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## Heavy Metals within the Hawaiian Islands Archipelago as Evidenced in the Hawaiian Monk Seal and Their Prey

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# Thesis of Yvanna M. Strait

Submitted in Partial Fulfillment of the Requirements for the Degree of

## Master of Science Marine Science

Nova Southeastern University  
Halmos College of Arts and Sciences

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Approved:  
Thesis Committee

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NOVA SOUTHEASTERN UNIVERSITY  
HALMOS COLLEGE OF ARTS AND SCIENCES

Heavy Metals within the Hawaiian Islands Archipelago as Evidenced in the  
Hawaiian Monk Seal and Their Prey

By

Yvanna M. Strait

Submitted to the Faculty of  
Halmos College of Arts and Sciences  
in partial fulfillment of the requirements for  
the degree of Master of Science with a specialty in:

Marine Science

Nova Southeastern University

## **Abstract**

The Hawaiian monk seal (HMS) is an endangered, endemic seal native to the Northwestern Hawaiian Islands (NWHI). During the 21<sup>st</sup> century, members of the HMS population have established residency within the Main Hawaiian Islands (MHI). This habitat shift may increase exposure of the animals to greater anthropogenic (urban industrialization, agricultural practices, and military activity) and natural (volcanic activity) heavy metal contaminants. Induced coupled plasma mass spectroscopy (ICPMS) analysis compared 16 heavy metal concentrations in HMS bone segregated by region, age, and sex. In addition, metal concentrations from potential prey items from the southern extent of the Northwestern Hawaiian Islands were analyzed relative to temporospatial distribution and species' biometrics. The MHI and NWHI seals and potential prey contain all 16 heavy metals studied, essential and nonessential metals. The HMS bone was found to have the highest concentrations of Zn and Fe, both elements used structurally in bone. The MHI had significantly lower concentration than NWHI when a significance was found. Anthropogenic sources of heavy metals might be sinking near their sources instead of dispersing out into the marine environment. Concentration differences of Cu and Fe found among ages and between sexes showed evidence of potential maternal offloading while concentration differences of Cd found among ages showed potential evidence bioaccumulation and/or biomagnification. The HMS had significantly higher concentrations of Sn, Zn, and Fe than the potential prey items. Tin may be biomagnifying within the food web while Zn and Fe are related sample tissue differences (bone vs whole organism) as heavy metals do not equally bind to all tissues.

Keywords: Heavy metals, Hawaiian monk seal, *Neomonachus schauinslandi*, Hawaiian Island Archipelago, bone, SECLER

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# 1. INTRODUCTION

## 1.1 Preface

The Hawaiian Island Archipelago is comprised largely of uninhabited islands and atolls except for the eight islands that make up the U. S. State of Hawai'i, making it a unique place for a comparative study on contaminants. Inorganic contaminants, specifically heavy metals, can be found naturally in the basalt that was created from volcanic activity and be added to the environment from anthropogenic sources such as agricultural practices, military activity, and industrialization (Hinkley et al., 1999; Hunter et al., 1995). As apex predators, marine mammals can be used as biomonitors to indicate potential contaminant concentrations within the environment (Ikemoto et al., 2004). Due to its slow turnover rate and lipid content, bone acts as a concentrator of long-term pollutants, including heavy metals. Additionally, bones can be scavenged from a carcass and accessed in archived collections (Gdula-Argasinska et al., 2004). Hawaiian monk seals (HMS) are endemic to the Northwest Hawaiian Islands (NWHI) that are uninhabited and undeveloped, but the seals have recently established a colony within the Main Hawaiian Islands (MHI), also known as the State of Hawai'i which is developed and populated (Wilson et al., 2017). HMS stay within shallow waters, normally within 200 m depth, when foraging and then spend the rest of their time on land associated with their breeding colony (Littnan et al., 2017). The seals do not tend to travel to other colonies making them a good species for a regional contaminant study (Stewart et al., 2006).

## 1.2 Hawaiian Monk Seal

The Hawaiian monk seal (*Neomonachus schauinslandi*) is the only true tropical pinniped alive. Endemic to the Hawaiian Island chain, they are listed as not just an endangered organism, but the second most endangered pinniped on the planet, with an estimated population of 1,437 in 2021 (Johanos, 2021a, b, c). The original breeding range of the HMS consisted of the islands and atolls of the NWHI in the central Pacific Ocean, more than 3600 km, or 2000 mi, from the nearest continental land mass. The chain extends 1,200 miles from the Main Hawaiian Islands, also known as the State of Hawai'i. The HMS prefer to haul out onto sandy beaches versus high rocky islets; they can also be found on shelving reef rocks. Pupping areas are on permanent islands or islets away from the high tide line (Kenyon & Rice, 1959).

In the middle of the 19th century, sealing expeditions reduced the monk seal population to near extinction. In the late 19th century and early 20th century, more expeditions traveled to

the NWHI, including guano diggers, bird hunters, and whalers. These expeditions also contributed to the reduction of the HMS population (Kenyon & Rice, 1959). Since then, growth rate of the seals' population has increased by 2 % per year from 2013-2018 (Carretta et al., 2021).

The HMS currently has only nine breeding colonies, eight within the NWHI and one within the MHI. The colonies within the NWHI, from the farthest north to south, include Kure Atoll, Midway Atoll, Pearl and Hermes Reef, Lisianski Island, Laysan Island, French Frigate Shoal, Necker Island, and Nihoa Island (Figure 1). Nihoa and Necker islands are high volcanic islands while the remaining land outcrops are atolls, some with multiple sand islets (Kenyon & Rice, 1959). These islands and atolls are subsiding as sea level increases and storm events enhance the effects of subsiding. One example of storm effects is East Island which was part of French Frigate Shoal until Hurricane Walaka in October 2018 swamped the island. These types of storm events are also decreasing valuable habitat for the monk seals (Baker et al., 2020). The size of the NWHI colonies is decreasing 3.3% annually while the MHI colony is increasing 6.5% annually (Carretta et al., 2014; Carretta et al., 2015). The MHI subpopulation began forming approximately 20 years ago when a small group of seals slowly moved into the islands (Wilson et al., 2017).

Legal protection of the environment in the NWHI began in 1909 when then President Theodore Roosevelt established the Hawaiian Islands Reservation, renamed the Hawaiian Islands National Wildlife Refuge years later (Executive Order 1019, 1909; Presidential Proclamation 2416, 1940). The NWHI Coral Reef Ecosystem Reserve was established in 2000 by Executive Order 13178 (2000). This region become a designated National Marine Sanctuary in 2001 by Executive Order 13196 (2001). The State of Hawaii established the Northwestern Hawaiian Islands State Marine Refuge to also recognize the significance of the NWHI (DLNR, 2005). The current protected area name of the NWHI is the Papahānaumokuākea Marine National Monument created under the Antiquities Act of 1906 and issued as a Presidential Proclamation by President George W. Bush (American Antiquities Act of 1906; Department of Commerce, 2006; Presidential Proclamation 8031, 2006). The Monument is run by co-Trustees including the National Oceanic and Atmospheric Administration (NOAA), U.S. Fish and Wildlife Service (FWS), and the State of Hawaii (Memorandum of Agreement, 2006).

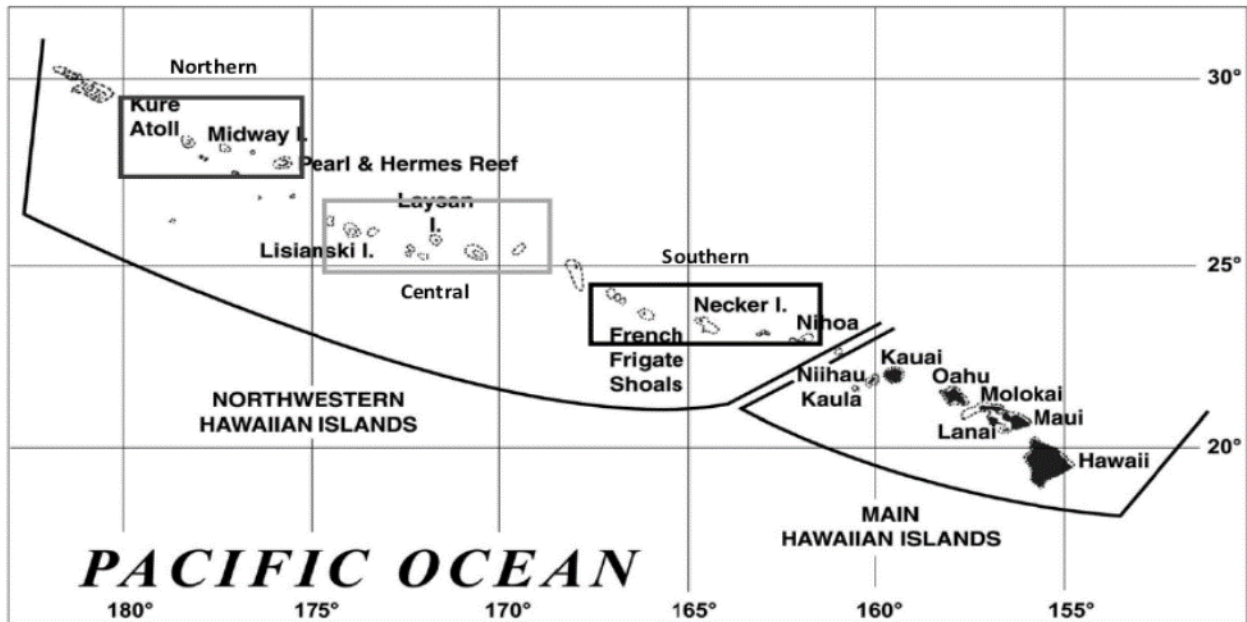


Figure 1. Hawaiian Island Archipelago separated into Northwest and Main Hawaiian Islands. The Northwest Hawaiian Islands are subdivided into three regions: Northern, Central, and Southern and designated by boxes (Polovina & Haight, 1999; Thompson, 2011).

Hawaiian monk seals are benthic foragers that spend most of their time diving and only come ashore for rest, molting, and pupping (Wilson et al., 2017). In the NWHI, the seals forage within the atoll lagoons or around the island of their associated colony (Stewart et al., 2006). While foraging, the seals search for mainly cryptic and solitary prey found in or under coral rubble or in high relief areas. While foraging for fish associated with the reef, HMS will dive down 18-90 m while close to land (Littnan et al., 2017). These habitats of coral rubble are dispersed in the MHI. The MHI seals have shorter foraging trips both in time and range compared to those within the NWHI. A HMS foraging trip in the MHI is typically short, lasting one-half to a whole day but longer trips of two to four days do occur. The home ranges of the HMS in the MHI varies by island. Molokai seals have fidelity to their island while Kauai seals might also go to nearby Niihau and back; Oahu has some faithful seals and some that travel to nearby islands (Wilson et al., 2017).

### **1.3 Prey of Hawaiian monk seal**

The HMS diet includes solitary, cryptic prey found near the sea floor. Their prey choice in the NWHI is largely separated into teleosts (78.6 %), cephalopods (15.7 %), and crustaceans (5.7 %). Of the teleost families identified, 30 out of 31 were reef-associated fishes. The most common teleost fish families of the diet were marine eels (Muraenidae, Congridae, and Ophichthidae) followed by Labridae (wrasses), Holocentridae (squirrelfishes), Balistidae (triggerfishes), and Scaridae (parrotfishes). No general differences in seal diets were observed throughout the extent of the NWHI, indicating the seals are generalist foragers and feeders (Goodman-Lowe, 1998). Cahoon et al. (2013) identified seven dominant fish families that comprised the majority of the diet within the MHI: Balistidae, Acanthuridae (surgeonfishes), Muraenidae (moray eels), Serranidae (groupers), Holocentridae (soldierfishes and squirrelfishes), Labridae, and Scaridae. Cephalopods comprise 18.3% of the seal diet within the MHI and this component is dominated by the two species *Octopus cyanea* and *O. ornatus*. MHI seals feed on less diversity in prey compared with that of NWHI seals (20 versus 31 families) but have significantly more diversity in prey choice (Cahoon et al., 2013; Carretta et al., 2021; Goodman-Lowe, 1998). Juvenile seals preferentially feed on small, slow-moving nocturnal teleosts and cephalopods while adults feed on larger diurnal or nocturnal prey (Goodman-Lowe, 1998; Iverson et al., 2011).



Acanthuridae (surgeonfish) fishes are named for their scalpel-like spine or spines that can be found on the sides of their caudal peduncle. Acanthurid fishes all have deep compressed bodies with eyes positioned high on the head and a single unnotched dorsal fin. There are 79 different species within the Pacific and Indian oceans, mainly reef associated. Their mouth is small and terminal with a single row of close-set teeth (Nelson, 1994). Surgeonfish can be algae grazers, feed on zooplankton, or detritivores (Randall, 2007).

Balistidae (triggerfish) come by their name from the first dorsal spine that will be locked in an erect position by the smaller second spine. Balistid fishes have a deep and moderately compressed body. These fish are found in the Atlantic, Indian, and Pacific oceans in shallow waters, mainly at coral reefs (Matsuura, 2015). Triggerfish are usually solitary fish. They will use holes in the reef to escape from threats and use the same hole as a place to sleep (Randall, 2007). Their mouth is small and, in a terminal, or almost terminal position with strong teeth (Matsuura, 2015). Most of the triggerfish are diurnal carnivores feeding on invertebrates such as crabs, mollusks, and sea urchins. Additional prey items could include zooplankton, benthic algae, or excrement of other fish, depending on the species (Randall, 2007).

Congidae (conger and garden eel) is a family characterized by having near-cylindrical bodies. Congidae has two subfamilies, the Congrinae (conger eels) and Heterocongrinae (garden eels) (Randall, 2007). These fish are found in the Atlantic, Indian, and Pacific oceans, mainly in deep or temperate waters. (Nelson, 1994). Conger eels are found in coral reefs or in rocky substrata. During the day they are normally hidden while at night they actively forage for crustaceans and sleeping reef fish. The garden eels are diurnal. They can generally be found in large colonies together but each in separate burrows in the sand. They will rise for their burrow to feed on zooplankton (Randall, 2007).

Holocentridae (squirrelfish and soldierfish) is a family of fish characterized by being red and having large eyes. These fish are found in the tropical Atlantic, Indian, and Pacific oceans in shallow waters with a depth from 0 to 100 m normal. The holocentrids are nocturnal and are cryptic during the day, lurking under ledges or in crevices of reefs (Nelson, 1994). There are two subfamilies in Holocentridae, Holocentrinae (squirrelfish) and Myripristinae (soldierfishes). The squirrelfish prey on benthic crustaceans while the soldierfish prey on larger zooplankton (Randall, 2007).

Labridae (wrasse) is the second largest marine fish family. This has led to the family being diverse with fish having a moderately deep body shape to a slender body and a short or long snout. These fish are found in the Atlantic, Indian, and Pacific oceans. They have a protrusible mouth with gaps between teeth that usually are jutting outward. Most of the species are sand burrowers (Nelson, 1994). All labrid fishes are carnivores. Their food choices do vary but most feed on invertebrates such as crabs, mollusks, sea urchins, and brittle stars. They use pharyngeal teeth to be able to crush their prey. Labrids are diurnal with smaller species buried in sand at night and large species sleeping deep within the reef (Randall, 2007).

Muraenidae (moray eel) is a family of eel characterized by very elongated, compressed bodies. They are found worldwide in tropical and temperate seas. These fish have a large mouth with numerous teeth, often having a canine shape (Nelson, 1994). The moray eels with long canines mainly prey on fish with the occasional crustacean or octopus. Moray eels with blunt teeth prey on crustaceans. Dentition can depend on size or potentially sex. Most species of moray eels stay hidden in the reef (Randall, 2007). These fish are benthic as adults, generally staying in shallow water among rocks and corals. Many of the species hide in holes or crevices during daytime leading them to be more active at night (Nelson, 1994).

Scaridae (parrotfish) are known for being bright in color and having fused teeth. These fish are chiefly tropical and found in the Atlantic, Indian, and Pacific oceans (Nelson, 1994). Parrotfish are mainly herbivores, feeding by scraping algae off rocks or dead coral. They are shallow-water fish primarily associated with coral reefs, but some species are found in seagrass beds or algal flats (Randall, 2007). Some species, at night, rest in a mucoid secreted cocoon (Nelson, 1994).

Serranidae (groupers) is a large, diverse family of fish. These fish are found in tropical and temperate oceans. The tip of their maxilla is exposed even when the mouth is closed. All the serranids are carnivores but can be small or large, up to 3 m in length (Nelson, 1994). Their size determines if they eat small prey like zooplankton or large prey like fish and crustaceans and the occasional cephalopod (Randall, 2007).

Palinuridae, or spiny lobsters, consists of species lacking massive pincers on the first set of legs. Their names derive from forward pointing spines on their carapace and antennae. The spiny lobsters can be found inhabiting crevices and caves. These lobsters are night feeders that forage on sandy bottoms adjacent to the reef (Hoover, 2006).

Octopodidae (octopus) is a family of octopuses that have eight sucker-lined tentacles and a beak for a mouth. These organisms have excellent sight and a complex nervous system. They are cryptic species with the ability to camouflage within their surrounds using specialized cells called chromatophores. They also have the ability to eject ink that is mixed with mucus as a form of escape from a predator. As a predator, octopuses feed on crabs, other mollusks, and fish (Hoover, 2006).

Commercial fishing in the NWHI has a long history with the extant fisheries beginning after World War II and landings reaching a peak in the 1960s and 1970s. Their target species were high-value pelagic fish (Pooley, 1993a; Schug, 2001). The bottom fish fishery started in 1945 and included groupers which are prey of the HMS. This deepwater fishing takes place at depths of 60-340 m using baited hook-and-line gear with powered mechanical line-haulers. Only one vessel operated in the NWHI until the 1980s. The palinurid spiny lobster fishery, a prey of the HMS, began in 1975 in the NWHI with an initial commercial catch of 2,000 kg but grew to an estimated 300,000 kg in 1981 (Polovina et al., 1982).

During the 1980s, the nearshore marine fisheries targeted bottom fish and lobsters but so did recreational and subsistence fishing (Pooley, 1993b). The National Marine Fisheries Service formed fishery management plans in accordance with the federal Fisheries Conservation and Management Act (known as the Magnuson Act, then the Magnuson-Stevens Act after reauthorization; Kittinger et al., 2010). From the 1980s to the 2000s, multiple fisheries were established, each with their own fishery management plans (Kittinger et al., 2010). A climatic event affecting the Subtropical Counter Current along the archipelago from 1977 to the early 1990s combined with commercial demand created a lobster population crash in 1989 (Polovina & Haight, 1999). Declines in HMS pup survival have been associated with declines in prey abundance during the late 1980s (e.g., Polovina et al., 1994). The lobster fishery closure occurred in 2000 when the NWHI Coral Reef Ecosystem Reserve was established (DiNardo & Marshall, 2001). With the closure of all the fisheries in Papahānaumokuākea National Marine Monument by 2010, the Ecosim model constructed by Parrish et al. (2012) anticipated an increase in prey availability to the monk seals.

#### **1.4 Environment**

The Hawaiian Island Chain is located in the central North Pacific Ocean. These islands were all formed at the same geologic hotspot as the Pacific tectonic plate moves to the northwest.

The age of the islands within the Hawaiian archipelago follows the plate's movement. The farthest northwest Hawaiian island, Kure, is estimated at 49 Ma while the farthest southeastern island of the NWHI, Nihoa Island, is estimated at 6 Ma. From there, the MHI date from 6 Ma (Niihau and Kauai) and to the continuously growing Island of Hawaii (Jicha et al., 2018). The distance from the Midway Atoll in the NWHI and the Island of Hawaii is approximately 1,600 miles (Tilling et al., 2010).

Island formation begins at a hotspot along the ocean floor, magma/lava building a seamount until it breaks the sea surface as a volcanic island. Erosion and subduction of the island landmass through time exposes the top of the island's fringing reef, forming a coral atoll. Stearns (1940) initially defined four stages of volcanism and then expanded it to 8 stages to incorporate the growth, erosion, submergence, reef development, and post erosion volcanism for the islands within the Hawaiian Island ridge and other islands found in the central Pacific Ocean (Figure 2). Stage 1 begins with magma eruption from the ocean floor to sea level. At this stage, the volcano is producing mainly pillow lava. The initial emergence of the mound will be poorly consolidated ash that will rapidly erode. That will last until the lava flow itself is subaerial, Stage 2 eruptions from rift zones and a central crater build the volcano with thin sheets of highly fluid, primitive olivine basalt that is compositionally uniform. Stage 3 yields the collapse around the vent, forming a caldera, and subsidence along the rift zones forming craters and grabens with uniform lava. Stage 4 starts when the lava composition changes from basalt to more alkali types. This lava is more viscous and fills any caldera, craters, and grabens. Stage 5 is the start of the island erosion process with stream and marine erosion dominating. The rate of erosion depends on the local climate, the prevailing winds, and ocean currents. Stage 6 is the start of overall subsidence and reef building. Stage 7 is when renewed volcanism may happen. Finally, stage 8 is when only an atoll remains, and subsidence of the land mass occurs. Mauna Loa and Kilauea on the Island of Hawaii are at stage 3 while the other volcanos on the island are at stage 4. The other islands within the MHI are at stage 5 while the NWHI are between stages 6 and 8 (Stearns, 1940; 1946; 1966).

Peterson and Moore (1987) proposed some changes to Stearns initial stages of volcanism for the evolution of the Hawaiian volcanos. For their initial stage, the lava is a variety of differentiated, alkali-type lavas. Stage 2 is the shield-building stage that produces tholeiitic basalt and tholeiitic picrite. The capping is the third stage where magma becomes differentiated and

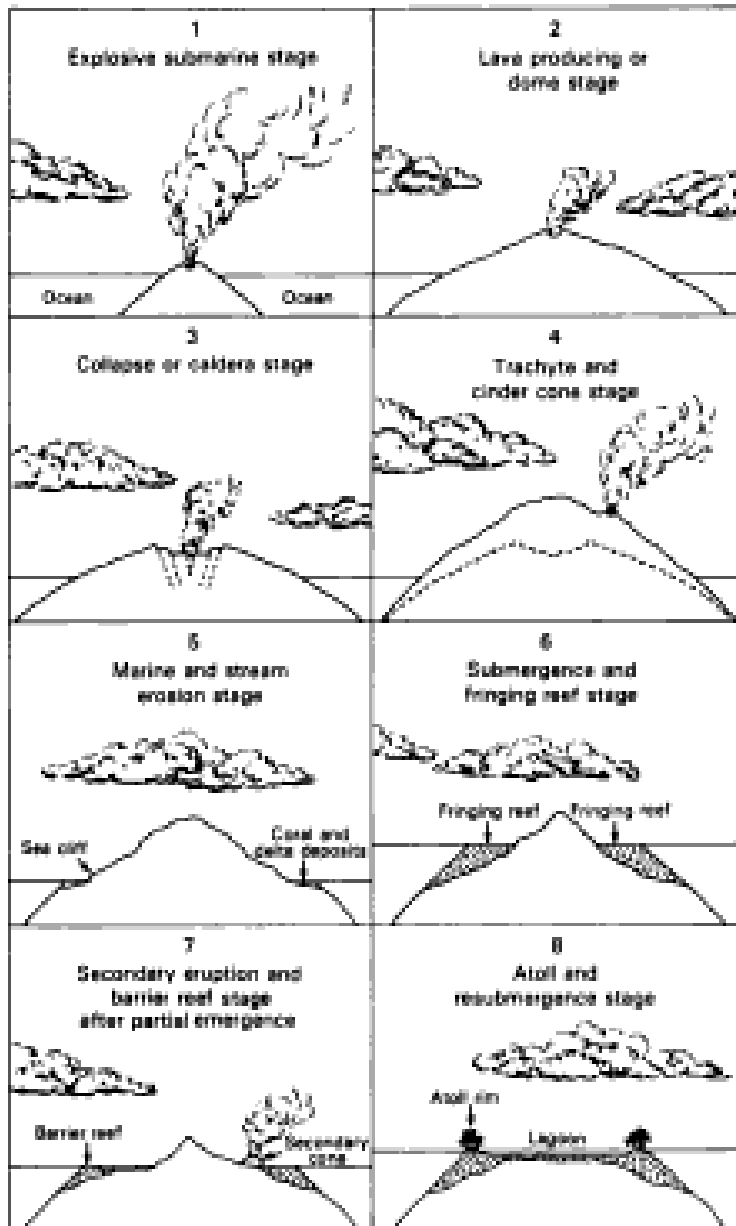


Figure 2. The eight stages of volcanism for oceanic islands in the central Pacific Ocean (Stearns, 1946)

yields alkali basalt and other alkalic rocks. Erosion starts in stage 4, named the erosional stage. Erosion begins in stages 2 and 3 but takes over during stage 4 as eruptions end. This cessation allows coral colonies to begin growth in the shallow water and build into larger reefs. Stage 5 is the renewed volcanism stage where, after thousands or millions of years, eruptions may intermittently occur. Stage 6 is when erosion has reduced the volcano to sea level, leaving the remaining fringing reef. The final stage is when the island subsides below sea level and is classified a guyot (Peterson & Moore, 1987).

The Island of Hawaii was created by five shield volcanos: Kohala, Mauna Kea, Huaalai, Mauna Loa, and Kilauea. The Kama'ehuakanaloa volcano, formally known as seamount Loihi, is actively growing over the hotspot 25-30 km south of the Island of Hawai'i (Peterson & Moore, 1987; USGS, 2022). Within the past 200 years Mauna Loa and Kilauea have remained active, in fact two of the most active volcanoes in the world. Mauna Loa's most recent eruptions were in 1975, 1984, and 2022. Kilauea has been continuously erupting since 1983 from Pu'u o'o - Kupaianaha and Halema'uma'u craters. The activity at the Hawaiian hotspot has increased in eruption rate within the last few centuries compared to the long-term arithmetic mean (Tilling et al., 2010; USGS, 2022).

Hawaii volcanoes are normally weakly explosive or nonexplosive. The less common, violent eruptions from the Hawaii volcanoes means that an ash cloud is unlikely to form. The nonexplosive eruption yields highly fluid magma and lava; the high fluidity is driven from the basaltic composition of the lava which is characterized by more iron (Fe), magnesium (Mg), calcium (Ca), and titanium (Ti) compared to viscous lava. The arithmetic mean chemical composition of lava from the Hawaii volcanoes is: 48.4 % Si, 13.2 % Al, 11.2 % Fe, 10.3 % Ca, 9.7 % Mg, 2.8 % Ti, 2.4 % Na, 0.6 % K, 1.4% other elements (Tilling et al., 2010).

Hawaiian volcanoes produce both tholeiitic and alkali basalts. The tholeiitic basalt from the rejuvenated stage has different element abundances compared to the shield tholeiitic basalt (Fodor et al., 1992). The NWHI have both tholeiitic and alkali basalts but, interestingly, only a few of the seamounts were found to have rejuvenated lavas. The few rejuvenated lavas could come from previous erosions or covered by reef growth (Garica et al., 2015).

The Hawaiian Island chain sits within the North Pacific gyre where ocean circulation is driven by the easterly trade winds (Lumpkin, 1998). The North Equatorial Current moves westward towards the islands and diverges to form the North Hawaiian Ridge Current that

travels northwesterly along the north side of the island chain and the Hawaii Lee Current that travels along the south side of the chain. The Hawaiian Lee Countercurrent flows eastward south of the islands (Chavanne et al., 2002). The Subtropical Countercurrent also flows easterly and bisects the middle of the NWHI (Toonen et al., 2011; Figure 3). The winds generate mesoscale eddies along the leeward side of the islands (Jia et al., 2011; Lumpkin, 1998; Patzert, 1969). The cyclonic, cold core eddies upwell nutrient-rich water as it moves northeasterly along the southern side of the islands (Seki et al., 2002). The anti-cyclonic, warm core eddies form off the Island of Hawaii moving southwest and away from the island chain (Bidigare et al., 2003). The eddies, therefore, are a form of water dispersal along the island chain as well as water retention in the area (Seki et al., 2002).

### **1.5 Heavy Metals**

Heavy metals are made up of all metal and metalloid type elements. They are normally found within the environment at relatively low (<0.1 %) concentrations. Concentrations of heavy metals can be found naturally and in man-made substances. Industrial waste products like sewage sludge contain sizeable quantities of heavy metals (Pais & Jones, 1997). As concentrations increase, they can pass the threshold to become toxic to the body (Baraj et al., 2009). Some elements (e.g., As, Cd, Hg, and Pb) are toxic even at low concentrations as they are non-essential and do not have any biological functions (Thompson, 1990). Examples of toxic effects can include immune toxic effects, genotoxic effects, cytotoxic effects, reproduction impairment, and endocrine alterations (Cardellicchio et al., 2000; Das et al., 2003).

Aluminum (Al) is the third most abundant element in the lithosphere and the most abundant trace element. The total content of Al in soils is 0.45-10%. The content of Al found in sea water is  $0.13 \times 10^{-4}$  to  $9.7 \times 10^{-4}$  mg/L while fresh water has 0.1-1200  $\mu\text{g/L}$ . Within a human bone, the content of Al is 4-27  $\mu\text{g/g}$  (Pais & Jones, 1997). Al is Nonessential for animals. For a human, the toxic intake is 5 g of Al. Most naturally occurring Al compounds are insoluble, leading to small amounts found in biological systems. Aluminum within a food web can transfer between animals by the calcium channels with the help of transferrin (Pais & Jones, 1997). Aluminum is used within foil, aluminum cookware, cans, ceramics, and fireworks (Baby et al., 2010).

Arsenic (As) was once widely used in pesticides, herbicides, and soil sterilants. The total content of As in soils is 0.1-48  $\mu\text{g/g}$  with a mean of 3.6-8.8  $\mu\text{g/g}$ . The average content of As

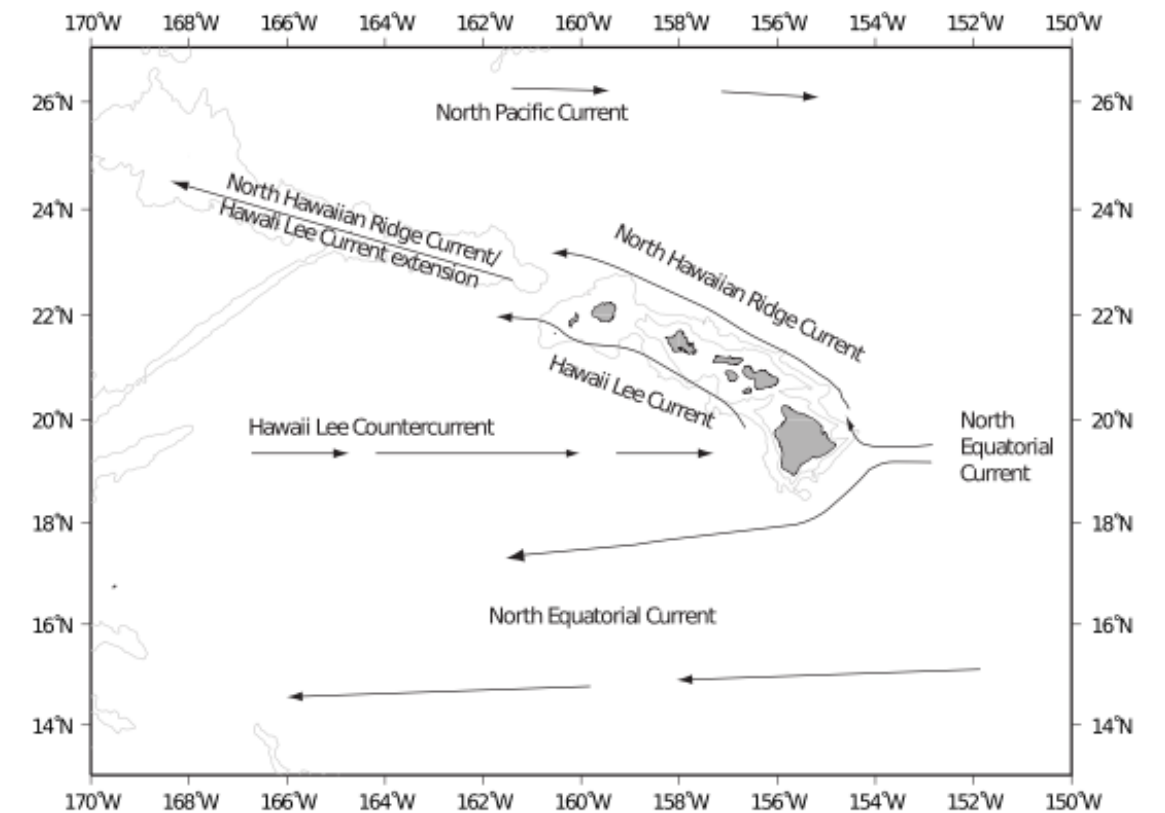


Figure 3. North Equatorial Current, North Hawaiian Ridge Current, Hawaii Lee Current, Hawaii Lee Countercurrent, and North Pacific Current surrounding the MHI and NWHI (Wren and Kobayashi, 2016).



found in sea water is  $1.45 \times 10^{-3} \mu\text{g/g}$  while the fresh water is content is around 0-0.5  $\mu\text{g/L}$  (Pais & Jones, 1997); As can be found worldwide within water supplies (Baby et al., 2010). Within a human bone, the mean content of As is 0.08-1.6  $\mu\text{g/g}$ . The toxic intake of As in humans is 5 to 50 mg/day while the lethal intake is 50 to 340 mg/day. The major source of As within the food web is from physical deposition or consumption. Arsenic is not expected to accumulate within the food web as it is not a highly mobile element. Its bioaccumulation index is moderate when compared to other trace elements (Pais & Jones, 1997). Arsenic is released by the smelting process of Cu, Pb, and Zn into the environment plus the manufacturing of chemicals and glasses. Other sources include wood preservatives, fungicides, paints, and rat poisoning (Baby et al., 2010).

Cadmium (Cd) is an element of concern because of the amount found in waste products, mainly sewage sludge. The total content of Cd in soils is 0.01-3.0  $\mu\text{g/g}$  (Pais & Jones, 1997). This can be due to Cd in insecticides, fungicides, sludge, and commercial fertilizers, all used in agriculture (Baby et al., 2010). The mean content of Cd found in sea water is  $1.1 \times 10^{-6}$  to  $38 \times 10^{-6}$  mg/L while fresh water is around 0.2  $\mu\text{g/L}$ . Within a human bone, the content of Cd is 1.8  $\mu\text{g/g}$ . Being nonessential for humans, the toxic intake is 30 to 330 mg while the lethal intake is 1.5-9g, and cadmium is known to accumulate in some environments (Pais & Jones, 1997). Sources of Cd include welding, electroplating, nuclear fission plants, and Cd and Ni batteries (Singh et al., 2011).

Chromium (Cr) occurs in multiple forms in the environment based on its valent state.  $\text{Cr}^{3+}$  is the stable form that is most commonly found in the environment and is also the essential form for some animal biological functions.  $\text{Cr}^{6+}$  is a toxic form to animals but is not common in the environment. The total content of Cr in soils is 5-1000  $\mu\text{g/g}$  with a mean of 65  $\mu\text{g/g}$ . The mean content of Cr found in sea water is 0.16  $\mu\text{g/L}$  while the fresh water is around 0.18  $\mu\text{g/L}$ . The content of Cr typically found in marine animals is 0.2-1.0  $\mu\text{g/g}$ . Within human bone, the content of Cr is 0.1-33  $\mu\text{g/g}$ . Being essential element for humans, the toxic intake is 200 mg while the lethal intake is greater than 3 g. Within the food web, chromium's bioaccumulation index is moderate (Pais & Jones, 1997). Sources of Cr include mines, mineral sources, paints, steel including stainless steel manufacturing, metal finishes, chrome and wood treatment, and alloy cast irons (Shrivastava et al., 2002; Singh et al., 2011).

Cobalt (Co) has geochemical characteristics similar to iron and manganese; its distribution is based on soil organic matter and clay content. Within soil, the parent material is the determining factor for the cobalt content. The total content of Co in soils is 1-40  $\mu\text{g/g}$ . The mean content of Co found in sea water is  $6.9 \times 10^{-6}$  mg/L while the fresh water is 0.01-0.18 mg/L. Within a human bone, the content of Co is 0.01-0.04  $\mu\text{g/g}$ . Being essential for a human, the toxic intake is 500 mg. Within the food web, cobalt is fairly mobile with a moderately high bioaccumulation index (Pais & Jones, 1997). Sources of Co include mining, rechargeable batteries, and turbine engines in jet aircrafts (Slack et al., 2017).

