J. A., Bauman, A. G., Cavalcante, G. H., Drouillard, K. G., Kjerfve, B., Marquis, E., Trick, C. G., & Usseglio, P. (2011). The growing need for sustainable ecological management of marine communities of the Persian Gulf. *Ambio*, 40(1), 4–17.
- Sarasiab, A. R., & Hosseini, M. (2014). Study on PCB 101, PCB 153, mercury and methyl mercury content in blue crab *Portunus Pelagicus* from Khuzestan shore (Persian Gulf). *Toxicology and Environmental Health Sciences*, 6(2), 81–86.
- Saravanan, R., Sugumar, V., & Mahin, M. I. B. (2018). Heavy metal stress induced hyperglycemia in blue swimmer crab, *Portunus pelagicus*. *Acta Oceanologica Sinica*, 37(5), 47–53.
- Sauliutė, G., & Svecevičius, G. (2015). Heavy metal interactions during accumulation via direct route in fish: a review. *Zoology and Ecology*, 25(1), 77–86.
- Scawarz, K., & Mertz, W. (1959). Chromium (III) and the glucose tolerance factor. *Archives of Biochemistry*, 85, 292–295.

- SCE. (2015). *A Protected Areas Strategy for the Kingdom of Bahrain*. Supreme Council for Environment (SCE).
- SCE. (2018). *Marine Water Quality Monitoring in Bahrain (2007-2017)*. Supreme Council for Environment (SCE).
- SCE. (2020). *Marine Environment Quality of Tubli Bay (2009-2020)*. Supreme Council for Environment (SCE).
- Schiffer, R. B., William Sunderman, F., Baggs, R. B., & Moynihan, J. A. (1991). The effects of exposure to dietary nickel and zinc upon humoral and cellular immunity in SJL mice. *Journal of Neuroimmunology*, 34(2), 229–239.
- Schintu, M., Marras, B., Durante, L., Meloni, P., & Contu, A. (2010). Macroalgae and DGT as indicators of available trace metals in marine coastal waters near a lead–zinc smelter. *Environmental Monitoring and Assessment*, 167(1–4), 653–661.
- Sheppard, C. (1993). Physical environment of the Gulf relevant to marine pollution: an overview. *Marine Pollution Bulletin*, 27, 3–8. [https://doi.org/10.1016/0025-326X\(93\)90003-3](https://doi.org/10.1016/0025-326X(93)90003-3)
- Sheppard, C., Al-Husiani, M., Al-Jamali, F., Al-Yamani, F., Baldwin, R., Bishop, J., Benzoni, F., Dutrieux, E., Dulvy, N. K., Durvasula, S. R. V, Jones, D. A., Loughland, R., Medio, D., Nithyanandan, M., Pilling, G. M., Polikarpov, I., Price, A. R. G., Purkis, S., Riegl, B., ... Zainal, K. (2010). The Gulf: A young sea in decline. *Marine Pollution Bulletin*, 60(1), 13–38.
- Sheppard, C., & Loughland, R. (2002). Coral mortality and recovery in response to increasing temperature in the southern Arabian Gulf. *Aquatic Ecosystem Health & Management*, 5(4), 395–402. <https://doi.org/10.1080/14634980290002020>
- Sheppard, C., Price, A., & Roberts, C. (1992). *Marine ecology of the Arabian region: patterns and processes in extreme tropical environments* (1 edition). London (United Kingdom) Academic Press.
- Shukla, G. S., & Singhal, R. L. (1984). The present status of biological effects of toxic metals in the environment: lead, cadmium, and manganese. *Canadian Journal of Physiology and Pharmacology*, 62(8), 1015–1031.
- Siddiqui, M. F. (2010). Cadmium induced renal toxicity in male rats, *Rattus rattus*. *Eastern Journal of Medicine*, 15(3), 93.
- Sigel, A., Sigel, H., & Sigel, R. K. O. (2013). *Interrelations between*

essential metal ions and human diseases (Vol. 13). Springer.

