

**Metal and metalloid bioaccumulation in
the Cape white seabream (*Diplodus
capensis*) and the tropical halfbeak
(*Hyporhamphus affinis*)**

W Neethling

 **orcid.org 0000-0002-6174-5511**

Dissertation accepted in fulfilment of the requirements for the
degree *Master of Science in Environmental Sciences* at the
North-West University

Supervisor:	Prof V Wepener
Co-supervisor:	Prof NJ Smit
Co-supervisor:	Dr JH Erasmus

Graduation May 2024

Acknowledgements

Thank you to our Lord and saviour Jesus Christ for the ability and knowledge to complete this dissertation. Thank you for blessing me throughout my studies even in times of uncertainty and providing me with so many opportunities.

I would like to express my profound gratitude to the following individuals and organisations who played pivotal roles at various stages of this project.

- First and foremost, I extend my heartfelt thanks to my supervisors, Prof. Victor Wepener, Prof. Nico Smit, and Dr. Hannes Erasmus. Their support and guidance throughout this project were truly invaluable. I appreciate the opportunities they provided me, not only for my involvement in this project but also for the acquisition of numerous new skills that will undoubtedly contribute to a successful career in this field.
- I would like to express my gratitude to the North-West University (NWU), Unit of Environmental Sciences and Management and the Water Research Group (WRG) for granting me with access to equipment and facilities that were instrumental in the successful execution of this project. Furthermore, I appreciate the opportunities I was given to showcase this research on a national stage at multiple conferences.
- My sincere appreciation goes to the Faculty of Natural and Agricultural Sciences Research Ethics Committee at NWU, whose provision of ethical clearance was essential in making this project successful (Ethics no: NWU-01425-20-A9).
- I wish to acknowledge Dr. Anja Erasmus for her assistance in creating the maps used in this dissertation.
- This work is based on the research and researchers supported by The Department of Science and Innovation (DSI) and the National Research Foundation (NRF) of South Africa (Grant number PMDS22060920723, Grant number 138544, JH Erasmus) in collaboration with the German Academic Exchange Service (DAAD). Opinions, findings, conclusions, and recommendations in this publication are that of the authors, and the NRF and DAAD accepts no liability whatsoever in this regard.
- Lastly, thank you to my parents (Wiets and Sanet Neethling) for your unwavering support and giving me the opportunity to further educate myself. Thank you for your motivation and encouragement throughout my studies.

Abstract

Metal and metalloid (collectively referred to as elements) concentrations in fish muscle and liver (tissues) along the South African coastline result from complex interactions of anthropogenic sources and natural processes. Anthropogenic sources, originating from human activities, introduce elements into the marine environment. Industrial discharges are known to contain Cd, Hg, and Pb, posing health risks to marine ecosystems and regular human consumers. Conversely, natural processes such as geological weathering and erosion can release elements in the marine environment leading to increased concentrations in fish tissue. Understanding anthropogenic sources and natural processes is vital for maintaining a healthy marine ecosystem and ensuring safe consumption of marine fish meat.

The present study aimed to investigate the element concentrations in tissues of *Hyporhamphus affinis* (tropical halfbeak) from Sodwana Bay during two surveys and assess the associated human health risks from consuming contaminated *H. affinis*. The hypothesis was that there would be differences in element concentrations in tropical halfbeaks between the pre-COVID (2018) and post-COVID (2022) periods at Sodwana Bay. This was tested by comparing element concentrations in *H. affinis* muscle and liver tissues collected in 2018 and 2022 and evaluating the potential non-carcinogenic and carcinogenic health risks. Similarly, element concentrations in tissues of *Diplodus capensis* (Cape white seabream) from both minimally impacted and anthropogenically impacted sites along the South African coastline were investigated from 2020 until 2022. The aim was to assess the human health risks related to the bioaccumulation of elements in *D. capensis*. The assumption was that element accumulation in the Cape white seabream would reflect land-use activities, leading to higher health risks for consumers in contaminated areas. This assumption was tested by collecting specimens from different sites, analysing element concentrations in *D. capensis* tissues, determining element bioavailability in the marine ecosystem, as well as calculating non-carcinogenic and carcinogenic health risks associated with consuming contaminated *D. capensis*.

The results indicated that recreational activities decreased at the beginning of the COVID-19 pandemic in early 2020 at Sodwana Bay and continued to be impacted by lockdown restrictions until the end of 2021, in contrast to the higher levels of activity seen before the pandemic. Analysis of element concentrations in the tissues of *H. affinis* revealed distinct trends. Notably, there was a significant decrease in the levels of elements such as As, Cd, Cu, Fe, Hg, and Ni in tissues. Conversely, there was a notable increase in the

concentrations of Cr, Mn, Pb, Sb and Sn in tissues. The present study observed that along the South African coastline, Chintsa had significantly higher element concentrations in muscle tissues compared to most other sites, while other sites showed relatively similar element concentrations in *D. capensis*. Among the different time intervals, Tsitsikamma (2021) and Chintsa (2022) had the highest element concentrations in muscle tissue. Elements in liver tissue consistently showed significant increases in most elements at Chintsa (2022) and Witsand (2021), except for Pb, which had a higher concentration in Mossel Bay. Furthermore, De Hoop (2021) and Witsand (2021) had significantly higher levels of Sn compared to the other study sites along the coast. Again, fish from Tsitsikamma (2021) and Chintsa (2022) presented the highest concentration levels within liver tissue when comparing different time intervals.

The analysed elements had the potential to present carcinogenic or non-carcinogenic risks to regular human consumers. Specifically, in *H. affinis* muscle tissue, non-carcinogenic risks were associated with As and Hg, while carcinogenic risks were linked to As, Cr, Ni and Pb. It is important to highlight that only the concentrations of As and Hg exceeded the acceptable thresholds for non-carcinogenic risks in *D. capensis* across all sites along the South African coastline. The assessment of the carcinogenic risks associated with these elements in *D. capensis* from the surveyed locations along the South African coastline revealed that As presented a risk to people consuming *D. capensis* at all sites examined.

The present study concluded that variations in element concentrations between the pre-COVID and post-COVID periods in *H. affinis* were primarily due to reduced human activities in Sodwana Bay. Natural processes, such as regular upwelling events and wind-driven erosion, also contributed to the observed element concentrations, as well as aerial Hg depositions from the Mpumalanga highveld led to increased Hg levels in tissues. Furthermore, Chintsa (2022) and Witsand (2021) showed the highest element concentrations, even though there are no known pollution sources. Mossel Bay (2022) had elevated Pb levels due to increased maritime traffic. The hypothesis of higher non-carcinogenic and carcinogenic risks in impacted sites was not supported, as risks were present in relatively unimpacted sites. Element concentrations in *D. capensis* were influenced by regular upwelling events and dispersed sources, such as geological weathering.

Keywords:

Bioaccumulation, bioavailability, element concentrations, human health risk, marine fish, South African coastline, upwelling

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List of Abbreviations

<i>Full term</i>	<i>Abbreviation</i>
Average Daily Dosage	ADD
Analysis of Variance	ANOVA
Cancer Risk	CR
Certified Reference Material	CRM
Department of Agriculture, Forestry and Fisheries	DAFF
Department of Environmental Affairs and Tourism	DEAT
Estimated Daily Intake	EDI
Element	Elem.
Hazard Quotient	HQ
KwaZulu-Natal	KZN
Level of Concern	L.C.
Marine Protected Area	MPA
Not certified	N.c.
Not detected	N.d.
Reference dose	RfD
South African National Park	SANPark
Standard length	SL
Total length	TL
United Nations Educational, Scientific and Cultural Organization	UNESCO
World Health Organization	WHO

Chapter 1: General Introduction

1.1 Background

1.1.1 The South African coastline

According to Attwood *et al.* (2002), the marine environment along the South African coastline is a rich and diverse national asset that not only offers economic and social opportunities but also sustains the livelihoods of its population. However, the reliance on the marine environment for resources, products, transportation, and economic gain has exerted substantial pressure on South Africa's marine ecosystem (Taljaard *et al.*, 2006; DEAT, 2008). Therefore, the sustainable use and management of coastal resources are of paramount importance to ensure the future development of the country (DEAT, 2008). Although South Africa has placed high priority on raising awareness of marine conservation, there is a notable gap in marine pollution research within the country, which hinders these conservation efforts. South Africa's coastline stretches more than 3100 km, with two major ocean currents, the warm Agulhas on the east coast and the cold Benguela on the west coast, resulting in a diverse range of habitats and biodiversity (Griffiths *et al.*, 2010; Wepener and Degger, 2019). This unique ecological setting suggests that the influx of pollutants into the Southern Hemisphere's oceans is expected to be significantly lower compared to the heavily industrialised Northern Hemisphere. South Africa has 23 designated Marine Protected Areas (MPAs), which play a vital role in ensuring the sustainable management of line fish resources. These areas provide safety for both juvenile fish and spawning stocks of reef-associated species. Research by Mann (2013) has shown the positive impact of these MPA's on both line fish species and recreational anglers. However, the South African coastline is currently under increasing pressure from the development of new towns and industries. The surge in urbanisation and industrialisation is contributing to an increased discharge of pollutants into the marine environment, consequently raising the overall pollutants delivered to the ocean (McGlashan, 1989).

1.1.2 Sources of element contamination in the marine environment

Naturally occurring elements within the marine environment can have adverse ecological consequences. While certain elemental contaminants enter the South African coastal ecosystems through natural processes (Gregory *et al.*, 2002), the majority results from human activities in river catchment areas that flow into the marine environment (Mzimela *et al.*, 2003; Wepener and Vermeulen 2005; Vogt *et al.*, 2019). Highly productive regions

worldwide, such as harbours, experience an influx of elements, primarily due to their proximity to pollutant sources and their location at the end of catchments that receive metal inputs (Degger *et al.*, 2011; Degger *et al.*, 2016; Moodley *et al.*, 2016). Harbours, which serve a crucial economic role, also function as essential ecosystems providing shelter, feeding grounds, and nursery areas for a diverse range of organisms (Wepener, 2008). Elements known for their stability are persistent environmental contaminants in coastal waters. South African rivers and estuaries along the coast exhibit elevated levels of elements such as Cd, Cu, Hg and Pb, indicating the influence of industrial activity and urbanisation near rivers on the metal content of aquatic environments (Binning and Baird, 2001; Learner *et al.*, 2009; Hutchings and Clark, 2010; Jooste *et al.*, 2014; Olaniran *et al.*, 2014). These contaminants can be transported downstream, eventually reaching marine systems (Oosthuizen and Ehrlich, 2001). The interest in elements such as Cu, Fe, Mn and Zn, which are essential for metabolic activity in fish, lies within the delicate balance between their essentiality and toxicity. In contrast, other elements such as Cd, Cr, Hg and Pb may exhibit extreme toxicity, even at low concentrations under certain conditions, necessitating regular monitoring of sensitive aquatic environments (Peerzada *et al.*, 1990).

Element sources can be both from anthropogenic activities and natural weathering processes associated with adjacent geological formations (Song *et al.*, 2014). It is well-documented in scientific literature that geological processes alone can lead to increased element concentrations even in the absence of industrial or human interventions. These natural sources include occurrences such as forest fires, weathering, erosion, and rock abrasion. Furthermore, the partitioning of polluting elements, often concentrated within clay minerals characterised by high absorption capacities, plays a role in element release. This partitioning occurs between sedimentary rocks, their precursor sediments, and the surrounding water. These elements are deposited within beach sediments through adsorption, hydrolysis, and coprecipitation mechanisms, with a smaller fraction remaining as free metal ions within the water column (Bartoli *et al.*, 2012). On South Africa's east coast, the warm Agulhas Current contains approximately 90% of the estuaries that serve as outlets for freshwater basins into the ocean. The occurrence of water upwelling along the eastern coast of South Africa, at the edge of the Agulhas Current, is a regular occurrence that extends along its entire span where water moves from the coastal side to the continental slope (Lutjeharms *et al.*, 2000). According to Roberts and Nieuwenhuys (2016), the Agulhas Current showed that nearly all significant and minor instances of cold-water influx aligned with north-easterly winds are conducive to upwelling. According to

Lehmann and Myrberg (2008), upwelling typically results from horizontal divergence in the ocean's surface layer. This process involves wind-driven movement of cooler, denser, and nutrient-rich water towards the ocean surface, displacing the warmer, mostly nutrient-depleted surface waters. In monitoring the pollution status of marine environments, bioindicator organisms such as fish have proven to be effective. The suitability of a particular fish species as a bioindicator species is dependent on factors such as its mobility, commercial value, distribution, habitat, and prevalence (Zhang *et al.*, 2020).

1.1.3 Fish as a bioindicator organisms

As established, given the importance of the marine ecosystem in supplying food globally and locally, the content of pollutants in consumed marine species needs to be investigated (O'Donoghue and Marshall, 2003). *Diplodus capensis* (Cape white seabream) is native to South African waters and is frequently caught in harbours or shallow rocky habitats at depths less than 60 m (Coetzee, 1986; Mann, 2013), while *Hyporhamphus affinis* (tropical halfbeak) is a reef fish found at depths between 0-6 m. Both species may be susceptible to the influence of element pollution in coastal waters. Bioindicator species serve as repositories for contaminants in their tissue and are valuable tools for assessing the bioavailability of pollutants in natural environments (Rainbow and Phillips, 1993; de Silva *et al.*, 2009). Among these bioindicators, nearshore fish species such as *D. capensis* and *H. affinis* are known to accumulate elevated concentrations of elements, primarily due to their dietary preferences and their occupancy of diverse habitats (Authman *et al.*, 2015). The selection of bioindicator species is based on their responsiveness to environmental shifts and their ability to accumulate elements within their tissues. From an ecological perspective, fish occupy a prominent position in the aquatic food chain, making them highly susceptible to contamination by elements, which, in turn, reflects the biological and environmental conditions of their aquatic surroundings (Dragun *et al.*, 2009). *Diplodus capensis* and *H. affinis* are omnivorous species, feeding on benthic prey, zooplankton, smaller fish, and detritus organic matter, making them more susceptible to the bioaccumulation of elements in their tissues. Despite the importance of *D. capensis* and *H. affinis* in small-scale commercial and recreational fishing within South Africa (Smale, 1992; Penney *et al.*, 1999; Mann, 2013), there is limited information concerning the safety of consuming these species in terms of metal toxicity and potential risks it poses to regular human consumers.

1.1.4 Human health risks

Naturally, elements are present in the marine environment at low concentrations. However, due to the proximity of coastal waters to human activities such as harbours, urban development, and recreation, element concentrations in marine environments are steadily increasing (Greenfield *et al.*, 2014). Since fish are a significant part of the human diet, their consumption, if contaminated, poses a risk to public health. Element contamination is a major concern due to the persistence, non-biodegradability, biogeochemical cycling, and ecotoxicological hazards associated with these substances (Xu *et al.*, 2016). Gu *et al.* (2015) demonstrated that toxic elements can have adverse effects on human health when consumed over extended periods. Similarly, essential elements can also become toxic when ingested in excessive amounts. Fish have been found to accumulate elements in both high and low concentrations, which can potentially be transferred to humans who consume them and to other organisms in the subsequent trophic levels of the food web. From a nutritional standpoint, elements can be categorised as either essential or non-essential. Essential elements such as Cu, Cr, Fe, Mn, Se and Zn are critical for maintaining proper human metabolism but in trace amounts. Non-essential elements, such as Cd, Hg and Pb, lack an essential role in the human body and may cause harm even at trace concentrations (Rahmani *et al.*, 2018). Therefore, the potential health benefits associated with fish consumption may be compromised by the presence of toxic elements, which can have adverse effects on the human body if consumed in excessive quantities (Bosch *et al.*, 2016). Overexposure to various trace elements can increase health risks, encompassing both non-carcinogenic effects like damage and impaired function of organs such as liver, kidney, and thyroid, as well as carcinogenic effects, leading to conditions such as thyroid, kidney, liver, and bone cancer (Pheiffer *et al.*, 2018; Volschenk *et al.*, 2019; Balali-Mood *et al.*, 2021). In numerous developing countries, including South Africa, many rural communities and low-income groups depend on fish harvested by subsistence fishers from aquatic environments as a vital source of nutritional protein (Ellender *et al.*, 2009; Kwarazuka and Béné, 2011; McCafferty *et al.*, 2012; Coetzee *et al.*, 2015; Zereini *et al.*, 2015; Tran *et al.*, 2019; Weyl *et al.*, 2021). However, communities that regularly consume contaminated fish face chronic exposure to pollutants. People in developing countries are not only exposed to contaminated dust, water, and soil, but also to the health risks associated with consuming fish from polluted aquatic ecosystems (Erasmus *et al.*, 2022b)

1.2 Problem statement, hypotheses, and aims.

Research on element contamination in marine fish around the African continent is currently limited, and there is a pressing need for further investigations. Understanding the potential sources of elevated element levels in marine fish, whether they derive from human activities or natural events, is crucial, as it can pose environmental and health risks for both ecosystems and humans. This is essential to ensure that fish, widely recognised as a nutritious food source, does not unexpectedly pose health risks to consumers (Bosch *et al.*, 2016). Despite routine monitoring of commercial fish, the absence of comprehensive information on elements, particularly Hg, in South African marine fish raises significant concern for both consumer health and the economic stability of the fishing industry. This knowledge gap leaves consumers without guidelines on which fish to avoid or which areas to stay away from to reduce the risk of consuming fish with Hg levels exceeding permissible limits (Bosch *et al.*, 2016). Research on element concentrations in frequently consumed fish species, particularly in the African continent, is still ongoing. Such studies are crucial to ascertain the actual toxicity levels and potential impacts on consumers.

1.2.1 General hypotheses

The hypotheses tested in the present study:

1. There will be differences in element concentrations of *H. affinis* between the periods before and after COVID-19 (2018 and 2022), due to the decrease in economic and recreational activities at the Sodwana Bay site.
2. The bioaccumulation of elements in *D. capensis* will reflect element concentrations associated with land-use activities, and specimens from these areas would present higher non-carcinogenic and carcinogenic risks to regular human consumers when compared to fish from the sites with less land-use activities such as Chintsa and Tsitsikamma.

1.2.2 Aims

To test the above hypotheses, the main aims of the present study were to:

- Determine the concentrations of elements in the muscle and liver tissues of *H. affinis* from Sodwana Bay during two surveys.
- Determine whether the COVID lockdown period had an influence on element levels in *H. affinis*.
- Assess the potential health risks the elements pose to human consumers.

- Evaluate the element concentrations in muscle and liver tissues of *D. capensis* collected from both relatively unaffected (Chintsa, De Hoop and Tsitsikamma) and human impacted (Mossel Bay and Witsand) sites along the South African coast.
- Compare element concentrations in muscle and liver tissue of *D. capensis* collected from Chintsa and Tsitsikamma between different time periods.
- Assess the potential human health risks related to the bioaccumulation of elements in muscle tissue of *D. capensis*.

1.3 Chapter layout

The dissertation has been divided into four chapters:

Chapter 1: General introduction providing background information on the rationale of the study, as well as the specified hypotheses and aims.

Chapter 2: An assessment in Sodwana Bay to examine the bioaccumulation of elements in muscle and liver tissue of *H. affinis* was carried out during two surveys, one conducted in 2018, characterised by high recreational activities (pre-COVID), and the other in 2022, marked by a reduction in recreational activities (post-COVID). Additionally, a human health risk assessment was performed to assess potential health risks associated with element overexposure.

Chapter 3: An assessment done along the South African coastline to investigate the bioaccumulation of elements in muscle and liver tissues of *D. capensis*, comparing tissues from relatively unimpacted sites and impacted sites. In addition, a human health risk assessment was conducted to examine the potential risks associated with overexposure to elements.

Chapter 4: The study's conclusions and suggestions for future research pertaining to the bioaccumulation of elements in fish species along the South African coastline and human health risk assessments.

Chapter 2: Element bioaccumulation in the tropical halfbeak (*Hyporhamphus affinis*) from Sodwana Bay

2.1 Introduction

Sodwana Bay is located on the Maputoland coast of South Africa's KwaZulu-Natal (KZN) province. Sodwana Bay is an integral part of Isimangaliso Wetland Park, which is recognised as South Africa's first world heritage site (UNESCO, 1999; Whitfield, *et al.*, 2013). It is interconnected with the adjacent Maputoland Marine Reserve as a marine protected area (MPA). In 2016, a total of 67 325 dives were recorded in Sodwana Bay alone (van der Schyff *et al.*, 2020). This region encompasses a stretch of approximately 179 km along the KZN coastline, extending from the mouth of the St. Lucia estuary to the border between South Africa and Mozambique. It is characterised by vast interconnected lakes, forested dunes, sandy beaches, and low cliffs. The area is predominantly dominated by expansive wetlands that host a diverse range of fauna. These waters are unique to South Africa, as they are within the tropical region, resulting in their warm temperature (Mograbi and Rogerson, 2007). Sodwana Bay is distinguished as the southernmost equatorial point where tropical species and coral reefs can be found in South Africa. The combination of distinctive natural features and cultural allure makes Sodwana Bay a preferred destination for tourists, offering abundant aquatic species and plant diversity. *Hyporhamphus affinis* is a popular fish species in this area and is used by local fishermen as bait and a source of protein as it is highly abundant.