Copper (Cu) is, in general, immobile in soil but will become mobile in acidic soils. The distribution of Cu in soil is fairly uniform but can bioaccumulate at the surface. In areas where copper-containing chemicals are used (such as agricultural land), copper contamination is not uncommon. Contamination can also occur from industrial pollution and/or industrial waste and sewage sludge. The total content of Cu in soils is 2-100  $\mu\text{g/g}$  with a mean of 18  $\mu\text{g/g}$ . The mean content of Cu found in sea water is  $8.0 \times 10^{-5}$  mg/L while the fresh water is 0.01-2.8 mg/L. Within a human bone, the content of Cu is 1-26  $\mu\text{g/g}$ . Being essential for a human, the toxic intake is greater than 250 mg. Within the food web, copper's bioaccumulation index is high (Pais & Jones, 1997). Sources of Cu include metal piping, chemical industry, pesticide production, and mining (Singh et al., 2011).

Iron (Fe) is a major constituent of the lithosphere. The geochemistry of Fe is complex and dependent on its valence state. The total content of Fe in soils is 38  $\mu\text{g/g}$ . The mean content of Fe found in sea water is  $1 \times 10^{-4}$  to  $4 \times 10^{-4}$  mg/L while the fresh water is 0.04-6200 mg/L. The content of Fe typically found in marine animals is 400  $\mu\text{g/g}$ . Within a human bone, the content of Fe is 3-380  $\mu\text{g/g}$ . Being essential for a human, the toxic intake is 200 mg of Fe while lethal intake is 7-35 g. Within the food web, iron's bioaccumulation index is low (Pais & Jones, 1997). Within the oceans, Fe is important for phytoplankton growth as it is required for proteins involved in photosynthesis and respiration (Boyd et al., 2007; Martin & Fitzwater, 1988; Raven et al., 1999).

Lead (Pb) is a well-known major pollutant. Its main source into the atmosphere was lead-containing gasoline that allowed the Pb to be introduced into the food web upon settling onto plants and soils. The total content of Pb in soils is 3-189  $\mu\text{g/g}$  with a mean of 32  $\mu\text{g/g}$ . The mean content of Pb found in sea water is  $30 \times 10^{-6}$  mg/L at the surface and  $4.0 \times 10^{-6}$  mg/L in deep water. In fresh water, Pb content is 0.01-5.6 mg/L. Within a human bone, the content of Pb is 3.6-30

$\mu\text{g/g}$ . Being nonessential for a human, the toxic intake is 1 mg of Pb while lethal intake is 10 g. Within the food web, lead's bioaccumulation index is moderate with its movement based largely on anthropogenic activity (Pais & Jones, 1997). Lead is commonly used in pipes, drains, and soldering materials for years as well as paints used on homes. Additionally, it's used for batteries, fuel additives, PVC plastics, and pesticides (Baby et al., 2010).

Manganese (Mn) is among the most abundant trace elements found in the lithosphere. The total content of Mn in soils is 200-3000  $\mu\text{g/g}$  with a mean of 545  $\mu\text{g/g}$ . The mean content of Mn found in sea water is  $0.4 \cdot 10^{-4}$  to  $1.0 \cdot 10^{-4}$  mg/L while in fresh water, Mn content is 0.1-110 mg/L. The content of Mn typically found in marine animals is 1-60  $\mu\text{g/g}$  (lowest in fish). Within a human bone, the content of Mn is 0.2-100  $\mu\text{g/g}$ . Being essential for a rat, the toxic intake is 10 to 20 mg of Mn. Within the food web, manganese's bioaccumulation index is low (Pais & Jones, 1997). Sources of Mn include welding ferromanganese production and fuel addition (Singh et al., 2011).

Mercury (Hg) in the environment is mainly based on anthropogenic activity. The most toxic forms of Hg are volatile and methylated. The total content of Hg in soils is 0.1-1.0  $\mu\text{g/g}$  with a mean of 0.3  $\mu\text{g/g}$ . The mean content of Hg found in sea water is  $4.9 \cdot 10^{-7}$  mg/L while in fresh water Hg content is <0.1-6.0  $\mu\text{g/kg}$  (Pais & Jones, 1997). Naturally, Hg occurs in the environment from degassing of the earth's crust, and atmospheric Hg is dispersed globally through winds and rainfall (Baby et al., 2010). Within a human bone, the content of Hg is 0.04-1.04  $\mu\text{g/g}$ . Being nonessential for a human, the toxic intake is 0.4 mg of Hg while the lethal intake is 150 to 300 mg. Within the food web, mercury typically enters through atmospheric deposition from coal combustion, smelting, and volcanic activity (Pais & Jones, 1997). Additionally, mining, chloralkali plants, and paper industries produce significant Hg (Baby et al., 2010).

Molybdenum (Mo) is mainly found in the anionic form but still can be found in cationic form. Molybdenum is the only essential element (plant and animal) that is on the second line of the periodic table in the transition metals. The total content of Mo in soils is 0.5-40  $\mu\text{g/g}$  with a mean of 2  $\mu\text{g/g}$ . The mean content of Mo found in sea water is 0.01 mg/L while in fresh water, Mo content is 0.3  $\mu\text{g/L}$ . Within a human bone, the content of Mo is <0.7  $\mu\text{g/g}$ . Being essential for a rat, the toxic intake is 5 mg of Mo while the lethal intake is 50 mg. Within the food web, molybdenum's bioaccumulation index is moderately high (Pais & Jones, 1997). Sources of Mo

include steel production, catalysts, flame retardants, lubricants, corrosion inhibitors, smoke suppressant, fertilizers, pigments, and electroplating techniques (Anke, 2004).

Nickel (Ni) was once thought to be a toxic element as the concentration of it found in various food was higher than what was needed for living organisms. The total content of Ni in soils is 1-200  $\mu\text{g/g}$  with a mean of 20  $\mu\text{g/g}$ . The mean content of Ni found in sea water is 236 ng/L while in fresh water, Ni content is 10  $\mu\text{g/L}$ . Within a human bone, the content of Ni is  $<0.7 \mu\text{g/g}$ . Being essential for a rat, the toxic intake is 50 mg of Ni. Within the food web, nickel's bioaccumulation index is moderate (Pais & Jones, 1997). Sources of Ni include production of nickel steel, iron alloys, and electroplating (Harasim & Filipek, 2015).

Selenium (Se) has different oxidation forms that will determine its soil and plant chemistry. The total content of Se in soils is 0.1-2.0  $\mu\text{g/g}$ . The mean content of Se found in sea water is  $0.47 \cdot 10^{-7}$  to  $1.8 \cdot 10^{-7}$  mg/L while in fresh water, Se content is 0.6-20  $\mu\text{g/L}$ . The content of Se typically found in marine animals is 4-5  $\mu\text{g/g}$ . Within a human bone, the content of Se is 1-9  $\mu\text{g/g}$ . Being essential for a human, the toxic intake is 5 mg of Se. Within the food web, selenium's bioaccumulation index is moderate (Pais & Jones, 1997). Selenium has an antagonistic relationship with mercury, meaning it can detoxify mercury within a body (Palmisano et al., 1995). Sources of Se include hard coal and crude oil combustion and processing of elements such as Cu, P, Pb, U, Zn (Yudovich & Ketris, 2006).

Tin (Sn) is similar to iron and aluminum in that mobility in soil is dependent on pH. The total content of Sn in soils is 0.3-200  $\mu\text{g/g}$  with a mean of 1.1  $\mu\text{g/g}$ . The mean content of Sn found in sea water is 4 ng/L while in fresh water, Sn content is 0.3-17 ng/g. Within a human bone, the content of Sn is 1.4  $\mu\text{g/g}$ . Being nonessential for a human, the toxic intake is 2 g of Sn. Within the food web, the mobility is not known (Pais & Jones, 1997). Sources of tin includes pesticides, antifouling paints, tin dishes, plastic stabilizers (Rudel, 2003).

Vanadium (V) is found in varied amounts in rocks. The geochemical properties are dependent on the oxidation state and acidity of the media. The total content of V in soils is 3-230  $\mu\text{g/g}$  with a mean of 90  $\mu\text{g/g}$ . The mean content of V found in sea water is 2.4  $\mu\text{g/L}$  while in fresh water, V content is 0.3-20  $\mu\text{g/L}$ . The content of V typically found in marine animals is 1  $\mu\text{g/g}$  with high levels of accumulation in crustaceans, shellfish, and fish. Within a human bone, the content of V is 0.8-8.3 ng/g. Being nonessential for a human, V is not normally toxic if ingested but can be toxic if in the respiratory system. Within the food web, vanadium's

bioaccumulation index is very low (Pais & Jones, 1997). Sources of V include production of special steels, glass industry, temperature-resistant alloys, manufacturing paints and pigments, lining arc welding electrodes, air-craft construction, atomic energy industry, space technology, and burning fossil fuels (Pyrzynska & Wierzbicki, 2004).

Zinc (Zn) has a uniform distribution in soil and is easily absorbed by minerals and organic substances. The total content of Zn in soils is 10-300  $\mu\text{g/g}$ . The mean content of Zn found in sea water is  $0.5 \times 10^{-4}$  to  $1.0 \times 10^{-5}$  mg/L while in fresh water, Zn content is 0.1-240  $\mu\text{g/L}$ . The content of Zn typically found in marine animals is 6-1500  $\mu\text{g/g}$  with high levels of accumulation in Radiolaria and Mollusca. Within a human bone, the content of Zn is 75-170  $\mu\text{g/g}$ . Being essential for a human, the toxic intake is 150-600 mg of Zn while the lethal intake is 6 g. Within the food web, zinc's bioaccumulation index is high (Pais & Jones, 1997). Sources of Zn include refineries, plumbing, metal plating, and brass manufacture (Singh et al., 2011).

### **1.6 Heavy metals in Hawaiian Islands**

The largest natural input of heavy metal contaminants in Hawaii is from volcanic activity. The breakdown, or weathering, of the volcanic islands will also add to the environmental load (Hinkley et al., 1999). Table 1 illustrates the potential anthropogenic sources found in the Hawaiian Archipelago with different levels of impact to the environment (Hunter et al., 1995). Buat-Menard and Arnold (1978) found particulate Cd, Cu, Hg, Se, and Zn emitted by Mt. Etna (Italy) had amounts of heavy metal concentrations released comparable to that from anthropogenic sources within the Mediterranean area. In a soil study from the volcanic Fernando de Noronha archipelago in the Atlantic Ocean, concentrations of Ba, Cr, Cu, Ni, V, and Zn were higher on the archipelago than the concentrations of continental soils (Neta et al., 2018). A soil study from Réunion, a volcanic island in the Indian Ocean, revealed that high Cd concentrations were from agricultural practices, high Hg concentrations were from volcanic activity, and high Pb concentrations were from other anthropogenic inputs. The high soil concentrations of Cr, Cu, Ni, and Zn were from the natural pedo-geochemical background (Doelsch et al., 2006).

Active volcanic studies in Hawaii are only possible on the Island of Hawaii but all the islands have comparable base materials of basalt. Table 2 shows some concentrations of elements found within material related to volcanic activity. The important metals from the plume of Pu'u O'o are Pb, Cd, Cu, and Zn with an arithmetic mean abundance relationship of 1-part Pb to 3-parts Cd to 4 parts Cu to 30 parts Zn (Hinkley et al., 1999). Copper, Mn, P, Si, and Zn leach

Table 1. Sources of anthropogenic heavy metal contaminants within Hawaii soil.

<b>Element</b>	<b>Potential Anthropogenic Sources</b>	<b>Citation</b>
Ag	Photography Claims there's none	Hunter et al., 1995 HDOH, 2011
As	Pesticides Herbicides, fungicides, and rodenticides Wood preservative	Hunter et al., 1995 HDOH, 2011 Ali et al., 2013
Cd	Rubber Fossil fuel combustion, manure, and phosphate fertilizers Lubricating oils, diesel oils, tires, phosphate fertilizers, sewage sludge, insecticides, electroplating, batteries, coal and oil combustion, non-ferrous metal production, refuse incineration, iron, and steel manufacturing Paints and pigments, plastic stabilizers, and electroplating of cadmium containing plastics	Hunter et al., 1995 HDOH, 2011  Sutherland, 2000  Ali et al., 2013
Cr	Plating Tanneries, steel industries, fly ash, and wood preservatives	Hunter et al., 1995 Ali et al., 2013
Cu	Wood preservative Fertilizers and industrial emissions Metal plating, bearing and brushing wear, moving engine parts, brake-lining wear, fungicides and insecticides, antifoulants, corrosion of Cu plumbing, algacides, concrete and asphalt, rubber, phosphate fertilizers, and sewage sludge	Hunter et al., 1995 HDOH, 2011  Sutherland, 2000
Hg	Marine antifouling Health service Insecticides, fungicides, electrical equipment, paint, plastics, cosmetics, mildew-proofing paints, phosphate fertilizers, batteries, and fireworks Coal combustion	Raine et al., 1995 Hunter et al., 1995  Sutherland, 2000
Ni	Plating Diesel fuel and vehicle exhaust, lubricating oil, brushing wear, brake lining wear, asphalt paving, phosphate fertilizers, and storage batteries Industrial effluents, kitchen appliances, surgical instruments, steel alloys, and automobile batteries	Ali et al., 2013 Hunter et al., 1995  Sutherland, 2000
Pb	Batteries and gasoline Automobile exhaust, tire wear, lubricating oil and grease, bearing wear, brake linings, rubber, concrete, paint manufacturing, insecticides, phosphate fertilizers, and sewage sludge Battery manufacture and herbicides	Hunter et al., 1995  Sutherland, 2000 Ali et al., 2013
Se	Electrical and shampoo Pesticides	Hunter et al., 1995 HDOH, 2011

Zn	<p>Rubber and plating</p> <p>Sewage sludge, ash from incinerators, and industrial facilities</p> <p>Vulcanization of rubber and tire wear, motor oil, grease, batteries, galvanizing, air-conditioning ducts, pesticides, phosphate fertilizers, sewage sludges, transmission fluid, under coating, brake linings, asphalt, concrete, coal combustion, smelting operations, incineration, and wood combustion</p>	<p>Hunter et al., 1995</p> <p>HDOH, 2011</p> <p>Sutherland, 2000</p>
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Table 2. Heavy metal concentrations from different natural sources such as volcanic gas emissions, and basalt weathering within the Main Hawaiian Islands.

<b>Element</b>	<b>Proportion (based off Pb) of arithmetic mean emissions from Kilauea<sup>a</sup></b>	<b>Tholeiitic basalt, Kilauea (µg/g)<sup>b</sup></b>	<b>Tholeiitic basalt, Waianae range, Oahu (µg/g)<sup>b</sup></b>	<b>Alkalic olivine basalt, Waianae range, Oahu (µg/g)<sup>b</sup></b>
Ag	0.11	-	-	-
As	0.94	-	-	-
Bi	3.9	-	-	-
Cd	2.9	0.41	0.36	0.13
Cr	-	385	628	388
Co	-	-	-	-
Cu	4	126	156	111
Fe	-	-	-	-
Hg	-	0.067	0.017	0.039
In	0.094	-	-	-
Mn	-	-	-	-
Mo	-	-	-	-
Ni	-	181	401	237
Pb	1	5	4.4	4.9
Sb	0.16	-	-	-
Se	6.3	-	-	-
Sn	1.1	-	-	-
Te	1.1	-	-	-
Tl	0.45	-	-	-
V	-	-	-	-
Zn	33	135	99	145

<sup>a</sup>Hinkley et al., 1999

<sup>b</sup>McMurtry et al., 1995



slowly and uniformly during basalt weathering while Cr, Fe, Ni, Nb, Ti, V, and Zr are essentially immobile (Eggleton et al., 1987). De Carlo and Spencer (1995) reported that about 50% of Cu in near-surface sediments is from natural inputs in the MHI. The different land uses reflected the distribution and abundance of heavy metals. Barium, Co, Cr, and Ni were found to be controlled by natural inputs (De Carlo & Anthony, 2002).

Distinct differences segregate the remote NWHI from the developed islands that define much of the MHI. The human population of the MHI is approximately 1.4 million people while the NWHI is occupied periodically by less than 100 people (Carretta et al., 2019). The population in the MHI went from 1,211,537 in 2000 to 1,360,301 in 2010 with an increase of about 15,000 people per year (Tian, 2012). The U.S. military has had a presence in the Hawaiian Island chain since the start of the twentieth century which vastly increased after the start of World War II. In the NWHI, the U.S. Navy maintained a military base on Midway Atoll's Sand Island and Eastern Island, while French Frigate Shoal maintained a US Coast Guard unit. Tern Island, part of French Frigate Shoal, also had a U.S. Coast Guard Long Range Navigation station and was a refueling station for U. S. Navy planes transiting between the MHI and Midway. The U.S. Coast Guard relinquished control of their NWHI bases in 1979 (Kenyon & Rice, 1959; Miao et al., 2001).

Military activities and training include the use of heavy metals as additives, explosives, and ingredients in chemical weapons (Clausen et al., 2004). Ammunition contains As, Ni, Pb, and Sb (Rooney et al., 1999). When the Pb in bullets is low-quality, they may also contain Ag and Bi (Johnson et al., 2005). To improve ballistic properties, Cu and Zn covers are added to high velocity rounds. Tracer and incendiary bullets contain Ba, Sr, and Zn (Nelson, 1997). Robinson et al. (2008) found elevated Cu, Ni, Pb, and Sb concentrations in plant leaves surrounding a military shooting range while a study of soil on military sites in Latvia found elevated concentrations of Cd, Cr, Cu, and Pb (Kokorite et al., 2008). After the bombing of Pearl Harbor in December 1941, Cr, Cu, Pb, and Zn were found in increased concentrations in Oahu sediment (Ashwood & Olsen, 1988).

In harbors, the antifouling paint from boats and ships can add to heavy metal concentrations in the environment. An analysis done on constituents of paints (including antifouling) from U.S. Navy ships in Pearl Harbor found Cd, Cr, Cu, Ni, Pb, and Zn (Ashwood et al., 1989). They also reported that inactive fleet maintenance operations can add Cu, Pb, and Zn

to the environment at measurable amounts. Even small boats within a marina can leak Cu into the water (Hunter et al., 1995).

Numerous agricultural activities abound within the MHI, including the commercial propagation of taro, rice, pineapple, sugarcane, cattle grazing, and various horticulture, all assisted by large machinery and vehicles (Hunter et al., 1995). The agriculture, forestry, fishing, and hunting industries combined in the MHI earned \$400 million in 2010 (Tian, 2012). Many heavy metal contaminants used in agricultural practices are found in herbicides, pesticides, or insecticides. Arsenic-based herbicides were used from the 1913 to 1950 for weed control on Hawaiian sugar cane fields (Cutler et al., 2013). Mercury is found in mercury-containing fungicides used for agriculture. Rock phosphates and superphosphates contain high amounts of Cd and Zn as impurities (McMurtry et al., 1995).

Urban developmental sets the MHI distinctly apart from the NWHI. With a large tourist industry and a continuously growing population, many heavy metal inputs are related to urban development. The tourism industry in the MHI earned \$10.9 billion in 2010 while construction accounted for \$3.4 billion also in 2010 (Tian, 2012). Copper, Pb, and Zn were found in higher concentrations in soils in urban areas compared to conservation lands (De Carlo & Anthony, 2002). Arsenic, Cd, Cu, Pb, and Zn were all elements with anthropogenic inputs while V only slightly displayed anthropogenic contribution after examining soil and sediment from multiple locations around Oahu (De Carlo & Anthony, 2002). De Carlo and Spencer (1995) claim that one-half to two-thirds of Zn concentrations were from anthropogenic inputs while Cd may be 90% from anthropogenic sources. Copper concentrations are less from automobiles and more likely from other anthropogenic sources such as plumbing, gutter, or antifouling paints.

Contaminants from automobile and military transport use began in the 1920s and have logarithmically increased to today (Andrews & Sutherland, 2004). Hunter et al. (1995) noted elements from automobiles included Cr, Pb, and Zn while Sutherland (2000) also noted Ba, Cd, Cu, and Ni. Automobiles accounted for 97% of the Pb burden within the Oahu coastal sediments. Concentrations of Pb have decreased since lead-alkyl fuel additives in automobiles were restricted in 1975, while concentrations of Cd and Zn have increased, reflecting the increase of traffic on Oahu since the 1960s (De Carlo & Spencer, 1995).

Primary inputs of the heavy metals in Hawaii come from volcanic emissions, agricultural inputs (fertilizers and pesticides), vehicle emissions (gaseous and fluid), and vehicle-associated

wear. Military activities are also a source found in specific locations. Hawaii's position in the middle of the Pacific Ocean places it away from major industries such as coal combustion, non-ferrous metal production, and steel and iron manufacturing (Sutherland, 2000). Yet the North Pacific surface water concentrations of lead are 2-fold greater than the natural concentration of lead due to industrial emissions into the atmosphere (Flegal & Patterson, 1983).

Kaneohe Bay on Oahu had a 2000% increase in population from 1940 to 1990, making it a model for the impacts that urbanization has had on tropical Pacific ecosystems (Smith et al., 1973). The oyster tissue collected in the coastal waters had elevated Cr, Cu, Pb, and Zn concentrations (Hunter et al., 1995). Increased heavy metal concentrations were found in locations associated with urban or agricultural inputs; however, island weathering has not been shown to impact elemental concentrations (Bienfang et al., 2009). As increases in elemental concentrations are always possible, the Hawaii Department of Health has set limits where elemental concentrations start to become a concern (Table 3).

As these heavy elements are added into the environment, they make their way from the soil into the streams and then into the ocean. Cadmium, Cu, Hg, Pb, and Zn were all enriched in sediment at Pearl and Honolulu harbors (McMurtry et al., 1995). Within the watersheds of Oahu, Pb was the most enriched element, associated with high population densities, significant traffic densities, and commercial/manufacturing activities. The bed sediment collected below storm drain outlets was found to be higher in Cu, Pb, and Zn concentrations (Andrews & Sutherland, 2004). Fish in the Manoa Stream on Oahu showed high concentrations of Pb. Minor concentrations of Ba, Cd, Cu, Hg, and Zn, also found in the Manoa Stream, were connected to anthropogenic contamination (Sutherland, 2000). Heavy metal contamination was found to be a significant problem within the Nuuanu watershed (Oahu), especially for Cu, Pb, and Zn. These contaminants were linked to high traffic densities, high population densities, and dense commercial/manufacturing activities (Andrews & Sutherland, 2004).

### **1.7 Heavy Metal Concentrations in Marine Mammals**

Many marine mammals are apex predators and incorporated heavy metals biomagnify to the upper levels of the food web, making these animals excellent biomonitors (Ikemoto et al., 2004). Heavy metals can be taken in through water, ingestion of food, absorption through the skin, or respiration (Skoch, 1990). Multiple studies on marine mammals have incorporated variables such as tissue choice, species, age, and sex differences, and the results are varied. For

Table 3. The Hawaii Department of Health (HDOH) established the elemental action level based on potential concentrations ( $\mu\text{g/g}$ ) in island soil. Elemental action levels are used in the decision making during an environmental hazard evaluation (Hawaii Department of Health, 2012).

<b>Element</b>	<b>HDOH Element Action Level (<math>\mu\text{g/g}</math>)</b>
Antimony	8.2
Arsenic	23
Barium	15,000
Beryllium	160
Cadmium	70
Chromium	None set for total Cr, only Cr III and VI
Cobalt	23
Copper	3,100
Lead	200
Mercury	23
Molybdenum	390
Nickel	3,800
Selenium	390
Silver	390
Thallium	0.78
Vanadium	390
Zinc	23,000

example, positive correlations between age and Ag, Fe, Hg, Pb, Se, Sr, and V concentrations were found in the livers of striped dolphins. While O'Shea (1999) writes that a general acceptance exists that sex differences do not exist, Kooyomjian (2022) found otherwise in Peruvian otariid pinnipeds.

Heavy metal concentrations were generally low and non-acutely toxic in Mediterranean monk seal (*Monachus monachus*). However, there was still a potential for immune and endocrine impacts on the seals from elements like As, Cd, Cr, Ni, and Se. An increase in As was found as pups weaned and developed a juvenile diet of fish and cephalopods (Formigaro et al., 2017). Agusa et al. (2008) suggested that Cd is associated with an invertebrate-based diet in marine mammals while Hg is associated with a fish diet. Female seals generally have higher Hg levels than the males, potentially from females having hyperactive metabolism and the differences of reproduction plus targeted food intake (Formigaro et al., 2017). Concentrations of Cd and Fe were higher in female seals while males had higher Pb and Ni; Honda et al. (1986) related these differences to their excretion during parturition and lactation. In a mother-pup transfer study of harp seals, Hg transferred across the placenta while Cd did not. Copper and Se had a positive correlation between mother and pup, indicating a transfer of the metals (Wagemann et al., 1988). Baikal seals, Caspian seals, and northern fur seals all had an increase in Ag, Hg, Se, and V in liver tissues with an increase in age (Ikemoto et al., 2004). Selenium is essential for growth and reproduction plus has the capacity to detoxify metals like As, Cd, Cu, and Hg (Venugopal & Luckey, 1978). There is limited knowledge on actual toxic effects of heavy metals on seals. The best current practice is to compare the concentration levels to humans and terrestrial mammals normally for overt toxicity versus subchronic effects (Formigaro et al., 2017). Marine mammal heavy metal concentrations are represented in Table 4 and potential prey heavy metal concentrations are in Table 5.

Bones are useful to study long-term pollution changes as they are preserved during biogenesis (Cáceres-Saez et al., 2016; Lavery et al., 2009). Bones are deposit sites for essential and non-essential elements (Takata et al., 2005). Compact bones have a low turnover rate, meaning they reflect elemental accumulation over many years of an animal's life. This makes it a suitable long-term bioindicators of the animal's environmental exposure (Gdula-Argasinska et al., 2004). Large mammals, including marine mammals, have a turnover rate of approximately 10 years (Carvalho et al., 2004). Bone serves as a major reservoir for ingested trace elements;

Table 4. Heavy metal concentrations ( $\mu\text{g/g}$ ) in marine mammal species with bone type when available. Concentrations are reported for a single specimen for Yang et al. (2006) and Simokon & Trukhin (2021), mean for Agusa et al. (2011), mean  $\pm$  SD for Honda et al. (1986), Szteren & Auriolles-Gamboa (2013), De Maria et al. (2021), and Garcia-Garin et al. (2021), mean  $\pm$  CV for Hao et al. (2020), and geometric mean  $\pm$  95% CI for this study.

Element	Species	Bone Type	Concentration	Reference
Al	<i>Pontoporia blainvillei</i>	Maxillary bone	100.1 $\pm$ 279.1	Garcia-Garin et al., 2021
	<i>Zalophus californianus</i>	Tympanic bulla	62.73 $\pm$ 44.46	Szteren & Auriolles-Gamboa, 2013
	<i>Arctocephalus australis</i>	Teeth	29.6 $\pm$ 42.6	De Maria et al., 2021
	<b><i>Neomonachus schauinslandi</i></b>	<b>Skull</b>	<b>3.19 <math>\pm</math> 2.27</b>	<b>This study</b>
As	<i>Pontoporia blainvillei</i>	Maxillary bone	0.7 $\pm$ 1.2	Garcia-Garin et al., 2021
	<i>Zalophus californianus</i>	Tympanic bulla	16.48 $\pm$ 39.92	Szteren & Auriolles-Gamboa, 2013
	<i>Neophocaena asaeorientalis</i>	3rd/4th Rib	6.7 $\pm$ 0.72	Hao et al., 2020
	<b><i>Neomonachus schauinslandi</i></b>	<b>Skull</b>	<b>0.0377 <math>\pm</math> 0.0263</b>	<b>This study</b>
Cd	<i>Pontoporia blainvillei</i>	Maxillary bone	0.0 $\pm$ 0.2	Garcia-Garin et al., 2021
	<i>Zalophus californianus</i>	Tympanic bulla	2.59 $\pm$ 1.84	Szteren & Auriolles-Gamboa, 2013
	<i>Neophocaena asaeorientalis</i>	3rd/4th Rib	4.3 $\pm$ 0.38	Hao et al., 2020
	<b><i>Neomonachus schauinslandi</i></b>	<b>Skull</b>	<b>0.0417 <math>\pm</math> 0.0784</b>	<b>This study</b>
Co	<i>Zalophus californianus</i>	Tympanic bulla	30.48 $\pm$ 35.93	Szteren & Auriolles-Gamboa, 2013
	<i>Phoca vitulina</i>	Bone	0.094	Agusa et al., 2011
	<i>Phoca largha</i>	Bone	0.017	Simokon & Trukhin, 2021
	<b><i>Neomonachus schauinslandi</i></b>	<b>Skull</b>	<b>0.00833 <math>\pm</math> 0.00240</b>	<b>This study</b>

Cr	<i>Pontoporia blainvillei</i>	Maxillary bone	1.2 ± 2.3	Garcia-Garin et al., 2021
	<i>Neophocaena asaeorientalis</i>	3rd/4th Rib	0.56 ± 0.17	Hao et al., 2020
	<i>Arctocephalus australis</i>	Teeth	0.17 ± 0.17	De Maria et al., 2021
	<b><i>Neomonachus schauinslandi</i></b>	<b>Skull</b>	<b>0.234 ± 0.0908</b>	<b>This study</b>
Cu	<i>Pontoporia blainvillei</i>	Maxillary bone	4.4 ± 13.3	Garcia-Garin et al., 2021
	<i>Zalophus californianus</i>	Tympanic bulla	4.03 ± 2.29	Szteren & Auriolles-Gamboa, 2013
	<i>Neophocaena asaeorientalis</i>	3rd/4th Rib	7.9 ± 0.42	Hao et al., 2020
	<b><i>Neomonachus schauinslandi</i></b>	<b>Skull</b>	<b>0.580 ± 5.076</b>	<b>This study</b>
Fe	<i>Pontoporia blainvillei</i>	Maxillary bone	130.5 ± 373.4	Garcia-Garin et al., 2021
	<i>Zalophus californianus</i>	Tympanic bulla	70.51 ± 68.40	Szteren & Auriolles-Gamboa, 2013
	<i>Neophocaena asaeorientalis</i>	3rd/4th Rib	247.6 ± 11.32	Hao et al., 2020
	<b><i>Neomonachus schauinslandi</i></b>	<b>Skull</b>	<b>81.7 ± 16.0</b>	<b>This study</b>
Hg	<i>Pontoporia blainvillei</i>	Maxillary bone	0.1 ± 0.1	Garcia-Garin et al., 2021
	<i>Zalophus californianus</i>	Tympanic bulla	0.04 ± 0.04	Szteren & Auriolles-Gamboa, 2013
	<i>Neophocaena asaeorientalis</i>	3rd/4th Rib	0.056 ± 0.013	Hao et al., 2020
	<b><i>Neomonachus schauinslandi</i></b>	<b>Skull</b>	<b>0.0230 ± 0.148</b>	<b>This study</b>
Mn	<i>Pontoporia blainvillei</i>	Maxillary bone	32.8 ± 265.4	Garcia-Garin et al., 2021
	<i>Neophocaena asaeorientalis</i>	3 <sup>rd</sup> /4 <sup>th</sup> Rib bone	10.7 ± 0.43	Hao et al., 2020
	<i>Stenella coeruleoalba</i>	10 <sup>th</sup> Dorsal	0.82 ± 0.22	Honda et al., 1986
	<b><i>Neomonachus schauinslandi</i></b>	<b>Skull</b>	<b>0.792 ± 0.0851</b>	<b>This study</b>

Mo	<i>Phoca vitulina</i>	Bone	0.061	Agusa et al., 2011
	<i>Phoca largha</i>	Bone	0.034	Simokon & Trukhin, 2021
	<i>Phocoenoides dalli</i>	Bone	0.01	Yang et al., 2006
	<b><i>Neomonachus schauinslandi</i></b>	<b>Skull</b>	<b>0.0282 ± 0.00702</b>	<b>This study</b>
Ni	<i>Pontoporia blainvillei</i>	Maxillary bone	1.0 ± 2.3	Garcia-Garin et al., 2021
	<i>Zalophus californianus</i>	Maxillary bone	45.33 ± 25.26	Szteren & Auriolles-Gamboa, 2013
	<i>Neophocaena asaeorientalis</i>	3 <sup>rd</sup> /4 <sup>th</sup> Rib bone	1.12 ± 0.14	Hao et al., 2020
	<b><i>Neomonachus schauinslandi</i></b>	<b>Skull</b>	<b>0.155 ± 0.0837</b>	<b>This study</b>
Pb	<i>Pontoporia blainvillei</i>	Maxillary bone	1.6 ± 3.5	Garcia-Garin et al., 2021
	<i>Zalophus californianus</i>	Tympanic bulla	25.67 ± 12.87	Szteren & Auriolles-Gamboa, 2013
	<i>Neophocaena asaeorientalis</i>	3 <sup>rd</sup> /4 <sup>th</sup> Rib bone	1.87 ± 0.11	Hao et al., 2020
	<b><i>Neomonachus schauinslandi</i></b>	<b>Skull</b>	<b>0.131 ± 0.287</b>	<b>This study</b>
Se	<i>Pontoporia blainvillei</i>	Maxillary bone	0.0 ± 0.2	Garcia-Garin et al., 2021
	<i>Zalophus californianus</i>	Tympanic bulla	48.74 ± 116.82	Szteren & Auriolles-Gamboa, 2013
	<i>Stenella coeruleoalba</i>	10 <sup>th</sup> Dorsal	0.75 ± 0.08	Honda et al., 1986
	<b><i>Neomonachus schauinslandi</i></b>	<b>Skull</b>	<b>0.167 ± 0.0254</b>	<b>This study</b>
Sn	<i>Phoca vitulina</i>	Bone	0.104	Agusa et al., 2011
	<b><i>Neomonachus schauinslandi</i></b>	<b>Skull</b>	<b>1.48 ± 0.0911</b>	<b>This study</b>
V	<i>Phoca vitulina</i>	Bone	0.046	Agusa et al., 2011
	<i>Phoca largha</i>	Bone	0.038	Simokon & Trukhin, 2021



	<i>Phocoenoides dalli</i>	Bone	0.26	Yang et al., 2006
	<b><i>Neomonachus schauinslandi</i></b>	<b>Skull</b>	<b>0.0580 ± 0.0145</b>	<b>This study</b>
Zn	<i>Pontoporia blainvillei</i>	Maxillary bone	251.2 ± 74.4	Garcia-Garin et al., 2021
	<i>Zalophus californianus</i>	Tympanic bulla	44.17 ± 38.65	Szteren & Aurioles-Gamboa, 2013
	<i>Stenella coeruleoalba</i>	10 <sup>th</sup> Dorsal	382.0 ± 23.9	Honda et al., 1986
	<b><i>Neomonachus schauinslandi</i></b>	<b>Skull</b>	<b>102 ± 6.30</b>	<b>This study</b>

Table 5. Heavy metal concentrations ( $\mu\text{g/g}$ ) in tissues of potential prey (family and species). Concentrations are reported for single specimen for Miao et al. (2001), single specimen and mean  $\pm$  SD for Metian et al. (2013), mean  $\pm$  SD for Briand et al. (2018), and geometric mean  $\pm$  95% CI for this study.