- Simonetti, P., Botté, S. E., Fiori, S. M., & Marcovecchio, J. E. (2013). Burrowing Crab (*Neohelice granulata*) as a Potential Bioindicator of Heavy Metals in the Bahía Blanca Estuary, Argentina. *Archives of Environmental Contamination and Toxicology*, 64(1), 110–118.
- Singh, R., Gautam, N., Mishra, A., & Gupta, R. (2011). Heavy metals and living systems: An overview. *Indian Journal of Pharmacology*, 43(3), 246.
- Sinicropi, M. S., Amantea, D., Caruso, A., & Saturnino, C. (2010). Chemical and biological properties of toxic metals and use of chelating agents for the pharmacological treatment of metal poisoning. *Archives of Toxicology*, 84(7), 501–520.
- Smith, G. S., & Sumpton, W. D. (1987). Sand crabs a valuable fishery in southeast Queensland. *Qld Fisherman*, 5, 13–15.
- Stankovic, S., Kalaba, P., & Stankovic, A. R. (2014). Biota as toxic metal indicators. *Environmental Chemistry Letters*, 12(1), 63–84.
- Stankovic, S., & Stankovic, A. R. (2013). Bioindicators of toxic metals. In *Green materials for energy, products and depollution* (pp. 151–228). Springer.
- Stern, B. R., Solioz, M., Krewski, D., Aggett, P., Aw, T.-C., Baker, S., Crump, K., Dourson, M., Haber, L., & Hertzberg, R. (2007). Copper and human health: biochemistry, genetics, and strategies for modeling dose-response relationships. *Journal of Toxicology and Environmental Health, Part B*, 10(3), 157–222.
- Sukumaran, K. K., & Neelakantan, B. (1997). Age and growth in two marine portunid crabs, *Portunus* (*Portunus*) *sanguinolentus* (Herbst) and *Portunus* (*Portunus*) *pelagicus* (Linnaeus) along the southwest coast of India. *Indian Journal of Fisheries*, 44(2), 111–131.
- Swift, S. A. (2003). Formation and circulation of dense water in the Persian/Arabian Gulf. *Journal of Geophysical Research*, 108(C1), 4-1-4–21.
- Tchounwou, P. B., Yedjou, C. G., Patlolla, A. K., & Sutton, D. J. (2012). Heavy metal toxicity and the environment. In *Molecular, clinical and environmental toxicology* (pp. 133–164). Springer.
- Truchet, D. M., Buzzi, N. S., Simonetti, P., & Marcovecchio, J. E. (2020). Uptake and detoxification of trace metals in estuarine crabs: insights into the role of metallothioneins. *Environmental Science and Pollution*

Research, 27(25), 31905–31917.

- Tsutsumi, T., Ishihara, A., Yamamoto, A., Asaji, H., Yamakawa, S., & Tokumura, A. (2014). The potential protective role of lysophospholipid mediators in nephrotoxicity induced by chronically exposed cadmium. *Food and Chemical Toxicology*, 65, 52–62.
- Turner, A. (2010). Marine pollution from antifouling paint particles. *Marine Pollution Bulletin*, 60(2), 159–171.
- Uma Devi, V., & Prabhakara Rao, Y. (1989). Zinc accumulation in fiddler crabs *Uca annulipes latreille* and *Uca triangularis* (milne edwards). *Ecotoxicology and Environmental Safety*, 18(2), 129–140.
- UNEP. (1999). *Overview on Land-Based sources and activities affecting the marine environment in ROPME Sea Area*.
- Valavanidis, A., & Vlachogianni, T. (2010). Metal pollution in ecosystems. Ecotoxicology studies and risk assessment in the marine environment. *Dept. of Chemistry, University of Athens University Campus Zografou*, 15784.
- Verboost, P. M., Van Rooij, J., Flik, G., Lock, R. A. C., & Bonga, S. E. W. (1989). The movement of cadmium through freshwater trout branchial epithelium and its interference with calcium transport. *Journal of Experimental Biology*, 145(1), 185–197.
- Viswanathan, C., Azhaguraj, R., Selvanayagam, M., & Raffi, S. M. (2013). Heavy metal levels in different tissues of the Blue Swimming crab (*Portunus pelagicus*, *Portunidae*) collected from Ennore Estuary. *International Journal of Research in Fisheries and Aquaculture*, 3(1), 1–6.
- Waiwood, B. A., Zitko, V., Haya, K., Burrige, L. E., & Mcleese, D. W. (1987). Uptake and excretion of zinc by several tissues of the lobster (*Homarus Americanus*). *Environmental Toxicology and Chemistry*, 6(1), 27–32.
- White, S. L., & Rainbow, P. S. (1982). Regulation and accumulation of copper, zinc and cadmium by the shrimp *Palaemon elegans*. *Mar. Ecol. Prog. Ser.*, 8(95), 101. <https://www.jstor.org/stable/24814627>
- WHO. (1996). *Trace elements in human nutrition and health*. World Health Organization. <https://apps.who.int/iris/handle/10665/37931>
- Williams, M. J. (1981). Methods for analysis of natural diet in portunid crabs (Crustacea: Decapoda: Portunidae). *Journal of Experimental Marine Biology and Ecology*, 52(1), 103–113.