Hyporhamphus affinis (38 cm SL) is a species of marine fish that thrives in coastal, estuarine, and marine environments (Kottelat *et al.*, 1993). They have posterior dorsal and anal fins, a forked caudal fin with an elongated lower lobe, and an elongated body form that is usually compressed (Kara *et al.*, 2012). In particular, the lower jaw of *H. affinis* is often longer than the upper jaw. Belonging to the Hemiramphidae family, which is one of the five families within the Beloniformes order, there are 109 species distributed across 12 genera (Nelson, 2016). Primarily inhabiting coral reefs, *H. affinis* feeds on small fish, zooplankton and detritus. *Hyporhamphus affinis* is an omnivorous species and can therefore be susceptible to the accumulation of elements through their diet and the aquatic environment.

Based on a recent comprehensive analysis of coastal regions worldwide conducted by Sheppard (2019), it has been observed that the condition of marine environments and habitats has been steadily deteriorating. Although trace amounts of elements occur

naturally in marine environments, human activities such as urban development, recreational activities, and the presence of harbours near coastal waters have contributed to elevated concentrations of elements in marine waters (Greenfield *et al.*, 2014). The coastal zones, which occupy more than 70% of the global coastline, are experiencing increased anthropogenic pressures (Sink *et al.*, 2012). These activities, essential for sustaining the ocean economy, can directly contribute to metal pollution along the coastline of South Africa, necessitating monitoring efforts to protect the environment (Erasmus *et al.*, 2022a). Recreational boating activities have various environmental impacts such as operational hazards, petroleum products, and pollutants associated with boat maintenance (Fields, 2003). Coastal areas are susceptible to contamination from various substances used in boat maintenance, including gasoline oil, diesel fuel, and battery acids. During boat operation, especially with two-stroke engines (mainly used in Sodwana Bay), gasoline spills, sometimes mixed with oil, can occur. Additionally, petroleum-based substances such as kerosene and diesel fuel can leak from boats while in the water (Fields, 2003).

According to Song *et al.* (2014), both natural weathering of surrounding geological formations and urban development contribute to the release of elements and contaminants into the coastal environment. Metals and metalloids (collectively referred to as elements), which are persistent and stable pollutants, can accumulate in coastal waters (Fatoki and Mathabatha, 2001). These environmental pollutants, beneficial or toxic, can be absorbed by organisms through water, sediment, and nutrition, potentially leading to adverse effects on the ecosystem. Elements can be classified into two categories: essential and non-essential. Essential elements are depleted by various energy-consuming metabolic activities and are important for optimal physiological functioning in organisms (Madgett *et al.*, 2021). However, non-essential elements have been present in marine environments for an extended period, undergoing multiple biogeochemical cycles. Non-essential elements have no biological function within organisms. Unlike certain organic substances, most elements are difficult to transform into less hazardous compounds (Morel and Price, 2003), thus posing a threat to the health of marine organisms. Due to the dense coastal population along the eastern coast of South Africa, effluents containing nutrients and pollutants enter the water through numerous estuaries, posing a risk to fish populations and other components of the marine-ecosystem (Wepener, 2007; Orr *et al.*, 2008; Whitfield and Cowley, 2010). High levels of elements in tissues of fish can pose a human health risk, possibly leading to carcinogenic or non-carcinogenic effects.

Individuals who live along the coast depend on fish as a protein source and rural communities that regularly eat contaminated fish are exposed to potentially harmful contaminants over an extended period. Overexposure to various trace elements can increase both non-carcinogenic (such as liver, thyroid, liver, and kidney damage and dysfunction) and carcinogenic (such as bone, thyroid, liver, and kidney cancer) health risks (Pheiffer *et al.*, 2018; Volschenk *et al.*, 2019; Balali-Mood *et al.*, 2021). These fish are typically contaminated with a combination of contaminants rather than just one, which may increase toxicity (Erasmus *et al.*, 2022b).

The aims of the present study were to determine the element concentrations in muscle and liver tissues of *H. affinis* from Sodwana Bay sampled during two surveys, as well as to determine the human health risks associated with consumption of contaminated *H. affinis*. The assumption was that there would be differences in element concentrations in *H. affinis* between the pre-COVID (2018) and post-COVID (2022) periods. This hypothesis was tested by analysing the element concentrations in *H. affinis* muscle and liver tissue collected during 2018 and 2022, as well as assessing the possible non-carcinogenic and carcinogenic human health risks associated with consumption of contaminated *H. affinis*.

2.2 Materials and Methods

2.2.1 Site description

Sodwana Bay, located on the northeast coast of South Africa, is part of the Maputoland marine protected area (MPA) and was proclaimed a World Heritage site in 2000 (Dicken, 2014). It is part of the iSimangaliso Wetland Park (Tunley, 2009), which was created to ensure the protection of the unique coastal region.

2.2.2 Collection of samples

Before sampling, permits were obtained from the Department of Agriculture, Forestry, and Fisheries (DAFF) and South African National Parks (SANParks), along with ethical approval from AnimCare at North-West University (NWU-01425-20-A9). *Hyporhamphus affinis* sampling was carried out during two separate surveys in Sodwana Bay (Figure 2.1). Samples were collected in August/September of 2018 and in September of 2022 using rod and reel. Live *H. affinis* was transported to a field laboratory and kept in an oxygenated tank until dissection could be performed. The fish were identified, and the total body weight and length were measured and photographed. Fish were stunned by blunt force percussion to the head and then euthanised by cervical transection followed by pithing. Tissue samples (± 200 mg of muscle without skin and liver) were obtained from each individual fish and placed in 15 mL pre-cleaned acid-washed polypropylene Falcon tubes and frozen at -20°C until further analyses. Muscle tissue represents the longer-term bioaccumulation, whereas liver tissue represents the shorter-term bioaccumulation due to metal exposure.

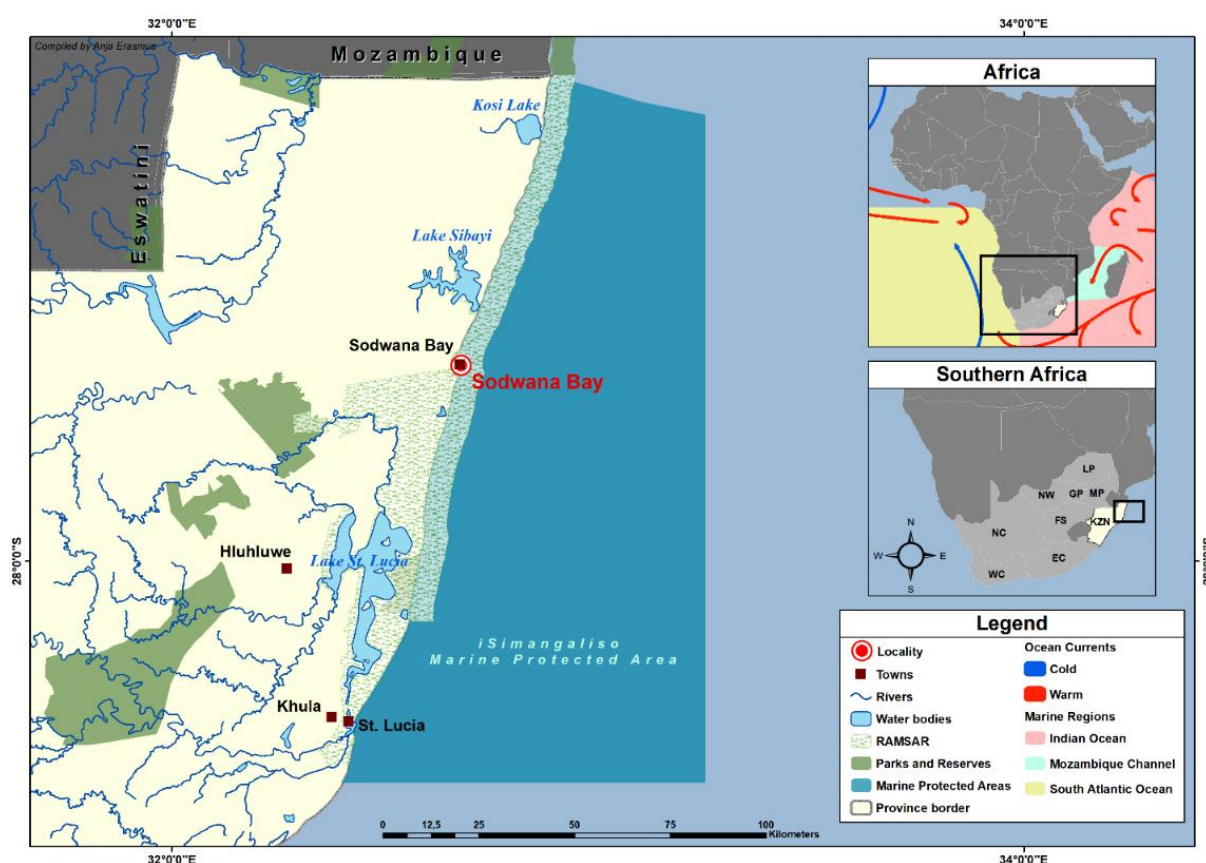


Figure 2.1: Map of the study area, indicating the sampling site (Sodwana Bay).

2.2.3 Sample preparation

Samples were thawed and the wet weight was recorded. Approximately 200 mg tissue was placed in a freeze dryer (FreeZone® 6, Labconco) for 96 h. Thereafter samples were reweighed to determine the moisture content (Erasmus *et al.*, 2020). Dried homogenised samples (± 170 mg muscle) (± 140 mg liver) were microwave digested. Acid digestions were carried out according to methods from Erasmus *et al.* (2022a) using an Ethos MAXI-44 Advanced Microwave Digestion System with Teflon vessels (Magna Analytical), ± 0.2 g dried sample, 7.5 mL 65% suprapur nitric acid (HNO_3) (Sigma-Aldrich Corporation), and 2.5 mL hydrochloric acid (HCl) (Rochelle Chemicals & Lab equipment), digested at 180°C for 30 min (Erasmus *et al.*, 2020). Samples were transferred to 50 mL volumetric flask and reconstituted to volume with 1 % HNO_3 made up in MilliQ water and subsequently transferred to pre-cleaned polypropylene containers for storage at room temperature until trace element analysis.

2.2.4 Trace element analysis

Multi-element analysis of arsenic (As), cadmium (Cd), copper (Cu), iron (Fe), manganese (Mn), nickel (Ni), lead (Pb), antimony (Sb), tin (Sn), and zinc (Zn) was done on tissue samples, using inductively coupled plasma mass spectroscopy (ICP-MS) (PerkinElmer, Elan 6000). Mercury (Hg) concentrations in tissue were determined using a flow injection mercury system (FIMS) and Chrome (Cr) concentrations in tissue were determined using a graphite furnace atomic absorption spectrometer (GF-AAS) (PerkinElmer, AAnalyst 600) equipped with Zeeman-effect background correction. The instrument operational settings, calibration methods, sample preparation and quality were as described in Erasmus *et al.* (2020; 2022c). Analytical efficiency was ensured by using sample blanks consisting of the 5 % HNO₃ dilutant. Certified reference materials for fish tissue (DORM-4) were prepared in triplicate in the same manner as the halfbeak tissues.

2.2.5 Human health risk assessment

2.2.5.1 Non-carcinogenic

For each analysed trace element, the average daily dosage (ADD) expressed in mg/kg body weight per day was determined to assess the risk of chronic non-cancer health effects from oral exposure.

ADD =

$$\frac{(\text{average trace element concentration in fish muscle (WW)} \times (\text{weight of portion}))}{(\text{adult body mass}) \times (\text{no. of days between fish meals})}$$

where the fish consumed weighed 150 g, the average adult weight was 60 kg, and there were three days between fish meals (Heath *et al.*, 2004). The moisture content of *H. affinis* in 2018 and 2022 was 72% and 64%, respectively.

Reference doses (RfDs) are used in risk assessments to determine the toxic effects of pollutants that are non-carcinogenic and to set thresholds at which detrimental health consequences might be anticipated. For each trace element, the danger to human health was evaluated using a hazard quotient (HQ).

$$HQ = \frac{ADD}{RfD}$$

Where HQ > 1 suggests a high probability of adverse health effects and HQ < 1 present a low probability of adverse health effects. The RfD levels (µg/kg/day) that were used in this study were as follow: As (0.3); Cd (1); Cr (3); Cu (40); Fe (700); Hg (0.1); Mn (140); Ni (20); Sb (0.4); Sn (600); Zn (300) (USEPA, 2000) and Pb (3.5) (Djedjibegovic *et al.*, 2020).

2.2.5.2 Carcinogenic

According to the USEPA (2005), the carcinogenic trace elements As, Cd, Cr, and Ni have a cancer risk (CR). The following formula was used to determine the estimated daily intake (EDI) of trace elements from eating fish:

$$EDI = \frac{(C \times DR)}{BM}$$

where BM is the average body weight of the consumer set at 60 kg, DR is the daily consumption of fish (g/day), and C is the average concentration of the trace element (mg/kg wet weight).

The CR was calculated as follows:

$$CR = EDI \times CSF$$

where the cancer slope factor of the trace elements (in mg/kg/day) is the CSF, which was as follows: As (1.5); Cd (0.001); Cr (0.5); Ni (0.84) (USEPA, 2000; IRIS, 2017) and Pb (0.00357) (Bacigalupo and Hale, 2012). A carcinogenic risk level of 10⁻⁶ is used as the public screening threshold for carcinogens. Therefore, if the CR is above 10⁻⁴ it is considered unacceptable, if the CR is between 10⁻⁶ and 10⁻⁴ a level of concern and a CR value below 10⁻⁶ is an acceptable risk (USEPA, 2005).

2.2.5.3 Maximum safe consumption limit

The maximum daily intake of fish that can be ingested without causing any harm to human health was determined to be:

$$Y = \frac{RfD \text{ or } CSF \times BM}{C}$$

where Y is the number of fish that can be eaten without causing harm, RfD is the reference dose for non-carcinogenic risk, CSF is the cancer slope factor for carcinogenic risk, BM is

the average consumer body weight, and C is the average trace element content in fish tissue.

2.2.6 Statistical analysis

Univariate statistical analyses were performed using GraphPad Prims v9. The statistical significances between elements in tissue off halfbeaks collected during the two sampling periods were determined at $p < 0.05$. Element concentration data were tested using the D'Agostino and Pearson omnibus normality test, as well as the Shapiro-Wilk normality test to examine the normality and homogeneity of the variance. When the data were parametric, an unpaired t-test was performed. In the case of non-parametric data, a Man-Whitney U test was performed.

2.3 Results

2.3.1 Element bioaccumulation in tissues of *Hyporhamphus affinis*

The recovery rate, detection limit, and quantification limit were calculated for the different elements of interest obtained from the certified reference materials (CRM) (DORM-4) and blanks, respectively (Table 2.1). Weights and lengths were recorded for each *H. affinis* sampled during the respective time periods (2018 and 2022) (Table 2.2). The element analysis revealed significant differences ($p < 0.05$) for several elements accumulated in tissue and between the two sampling periods (2018 and 2022) (Figures 2.2 and 2.3). There was a significant ($p < 0.05$) increase in concentrations of Cr (Figure 2.2C), Mn (Figure 2.3A), Pb (Figure 2.3C), Sb (Figure 2.3D) and Sn (Figure 2.3E) in muscle tissues between 2018 and 2022. There was a significant ($p < 0.05$) increase in concentrations of Pb (Figure 2.3C), Sb (Figure 2.3D) and Sn (Figure 2.3E) in liver tissue between 2018 and 2022. On the contrary, there was a significant decrease ($p < 0.05$) in As (Figure 2.2A), Cu (Figure 2.2D), Fe (Figure 2.2E) and Hg (Figure 2.2F) during 2022 compared to 2018 in muscle tissue. Lastly, there was a significant decrease ($p < 0.05$) in As (Figure 2.2A), Cd (Figure 2.2B), Cr (Figure 2.2C), Fe (Figure 2.2E), Hg (Figure 2.2F), Ni (Figure 2.3B) and Zn (Figure 2.3F) in liver tissues between 2018 and 2022.

Table 2.1: The recovery rate (%), the detection limit (mg/kg) and the quantification limit (mg/kg) of the elements of interest obtained from the certified reference material (DORM-4) and blanks.

CRM (DORM-4)		Blanks	
<i>Element</i>	<i>Recovery rate (RR) (%)</i>	<i>Limit of detection (LoD) (mg/kg)</i>	<i>Limit of quantification (LoQ) (mg/kg)</i>
As	83.9	5.21E-07	1.56E-06
Cd	99.1	5.95E-07	1.78E-06
Cr	103.9	1.16E-05	3.48E-05
Cu	87.9	n.d.	n.d.
Fe	110.7	4.85E-04	1.45E-03
Hg	103.1	5.97E-06	1.79E-05
Mn	115.8	3.48E-05	1.04E-04
Ni	111.5	1.36E-05	4.08E-05
Pb	117.5	2.75E-05	8.26E-05
Sb	n.c.	n.d.	n.d.
Sn	95.4	4.34E-06	1.30E-05
Zn	83.6	1.00E-04	3.01E-04

*n.c. - Not certified

*n.d. - Not detected

Table 2.2: Length and weight of *Hyporhamphus affinis* sampled from Sodwana Bay (2018 and 2022).

<i>Year</i>	<i>Total individuals (n)</i>	<i>Total length (TL in mm)</i>	<i>Total weight (g)</i>
2018	18	296.7 ± 20.8	78.2 ± 15.2
2022	15	307.7 ± 28.3	87.3 ± 27.1

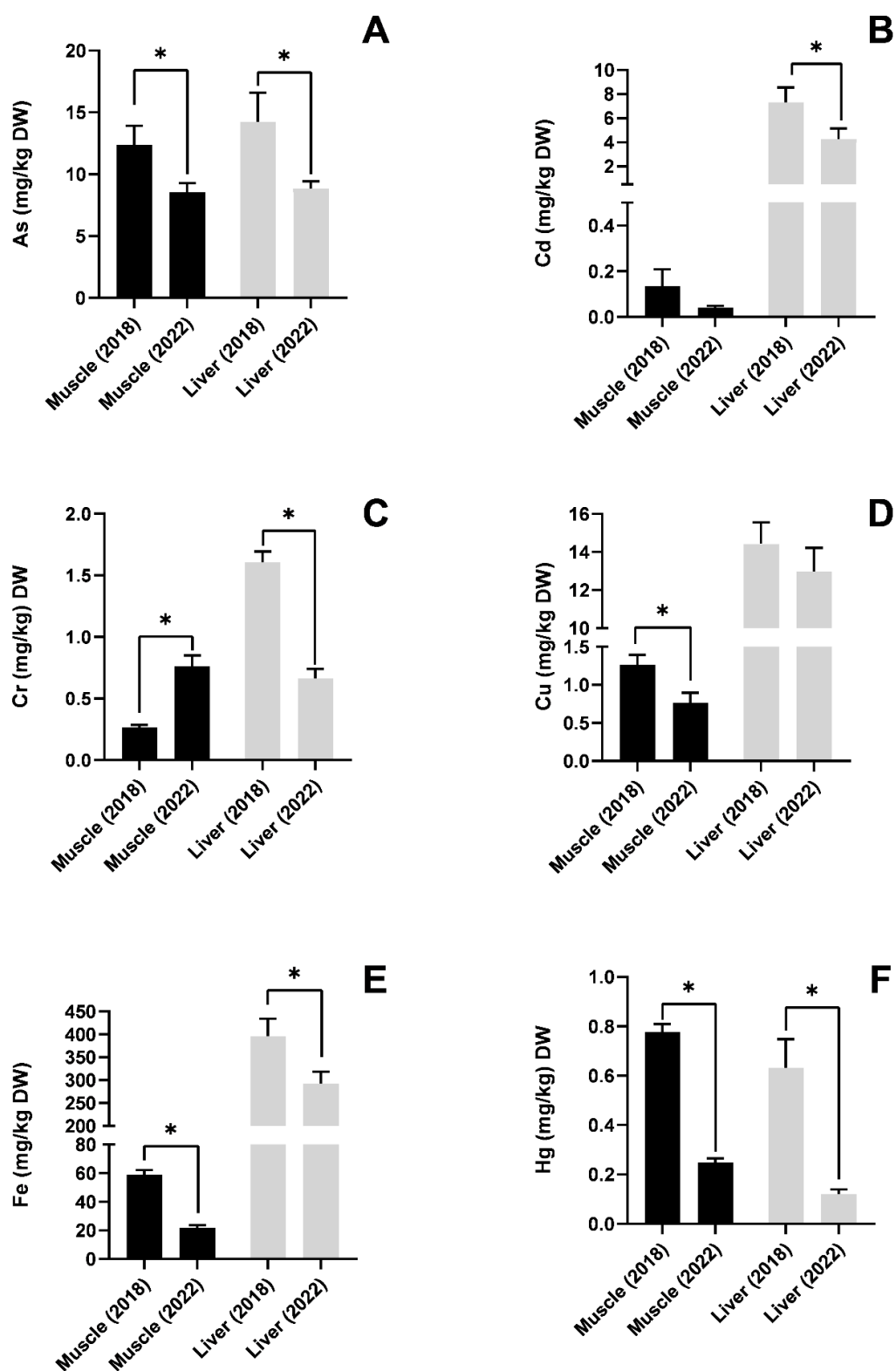


Figure 2.2: Mean $\pm$ standard deviation of concentrations of As (A), Cd (B), Cr (C), Cu (D), Fe (E), and Hg (F) concentrations (mg/kg dry weight) in muscle and liver tissue of *Hyporhamphus affinis* collected during two different surveys (2018 and 2022) in Sodwana

Bay, South Africa. Significant differences ($p<0.05$) between tissue surveys are indicated with asterisk superscripts.