Element	Family	Species	Tissue Type	Concentration ( $\mu\text{g/g}$ )	Reference
As	Acanthuridae	<i>Acanthurus triostegus</i>	Whole	24	Miao et al., 2001
	Acanthuridae	<i>Naso unicornis</i>	Liver	10.2	Metian et al., 2013
	<b>Acanthuridae</b>	-	<b>Whole</b>	<b>3.38<math>\pm</math>2.83</b>	<b>This study</b>
	Congidae	<i>Conger cinereus</i>	Whole	56	Miao et al., 2001
	Congidae	<i>Conger laticauda laticudata</i>	Muscle	31.71 $\pm$ 26.67	Briand et al., 2018
	<b>Congidae</b>	-	<b>Whole</b>	<b>25.7<math>\pm</math>4.27</b>	<b>This study</b>
	Labridae	<i>Bodianus perditio</i>	Liver	43.8	Metian et al., 2013
	Labridae	<i>Cheilinus chlorourus</i>	Liver	10.4	Metian et al., 2013
	<b>Labridae</b>	-	<b>Whole</b>	<b>14.3<math>\pm</math>7.50</b>	<b>This study</b>
	Muraenidae	<i>Gymnothorax flavimarginatus</i>	Whole	81	Miao et al., 2001
	Muraenidae	<i>Gymnothorax undulatus</i>	Muscle	23.27 $\pm$ 12.21	Briand et al., 2018
	<b>Muraenidae</b>	-	<b>Whole</b>	<b>8.71<math>\pm</math>13.5</b>	<b>This study</b>
	Palinuridae	<i>Panulirus marginatus</i>	Whole	116	Miao et al., 2001
	<b>Palinuridae</b>	-	<b>Whole</b>	<b>101<math>\pm</math>22.9</b>	<b>This study</b>
	Scaridae	<i>Scarus ghobban</i>	Liver	<9.41	Metian et al., 2013
	Scaridae	<i>Scarus microrhinos</i>	Liver	<7.74	Metian et al., 2013
	<b>Scaridae</b>	-	<b>Whole</b>	<b>2.62<math>\pm</math>0.467</b>	<b>This study</b>
	Serranidae	<i>Epinephelus coeruleopunctatus</i>	Liver	<6.31	Metian et al., 2013
Serranidae	<i>Plectropomus leopardus</i>	Liver	<8.10	Metian et al., 2013	
<b>Serranidae</b>	-	<b>Whole</b>	<b>4.49<math>\pm</math>2.68</b>	<b>This study</b>	
Cd	Acanthuridae	<i>Acanthurus triostegus</i>	Whole	2.7	Miao et al., 2001
	Acanthuridae	<i>Naso unicornis</i>	Liver	8.92	Metian et al., 2013
	<b>Acanthuridae</b>	-	<b>Whole</b>	<b>0.376<math>\pm</math>0.941</b>	<b>This study</b>
	Congidae	<i>Conger cinereus</i>	Whole	3	Miao et al., 2001

	Congidae	<i>Conger Laticauda laticudata</i>	Muscle	0.034±0.04	Briand et al., 2018
	<b>Congidae</b>	-	<b>Whole</b>	<b>0.151±0.102</b>	<b>This study</b>
	Labridae	<i>Bodianus perditio</i>	Liver	0.25	Metian et al., 2013
	Labridae	<i>Cheilinus chlorourus</i>	Liver	3.39	Metian et al., 2013
	<b>Labridae</b>	-	<b>Whole</b>	<b>0.181±0.0933</b>	<b>This study</b>
	Muraenidae	<i>Gymnothorax flavimarginatus</i>	Whole	4	Miao et al., 2001
	Muraenidae	<i>Gymnothorax undulatus</i>	Muscle	<dl	Briand et al., 2018
	<b>Muraenidae</b>	-	<b>Whole</b>	<b>0.593±0.465</b>	<b>This study</b>
	Palinuridae	<i>Panulirus marginatus</i>	Whole	6	Miao et al., 2001
	<b>Palinuridae</b>	-	<b>Whole</b>	<b>3.58±2.48</b>	<b>This study</b>
	Scaridae	<i>Scarus ghobban</i>	Liver	0.06	Metian et al., 2013
	Scaridae	<i>Scarus microrhinos</i>	Liver	0.88	Metian et al., 2013
	<b>Scaridae</b>	-	<b>Whole</b>	<b>0.379±0.133</b>	<b>This study</b>
	Serranidae	<i>Epinephelus coeruleopunctatus</i>	Liver	0.18	Metian et al., 2013
	Serranidae	<i>Plectropomus leopardus</i>	Liver	0.24	Metian et al., 2013
	<b>Serranidae</b>	-	<b>Whole</b>	<b>0.0741±0.0938</b>	<b>This study</b>
Co	Acanthuridae	<i>Naso unicornis</i>	Liver	5.62	Metian et al., 2013
	<b>Acanthuridae</b>	-	<b>Whole</b>	<b>0.0562±0.0207</b>	<b>This study</b>
	Congidae	<i>Conger Laticauda laticudata</i>	Muscle	0.14±0.14	Briand et al., 2018
	<b>Congidae</b>	-	<b>Whole</b>	<b>0.0234±0.00269</b>	<b>This study</b>
	Labridae	<i>Bodianus perditio</i>	Liver	0.8	Metian et al., 2013
	Labridae	<i>Cheilinus chlorourus</i>	Liver	6.25	Metian et al., 2013
	<b>Labridae</b>	-	<b>Whole</b>	<b>0.0283±0.00821</b>	<b>This study</b>
	Muraenidae	<i>Gymnothorax undulatus</i>	Muscle	0.05±0.01	Briand et al., 2018
	<b>Muraenidae</b>	-	<b>Whole</b>	<b>0.0193±0.00742</b>	<b>This study</b>
	Scaridae	<i>Scarus ghobban</i>	Liver	1.24	Metian et al., 2013
	Scaridae	<i>Scarus microrhinos</i>	Liver	1.34	Metian et al., 2013
	<b>Scaridae</b>	-	<b>Whole</b>	<b>0.0446±0.0151</b>	<b>This study</b>

	Serranidae	<i>Epinephelus coeruleopunctatus</i>	Liver	0.42	Metian et al., 2013
	Serranidae	<i>Plectropomus leopardus</i>	Liver	1.18	Metian et al., 2013
Cr	<b>Serranidae</b>	-	<b>Whole</b>	<b>0.0185±0.0205</b>	<b>This study</b>
	Acanthuridae	<i>Acanthurus triostegus</i>	Whole	8.2	Miao et al., 2001
	Acanthuridae	<i>Naso unicornis</i>	Liver	0.82	Metian et al., 2013
	<b>Acanthuridae</b>	-	<b>Whole</b>	<b>2.31±0.730</b>	<b>This study</b>
	Congidae	<i>Conger cinereus</i>	Whole	5	Miao et al., 2001
	Congidae	<i>Conger Laticauda laticudata</i>	Muscle	4.75±5.91	Briand et al., 2018
	<b>Congidae</b>	-	<b>Whole</b>	<b>2.08±0.551</b>	<b>This study</b>
	Labridae	<i>Bodianus perditio</i>	Liver	0.83	Metian et al., 2013
	Labridae	<i>Cheilinus chlorourus</i>	Liver	0.80	Metian et al., 2013
	<b>Labridae</b>	-	<b>Whole</b>	<b>2.70±1.17</b>	<b>This study</b>
Muraenidae	<i>Gymnothorax flavimarginatus</i>	Whole	5	Miao et al., 2001	
Muraenidae	<i>Gymnothorax undulatus</i>	Muscle	0.56±0.09	Briand et al., 2018	
<b>Muraenidae</b>	-	<b>Whole</b>	<b>1.14±0.234</b>	<b>This study</b>	
Palinuridae	<i>Panulirus marginatus</i>	Whole	5	Miao et al., 2001	
<b>Palinuridae</b>	-	<b>Whole</b>	<b>1.51±0.550</b>	<b>This study</b>	
Scaridae	<i>Scarus ghobban</i>	Liver	0.94	Metian et al., 2013	
Scaridae	<i>Scarus microrhinos</i>	Liver	<0.77	Metian et al., 2013	
<b>Scaridae</b>	-	<b>Whole</b>	<b>2.08±0.670</b>	<b>This study</b>	
Serranidae	<i>Epinephelus coeruleopunctatus</i>	Liver	<0.63	Metian et al., 2013	
Serranidae	<i>Plectropomus leopardus</i>	Liver	0.81	Metian et al., 2013	
<b>Serranidae</b>	-	<b>Whole</b>	<b>0.494±0.200</b>	<b>This study</b>	
Cu	Acanthuridae	<i>Acanthurus triostegus</i>	Whole	80.7	Miao et al., 2001
	Acanthuridae	<i>Naso unicornis</i>	Liver	9.44	Metian et al., 2013
	<b>Acanthuridae</b>	-	<b>Whole</b>	<b>1.72±0.464</b>	<b>This study</b>
	Congidae	<i>Conger cinereus</i>	Whole	22	Miao et al., 2001
	Congidae	<i>Conger Laticauda</i>	Muscle	1.88±0.93	Briand et al., 2018

		<i>laticudata</i>			
	<b>Congidae</b>	-	<b>Whole</b>	<b>3.76±5.62</b>	<b>This study</b>
	Labridae	<i>Bodianus perditio</i>	Liver	8.78	Metian et al., 2013
	Labridae	<i>Cheilinus chlorourus</i>	Liver	53.0	Metian et al., 2013
	<b>Labridae</b>	-	<b>Whole</b>	<b>1.38±0.255</b>	<b>This study</b>
	Muraenidae	<i>Gymnothorax</i>	Whole	14	Miao et al., 2001
		<i>flavimarginatus</i>			
	Muraenidae	<i>Gymnothorax undulatus</i>	Muscle	1.02±0.48	Briand et al., 2018
	<b>Muraenidae</b>	-	<b>Whole</b>	<b>3.05±2.16</b>	<b>This study</b>
	Palinuridae	<i>Panulirus marginatus</i>	Whole	110	Miao et al., 2001
	<b>Palinuridae</b>	-	<b>Whole</b>	<b>97.9±13.4</b>	<b>This study</b>
	Scaridae	<i>Scarus ghobban</i>	Liver	1.51	Metian et al., 2013
	Scaridae	<i>Scarus microrhinos</i>	Liver	1.11	Metian et al., 2013
	<b>Scaridae</b>	-	<b>Whole</b>	<b>1.98±0.505</b>	<b>This study</b>
	Serranidae	<i>Epinephelus</i>	Liver	55.7	Metian et al., 2013
		<i>coeruleopunctatus</i>			
	Serranidae	<i>Plectropomus leopardus</i>	Liver	111±27.0	Metian et al., 2013
	<b>Serranidae</b>	-	<b>Whole</b>	<b>0.912±0.412</b>	<b>This study</b>
Fe	Acanthuridae	<i>Naso unicornis</i>	Liver	4620	Metian et al., 2013
	<b>Acanthuridae</b>	-	<b>Whole</b>	<b>92.9±192</b>	<b>This study</b>
	Congidae	<i>Conger Laticauda</i>	Muscle	38±35	Briand et al., 2018
		<i>laticudata</i>			
	<b>Congidae</b>	-	<b>Whole</b>	<b>88.2±18.1</b>	<b>This study</b>
	Labridae	<i>Bodianus perditio</i>	Liver	1600	Metian et al., 2013
	Labridae	<i>Cheilinus chlorourus</i>	Liver	7080	Metian et al., 2013
	<b>Labridae</b>	-	<b>Whole</b>	<b>77.6±17.6</b>	<b>This study</b>
	Muraenidae	<i>Gymnothorax undulatus</i>	Muscle	7.2±0.5	Briand et al., 2018
	<b>Muraenidae</b>	-	<b>Whole</b>	<b>53.0±16.7</b>	<b>This study</b>
	Scaridae	<i>Scarus ghobban</i>	Liver	250	Metian et al., 2013
	Scaridae	<i>Scarus microrhinos</i>	Liver	240	Metian et al., 2013
	<b>Scaridae</b>	-	<b>Whole</b>	<b>86.5±158</b>	<b>This study</b>
	Serranidae	<i>Epinephelus</i>	Liver	2240	Metian et al., 2013

		<i>coeruleopunctatus</i>			
	Serranidae	<i>Plectropomus leopardus</i>	Liver	2500±350	Metian et al., 2013
	<b>Serranidae</b>	-	<b>Whole</b>	<b>38.0±17.4</b>	<b>This study</b>
Hg	Acanthuridae	<i>Naso unicornis</i>	Liver	0.31	Metian et al., 2013
	<b>Acanthuridae</b>	-	<b>Whole</b>	<b>0.0293±0.0222</b>	<b>This study</b>
	Congidae	<i>Conger Laticauda laticudata</i>	Muscle	0.095±0.06	Briand et al., 2018
	<b>Congidae</b>	-	<b>Whole</b>	<b>0.367±0.0437</b>	<b>This study</b>
	Labridae	<i>Bodianus perditio</i>	Liver	5.04	Metian et al., 2013
	Labridae	<i>Cheilinus chlorourus</i>	Liver	2.69	Metian et al., 2013
	<b>Labridae</b>	-	<b>Whole</b>	<b>0.0471±0.0132</b>	<b>This study</b>
	Muraenidae	<i>Gymnothorax flavimarginatus</i>	Whole	0.34	Miao et al., 2001
	Muraenidae	<i>Gymnothorax undulatus</i>	Muscle	0.161±0.019	Briand et al., 2018
	<b>Muraenidae</b>	-	<b>Whole</b>	<b>0.550±0.362</b>	<b>This study</b>
	Scaridae	<i>Scarus ghobban</i>	Liver	0.06	Metian et al., 2013
	Scaridae	<i>Scarus microrhinos</i>	Liver	0.03	Metian et al., 2013
	<b>Scaridae</b>	-	<b>Whole</b>	<b>0.0190±0.00437</b>	<b>This study</b>
	Serranidae	<i>Epinephelus coeruleopunctatus</i>	Liver	0.88	Metian et al., 2013
	Serranidae	<i>Plectropomus leopardus</i>	Liver	5.25±0.73	Metian et al., 2013
	<b>Serranidae</b>	-	<b>Whole</b>	<b>0.104±0.349</b>	<b>This study</b>
Mn	Acanthuridae	<i>Naso unicornis</i>	Liver	3.92	Metian et al., 2013
	<b>Acanthuridae</b>	-	<b>Whole</b>	<b>2.56±0.716</b>	<b>This study</b>
	Congidae	<i>Conger Laticauda laticudata</i>	Muscle	2.42±1.89	Briand et al., 2018
	<b>Congidae</b>	-	<b>Whole</b>	<b>2.46±0.686</b>	<b>This study</b>
	Labridae	<i>Bodianus perditio</i>	Liver	2.42	Metian et al., 2013
	Labridae	<i>Cheilinus chlorourus</i>	Liver	4.27	Metian et al., 2013
	<b>Labridae</b>	-	<b>Whole</b>	<b>5.09±2.03</b>	<b>This study</b>
	Muraenidae	<i>Gymnothorax undulatus</i>	Muscle	3.69±0.76	Briand et al., 2018
	<b>Muraenidae</b>	-	<b>Whole</b>	<b>9.98±14.2</b>	<b>This study</b>

	Scaridae	<i>Scarus ghobban</i>	Liver	2.63	Metian et al., 2013
	Scaridae	<i>Scarus microrhinos</i>	Liver	0.93	Metian et al., 2013
	<b>Scaridae</b>	-	<b>Whole</b>	<b>5.47±1.96</b>	<b>This study</b>
	Serranidae	<i>Epinephelus coeruleopunctatus</i>	Liver	1.52	Metian et al., 2013
	Serranidae	<i>Plectropomus leopardus</i>	Liver	2.91±0.37	Metian et al., 2013
	<b>Serranidae</b>	-	<b>Whole</b>	<b>1.22±2.32</b>	<b>This study</b>
Ni	Acanthuridae	<i>Naso unicornis</i>	Liver	<1.64	Metian et al., 2013
	<b>Acanthuridae</b>	-	<b>Whole</b>	<b>0.628±0.254</b>	<b>This study</b>
	Congidae	<i>Conger Laticauda laticudata</i>	Muscle	1.69±1.94	Briand et al., 2018
	<b>Congidae</b>	-	<b>Whole</b>	<b>0.210±0.157</b>	<b>This study</b>
	Labridae	<i>Bodianus perditio</i>	Liver	<1.65	Metian et al., 2013
	Labridae	<i>Cheilinus chlorourus</i>	Liver	1.62	Metian et al., 2013
	<b>Labridae</b>	-	<b>Whole</b>	<b>0.258±0.0646</b>	<b>This study</b>
	Muraenidae	<i>Gymnothorax undulatus</i>	Muscle	0.34±0.06	Briand et al., 2018
	<b>Muraenidae</b>	-	<b>Whole</b>	<b>0.155±0.0257</b>	<b>This study</b>
	Scaridae	<i>Scarus ghobban</i>	Liver	<1.88	Metian et al., 2013
	Scaridae	<i>Scarus microrhinos</i>	Liver	<1.55	Metian et al., 2013
	<b>Scaridae</b>	-	<b>Whole</b>	<b>0.440±0.0859</b>	<b>This study</b>
	Serranidae	<i>Epinephelus coeruleopunctatus</i>	Liver	<1.26	Metian et al., 2013
	Serranidae	<i>Plectropomus leopardus</i>	Liver	<1.62	Metian et al., 2013
	<b>Serranidae</b>	-	<b>Whole</b>	<b>0.0806±0.0472</b>	<b>This study</b>
Pb	Acanthuridae	<i>Acanthurus triostegus</i>	Whole	13.5	Miao et al., 2001
	Acanthuridae	<i>Naso unicornis</i>	Liver	<0.08	Metian et al., 2013
	<b>Acanthuridae</b>	-	<b>Whole</b>	<b>0.538±1.78</b>	<b>This study</b>
	Congidae	<i>Conger cinereus</i>	Whole	6	Miao et al., 2001
	Congidae	<i>Conger Laticauda laticudata</i>	Muscle	0.07±0.10	Briand et al., 2018
	<b>Congidae</b>	-	<b>Whole</b>	<b>0.0929±0.139</b>	<b>This study</b>
	Labridae	<i>Bodianus perditio</i>	Liver	0.21	Metian et al., 2013

	Labridae	<i>Cheilinus chlorourus</i>	Liver	0.24	Metian et al., 2013
	<b>Labridae</b>	-	<b>Whole</b>	<b>0.335±1.84</b>	<b>This study</b>
	Muraenidae	<i>Gymnothorax flavimarginatus</i>	Whole	7	Miao et al., 2001
	Muraenidae	<i>Gymnothorax undulatus</i>	Muscle	0.04±0.003	Briand et al., 2018
	<b>Muraenidae</b>	-	<b>Whole</b>	<b>0.174±0.222</b>	<b>This study</b>
	Palinuridae	<i>Panulirus marginatus</i>	Whole	11	Miao et al., 2001
	<b>Palinuridae</b>	-	<b>Whole</b>	<b>0.217±0.164</b>	<b>This study</b>
	Scaridae	<i>Scarus ghobban</i>	Liver	0.14	Metian et al., 2013
	Scaridae	<i>Scarus microrhinos</i>	Liver	<0.08	Metian et al., 2013
	<b>Scaridae</b>	-	<b>Whole</b>	<b>0.336±0.508</b>	<b>This study</b>
	Serranidae	<i>Epinephelus coeruleopunctatus</i>	Liver	<0.06	Metian et al., 2013
	Serranidae	<i>Plectropomus leopardus</i>	Liver	0.24±0.05	Metian et al., 2013
	<b>Serranidae</b>	-	<b>Whole</b>	<b>0.0523±0.0806</b>	<b>This study</b>
Se	Acanthuridae	<i>Acanthurus triostegus</i>	Whole	21.5	Miao et al., 2001
	Acanthuridae	<i>Naso unicornis</i>	Liver	<16.4	Metian et al., 2013
	<b>Acanthuridae</b>	-	<b>Whole</b>	<b>0.407±0.122</b>	<b>This study</b>
	Congidae	<i>Conger cinereus</i>	Whole	22	Miao et al., 2001
	Congidae	<i>Conger Laticauda laticudata</i>	Muscle	1.90±0.85	Briand et al., 2018
	<b>Congidae</b>	-	<b>Whole</b>	<b>1.20±0.217</b>	<b>This study</b>
	Labridae	<i>Bodianus perditio</i>	Liver	<16.5	Metian et al., 2013
	Labridae	<i>Cheilinus chlorourus</i>	Liver	19.1	Metian et al., 2013
	<b>Labridae</b>	-	<b>Whole</b>	<b>1.15±0.475</b>	<b>This study</b>
	Muraenidae	<i>Gymnothorax flavimarginatus</i>	Whole	25	Miao et al., 2001
	Muraenidae	<i>Gymnothorax undulatus</i>	Muscle	1.95±0.01	Briand et al., 2018
	<b>Muraenidae</b>	-	<b>Whole</b>	<b>1.65±0.380</b>	<b>This study</b>
	Palinuridae	<i>Panulirus marginatus</i>	Whole	27	Miao et al., 2001
	<b>Palinuridae</b>	-	<b>Whole</b>	<b>1.25±0.128</b>	<b>This study</b>
	Scaridae	<i>Scarus ghobban</i>	Liver	<18.8	Metian et al., 2013



	Scaridae	<i>Scarus microrhinos</i>	Liver	<15.5	Metian et al., 2013
	<b>Scaridae</b>	-	<b>Whole</b>	<b>0.907±0.273</b>	<b>This study</b>
	Serranidae	<i>Epinephelus coeruleopunctatus</i>	Liver	<12.6	Metian et al., 2013
	Serranidae	<i>Plectropomus leopardus</i>	Liver	<16.2	Metian et al., 2013
	<b>Serranidae</b>	-	<b>Whole</b>	<b>1.47±0.327</b>	<b>This study</b>
V	Acanthuridae	<i>Naso unicornis</i>	Liver	7.22	Metian et al., 2013
	<b>Acanthuridae</b>	-	<b>Whole</b>	<b>1.03±0.387</b>	<b>This study</b>
	Congidae	<i>Conger Laticauda laticudata</i>	Muscle	<dl	Briand et al., 2018
	<b>Congidae</b>	-	<b>Whole</b>	<b>0.0420±0.0148</b>	<b>This study</b>
	Labridae	<i>Bodianus perditio</i>	Liver	<1.65	Metian et al., 2013
	Labridae	<i>Cheilinus chlorourus</i>	Liver	3.83	Metian et al., 2013
	<b>Labridae</b>	-	<b>Whole</b>	<b>0.307±0.457</b>	<b>This study</b>
	Muraenidae	<i>Gymnothorax undulatus</i>	Muscle	<dl	Briand et al., 2018
	<b>Muraenidae</b>	-	<b>Whole</b>	<b>0.276±0.593</b>	<b>This study</b>
	Scaridae	<i>Scarus ghobban</i>	Liver	<1.88	Metian et al., 2013
	Scaridae	<i>Scarus microrhinos</i>	Liver	<1.55	Metian et al., 2013
	<b>Scaridae</b>	-	<b>Whole</b>	<b>1.23±0.554</b>	<b>This study</b>
	Serranidae	<i>Epinephelus coeruleopunctatus</i>	Liver	<1.26	Metian et al., 2013
	Serranidae	<i>Plectropomus leopardus</i>	Liver	5.66±2.40	Metian et al., 2013
	<b>Serranidae</b>	-	<b>Whole</b>	<b>0.0442±0.0997</b>	<b>This study</b>
Zn	Acanthuridae	<i>Acanthurus triostegus</i>	Whole	118	Miao et al., 2001
	Acanthuridae	<i>Naso unicornis</i>	Liver	110	Metian et al., 2013
	<b>Acanthuridae</b>	-	<b>Whole</b>	<b>25.6±11.6</b>	<b>This study</b>
	Congidae	<i>Conger cinereus</i>	Whole	92	Miao et al., 2001
	Congidae	<i>Conger Laticauda laticudata</i>	Muscle	43±23	Briand et al., 2018
	<b>Congidae</b>	-	<b>Whole</b>	<b>40.7±2.11</b>	<b>This study</b>
	Labridae	<i>Bodianus perditio</i>	Liver	63.5	Metian et al., 2013
	Labridae	<i>Cheilinus chlorourus</i>	Liver	122	Metian et al., 2013

<b>Labridae</b>	-	<b>Whole</b>	<b>46.5±7.02</b>	<b>This study</b>
Muraenidae	<i>Gymnothorax flavimarginatus</i>	Whole	104	Miao et al., 2001
Muraenidae	<i>Gymnothorax undulatus</i>	Muscle	58±5.3	Briand et al., 2018
<b>Muraenidae</b>	-	<b>Whole</b>	<b>51.6±22.6</b>	<b>This study</b>
Palinuridae	<i>Panulirus marginatus</i>	Whole	129	Miao et al., 2001
<b>Palinuridae</b>	-	<b>Whole</b>	<b>61.1±8.50</b>	<b>This study</b>
Scaridae	<i>Scarus ghobban</i>	Liver	19.6	Metian et al., 2013
Scaridae	<i>Scarus microrhinos</i>	Liver	23.4	Metian et al., 2013
<b>Scaridae</b>	-	<b>Whole</b>	<b>18.6±2.86</b>	<b>This study</b>
Serranidae	<i>Epinephelus coeruleopunctatus</i>	Liver	1034	Metian et al., 2013
Serranidae	<i>Plectropomus leopardus</i>	Liver	393±167	Metian et al., 2013
<b>Serranidae</b>	-	<b>Whole</b>	<b>25.8±8.54</b>	<b>This study</b>

those transported in blood get fixed to bone cells, osteocytes (Nechifor et al., 2009). The elements are then integrated into the bone matrix during calcification and will remain until the bone is remodeled or resorbed (Helliwell et al., 1996). Since cortical (compact, 80% of bone mass) bone and trabecular (spongy, 20% of bone mass) bone are formed by bone packets and individual osteons are produced at different times, the content of the bone depends on the time of deposition (Ott, 2018; Roschger et al., 2008). As bones form, the mineral crystals can be changed from the normal composition based on the surrounding matrix at that time if other competing ions are there. An example is Ca substitution by Ba, Co, Pb, Mg, Mn, Ni, or Zn as they are bivalent metal ions (Aaseth et al., 2012). Post-mortem changes are possible with bone as diagenetic process can alter the original composition of the bone through activities such as exposure to ground water, decomposition, and leaching. The original elements of the bone can be enriched, depleted, or substituted through these activities (Price et al., 1985). Compact bone as opposed to trabecular bone is more likely to preserve environmental signals (Sillen, 1990). Elements involved with the mineral portion of cortical bone include Ca, C, Cl, F, K, Mg, Na, P, and Sr (Armstrong & Singer, 1965). Humans deposit 75 % of trace elements into bone until adolescence, where it increases to 90-95 % with exposure (Lobinski et al., 2006). Honda et al. (1982, 1984a, b) found marine mammal bone had high concentrations of Mn, Pb, and Zn, of which Mn and Zn are essential for bone formation. Zinc accumulates in bone with calcification while Cu is associated with the formation of collagen in cartilage.

## **2. HYPOTHESES AND OBJECTIVES**

### **1. Do heavy metal concentrations in Hawaiian monk seals vary between the Northwestern Hawaiian Islands (NWHI) and the Main Hawaiian Islands (MHI)?**

The seals originate from the NWHI but within the past century have established a colony within the MHI. The different regions will have experienced different inputs of naturally sourced metals, such as via volcanism, and anthropogenic inputs of metals from military, shipping, agriculture, and urban development. The natural input of heavy metals via volcanism and weathering are believed to be the same for all islands but the remaining atolls in the NWHI (coral exoskeleton) may have different concentrations. Anthropogenic inputs will be different for each region as human activity varies between the two regions, but ocean current patterns are consistent.

**2. Are there sex differences in heavy metal concentrations in Hawaiian monk seals?**

Male and female seals may have foraging differences that would be reflected in their diet. Adult female seals may also offload heavy metals during gestation and lactation that males would not.

**3. Are there age class differences in heavy metal concentrations in the Hawaiian monk seal?**

Pup, juvenile, and adult seals have foraging differences due to experience that would be reflected in their diet.

**4. Do heavy metal concentrations vary among potential prey?**

While the composition of prey in the seals' diet is expected to be similar, the concentration of heavy metals likely varies among taxa. Biomagnification from the prey to the seals will also vary with the prey's trophic levels. The habitat and location of the prey could represent bioaccumulation in certain areas.

**3. MATERIALS AND METHODS**

**3.1 Archived Tissue Samples**

Bone samples (n=102) opportunistically collected from deceased HMS and prey items (n=81) were collected by the Pacific Islands Fisheries Science Center (PIFSC), NOAA in Hawaii under the authorized US Marine Mammal Protection Act Permit 764-1703-01, National Museum of Natural History and the Museum Support Center, Smithsonian Institution issued by the National Marine Fisheries Service. Bone samples were collected throughout the NWHI and MHI during the 2000s and 2010s. Bone samples were from the following locations: NWHI - Northern (n=21), Central (n=40), and Southern (n=30), and the MHI (n=11). The spatial range of the Northern NWHI extends from 30° N, 180° W to 27° N, 175° W, including Kure Atoll, Midway Island, and Pearl and Hermes Reef. The Central NWHI ranges from 27° N, 175° W to 25° N, 167° W, including Laysan and Lisianski islands. The Southern NWHI ranges from 25° N, 167° W to 23° N, 161° W, including French Frigate Shoal and Necker and Nihoa islands. Bones were from three sex categories: female (n=37), male (n=31), unknown (n=34), and further categorized by age classes: adult (n=19), juvenile (n=32), and pup (n=35). Adult seals were of reproductive age while the juveniles were not (Table 6). HMS potential prey samples (n=81) were collected

Table 6. Hawaiian monk seal bone samples analyzed in this study, including separation by age class and sex. Regions include the MHI referred to as Main and the NWHI being separated into Southern, Central, and Northern regions.

<b>Region</b>	<b>Age Class</b>	<b>Sex</b>	<b>N</b>	
<b>Main</b>	Adult	Male	1	
		Unknown	1	
	Juvenile	Female	2	
		Unknown	2	
	Pup	Female	1	
		Unknown	1	
Unknown	Unknown	3		
<b>Southern</b>	Adult	Female	2	
		Male	2	
		Unknown	1	
	Juvenile	Female	3	
		Unknown	1	
	Pup	Female	5	
		Male	9	
		Unknown	2	
	Unknown	Unknown	5	
	<b>Central</b>	Adult	Female	3
Male			1	
Unknown			2	
Juvenile		Female	7	
		Male	9	
		Unknown	5	
Pup		Female	4	
		Male	6	
Unknown		Female	1	
		Unknown	2	
<b>Northern</b>		Adult	Female	3
			Male	1
	Unknown		2	
	Juvenile	Female	3	
		Pup	Female	3
	Unknown	Male	2	
		Unknown	2	
		Unknown	2	
		Unknown	5	

from the Southern region of the NWHI during 2000 to 2006 and represent 10 different taxonomic groups: Muraenidae (moray eel), Palinuridae (lobster), Congidae (Conger eel), Labridae (wrasse), Holocentridae (squirrelfish), Acanthuridae (surgeonfish), Serranidae (groupers), Balistidae (triggerfish), Scaridae (parrotfish), and Octopodidae (octopus) (Table 7).

### **3.2 Heavy Metal Analysis**

Archived, whole prey samples were homogenized with a clean, commercial blender while whole, frozen at -20° C, 1-2 g wet weight subsampled, and then dried at 60° C for a minimum of 72 hours or until completely dry. A dental amalgamator (brand Wig-L-Bug) was used to pulverize the prey samples into a fine powder.

Whole and fractured pieces of cortical bone samples were subsampled using a Dremel (Mt. Prospect, IL) rotary cutting tool for a targeted mass of over 0.2 g. Bone pieces were then cleaned by soaking in 10% H<sub>2</sub>O<sub>2</sub> for 48 hours, sonicated in deionized water for 30 minutes, and then dried at 60° C for a minimum of 72 hours until completely dried.

All seal bone and prey samples were analyzed via elemental analysis for 16 heavy metals: aluminum (Al), arsenic (As), cadmium (Cd), cobalt (Co), chromium (Cr), copper (Cu), iron (Fe), mercury (Hg), manganese (Mn), molybdenum (Mo), nickel (Ni), lead (Pb), selenium (Se), tin (Sn), vanadium (V), and zinc (Zn). Each sample (bone and prey) was weighed between 0.2 - 0.3 g dry weight before placement in a 100 ml Teflon PTFE tube and digested. Bone samples were digested with 3 ml 70 % trace metal basis nitric acid (CAS Number 7697-37-2) and 1 ml 30 % hydrogen peroxide (CAS Number 7722-84-1) while the prey samples, between 0.2 g - 0.3 g, were digested with 4 ml 70 % trace metal basis nitric acid (CAS Number 7697-37-2) and 2 ml 30 % hydrogen peroxide (CAS Number 7722-84-1). The samples were digested at room temperature over night or until completely digested. Heat (40° C) was added, using a ModBlock to samples that were not initially digesting. All samples were then diluted to 25 ml in a volumetric flask with ultrapure deionized water (18.2 megohm). Five ml samples were added to a 15 ml PTFE bottle and shipped to the University of Southern Mississippi's Center for Trace Analysis. Samples were then analyzed using a sector-field inductively coupled plasma mass spectrometer (ThermoFisher Element XR) with a Peltier-cooler spray chamber (PC-3; Elemental Scientific, Inc.) to determine the concentrations of the metals.

### **3.3 Statistical Analysis**

Table 7. Total number of whole potential prey samples from the Southern Northwest Hawaiian Island region analyzed in this study, including taxonomic family and region.

<b>Family</b>	<b>Region</b>	<b>N</b>
<b>Acanthuridae</b>	Southern NWHI	9
<b>Balistidae</b>	Southern NWHI	9
<b>Congidae</b>	Southern NWHI	2
<b>Holocentridae</b>	Southern NWHI	10
<b>Labridae</b>	Southern NWHI	9
<b>Muraenidae</b>	Southern NWHI	10
<b>Octopodidae</b>	Southern NWHI	8
<b>Palinuridae</b>	Southern NWHI	10
<b>Scaridae</b>	Southern NWHI	10
<b>Serranidae</b>	Southern NWHI	4

Descriptive statistics for all data, including arithmetic mean, standard deviation, geometric mean, confidence interval 95 %, and median for heavy metal concentrations, were calculated using Microsoft Excel (Version 2205 Build 16.0.15225.20278; Microsoft Corporation). Arithmetic mean represents all samples equally while geometric mean accounts for outliers by focusing on the central tendency of the set of numbers. For concentrations below the detection range, the lowest detected concentration was divided in half and used to replace the below the detection range values; this was only used for a few As samples. Bone meal and fish protein certified reference materials were used to check for digestion yield for the specific heavy metals they contained.

Certified reference material was analyzed along with the samples: Bone meal (Standard Reference Material 1486) and fish protein (DORM-5 Fish Protein Certified Reference Material). The recovery percentage for the bone had a high of 109 % (Cd) and a low of 73.8 % (Hg) with outliers of 216% (Al) and 22.4% (As) (Table 8). The recovery percentage for fish protein had a high of 97% (Cd) and a low of 61% (Pb) (Table 8).

The molar ratio of Se to Hg was calculated for each seal bone and potential prey sample via:

$$\frac{\text{Se}}{\text{Hg}} = \frac{(\text{Se}/78.96)}{(\frac{\text{Hg}}{200.59})}$$

where 78.96 g/mol and 200.59 g/mol are the atomic masses of Se and Hg, respectively. Molar ratios are needed to more accurately state the proportion of Se and Hg compared to concentration ratios. A Kendall's tau correlation was used to determine the strength of the statistical relationship between Se and Hg concentrations for each seal bone and whole potential prey.

To determine the differences among the heavy metal concentrations, t-test, one-way analysis of variance (ANOVA), and two-way ANOVA were utilized using RStudio (Version 2022.02.2 Build 485; RStudio, PBC). All data was tested for normality using a Shapiro-Wilk test and Bartlett's test. A non-parametric Mann-Whitney Wilcoxon test was used to test for each heavy metal concentration variance between seal sex and between Southern region seals and potential prey. A Kruskal Wallis non-parametric test with a non-parametric multiple comparisons post-hoc test was used to test for each heavy metal concentration variance among seal region, age class, and sex plus heavy metal concentration variance among prey families, prey taxa, and



Table 8. The percentage yield of elements with certified concentrations found within the reference materials: bone meal and fish protein.