- Wolfe, M. F., Schwarzbach, S., & Sulaiman, R. A. (1998). Effects of mercury on wildlife: a comprehensive review. *Environmental Toxicology and Chemistry: An International Journal*, 17(2), 146–160.
- Wright, D. A. (1995). Trace metal and major ion interactions in aquatic animals. *Marine Pollution Bulletin*, 31(1–3), 8–18.
- Zainal, K., Al-Sayed, H., Ghanem, E., Butti, E., & Nasser, H. (2007). Baseline ecological survey of Huwar Islands, The Kingdom of Bahrain. *Aquatic Ecosystem Health & Management*, 10(3), 290–300.
- Zainal, Khadija. (2013). Natural food and feeding of the commercial blue swimmer crab, *Portunus pelagicus* (Linnaeus, 1758) along the coastal waters of the Kingdom of Bahrain. *Journal of the Association of Arab Universities for Basic and Applied Sciences*, 13(1), 1–7.
- Zainal, Khadija. (2017). Relative growth and heterochely in the blue swimmer crab *Portunus pelagicus* (Linnaeus 1758) from the Kingdom of Bahrain. *Arabian Journal for Science and Engineering*, 42(1), 75–84.
- Zainal, Khadija, Al-Sayed, H., & Al-Madany, I. (2008). Coastal pollution in Bahrain and its management. In A. H. Abuzinada, H.-J. Barth, F. Krupp, B. Böer, & T. Z. Al Abdessalaam (Eds.), *Protecting the Gulf's marine ecosystems from pollution* (pp. 147–162). Springer Science & Business Media.
- Zhao, J., Temimi, M., Al Azhar, M., & Ghedira, H. (2015). Satellite-based tracking of oil pollution in the Arabian Gulf and the Sea of Oman. *Canadian Journal of Remote Sensing*, 41(2), 113–125.
- Zhao, S., Feng, C., Quan, W., Chen, X., Niu, J., & Shen, Z. (2012). Role of living environments in the accumulation characteristics of heavy metals in fishes and crabs in the Yangtze River Estuary, China. *Marine Pollution Bulletin*, 64(6), 1163–1171.

الخلاصة

يتواجد سرطان البحر الأزرق السابح (*Portunus pelagicus*) بكثرة في المحيطين الهندي والهادي، والعديد من المناطق البحرية الأخرى بما في ذلك منطقة الخليج العربي. يراكم سرطان البحر الأزرق السابح أيونات العناصر النزرة في أنسجة جسمه المختلفة كباقي القشريات القاعية التي تعيش في البيئة البحرية. ويعتبر سرطان البحر الأزرق السابح من الأطعمة البحرية المتداولة في البحرين، لذا من المهم دراسة مستوى أيونات العناصر النزرة في أنسجته.