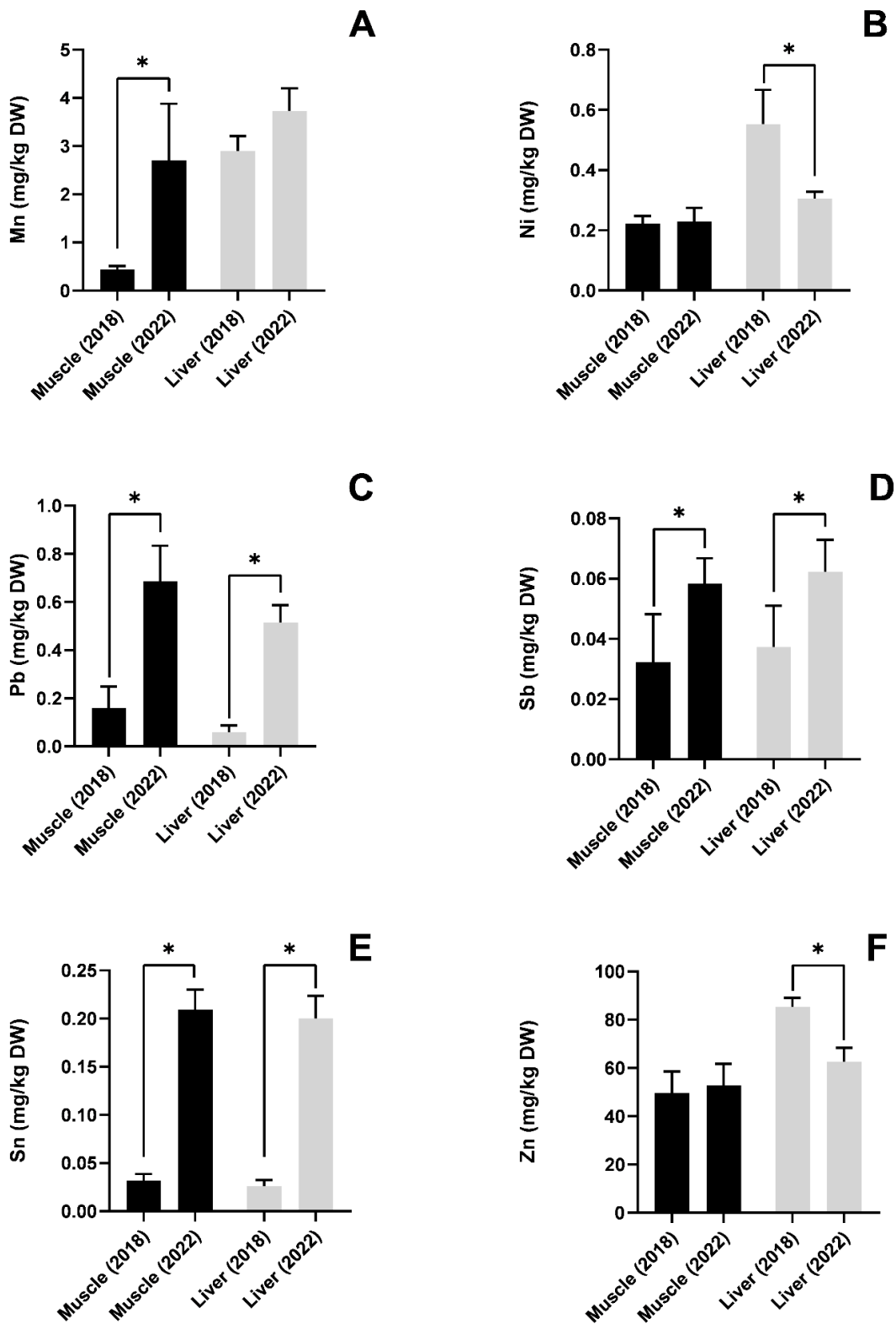


Figure 2.3: Mean $\pm$ standard deviation of concentrations of Mn (A), Ni (B), Pb (C), Sb (D), Sn (E), and Zn (F) concentrations (mg/kg dry weight) in muscle and liver tissue of

Hyporhamphus affinis collected during two different surveys (2018 and 2022) in Sodwana Bay, South Africa. Significant differences ($p < 0.05$) between surveys for the tissues are indicated with asterisk superscripts.

2.3.2 Human health risk assessment

The hazard quotient and cancer risk were calculated from the average daily dose and the estimated daily intake. Furthermore, reference doses and cancer slope factor were used, respectively and this assessed the non-carcinogenic and carcinogenic human health risks associated with consumption of contaminated fish (Table 2.3). Over the two time periods the hazard quotients (HQ) were < 1 for most investigated elements (see Appendix A) except for As that had a HQ value of 4.8 and 7.6 for 2018 and 2022 respectively, furthermore Hg had a value that was > 1 for 2018. Therefore, As and Hg poses a high risk of non-carcinogenic, adverse human health effects when contaminated fish are frequently consumed. Furthermore, an unacceptable cancer risk with CR values $> 10^{-4}$ was found during the two time periods for As, Cr, Ni, and Pb. In general, the non-carcinogenic risk is higher in 2022 compared to 2018 when comparing As values. The non-carcinogenic risk for Hg was higher in 2018 compared to 2022. The carcinogenic risk for As and Pb was higher in 2018 compared to 2022, while the carcinogenic risks for Cr and Ni were higher in 2022 compared to 2018.

Table 2.3: The average concentration of trace elements in muscle tissue was used to calculate the mean and standard deviation of hazard quotients (HQs), cancer risk (CR), and the maximum safe daily consumption of *Hyporhamphus affinis* from 2018 (clear column) and 2022 (shaded column), assuming a person of 60 kg consumes a fish meal (150 g) twice a week. Only values of HQ > 1, indicating a high chance of adverse health effects, as well as CR > 10⁻⁴, indicating an unacceptable risk to people who consume these fish, are included in the table. Hazard quotients and CR values for the majority of the trace elements investigated were significantly below zero.

Trace elements	<i>H. affinis</i>	
	2018	2022
Hazard quotients for non-carcinogenic risk (HQ)		
As	4.8 ± 9.5	7.6 ± 2.5
Hg	1.8 ± 0.4	0.7 ± 0.2
Cancer risk (CR) (× 10 ⁻⁴)		
As	126 ± 64	103 ± 34
Cr	0.9 ± 0.4	3.8 ± 3.4
Ni	0.4 ± 0.3	1.5 ± 1.2
Pb	1.2 ± 0.9	0.02 ± 0.01
Maximum safe consumption limit per day (g)		
As (CR)	30 ± 10	36 ± 10
As (NC)	6.1 ± 2	7.2 ± 2.1
Cr (CR)	471 ± 143	130 ± 48
Pb (CR)	19 ± 17	1.9 ± 1.9
Hg (NC)	30 ± 8.5	79 ± 19

Reference dose (RfD) (µg/kg/day): As (0.3); Cd (1); Cr (3); Cu (40); Ni (20); Zn (300) (USEPA, 2000); Pb (3.5) (Djedjibegovic *et al.*, 2020). Cancer slope factors (mg/kg/day): As (1.5); Cd (0.001); Cr (0.5); Ni (0.84) (USEPA, 2000; IRIS, 2017); Pb (0.00357) (Bacigalupo and Hale, 2012).

2.4 Discussion

In South Africa, COVID lockdown restrictions between early 2020 and the end of 2021 resulted in the cessation of coastal recreational activities. This contrasts with the high degree of activity at Sodwana Bay in the year preceding the COVID pandemic. It was clear from the bioaccumulation results that notable trends in element concentrations between these periods in tissues were evident. There were significant decreases in As, Cd, Cu, Fe, Hg, Ni, and Zn in the tissues. Conversely, there was a notable increase in Cr, Mn, Pb, Sb, and Sn concentrations in the tropical halfbeak tissues from the 2022 survey. The element concentrations exhibited higher levels for most elements in liver tissue compared to muscle tissue, except for Hg and Pb. The human health risk assessment indicated non-carcinogenic risks for As and Hg, while carcinogenic risks were associated with As, Cr, Ni, and Pb.

In the present study, it was found that the element concentrations in liver tissue were mostly higher or relatively similar when compared to muscle tissue, except for Hg and Pb. A study conducted by Bashir and Alhemkali (2015) in the Kapar coastal waters of Malaysia had similar findings in two marine fish species, *Arius maculatus* (spotted sea catfish) and *Pennahia anea* (bigeye croaker) with Cu, Zn and Cd concentrations being significantly higher in liver tissue compared to muscle tissue. Furthermore, a study by Ramos-Miras *et al.* (2019), conducted in the western Mediterranean Sea, found higher concentrations of As, Cr and Ni in liver tissue compared to muscle tissue in four species of marine fish, including *Mullus surmuletus* (striped red mullet), *Merluccius merluccius* (European hake), *Auxis rochei* (bullet tuna) and *Scomber japonicus* (Pacific mackerel), which supports the findings in the present study. The Pb concentrations were found to be higher in muscle tissue compared to liver tissue of *A. rochei* (Ramos-Miras *et al.*, 2019), which was also evident in the present study with *H. affinis*. This disparity can be attributed to the low affinity of Pb for binding to metallothionein, a compound that exhibits limited binding capabilities with Pb (Ploetz *et al.*, 2007). Furthermore, a study by Mzimela *et al.* (2003) in the Mhlathuze Estuary in South Africa found elevated concentrations of Fe and Mn in *Liza dumerelli* (thinlip grey mullet) liver tissue compared to muscle tissue, supporting the findings of the present study. Higher concentrations in liver tissue are predominantly attributed to the role of the liver as a primary organ responsible for the detoxification of metabolic processes of various elements. The liver has enzymatic mechanisms that convert toxic forms of elements to less harmful products (Sevcikova *et al.*, 2011). During this process, elements may accumulate in liver tissue. The liver is highly vascularised,

allowing this organ to receive a substantial portion of the bloodstream, including blood that contains different elements (Rajeshkumar *et al.*, 2013). This elevated blood perfusion facilitates the accumulation of elements in liver compared to less vascularised tissues, such as muscle. Furthermore, liver tissues have a greater ability to sequester and store elements within various compounds such as metallothionein and glutathione (Fernandes *et al.*, 2007). A study by Hajeb *et al.* (2009) in Malaysia observed higher concentrations of Hg in muscle tissue compared to liver tissue in the marine fishes *Rastrelliger branchsoma* (short mackerel) and *Alepes vari* (herring scad), consistent with the findings of the current study. Methylmercury (MeHg), a lipophilic and toxic form of Hg, tends to accumulate more in fish muscle tissue due to its higher lipid content relative to liver tissue. In addition to MeHg's affinity for lipids, it can also interact with proteins in the fish body (Charette *et al.*, 2021). Muscle and liver tissue exhibit distinct protein compositions, which can influence the distribution of MeHg within these tissues (Golovanova, 2008). The disparities observed between the two sampling surveys can be attributed to a combination of natural processes and localised inputs specific to Sodwana Bay.

The decrease in As, Cu, Ni and Zn concentrations observed in 2022 compared to 2018 can primarily be attributed to the reduced rates of boat activity and the decrease in localised input. According to the findings of Verma and Dwivedi (2013), Ni can be associated with batteries and combustion fuels, while Cu is mainly associated with batteries. Although the Ni content in fuel is typically low, the cumulative effect of numerous boats operating in the same area can lead to a minor release of Ni into the water. Zinc coating, often referred to as galvanising, is a contemporary technique used to protect steel compartments from corrosion in marine environments (Fields, 2003). By serving as a sacrificial layer that corrodes in place of the underlying steel, it protects the metal from direct contact with seawater. Consequently, the decrease in Zn concentrations in liver tissue may plausibly be related to a decrease in recreational activities during the period of reduced activity. The observed decline in Cd concentrations in 2022 compared to 2018 may be due to the reduced levels of agricultural runoff from the Mbazwana plantation (± 8 km northwest of Sodwana Bay) and Manzengwenya plantation (± 33 km northwest of Sodwana Bay). The decrease element input can be attributed to the drying out of Lake Sibhayi, resulting from drought conditions, as plantations require significant amounts of water for their production. Elevated levels of Hg were found in sediments from a study conducted by van Rooyen *et al.* (2023) in the Phongolo River Floodplain (PRF), where there is no gold mining or industrial activities present in the catchments. Sodwana Bay is

situated on the same atmospheric pathway as the PRF, making aerial Hg depositions imminent. Therefore, elevated levels of Hg in tropical halfbeak tissues are linked to high levels of aerial depositions originating in the Mpumalanga highveld during 2018 when economic activity was not restricted. Conversely, the concentrations of Cr, Mn, Pb, Sb and Sn in the tropical halfbeak tissues from the post-lockdown (2022) were elevated. Vetrimurugan *et al.* (2018) reported elevated levels of Cr in beach sediments at Sodwana Bay, which they attributed to the enrichment caused by the presence of Quaternary beach placer deposits containing ilmenite, rutile, and zircon. It is possible that the increased levels of Cr in muscle tissue of *H. affinis* are due to elevated levels of coastal erosion and wind-driven erosion, which could have led to higher levels of beach sediments entering the marine environment. Sturve *et al.* (2021) found no indication of elevated levels of elements at Maputo Bay. The Agulhas Current mainly affects sediment distribution and deposition in ocean areas further from the coast (Hahn *et al.*, 2018). Tropical halfbeaks are shoreline fish; therefore, the Agulhas Current is unlikely to be a major contributor to the observed element concentrations in their tissues. Elevated concentrations of Mn, Pb, Sb, and Sn are more likely linked to upwelling events that occur along the eastern coast of South Africa each year (Hanekom *et al.*, 2009). Upwelling regimes are well-established in pristine marine areas such as Sodwana Bay, and they are often associated with elevated levels of elements (Lares *et al.*, 2002). Wepener and Degger (2020) have shown that upwelling regimes can lead to element accumulation in tissues. There are seasonal changes associated with Pb concentrations (Wepener and Degger, 2012), which may explain the lower levels of Pb observed. According to Kumar *et al.* (2021), there is limited information available on Sb and Sn concentrations in marine ecosystems, making them distinctive elements of investigation.

In the present study, it was evident that lockdown restrictions caused a significant decrease in As, Cu, Ni and Zn concentrations in the tissues of *H. affinis*. In South Africa, a significant percentage of rivers reach the ocean, since South Africa has an extensive coastline along the Atlantic and Indian Oceans. The observed decrease in the concentration of certain elements in *H. affinis* is likely due to the reduced recreational and economic activity during the imposed lockdown. The tropical halfbeak, being a shoreline fish, serves as an ideal indicator of elemental level variations, particularly in proximity to estuaries. Typically, industries discharge significant quantities of untreated or inadequately treated wastewater into the environment, predominantly directed into rivers and streams (Ashan *et al.*, 2019; Kubra *et al.*, 2022). However, the implementation of COVID-19

lockdown measures led to a decrease in industrial activities, resulting in a reduction in the release of industrial effluents into surface water bodies. Consequently, this can contribute to improved surface water quality (Aman *et al.*, 2020; Arif *et al.*, 2020; Dutta *et al.*, 2020; Yunus *et al.*, 2020; Khan *et al.*, 2021; Karunanidhi *et al.*, 2021; Rendana *et al.*, 2022). A study conducted by Siddique *et al.* (2023) observed a decline in water pollution in Bangladesh during the lockdown period compared to the pre-lockdown period. This improvement is attributed to the complete cessation of activities in the industrial, recreational and business sectors, preventing the direct discharge of waste into rivers. Chakraborty *et al.* (2021) found that the lockdown of industrial and recreational activities in India led to a decrease in element concentrations in river water and lower mean concentration levels of water pollution during the lockdown period. However, certain elements remained at elevated levels, possibly due to natural processes such as upwelling, erosion, wind-driven erosion, etc. These elevated levels could pose a human health risk to regular consumers of *H. affinis*.

Due to its high abundance and easy accessibility, *H. affinis* can be consumed by local fisherman and rural communities residing in the Sodwana Bay area. The Sodwana Bay shore lacks data on real fish consumption, hence an estimate of the risk to human health has been established using adults weighing 60 kg. The human health risk assessment did not consider those who would be more sensitive to chronic exposure to trace elements, such as children, pregnant women, and mothers who breastfeed and their infants (Javed and Usmani, 2019). The concentrations of As and Hg in *H. affinis* muscle tissue were unsafe for human consumption in 2018 and 2022. Long-term exposure to As in food and drinking water can cause cancer and skin irritation. In adults, Hg poisoning can damage the visual cortex and kill neurons in the cerebellar granule layer (Krishna *et al.*, 2014). When comparing pre-lockdown and post-lockdown human health risk results, the non-carcinogenic risks for As slightly decreased over the four year period. Mercury levels in *H. affinis* decreased to safe levels for human consumption post-lockdown, compared to pre-lockdown (Table 2.3). However, the safe daily consumption limits calculated to avoid non-carcinogenic effects are too low to support people who rely on fish as a cheap and convenient source of protein. The cancer risk assessment for trace elements in *H. affinis* showed that the risk of As exposure was higher post-lockdown than pre-lockdown, similar to findings from Siddique *et al.* (2023) via water ingestion in Bangladesh. Chromium levels were at a level of concern pre-lockdown and posed a human health risk to consumers post-lockdown. According to Budiawan and Auerkari (2017), when Cr is present in

sufficiently high amounts in the environment, it can have toxic, and genotoxic effects on humans. The Ni concentrations were at a level of concern in pre-lockdown and posed a higher risk in post-lockdown. When used as a direct ingredient in human food, Ni compounds are generally considered safe because they are believed to be a normal component of the diet. The carcinogenic risks associated with overexposure to Pb increased over four years between the pre- and post-lockdown periods. According to the World Health Organisation (2023), Pb is believed to have permanent impacts on the brain and behaviour. Limited data is available on the human health risks associated with carcinogenic risks when consuming contaminated fish tissue during the COVID-pandemic. However, a study by Siddique *et al.* (2023) assessed carcinogenic risks associated with water consumption during the COVID-pandemic. The findings indicated that Cr and Pb posed a higher risk in the pre-lockdown period when compared to the post-lockdown period when ingesting water from rivers, they attributed this to industrial influences and urban pollution. The reduction in pollution levels occurred due to the decrease in industrial activity during the COVID-19 lockdown (Siddique *et al.*, 2023).

2.5 Conclusions

The decline in tissue concentrations of tropical halfbeaks from Sodwana Bay was primarily ascribed to the reduction in recreational activities resulting from the COVID-19 lockdown measures. The elevated concentrations observed were related to known natural processes along the east coast of South Africa, such as periodic upwelling events and wind-driven erosion. Notably, increased Hg levels were associated with aerial depositions from coal-fired power stations in the Mpumalanga Highveld. The assessment of human health risks revealed both non-carcinogenic and carcinogenic risks at Sodwana Bay. The maximum safe daily consumption limit suggested that relying on tropical halfbeaks as the primary protein source would be an unreliable source, as the maximum consumption limits were insufficient. Future studies can investigate the physiological aspects of tropical halfbeaks, such as size, sex, and length. These physiological factors can be used to examine the potential positive or negative relationships with recorded element concentrations at Sodwana Bay. It is also crucial that future studies consider not only adults when evaluating human health risks associated with element overexposure, but also vulnerable groups such as pregnant women and infants. The present study can serve as a foundational base for subsequent research focusing on human health risks over time in Sodwana Bay.

Lastly, future studies can assess the interactions between environmental factors and element concentrations in tropical halfbeaks in Sodwana Bay. Focusing on the influence of habitat characteristics and water quality on the observed element concentrations, thus providing a more comprehensive understanding of the ecosystem dynamics.

Chapter 3: Element bioaccumulation in the Cape white seabream (*Diplodus capensis*) along the South African coastline

3.1 Introduction

Metals and metalloids (collectively referred to as elements), represent stable and persistent environmental pollutants within coastal waters (Fatoki and Mathabatha, 2001). The marine environment is influenced by the presence of minerals and elements, which can have beneficial and toxic characteristics. Organisms can assimilate these elements through various pathways, including water, sediment, and dietary sources. Although elements naturally occur in trace concentrations in marine ecosystems, the proximity of coastal waters to anthropogenic activities such as harbours, urban developments, and recreational areas makes them particularly susceptible to elevated element concentrations (Greenfield *et al.*, 2014). Recent meta-analyses of coastal regions in multiple geographical areas and countries have indicated a steady decline in the quality of shallow marine environments and habitats throughout the world (Wepener and Degger, 2019). This concerning trend can be attributed to the increasing prevalence of industrial activities and the widespread adoption of consumerist lifestyles (Verma and Dwivedi, 2013). The presence of elements in the environment raises concerns, particularly along the South African coastline, where more than 70% of the coastal zone is developed and inhabited, thus intensifying anthropogenic pressures on the nearshore marine environment (Taljaard *et al.*, 2006).