Tissue	Element	Recovery
		Percentage
<b>Bone</b>	Al	216
	As	22.4
	Cd	109
	Cu	86.6
	Fe	96.0
	Hg	73.8
	Mn	95.2
	Pb	100
	Se	85.8
	Zn	97.3
<b>Fish Protein</b>	As	80.9
	Cd	96.9
	Cr	84.6
	Cu	90.9
	Fe	94.7
	Hg	90.0
	Mn	83.9
	Ni	82.1
	Pb	61.4
	Se	71.8
	V	86.2
	Zn	83.9

Southern region seals. A statistically significant difference between two groups was found when the p-value was less than 0.05 leaving a 5% chance that the difference occurred by chance.

## **4. RESULTS**

### **4.1 Hawaiian Monk Seal**

A total of 102 Hawaiian monk seal bones were analyzed for 16 heavy metals. On average, Zn (269 – 67.8  $\mu\text{g/g}$ ) and Fe (494 – 14.6  $\mu\text{g/g}$ ) had the highest concentrations of all 16 metals, often two to three orders of magnitude higher. There were a few major concentration outliers among the seals. One Southern male pup had a Cu concentration of 264  $\mu\text{g/g}$  which was two to three orders of magnitude larger than the rest of the seals' Cu concentrations. One Northern seal of unknown age and sex had a Pb concentration of 14.9  $\mu\text{g/g}$  which was two orders of magnitude larger than the rest of the seals. One Central male pup had a Hg concentration of 7.70  $\mu\text{g/g}$  which was two to three orders of magnitude larger than majority of the rest of the seals' Hg concentrations.

Region significantly affected the heavy metal concentrations of As, Cd, Cr, Cu, and Se (Table 9). The Main (MHI) region had significantly lower concentration than Cu in the Southern region, Se in the Southern and Central regions, As and Cd in the Central and Northern regions, and Cr in all three NWHI regions. Selenium was significantly lower in the Central region compared to the Northern region (Table 10 and Figure 4).

Female Co and Pb concentrations were significantly greater than male concentrations across all seal bones while male Cu and Se concentrations were significantly greater than female concentrations (Tables 9 and 11 and Figure 5). Sex categories by region had no significant effects on the heavy metal concentrations (Tables 9 and 12 and Figure 6).

Age class significantly affected the heavy metal concentrations of Cd, Cu, Fe, Ni, Sn, and V (Table 9). Pup Cu and Fe concentrations were significantly greater than adult and juvenile concentrations. Pup Sn concentration was significantly greater than adult while pup Ni concentration was significantly greater than juvenile. Juvenile V concentration was significantly greater than pup, and adult Cd concentration was significantly greater than pup (Table 13 and Figure 7). Age class by region significantly affected the following heavy metal concentrations of Cd and Fe (Table 9). In the Southern region, adult Cd concentration was significantly greater than pup. In the Central region, pup Fe concentration was significantly greater than adult (Table 14 and Figure 8).

Table 9. Statistical results for all heavy metals under all comparisons.

Heavy Metals		HMS Region	HMS Sex	HMS Sex - Region	HMS Age Class	HMS Age Class - Region	HMS Age Class - Sex	Prey Taxa	Prey Family	Seal - Prey	Seal - Prey Taxa
<b>Al</b>	Statistical Test	X <sup>2</sup> (3) = 0.98794	W = 621	X <sup>2</sup> (11) = 7.1761	X <sup>2</sup> (2) = 4.7641	X <sup>2</sup> (15) = 16.943	X <sup>2</sup> (10) = 13.018	X <sup>2</sup> (2) = 5.1043	X <sup>2</sup> (9) = 20.556	W = 7190	X <sup>2</sup> (3) = 76.149
	p-value	p = 0.804	p = 0.565	p = 0.785	p = 0.092	p = 0.322	p = 0.223	p = 0.078	p = 0.015	p < 0.001	p < 0.001
<b>As</b>	Statistical Test	X <sup>2</sup> (3) = 15.588	W = 476	X <sup>2</sup> (11) = 23.869	X <sup>2</sup> (2) = 2.6167	X <sup>2</sup> (15) = 29.248	X <sup>2</sup> (10) = 10.023	X <sup>2</sup> (2) = 40.476	X <sup>2</sup> (9) = 58.956	W = 8262	X <sup>2</sup> (3) = 142.69
	p-value	p = 0.001	p = 0.232	p = 0.013	p = 0.270	p = 0.015	p = 0.439	p < 0.001	p < 0.001	p < 0.001	p < 0.001
<b>Cd</b>	Statistical Test	X <sup>2</sup> (3) = 17.778	W = 674	X <sup>2</sup> (11) = 25.41	X <sup>2</sup> (2) = 16.211	X <sup>2</sup> (15) = 42.039	X <sup>2</sup> (10) = 22.235	X <sup>2</sup> (2) = 40.259	X <sup>2</sup> (9) = 51.102	W = 7574	X <sup>2</sup> (3) = 116.34
	p-value	p < 0.001	p = 0.220	p = 0.008	p < 0.001	p < 0.001	p = 0.014	p < 0.001	p < 0.001	p < 0.001	p < 0.001
<b>Co</b>	Statistical Test	X <sup>2</sup> (3) = 4.8732	W = 727	X <sup>2</sup> (11) = 15.482	X <sup>2</sup> (2) = 1.8962	X <sup>2</sup> (15) = 11.672	X <sup>2</sup> (10) = 17.934	X <sup>2</sup> (2) = 18.427	X <sup>2</sup> (9) = 39.254	W = 7545	X <sup>2</sup> (3) = 98.208
	p-value	p = 0.181	p = 0.030	p = 0.162	p = 0.388	p = 0.7037	p = 0.056	p < 0.001	p < 0.001	p < 0.001	p < 0.001
<b>Cr</b>	Statistical Test	X <sup>2</sup> (3) = 15.894	W = 637	X <sup>2</sup> (11) = 21.676	X <sup>2</sup> (2) = 3.0008	X <sup>2</sup> (15) = 28.099	X <sup>2</sup> (10) = 13.262	X <sup>2</sup> (2) = 3.9433	X <sup>2</sup> (9) = 27.96	W = 7970	X <sup>2</sup> (3) = 117.23
	p-value	p = 0.001	p = 0.440	p = 0.027	p = 0.223	p = 0.021	p = 0.209	p = 0.139	p < 0.001	p < 0.001	p < 0.001
<b>Cu</b>	Statistical Test	X <sup>2</sup> (3) = 12.172	W = 409	X <sup>2</sup> (11) = 18.582	X <sup>2</sup> (2) = 23.088	X <sup>2</sup> (15) = 48.304	X <sup>2</sup> (10) = 31.8	X <sup>2</sup> (2) = 41.494	X <sup>2</sup> (9) = 52.673	W = 7380	X <sup>2</sup> (3) = 97.455
	p-value	p = 0.007	p = 0.021	p = 0.069	p < 0.001	p < 0.001	p < 0.001	p < 0.001	p < 0.001	p < 0.001	p < 0.001
<b>Fe</b>	Statistical Test	X <sup>2</sup> (3) = 5.3071	W = 475	X <sup>2</sup> (11) = 13.179	X <sup>2</sup> (2) = 17.4	X <sup>2</sup> (15) = 40.903	X <sup>2</sup> (10) = 33.204	X <sup>2</sup> (2) = 28.543	X <sup>2</sup> (9) = 44.593	W = 3340	X <sup>2</sup> (3) = 29.632
	p-value	p = 0.151	p = 0.229	p = 0.282	p < 0.001	p < 0.001	p < 0.001	p < 0.001	p < 0.001	p = 0.013	p < 0.001
<b>Hg</b>	Statistical Test	X <sup>2</sup> (3) = 3.0719	W = 454	X <sup>2</sup> (11) = 11.234	X <sup>2</sup> (2) = 3.4581	X <sup>2</sup> (15) = 16.025	X <sup>2</sup> (10) = 10.698	X <sup>2</sup> (2) = 4.2707	X <sup>2</sup> (9) = 55.961	W = 6378	X <sup>2</sup> (3) = 44.355
	p-value	p = 0.381	p = 0.144	p = 0.424	p = 0.178	p = 0.380	p = 0.382	p = 0.1182	p < 0.001	p < 0.001	p < 0.001
<b>Mn</b>	Statistical Test	X <sup>2</sup> (3) = 3.4677	W = 525	X <sup>2</sup> (11) = 8.755	X <sup>2</sup> (2) = 4.1417	X <sup>2</sup> (15) = 26.992	X <sup>2</sup> (10) = 14.638	X <sup>2</sup> (2) = 24.928	X <sup>2</sup> (9) = 56.573	W = 7450	X <sup>2</sup> (3) = 98.429
	p-value	p = 0.325	p = 0.557	p = 0.645	p = 0.126	p = 0.029	p = 0.146	p < 0.001	p < 0.001	p < 0.001	p < 0.001
<b>Mo</b>	Statistical Test	X <sup>2</sup> (3) = 2.7201	W = 617	X <sup>2</sup> (11) = 7.4972	X <sup>2</sup> (2) = 5.6123	X <sup>2</sup> (15) = 16.32	X <sup>2</sup> (10) = 15.471	X <sup>2</sup> (2) = 30.51	X <sup>2</sup> (9) = 56.176	W = 7795	X <sup>2</sup> (3) = 114.46
	p-value	p = 0.437	p = 0.599	p = 0.758	p = 0.060	p = 0.361	p = 0.116	p < 0.001	p < 0.001	p < 0.001	p < 0.001
<b>Ni</b>	Statistical Test	X <sup>2</sup> (3) = 5.2283	W = 599	X <sup>2</sup> (11) = 8.1061	X <sup>2</sup> (2) = 7.0266	X <sup>2</sup> (15) = 21.588	X <sup>2</sup> (10) = 15.599	X <sup>2</sup> (2) = 30.73	X <sup>2</sup> (9) = 64.321	W = 6222	X <sup>2</sup> (3) = 52.151
	p-value	p = 0.156	p = 0.760	p = 0.704	p = 0.030	p = 0.119	p = 0.112	p < 0.001	p < 0.001	p < 0.001	p < 0.001
<b>Pb</b>	Statistical Test	X <sup>2</sup> (3) = 7.2702	W = 783	X <sup>2</sup> (11) = 23.52	X <sup>2</sup> (2) = 1.1997	X <sup>2</sup> (15) = 22.687	X <sup>2</sup> (10) = 20.001	X <sup>2</sup> (2) = 0.26148	X <sup>2</sup> (9) = 15.867	W = 5719	X <sup>2</sup> (3) = 19.973
	p-value	p = 0.064	p = 0.005	p = 0.015	p = 0.549	p = 0.091	p = 0.029	p = 0.877	p = 0.070	p < 0.001	p < 0.001
<b>Se</b>	Statistical Test	X <sup>2</sup> (3) = 19.917	W = 413	X <sup>2</sup> (11) = 23.986	X <sup>2</sup> (2) = 3.8685	X <sup>2</sup> (15) = 31.099	X <sup>2</sup> (10) = 19.201	X <sup>2</sup> (2) = 5.2525	X <sup>2</sup> (9) = 43.294	W = 8127	X <sup>2</sup> (3) = 127.36
	p-value	p < 0.001	p = 0.024	p = 0.013	p = 0.145	p = 0.009	p = 0.378	p = 0.072	p < 0.001	p < 0.001	p < 0.001
<b>Sn</b>	Statistical Test	X <sup>2</sup> (3) = 1.602	W = 472	X <sup>2</sup> (11) = 8.5094	X <sup>2</sup> (2) = 8.4288	X <sup>2</sup> (15) = 26.998	X <sup>2</sup> (10) = 25.274	X <sup>2</sup> (2) = 28.217	X <sup>2</sup> (9) = 40.53	W = 2389	X <sup>2</sup> (3) = 44.331
	p-value	p = 0.659	p = 0.215	p = 0.667	p = 0.015	p = 0.029	p = 0.005	p < 0.001	p < 0.001	p < 0.001	p < 0.001
<b>V</b>	Statistical Test	X <sup>2</sup> (3) = 3.666	W = 698	X <sup>2</sup> (11) = 16.99	X <sup>2</sup> (2) = 16.22	X <sup>2</sup> (15) = 29.92	X <sup>2</sup> (10) = 33.13	X <sup>2</sup> (2) = 1.5372	X <sup>2</sup> (9) = 48.316	W = 7648	X <sup>2</sup> (3) = 98.508
	p-value	p = 0.300	p = 0.127	p = 0.108	p < 0.001	p = 0.012	p < 0.001	p = 0.464	p < 0.001	p < 0.001	p < 0.001
<b>Zn</b> <sup>47</sup>	Statistical Test	X <sup>2</sup> (3) = 5.0912	W = 481	X <sup>2</sup> (11) = 10.434	X <sup>2</sup> (2) = 2.5246	X <sup>2</sup> (15) = 26.062	X <sup>2</sup> (10) = 7.0303	X <sup>2</sup> (2) = 15.385	X <sup>2</sup> (9) = 55.298	W = 965	X <sup>2</sup> (3) = 84.382
	p-value	p = 0.1652	p = 0.259	p = 0.492	p = 0.283	p = 0.037	p = 0.723	p < 0.001	p < 0.001	p < 0.001	p < 0.001

Table 10a. Arithmetic mean and standard deviation of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by region: MHI (Main) and NWHI (Southern, Central, Northern).

Region	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
<b>Main</b>	2.54 $\pm$	0.0166	0.0191 $\pm$	0.0139 $\pm$	0.120 $\pm$	0.371 $\pm$	79.3 $\pm$	0.0489 $\pm$	0.680 $\pm$	0.0346 $\pm$	0.368 $\pm$	0.0820 $\pm$	0.106 $\pm$	1.46 $\pm$	0.119 $\pm$	96.0 $\pm$
	0.786	$\pm$ 0.0202	0.0178	0.0095	0.0491	0.402	33.3	0.102	0.248	0.0439	0.296	0.0364	0.0556	0.313	0.0954	19.7
<b>Southern</b>	6.24 $\pm$	0.0761	0.0657 $\pm$	0.0137 $\pm$	0.298 $\pm$	10.1 $\pm$	145 $\pm$	0.0477 $\pm$	0.895 $\pm$	0.0315 $\pm$	0.407 $\pm$	0.277 $\pm$	0.239 $\pm$	1.48 $\pm$	0.0719 $\pm$	114 $\pm$
	6.96	$\pm$ 0.112	0.0906	0.0162	0.264	48.1	110	0.0947	0.422	0.0206	0.519	0.323	0.181	0.262	0.0705	39.2
<b>Central</b>	6.62 $\pm$	0.117 $\pm$	0.195 $\pm$	0.0090 $\pm$	0.326 $\pm$	0.767 $\pm$	95.7 $\pm$	0.234 $\pm$	0.839 $\pm$	0.0417 $\pm$	0.269 $\pm$	0.142 $\pm$	0.218 $\pm$	1.64 $\pm$	0.0856 $\pm$	114 $\pm$
	15.6	0.153	0.628	0.0081	0.250	0.603	69.2	1.21	0.356	0.0485	0.448	0.127	0.108	0.633	0.0740	30.4
<b>Northern</b>	6.43 $\pm$	0.112 $\pm$	0.114 $\pm$	0.0140 $\pm$	0.549 $\pm$	0.806 $\pm$	85.8 $\pm$	0.0302 $\pm$	0.999 $\pm$	0.0323 $\pm$	0.218 $\pm$	0.909 $\pm$	0.143 $\pm$	1.45 $\pm$	0.0947 $\pm$	98 $\pm$
	11.9	0.154	0.153	0.0136	0.897	1.30	59.4	0.0349	0.628	0.0181	0.293	3.22	0.0601	0.379	0.0689	28.1

Table 10b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by region: MHI (Main) and NWHI (Southern, Central, Northern).

Region	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
<b>Main</b>	2.43 $\pm$	0.0099 $\pm$	0.0121 $\pm$	0.0107 $\pm$	0.113 $\pm$	0.263 $\pm$	73.6 $\pm$	0.0186 $\pm$	0.642 $\pm$	0.0238 $\pm$	0.222 $\pm$	0.0732 $\pm$	0.0948 $\pm$	1.43 $\pm$	0.0782 $\pm$	94.2 $\pm$
	0.465	0.0119	0.0105	0.0056	0.0290	0.238	19.7	0.0602	0.147	0.0259	0.175	0.0215	0.0329	0.185	0.0564	11.7
<b>Southern</b>	3.75 $\pm$	0.0293 $\pm$	0.0285 $\pm$	0.0090 $\pm$	0.219 $\pm$	0.950 $\pm$	108 $\pm$	0.0191 $\pm$	0.804 $\pm$	0.0263 $\pm$	0.201 $\pm$	0.164 $\pm$	0.200 $\pm$	1.46 $\pm$	0.0388 $\pm$	109 $\pm$
	2.49	0.0401	0.0324	0.0058	0.0946	17.2	39.5	0.0339	0.151	0.0074	0.186	0.115	0.0649	0.0938	0.0252	14.0
<b>Central</b>	3.01 $\pm$	0.0601 $\pm$	0.0670 $\pm$	0.0068 $\pm$	0.260 $\pm$	0.563 $\pm$	75.6 $\pm$	0.0314 $\pm$	0.786 $\pm$	0.0306 $\pm$	0.125 $\pm$	0.110 $\pm$	0.195 $\pm$	1.56 $\pm$	0.0664 $\pm$	111 $\pm$
	4.83	0.0473	0.194	0.0025	0.0774	0.187	21.4	0.376	0.110	0.0150	0.139	0.0334	0.0334	0.196	0.0229	9.42
<b>Northern</b>	3.26 $\pm$	0.0449 $\pm$	0.0555 $\pm$	0.0098 $\pm$	0.308 $\pm$	0.460 $\pm$	67.3 $\pm$	0.0186 $\pm$	0.878 $\pm$	0.0291 $\pm$	0.136 $\pm$	0.177 $\pm$	0.132 $\pm$	1.40 $\pm$	0.0681 $\pm$	85.8 $\pm$
	5.07	0.0658	0.0656	0.0058	0.384	0.556	25.4	0.0149	0.269	0.0078	0.125	1.38	0.162	0.162	0.0295	12.0

Table 10c. Median of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by region: MHI (Main) and NWHI (Southern, Central, Northern).

Region	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
<b>Main</b>	2.32	0.0119	0.0122	0.0109	0.111	0.213	66.6	0.0167	0.690	0.0199	0.305	0.0860	0.0923	1.39	0.0727	85.4
<b>Southern</b>	2.44	0.0282	0.0287	0.0084	0.209	0.772	128	0.0218	0.778	0.0272	0.127	0.140	0.182	1.50	0.0543	104
<b>Central</b>	2.23	0.0495	0.0579	0.0057	0.244	0.473	72.1	0.0247	0.741	0.0250	0.0987	0.0948	0.199	1.51	0.0675	103
<b>Northern</b>	2.82	0.0526	0.0839	0.0098	0.193	0.447	62.3	0.0236	0.808	0.0278	0.107	0.128	0.135	1.43	0.0909	98.5

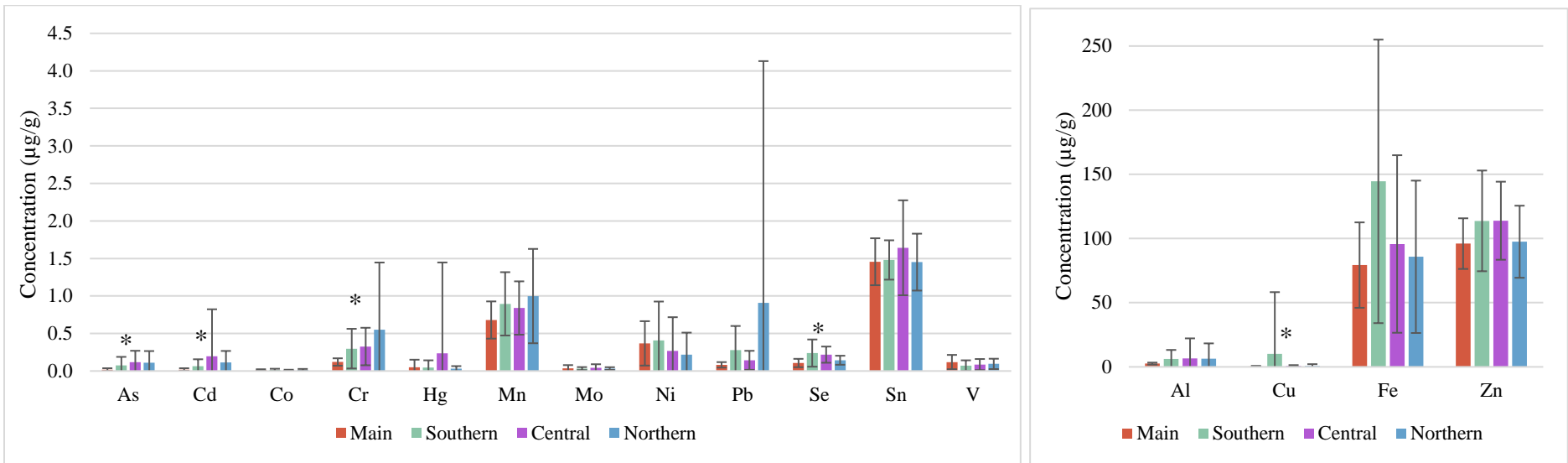


Figure 4a. Arithmetic mean and standard deviation of 16 heavy metal concentrations (µg/g) in HMS bone by region: MHI (Main) and NWHI (Southern, Central, Northern). Metals with a significant difference among regions are indicated with an asterisk.

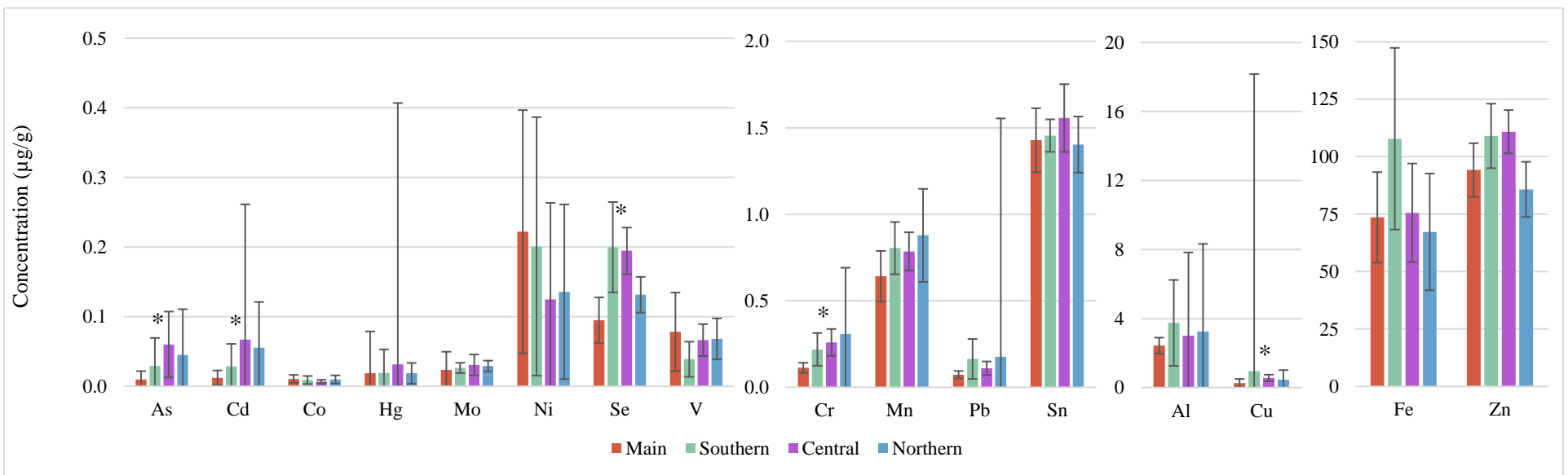


Figure 4b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations (µg/g) in HMS bone by region: MHI (Main) and NWHI (Southern, Central, Northern). Metals with a significant difference among regions are indicated with an asterisk.

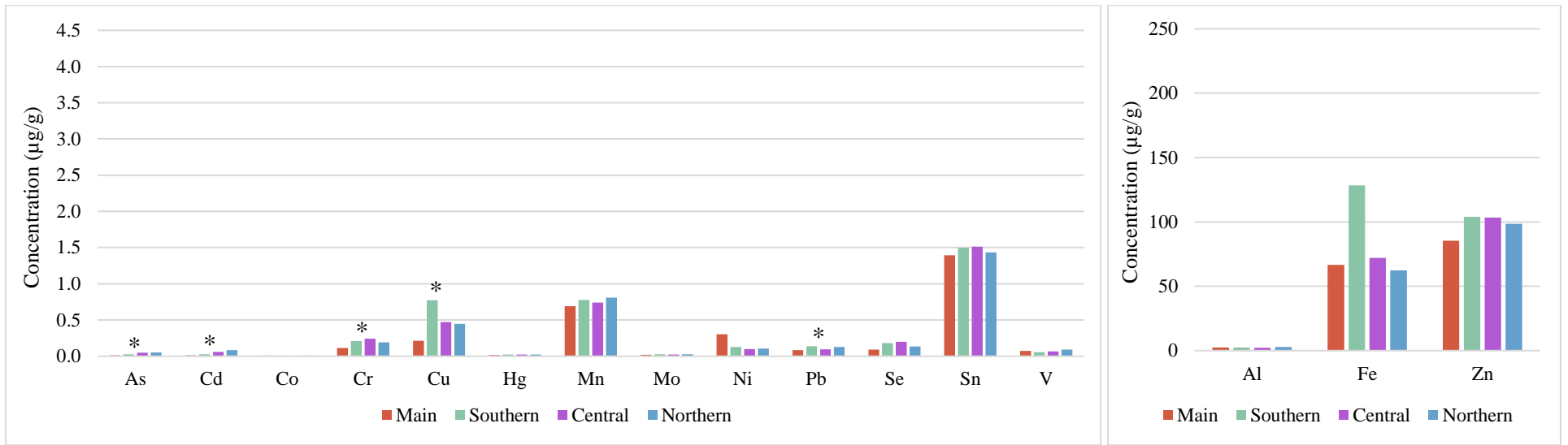


Figure 4c. Median of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by region: MHI (Main) and NWHI (Southern, Central, Northern). Metals with a significant difference among regions are indicated with an asterisk.

Table 11a. Arithmetic mean and standard deviation of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by sex.

Sex	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
Female	5.68 $\pm$	0.0951 $\pm$	0.208 $\pm$	0.0148 $\pm$	0.368 $\pm$	0.731 $\pm$	92.1 $\pm$	0.0376 $\pm$	0.810 $\pm$	0.0419 $\pm$	0.350 $\pm$	0.213 $\pm$	0.193 $\pm$	1.46 $\pm$	0.0877 $\pm$	104 $\pm$
	9.01	0.166	0.657	0.0158	0.350	0.804	75.0	0.0575	0.333	0.0490	0.510	0.189	0.158	0.426	0.0647	24.2
Male	4.85 $\pm$	0.0970 $\pm$	0.0667 $\pm$	0.0080 $\pm$	0.265 $\pm$	9.53 $\pm$	116 $\pm$	0.299 $\pm$	0.849 $\pm$	0.0312 $\pm$	0.321 $\pm$	0.137 $\pm$	0.231 $\pm$	1.64 $\pm$	0.0702 $\pm$	113 $\pm$
	6.73	0.0994	0.102	0.0057	0.194	47.4	97.0	1.38	0.345	0.0214	0.465	0.175	0.131	0.607	0.0812	38.3

Table 11b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by sex.

Sex	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
Female	3.30 $\pm$	0.0332 $\pm$	0.0505 $\pm$	0.0097 $\pm$	0.256 $\pm$	0.471 $\pm$	69.9 $\pm$	0.0168	0.752 $\pm$	0.0301 $\pm$	0.172 $\pm$	0.158 $\pm$	0.159 $\pm$	1.41 $\pm$	0.0634 $\pm$	102 $\pm$
	2.90	0.0533	0.212	0.0051	0.113	0.259	24.2	$\pm$ 0.185	0.107	0.0158	0.164	0.0610	0.0510	0.137	0.0209	7.78
Male	2.92 $\pm$	0.0486 $\pm$	0.0316 $\pm$	0.0066 $\pm$	0.213 $\pm$	0.833 $\pm$	87.9 $\pm$	0.0310	0.793 $\pm$	0.0269 $\pm$	0.157 $\pm$	0.0956 $\pm$	0.202 $\pm$	1.57 $\pm$	0.0408 $\pm$	109 $\pm$
	2.37	0.0350	0.0357	0.0020	0.0683	16.7	34.1	$\pm$ 0.485	0.121	0.0075	0.164	0.0617	0.0462	0.214	0.0286	13.5

Table 11c. Median of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by sex.

Sex	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
Female	2.40	0.0279	0.0585	0.0103	0.231	0.368	60.1	0.0168	0.707	0.0265	0.137	0.133	0.135	1.43	0.0662	97.7
Male	2.31	0.0726	0.0306	0.0070	0.192	0.702	86.8	0.0265	0.752	0.0261	0.119	0.0851	0.199	1.53	0.0465	103

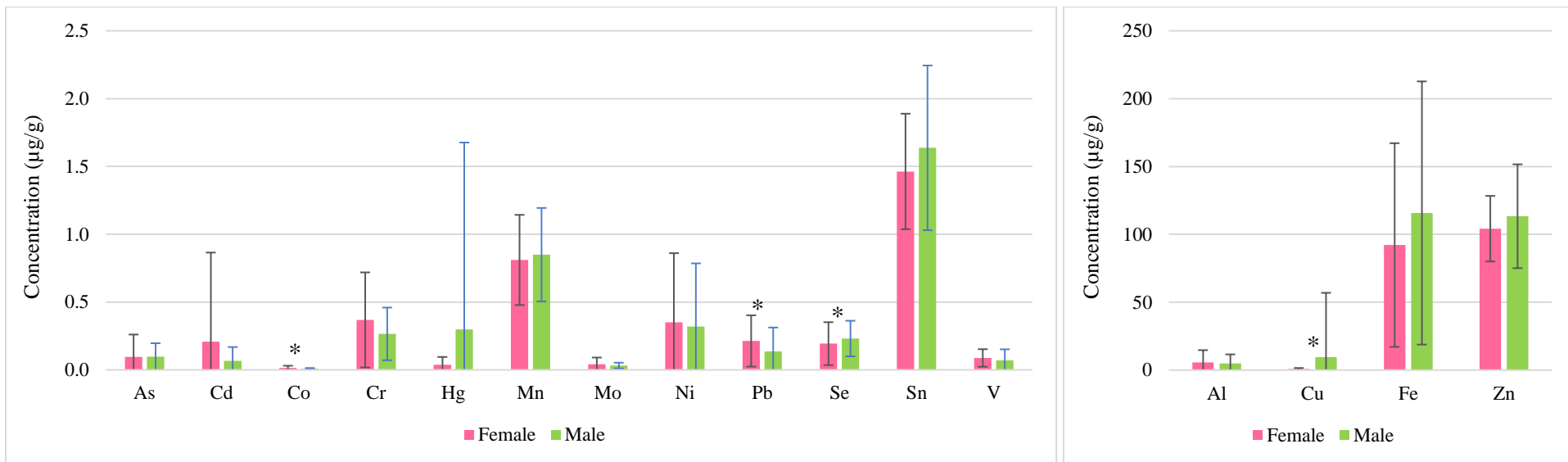


Figure 5a. Arithmetic mean and standard deviation of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by sex. Metals with a significant difference between sexes are indicated with an asterisk.

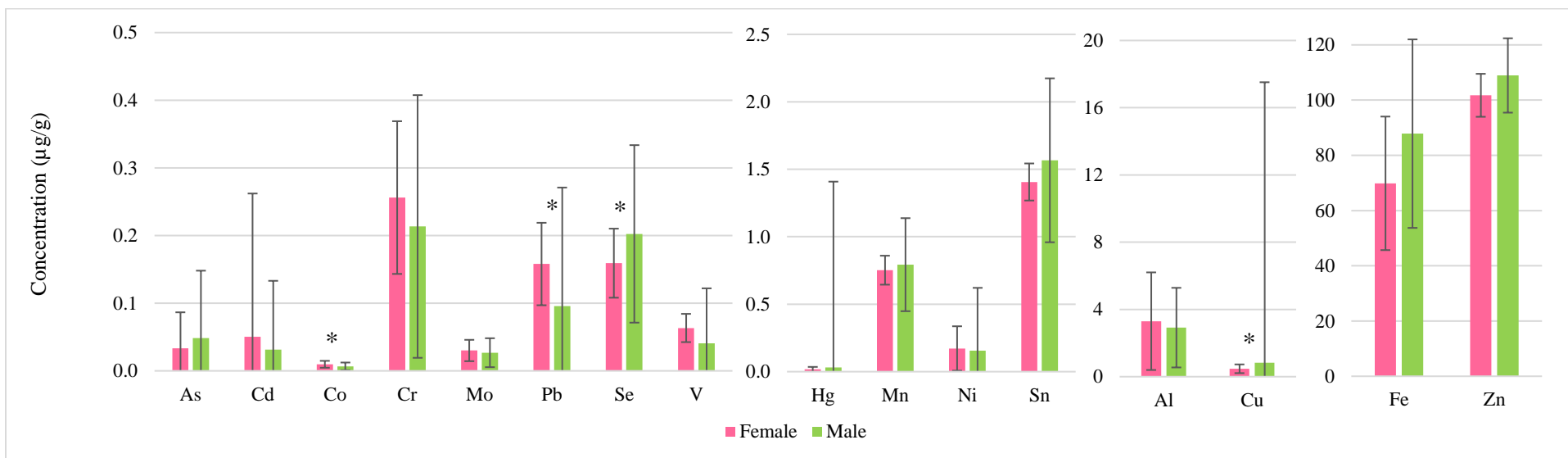


Figure 5b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by sex. Metals with a significant difference between sexes are indicated with an asterisk.



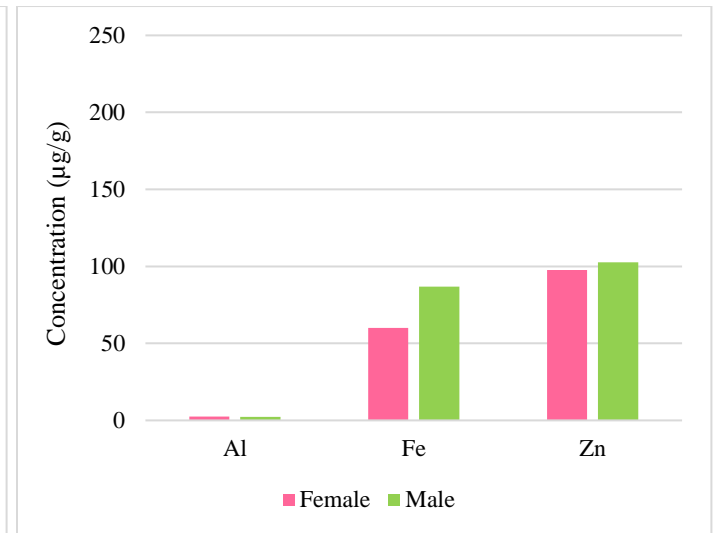
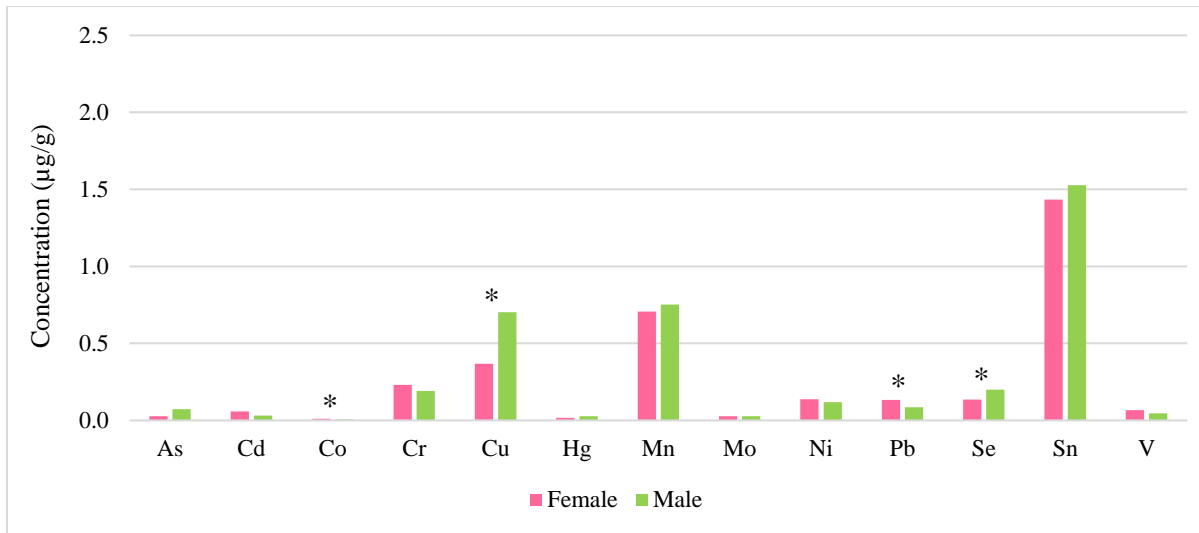


Figure 5c. Median of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by sex. Metals with a significant difference between sexes are indicated with an asterisk.