في هذه الدراسة تم قياس تراكيز ستة أيونات للعناصر النزرة في أنسجة ذكور وإناث سرطان البحر الأزرق السابح التي تم جمعها من أربعة مواقع بحرية في المياه الإقليمية لمملكة البحرين، وهي رأس المطة (الساحل الغربي)، ومرودة (الساحل الشمالي)، وخليج تولبي، وعسكر (الساحل الشرقي). نُفذت هذه الدراسة على مرحلتين، في المرحلة الأولى تم قياس تراكيز كل من Zn^{2+} و Cu^{2+} و Cr^{3+} و Ni^{2+} و Pb^{2+} و Cd^{2+} في كل من اللحم الأبيض (عضلات الأرجل والمخالب) واللحم البني (الجهاز الهضمي) في ذكور وإناث سرطان البحر الأزرق السابح على أساس الوزن الجاف، حيث تم جمع عينة تضم أنسجة 15 سرطاناً من الذكور وأخرى من الإناث من كل موقع. وبناءً على نتائج المرحلة الأولى لهذه الدراسة تم اختيار أيونات ثلاثة عناصر وهي Zn^{2+} و Cu^{2+} و Cd^{2+} لتكون ضمن المرحلة الثانية لهذه الدراسة التي اعتمدت على الوزن الرطب في الكشف عن الفروق ذات الدلالة الإحصائية بناءً على نوع اللحم والجنس والموقع. حيث تم قياس تراكيز أيونات هذه العناصر النزرة في أنسجة اللحم الأبيض واللحم البني في 10 من سرطان البحر بشكل منفرد لكل من الذكور والإناث وفي المواقع الأربعة.

تراوحت تراكيز أيونات Zn^{2+} و Cu^{2+} و Cr^{3+} و Ni^{2+} و Cd^{2+} في حدود 325.7-33.3، 300.4-174.6، 10.3 - 0.02، 3.03-0.11، 0.25-0.025 ملجم/كجم ووزن جاف، على التوالي، في حين كان مستوى Pb^{2+} أقل من حد القياس الممكن لجهاز التحليل. وكان الترتيب السائد لأيونات العناصر النزرة على الشكل التالي $Zn^{2+} > Cu^{2+} > Cd^{2+} > Ni^{2+} > Cr^{3+}$. أظهرت نتائج هذه الدراسة أن سرطان البحر الأزرق السابح يراكم أيونات كل من Cu^{2+} و Cd^{2+} في أنسجة اللحم البني أكثر من اللحم الأبيض، بينما لم يتم رصد فروق ذات دلالة إحصائية في مراكمة Zn^{2+} بين أنسجة اللحم الأبيض والبني. وأظهرت النتائج أيضاً أن إناث سرطان البحر الأزرق السابح تراكم Zn^{2+} في لحومها البيضاء والبنية أكثر من الذكور، وكذلك بالنسبة إلى Cu^{2+} في أنسجة اللحم الأبيض، في حين لم يتم رصد فروق ذات دلالة إحصائية في مراكمة Cd^{2+} في كل من اللحم الأبيض والبني وكذلك Cu^{2+} في أنسجة اللحم البني بين الذكور والإناث. أما فيما يخص الفروق بين المواقع فقد أظهرت النتائج أن السرطانات التي تعيش في موقع رأس المطة تراكم Cd^{2+} و Cu^{2+} في اللحم البني أعلى من باقي السرطانات التي تعيش في المواقع الأخرى. في حين أن السرطانات التي تعيش في موقع مرودة تحتوي على مستويات من Zn^{2+} في اللحم الأبيض أعلى من باقي المواقع، وأن السرطانات التي تعيش في موقع مرودة وخليج تولبي تحتوي على مستويات من Zn^{2+} في اللحم البني أعلى من باقي المواقع.

في ضوء نتائج هذه الدراسة، تعتبر مستويات Pb^{2+} و Cd^{2+} في اللحم الأبيض من أنسجة سرطان البحر الأزرق السابح في البحرين آمنة للاستهلاك البشري وفقاً للمعايير المعمول بها في مملكة البحرين وكذلك في الاتحاد الأوروبي. ومع ذلك، يوصى بالامتناع عن تناول اللحم البني لتجنب الآثار السلبية الناتجة عن المستوى العالي لأيون Cd^{2+} .

جامعة البحرين

برنامج البيئة والتنمية المستدامة

كلية العلوم



تراكم ملوثات العناصر النزرة في سرطان البحر الأزرق الساحب في البحرين

بحث مقدم استكمالاً لمتطلبات الحصول على درجة الماجستير في البيئة والتنمية المستدامة

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