The South African coastline, characterised by its dynamic nature, is influenced by two prominent ocean currents, each contributing to distinct local nearshore circulation patterns. The Benguela Current, which transports cold and nutrient-rich waters, flows north along the western coast, while the Agulhas Current transports warm waters southward along the eastern coast (Van As *et al.*, 2012). The comprehension of various contaminants, originating from the surrounding landscape and transported by ocean currents, which impact coastal biodiversity and ecosystem health in South Africa, remains limited (Erasmus *et al.*, 2021). Approximately 30% of South Africa's population resides near coastal areas, leading to a widespread trend of urbanisation and industrialisation within coastal communities (Taljaard *et al.*, 2006). An emerging concern revolves around the contamination of South Africa's coastal environment with elements, a direct consequence of activities that support the maritime economy. Consequently, rigorous monitoring

becomes imperative to ensure the health of both human populations and ecological equilibrium (Erasmus *et al.*, 2022b). Although a fraction of the element presence in South African coastal marine environments is of natural origin (Gregory *et al.*, 2002), the predominant percentile arises from anthropogenic activities within river catchments that ultimately discharge into the ocean (Mzimela *et al.*, 2003; Wepener and Vermeulen, 2005; Vogt *et al.*, 2019). As a result, contaminants can become bioavailable in the marine environment and accumulate within the tissues of marine organisms.

The bioaccumulation and toxicity of elements are intricately linked to environmental factors that influence the speciation and bioavailability of elements. These factors include temperature, salinity, and pH, along with biological variables such as size, sex, age, feeding habits, and life cycles (Erasmus *et al.*, 2021). Upon element uptake, the liver plays a pivotal role as the primary detoxification and elimination organ, while muscle tissues become predominant for longer-term element bioaccumulation due to sequestration mechanisms (Erasmus *et al.*, 2021). Fish tissue exhibits a propensity to accumulate quantities of elements present in the aquatic environment, rendering them vital indicators of ecosystem contamination (Moodley *et al.*, 2021). Elevated levels of elements (essential and non-essential) have the potential to pose a threat to ecological functioning (Moodley *et al.*, 2021). Element concentrations in regularly consumed fish species are still being researched, particularly in Africa, but such research is necessary to determine true toxicity and eventual consumer impacts (Bosch *et al.*, 2016). Although fish muscle is not primary tissue associated with element bioaccumulation, they must be examined due to the health risks they can pose to humans who consume this portion (Moodley *et al.*, 2021).

Despite the various health benefits of fish to human consumers, elements can pose a serious threat to consumers' health (Bosch *et al.*, 2016). Furthermore, fish is a staple in the human diet; consuming contaminated fish poses a public health risk (Moodley *et al.*, 2021). Due to the persistence of elements, inherent toxicity, subsequent bioaccumulation, and biomagnification in the food chain, higher element concentrations pose a health hazard to human health and the environment (Rainbow and Luoma, 2011; Li *et al.*, 2013; Cobbina *et al.*, 2015; Nan *et al.*, 2016). The lack of reported data on elements, particularly mercury, in South African marine fish is a major concern for consumer health and industry economics. The lack of available data leaves the fishing industry without clear directives to avoid contaminated fish and areas, thus posing potential health risks to human consumers (Bosch *et al.*, 2016). Research on element contamination in marine fish around the African

continent is limited, and further research is needed to ensure that regularly consumed fish, which is widely considered a nutritious food source, does not pose unexpected health risks to consumers. To better understand the potential effects of element contamination on the South African coastline, bioindicator species can be used to gain more insight.

Bioindicator species serve as repositories of various contaminants within their tissues, offering valuable means to assess the accessibility and presence of pollutants within ecosystems (Rainbow and Phillips, 1993; de Silva *et al.*, 2009). Predatory fish, which are at the top of the food chain, are particularly vulnerable to element contamination reflecting both biotic and abiotic conditions in their surroundings (Dragun *et al.*, 2009). Certain fish species such as *Diplodus capensis* (Cape white seabream) can exhibit element concentrations due to their feeding habits and the various habitats they inhabit (Authman *et al.*, 2015). *Diplodus capensis* is widely distributed along the southern African coastline and thrives in a variety of habitats, including rocky and sandy substrates (Coetzee, 1986). In southern Africa, *D. capensis* represents a prevalent inshore species of substantial interest to recreational and subsistence fishermen (Smith and Heemstra, 1995). This species of marine fish mainly prefers turbulent waters and rocky shorelines for its habitat (Patrick and Strydom, 2009). Additionally, instances of *D. capensis* have been documented within the surf zone adjacent to sandy beaches and estuarine environments (Whitfield, 1985; Lasiak, 1986; Heemstra and Heemstra, 2004). The average length of a mature cape white seabream measures approximately 45 cm (Heemstra and Heemstra, 2004). *Diplodus capensis* exhibits a wide geographical range, boasting high population densities, attaining weights of up to 3 kg.

The aim of this chapter was to assess the element concentrations in muscle and liver (tissues) of *D. capensis* collected from both relatively non-impacted and anthropogenically impacted sites along the South African coastline during 2020 and 2022. Furthermore, the second aim of this chapter was to evaluate the potential human health risks associated with eating *D. capensis* muscle tissue. The assumption was that element accumulation in *D. capensis* will reflect element concentrations associated with land-use activities in the region they were collected in and that specimens from contaminated areas will pose a higher non-carcinogenic and carcinogenic health risk to human consumers. The assumption was tested by collecting specimens of *D. capensis* from relatively non-impacted and anthropogenically impacted sites along the South African coastline. Furthermore, performing analytical assessments of element concentrations within *D.*

capensis tissue samples, elucidating the bioavailability of elements within the marine ecosystem. Lastly, the non-carcinogenic and carcinogenic health risks associated with consumption of contaminated fish were calculated.

3.2 Materials and Methods

All materials and methods for sample collection, sample preparation, element analysis, and human health risk calculations used in this chapter are provided in full in Chapter 2.

3.2.1 Description of the study area and sampling sites

Sampling was carried out between 2020 and 2022 at five localities along the coastline of South Africa. The sites were categorised into two groups, either the site was anthropogenically impacted or relatively non-impacted. The impacted sites included Witsand (2021) and Mossel Bay (2022), whereas the relatively non-impacted sites included De Hoop (2021), Tsitsikamma (2020, 2021 and 2022) and Chintsa (2020 and 2022). Chintsa is a coastal town in the Eastern Cape Province and is an example of a coastal environment with minimal point source pollution and proximity to essential estuarine systems, including the Great Kei River (Erasmus *et al.*, 2021). The convergence of the cold, nutrient rich Benguela current and the warmer Agulhas current at Chintsa along with Tsitsikamma creates various environmental conditions and habitats (Vermaak *et al.*, 2023). Tsitsikamma which is a section of the Garden Route National Park, is located on the southeastern coast and is a UNESCO-designated marine protected area that encompasses an 80 km stretch of coastline. This park showcases a variety of habitats, including rocky shores and intertidal zones, and plays a key role in preserving the ecological balance of the Storms River Mouth and adjacent marine ecosystems. De Hoop, located along South Africa's southern shore within the Western Cape Province, is situated at the confluence of freshwater and marine environments. This unique position gives rise to complex ecological interactions and shows the interaction of various ecosystems. Mossel Bay is a harbour town also situated in the Western Cape Province and serves as both an important tourism hub and agricultural region. According to Gubili *et al.* (2009), it is a strategic location as the gateway to the Garden Route National Park, which spans terrestrial and marine environments, underscores its significance in regional biodiversity conservation. Witsand is situated on the southern coastline in the Western Cape Province. Although not directly within De Hoop, the nearby De Hoop Marine Protected Area significantly influences the coastal ecology of the region.

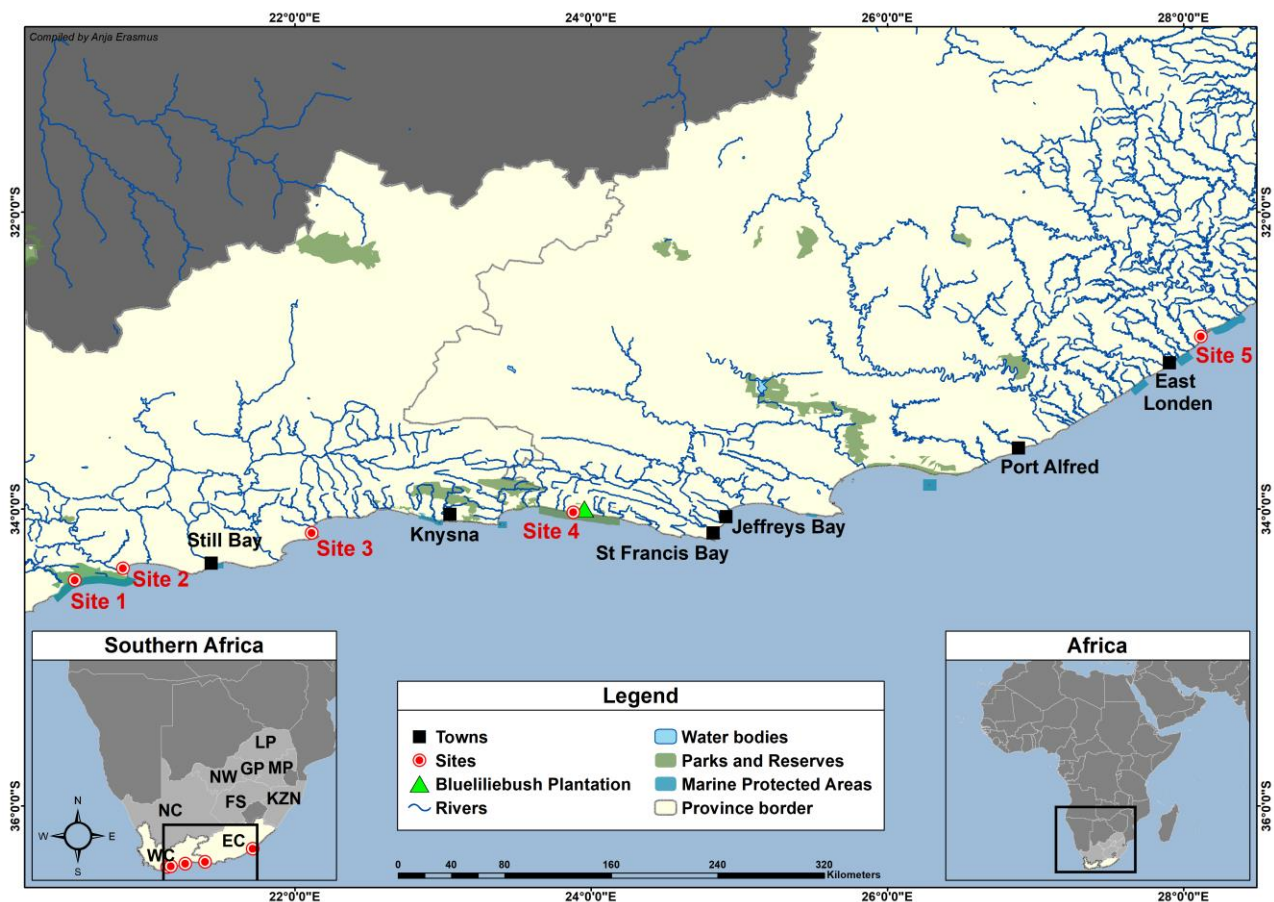


Figure 3.1: Map of the sites sampled along the South African coastline.

3.2.2 Statistical data analyses

Univariate analysis of variance (two-way ANOVA) was performed to determine the statistical significance ($p < 0.05$) of the elements in the tissue between sites and different time periods. Element concentration data were tested using the D'Agostino and Pearson omnibus normality test as well as the Shapiro-Wilk normality test was used to examine the normality and homogeneity of variance. If data had a normal distribution, parametric tests were used. If data was not normally distributed, non-parametric tests were used.

3.3 Results

3.3.1 Element bioaccumulation in tissues of *Diplodus capensis*

The recovery rate, detection limit and quantification limit were calculated for the different elements of interest obtained from certified reference material (CRM) (DOLT-5) and blanks, respectively (Table 3.1). All recoveries were within 20% of the certified values and therefore no corrections were applied. Furthermore, weight and length measurements were recorded for each specimen of *D. capensis* collected from locations along the South African coast and different time periods (Table 3.2). All fish were generally of a similar size with the smallest fish collected from Witsand (2021).

Table 3.1: The recovery rate (%), the detection limit (mg/kg) and the quantification limit (mg/kg) of the elements of interest obtained from the certified reference material (DOLT-5) and blanks.

CRM (DOLT-5)		Blanks	
<i>Element</i>	<i>Recovery rate (RR) (%)</i>	<i>Limit of detection (LoD) (mg/kg)</i>	<i>Limit of quantification (LoQ) (mg/kg)</i>
As	93	5.21E-07	1.56E-06
Cd	87	5.95E-07	1.78E-06
Cr	104	1.16E-05	3.48E-05
Cu	109	n.d.	n.d.
Fe	102	4.85E-04	1.45E-03
Hg	103	5.97E-06	1.79E-05
Mn	106	3.48E-05	1.04E-04
Ni	109	1.36E-05	4.08E-05
Pb	118	2.75E-05	8.26E-05
Sb	97	n.d.	n.d.
Sn	94	4.34E-06	1.30E-05
Zn	93	1.00E-04	3.01E-04

*n.d. - Not detected

Element analysis revealed significant differences ($p < 0.05$) in the accumulation of several elements in tissues between sites and surveys (Figures 3.2-3.5). For muscle tissue significant ($p < 0.05$) differences were only recorded in concentrations of As (Figure 3.2A) and Cr (Figure 3.2C) between the different sites. The As and Cr levels were the highest at Chintsa in 2022. There were more significant ($p < 0.05$) differences for liver concentrations with As (Figure 3.2A), Cr (Figure 3.2C), Mn (Figure 3.3A), Ni (Figure 3.3B), Pb (Figure 3.3C), Sn (Figure 3.3E), and Zn (Figure 3.3F) that differed in *D. capensis* between sites. The Cr and Ni levels in liver tissue were the highest at Chintsa in 2022, whereas As, Mn and Sn concentrations in liver tissue was the highest at Witsand during 2021. The highest concentrations of Pb and Zn in liver tissue were recorded at Mossel Bay (2022) and De Hoop (2021), respectively. Furthermore, significant ($p < 0.05$) differences were observed between the different time periods. Significant ($p < 0.05$) differences of Cd (Figure 3.4B), Cr (Figure 3.4C) and Sn (Figure 3.5E) concentrations were observed within muscle tissue between different time periods. Cadmium and Sn levels in muscle tissue were the highest in Tsitsikamma during 2020, while Cr in muscle tissue was recorded in muscle tissue in Chintsa during 2022. Lastly, significant ($p < 0.05$) differences were observed in the concentrations of As (Figure 3.4A), Cd (Figure 3.4B), Cr (Figure 3.4C), Cu (Figure 3.4D), Fe (Figure 3.4E), Mn (Figure 3.5A), Ni (Figure 3.5B), and Zn (Figure 3.5F) within *D. capensis* liver tissue between respective time periods. The As, Cu, Fe, and Zn levels in liver tissue were the highest in Tsitsikamma fish during 2021. The Cd levels in liver tissue were the highest in Tsitsikamma fish during 2020, whereas, at Chintsa during 2022 the highest levels in liver tissue were recorded for Cr, Mn and Ni.

Table 3.2: Length and weight of *Diplodus capensis* sampled along the South African coastline.

<i>Site</i>	<i>Total specimens (n)</i>	<i>Total length (TL in mm)</i>	<i>Total weight (g)</i>
De Hoop (2021)	12	254 ± 64	384 ± 154
Chintsa (2020)	3	231 ± 53	68 ± 13
Chintsa (2022)	10	209 ± 69	203 ± 226
Mossel Bay (2022)	5	209 ± 42	182 ± 70
Tsitsikamma (2020)	3	336 ± 11	753 ± 179
Tsitsikamma (2021)	12	287 ± 42	432 ± 183
Tsitsikamma (2022)	10	286 ± 44	448 ± 196
Witsand (2021)	3	186 ± 41	136 ± 61

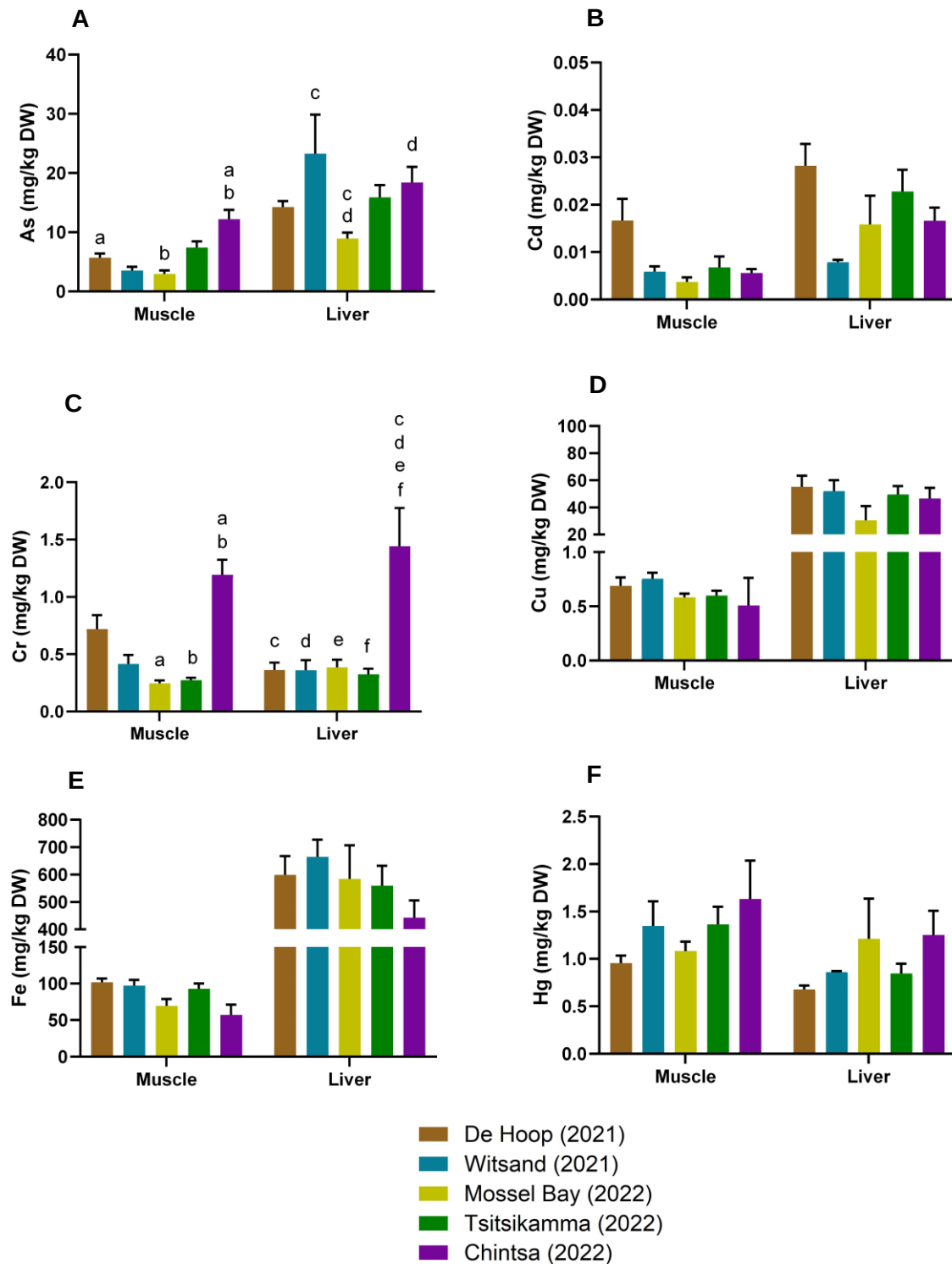


Figure 3.2: Mean $\pm$ standard deviation of the concentrations of As (A), Cd (B), Cr (C), Cu (D), Fe (E) and Hg (F) concentrations (mg/kg dry weight) in muscle and liver tissue of *Diplodus capensis* collected at different sites along the South African coast. Significant differences ($p < 0.05$) between tissue samples from the different sites are indicated with common symbolic superscripts.