Table 12a. Arithmetic mean and standard deviation of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by NWHI region (Southern, Central, Northern) and sex.

Region - Sex	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
Southern Female	5.01 $\pm$	0.0290 $\pm$	0.0622 $\pm$	0.0165 $\pm$	0.351 $\pm$	0.893 $\pm$	133 $\pm$	0.0465 $\pm$	0.740 $\pm$	0.0324 $\pm$	0.408 $\pm$	0.169 $\pm$	0.289 $\pm$	1.38 $\pm$	0.0635 $\pm$	101 $\pm$
	5.49	0.0342	0.0818	0.0254	0.369	1.20	86.7	0.0906	0.360	0.0259	0.545	0.183	0.264	0.297	0.0537	21.1
Southern Male	8.01 $\pm$	0.0760 $\pm$	0.0707 $\pm$	0.0106 $\pm$	0.310 $\pm$	25.5 $\pm$	154 $\pm$	0.0641 $\pm$	0.976 $\pm$	0.0302 $\pm$	0.474 $\pm$	0.221 $\pm$	0.233 $\pm$	1.56 $\pm$	0.0508 $\pm$	129 $\pm$
	9.24	0.0988	0.123	0.0072	0.250	79.4	141	0.133	0.484	0.0131	0.627	0.274	0.159	0.286	0.0646	57.5
Central Female	6.81 $\pm$	0.122 $\pm$	0.374 $\pm$	0.0121 $\pm$	0.389 $\pm$	0.776 $\pm$	89.1 $\pm$	0.0390 $\pm$	0.860 $\pm$	0.0539 $\pm$	0.335 $\pm$	0.218 $\pm$	0.180 $\pm$	1.55 $\pm$	0.0830 $\pm$	115 $\pm$
	12.4	0.203	1.01	0.0107	0.294	0.722	82.9	0.0371	0.346	0.0710	0.603	0.170	0.0823	0.520	0.0642	26.9
Central Male	3.37 $\pm$	0.118 $\pm$	0.0665 $\pm$	0.0068 $\pm$	0.271 $\pm$	0.755 $\pm$	97.2 $\pm$	0.562 $\pm$	0.760 $\pm$	0.0296 $\pm$	0.254 $\pm$	0.0858 $\pm$	0.254 $\pm$	1.75 $\pm$	0.0888 $\pm$	106 $\pm$
	4.82	0.108	0.100	0.0044	0.172	0.456	55.4	1.98	0.220	0.0165	0.387	0.0417	0.125	0.821	0.100	20.3
Northern Female	5.57 $\pm$	0.143 $\pm$	0.161 $\pm$	0.0174 $\pm$	0.448 $\pm$	0.509 $\pm$	56.2 $\pm$	0.0356 $\pm$	0.810 $\pm$	0.0390 $\pm$	0.284 $\pm$	0.292 $\pm$	0.148 $\pm$	1.48 $\pm$	0.0845 $\pm$	95.5 $\pm$
	7.53	0.198	0.224	0.0120	0.457	0.382	32.7	0.0526	0.308	0.0252	0.386	0.242	0.0603	0.438	0.0468	17.7
Northern Male	2.56 $\pm$	0.0956 $\pm$	0.0845 $\pm$	0.0051 $\pm$	0.142 $\pm$	0.437 $\pm$	72.1 $\pm$	0.0332 $\pm$	0.884 $\pm$	0.0197 $\pm$	0.137 $\pm$	0.128 $\pm$	0.144 $\pm$	1.43 $\pm$	0.0470 $\pm$	102 $\pm$
	0.744	0.0969	0.0779	0.0041	0.0196	0.0557	33.7	0.0220	0.333	0.0056	0.0210	0.105	0.0647	0.287	0.0494	13.2

Table 12b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by NWHI region (Southern, Central, Northern) and sex.

Region - Sex	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
Southern Female	3.10 $\pm$	0.0169 $\pm$	0.0269 $\pm$	0.0075 $\pm$	0.224 $\pm$	0.516 $\pm$	109 $\pm$	0.0135 $\pm$	0.675 $\pm$	0.0250 $\pm$	0.181 $\pm$	0.122 $\pm$	0.220 $\pm$	1.35 $\pm$	0.0404 $\pm$	99.1 $\pm$
	3.40	0.0212	0.0507	0.0158	0.229	0.744	53.7	0.0562	0.223	0.0161	0.338	0.113	0.163	0.184	0.0333	13.1
Southern Male	4.59 $\pm$	0.0256 $\pm$	0.0228 $\pm$	0.0090 $\pm$	0.234 $\pm$	1.64 $\pm$	100 $\pm$	0.0264 $\pm$	0.868 $\pm$	0.0279 $\pm$	0.222 $\pm$	0.134 $\pm$	0.199 $\pm$	1.53 $\pm$	0.0214 $\pm$	120 $\pm$
	5.46	0.0584	0.0725	0.0043	0.148	46.9	83.2	0.0785	0.286	0.0077	0.371	0.162	0.0937	0.169	0.0382	34.0
Central Female	3.61 $\pm$	0.0456 $\pm$	0.0891 $\pm$	0.0085 $\pm$	0.314 $\pm$	0.524 $\pm$	63.1 $\pm$	0.0258 $\pm$	0.805 $\pm$	0.0346 $\pm$	0.151 $\pm$	0.174 $\pm$	0.163 $\pm$	1.48 $\pm$	0.0632 $\pm$	112 $\pm$
	6.26	0.103	0.511	0.0054	0.149	0.365	41.9	0.0188	0.175	0.0359	0.305	0.0861	0.0416	0.263	0.0325	13.6
Central Male	2.32 $\pm$	0.0798 $\pm$	0.0425 $\pm$	0.0058 $\pm$	0.226 $\pm$	0.614 $\pm$	85.6 $\pm$	0.0397 $\pm$	0.736 $\pm$	0.0263 $\pm$	0.117 $\pm$	0.0771 $\pm$	0.227 $\pm$	1.64 $\pm$	0.0635 $\pm$	104 $\pm$
	2.44	0.0544	0.0508	0.0022	0.0868	0.231	28.0	1.00	0.111	0.0083	0.196	0.0211	0.0632	0.415	0.0507	10.3
Northern Female	3.41 $\pm$	0.0458 $\pm$	0.0766 $\pm$	0.0147 $\pm$	0.307 $\pm$	0.375 $\pm$	48.8 $\pm$	0.0142 $\pm$	0.764 $\pm$	0.0336 $\pm$	0.165 $\pm$	0.211 $\pm$	0.138 $\pm$	1.42 $\pm$	0.0718 $\pm$	94.2 $\pm$
	4.92	0.129	0.146	0.0079	0.299	0.250	21.4	0.0344	0.201	0.0165	0.252	0.158	0.0394	0.286	0.0306	11.6
Northern Male	2.48 $\pm$	0.0503 $\pm$	0.0327 $\pm$	0.0041 $\pm$	0.141 $\pm$	0.434 $\pm$	67.2 $\pm$	0.0255 $\pm$	0.844 $\pm$	0.0192 $\pm$	0.136 $\pm$	0.104 $\pm$	0.135 $\pm$	1.41 $\pm$	0.0321 $\pm$	101 $\pm$
	0.842	0.110	0.0881	0.0047	0.0222	0.0631	38.1	0.0249	0.377	0.0063	0.0237	0.118	0.0732	0.325	0.0559	14.9

Table 12c. Median of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by NWHI region (Southern, Central, Northern) and sex.

Region - Sex	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
Southern Female	1.98	0.0145	0.0137	0.0063	0.197	0.392	125	0.0103	0.586	0.0240	0.102	0.125	0.203	1.46	0.0577	98.9
Southern Male	2.80	0.0171	0.0180	0.0082	0.212	0.880	130	0.0265	1.05	0.0306	0.216	0.168	0.172	1.59	0.0365	103
Central Female	2.89	0.0324	0.0672	0.0090	0.276	0.310	56.9	0.0217	0.740	0.0253	0.120	0.151	0.133	1.49	0.0615	103
Central Male	1.90	0.0776	0.0371	0.0050	0.200	0.701	86.8	0.0193	0.685	0.0219	0.0972	0.0833	0.250	1.53	0.0666	102
Northern Female	2.99	0.0526	0.0672	0.0149	0.193	0.447	49.4	0.0175	0.707	0.0386	0.169	0.192	0.135	1.56	0.0909	91.4
Northern Male	2.70	0.0790	0.0939	0.0035	0.133	0.413	62.3	0.0426	0.808	0.0172	0.136	0.0786	0.115	1.28	0.0240	106

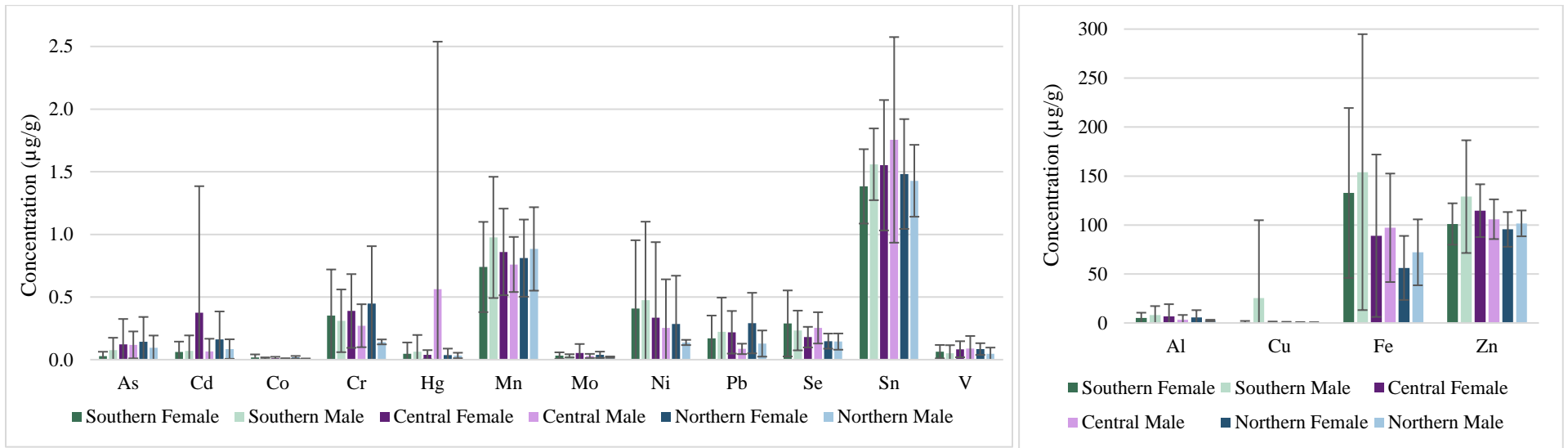


Figure 6a. Arithmetic mean and standard deviation of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by NWHI region (Southern, Central, and Northern) and sex. No metals are indicated with an asterisk as there was no significant difference found.

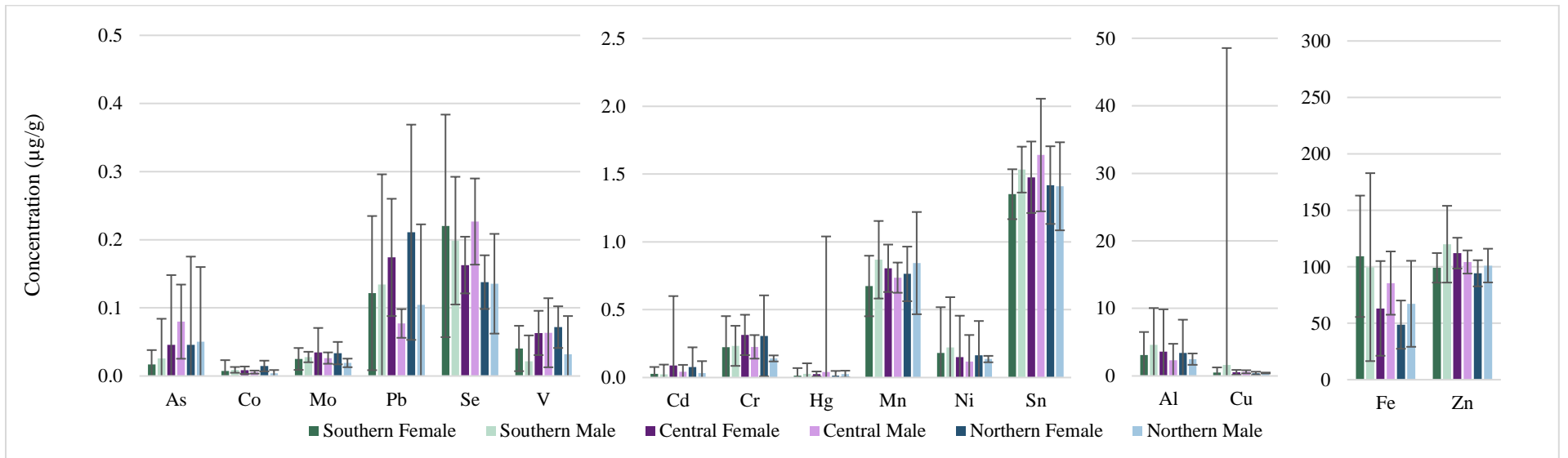


Figure 6b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by NWHI region (Southern, Central, and Northern) and sex. No metals are indicated with an asterisk as there was no significant difference found.

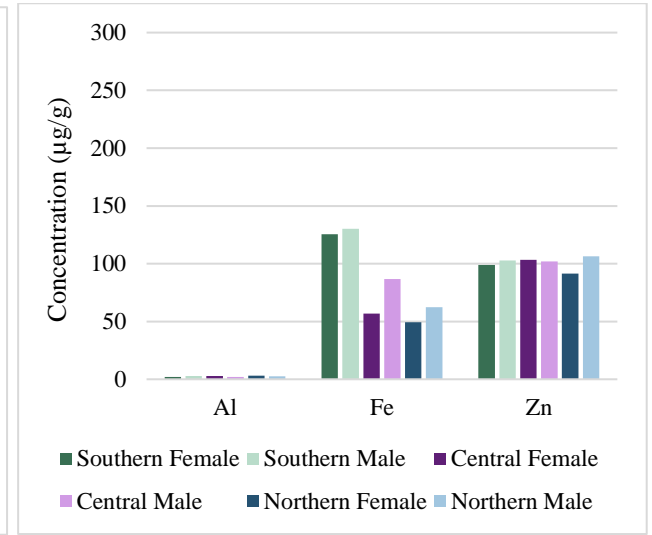
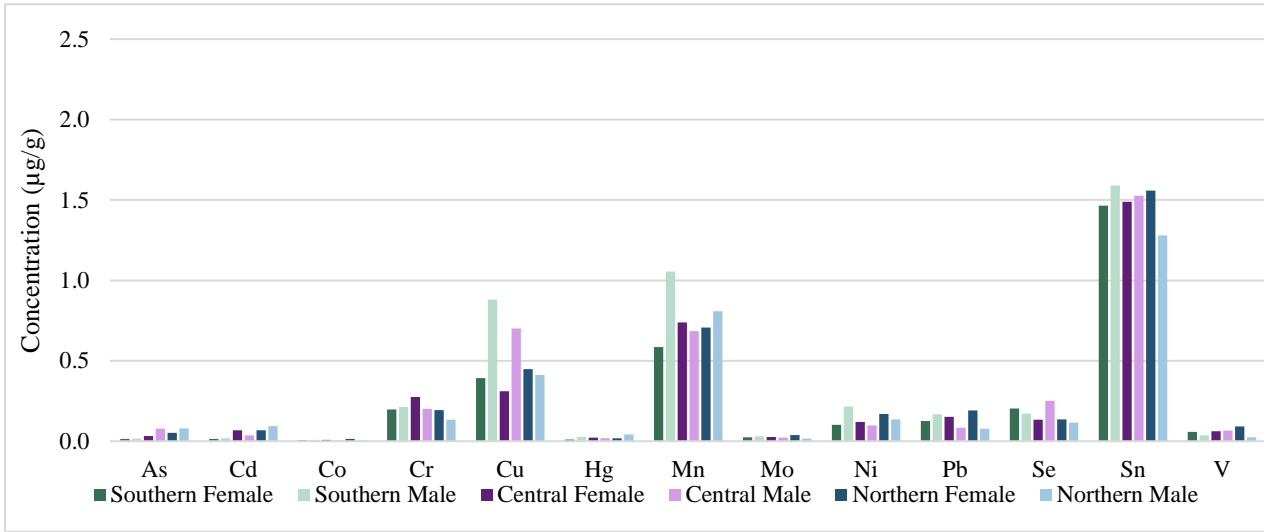


Figure 6c. Median of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by NWHI region (Southern, Central, and Northern) and sex. No metals are indicated with an asterisk as there was no significant difference found.

Table 13a. Arithmetic mean and standard deviation of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by age class (adult, juvenile, pup).

Age Class	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
<b>Adult</b>	4.60 $\pm$	0.0542 $\pm$	0.133 $\pm$	0.0142 $\pm$	0.234 $\pm$	0.562 $\pm$	69.0 $\pm$	0.0786 $\pm$	0.773 $\pm$	0.0308 $\pm$	0.273 $\pm$	0.220 $\pm$	0.185 $\pm$	1.31 $\pm$	0.0679 $\pm$	104 $\pm$
	4.15	0.0673	0.114	0.0197	0.162	1.12	62.8	0.117	0.280	0.0239	0.411	0.237	0.156	0.296	0.0393	47.8
<b>Juvenile</b>	4.98 $\pm$	0.100 $\pm$	0.0963 $\pm$	0.0081 $\pm$	0.229 $\pm$	0.524 $\pm$	73.9 $\pm$	0.0358 $\pm$	0.740 $\pm$	0.0275 $\pm$	0.198 $\pm$	0.129 $\pm$	0.205 $\pm$	1.55 $\pm$	0.109 $\pm$	104 $\pm$
	9.77	0.133	0.151	0.0057	0.145	0.423	46.4	0.0513	0.301	0.0150	0.275	0.107	0.0890	0.651	0.0806	23.8
<b>Pup</b>	4.72 $\pm$	0.0842 $\pm$	0.163 $\pm$	0.0119 $\pm$	0.397 $\pm$	8.77 $\pm$	137 $\pm$	0.246 $\pm$	0.889 $\pm$	0.0480 $\pm$	0.417 $\pm$	0.185 $\pm$	0.195 $\pm$	1.58 $\pm$	0.0519 $\pm$	111 $\pm$
	5.93	0.143	0.673	0.0096	0.366	44.6	95.5	1.30	0.357	0.0534	0.565	0.215	0.164	0.333	0.0543	25.5

Table 13b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by age class (adult, juvenile, pup).

Age Class	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
<b>Adult</b>	3.55 $\pm$	0.0296 $\pm$	0.0955 $\pm$	0.0088 $\pm$	0.195 $\pm$	0.317 $\pm$	49.5 $\pm$	0.0365 $\pm$	0.728 $\pm$	0.0251 $\pm$	0.132 $\pm$	0.143 $\pm$	0.148 $\pm$	1.27 $\pm$	0.0556 $\pm$	87.8 $\pm$
	1.87	0.0303	0.0510	0.0089	0.0728	0.506	28.2	0.0526	0.126	0.0108	0.185	0.107	0.0701	0.133	0.0176	21.5
<b>Juvenile</b>	2.62 $\pm$	0.0473 $\pm$	0.0487 $\pm$	0.0065 $\pm$	0.192 $\pm$	0.385 $\pm$	64.4 $\pm$	0.0212 $\pm$	0.691 $\pm$	0.0243 $\pm$	0.105 $\pm$	0.106 $\pm$	0.186 $\pm$	1.46 $\pm$	0.0847 $\pm$	102 $\pm$
	3.39	0.0462	0.0522	0.0020	0.0501	0.146	16.1	0.0178	0.104	0.0052	0.0952	0.0372	0.0308	0.226	0.0279	8.23
<b>Pup</b>	2.96 $\pm$	0.0299 $\pm$	0.0221 $\pm$	0.0087 $\pm$	0.275 $\pm$	1.03 $\pm$	112 $\pm$	0.0201 $\pm$	0.827 $\pm$	0.0351 $\pm$	0.218 $\pm$	0.115 $\pm$	0.157 $\pm$	1.55 $\pm$	0.0311 $\pm$	108 $\pm$
	1.97	0.0473	0.223	0.0032	0.121	14.8	31.6	0.430	0.118	0.0177	0.187	0.0711	0.0545	0.110	0.0180	8.45

Table 13c. Median of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by age class (adult, juvenile, pup).

Age Class	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
<b>Adult</b>	2.84	0.0303	0.0839	0.0079	0.184	0.294	49.1	0.0335	0.702	0.0231	0.0923	0.131	0.133	1.34	0.0670	99.0
<b>Juvenile</b>	2.12	0.0414	0.0529	0.0059	0.192	0.361	61.0	0.0182	0.651	0.0224	0.0869	0.0983	0.183	1.38	0.0897	98.9
<b>Pup</b>	2.25	0.0333	0.0180	0.0085	0.251	0.817	123	0.0228	0.781	0.0302	0.196	0.104	0.162	1.57	0.0365	105

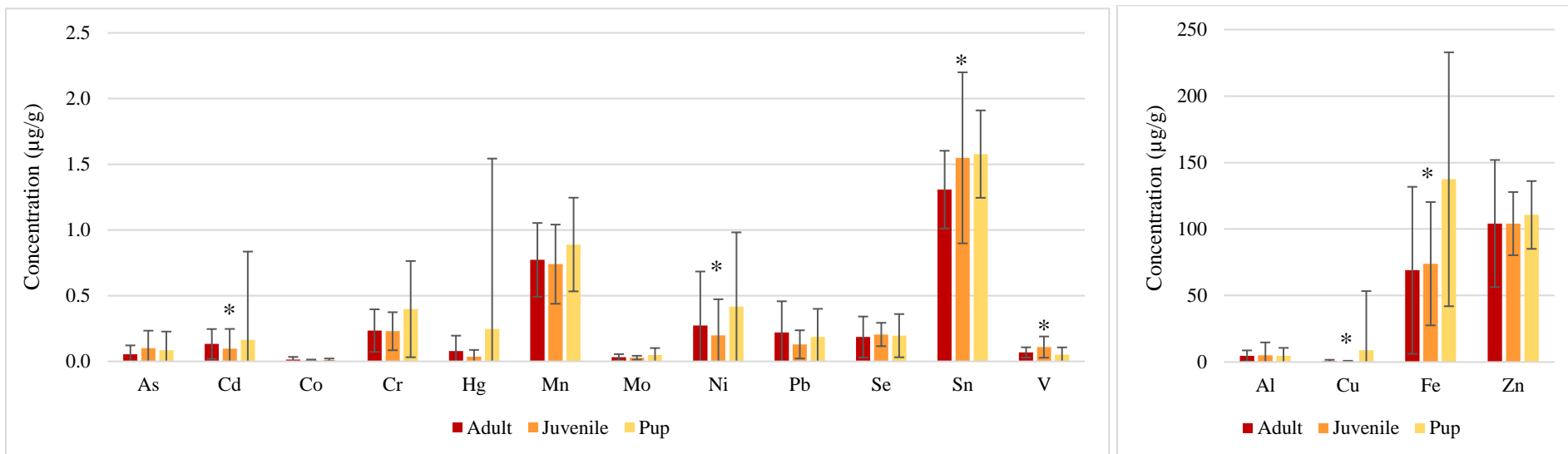


Figure 7a. Arithmetic mean and standard deviation of 16 heavy metal concentrations (µg/g) in HMS bone by age class (adult, juvenile, pup). Metals with a significant difference among age classes are indicated with an asterisk.

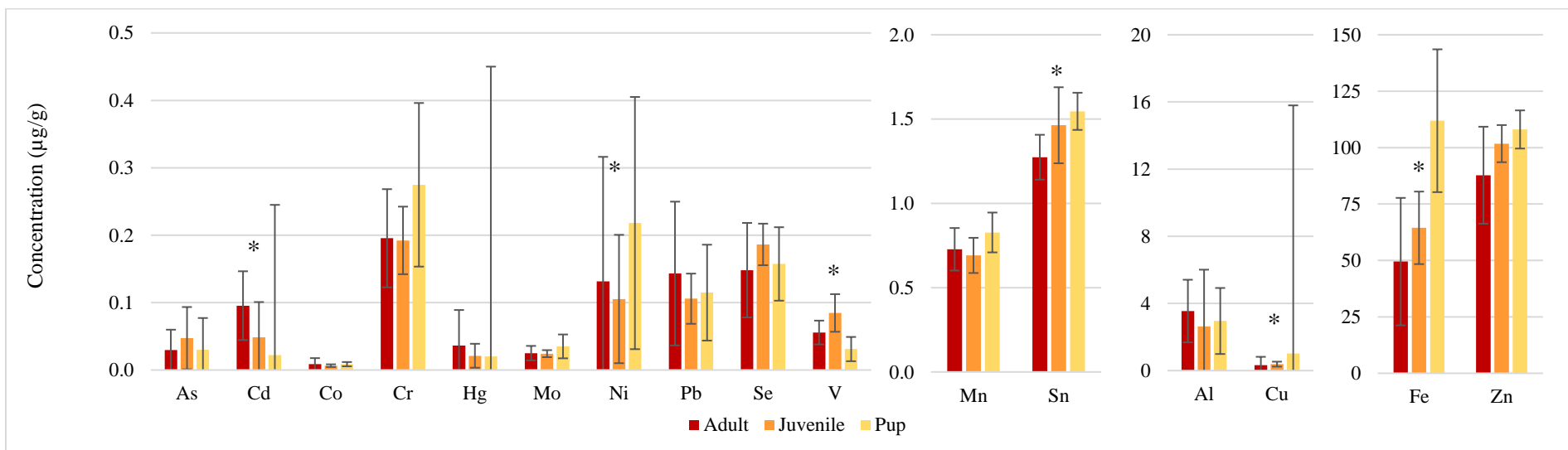


Figure 7b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations (µg/g) in HMS bone by age class (adult, juvenile, pup). Metals with a significant difference among age classes are indicated with an asterisk.

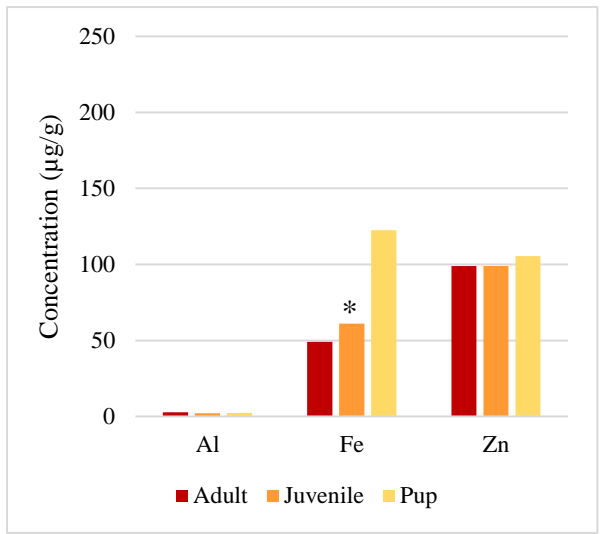
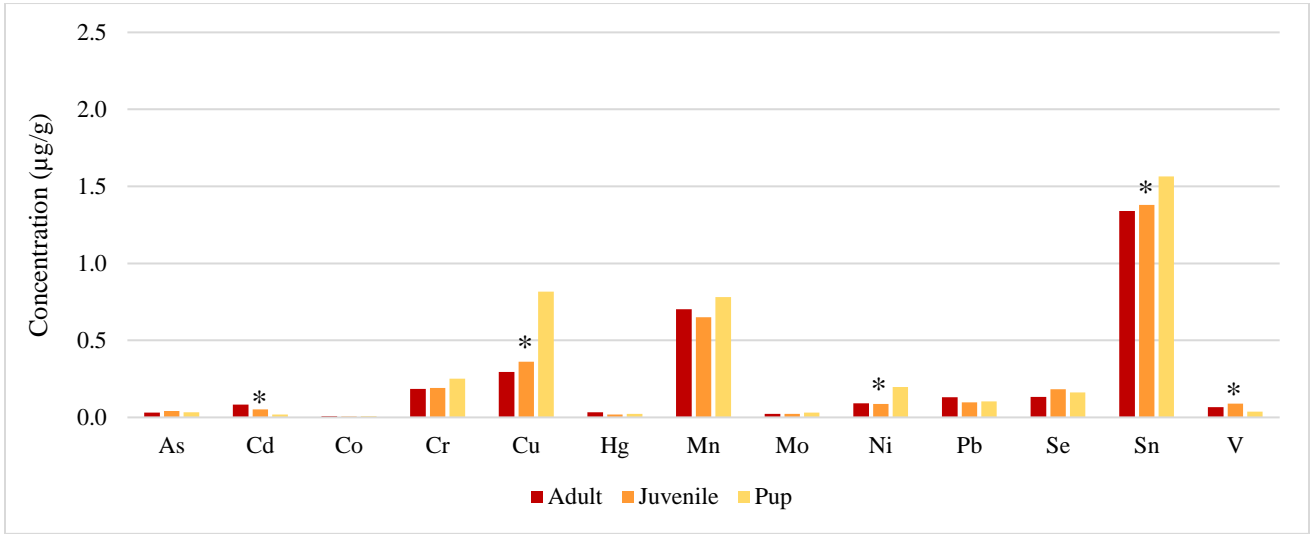


Figure 7c. Median of 16 heavy metal concentration (µg/g) in HMS bone by age class (adult, juvenile, pup). Metals with a significant difference among age classes are indicated with an asterisk.

Table 14a. Arithmetic mean and standard deviation of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by NWHI region (Southern, Central, Northern) and age class (adult, juvenile, pup).

Region – Age class	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
<b>Southern Adult</b>	8.98 ± 6.36	0.0703 ± 0.104	0.170 ± 0.0691	0.0243 ± 0.0034	0.240 ± 0.122	1.41 ± 2.10	132 ± 90.8	0.162 ± 0.207	0.969 ± 0.403	0.0210 ± 0.0106	0.597 ± 0.663	0.279 ± 0.318	0.322 ± 0.252	1.46 ± 0.214	0.0835 ± 0.0662	147 ± 69.1
	<b>Southern Juvenile</b>	1.68 ± 0.212	0.0241 ± 0.0154	0.0143 ± 0.0044	0.0055 ± 0.0036	0.125 ± 0.0437	0.216 ± 0.0309	47.9 ± 9.59	0.0221 ± 0.0261	0.478 ± 0.0620	0.0240 ± 0.0191	0.0699 ± 0.0198	0.0719 ± 0.0349	0.203 ± 0.0742	1.09 ± 0.187	0.0913 ± 0.0535
<b>Southern Pup</b>	6.48 ± 8.05	0.0536 ± 0.0697	0.0467 ± 0.0971	0.0112 ± 0.0085	0.386 ± 0.328	18.0 ± 65.8	159 ± 119	0.0224 ± 0.0160	0.924 ± 0.442	0.0369 ± 0.0212	0.413 ± 0.551	0.246 ± 0.266	0.224 ± 0.205	1.55 ± 0.244	0.0435 ± 0.0495	110 ± 31.6
<b>Central Adult</b>	2.89 ± 1.35	0.0409 ± 0.0315	0.182 ± 0.170	0.0086 ± 0.0072	0.175 ± 0.0697	0.297 ± 0.0688	37.0 ± 14.1	0.0567 ± 0.0324	0.688 ± 0.191	0.0342 ± 0.0273	0.0763 ± 0.0534	0.218 ± 0.238	0.134 ± 0.0642	1.27 ± 0.321	0.0543 ± 0.0290	102 ± 10.5
	<b>Central Juvenile</b>	5.35 ± 11.1	0.0957 ± 0.106	0.101 ± 0.116	0.0069 ± 0.0042	0.265 ± 0.139	0.558 ± 0.404	80.3 ± 51.6	0.0452 ± 0.0606	0.793 ± 0.288	0.0291 ± 0.0154	0.147 ± 0.154	0.126 ± 0.0799	0.232 ± 0.0884	1.63 ± 0.749	0.111 ± 0.0900
<b>Central Pup</b>	4.05 ± 3.23	0.167 ± 0.231	0.438 ± 1.25	0.0121 ± 0.0125	0.450 ± 0.355	1.36 ± 0.592	150 ± 73.4	0.805 ± 2.42	0.866 ± 0.284	0.0710 ± 0.0853	0.625 ± 0.766	0.132 ± 0.128	0.233 ± 0.134	1.79 ± 0.373	0.0484 ± 0.0299	116 ± 25.0
<b>Northern Adult</b>	3.05 ± 1.27	0.0685 ± 0.0725	0.0849 ± 0.0509	0.0129 ± 0.0165	0.331 ± 0.235	0.213 ± 0.108	48.0 ± 34.2	0.0511 ± 0.0610	0.671 ± 0.213	0.0407 ± 0.0302	0.102 ± 0.0512	0.216 ± 0.233	0.146 ± 0.0811	1.22 ± 0.369	0.0712 ± 0.0251	72.5 ± 34.6
	<b>Northern Juvenile</b>	9.62 ± 13.4	0.327 ± 0.253	0.280 ± 0.373	0.0144 ± 0.0082	0.304 ± 0.216	0.609 ± 0.532	46.9 ± 22.0	0.0130 ± 0.0069	0.837 ± 0.414	0.0322 ± 0.0131	0.473 ± 0.693	0.268 ± 0.270	0.138 ± 0.0542	1.68 ± 0.304	0.131 ± 0.0313
<b>Northern Pup</b>	2.51 ± 1.98	0.058 ± 0.0776	0.0800 ± 0.110	0.0116 ± 0.0069	0.430 ± 0.519	0.597 ± 0.283	82.0 ± 49.2	0.0241 ± 0.0158	0.885 ± 0.315	0.0277 ± 0.0119	0.171 ± 0.116	0.160 ± 0.194	0.120 ± 0.0443	1.40 ± 0.375	0.0490 ± 0.0463	104 ± 12.2

Table 14b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by NWHI region (Southern, Central, Northern) and age class (adult, juvenile, pup).