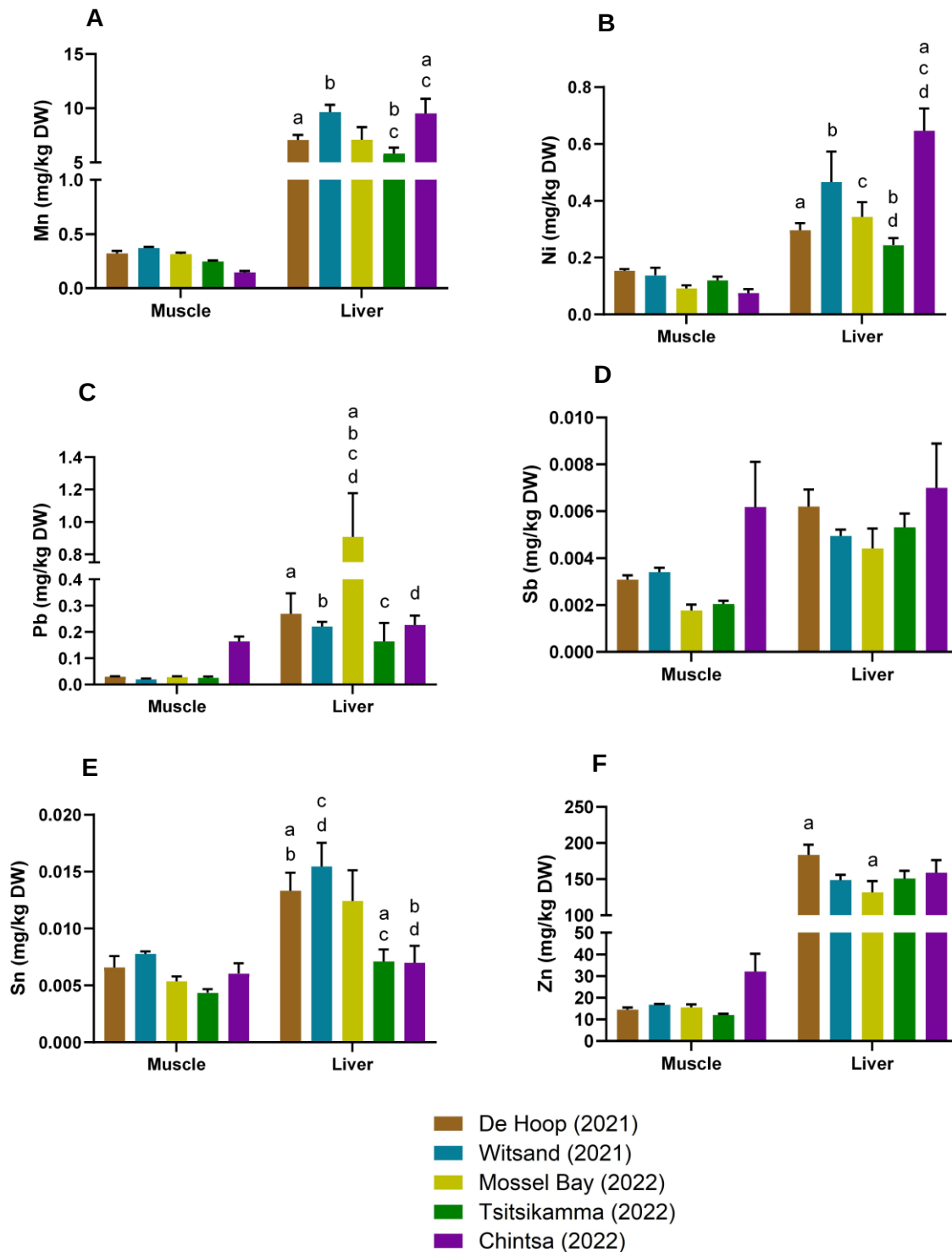


Figure 3.3: Mean $\pm$ standard deviation of the concentrations of Mn (A), Ni (B), Pb (C), Sb (D), Sn (E) and Zn (F) concentrations (mg/kg dry weight) in muscle and liver tissue of *Diplodus capensis* collected at different sites along the South African coast. Significant differences ($p < 0.05$) between tissue samples from the different sites are indicated with common symbolic superscripts.

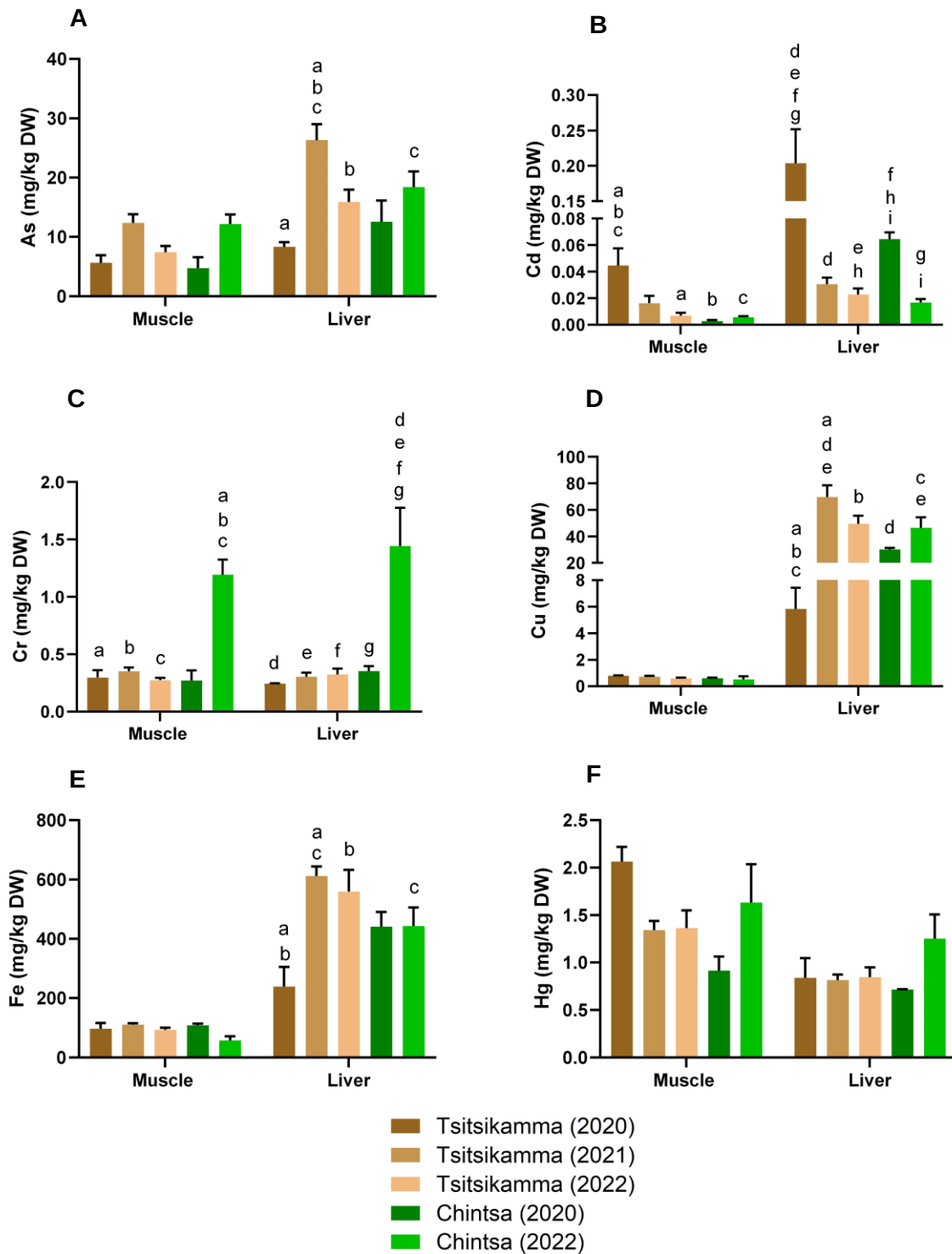


Figure 3.4: Mean $\pm$ standard deviation of the concentrations of As (A), Cd (B), Cr (C), Cu (D), Fe (E), and Hg (F) concentrations (mg/kg dry weight) in muscle and liver tissue of *Diplodus capensis* collected at different sites and periods along the South African coast. Significant differences ($p < 0.05$) between tissue samples from the different sites are indicated with common symbolic superscripts.

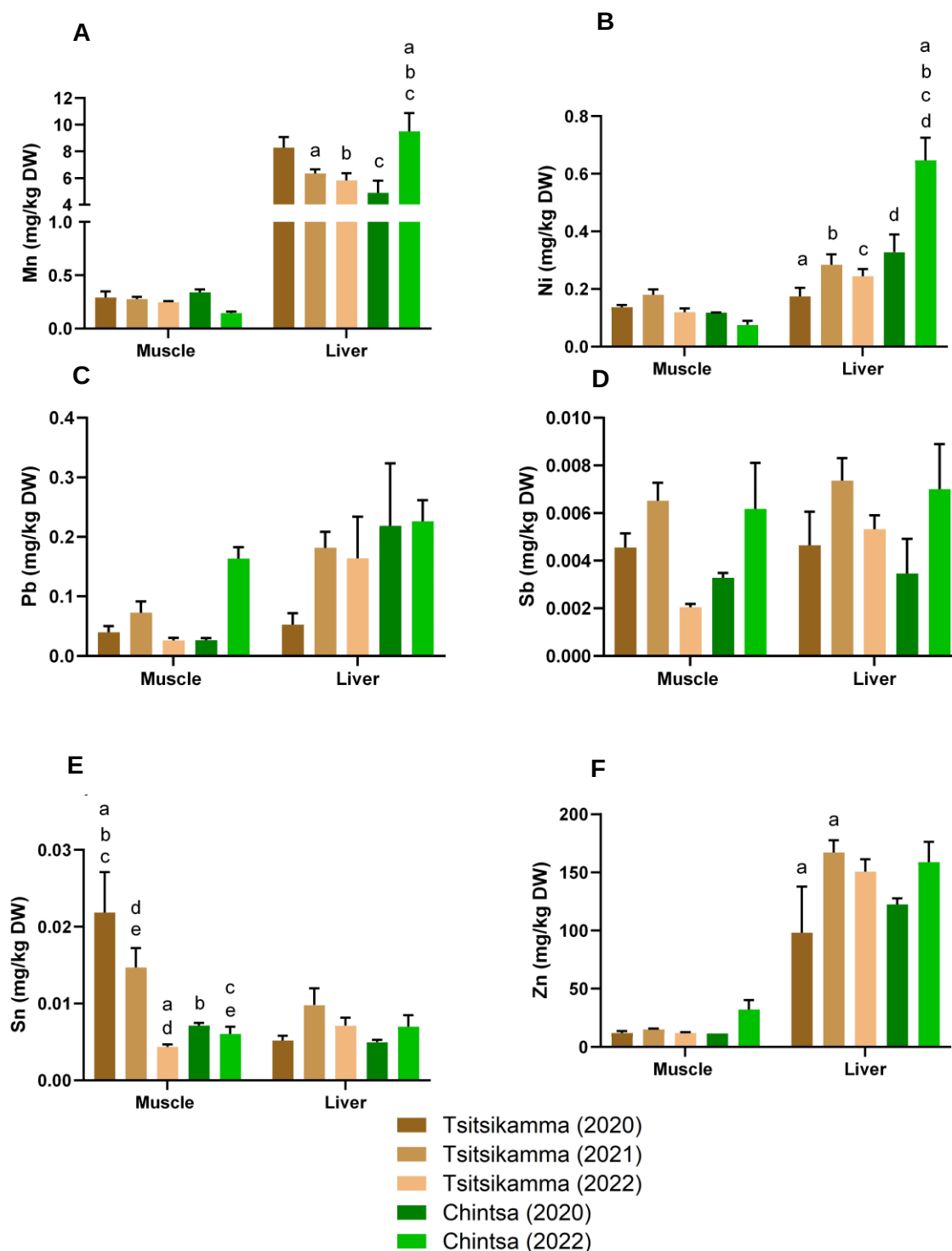


Figure 3.5: Mean $\pm$ standard deviation of the concentrations of Mn (A), Ni (B), Pb (C), Sb (D), Sn (E), and Zn (F) concentrations (mg/kg dry weight) in muscle and liver tissue of *Diplodus capensis* collected at different sites and periods along the South African coast. Significant differences ($p < 0.05$) between tissue samples from the different sites are indicated with common symbolic superscripts.

3.3.2 Human health risk assessment

The hazard quotient (HQ) and cancer risk (CR) were calculated using the average daily dose and estimated daily intake. Reference doses and cancer slope factors were used to evaluate the non-carcinogenic and carcinogenic human health risks linked to the consumption of contaminated fish (Table 3.3). When comparing the HQ between the different sites, most elements were < 1 (see Appendix B), except for As and Hg which were > 1 for all the respective sites. Elevated cancer risks, as indicated by CR values exceeding 10^{-4} , were identified at all respective sites for As. Furthermore, Cr posed an unacceptable non-carcinogenic risk in fish from De Hoop (2022), Witsand (2021) and Chintsa (2022). The CR for Ni exceeded the unacceptable threshold of 10^{-4} at De Hoop (2021) and Witsand (2021). In the case of Cr, the levels were between 10^{-4} and 10^{-6} , which is at a level of concern for Tsitsikamma (2021), Mossel Bay (2022), and Tsitsikamma (2022). Furthermore, Ni levels were at a level of concern at Tsitsikamma (2021), Chintsa (2022), Mossel Bay (2022) and Tsitsikamma (2022).

Table 3.3: The average concentration of trace elements in muscle tissue was used to calculate the mean and standard deviation of hazard quotients (HQs), cancer risk (CR), and the maximum safe daily consumption of *Diplodus capensis* from the sites along the South African coastline, assuming a person of 60 kg consumes a fish meal (150 g) twice a week. Only values of HQ > 1, indicating a high chance of adverse health effects, as well as CR > 10⁻⁴, indicating an unacceptable risk to people who consume these fish, are included in the table. Hazard quotients and CR values for the majority of the investigated trace elements were below zero.

Trace elements	<i>Diplodus capensis</i>					
	De Hoop (2021)	Tsitsikamma (2021)	Witsand (2021)	Chintsa (2022)	Mossel Bay (2022)	Tsitsikamma (2022)
Hazard quotients for non-carcinogenic risk (HQ)						
As	5 ± 2.2	7.5 ± 3.1	3.4 ± 1	7.2 ± 3	1.9 ± 0.9	5.6 ± 2.5
Hg	2.5 ± 0.8	2.5 ± 0.6	3.9 ± 1.3	2.9 ± 2.3	3.3 ± 2.7	3.1 ± 1.3
Cancer risk (CR) (× 10 ⁻⁴)						
As	67 ± 30	101 ± 42	45 ± 14	97 ± 41	25 ± 11	75 ± 34
Cr	6.3 ± 13	0.93 ± 0.31	1.8 ± 0.6	7.5 ± 7.6	0.71 ± 0.17	0.93 ± 0.22
Ni	1.1 ± 0.2	0.83 ± 0.31	1.1 ± 0.33	0.33 ± 0.27	0.45 ± 0.13	0.68 ± 0.23
Maximum safe consumption limit per day (g)						
As (CR)	59 ± 25	36 ± 10	78 ± 23	41 ± 18	161 ± 86	55 ± 26
As (NC)	11 ± 5.1	7.3 ± 2.1	15 ± 4.6	8.3 ± 3.7	32 ± 17	11 ± 5.3
Cr (CR)	160 ± 81	424 ± 120	224 ± 71	101 ± 65	553 ± 146	420 ± 95

Hg (NC)	21 $\pm$ 7.3	21 $\pm$ 5.3	13 $\pm$ 3.9	26 $\pm$ 14	20 $\pm$ 9.6	19 $\pm$ 10
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Reference dose (RfD) ($\mu\text{g/kg/day}$): As (0.3); Cd (1); Cr (3); Cu (40); Ni (20); Zn (300) (USEPA, 2000); Pb (3.5) (Djedjibegovic *et al.*, 2020). Cancer slope factors (mg/kg/day): As (1.5); Cd (0.001); Cr (0.5); Ni (0.84) (USEPA, 2000; IRIS, 2017); Pb (0.00357) (Bacigalupo and Hale, 2012).

3.4 Discussion

In the present study, the different sites along the South African coastline had different sources of observed variations in element concentrations within the tissues of Cape white seabream. There were significant differences in element concentrations within tissues readily discerned at various sites along the South African coastline, as well as between time intervals at Chintsa and Tsitsikamma. It was observed that, for most elements, higher concentrations were found in liver tissue compared to muscle tissue, with the exception of Hg. Most of the element concentrations in muscle tissue were significantly higher at Chintsa (2022) compared to other sites, while other sites exhibited relatively comparable element concentrations. The level of element concentrations within liver tissues at Chintsa (2022) and Witsand (2021) consistently showed significantly higher levels for most elements, except Pb, which had a higher concentration at Mossel Bay (2022). In addition, De Hoop (2021) and Witsand (2021) fish had significantly higher levels of Sn compared to fish from the other study sites. When comparing the different time intervals at Chintsa and Tsitsikamma, for most element concentrations in muscle tissue, the highest levels were recorded at Tsitsikamma during 2021 and Chintsa during 2022. Only Cd and Sn concentrations were higher at Tsitsikamma during 2020 for muscle tissue. Majority of element concentrations in liver tissue indicated higher levels at Tsitsikamma during 2021 and Chintsa during 2022 with only Cd levels being higher at Tsitsikamma during 2020 and Chintsa during 2020.

Elevated levels of As, Cd, Cr, Cu, Fe, Mn, Ni, Pb, Sb, Sn and Zn were observed in liver tissue compared to muscle tissue in Cape white seabream. Mercury exhibited higher concentrations in muscle tissue than in liver tissue. The selective accumulation of essential elements such as Fe, Cu, Mn and Zn is specific to certain organs. The findings of a study by Jia *et al.* (2017) demonstrated higher levels of As, Cd, Cu, Fe, Mn, Pb and Zn in liver tissue compared to muscle tissue in three species of fish (*Carassius auratus*, *Pelteobagrus fulvidraco*, and *Squaliobarbus curriculus*), consistent with the results of the present study. A study by Licata *et al.* (2005) from the Straits of Messina, Italy on *Thunnus thynnus* (Atlantic bluefin tuna) found Cd, Cu, Mn and Pb levels higher in liver tissue compared to muscle tissue, furthermore Gutiérrez-Ravelo *et al.* (2020) found Fe and Ni levels higher in liver tissue compared to muscle tissue in *Sarpa salpa* (salema porgy) from the North-Eastern Atlantic Ocean. This pattern is attributed to the presence of metallothionein protein, which is capable of binding certain elements such as Cd and Cu for detoxification (Ploetz *et al.*, 2007). The significance of Fe as a vital element for various organisms is

known by its role in the transport of oxygen through the blood. The liver, which is a highly vascularised organ, tends to accumulate substantial amounts of essential elements, such as Cu, Fe, Mn and Zn. The liver consistently accumulates elevated concentrations of elements, regardless of the uptake route. Consequently, liver tissue emerges as a reliable indicator of water pollution with elements, as the concentrations stored in this organ often reflect those present in the surrounding environment (Jezierska and Witeska, 2006). In a study conducted by Afandi *et al.* (2018) along the North-West African coast (South Atlantic Moroccan coast), higher concentrations of Hg were observed in muscle tissue compared to *Trachurus trachurus* (Atlantic horse mackerel) liver tissue during the summer season, similar to the findings of the present study. According to Peng *et al.* (2016), muscle tissue serves as a significant reservoir for Hg bioaccumulation. Methylmercury, a form of toxic Hg, exhibits a strong affinity to bind with proteins rather than fat, and since fish muscle tissue is protein-rich (Mason *et al.*, 1995), it became a major site for Hg bioaccumulation. The amino acid cysteine, characterised by a sulphur group, is particularly attractive to MeHg. Methylmercury forms stable complexes with thiols, such as those present in cysteine-containing proteins (Harris and Pickering, 2003; Lemes and Wang, 2009; Huang *et al.*, 2013). Since muscle tissue constitutes a substantial portion of fish and contains abundant proteins, it provides a favourable environment for the binding and retention of MeHg. Unlike certain organs, muscle tissue tends to be less metabolically active (Peng *et al.*, 2016). This implies that once MeHg is deposited in muscle tissue, it can remain relatively stable over time compared to organs with higher metabolic activity. Differences in element concentrations observed between different sites and time periods along the South African coastline can be attributed to a combination of several natural processes, such as upwelling events (Wepener and Degger, 2020) as well as localised input through land-based activities and releases through streams and stormwater into the nearshore marine environment (Erasmus *et al.*, 2021).

The concentration of As, Cr and Ni in tissues was the highest at Chintsa (2022) compared to the other sites. Chintsa is a small coastal town situated between the Gonubie and Kei Marine Protected Areas within the Amathole Marine Protected Area, where there are minimal to no identifiable point sources of pollution (Erasmus *et al.*, 2021). Chintsa is positioned south of the Great Kei River and is surrounded by numerous estuaries (Erasmus *et al.*, 2021). The cumulative higher levels of Cr and Ni concentrations observed at Chintsa (2022) compared to the rest of the sites including De Hoop (2021), Witsand (2021), Tsitsikamma (2022) and Mossel Bay (2022) can be attributed to increased

mobilisation, potentially resulting from illegal sand mining operations in the Eastern Cape (Chevallier, 2014). As established by Hanekom *et al.* (2009), the eastern coast of South Africa encounters an average of around eight upwelling events annually along its shoreline. According to Erasmus *et al.* (2020), high levels of As and Cr can be related to element-rich phytoplankton originating from these frequent upwelling events. The concentrations of As, Mn and Sn in liver tissue were the highest at Witsand. A study done by Van der Spuy *et al.* (2023) conducted at Witsand, found much lower levels of As and Mn in liver tissue of evileye puffer fish (*Amblyrhynchotes honckenii*) compared to the present study. It is important to note that the process of element uptake and accumulation in organisms is intricately influenced by a range of physiological and biochemical factors (Mubiana *et al.* 2006). Any alterations in these factors can contribute to variations between individuals. Such influential factors include body size, sex, reproductive status, and nutritional status. The unexpectedly high concentrations observed in Witsand (2022) fish raised concerns, since the Breede River Estuary at Witsand is traditionally recognised as a pristine and uncontaminated site (Breede River Estuarine Management Plan, 2016). A study by Erasmus *et al.* (2021) also identified elevated element levels in klipfish, *Clinus superciliosus*, from unimpacted sites located along the same coastline. These findings were attributed to prolonged exposure to elements originating from natural geogenic weathering, introduced by freshwater runoff and frequent upwelling events. Supporting studies for this phenomenon can be found in the levels of elements detected in the Brown mussel, *Perna perna*, at various sites along the South African south coast (Degger *et al.*, 2011; Wepener and Degger, 2020). These studies collectively underline the influence of natural processes, such as geogenic weathering and upwelling regimes, on element concentrations in marine organisms, even in areas traditionally considered uncontaminated. The high levels of Pb in *D. capensis* liver tissue at Mossel Bay (2022) can be due to the increasing rate with which this location is used by service crafts associated with the burgeoning oil industry operating off the Mossel Bay coastline (Ports and Ships, 2019). It is known that maritime traffic and shipping operations in and around harbours can lead to the release of Pb-containing paints and fuels into the water, contributing to Pb pollution. A study by Gardner *et al.* (1983) had previously raised concern about potential pollutant impacts stemming from the discharge of raw sewage into the harbour area and factory effluents originating from the Voorbaai region. According to Gardner *et al.* (1983), it is important to note that two additional rivers, namely the Little Brak and Hartenbos Rivers, discharge into the Mossel Bay area. These rivers host human

settlements that can contribute to the influx of pollutants within the estuary. In a study conducted by Wepener and Degger (2020), the concentration of Pb in the marine bivalve *P. perna* was reported at a level of 0.60 mg/kg DW. In the present study, we observed a higher concentration, with *D. capensis* recorded at 0.91 mg/kg DW. These findings suggest that Pb pollution in Mossel Bay is still present today. The study by Van der Spuy *et al.* (2023) in Mossel Bay found that Pb levels in *A. honckenii* within liver tissue were 0.25 mg/kg DW, indicating a relatively lower concentration compared to the present study. The concentration of Zn in *D. capensis* liver tissue was the highest at De Hoop (2021). Zinc is an essential element that plays a distinct role in fish metabolism. However, the presence of significantly elevated levels of Zn can pose risks and be hazardous to fish. According to Swales *et al.* (1998), the accumulation of Zn in liver tissue is attributed to the regulatory and excretory functions of the organ.

The decrease in activities at coastal areas during the COVID-19 pandemic lock-down periods can provide an explanation for the differences observed in element concentrations of tissues during the sampling periods. The concentrations of Cr, Mn and Ni were significantly higher in fish collected at Chintsa during 2022, compared to 2020. A study by Selvam *et al.* (2020) in South India found a decrease in trace elements in groundwater from coastal industrial cities as a result of lockdown restrictions. The decline in trace elements in the surface water system can be attributed to the non-operation of both small- and large-scale industries (Karunanidhi *et al.*, 2021). The three elevated element concentrations at Chintsa during 2022 are known to be associated with anthropogenic input, however as mentioned, Chintsa is known to have very limited to no known point sources of pollution. The increased concentrations of Cr, Mn and Ni observed at Chintsa in tissues during 2022 when compared over the different time periods can be due to the east coast of South Africa being subjected to occasional deep-water upwelling occurrences (Diaz-Pulido and Garzón-Ferreira, 2002). The highest concentrations of Cd and Sn in tissues were recorded at Tsitsikamma during 2020. According to Bosch *et al.* (2016), Cd can enter the environment through natural processes such as rock abrasion. Although Cd is a prevalent contaminant in marine fish tissues, its accumulation is not uniform. As a result, there is a need for extensive investigations on various fish species to determine the actual risks that Cd can present to fish (Heath, 1987). Naturally, due to industrial activities, Cd accumulates in soils and easily pollutes surface water and groundwater through runoff and infiltrations after precipitation (Karunanidhi *et al.*, 2021). The concentrations of As, Cu, Fe, Mn, and Zn in liver tissue at Tsitsikamma during 2021 exhibited the highest

concentrations. Elevated concentrations recorded in Tsitsikamma fish during 2021 can be attributed to the prevalence of high elemental levels resulting from geogenic weathering and broader oceanographic processes, rather than stemming from diffuse or point sources (Degger *et al.*, 2011; Wepener and Degger, 2020; Erasmus *et al.*, 2021). Consequently, it can be inferred that, depending on the intensity of these naturally occurring element concentrations, fish could have been exposed to these environmental conditions for an extended period, allowing them to develop a tolerance to these elements. In contrast, the increased impulse of anthropogenic contaminations in the area is relatively recent and thus fish have not yet had the time to develop a tolerance to these elements (Erasmus *et al.*, 2020; Erasmus *et al.*, 2022a). High element concentrations in muscle tissue of Cape white seabream can pose risks to individuals who regularly consume them.

In Africa, fish protein contributes substantially to the overall supply of animal protein, accounting for an average of 19.4% (Bosch *et al.*, 2016). Due to its high abundance and easy accessibility, *D. capensis* serves as a source of nutrition for local fisherman and rural communities that live along the South African coastline. It is essential to note that there is a lack of accurate data on actual fish consumption data along the South African coastline. Consequently, the potential health risk calculations assumed that an average adult weighing 60 kg consumed a 150 g portion of fish twice a week. The bioaccumulation results in muscle tissue have been used to assess the potential risks posed to individuals who regularly consume contaminated *D. capensis* muscle tissue. According to the human health risk assessment (USEPA, 2000; USEPA, 2005), the concentrations of As and Hg in *D. capensis* were found to exceed safe levels for human consumption (Table 3.3). Only concentrations of As and Hg exceeded acceptable thresholds for non-carcinogenic risks ($HQ > 1$). It is important to note that *D. capensis* can contain a mixture of various trace elements, which could potentially even further amplify the risks for individuals who consume *D. capensis*. Early symptoms of As overexposure in humans include vomiting, abdominal pain, muscle weakness, diarrhoea, and skin flushing (Bosch *et al.*, 2016). According to Guynup and Safina (2012), even low-level exposure to Hg can adversely affect the cardiovascular system, although chronic exposure to Hg can affect the pituitary gland and the liver, ultimately compromising the immune system. According to Bosch *et al.* (2016), organic As is primarily prevalent in fish and meat, but it does not necessarily accumulate in the body due to its rapid excretion. A study by Nyarko *et al.* (2023) in the coastal waters of Ghana revealed non-carcinogenic risks in the sparid, *Dentex angolensis* for Hg, and similarly, non-carcinogenic risks were observed for As in the prawn, *Penaeus*

notialis, reflecting the results of the present study. The study calculated the maximum safe daily consumption limits necessary to prevent non-carcinogenic health risks associated with As and Hg contamination in *D. capensis* (Table 3.3). These limits were neither realistic nor sufficient to sustain individuals who rely on Cape white seabream as an affordable and easily accessible protein source to avoid non-carcinogenic risks. Assessment of the carcinogenic risks associated with trace elements in *D. capensis* from sites along the South African coastline revealed that As posed a risk at all sites surveyed, while Cr was at a level of concern at all the sites. Arsenic is known to be associated with several cancers, including lung, bladder, and skin cancer (Erasmus *et al.*, 2022b). A study by Moodley *et al.* (2021) at three localities namely Isipingo Beach, Cuttings Beach and Amanzimtoti Beach from South Durban in South Africa found CR values for As $>1.8 \times 10^{-3}$. These values are similar to the present study, as the lowest CR value for As was 25×10^{-4} at Mossel Bay. The CR for Cr ranged between 10^{-6} and 10^{-4} , which signifies a level of concern for regular consumers. Ingestion of Cr can lead to the development of kidney, liver and stomach cancers (Sun *et al.*, 2015). A study by Atique Ullah *et al.* (2019) in Dhaka Bangladesh, on two species of marine fish *Tenualosa ilisha* (hilsa herring) and *Dorosoma cepedianum* (gizzard shad) found CR values for Cr at 1.53×10^{-4} , similar to the results of the present study. As for Ni, the CR was $>10^{-4}$ at both De Hoop (2021) and Witsand (2021). At Tsitsikamma (2021), Chintsa (2022), Mossel Bay (2022), and Tsitsikamma during 2022, the CR for Ni ranged between 10^{-6} and 10^{-4} , indicating a level of concern for regular fish consumers. According to a study conducted by Cameron *et al.* (2011), Ni is primarily associated with lung and nasal cancer, primarily through inhalation exposure routes. According to Mathews and Fisher (2009), regulatory guidelines currently disregard the bioavailability of dietary exposure from elements; however, it is crucial to understand that dietary exposure could be a significant pathway for the absorption of various potentially harmful elements in marine fish. The daily safe consumption limit that can pose a CR for As was too low to sustain consumers who primarily depend on fish as their main source protein, whereas the consumption limit for Cr was at a more suitable range for regular consumers to avoid possible health risks.

3.5 Conclusions

The higher concentrations found in *D. capensis* at Chintsa (2022) were primarily associated with natural processes, particularly upwelling events along the east coast of

South Africa. Witsand (2021) and De Hoop (2021) fish experienced increased concentrations due to prolonged exposure to elements resulting from natural geogenic weathering, received by freshwater runoff and frequent upwelling events. At Tsitstikamma, the decrease in fish element concentration levels during different time periods was mainly due to the decline in the operations of both small- and large-scale industries, which typically contribute to the levels of coastal elements. The increased concentrations in different time periods at Chintsa are also attributed to occasional deep-water upwelling events along the east coast of South Africa and natural processes such as rock abrasion. The human health risk assessment showed both non-carcinogenic and carcinogenic risks for people who consume *D. capensis* along the South African coastline. Non-carcinogenic risks for As and Hg were found at all sites, while carcinogenic risks were observed for As at all sites, Cr at Chintsa (2022), De Hoop (2021) and Witsand (2021), and Ni at De Hoop (2021) and Witsand (2022). Regarding safe daily consumption limits, relying on Cape white seabream as the primary source of food was deemed unreliable due to insufficient maximum consumption limits. Future studies should not only consider adults when assessing human health risks linked to element overexposure but should include vulnerable groups such as pregnant women and infants. This study serves as a baseline for future research focussing on long-term human health risks along the South African coastline. Further research can include the evaluation of seasonal and temporal variations in element concentrations of *D. capensis*. To understand variations influenced by environmental factors, these studies could include different seasons and span multiple years. Future studies can also investigate different fish species along the same coastline to understand species-specific differences in element concentrations. This comparative analysis can provide valuable information on species-specific bioaccumulation patterns. Lastly, future research can look at other organs within *D. capensis* to understand whether certain elements accumulate more in particular parts of the fish. This can help identify potential bioaccumulation patterns.

Chapter 4: Conclusions and recommendations

Research on the levels of element concentrations in commonly consumed fish species is imperative, particularly in Africa (Bosch *et al.*, 2016). This research is important to gain an understanding of element exposure in marine fish and potential risks to regular consumers. The data from the study supported the hypothesis that variations in concentrations of elements were detected when comparing pre-lockdown (2018) and post-lockdown (2022) time intervals of *H. affinis*. These differences were mainly linked to reduced levels of recreational and economic activities that took place in Sodwana Bay, however, natural processes such as upwelling events and wind-driven erosion could most probably influenced the difference in concentrations that were observed. The increased Hg levels observed in *H. affinis* at Sodwana Bay were attributed to aerial depositions from the Mpumalanga highveld. The data did, however, not support the second hypothesis that high element concentrations will be associated with land-use activities, since high concentrations were observed at non-impacted sites such as Chintsa and Witsand. Chintsa did not have any known sources of point pollution, and according to literature Witsand was considered a relatively pristine site, thus did not reflect the accumulation of elements in *D. capensis* linked to land-use activities. As expected, the increased maritime traffic in Mossel Bay led to elevated levels of Pb. Moreover, the hypothesis suggesting that non-carcinogenic and carcinogenic risks would mostly be associated with impacted sites was not supported by the data, as risks were evident in relatively unimpacted sites along the South African coastline as well. The determinants that influence element concentrations in *D. capensis* were primarily associated with large-scale oceanographic processes and dispersed sources, including upwelling and geological weathering. Although the current study focused on assessing the bioaccumulation in fish, it is crucial to conduct studies on primary producers such as algae, phytoplankton, periphyton and zooplankton. These studies are essential to evaluate the accumulation and potential impacts of elements at lower trophic levels and how they can contribute to elevated levels in fish, enabling a comprehensive understanding of exposure through food webs. Sampling of *H. affinis* in Sodwana Bay revealed increased levels of elements (As, Cr, Pb, and Hg), which have the potential to cause adverse health effects in humans. Similarly, consumption of *D. capensis* from the respective sites along the South African coastline revealed that concentrations of As, Cr and Hg could result in adverse human health effects. The maximum safe daily consumption limit was found to be as low as 7.3 g for non-carcinogenic risks associated with *D. capensis*, while for *H. affinis*, non-carcinogenic

effects were evident at levels as low as 6.1 g in Sodwana Bay. It is important to note that this study did not specifically include infants or pregnant women in the human health assessment. These groups could potentially be more vulnerable to the development of non-carcinogenic and carcinogenic symptoms. Therefore, this study served primarily as a basic assessment of human health risks. However, it offered valuable information on understanding the health risks associated with trace element concentrations through fish consumption. The study's findings further underscore the scarcity of data on element bioaccumulation patterns in fish species along the South African coast, emphasising the necessity for ongoing, long-term monitoring (Erasmus *et al.*, 2021).

References

- Afandi, I., Talba, S., Benhra, A., Benbrahim, S., Chfiri, R., Labonne, M., Masski, H., Laë, R., Tito De Moraes, L., Bekkali, M. & Bouthir, F.Z. 2018. Trace metal distribution in pelagic fish species from the north-west African coast (Morocco). *International Aquatic Research*, 10: 191-205.
- Aman, A.A., Salman, M.S. & Yunus, A.P. 2020. COVID-19 and its impact on the environment: improved pollution levels during the lockdown period – a case from Ahmedabad, India. *Remote Sensing Applications: Society and Environment*, 20: 100382.
- Arif, M., Kumar, R. & Parveen, S. 2020. Reduction in water pollution in Yamuna River due to lockdown under COVID-19 pandemic. *ChemRxiv*, 22: 3389-3406.
- Ashan, M.A., Satter, F., Siddique, M.A.B., Akbor, M.A., Ahmed, S., Shajahan, M. & Khan, R., 2019. Chemical and physicochemical characterization of effluents from the tanning and textile industries in Bangladesh with multivariate statistical approach. *Environmental Monitoring and Assessment*, 191: 1-24.
- Atique Ullah, A.K.M., Akter, M., Musarrat, M. & Quraishi, S.B. 2019. Evaluation of possible human health risk of heavy metals from the consumption of two marine fish species *Tenualosa ilisha* and *Dorosoma cepedianum*. *Biological Trace Element Research*, 191: 485-494.
- Attwood, C.G., Allen, J. & Claassen, P.J. 2002. Nearshore surface current patterns in the Tsitsikamma National Park, South Africa. *African Journal of Marine Science*, 24: 151-160.
- Authman, M.M.N., Zaki, M.S., Khallaf, E.A. & Abbas, H.H. 2015. Use of fish as bioindicator of the effects of heavy metals pollution. *Journal of Aquaculture. Research and Development*, 6: 1-13.
- Bacigalupo, C. & Hale, B. 2012. Human health risks of Pb and As exposure via consumption of 651 home garden vegetables and incidental soil and dust ingestion: A probabilistic screening tool. *Science Total Environment*, 423: 27-38.
- Balali-Mood, M., Naseri, K., Tahergorabi, Z., Khazdair, M.R. & Sadeghi, M. 2021. Toxic mechanisms of five heavy metals: mercury, lead, chromium, cadmium, and arsenic. *Frontiers in Pharmacology*, 12: 643-972.

- Bartoli, G., Papa, S., Sagnella, E. & Fioretto, A. 2012. Heavy metal content in sediments along the Calore River: relationships with physical–chemical characteristics. *Journal of Environmental Management*, 95: S9-S14.
- Bashir, F.A. Alhemmal, E.M. 2015. Analysis of some heavy metal in marine fish in muscle, liver, and gill tissue in two marine fish species from Kapar coastal waters, Malaysia. In *The Second Symposium on Theories and Applications of Basic and Biosciences*, 2: 1-15.
- Binning, K. & Baird, D. 2001. Survey of heavy metals in the sediments of the Swartkops River Estuary, Port Elizabeth South Africa. *Water SA*, 27: 461-466.
- Bosch, A.C., O'Neill, B., Sigge, G.O., Kerwath, S.E. & Hoffman, L.C. 2016. Heavy metals in marine fish meat and consumer health: a review. *Journal of the Science of Food and Agriculture*, 96: 32-48.
- Breede River Estuarine Management Plan, 2016. Western Cape Estuary Management Framework and Implementation Strategy. SSI Environmental, 1st edition. Royal Haskoning D.H.V. (revised 2011, 2nd Edition, 2016).
- Budiawan, R.T.A. & Auerkari, E.I. 2017. Effects of Chromium on Human Body. *Annual Research & Review in Biology*, 13: 1-8.
- Cameron, K.S., Buchner, V. & Tchounwou, P.B. 2011. Exploring the molecular mechanisms of nickel-induced genotoxicity and carcinogenicity: a literature review. *Reviews on Environmental Health*, 26: 81-92.
- Chakraborty, B., Bera, B., Adhikary, P.P., Bhattacharjee, S., Roy, S., Saha, S., Ghosh, A., Sengupta, D. & Shit, P.K. 2021. Positive effects of COVID-19 lockdown on river water quality: evidence from River Damodar, India. *Scientific Reports*, 11: 20140.
- Charette, T., Rosabal, M. & Amyot, M. 2021. Mapping metal (Hg, As, Se), lipid and protein levels within fish muscular system in two fish species (Striped Bass and Northern Pike). *Chemosphere*, 265: 129036.
- Chevallier, R. 2014. Illegal sand mining in South Africa. <https://africaportal.org/publication/illegal-sand-mining-in-south-africa/>
- Cobbina, S.J., Chen, Y., Zhou, Z., Wu, X., Zhao, T., Zhang, Z., Feng, W., Wang, W., Li, Q. & Wu, X. 2015. Toxicity assessment due to sub-chronic exposure to individual and mixtures of four toxic heavy metals. *Journal of Hazardous Materials*, 294: 109-120.

- Coetzee, H.C., Nell, W., Van Eeden, E.S. & De Crom, E.P. 2015. Artisanal fisheries in the Ndumo area of the lower Phongolo River floodplain, South Africa. *Koedoe: African Protected Area Conservation and Science*, 57: 1-6.
- Coetzee, P.S. 1986. Diet composition and breeding cycle of blacktail, *Diplodus sargus capensis* (Pisces: Sparidae), caught off St Croix Island, Algoa Bay, South Africa. *South African Journal of Zoology*, 21: 237-243.
- de Silva, E.T., Klumpp, D. & Ridd, M. 2009. The barnacle *Balanus amphitrite* as a bioindicator for Cd: development and application of a simulation model. *Estuarine, Coastal and Shelf Science*, 82: 171-179.
- Degger, N., Chiu, J.M., Po, B.H., Anna, C.K., Zheng, G.J., Zhao, D.M., Xu, D., Cheng, Y.S., Wang, X.H., Liu, W.H. & Lau, T.C. 2016. Heavy metal contamination along the China coastline: A comprehensive study using Artificial Mussels and native mussels. *Journal of Environmental Management*, 180: 238-246.
- Degger, N., Wepener, V., Richardson, B.J. & Wu, R.S.S. 2011. Application of artificial mussels (AMs) under South African marine conditions: A validation study. *Marine Pollution Bulletin*, 63: 108-118.
- Department of Environmental Affairs and Tourism (DEAT), 2008. South Africa's National Programme of Action for Protection of the Marine Environment from Land-based Activities. First ed. Cape Town.
- Diaz-Pulido, G. & Garzon-Ferreira, J. 2002. Seasonality in algal assemblages on upwelling-influenced coral reefs in the Colombian Caribbean. *Botanica Marina*, 45: 284-292.
- Dicken, M.L. 2014. Socio-economic aspects of the Sodwana Bay SCUBA diving industry, with a specific focus on sharks. *African Journal of Marine Science*, 36: 39-47.
- Djedjibegovic, J., Marjanovic, A., Tahirovic, D., Caklovica, K., Turalic, A., Lugusic, A., Omeragic, E., Sober, M. & Caklovica, F. 2020. Heavy metals in commercial fish and seafood products and risk assessment in adult population in Bosnia and Herzegovina. *Science Report*, 10: 13-238.
- Dragun, Z., Roje, V., Mikac, N. & Raspor, B. 2009. Preliminary assessment of total dissolved trace metal concentrations in Sava River water. *Environmental Monitoring and Assessment*, 99: 1-4.