Region – Age class	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
<b>Southern Adult</b>	6.56 ± 5.57	0.0256 ± 0.0910	0.151 ± 0.0606	0.0135 ± 0.0297	0.215 ± 0.107	0.710 ± 1.84	91.1 ± 79.6	0.0518 ± 0.182	0.892 ± 0.354	0.0184 ± 0.0093	0.285 ± 0.581	0.192 ± 0.279	0.248 ± 0.221	1.45 ± 0.188	0.0531 ± 0.0580	137 ± 60.6
	<b>Southern Juvenile</b>	1.68 ± 0.208	0.0203 ± 0.0151	0.0138 ± 0.0043	0.0045 ± 0.0035	0.118 ± 0.0428	0.214 ± 0.0303	47.2 ± 9.39	0.0135 ± 0.0256	0.475 ± 0.0608	0.0198 ± 0.0188	0.0677 ± 0.0194	0.0668 ± 0.0342	0.192 ± 0.0728	1.08 ± 0.183	0.0820 ± 0.0524
<b>Southern Pup</b>	3.68 ± 3.95	0.0232 ± 0.0342	0.0167 ± 0.0476	0.0084 ± 0.0042	0.272 ± 0.161	1.43 ± 32.3	123 ± 58.5	0.0156 ± 0.0078	0.831 ± 0.217	0.0325 ± 0.0104	0.213 ± 0.270	0.152 ± 0.130	0.181 ± 0.101	1.53 ± 0.119	0.0221 ± 0.0242	106 ± 15.5
<b>Central Adult</b>	2.68 ± 1.08	0.0346 ± 0.0252	0.126 ± 0.136	0.0064 ± 0.0057	0.164 ± 0.0558	0.290 ± 0.0550	34.5 ± 11.3	0.0457 ± 0.0259	0.666 ± 0.153	0.0280 ± 0.0218	0.0637 ± 0.0427	0.138 ± 0.191	0.123 ± 0.0514	1.23 ± 0.257	0.0475 ± 0.0232	102 ± 8.42
	<b>Central Juvenile</b>	2.64 ± 4.75	0.0540 ± 0.0451	0.0678 ± 0.0497	0.0059 ± 0.0018	0.234 ± 0.0593	0.440 ± 0.173	69.5 ± 22.1	0.0276 ± 0.0259	0.754 ± 0.123	0.0263 ± 0.0066	0.0928 ± 0.0659	0.110 ± 0.0342	0.216 ± 0.0378	1.53 ± 0.321	0.0877 ± 0.0385
<b>Central Pup</b>	3.05 ± 2.00	0.0767 ± 0.143	0.0503 ± 0.776	0.0083 ± 0.0078	0.335 ± 0.220	1.24 ± 0.367	136 ± 45.5	0.0456 ± 1.50	0.832 ± 0.176	0.0439 ± 0.0529	0.303 ± 0.475	0.0966 ± 0.0795	0.204 ± 0.0833	1.76 ± 0.231	0.0425 ± 0.0185	113 ± 15.5
<b>Northern Adult</b>	2.86 ± 1.01	0.0397 ± 0.0580	0.0711 ± 0.0407	0.0081 ± 0.0132	0.267 ± 0.188	0.193 ± 0.0864	39.5 ± 27.3	0.0290 ± 0.0488	0.642 ± 0.171	0.0339 ± 0.0242	0.0913 ± 0.0409	0.136 ± 0.187	0.128 ± 0.0649	1.18 ± 0.295	0.0667 ± 0.0201	50.3 ± 27.7
	<b>Northern Juvenile</b>	4.37 ± 15.2	0.251 ± 0.286	0.144 ± 0.422	0.0125 ± 0.0093	0.261 ± 0.244	0.416 ± 0.602	43.1 ± 24.9	0.0114 ± 0.0078	0.773 ± 0.468	0.0300 ± 0.0148	0.188 ± 0.784	0.192 ± 0.305	0.130 ± 0.0613	1.66 ± 0.344	0.129 ± 0.0354
<b>Northern Pup</b>	2.00 ± 1.47	0.0231 ± 0.0575	0.0237 ± 0.0816	0.0089 ± 0.0051	0.273 ± 0.384	0.529 ± 0.209	71.0 ± 36.5	0.0135 ± 0.0117	0.837 ± 0.233	0.0257 ± 0.0088	0.138 ± 0.0863	0.103 ± 0.144	0.113 ± 0.0328	1.36 ± 0.278	0.0315 ± 0.0343	103 ± 9.04



Table 14c. Median of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by NWHI region (Southern, Central, Northern) and age class (adult, juvenile, pup).

<b>Region – Age class</b>	<b>Al</b>	<b>As</b>	<b>Cd</b>	<b>Co</b>	<b>Cr</b>	<b>Cu</b>	<b>Fe</b>	<b>Hg</b>	<b>Mn</b>	<b>Mo</b>	<b>Ni</b>	<b>Pb</b>	<b>Se</b>	<b>Sn</b>	<b>V</b>	<b>Zn</b>
<b>Southern Adult</b>	11.2	0.0279	0.183	0.0082	0.192	0.387	146	0.0232	1.13	0.0204	0.193	0.133	0.174	1.44	0.109	116
<b>Southern Juvenile</b>	1.64	0.0220	0.0137	0.0058	0.135	0.215	49.2	0.0119	0.477	0.0154	0.0689	0.0581	0.201	1.09	0.0697	94.2
<b>Southern Pup</b>	2.68	0.0156	0.0128	0.0090	0.259	0.822	139	0.0211	0.823	0.0304	0.171	0.157	0.172	1.54	0.0287	101
<b>Central Adult</b>	2.61	0.0314	0.0886	0.0048	0.162	0.302	38.7	0.0631	0.645	0.0244	0.0605	0.119	0.119	1.31	0.0552	100
<b>Central Juvenile</b>	2.10	0.0695	0.0585	0.0056	0.236	0.389	63.4	0.0217	0.694	0.0244	0.0972	0.100	0.224	1.38	0.0873	102
<b>Central Pup</b>	3.09	0.0831	0.0322	0.0069	0.328	1.38	132	0.0287	0.786	0.0319	0.335	0.0961	0.210	1.73	0.0377	110
<b>Northern Adult</b>	2.91	0.0432	0.0752	0.0069	0.235	0.185	38.0	0.0360	0.662	0.0324	0.0880	0.140	0.117	1.13	0.0694	82.8
<b>Northern Juvenile</b>	2.40	0.300	0.0672	0.0151	0.193	0.543	48.4	0.0164	0.710	0.0386	0.0797	0.128	0.142	1.58	0.131	91.4
<b>Northern Pup</b>	1.76	0.0081	0.0468	0.0144	0.190	0.500	62.3	0.0237	0.808	0.0242	0.136	0.0786	0.115	1.28	0.0283	106

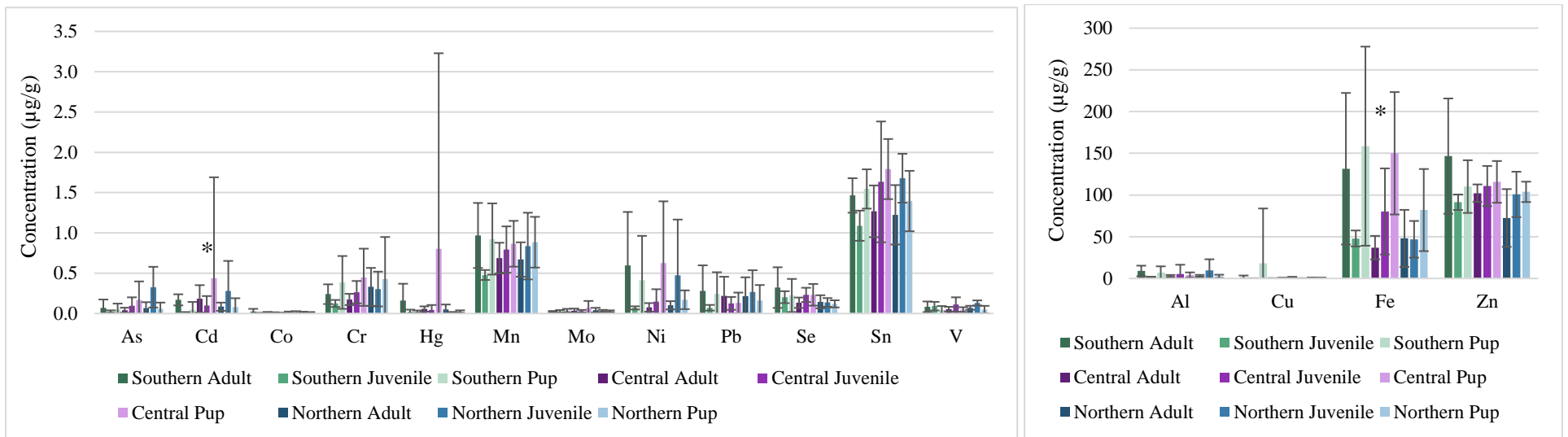


Figure 8a. Arithmetic mean and standard deviation of 16 heavy metal concentrations (µg/g) in HMS bone by NWHI region (Southern, Central, Northern) and age class (adult, juvenile, pup). Metals with a significant difference among regional-age classes are indicated with an asterisk.

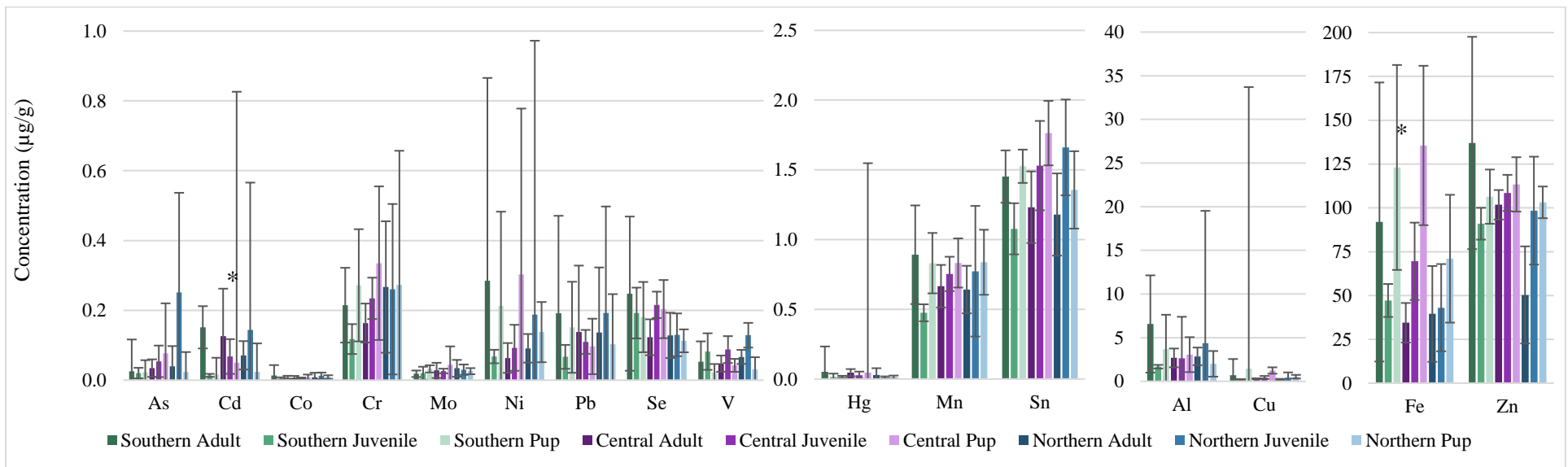


Figure 8b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations (µg/g) in HMS bone by NWHI region (Southern, Central, Northern) and age class (adult, juvenile, pup). Metals with a significant difference among regional-age classes are indicated with an asterisk.

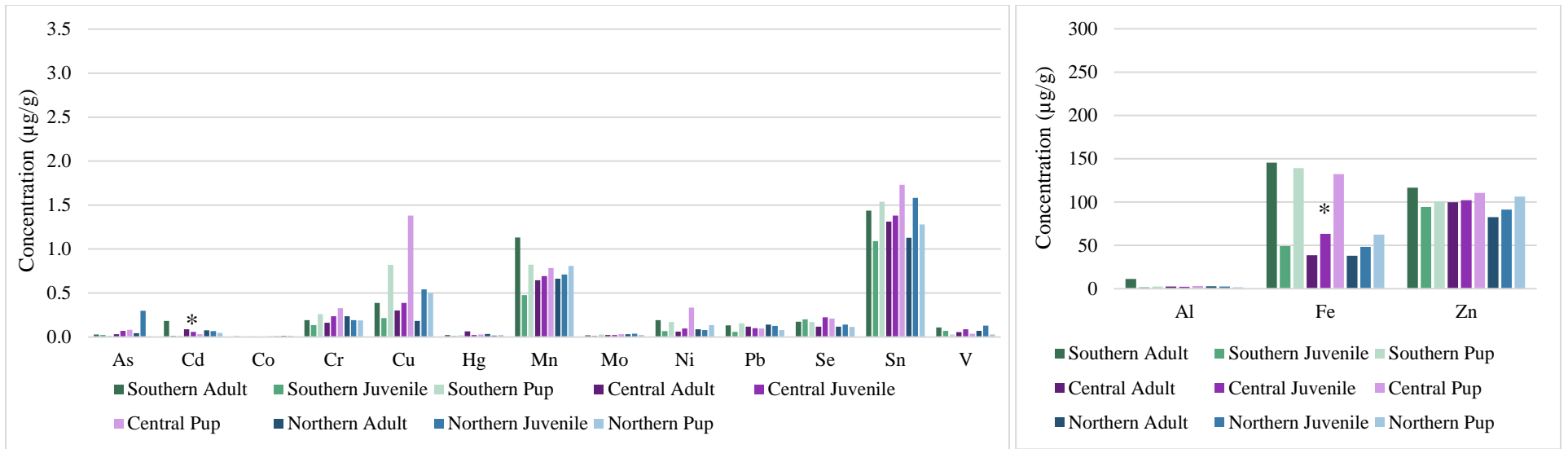


Figure 8c. Median of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by NWHI region (Southern, Central, Northern) and age class (adult, juvenile, pup). Metals with a significant difference among regional-age classes are indicated with an asterisk.

Age class by sex also significantly affected the heavy metal concentrations of Cd, Cu, Fe, Sn, and V, the same as age class alone minus Ni (Table 9). Adult female Cd concentration was significantly greater than pup male. Pup male and female Cu concentrations were significantly greater than adult female. Pup female Fe concentration was significantly greater than adult and juvenile female. Pup female Sn concentration was significantly greater than adult female. Juvenile female V concentration was significantly greater than pup male (Table 15 and Figure 9).

When compared to other studies (Table 4), the heavy metal concentrations in the HMS were lower than most other marine mammal bone studies. Aluminum, As, Cd, Cu, Ni, Se, and Pb were all at lower concentrations within the HMS compared to other marine mammal bones. Cobalt, Cr, Fe, Hg, Mn, Mo, V, and Zn were around similar concentrations between the HMS and other marine mammal bones. Tin concentration of HMS ( $1.48 \pm 0.0911 \mu\text{g/g}$ ) was one order of magnitude greater than *Phoca vitulina* (harbor seal) concentration ( $0.104 \mu\text{g/g}$ ) (Agusa et al. 2011). Zinc concentration of the HMS ( $102 \pm 6.30 \mu\text{g/g}$ ) was one order of magnitude greater than *Zalophus californianus* (Californian sea lion) ( $44.17 \pm 38.65 \mu\text{g/g}$ ) but not *Pontoporia blainvillei* (Franciscana dolphin) ( $251.2 \pm 74.4 \mu\text{g/g}$ ) or *Stenella coeruleoalba* (striped dolphin) ( $382.0 \pm 23.9 \mu\text{g/g}$ ) concentrations (Garcia-Garin et al., 2021; Honda et al., 1986; Szteren & Auriol-Gamboa 2013).

#### 4.2 Potential Prey

Prey taxa had a significant effect on 10 of the 16 heavy metal concentrations in the monk seal bones - As, Cd, Co, Cu, Fe, Mn, Mo, Ni, Sn, and Zn (Table 9). Teleost Fe, Mn, and Sn concentrations were significantly greater than cephalopod and crustacean. Teleost As, Cd, Cu, Mo, and Ni concentrations were significantly less than cephalopod and crustacean. Teleost Co and Zn concentrations were significantly less than cephalopod (Table 16 and Figure 10). All 10 of the prey families (Acanthuridae, Balistidae, Congridae, Holocentridae, Labridae, Muraenidae, Octopodidae, Palinuridae, Scaridae, and Serranidae) had a significant effect on 15 of the 16 the heavy metal concentrations - As, Cd, Co, Cr, Cu, Fe, Hg, Mn, Mo, Ni, Pb, Se, Sn, V, and Zn (Table 9). Aluminum concentrations were the only heavy metal not significantly affected by the prey family. Nickel concentrations had the most significant differences among families with 11 family pairs exhibiting significant difference. Cadmium, Cu, and Mo all had 10 family pairs that were significantly different. Octopodidae was significantly different 37 times (out of 720 combinations) from another family within all 16 metals. Palinuridae was significantly

Table 15a. Arithmetic mean and standard deviation of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by sex and age class (adult, juvenile, pup).

Sex – Age Class	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
<b>Female Adult</b>	5.01 $\pm$	0.0233 $\pm$	0.183 $\pm$	0.0243 $\pm$	0.232 $\pm$	0.257 $\pm$	57.0 $\pm$	0.0895 $\pm$	0.847 $\pm$	0.0366 $\pm$	0.248 $\pm$	0.286 $\pm$	0.190 $\pm$	1.17 $\pm$	0.0578 $\pm$	101 $\pm$
	4.64	0.0154	0.149	0.0278	0.155	0.0851	57.1	0.100	0.278	0.0357	0.437	0.242	0.153	0.321	0.0374	14.5
<b>Male Adult</b>	4.62 $\pm$	0.115 $\pm$	0.0983 $\pm$	0.0067 $\pm$	0.156 $\pm$	0.436 $\pm$	89.2 $\pm$	0.114 $\pm$	0.705 $\pm$	0.0228 $\pm$	0.384 $\pm$	0.143 $\pm$	0.279 $\pm$	1.50 $\pm$	0.0797 $\pm$	131 $\pm$
	4.77	0.110	0.0772	0.0032	0.0258	0.273	90.7	0.196	0.247	0.0083	0.533	0.0815	0.213	0.251	0.0593	78.2
<b>Female Juvenile</b>	6.80 $\pm$	0.122 $\pm$	0.0989 $\pm$	0.0096 $\pm$	0.221 $\pm$	0.514 $\pm$	59.5 $\pm$	0.0196 $\pm$	0.739 $\pm$	0.0275 $\pm$	0.259 $\pm$	0.160 $\pm$	0.178 $\pm$	1.41 $\pm$	0.122 $\pm$	100 $\pm$
	13.5	0.162	0.174	0.0069	0.145	0.459	29.7	0.0161	0.361	0.0142	0.362	0.143	0.0810	0.398	0.0626	25.7
<b>Male Juvenile</b>	4.12 $\pm$	0.124 $\pm$	0.0810 $\pm$	0.0081 $\pm$	0.302 $\pm$	0.683 $\pm$	93.8 $\pm$	0.0662 $\pm$	0.817 $\pm$	0.0341 $\pm$	0.134 $\pm$	0.100 $\pm$	0.253 $\pm$	1.88 $\pm$	0.125 $\pm$	109 $\pm$
	6.15	0.127	0.129	0.0053	0.177	0.466	64.5	0.0883	0.262	0.0196	0.122	0.0445	0.101	1.06	0.117	23.7
<b>Female Pup</b>	5.10 $\pm$	0.112 $\pm$	0.365 $\pm$	0.0159 $\pm$	0.630 $\pm$	1.31 $\pm$	157 $\pm$	0.0288 $\pm$	0.885 $\pm$	0.0637 $\pm$	0.540 $\pm$	0.242 $\pm$	0.217 $\pm$	1.73 $\pm$	0.0701 $\pm$	112 $\pm$
	3.95	0.216	1.10	0.0114	0.464	1.05	82.7	0.0375	0.343	0.0732	0.673	0.198	0.229	0.398	0.0680	27.4
<b>Male Pup</b>	5.30 $\pm$	0.0773 $\pm$	0.0499 $\pm$	0.0084 $\pm$	0.278 $\pm$	16.9 $\pm$	135 $\pm$	0.476 $\pm$	0.908 $\pm$	0.0322 $\pm$	0.401 $\pm$	0.154 $\pm$	0.205 $\pm$	1.55 $\pm$	0.0382 $\pm$	110 $\pm$
	7.73	0.0807	0.0940	0.0066	0.224	63.9	112	1.86	0.403	0.0249	0.547	0.232	0.119	0.255	0.0425	28.5

Table 15b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by sex and age class (adult, juvenile, pup).

Sex – Age Class	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
<b>Female Adult</b>	3.94 $\pm$	0.0181 $\pm$	0.133 $\pm$	0.0149 $\pm$	0.197 $\pm$	0.243 $\pm$	42.1 $\pm$	0.0471 $\pm$	0.809 $\pm$	0.0251 $\pm$	0.114 $\pm$	0.213 $\pm$	0.154 $\pm$	1.14 $\pm$	0.0469 $\pm$	100 $\pm$
	3.22	0.0107	0.103	0.0193	0.107	0.0590	39.6	0.0694	0.193	0.0247	0.303	0.168	0.106	0.222	0.0259	10.1
<b>Male Adult</b>	3.37 $\pm$	0.0340 $\pm$	0.0888 $\pm$	0.0086 $\pm$	0.156 $\pm$	0.273 $\pm$	50.3 $\pm$	0.0416 $\pm$	0.605 $\pm$	0.0223 $\pm$	0.176 $\pm$	0.143 $\pm$	0.187 $\pm$	1.09 $\pm$	0.0488 $\pm$	83.2 $\pm$
	4.18	0.0960	0.0676	0.0028	0.0226	0.239	79.5	0.172	0.217	0.0073	0.467	0.0714	0.187	0.220	0.0520	68.6
<b>Female Juvenile</b>	2.85 $\pm$	0.0540 $\pm$	0.0458 $\pm$	0.0072 $\pm$	0.182 $\pm$	0.373 $\pm$	54.8 $\pm$	0.0146 $\pm$	0.675 $\pm$	0.0244 $\pm$	0.130 $\pm$	0.126 $\pm$	0.162 $\pm$	1.36 $\pm$	0.103 $\pm$	97.1 $\pm$
	6.82	0.0819	0.0882	0.0035	0.0734	0.232	15.0	0.0082	0.183	0.0072	0.183	0.0724	0.0410	0.201	0.0317	13.0
<b>Male Juvenile</b>	2.55 $\pm$	0.0774 $\pm$	0.0454 $\pm$	0.0067 $\pm$	0.254 $\pm$	0.524 $\pm$	80.0 $\pm$	0.0317 $\pm$	0.787 $\pm$	0.0298 $\pm$	0.0931 $\pm$	0.0917 $\pm$	0.235 $\pm$	1.69 $\pm$	0.0988 $\pm$	107 $\pm$
	4.02	0.0831	0.0840	0.0034	0.115	0.305	42.2	0.0577	0.171	0.0128	0.0800	0.0291	0.0662	0.695	0.0765	15.5
<b>Female Pup</b>	3.68 $\pm$	0.0272 $\pm$	0.0351 $\pm$	0.0117 $\pm$	0.445 $\pm$	0.976 $\pm$	139 $\pm$	0.0112 $\pm$	0.828 $\pm$	0.0450 $\pm$	0.329 $\pm$	0.182 $\pm$	0.162 $\pm$	1.69 $\pm$	0.0448 $\pm$	109 $\pm$
	2.14	0.117	0.596	0.0062	0.252	0.569	44.9	0.0204	0.187	0.0398	0.366	0.108	0.125	0.217	0.0370	14.9
<b>Male Pup</b>	3.00 $\pm$	0.0371 $\pm$	0.0204 $\pm$	0.0068 $\pm$	0.214 $\pm$	1.35 $\pm$	104 $\pm$	0.0298 $\pm$	0.834 $\pm$	0.0271 $\pm$	0.193 $\pm$	0.0907 $\pm$	0.180 $\pm$	1.53 $\pm$	0.0232 $\pm$	107 $\pm$
	3.67	0.0384	0.0447	0.0031	0.107	30.4	53.4	0.885	0.192	0.0118	0.260	0.110	0.0564	0.121	0.0202	13.5

Table 15c. Median of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by sex and age class (adult, juvenile, pup).

	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
<b>Female Adult</b>	2.94	0.0226	0.133	0.0134	0.186	0.285	38.0	0.0606	0.813	0.0205	0.0922	0.173	0.134	1.11	0.0470	99.1
<b>Male Adult</b>	2.84	0.104	0.0588	0.0070	0.162	0.387	49.1	0.0434	0.599	0.0204	0.158	0.131	0.218	1.53	0.0727	100
<b>Female Juvenile</b>	1.97	0.0392	0.0585	0.0087	0.193	0.279	56.2	0.0165	0.638	0.0244	0.0797	0.122	0.142	1.38	0.130	94.9
<b>Male Juvenile</b>	2.10	0.0776	0.0327	0.0059	0.286	0.476	83.1	0.0193	0.752	0.0284	0.100	0.0851	0.258	1.53	0.0873	102
<b>Female Pup</b>	4.28	0.0141	0.0185	0.0144	0.602	0.911	126	0.0079	0.776	0.0405	0.324	0.151	0.140	1.73	0.0457	114
<b>Male Pup</b>	2.25	0.0413	0.0180	0.0073	0.194	0.831	92.5	0.0265	0.790	0.0278	0.136	0.0781	0.172	1.59	0.0355	105

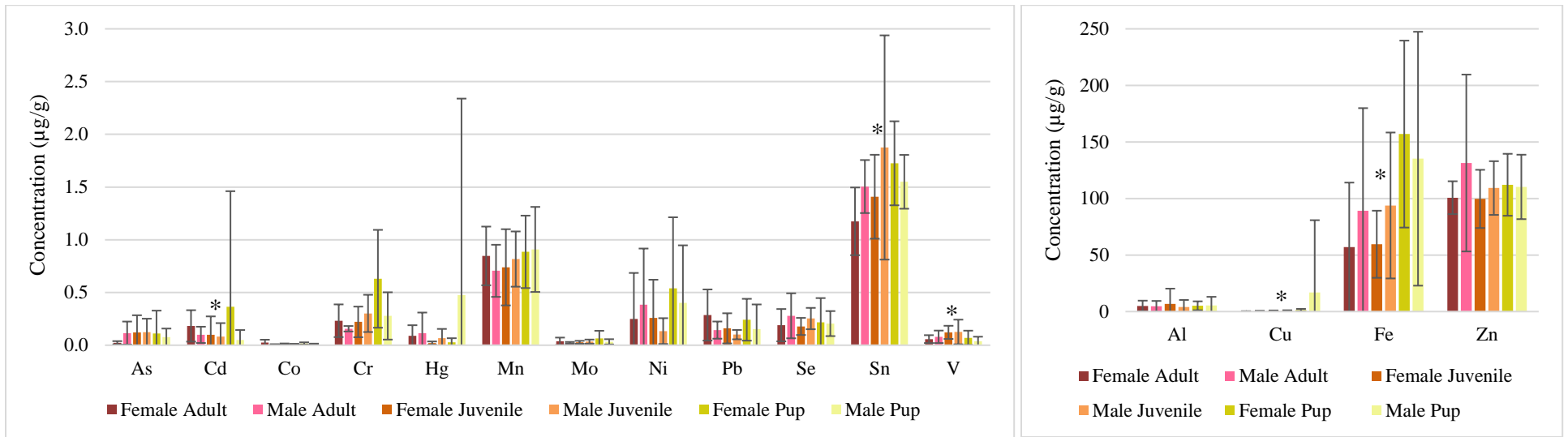


Figure 9a. Arithmetic mean and standard deviation of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by sex and age class (adult, juvenile, pup). Metals with a significant difference among sex-age classes are indicated with an asterisk.

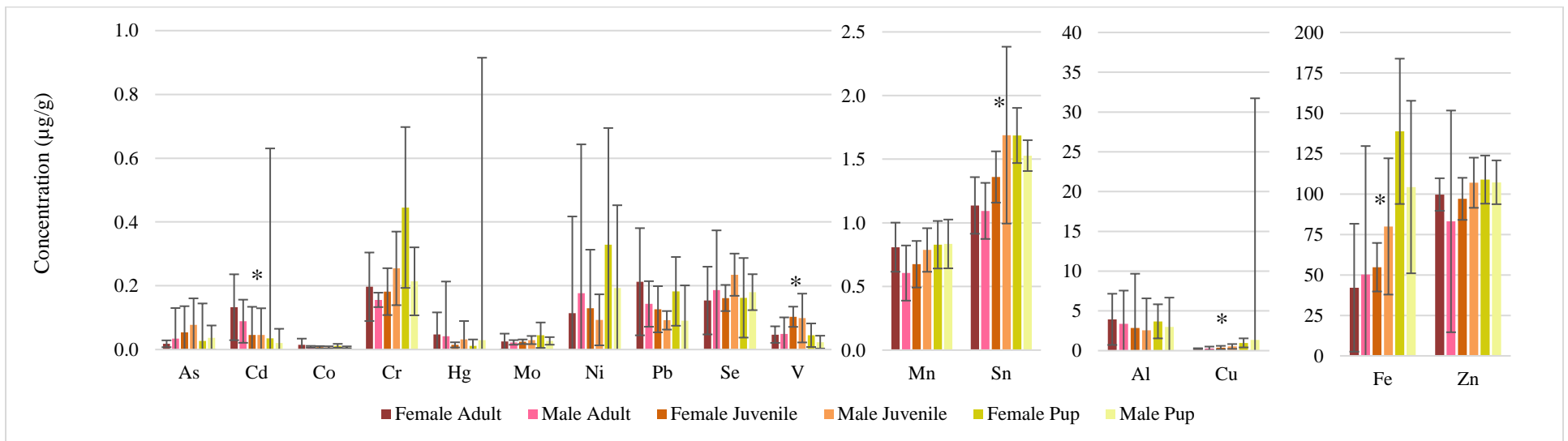


Figure 9b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by sex and age class (adult, juvenile, pup). Metals with a significant difference among sex-age classes are indicated with an asterisk.

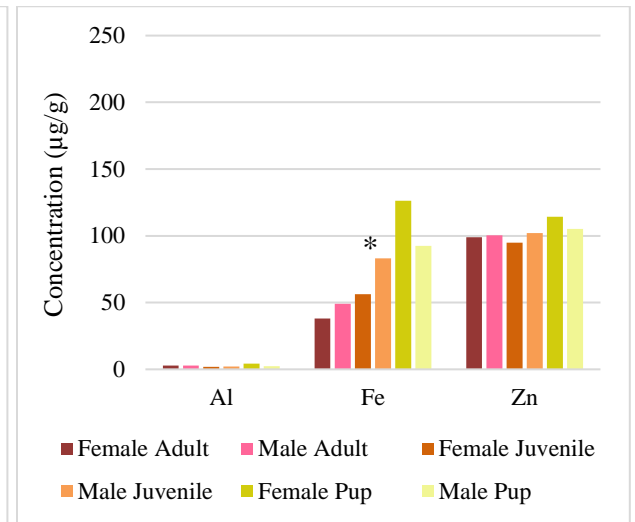
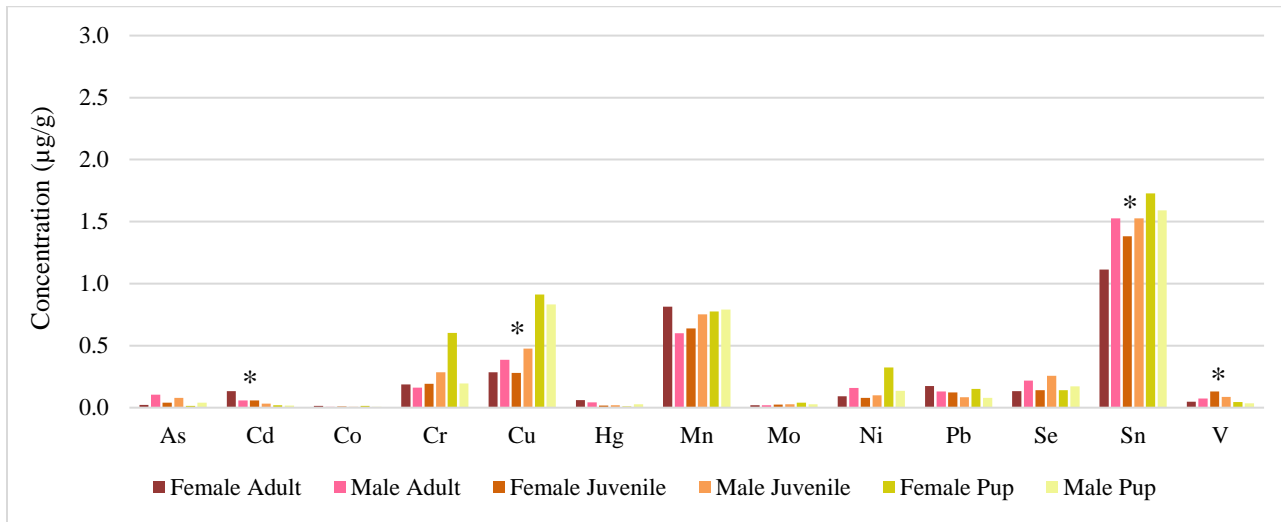


Figure 9c. Median of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone by sex and age class (adult, juvenile, pup). Metals with a significant difference among sex-age classes are indicated with an asterisk.

Table 16a. Arithmetic mean and standard deviation of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in whole prey by taxa in the NWHI Southern region.

Taxa	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
Teleosts	32.0 $\pm$	13.2 $\pm$	0.590 $\pm$	0.0381 $\pm$	2.37 $\pm$	2.88 $\pm$	107 $\pm$	0.196 $\pm$	7.55 $\pm$	0.126 $\pm$	0.881	0.822 $\pm$	1.27 $\pm$	1.26 $\pm$	0.939 $\pm$	57.7 $\pm$
	52.6	14.7	0.740	0.0248	1.76	5.01	150	0.351	11.1	0.0720	$\pm$ 3.77	1.92	0.764	0.736	1.01	62.0
Crustaceans	36.4 $\pm$	108 $\pm$	4.43 $\pm$	0.0424 $\pm$	1.69 $\pm$	100 $\pm$	23.6 $\pm$	0.139 $\pm$	0.998 $\pm$	0.201 $\pm$	1.01 $\pm$	0.280 $\pm$	1.26 $\pm$	0.358 $\pm$	0.566 $\pm$	62.5 $\pm$
	35.9	37.0	4.00	0.0110	0.887	21.7	8.39	0.102	0.217	0.0526	0.426	0.265	0.207	0.746	0.291	13.7
Cephalopods	66.9 $\pm$	126 $\pm$	20.4 $\pm$	0.157 $\pm$	1.35 $\pm$	98.5 $\pm$	48.0 $\pm$	0.114 $\pm$	1.83 $\pm$	0.834 $\pm$	2.25 $\pm$	0.365 $\pm$	3.51 $\pm$	0.166 $\pm$	0.829 $\pm$	95.7 $\pm$
	96.8	90.0	9.15	0.147	0.768	35.6	22.5	0.0799	0.271	0.322	1.08	0.352	5.65	0.370	0.209	49.2

Table 16b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in whole prey by taxa in the NWHI Southern region.

Taxa	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
Teleosts	13.2 $\pm$	7.55 $\pm$	0.347 $\pm$	0.0315 $\pm$	1.86 $\pm$	2.00 $\pm$	79.3 $\pm$	0.0714 $\pm$	4.27 $\pm$	0.108 $\pm$	0.324 $\pm$	0.268	1.05 $\pm$	0.988 $\pm$	0.495 $\pm$	41.8 $\pm$
	13.0	3.63	0.183	0.0061	0.434	1.24	37.2	0.0866	2.74	0.0178	0.932	$\pm$ 0.474	0.189	0.182	0.249	15.3
Crustaceans	26.1 $\pm$	101 $\pm$	3.58 $\pm$	0.0410 $\pm$	1.51 $\pm$	97.9 $\pm$	22.2 $\pm$	0.118 $\pm$	0.978 $\pm$	0.195 $\pm$	0.923 $\pm$	0.217 $\pm$	1.25 $\pm$	0.135 $\pm$	0.501 $\pm$	61.1 $\pm$
	22.2	22.9	2.48	0.0068	0.550	13.4	5.20	0.0635	0.134	0.0326	0.264	0.164	0.128	0.463	0.180	8.50
Cephalopods	23.4 $\pm$	106 $\pm$	19.0 $\pm$	0.118 $\pm$	1.18 $\pm$	93.3 $\pm$	43.8 $\pm$	0.0947 $\pm$	1.82 $\pm$	0.791 $\pm$	2.04 $\pm$	0.259 $\pm$	2.02 $\pm$	0.0459	0.805 $\pm$	87.4 $\pm$
	67.1	62.4	6.34	0.102	0.532	24.7	15.6	0.0554	0.187	0.223	0.746	0.244	3.91	$\pm$ 0.257	0.145	34.1

Table 16c. Median of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in whole prey by taxa in the NWHI Southern region.