- Du Preez, M., Nel, R. & Bouwman, H. 2018. First report of metallic elements in loggerhead and leatherback turtle eggs from the Indian Ocean. *Chemosphere*, 197: 716-728.
- Dutta, V., Dubey, D. & Kumar, S. 2020. Cleaning the River Ganga: impact of lockdown on water quality and future implications on river rejuvenation strategies. *Science of the Total Environment*, 743: 140756.
- Ellender, B.R., Weyl, O.L. & Winker, H. 2009. Who uses the fishery resources in South Africa's largest impoundment? Characterising subsistence and recreational fishing sectors on Lake Gariep. *Water SA*, 35: 679-684.
- Erasmus, A., Ikenaka, Y., Nakayama, S.M., Ishizuka, M., Smit, N.J. & Wepener, V. 2020. Trophic transfer of pollutants within two intertidal rocky shore ecosystems in different biogeographic regions of South Africa. *Marine Pollution Bulletin*, 157: 111-309.
- Erasmus, A., Smit, N.J., Zimmermann, S., Nachev, M., Sures, B. & Wepener, V. 2021. Metal and metalloid concentrations in the southern African endemic inter- and infratidal super klipfish, *Clinus superciliosus*, from the west and south coasts of South Africa. *Marine Pollution Bulletin*, 172: 1-11.
- Erasmus, A., Wepener, V., Zimmermann, S., Nachev, M., Hadfield, K.A., Smit, N.J. and Sures, B. 2022a. High element concentrations are not always equivalent to a stressful environment: differential responses of parasite taxa to natural and anthropogenic stressors. *Marine Pollution Bulletin*, 184: 114110.
- Erasmus, J.H., Smit, N.J., Gerber, R., Schaeffner, B.C., Nkabi, N. & Wepener, V. 2022c. Total mercury concentrations in sharks, skates and rays along the South African coast. *Marine Pollution Bulletin*, 184: 114142.
- Erasmus, J.H., Zimmermann, S., Smit, N.J., Malherbe, W., Nachev, M., Sures, B. & Wepener, V. 2022b. Human health risks associated with consumption of fish contaminated with trace elements from intensive mining activities in a peri-urban region. *Science of the Total Environment*, 825: 1-12.
- Fatoki, O.S. & Mathabatha, S. 2001. An assessment of heavy metal pollution in the East London and Port Elizabeth harbours. *Water SA*, 27: 233-240.
- Fernandes, C., Fontaínhas-Fernandes, A., Peixoto, F. & Salgado, M. A. 2007. Bioaccumulation of heavy metals in *Liza saliens* from the Esmoriz–Paramos coastal lagoon, Portugal. *Ecotoxicology and Environmental Safety*, 66: 426-431.

- Fields, S. 2003. The environmental pain of pleasure boating. *Environmental Health Perspectives*, 111: 216-223.
- Gardner, B.D., Connel, A.D., Eagle, G.A., Molden, A.G.S., Oliff, W.D., Orren, M.J. & Watling, R.J. 1983. South African marine pollution survey report 1976-1979. National Scientific Programmes Unit: Council of Scientific & Industrial Research, SANSP Report 73, 113.
- Golovanova, I. L. 2008. Effect of heavy metals on the physiological and biochemical status of fishes and aquatic invertebrates. *Inland Water Biology*, 1: 93-101.
- Greenfield, R., Brink, K., Degger, N. & Wepener, V. 2014. The usefulness of transplantation studies in monitoring of metals in the marine environment: South African experience. *Marine Pollution Bulletin*, 85: 566-573.
- Gregory, M.A., Marshall, D.J., George, R.C., Anandraj, A. & McClurg, T.P. 2002. Correlations between metal uptake in the soft tissue of *Perna perna* and gill filament pathology after exposure to mercury. *Marine Pollution Bulletin*, 45: 114-125.
- Griffiths, C.L., Robinson, T.B., Lange, L. & Mead, A. 2010. Marine biodiversity in South Africa: an evaluation of current states of knowledge. *PloS One*, 5: 12008.
- Gu, Y.G., Lin, Q., Wang, X.H., Du, F.Y., Yu, Z.L. & Huang, H.H. 2015. Heavy metal concentrations in wild fishes captured from the South China Sea and associated health risks. *Marine Pollution Bulletin*, 96: 508-512.
- Gubili, C., Johnson, R., Gennari, E., Oosthuizen, W.H., Kotze, D., Meÿer, M., Sims, D.W., Jones, C.S. & Noble, L.R. 2009. Concordance of genetic and fin photo identification in the great white shark, *Carcharodon carcharias*, off Mossel Bay, South Africa. *Marine Biology*, 156: 2199-2207.
- Gutiérrez-Ravelo, A., Gutiérrez, Á.J., Paz, S., Carrascosa-Iruzubieta, C., González-Weller, D., Caballero, J.M., Revert, C., Rubio, C. & Hardisson, A. 2020. Toxic metals (Al, Cd, Pb) and trace element (B, Ba, Co, Cu, Cr, Fe, Li, Mn, Mo, Ni, Sr, V, Zn) levels in *Sarpa salpa* from the north-eastern Atlantic Ocean region. *International Journal of Environmental Research and Public Health*, 17: 7212.
- Guynup, S. & Safina, B.C. 2012. Mercury: Sources in the environment, health effects, and politics. *Blue Ocean Institute*, 1: 7-54.

- Hahn, A., Miller, C., Andó, S., Bouimetarhan, I., Cawthra, H.C., Garzanti, E., Green, A.N., Radeff, G., Schefuß, E. & Zabel, M. 2018. The provenance of terrigenous components in marine sediments along the east coast of southern Africa. *Geochemistry, Geophysics, Geosystems*, 19: 1946-1962.
- Hajeb, P., Jinap, S., Ismail, A., Fatimah, A.B., Jamilah, B. & Rahim, M.A. 2009. Assessment of mercury level in commonly consumed marine fishes in Malaysia. *Food control*, 20: 79-84.
- Hanekom, N., Randall, R.M., Bower, D., Riley, A. & Kruger, N. 2009. Garden Route National Park: The Tsitsikamma Area–State of Knowledge. *South African National Parks*, 78.
- Harris, H.H., Pickering, I.J. & George, G.N. 2003. The chemical form of mercury in fish. *Science* 301: 1203.
- Heath, A.G. 1987. Behaviour and nervous system function. *Water Pollution and Fish Physiology*, 181-196.
- Heath, R., Du Preez, H., Genthe, B. & Avenant-Oldewage, A. 2004. Freshwater fish and human health reference guide. *Water Research Commission: Pretoria, South Africa*.
- Heemstra, P. & Heemstra, E. 2004. Coastal Fishes of Southern Africa. *Grahamstown (National Inquiry Service Centre & South African Institute for Aquatic Biodiversity)*, 488.
- Huang, S.S., Strathe, A.B., Fadel, J.G., Johnson, M.L., Lin, P., Liu, TY. & Hung, S.S.O. 2013. The interactive effects of selenomethionine and methylmercury on their absorption, disposition, and elimination in juvenile white sturgeon. *Aquatic Toxicology*, 126: 274-282.
- Hutchings, K. & Clark, B.M. 2010. Assessment of the Effects of Heavy Metal Accumulation from Land-based Sources on the Diep River Estuary. *Report# EADP/14 prepared by Anchor Environmental for DEA & DP*, 1-89.
- Integrated Risk Information System (IRIS). 2017. IRIS Assessments of United States Environmental Protection Agency. <https://www.epa.gov/iris> [Accessed 12 November 2023].
- Javed, M. & Usmani, N. 2019. An overview of the adverse effects of heavy metal contamination on fish health. *Proceedings of the National Academy of Science, India, Section B*, 89: 389-403.

- Jeziarska, B. & Witeska, M. 2006. The metal uptake and accumulation in fish living in polluted waters. Springer Netherlands. In Soil and water pollution monitoring, *Protection and Remediation*, 107-114.
- Jia, Y., Wang, L., Qu, Z., Wang, C. & Yang, Z. 2017. Effects on heavy metal accumulation in freshwater fishes: species, tissues, and sizes. *Environmental Science and Pollution Research*, 24: 9379-9386.
- Jooste, A., Marr, S.M., Addo-Bediako, A. & Luus-Powell, W.J. 2014. Metal bioaccumulation in the fish of the Olifants River, Limpopo province, South Africa, and the associated human health risk: a case study of rednose labeo *Labeo rosae* from two impoundments. *African Journal of Aquatic Science*, 39: 271-277.
- Kara, M.H., Rouag, F. & Laouira, L. 2012. Westward range expansion of the Lessepsian spotted halfbeak *Hemiramphus far* (Hemiramphidae) in the Mediterranean Sea. *Marine Biodiversity Records*, 1: 45.
- Karunanidhi, D., Aravinthasamy, P., Subramani, T. & Setia, R. 2021. Effects of COVID-19 pandemic lockdown on microbial and metals contaminations in a part of Thirumanimuthar River, South India: a comparative health hazard perspective. *Journal of Hazardous Materials*, 416: 125909.
- Kawarazuka, N. & Béné, C. 2011. The potential role of small fish species in improving micronutrient deficiencies in developing countries: building evidence. *Public Health Nutrition*, 14: 1927-1938.
- Khan, R., Islam, H.M.T. & Islam, A.R.M.T. 2021. Mechanism of elevated radioactivity in Teesta River basin from Bangladesh: radiochemical characterization, provenance and associated hazards. *Chemosphere*, 264: 128459.
- Kottelat, M., Whiten, A.J., Kartikasari, S.N. & Wirjoatmodjo, S. 1993. *Freshwater Fishes of Western Indonesia and Sulawesi*. Periplus Editions (HK) Ltd. collaboration with the Environmental Management Development in Indonesia (EMDI) Project Ministry of State for Population and Environment, Republic of Indonesia.
- Krishna, P.V., Jyothrimayi, V. & Madhussudhana Rao, K. 2014. Human health risk assessment of heavy metal accumulation through fish consumption from Machilipatnam Coast, Andhra Pradesh, India. *International Research Journal of Public and Environmental Health*, 1: 121-125.

- Kubra, K., Mondol, A.H., Ali, M.M., Palash, M.A.U., Islam, M.S., Ahmed, A.S., Masuda, M.A., Islam, A.R.M.T., Bhuyan, M.S., Rahman, M.Z. & Rahman, M.M. 2022. Pollution level of trace metals (As, Pb, Cr and Cd) in the sediment of Rupsha River, Bangladesh: assessment of ecological and human health risks. *Frontiers in Environmental Science*, 10: 778544.
- Kumar, N., Bhushan, S., Gupta, S.K., Kumar, P., Chandan, N.K., Singh, D.K. & Kumar, P. 2021. Metal determination and biochemical status of marine fishes facilitate the biomonitoring of marine pollution. *Marine Pollution Bulletin*, 170: 112682.
- Lares, M.L., Flores-Muñoz, G. & Lara-Lara, R. 2002. Temporal variability of bioavailable Cd, Hg, Zn, Mn, and Al in an upwelling regime. *Environmental Pollution*, 120: 595-608.
- Lasiak T.A. 1986. Juveniles, food, and the surf zone habitat: implications for teleost nursery areas. *South African Journal of Zoology*, 21: 51-56.
- Learner, J., Binedell, M., Oosthuizen, R., Matooane, M., Mason, R. & Macmillan, P. 2009. South Africa Risk Assessment of the Mercury Footprint in Selected Rivers. CSIR Natural Resources and the Environment (Report No: CSIR/NRE/WR/EXP/2008/ 0450/A).
- Lehmann, A. & Myrberg, K. 2008. Upwelling in the Baltic Sea—A review. *Journal of Marine Systems*, 74: S3-S12.
- Lemes, M. & Wang, F. 2009. Methylmercury speciation in fish muscle by HPLC-ICP-MS following enzymatic hydrolysis. *Journal of Analytical Atomic Spectrometry*, 24: 663-668.
- Li, F., Huang, J., Zeng, G., Yuan, X., Li, X., Liang, J., Wang, X., Tang, X. & Bai, B. 2013. Spatial risk assessment and sources identification of heavy metals in surface sediments from the Dongfing Lake, Middle China. *Journal of Geochemical Exploration*, 132: 75-83.
- Licata, P., Trombetta, D., Cristani, M., Naccari, C., Martino, D., Caló, M. & Naccari, F. 2005. Heavy metals in liver and muscle of bluefin tuna (*Thunnus thynnus*) caught in the straits of Messina (Sicily, Italy). *Environmental Monitoring and Assessment*, 107: 239-248.
- Lutjeharms, J.R.E., Cooper, J. & Roberts, M. 2000. Upwelling at the inshore edge of the Agulhas Current. *Continental Shelf Research*, 20: 737-761.
- Madgett, A.S., Yates, K., Webster, L., McKenzie, C. & Moffat, C.F. 2021. The concentration and biomagnification of trace metals and metalloids across four trophic levels in a marine food web. *Marine Pollution Bulletin*, 173: 112929.
- Mann, B. 2013. Southern African marine linefish species profiles. *Special publication*, 9.

- Mason, R.P., Reinfelder, J.R. & Morel, F.M.M. 1995. Bioaccumulation of mercury and methylmercury. *Water, Air and Soil Pollution*, 80: 915-921.
- Mathews, T. & Fisher, N.S. 2009. Dominance of dietary intake of metals in marine elasmobranch and teleost fish. *Science of the Total Environment*, 407: 5156-5161.
- McCafferty, J.R., Ellender, B.R., Weyl, O.L.F. & Britz, P.J. 2012. The use of water resources for inland fisheries in South Africa. *Water SA*, 38: 327-344.
- McGlashan, J.E. 1989. Characteristics of waste discharges. *Marine Pollution*, Report No. 161: 11-20.
- Mograbi, J. & Rogerson, C.M. 2007. Maximising the local pro-poor impacts of dive tourism: Sodwana Bay, South Africa. *Urban Forum*, 18: 85-104.
- Moodley, K., Pillay, S., Pather, K. & Ballabh, H. 2016. Seasonal discharge and chemical flux variations of rivers flowing into the Bayhead canal of Durban Harbour, South Africa. *Acta Geochimica*, 35: 340-353.
- Moodley, R., Mahlangeni, N.T. & Reddy, P. 2021. Determination of heavy metals in selected fish species and seawater from the South Durban Industrial Basin, KwaZulu-Natal, South Africa. *Environmental Monitoring and Assessment*, 193: 1-11.
- Morel, F.M. & Price, N.M. 2003. The biogeochemical cycles of trace metals in the oceans. *Science*, 300: 944-947.
- Mubiana, V.K., Vercauteren, K. & Blust, R. 2006. The influence of body size, condition index and tidal exposure on the variability in metal bioaccumulation in *Mytilus edulis*. *Environmental Pollution*, 144: 272-279.
- Mzimela, H.M., Wepener, V. & Cyrus, D.P. 2003. Seasonal variation of selected metals in sediments, water and tissues of the groovy mullet, *Liza dumerelii* (Mugilidae) from the Mhlathuze Estuary, South Africa. *Marine Pollution Bulletin*, 46: 659-664.
- Nan, Y., Liu, W.B., Xie, H.T., Gao, L.R., Han, Y., Wang, M.J. & Li, H.F. 2016. Distribution and assessment of heavy metals in the surface sediment of Yellow River China. *Journal of Environmental Science*, 39: 45-51.
- Nelson, J.S., Grande, T.C. & Wilson, M.V. 2016. *Fishes of the World*. John Wiley & Sons.
- Nyarko, E., Boateng, C.M., Asamoah, O., Edusei, M.O. & Mahu, E. 2023. Potential human health risks associated with ingestion of heavy metals through fish consumption in the Gulf of Guinea. *Toxicology Reports*, 10: 117-123.

- O'Donoghue, S. & Marshall, D.J. 2003. Marine pollution research in South Africa: a status report. *South African Journal of Science*, 99: 349-356.
- Olaniran, A.O., Naicker, K. & Pillay, B. 2014. Assessment of physico-chemical qualities and heavy metal concentrations of Umgeni and Umdloti Rivers in Durban, South Africa. *Environmental Monitoring and Assessment*, 186: 2629-2639.
- Oosthuizen, J. & Ehrlich, R. 2001. The impact of pollution from a mercury processing plant in KwaZulu-Natal, South Africa, on the health of fish-eating communities in the area: An environmental health risk assessment. *International Journal of Environmental Health Research*, 11: 41-50.
- Orr, K.K., Burgess, J.E. & Froneman, P.W. 2008. The effects of increased freshwater inflow on metal enrichment in selected Eastern Cape estuaries, South Africa. *Water SA* 34: 39-52.
- Patrick, P. & Strydom, N.A. 2009. Swimming abilities of wild-caught, late-stage larvae of *Diplodus capensis* and *Sarpa salpa* (Pisces: Sparidae) from temperate South Africa. *Estuarine, Coastal and Shelf Science*, 85: 547-554.
- Peerzada, N., McMorrow, L., Skiliros, S., Guinea, M. & Ryan, P. 1990. Distribution of heavy metals in Gove Harbour, Northern Territory, Australia. *Science of the Total Environment*, 92: 1-12.
- Peng, X., Liu, F. & Wang, W.X. 2016. Organ-specific accumulation, transportation, and elimination of methylmercury and inorganic mercury in a low Hg accumulating fish. *Environmental Toxicology and Chemistry*, 35: 2074-2083.
- Penney, A.J., Mann-Lang, J.B., Van Der Elst, R.P. & Wilke, C. 1999. Long-term trends in catch and effort in the KwaZulu-Natal nearshore line fisheries. *African Journal of Marine Science*, 21: 51-76.
- Pheiffer, W., Wolmarans, N.J., Gerber, R., Yohannes, Y.B., Ikenaka, Y., Ishizuka, M., Smit, N.J., Wepener, V. & Pieters, R. 2018. Fish consumption from urban impoundments: what are the health risks associated with DDTs and other organochlorine pesticides in fish to township residents of a major inland city. *Science of the Total Environment*, 628: 517-527.
- Ploetz, D. M., Fitts, B. E. & Rice, T. M. 2007. Differential accumulation of heavy metals in muscle and liver of a marine fish, (King Mackerel, *Scomberomorus cavalla* Cuvier) from

the northern Gulf of Mexico, USA. *Bulletin of Environmental Contamination and Toxicology*, 78: 134- 137.

Ports & Ships. 2019. Mossel Bay. Available at <https://ports.co.za/mossel-bay.php>. [Accessed 17 October 2023].

Rahmani, J., Fakhri, Y., Shahsavani, A., Bahmani, Z., Urbina, M.A., Chirumbolo, S., Keramati, H., Moradi, B., Bay, A. & Bjørklund, G. 2018. A systematic review and meta-analysis of metal concentrations in canned tuna fish in Iran and human health risk assessment. *Food and Chemical Toxicology*, 118: 753-765.

Rainbow, P.S. & Luoma, S.N. 2011. Trace metals in aquatic invertebrates. In: Beyer, W.N., Meador, J.P. (Eds.), *Environmental Contaminants in Biota: Interpreting Tissue Concentrations*. Taylor and Francis Books, Boca Raton, FL, USA, 231-252.

Rainbow, P.S. & Phillips, J.H. 1993. Review: cosmopolitan biomonitors of trace metals. *Marine Pollution Bulletin*, 26: 593-601.

Rajeshkumar, S., Mini, J. & Munuswamy, N. 2013. Effects of heavy metals on antioxidants and expression of HSP70 in different tissues of Milk fish (*Chanos chanos*) of Kaattuppalli Island, Chennai, India. *Ecotoxicology and Environmental Safety*, 98: 8-18.

Ramos-Miras, J.J., Sanchez-Muros, M.J., Morote, E., Torrijos, M., Gil, C., Zamani-Ahmadm Mahmoodi, R. & Martin, J.R. 2019. Potentially toxic elements in commonly consumed fish species from the western Mediterranean Sea (Almería Bay): bioaccumulation in liver and muscle tissues in relation to biometric parameters. *Science of the Total Environment*, 671: 280-287.

Rendana, M., Idris, W.M.R. & Rahim, S.A. 2022. Effect of COVID-19 Movement Control Order Policy on Water Quality Changes in Sungai Langat, Selangor, Malaysia within Distinct Land Use Areas. *Sains Malaysiana*, 51: 1587-1598.

Roberts, M.J. & Nieuwenhuys, C. 2016. Observations and mechanisms of upwelling in the northern KwaZulu-Natal Bight, South Africa. *African Journal of Marine Science*, 38: S43-S63.

Selvam, S., Jesuraja, K., Venkatramanan, S., Chung, S.Y., Roy, P.D., Muthukumar, P. & Kumar, M. 2020. Imprints of pandemic lockdown on subsurface water quality in the coastal industrial city of Tuticorin, South India: A revival perspective. *Science of the Total Environment*, 738: 139848.

- Sevcikova, M., Modra, H., Slaninova, A. Svobodova, Z. 2011. Metals as a cause of oxidative stress in fish: a review. *Veterinární Medicína*, 56: 537-546.
- Sheppard. C. Ed., 2019. *World Seas: An Environmental Evaluation* (57-78). London: Academic Press.
<https://www.sciencedirect.com/science/article/abs/pii/B9780081008539000063> [Accessed 12 November 2023]
- Siddique, M.A.B., Islam, M.S., Ali, M.M., Tokatli, C., Islam, A., Pal, S.C., Idris, A.M., Malafaia, G. & Islam, A.R.M.T. 2023. Effects of COVID-19 era on a subtropical river basin in Bangladesh: Heavy metal (loid) s distribution, sources, and probable human health risks. *Science of the Total Environment*, 857: 159383.
- Sigel, A., Sigel, H. & Sigel, R. K. O. 2009. Metallothioneins and related chelators. *Royal Society of Chemistry*, 545: 1-29.
- Sink, K., Holness, S., Harris, L., Majiedt, P., Atkinson, L., Robinson, T., Kirkman, S., Hutchings, L., Leslie, R., Lamberth, S., Kerwath, S., Von der Heyden, S., Lombard, A., Attwood, C., Branch, G., Fairweather, T., Taljaard, S., Weerts, S., Cowley, P., Awad, A., Halpern, B., Grantham, H. & Wolf, T. 2012. National Biodiversity Assessment 2011: Technical report. In: Marine and Coastal Component, 4. *South African National Biodiversity Institute*, 325.
- Smale, M.J. 1992. Predatory fish and their prey—an overview of trophic interactions in the fish communities of the west and south coasts of South Africa. *South African Journal of Marine Science*, 12: 803-821.
- Smith, M.M. & Heemstra, P.C. 1995. *Smith's Sea Fishes*. Macmillan, Johannesburg, South Africa, 1047.
- Song, Y., Choi, M.S., Lee, J.Y. & Jang, D.J. 2014. Regional background concentrations of heavy metals (Cr, Co, Ni, Cu, Zn, Pb) in coastal sediments of the South Sea of Korea. *Science of the Total Environment*, 484: 80-91.
- Sturve, J., Gustavsson, M., Moksnes, P.O. & de Abreu, D.C. 2021. Effects of pesticides and metals on penaeid shrimps in Maputo Bay, Mozambique—A field study. *Marine Pollution Bulletin*, 173: 112964.
- Sun, H., Brocato, J. & Costa, M. 2015. Oral chromium exposure and toxicity. *Current Environmental Health Reports*, 2: 295-303.

- Swales, S., Storey, A.W., Roderick, I.D., Figa, B.S., Bakowa, K.A. & Tenakanai, C.D. 1998. Biological monitoring of the impacts of the Ok Tedi copper mine on fish populations in the Fly River system, Papua New Guinea. *Science of the Total Environment*, 214: 99-111.
- Taljaard, S., Monteiro, P.M.S. & Botes, W.A.M. 2006. A structured ecosystem-scale approach to marine water quality management. *Water SA*, 32: 535-542.
- Tran, N., Chu, L., Chan, C.Y., Genschick, S., Phillips, M.J. & Kefi, A.S. 2019. Fish supply and demand for food security in Sub-Saharan Africa: An analysis of the Zambian fish sector. *Marine Policy*, 99: 343-350.
- Tunley K. 2009. State of management of South Africa's marine protected areas. WWF-South Africa Report Series – 2009/ Marine/001. Cape Town: WWF-SA.
- UNESCO,1999. World Communication and Information 1999-2000. Report Published by United Nations Educational, Scientific and Cultural Organisation.
- United States Environmental Protection Agency (USEPA), 2000. volume2.pdf (epa.gov) [Accessed 07 November 2023]
- United States Environmental Protection Agency (USEPA), 2005. Guidelines for Carcinogen Risk Assessment. Guidelines for Carcinogen Risk Assessment (epa.gov) [Accessed 07 November 2023]
- Van As, J., du Preez, J., Brown, L. & Smit, N. 2012. The story of Life and the Environment—an African perspective—Cape Town. 456.
- Van der Schyff, V., Yive, N.S.C.K. & Bouwman, H. 2020. Metal concentrations in corals from South Africa and the Mascarene Basin: a first assessment for the Western Indian Ocean. *Chemosphere*, 239 :124784.
- Van Der Spuy, L., Erasmus, J.H., Nachev, M., Schaeffner, B.C., Sures, B., Wepener, V. & Smit, N.J. 2023. The use of fish parasitic isopods as element accumulation indicators in marine pollution monitoring. *Marine Pollution Bulletin*, 194: 115385.
- Van Rooyen, D., Erasmus, J.H., Gerber, R., Nachev, M., Sures, B., Wepener, V. & Smit, N.J. 2023. Bioaccumulation and trophic transfer of total mercury through the aquatic food webs of an African sub-tropical wetland system. *Science of the Total Environment*, 889: 164210.

- Verma, R. & Dwivedi, P. 2013. Heavy metal water pollution-A case study. *Recent Research in Science and Technology*, 5: 98-99.
- Vermaak, A., Smit, N.J. & Kudlai, O. 2023. Molecular characterisation of three species of *Coitocaecum* (Digenea: Opecoelidae) infecting *Clinus superciliosus* (Clinidae) in South Africa, with description of *Coitocaecum brayi* sp. n. *Folia Parasitologica*, 70: 1-10.
- Vetrimurugan, E., Shruti, V.C., Jonathan, M.P., Roy, P.D., Rawlins, B.K. & Rivera-Rivera, D.M. 2018. Metals and their ecological impact on beach sediments near the marine protected sites of Sodwana Bay and St. Lucia, South Africa. *Marine Pollution Bulletin*, 127: 568-575.
- Vogt, T., Pieters, R., Giesy, J. & Newman, B.K. 2019. Biological toxicity estimates show involvement of a wider range of toxic compounds in sediments from Durban, South Africa than indicated from instrumental analyses. *Marine Pollution Bulletin*, 138: 49-57.
- Volschenk, C.M., Gerber, R., Mkhonto, M.T., Ikenaka, Y., Yohannes, Y.B., Nakayama, S.M.M., Ishizuka, M., van Vuren, J.H.J., Wepener, V. & Smit, N.J. 2019. Bioaccumulation of persistent organic pollutants and their trophic transfer through the food web: human health risks to the rural communities reliant on fish from South Africa's largest floodplain. *Science of the Total Environment*, 685: 1116-1126.
- Wepener V. & Degger N. 2019. South Africa. In: Sheppard C (Ed.), World seas: an environmental evaluation, Volume II: The Indian Ocean to the Pacific. (2nd edn). London: Elsevier, 101-119.
- Wepener, V. & Degger, N. 2012. Status of marine pollution research in South Africa (1960-present). *Marine Pollution Bulletin*, 64: 1508-1512.
- Wepener, V. & Vermeulen, L.A. 2005. A note on the concentrations and bioavailability of selected metals in sediments of Richards Bay Harbour, South Africa. *Water SA*, 34: 589-595.
- Wepener, V. 2008. Application of active biomonitoring within an integrated water resources management framework in South Africa. *South African Journal of Science*, 104: 367-373.
- Wepener, V., 2007. Carbon, nitrogen, and phosphorus fluxes in four sub-tropical estuaries of northern KwaZulu-Natal: case studies in the application of a mass balance approach. *Water SA*, 33: 203-214.

- Wepener, V.W. & Degger, N.D. 2020. Monitoring metals in South African harbours between 2008 and 2009, using resident mussels as indicator organisms. *African Zoology*, 55: 267-277.
- Weyl, O.L.F., Barkhuizen, L., Christison, K., Dalu, T., Hlungwani, H.A., Impson, D., Sankar, K., Mandrak, N.E., Marr, S.M., Sara, J.R. & Smit, N.J. 2021. Ten research questions to support South Africa's inland fisheries policy. *African Journal of Aquatic Science*, 46: 1-10.
- Whitfield, A.K. & Cowley, P.D. 2010. The status of fish conservation in South African estuaries. *Journal of Fish Biology*, 76: 2067-2089.
- Whitfield, A.K. 1985. The role of zooplankton in the feeding ecology of fish fry from some southern African estuaries. *South African Journal of Zoology*, 20: 166-171.
- Whitfield, A.K., Bate, G.C., Forbes, T. & Taylor, R.H. 2013. Relinkage of the Mfolozi River to the St Lucia estuarine system—urgent imperative for the long-term management of a Ramsar and World Heritage Site. *Aquatic Ecosystem Health Management*, 16: 104-108.
- World Health Organisation (WHO). 2023. Lead. Lead poisoning (who.int) [Accessed 10 November 2023].
- Xu, F., Tian, X., Yin, F., Zhao, Y. & Yin, X. 2016. Heavy metals in the surface sediments of the northern portion of the South China Sea shelf: distribution, contamination, and sources. *Environmental Science and Pollution Research*, 23: 8940-8950.
- Yunus, A.P., Masago, Y. & Hijioka, Y. 2020. COVID-19 and surface water quality: improved lake water quality during the lockdown. *Science of the Total Environment*, 731: 139012.
- Zereini, F., Müller, I. & Wiseman, C.L.S. 2015. The Influence of Anionic Species (Cl-NO₃-SO₄²⁻) on the Transformation and Solubility of Platinum in Platinum/Aluminium Oxide Model Substance. *Platinum Metals in the Environment*. New York: Springer, 277-288.
- Zhang, C., Wang, S., Pan, Z., Sun, D., Xie, S., Zhou, A., Wang, J. & Zou, J. 2020. Occurrence and distribution of microplastics in commercial fishes from estuarine areas of Guangdong, South China. *Chemosphere*, 260: 127656.

APPENDIX A

Table A. 1: The average concentration of trace elements in muscle tissue was used to calculate the mean and standard deviation of hazard quotients (HQs), of *Hyporhamphus affinis* from 2018 (clear column) and 2022 (shaded column), assuming a person of 60 kg consumes a fish meal (150 g) twice a week.

Trace elements	<i>Hyporhamphus affinis</i>	
	2018	2022
Hazard quotients for non-carcinogenic risk (HQ)		
As	4.8 $\pm$ 9.5	7.6 $\pm$ 2.5
Cd	0.06 $\pm$ 0.03	0.011 $\pm$ 0.010
Cr	0.02 $\pm$ 0.008	0.086 $\pm$ 0.075
Cu	0.003 $\pm$ 0.007	0.005 $\pm$ 0.003
Fe	0.004 $\pm$ 0.01	0.008 $\pm$ 0.003
Hg	1.8 $\pm$ 0.4	0.6 $\pm$ 0.1
Mn	0.0005 $\pm$ 0.0007	0.005 $\pm$ 0.009
Ni	0.001 $\pm$ 0.002	0.003 $\pm$ 0.002
Pb	0.02 $\pm$ 0.01	0.052 $\pm$ 0.044
Sb	Too small	0.039 $\pm$ 0.021
Sn	<10 ⁻⁴	<10 ⁻⁴
Zn	0.02 $\pm$ 0.03	0.047 $\pm$ 0.031

Reference dose (RfD) ($\mu\text{g/kg/day}$): As (0.3); Cd (1); Cr (3); Cu (40); Ni (20); Zn (300) (USEPA, 2000); Pb (3.5) (Djedjibegovic *et al.*, 2020).

Table A. 2: The average concentration of trace elements in muscle tissue was used to calculate the mean and standard deviation of cancer risk (CR) of *Hyporhamphus affinis* from 2018 (clear column) and 2022 (shaded column), assuming a person of 60 kg consumes a fish meal (150 g) twice a week.

Trace elements	<i>Hyporhamphus affinis</i>	
	2018	2022
Cancer risk (CR) ($\times 10^{-4}$)		
As	126 $\pm$ 64	103 $\pm$ 34
Cd	0.0009 $\pm$ 0.002	0.0003 $\pm$ 0.0002
Cr	0.9 $\pm$ 0.4	3.8 $\pm$ 3.4
Cu	Too small	Too small
Ni	0.4 $\pm$ 0.3	1.5 $\pm$ 1.2
Pb	1.2 $\pm$ 0.9	0.02 $\pm$ 0.01
Zn	Too small	Too small

Cancer slope factors (mg/kg/day): As (1.5); Cd (0.001); Cr (0.5); Ni (0.84) (USEPA, 2000; IRIS, 2017); Pb (0.00357) (Bacigalupo and Hale, 2012).

Table A. 3: The average concentration of trace elements in muscle tissue was used to calculate the mean and standard deviation of the maximum safe daily consumption of *Hyporhamphus affinis* from 2018 (clear column) and 2022 (shaded column), assuming a person of 60 kg consumes a fish meal (150 g) twice a week.

Trace elements	<i>Hyporhamphus affinis</i>	
	2018	2022
Maximum safe consumption limit per day (g)		
As (CR)	30 $\pm$ 10	36 $\pm$ 10
As (NC)	6.1 $\pm$ 2	7.2 $\pm$ 2.1
Cd (CR)	8.2 $\pm$ 6.6	7.3 $\pm$ 4.5
Cd (NC)	24628 $\pm$ 19833	21881 $\pm$ 13579
Cr (CR)	471 $\pm$ 143	130 $\pm$ 48
Cr (NC)	2762 $\pm$ 872	784 $\pm$ 289
Cu (NC)	8141 $\pm$ 3042	18869 $\pm$ 23352
Fe (NC)	2762 $\pm$ 620	6930 $\pm$ 2474
Hg (NC)	31 $\pm$ 8.1	79 $\pm$ 19
Mn (NC)	99498 $\pm$ 50687	28814 $\pm$ 17162
Ni (CR)	1001 $\pm$ 440	887 $\pm$ 339
Ni (NC)	23836 $\pm$ 10497	21137 $\pm$ 8082
Pb (CR)	19 $\pm$ 17	1.9 $\pm$ 1.9
Pb (NC)	332 $\pm$ 284	32 $\pm$ 33
Sb (NC)	10039 $\pm$ 6473	1754 $\pm$ 1123
Sn (NC)	>10 ⁵	>10 ⁵
Zn (NC)	1966 $\pm$ 1179	1545 $\pm$ 9

APPENDIX B

Table B. 1: The average concentration of trace elements in muscle tissue was used to calculate the mean and standard deviation of hazard quotients (HQs), of *Diplodus capensis* from sites along the South African coast, assuming a person of 60 kg consumes a fish meal (150 g) twice a week.

Elem.	<i>Diplodus capensis</i>					
	De Hoop (2021)	Tsitsikamma (2021)	Witsand (2021)	Chintsa (2022)	Mossel Bay (2022)	Tsitsikamma (2022)
Hazard quotients for non-carcinogenic risk (HQ)						
As	5 ± 2.2	7.5 ± 3.1	3.4 ± 1	7.2 ± 3	1.9 ± 0.9	5.6 ± 2.5
Cd	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²
Cr	0.1 ± 0.2	<10 ⁻²	0.03 ± 0.01	0.1 ± 0.1	0.01 ± 0.01	<10 ⁻²
Cu	<10 ⁻²	<10 ⁻²	<10 ⁻²	0.01 ± 0.03	<10 ⁻²	<10 ⁻²
Fe	0.03 ± 0.01	<10 ⁻²	0.04 ± 0.01	0.01 ± 0.01	0.01 ± 0.01	<10 ⁻²
Hg	2.5 ± 0.8	2.5 ± 0.6	3.9 ± 1.3	2.9 ± 2.3	3.3 ± 2.7	3.1 ± 1.3
Mn	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²
Ni	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²
Pb	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²
Sb	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²
Sn	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²	<10 ⁻²
Zn	0.01 ± 0.01	<10 ⁻²	<10 ⁻²	0.02 ± 0.02	0.01 ± 0.01	<10 ⁻²

Reference dose (RfD) (µg/kg/day): As (0.3); Cd (1); Cr (3); Cu (40); Ni (20); Zn (300) (USEPA, 2000); Pb (3.5) (Djedjibegovic *et al.*, 2020).

*Elem. - Element

Table B. 2: The average concentration of trace elements in muscle tissue was used to calculate the mean and standard deviation of cancer risk (CR) of *Diplodus capensis* from sites along the South African coast, assuming a person of 60 kg consumes a fish meal (150 g) twice a week.

Element	<i>Diplodus capensis</i>					
	De Hoop (2021)	Tsitsikamma (2021)	Witsand (2021)	Chintsa (2022)	Mossel Bay (2022)	Tsitsikamma (2022)
Cancer risk (CR) ($\times 10^{-4}$)						
As	67 $\pm$ 30	101 $\pm$ 42	45 $\pm$ 14	97 $\pm$ 41	25 $\pm$ 11	75 $\pm$ 34
Cd	L.C.	L.C.	L.C.	L.C.	L.C.	L.C.
Cr	6.3 $\pm$ 13	0.9 $\pm$ 0.3	1.8 $\pm$ 0.6	7.5 $\pm$ 7.6	0.7 $\pm$ 0.1	0.9 $\pm$ 0.2
Ni	1.1 $\pm$ 0.2	0.8 $\pm$ 0.3	1.1 $\pm$ 0.3	0.3 $\pm$ 0.2	0.4 $\pm$ 0.1	0.6 $\pm$ 0.2
Pb	L.C.	L.C.	L.C.	L.C.	L.C.	L.C.

Cancer slope factors (mg/kg/day): As (1.5); Cd (0.001); Cr (0.5); Ni (0.84) (USEPA, 2000; IRIS, 2017); Pb (0.00357) (Bacigalupo and Hale, 2012).

*L.C.- Level of Concern, between 10^{-4} and 10^{-6}

Table B. 3: The average concentration of trace elements in muscle tissue was used to calculate the mean and standard deviation of the maximum safe daily consumption of *Diplodus capensis* from sites along the South African coast, assuming a person of 60 kg consumes a fish meal (150 g) twice a week.

Element	<i>Diplodus capensis</i>					
	De Hoop (2021)	Tsitsikamma (2021)	Witsand (2021)	Chintsa (2022)	Mossel Bay (2022)	Tsitsikamma (2022)
Maximum safe consumption limit per day (g)						
As (CR)	59 $\pm$ 25	36 $\pm$ 10	78 $\pm$ 23	41 $\pm$ 18	161 $\pm$ 86	55 $\pm$ 26
As (NC)	11 $\pm$ 5.1	7.3 $\pm$ 2.1	15 $\pm$ 4.6	8.3 $\pm$ 3.7	32 $\pm$ 17	11 $\pm$ 5.3
Cd (CR)	17 $\pm$ 10	30 $\pm$ 19	31 $\pm$ 10	55 $\pm$ 27	89 $\pm$ 45	47 $\pm$ 20
Cd (NC)	>10 ³	>10 ³	>10 ³	>10 ³	>10 ³	>10 ³
Cr (CR)	160 $\pm$ 81	424 $\pm$ 120	224 $\pm$ 71	101 $\pm$ 65	553 $\pm$ 146	420 $\pm$ 95
Cr (NC)	>10 ³	>10 ³	>10 ³	611 $\pm$ 393	>10 ³	>10 ³
Cu (NC)	>10 ³	>10 ³	>10 ³	>10 ³	>10 ³	>10 ³
Fe (NC)	>10 ³	>10 ³	>10 ³	>10 ³	>10 ³	>10 ³
Hg (NC)	21 $\pm$ 7.3	21 $\pm$ 5.3	13 $\pm$ 3.9	26 $\pm$ 14	20 $\pm$ 9.6	19 $\pm$ 10
Mn (NC)	>10 ³	>10 ³	>10 ³	>10 ³	>10 ³	>10 ³
Ni (CR)	>10 ³	>10 ³	>10 ³	>10 ³	>10 ³	>10 ³
Ni (NC)	>10 ³	>10 ³	>10 ³	>10 ³	>10 ³	>10 ³
Pb (CR)	23 $\pm$ 3.8	20 $\pm$ 10	32 $\pm$ 8.8	7.1 $\pm$ 3	33 $\pm$ 8.2	38 $\pm$ 19
Pb (NC)	391 $\pm$ 64	343 $\pm$ 98	638 $\pm$ 28	119 $\pm$ 51	567 $\pm$ 138	642 $\pm$ 320
Sb (NC)	>10 ³	>10 ³	>10 ³	>10 ³	>10 ³	>10 ³
Sn (NC)	>10 ³	>10 ³	>10 ³	>10 ³	>10 ³	>10 ³

Zn (NC)	$>10^3$	$>10^3$	$>10^3$	$>10^3$	$>10^3$	$>10^3$
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