Taxa	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
Teleosts	8.67	6.11	0.345	0.0320	2.10	1.72	87.1	0.0530	4.15	0.114	0.305	0.212	1.11	1.13	0.626	43.1
Crustaceans	20.8	104	3.04	0.0440	1.50	95.1	25.1	0.119	0.930	0.199	0.964	0.194	1.25	0.0942	0.492	61.0
Cephalopods	17.4	90.1	18.7	0.0952	1.17	90.5	43.4	0.0699	1.75	0.729	1.84	0.191	1.53	0.0329	0.809	72.9



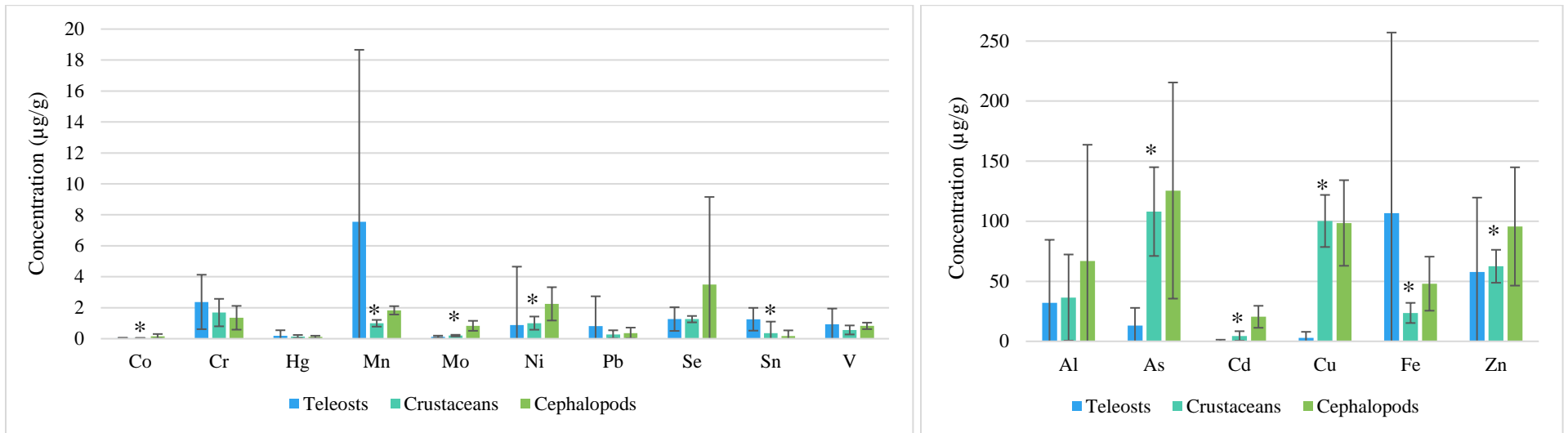


Figure 10a. Arithmetic mean and standard deviation of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in whole prey by taxa in the NWHI Southern region. Metals with a significant difference among taxa are indicated with an asterisk.

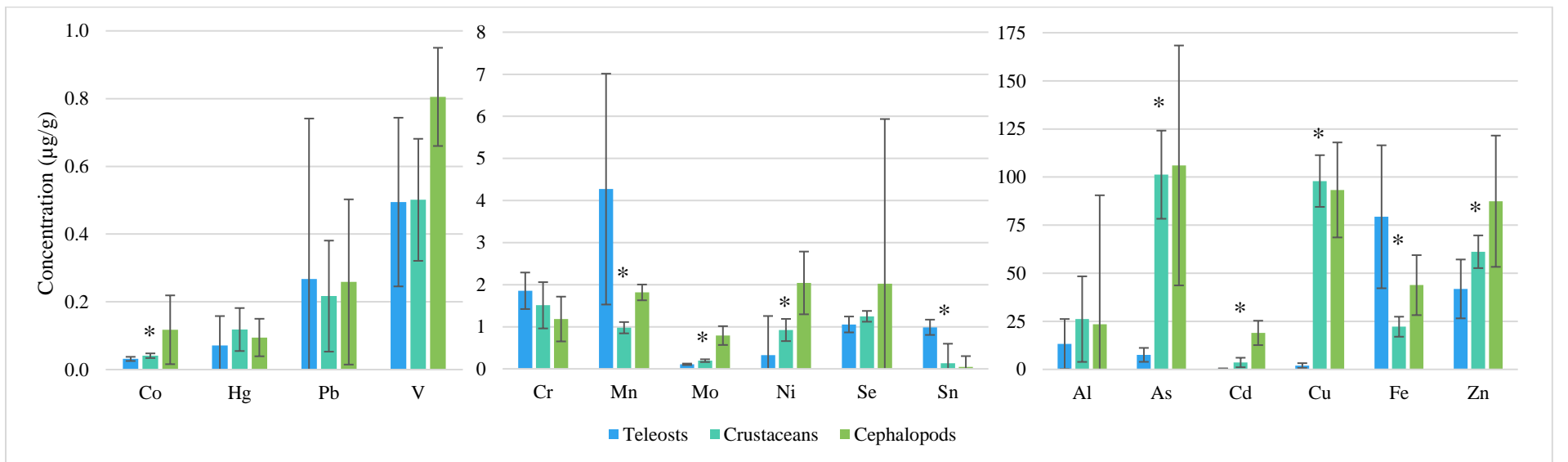


Figure 10b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in whole prey by taxa in the NWHI Southern region. Metals with a significant difference among taxa are indicated with an asterisk.

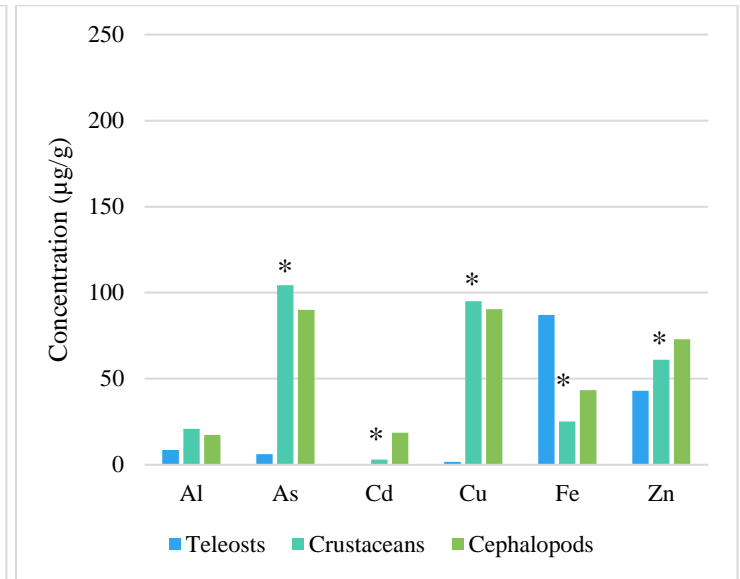
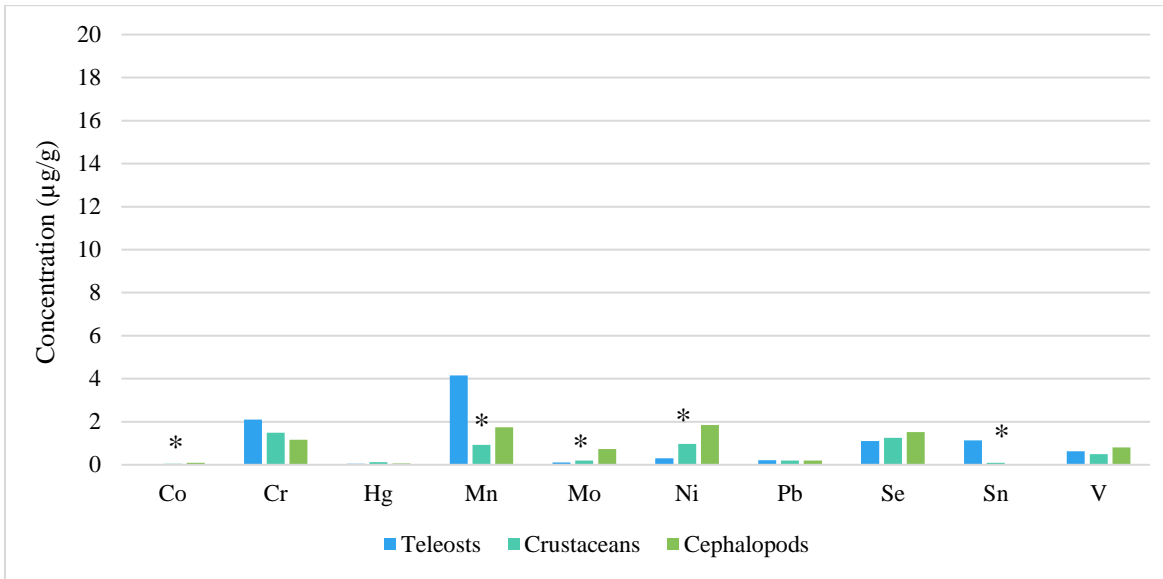


Figure 10c. Median of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in whole prey by taxa in the NWHI Southern region. Metals with a significant difference among taxa are indicated with an asterisk.

different 32 times from another family within the suite of 16 metals. Holocentridae – Octopodidae, Octopodidae – Serranidae, and Palinuridae – Scaridae all were significantly different within 7 heavy metals (Table 17, 18, and Figure 11).

When compared to other studies (Table 5), the heavy metal concentrations of the whole prey samples in this study encompassed the entire range of concentration values. Acanthuridae concentration from this study were an order of magnitude less than other studies for As, Cd, Hg, Se, and Zn and two orders of magnitude less for Co and Fe (Metian et al., 2013; Miao et al., 2001). The Miao et al. (2001) study used samples of whole organisms from French Frigate Shoals while the Metian et al. (2013) study used liver samples from New Caledonia, an island within the southeast Pacific Ocean. Labridae concentrations from this study were an order of magnitude less than other studies for Co, Ni, Se, and V and two orders of magnitude less for Fe and Hg, but an order or magnitude greater for Cr (Metian et al., 2013). Muraenidae concentrations from this study were an order of magnitude less than other studies for As, but an order of magnitude greater for Fe (Briand et al., 2018; Miao et al., 2001). The Briand et al. (2018) study used muscle samples from New Caledonia. Palinuridae concentration from this study were an order of magnitude less than other studies for Pb, Se, and Zn (Miao et al., 2001). Scaridae concentrations from this study were an order of magnitude less than other studies for Fe and Ni and two orders of magnitude less for Co and Se but an order of magnitude greater for Cr (Metian et al., 2013). Serranidae concentrations from this study were an order of magnitude less than other studies for Cd, Co, Se, and Zn and two orders of magnitude less for Cu, Fe, Ni, and V (Metian et al., 2013). The tissue choice difference between this study, whole organism, versus Metian et al. (2013) lipid rich liver may result in most of the differences.

#### **4.3 Potential Prey to Hawaiian Monk Seal**

The HMS bone concentrations were significantly different than the whole potential prey concentrations for all heavy metals (Table 9). Aluminum, As, Cd, Co, Cr, Cu, Hg, Mn, Mo, Ni, Pb, Se, and V concentrations in prey were significantly greater than the concentrations in the seal bone while Fe, Sn, and Zn concentrations were significantly less (Figure 12).

Prey taxa (seal, teleost, cephalopod, and crustacean) had a significant effect on heavy metal concentration for all 16 metals (Table 9). All three prey taxa concentrations were significantly greater than the seal for Al, As, Cd, Co, Cr, Cu, Hg, Mo, Ni, Se, and V. The teleost

Table 17a. Arithmetic mean and standard deviation of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in whole prey by family in the NWHI Southern region.

Family	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
Acanthuridae	78.7 $\pm$ 76.4	4.53 $\pm$ 4.34	0.870 $\pm$ 1.44	0.0626 $\pm$ 0.0317	2.64 $\pm$ 1.12	1.86 $\pm$ 0.710	174 $\pm$ 294	0.0392 $\pm$ 0.0339	2.77 $\pm$ 1.10	0.185 $\pm$ 0.0868	0.696 $\pm$ 0.389	1.35 $\pm$ 2.73	0.445 $\pm$ 0.187	1.33 $\pm$ 0.743	1.14 $\pm$ 0.593	29.7 $\pm$ 17.7
	Balistidae	42.5 $\pm$ 80.5	18.7 $\pm$ 8.99	0.577 $\pm$ 0.398	0.0453 $\pm$ 0.0242	3.02 $\pm$ 1.92	6.27 $\pm$ 12.3	95.9 $\pm$ 21.3	0.0410 $\pm$ 0.0255	12.1 $\pm$ 4.96	0.179 $\pm$ 0.0764	4.16 $\pm$ 9.79	1.47 $\pm$ 3.09	0.850 $\pm$ 0.320	1.16 $\pm$ 0.473	2.13 $\pm$ 1.37
Congridae	6.58 $\pm$ 0.0670	25.8 $\pm$ 3.08	0.160 $\pm$ 0.0739	0.0235 $\pm$ 0.0019	2.10 $\pm$ 0.397	4.73 $\pm$ 4.06	88.7 $\pm$ 13.0	0.368 $\pm$ 0.0315	2.48 $\pm$ 0.495	0.0949 $\pm$ 0.0044	0.225 $\pm$ 0.113	0.117 $\pm$ 0.100	1.20 $\pm$ 0.156	0.791 $\pm$ 0.731	0.0427 $\pm$ 0.0107	40.7 $\pm$ 1.52
	Holocentridae	24.8 $\pm$ 59.5	19.8 $\pm$ 20.6	0.748 $\pm$ 0.772	0.0311 $\pm$ 0.0214	2.84 $\pm$ 2.88	2.23 $\pm$ 1.66	112 $\pm$ 30.1	0.202 $\pm$ 0.208	1.64 $\pm$ 0.699	0.121 $\pm$ 0.0702	0.238 $\pm$ 0.117	0.243 $\pm$ 0.133	2.07 $\pm$ 0.993	2.10 $\pm$ 0.681	0.441 $\pm$ 0.249
Labridae	30.3 $\pm$ 28.2	17.9 $\pm$ 11.5	0.220 $\pm$ 0.143	0.0304 $\pm$ 0.0126	3.18 $\pm$ 1.79	1.43 $\pm$ 0.390	81.6 $\pm$ 26.9	0.0509 $\pm$ 0.0202	5.70 $\pm$ 3.10	0.111 $\pm$ 0.0362	0.275 $\pm$ 0.0989	1.59 $\pm$ 2.82	1.30 $\pm$ 0.727	0.771 $\pm$ 0.730	0.496 $\pm$ 0.700	47.7 $\pm$ 10.7
	Muraenidae	14.2 $\pm$ 16.3	16.3 $\pm$ 21.8	0.954 $\pm$ 0.751	0.0213 $\pm$ 0.0120	1.20 $\pm$ 0.378	3.91 $\pm$ 3.49	58.7 $\pm$ 26.9	0.722 $\pm$ 0.584	20.1 $\pm$ 22.9	0.0686 $\pm$ 0.0176	0.160 $\pm$ 0.0415	0.298 $\pm$ 0.358	1.76 $\pm$ 0.614	0.981 $\pm$ 0.724	0.612 $\pm$ 0.957
Octopodidae	66.9 $\pm$ 96.8	126 $\pm$ 90.0	20.4 $\pm$ 9.15	0.157 $\pm$ 0.147	1.35 $\pm$ 0.768	98.5 $\pm$ 35.6	48.0 $\pm$ 22.5	0.114 $\pm$ 0.0799	1.83 $\pm$ 0.271	0.834 $\pm$ 0.322	2.25 $\pm$ 1.08	0.365 $\pm$ 0.352	3.51 $\pm$ 5.65	0.166 $\pm$ 0.370	0.829 $\pm$ 0.209	95.7 $\pm$ 49.3
	Palinuridae	36.4 $\pm$ 35.9	108 $\pm$ 37.0	4.43 $\pm$ 4.00	0.0424 $\pm$ 0.0110	1.69 $\pm$ 0.887	100 $\pm$ 21.7	23.6 $\pm$ 8.39	0.139 $\pm$ 0.102	1.00 $\pm$ 0.217	0.201 $\pm$ 0.0526	1.01 $\pm$ 0.426	0.280 $\pm$ 0.265	1.26 $\pm$ 0.207	0.358 $\pm$ 0.746	0.566 $\pm$ 0.291
Scaridae	18.4 $\pm$ 15.8	2.72 $\pm$ 0.753	0.436 $\pm$ 0.215	0.0485 $\pm$ 0.0244	2.32 $\pm$ 1.08	2.10 $\pm$ 0.815	150 $\pm$ 254	0.0200 $\pm$ 0.0071	6.01 $\pm$ 3.16	0.147 $\pm$ 0.0520	0.458 $\pm$ 0.139	0.613 $\pm$ 0.820	0.984 $\pm$ 0.441	1.36 $\pm$ 0.523	1.42 $\pm$ 0.894	19.1 $\pm$ 4.62
	Serranidae	15.6 $\pm$ 23.1	4.93 $\pm$ 2.74	0.116 $\pm$ 0.0957	0.0244 $\pm$ 0.0209	0.524 $\pm$ 0.204	0.967 $\pm$ 0.420	41.6 $\pm$ 17.7	0.249 $\pm$ 0.356	2.30 $\pm$ 2.36	0.0335 $\pm$ 0.0129	0.0894 $\pm$ 0.0482	0.0753 $\pm$ 0.0823	1.50 $\pm$ 0.333	0.988 $\pm$ 0.312	0.0997 $\pm$ 0.102

Table 17b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in whole prey by family in the NWHI Southern region.

Family	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
Acanthuridae	38.5 $\pm$ 49.9	3.38 $\pm$ 2.83	0.376 $\pm$ 0.941	0.0562 $\pm$ 0.0207	2.31 $\pm$ 0.730	1.72 $\pm$ 0.464	92.9 $\pm$ 192	0.0293 $\pm$ 0.0222	2.56 $\pm$ 0.716	0.170 $\pm$ 0.0567	0.628 $\pm$ 0.254	0.538 $\pm$ 1.78	0.407 $\pm$ 0.122	1.14 $\pm$ 0.485	1.03 $\pm$ 0.387	25.6 $\pm$ 11.6
	Balistidae	11.2 $\pm$ 52.6	17.0 $\pm$ 5.87	0.449 $\pm$ 0.260	0.0403 $\pm$ 0.0158	2.52 $\pm$ 1.25	2.80 $\pm$ 8.07	93.6 $\pm$ 13.9	0.0348 $\pm$ 0.0167	11.2 $\pm$ 3.24	0.165 $\pm$ 0.0499	1.11 $\pm$ 6.40	0.456 $\pm$ 2.02	0.793 $\pm$ 0.209	1.06 $\pm$ 0.309	1.80 $\pm$ 0.897
Congridae	6.58 $\pm$ 0.0928	25.7 $\pm$ 4.27	0.151 $\pm$ 0.102	0.0234 $\pm$ 0.0027	2.08 $\pm$ 0.551	3.76 $\pm$ 5.62	88.2 $\pm$ 18.1	0.367 $\pm$ 0.0437	2.46 $\pm$ 0.686	0.0948 $\pm$ 0.0060	0.210 $\pm$ 0.157	0.0929 $\pm$ 0.139	1.20 $\pm$ 0.217	0.598 $\pm$ 1.01	0.0420 $\pm$ 0.0148	40.7 $\pm$ 2.11
	Holocentridae	7.26 $\pm$ 36.9	10.2 $\pm$ 12.8	0.540 $\pm$ 0.478	0.0253 $\pm$ 0.0133	1.98 $\pm$ 1.78	1.87 $\pm$ 1.03	109 $\pm$ 18.6	0.133 $\pm$ 0.129	1.50 $\pm$ 0.433	0.104 $\pm$ 0.0435	0.212 $\pm$ 0.0726	0.210 $\pm$ 0.0826	1.88 $\pm$ 0.579	1.92 $\pm$ 0.422	0.379 $\pm$ 0.155
Labridae	21.3 $\pm$ 18.4	14.3 $\pm$ 7.50	0.181 $\pm$ 0.0933	0.0283 $\pm$ 0.0082	2.70 $\pm$ 1.17	1.38 $\pm$ 0.255	77.6 $\pm$ 17.6	0.0471 $\pm$ 0.0132	5.09 $\pm$ 2.03	0.106 $\pm$ 0.0237	0.258 $\pm$ 0.0646	0.335 $\pm$ 1.84	1.15 $\pm$ 0.475	0.484 $\pm$ 0.477	0.307 $\pm$ 0.457	46.5 $\pm$ 7.02
	Muraenidae	9.67 $\pm$ 10.1	8.71 $\pm$ 13.5	0.593 $\pm$ 0.465	0.0193 $\pm$ 0.0074	1.14 $\pm$ 0.234	3.05 $\pm$ 2.16	53.0 $\pm$ 16.7	0.550 $\pm$ 0.362	9.98 $\pm$ 14.2	0.0666 $\pm$ 0.0109	0.155 $\pm$ 0.0257	0.174 $\pm$ 0.222	1.65 $\pm$ 0.380	0.700 $\pm$ 0.449	0.276 $\pm$ 0.593
Octopodidae	23.4 $\pm$ 67.1	106 $\pm$ 62.4	19.0 $\pm$ 6.34	0.118 $\pm$ 0.102	1.18 $\pm$ 0.532	93.3 $\pm$ 24.7	43.8 $\pm$ 15.6	0.0947 $\pm$ 0.0554	1.82 $\pm$ 0.187	0.791 $\pm$ 0.223	2.04 $\pm$ 0.746	0.259 $\pm$ 0.244	2.02 $\pm$ 3.91	0.0459 $\pm$ $\pm$ 0.257	0.805 $\pm$ 0.145	87.4 $\pm$ 34.1
	Palinuridae	26.1 $\pm$ 22.2	101 $\pm$ 22.9	3.58 $\pm$ 2.48	0.0410 $\pm$ 0.0068	1.51 $\pm$ 0.550	97.9 $\pm$ 13.4	22.2 $\pm$ 5.20	0.118 $\pm$ 0.0635	0.978 $\pm$ 0.134	0.195 $\pm$ 0.0326	0.923 $\pm$ 0.264	0.217 $\pm$ 0.164	1.25 $\pm$ 0.128	0.135 $\pm$ 0.463	0.501 $\pm$ 0.180
Scaridae	13.8 $\pm$ 9.77	2.62 $\pm$ 0.467	0.379 $\pm$ 0.133	0.0446 $\pm$ 0.0151	2.08 $\pm$ 0.670	1.98 $\pm$ 0.505	86.5 $\pm$ 158	0.0190 $\pm$ 0.0044	5.47 $\pm$ 1.96	0.141 $\pm$ 0.0322	0.440 $\pm$ 0.0859	0.336 $\pm$ 0.508	0.907 $\pm$ 0.273	1.26 $\pm$ 0.324	1.23 $\pm$ 0.554	18.6 $\pm$ 2.86
	Serranidae	7.31 $\pm$ 22.7	4.49 $\pm$ 2.68	0.0741 $\pm$ 0.0938	0.0185 $\pm$ 0.0205	0.494 $\pm$ 0.200	0.912 $\pm$ 0.412	38.0 $\pm$ 17.4	0.104 $\pm$ 0.349	1.22 $\pm$ 2.32	0.0318 $\pm$ 0.0126	0.0806 $\pm$ 0.0472	0.0523 $\pm$ 0.0806	1.47 $\pm$ 0.327	0.954 $\pm$ 0.306	0.0442 $\pm$ 0.0997

Table 17c. Median of 16 heavy metals concentration ( $\mu\text{g/g}$ ) of HMS whole prey samples by family in the Southern region of the NWHI.

Family	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
<b>Acanthuridae</b>	48.8	3.62	0.382	0.0534	2.65	2.13	90.2	0.0217	2.73	0.174	0.584	0.420	0.423	1.18	0.941	25.5
<b>Balistidae</b>	5.31	15.9	0.345	0.0407	3.14	1.91	90.6	0.0334	10.7	0.153	0.913	0.251	0.806	1.01	1.81	105
<b>Congridae</b>	6.58	25.8	0.160	0.0235	2.10	4.73	88.7	0.368	2.48	0.0949	0.225	0.117	1.20	0.791	0.0427	40.7
<b>Holocentridae</b>	4.45	5.66	0.490	0.0257	1.76	1.59	101	0.104	1.41	0.105	0.204	0.226	1.80	2.26	0.415	44.0
<b>Labridae</b>	29.3	15.5	0.216	0.0302	2.77	1.37	74.2	0.0530	4.73	0.114	0.267	0.230	1.11	0.540	0.236	46.7
<b>Muraenidae</b>	8.65	6.41	0.892	0.0188	1.21	2.59	53.1	0.526	9.54	0.0623	0.157	0.164	1.81	0.967	0.279	53.3
<b>Octopodidae</b>	17.4	90.1	18.7	0.0952	1.17	90.5	43.4	0.0699	1.75	0.729	1.84	0.191	1.53	0.0329	0.809	72.9
<b>Palinuridae</b>	20.8	104	3.04	0.0440	1.50	95.1	25.1	0.119	0.930	0.199	0.964	0.194	1.25	0.0942	0.492	61.0
<b>Scaridae</b>	13.4	2.82	0.377	0.0406	2.52	1.93	73.9	0.0206	4.89	0.136	0.472	0.204	0.889	1.44	1.28	18.4
<b>Serranidae</b>	4.79	3.80	0.118	0.0182	0.507	0.797	44.5	0.0983	1.83	0.0304	0.0756	0.0386	1.41	0.916	0.102	25.3

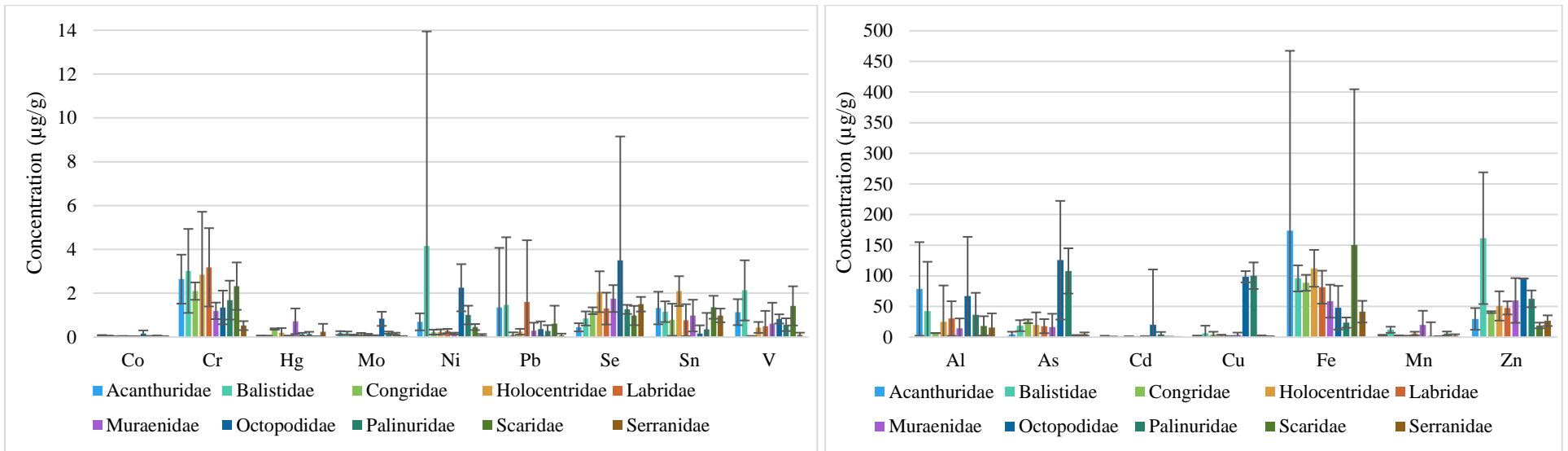


Figure 11a. Arithmetic mean and standard deviation of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in whole prey by family in the NWHI Southern region.

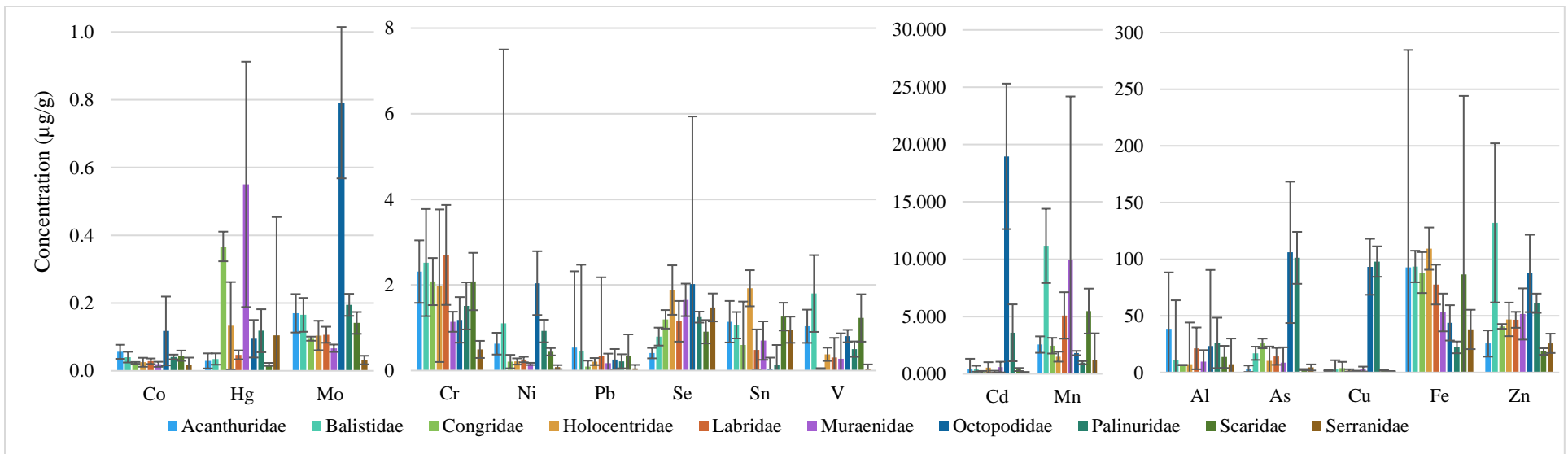


Figure 11b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in whole prey by family in the NWHI Southern region.

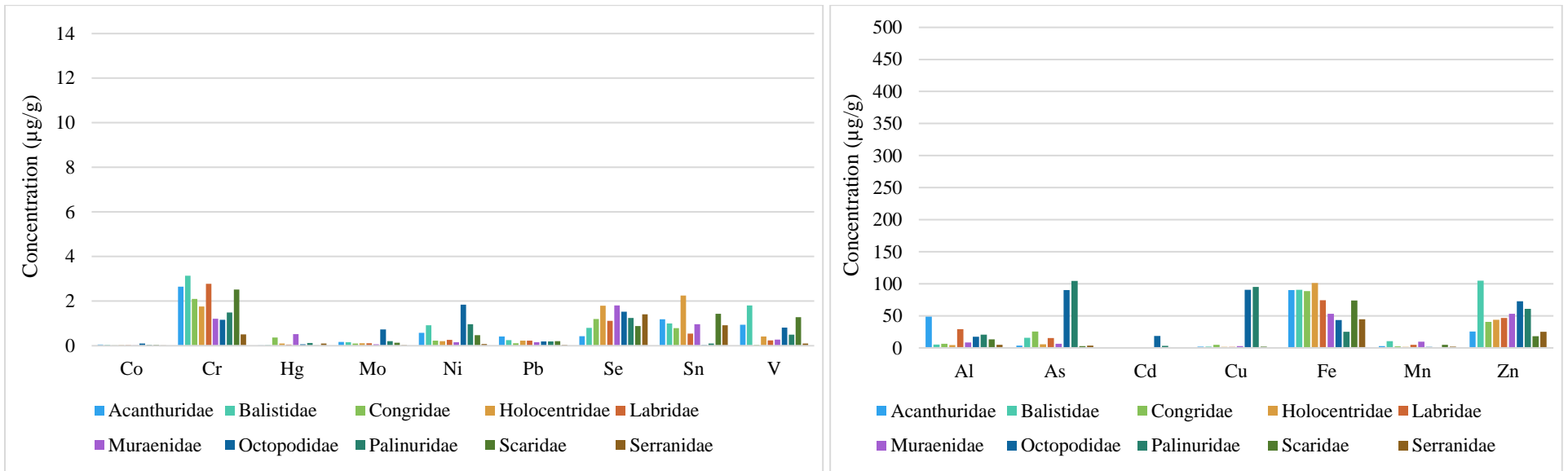


Figure 11c. Median of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in whole prey by family in the NWHI Southern region.

Table 18. Significant differences between prey families listed by heavy metal.

Family	Acanthuridae	Balistidae	Congridae	Holocentridae	Labridae	Muraenidae	Octopodidae	Palinuridae	Scaridae	Serranidae
<b>Acanthuridae</b>										
<b>Balistidae</b>	Zn									
<b>Congridae</b>		V								
<b>Holocentridae</b>	Se	Mn, Se, V								
<b>Labridae</b>		V								
<b>Muraenidae</b>	Co, Hg, Mo, Ni, Se	Hg, Mo, Ni, V		Mn	Hg					
<b>Octopodidae</b>	As, Cd, Cu, Se, Sn, Zn	Cd, Mn, Sn	Cd	Cd, Co, Cu, Fe, Mo, Ni, Sn	Cd, Co, Cu, Mo, Ni	Co, Mo, Ni				
<b>Palinuridae</b>	As, Cu, Fe, Se	Cu, Fe, Mn		As, Cu, Fe, Ni, Sn	Cd, Cu, Fe, Hg, Mn	As, Mn, Mo, Ni				
<b>Scaridae</b>		Zn		Hg	V	Hg, Zn	As, Cd, Hg, Sn, Zn	As, Cd, Cu, Fe, Hg, Mn, Zn		
<b>Serranidae</b>	Cr, Mo, Pb, Ni, V	Cr, Mo, Ni, V, Zn			Cr		As, Cd, Co, Cu, Mo, Ni, Zn	Cd, Cu, Mo, Ni	V	



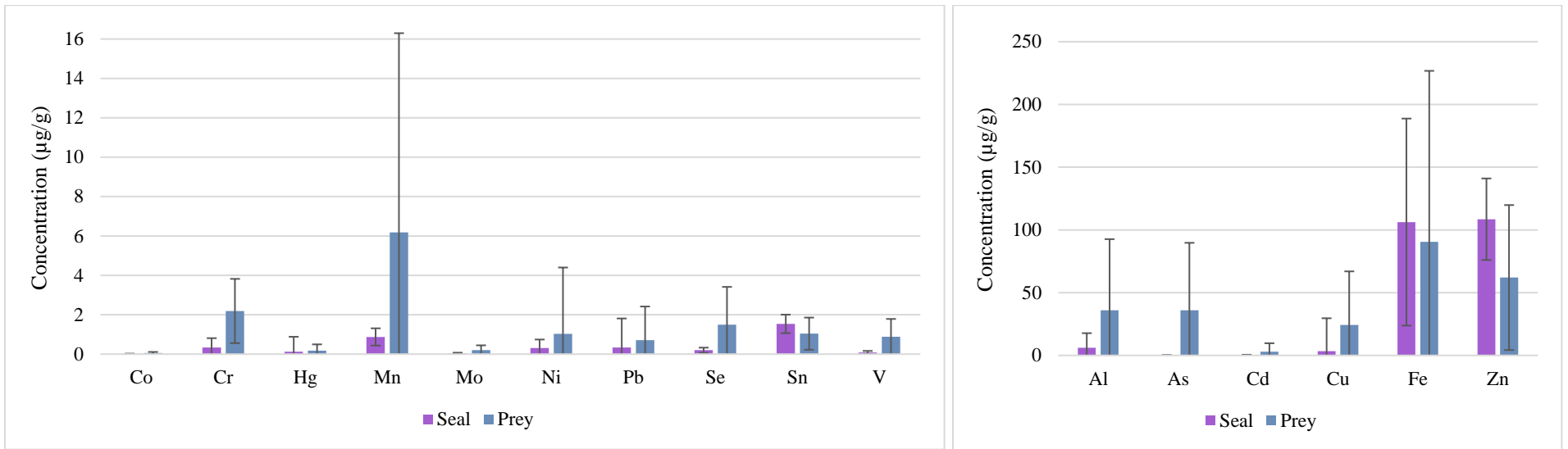


Figure 12a. Arithmetic mean and standard deviation of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone and whole potential prey.

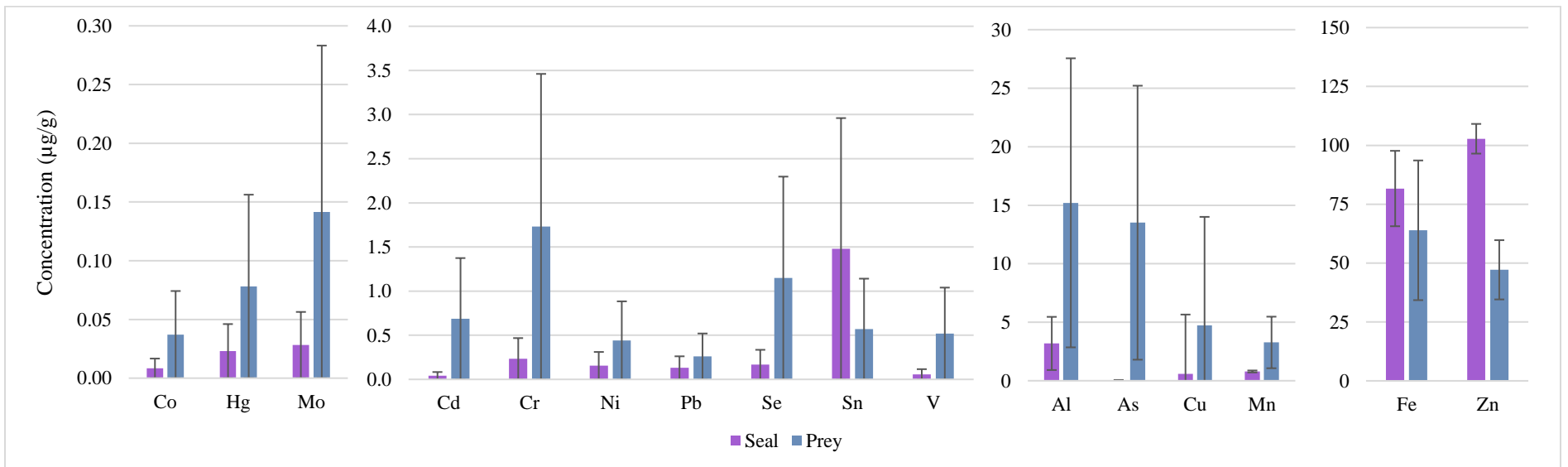


Figure 12b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone and whole potential prey.

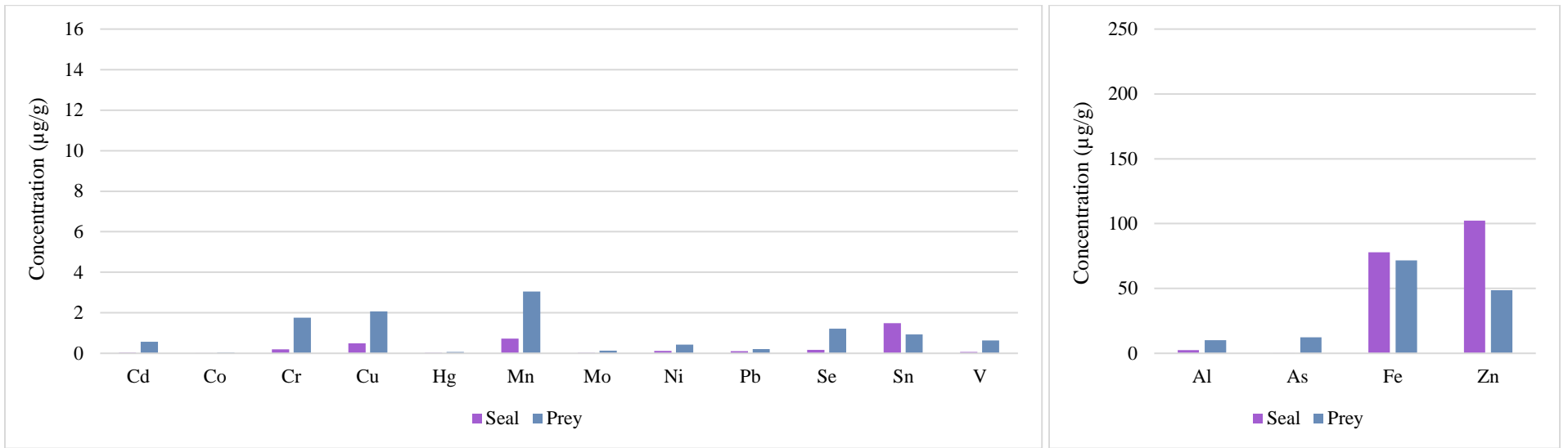


Figure 12c. Median of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone and whole potential prey.

and cephalopod Mn and the teleost Pb concentrations were significantly greater than the seal. The seal Sn concentration was significantly greater than all three prey taxa and seal Zn concentration was significantly greater than the teleost and crustacean; Fe concentration in seal was significantly greater than the crustacean (Figure 13).

#### **4.4 Se:Hg**

The Se to Hg molar ratio was tested because of the potential detoxifying effect since Se can sequester Hg in the toxicologically inert mercury selenide compounds if the ratio Se:Hg molar ratio is one or above (McCormack et al. 2020). Kendall's tau correlation confirmed a significant relationship between Se and Hg across all HMS bone samples ( $z=3.48$ ,  $p < 0.001$ , and  $\tau = 0.234$ , Figure 14). The equation that best describes their relation is  $Hg = 0.0469 + 0.359*Se$ . When the seals were analyzed independently, the molar ratio of each sample exceeded 1 except for one male pup, ARC1216, which exhibited potential Hg toxicity (Table 19).

Kendall's tau correlation confirmed a significant relationship between Se and Hg across all potential prey samples ( $z = 5.19$ ,  $p < 0.001$ , and  $\tau = 0.393$ , Figure 15). The equation that best describes their relation is  $Hg = 0.138 + 0.0288*Se$ . When the prey were analyzed independently, the molar ratio of each sample exceeded a 1:1 relationship between Se and Hg, indicative of Hg toxicity protection by Se (Table 20).

## **5. DISCUSSION**

### **5.1 Heavy Metals in Food Web**

Even with the location of the Hawaiian Island Archipelago in the middle of the Pacific Ocean, heavy metals are still found within the Hawaiian monk seal bone and whole prey living in the region, including essential and nonessential elements. Heavy metals enter the oceans through atmospheric deposition, erosion of geological matrix, and through anthropogenic activities (Muir et al., 1999). Once within the water, metal contaminants remain in soluble or suspension form until they either settle within the benthos or are incorporated by an organism; heavy metals are adsorbed by living and dead organic matter such as particulate organic carbon, lipids, and animal membranes (Baby et al., 2010; Muir et al., 1999).

The transfer of heavy metals across biological membranes are affected by metabolic rate of the organism and the differences in uptake due to speciation of the metals (influenced by water hardness, alkalinity, pH, temperature, and redox conditions in sediment; Heath, 1987).

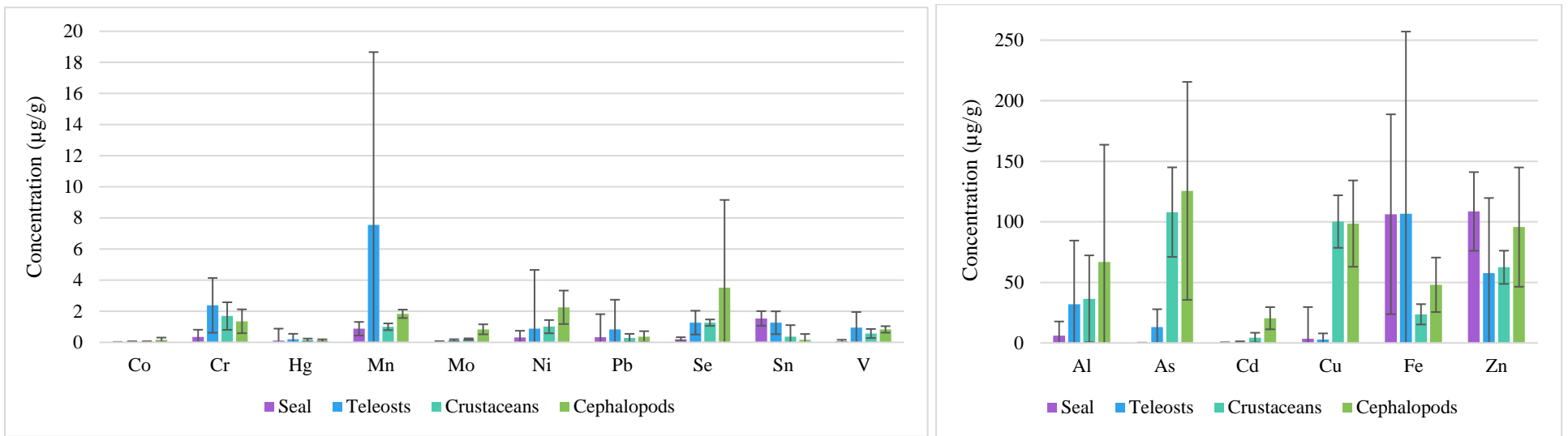


Figure 13a. Arithmetic mean and standard deviation of 16 heavy metal concentrations (µg/g) in HMS bone and whole prey by taxa.

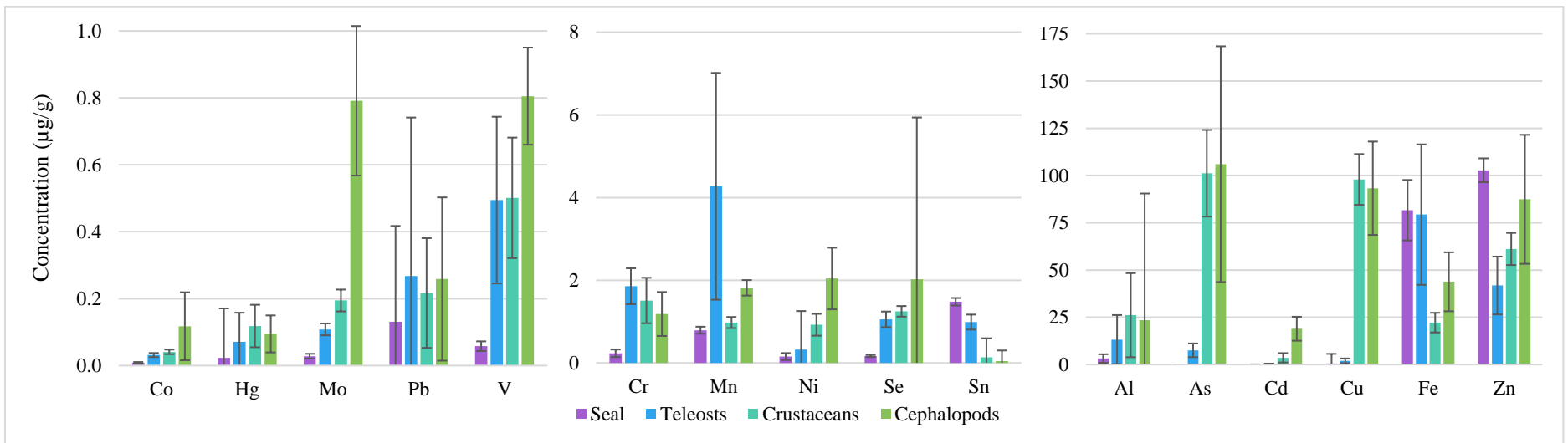


Figure 13b. Geometric mean and 95% confidence interval of 16 heavy metal concentrations (µg/g) in HMS bone and whole prey by taxa.

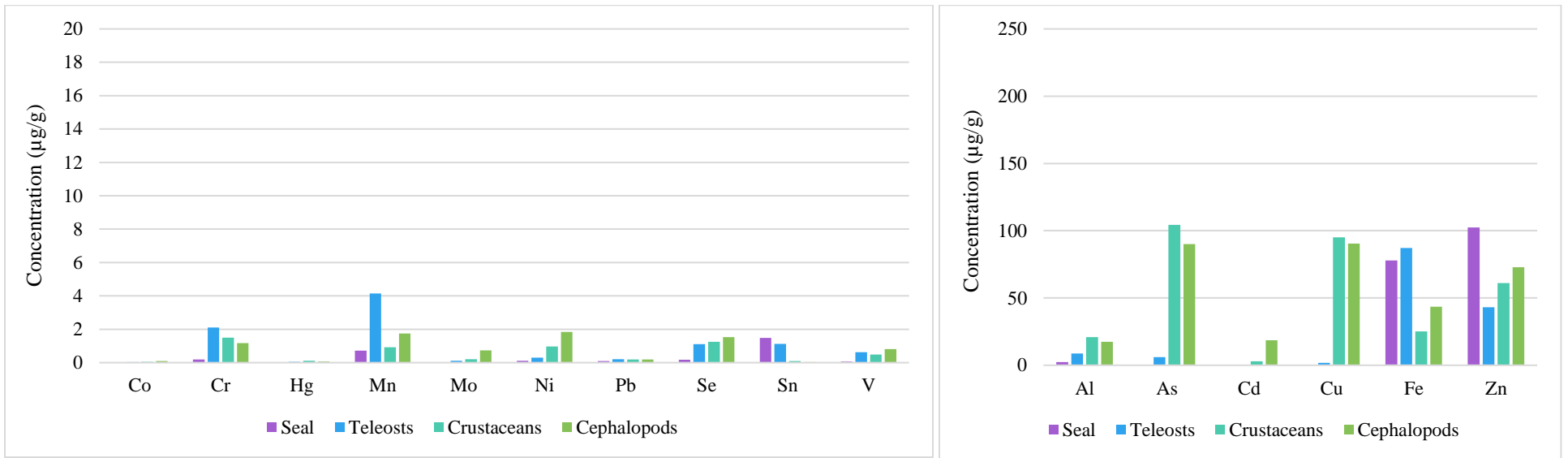


Figure 13c. Median of 16 heavy metal concentrations ( $\mu\text{g/g}$ ) in HMS bone and whole prey by taxa.

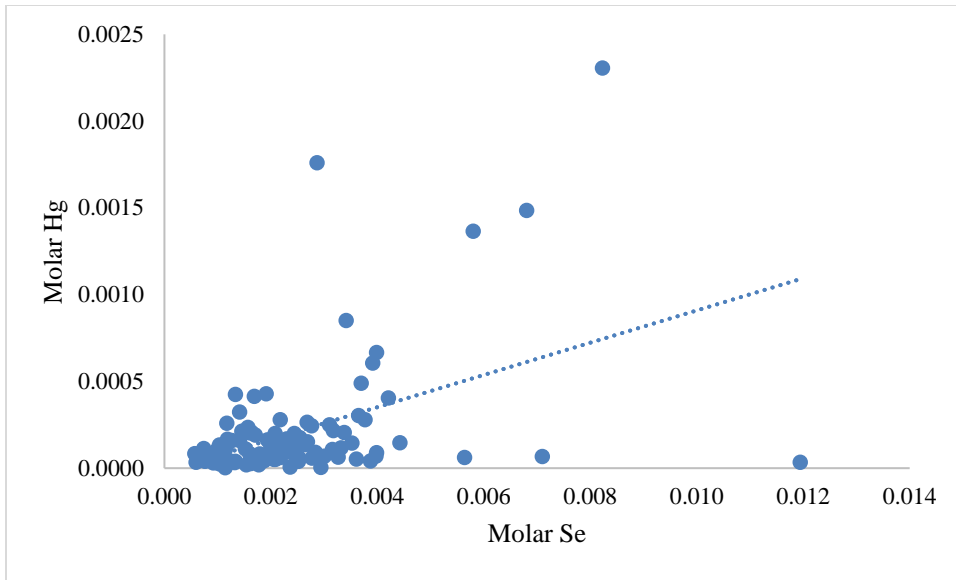


Figure 14. HMS bone molar Se and molar Hg without ARC1216 being the Hg outlier.

Table 19. HMS bone Se:Hg ratio.

<b>ARC-Bone</b>	<b>Molar Ratio Se:Hg</b>
ARC1174	4.09
ARC466	15.3
ARC541	3.14
ARC452	14.7
ARC449	4.46
ARC460	7.94
ARC1049	95.6
ARC1069	26.7
ARC1148	7.80
ARC444	19.1
ARC445	29.1
ARC464	7.90
ARC536	44.2
ARC1060	7.55
ARC1097	6.47
ARC1209	50.7
ARC434	4.25
ARC447	34.6
ARC451	68.6
ARC453	12.3
ARC465	41.8
ARC468	23.1
ARC1057	18.9
ARC450	24.5
ARC455	10.4
ARC461	31.0
ARC872	10.4
ARC1076	5.98
ARC1145	4.38
ARC1230	17.6
ARC435	63.9
ARC1103	19.4
ARC1164	106
ARC1165	24.5
ARC1216	0.0740
ARC1217	16.4
ARC1168	19.7
ARC474	51.6
ARC1059	91.8
ARC1067	25.3
ARC1195	42.8
ARC1162	7.75

ARC1138	40.0
ARC1210	34.3
ARC1055	16.2
ARC1088	8.72
ARC1124	17.7
ARC1082	6.88
ARC1128	1.63
ARC1192	4.50
ARC1213	30.3
ARC1052	35.8
ARC1101	4.01
ARC446	8.94
ARC442	11.4
ARC1092	19.5
ARC1105	7.07
ARC439	21.9
ARC454	40.9
ARC458	27.5
ARC1041	20.7
ARC1113	7.97
ARC1119	649
ARC1080	6.84
ARC1142	31.4
ARC1123	6.48
ARC1133	13.2
ARC1077	8.93
ARC1081	24.3
ARC1100	22.4
ARC1121	16.3
ARC1042	28.2
ARC1056	19.1
ARC545	4.58
ARC1116	3.57
ARC436	62.2
ARC1054	13.7
ARC1141	12.0
ARC1187	79.9
ARC540	44.0
ARC1115	41.2
ARC1045	35.5
ARC1093	6.67
ARC1196	562
ARC1220	88.8
ARC1234	345
ARC1066	9.44



ARC1070	41.0
ARC1075	11.9
ARC1130	7.81
ARC1131	17.5
ARC1155	35.4
ARC1191	30.2
ARC1197	57.6
ARC538	14.4
ARC1084	10.7
ARC1182	13.7
ARC1118	12.4
ARC1153	13.5
ARC1178	433
ARC1186	10.1
ARC1200	47.3

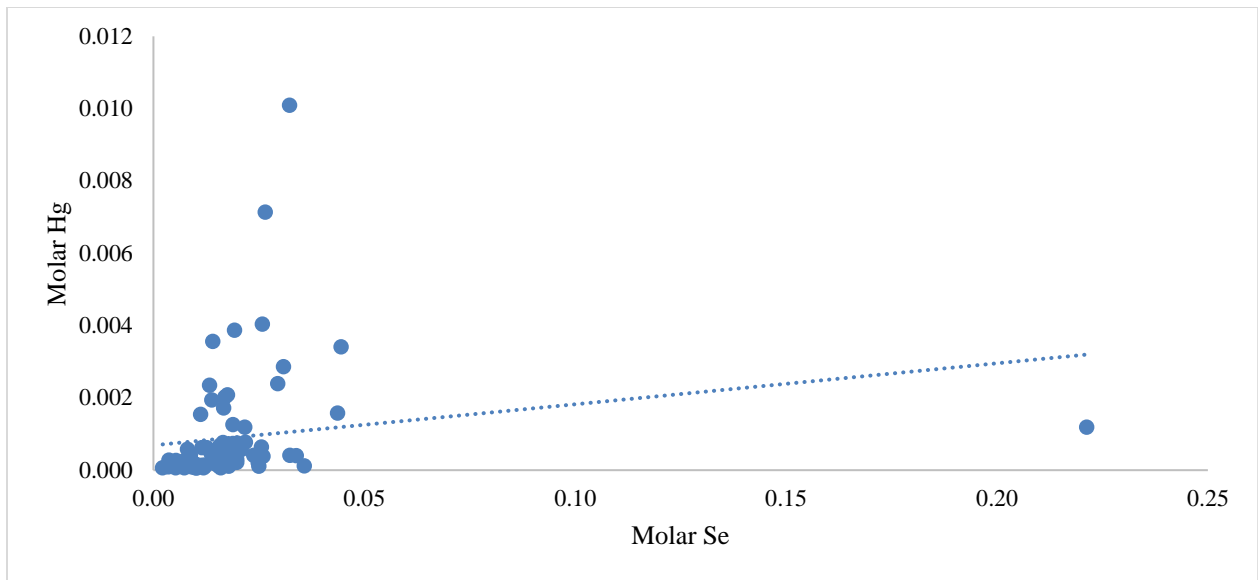


Figure 15. The molar Se to Hg ratio of the whole potential prey samples.

Table 20. Whole potential prey Se:Hg.

<b>MSP-whole</b>	<b>Family</b>	<b>Molar Ratio Se:Hg</b>
MSP 542	Acanthuridae	13.3
MSP 2977	Acanthuridae	139
MSP 2450	Acanthuridae	77.1
MSP 2686	Acanthuridae	55.5
MSP 635	Acanthuridae	14.0
MSP 44	Acanthuridae	37.4
MSP 1827	Acanthuridae	19.8
MSP 2444	Acanthuridae	29.5
MSP 2384	Acanthuridae	35.4
MSP 104	Balistidae	260
MSP 2433	Balistidae	30.6
MSP 2976	Balistidae	78.7
MSP 638	Balistidae	39.4
MSP 642	Balistidae	65.4
MSP 2779	Balistidae	30.4
MSP 2783	Balistidae	90.8
MSP 1314	Balistidae	50.6
MSP 1315	Balistidae	32.3
MSP 649	Congridae	9.65
MSP 650	Congridae	7.11
MSP 1290	Holocentridae	13.0
MSP 451	Holocentridae	85.2
MSP 1902	Holocentridae	27.7
MSP 1903	Holocentridae	79.4
MSP 1291	Holocentridae	8.42
MSP 22	Holocentridae	18.5
MSP 2466	Holocentridae	62.0
MSP 23	Holocentridae	28.2
MSP 242	Holocentridae	92.8
MSP 245	Holocentridae	57.6
MSP 728	Labridae	84.2
MSP 663	Labridae	60.6
MSP 1995	Labridae	297
MSP 1937	Labridae	53.3
MSP 1938	Labridae	63.7
MSP 2344	Labridae	61.3
MSP 2860	Labridae	29.5
MSP 36	Labridae	21.8
MSP 778	Labridae	67.0
MSP 651	Muraenidae	3.72
MSP 1880	Muraenidae	3.95

MSP 1789	Muraenidae	10.8
MSP 1810	Muraenidae	26.8
MSP 1794	Muraenidae	3.20
MSP 511	Muraenidae	6.40
MSP 512	Muraenidae	5.68
MSP 296	Muraenidae	12.4
MSP 257	Muraenidae	14.9
MSP 317	Muraenidae	7.27
MSP 1783	Octopodidae	68.5
MSP 1784	Octopodidae	56.4
MSP 1904	Octopodidae	18.3
MSP 2798	Octopodidae	54.7
MSP 2799	Octopodidae	187
MSP 515	Octopodidae	40.2
MSP 135	Octopodidae	38.4
MSP 137	Octopodidae	67.8
MSP 326	Palinuridae	23.2
MSP 329	Palinuridae	65.5
MSP 334	Palinuridae	25.7
MSP 335	Palinuridae	25.5
MSP 344	Palinuridae	36.9
MSP 345	Palinuridae	20.3
MSP 546	Palinuridae	25.4
MSP 1747	Palinuridae	8.42
MSP 621	Palinuridae	27.9
MSP 626	Palinuridae	43.3
MSP 48	Scaridae	193
MSP 495	Scaridae	55.3
MSP 497	Scaridae	108
MSP 1620	Scaridae	177
MSP 1621	Scaridae	120
MSP 1355	Scaridae	224
MSP 1440	Scaridae	159
MSP 2501	Scaridae	102
MSP 2861	Scaridae	96.9
MSP 2862	Scaridae	81.0
MSP 2966	Serranidae	21.5
MSP 2968	Serranidae	4.96
MSP 260	Serranidae	132
MSP 754	Serranidae	118

A steady state is established for metals within an organism between the dissolved forms in water and dissolved or protein-bound forms in blood and other tissues (Muir et al., 1999). With a high surface area to volume ratio, direct absorption through the integument is a major pathway of uptake for small organisms (Sheldon et al., 1972). Zooplankton can uptake heavy metals while grazing as they are adsorbed onto particle surfaces (Muir et al., 1999). Five of the prey families examined within this study include planktivorous fish.

In the marine environment, fish and invertebrates accumulate heavy metals via three major routes: gills, dermis, and diet (Muir et al., 1999). Baby et al. (2010) states that fish often accumulate large amounts of certain metals such as Cd, Cu, Fe, Mn, Pb, and Zn. Water-borne metals will first target gill surfaces as the micro-environment found on the epithelial membrane of the gill consists of phospholipid covered by a mucous layer (Bolis et al., 1984; Spicer & Weber, 1991; Van de Winkel et al., 1986). Seals have several different pathways to uptake heavy metals: atmospheric through lungs, absorption through skin, across the placenta during fetal development, via milk through lactating, and from ingestion of sea water and food (Das et al., 2003).

## 5.2 Highest Metal Concentrations in Bone

Zinc and Fe had the highest concentrations in seal bone out of all 16 heavy metals within this study. Zinc is essential for bone formation (Honda et al. 1982, 1984a,b; Underwood, 1971) and has been found to have a positive correlation with calcium leading Honda et al. (1984a) to suggest that Zn incorporates into bone hydroxyapatite and proceeds to accumulate in bone during calcification. Zinc accelerates bone formation and is essential for the correct ossification and mineralization of the skeleton (Lanocha et al., 2012). The high Zn concentration may be due to the necessity of the essential element within bone. When examining heavy metals within Weddell seals, Yamamoto et al. (1987) found Zn was mostly located in muscles and bones. The HMS bone Zn concentration (Table 4) of  $102 \pm 6.30 \mu\text{g/g}$  was greater than *Zalophus californianus* bone Zn concentration of  $44.17 \pm 38.65 \mu\text{g/g}$  but lower than Zn within bone of *Pontoporia blainvillei* ( $251.2 \pm 74.4 \mu\text{g/g}$ ) and *Stenella coeruleoalba* ( $382.0 \pm 23.9 \mu\text{g/g}$ ; Garcia-Garin et al., 2021; Honda et al., 1986; Szteren & Auriolles-Gamboa, 2013).

Iron is required for proper differentiation and function within all cells including bone cells (Steinbicker et al., 2011). Iron can also be found within the bone matrix as it is essential for the synthesis of collagen structure when bone mineralization occurs (Maciejewska et al., 2014;

Propckop 1971); therefore, the high Fe concentration is likely required during bone component formation. The HMS bone Fe concentration (Table 4) of  $81.7 \pm 16.0 \mu\text{g/g}$  was greater than *Zalophus californianus* bone Fe concentration of  $70.51 \pm 68.40 \mu\text{g/g}$  but lower than Fe within bone of *Pontoporia blainvillei* ( $130.5 \pm 373.4 \mu\text{g/g}$ ) and *Neophocaena asaeorientalis* ( $247.6 \pm 11.32 \mu\text{g/g}$ ; Garcia-Garin et al., 2021; Hao et al. 2020; Szteren & Aurioles-Gamboa, 2013). The variance within bone concentrations can be related to the difference in the type of bone analyzed. Honda et al. (1982) found wide variations of metal concentrations among skull, tympanic bulla, maxillary bone, and 3<sup>rd</sup>/4<sup>th</sup> rib, respectfully. within striped dolphins (*Stenella coeruleoalba*).

The third highest heavy metal concentration within the HMS bone was Al which is the most abundant metal in the earth's crust (Namiesnik & Rabajczyk, 2010). Aluminum exists within the bone matrix and is up taken by osteoclasts during resorption (Gdula-Argasinska et al., 2004; Priest, 2004). Aluminum is mainly deposited within human bones, (60% of body load) (Krewski et al., 2007) but the concentrations found within the soft body tissues are usually very low (less than 0.1%). Bone may, therefore, serve as the most abundant reservoir of Al within a mammal. A mammal can offload excessive Al through feces or urine excretion resulting in lower concentration than found elsewhere biologically in the environment (Schafer & Jahreis, 2006). Compared to other marine mammal bone Al concentrations (Table 4), the HMS concentration of  $3.19 \pm 2.27 \mu\text{g/g}$  was much lower than *Arctocephalus australis* with  $29.6 \pm 42.6 \mu\text{g/g}$ , *Zalophus californianus* with  $62.73 \pm 44.46 \mu\text{g/g}$ , and *Pontoporia blainvillei* with  $100.1 \pm 279.1 \mu\text{g/g}$  (De Marie et al., 2021; Garcia-Garin et al., 2021; Szteren & Aurioles-Gamboa, 2013). Continental crust with its high Al concentrations weathers from continental land masses and that sediment can be transported to coastal margins., Species foraging over continental shelves will likely have higher concentrations of Al than the mid Pacific basin (Hofmann, 1988; Wedepohl, 1995).

When compared to other marine mammal bone concentrations (Table 4), Sn was a magnitude higher in monk seal bone. The HMS bone Sn was  $1.48 \pm 0.0911 \mu\text{g/g}$  while harbor seal from the North Sea had a concentration of  $0.104 \mu\text{g/g}$  (Agusa et al., 2011). Anthropogenic sources of Sn are found in the form of organotins such as tributyltin whose global use began in the 1960s and became popular in antifouling agents for boats and paints in fishnets (Fent, 1996). while tributyltin was globally banned in 2008 its release in the marine environment continues through hull maintenance activities such as pressure hosing, scraping, and blasting (Eklund & Eklund, 2014; Eklund et al., 2014; Turner, 2010; Turner et al., 2015; Ytreberg et al., 2016). The

use of tributyltin has caused contamination and degradation of coastal environments worldwide through its rapidly increased application, high solubility, and toxicity. Toxic evidence of these contaminants have been found in gastropods and bivalves (Alzieu, 2006; Alzieu et al., 1981; Wolnickowski et al., 1987). Pougnet et al. (2014) identified the main source of butyl-Sn species within Toulon Bay sediments was from marinas and military shipyards. De Carlo et al. (2004) found elevated Cu and Zn concentrations related to boating activities within the Ala Wai Canal, on Oahu. While the authors did not measure Sn concentrations, they did find boating activities impacted the heavy metal concentrations examined.

Zinc concentrations found in Californian sea lion ( $44.17 \pm 38.65 \mu\text{g/g}$ ) bone was an order of magnitude less than the HMS concentration ( $102 \pm 6.30 \mu\text{g/g}$ ); however, this was not the same for Franciscana or striped dolphin (Table 4). The higher Zn concentration within the HMS could be related to proximity to volcanic activity as well as basalt. The soil from the volcanic Fernando de Noronha Archipelago in Brazil had higher Zn concentration than compared to continental soil (Neta et al., 2018). Hinkley et al. (1999) also found Zn to be an important metal within the volcanic plume of the cone Pu'u O'o on Hawaii island, ultimately becoming part of the Hawaiian archipelago. While the basalt weathers away, Zn leaches slowly and uniformly back into the environment and uptake by the biological organisms like HMS (Eggleton et al., 1987). The remaining 14 heavy metals examined were at lower concentrations within the HMS bone than other marine mammal bone concentrations (Table 4). The other marine mammals either live in neritic waters over the continental crust or are migratory organisms that travel near continental crust. HMS reside only in waters along basaltic islands and over oceanic crust (Carretta et al., 2015; Kenyon & Rice, 1959). Heavy metal concentrations do differ between the two crusts (Hofmann, 1988; Wedepohl, 1995). Additionally, continental land has the potential for greater anthropogenic sources with higher heavy metal concentrations than the islands of the NWHI.

### **5.3 Spatial Heavy Metal Differences**

Concentrations of As, Cd, Cr, Cu, and Se in Hawaiian monk seal bones differed significantly between the MHI and NWHI. Seal bone metal concentrations from the NWHI were higher for all five elements. These results were surprising as metal concentrations were expected to be higher in anthropogenically developed regions around the MHI. Diet choice could be a factor for the difference between the regions. This study found cephalopods and crustaceans had significantly greater concentrations of As, Cd, and Cu compared to teleosts. Cephalopods and

crustaceans have hemocyanin, instead of hemoglobin in teleosts, that uses Cu to bind oxygen within the blood cells (Jakimska et al., 2011a, b). Other studies have reported that total As concentrations found in crustaceans and mollusks (cephalopods) are generally higher than concentrations within marine fish (De Gieter et al., 2002; Phillips, 1990). High levels of As found within the muscles of bottom dwellers, such as crustaceans and mollusks (cephalopods), can be associated with the benthos with higher concentrations of As than surface waters (Anacleto et al., 2009; Storelli & Marcotrigiano, 2000). Cadmium is known to accumulate within marine invertebrates, especially mollusks (Bryan 1984). Within cephalopods, Cd is found to concentrate within the digestive glands (Bustamante et al., 1998; Finger & Smith 1987; Miramand & Bentley 1992; Miramand & Guary 1980; Smith et al., 1984). When released into the marine ecosystem, heavy metals will rapidly bind to particulates and sink down to the benthos (Hedge et al., 2009). This leads to benthic marine sediments acting as the ultimate sink for heavy metals within the environment (Ruilian et al., 2008). Within an aquatic ecosystem, heavy metal pollution is most often reflected in high concentrations within sediment, macrophytes, and benthic animals compared to elevated concentrations in water (Linnik & Zubenko 2000). The difference of diet choice and availability may impact the concentration differences found between the MHI and NWHI seal population.

A multitude of anthropogenic sources (military, petroleum) have focused on and around the MHI for more than a century; conversely, the NWHI remained undeveloped except for limited occupation military sites on Midway Atoll and French Frigate Shoal (Kenyon & Rice, 1959; Miao et al., 2001). One would expect anthropogenic sources from the MHI to have an impact on heavy metal concentrations found around the environment. Within the Ala Wai Canal watershed on Oahu in the MHI, peak heavy metal concentrations were associated with areas that had the highest urbanization (De Carlo et al., 2004). Bienfang et al. (2009) found that most heavy metal concentrations found in coastal water samples off Waikiki and Kaneohe were similar in concentration to samples from relatively unpopulated sites off Kauai. This demonstrates minimal impact of anthropogenic sources on heavy metal concentrations in adjoining oceanic water. Heavy metals in the environment were enriched by urban and agricultural inputs but declined sharply in concentration within a short distance from shore (Bienfang et al., 2009). This could support the idea that heavy metals within the State of Hawaii waters are not dispersing far from their source but, rather, are constrained within a sink near the source. The seals of this study



within the MHI may be far enough away from the anthropogenic sources that they do not uptake high concentrations of heavy metals. This does not mean that heavy metals are not a risk for HMS in the MHI, but rather considered a low-level risk. If any seal resides within an area near a heavy metal source, that seal could still be at risk for toxic effects.

Other studies have also confirmed that heavy metals do not disperse far from their source. Shriadah (1998) measured heavy metals within creeks along the coast of the United Arab Emirates and found that the spread of heavy metal concentration was highest where municipal and industrial wastewaters were deposited and decreased in concentration as these waters dispersed. Sadiq's study (2002) and Naser's (2013) study in Bahrain found an onshore-offshore spatial gradient of heavy metals with localized increases at outfalls of desalination plants and industrial facilities. Heavy metals were at the highest concentrations within the environment by their source.

#### **5.4 Seal Age Class and Sex Differences**

Copper concentrations were significantly lower in adult ( $0.317 \pm 0.506 \mu\text{g/g}$ ) and juvenile ( $0.385 \pm 0.146 \mu\text{g/g}$ ) monk seals compared to pups ( $1.03 \pm 14.8 \mu\text{g/g}$ ), lower in females ( $0.471 \pm 0.259 \mu\text{g/g}$ ) compared to males ( $0.833 \pm 16.7 \mu\text{g/g}$ ), and in adult females ( $0.243 \pm 0.0590 \mu\text{g/g}$ ) compared to male ( $1.35 \pm 30.4 \mu\text{g/g}$ ), and female pups ( $0.976 \pm 0.569 \mu\text{g/g}$ ) (Tables 11, 13, and 15). These data together support the idea that Cu may be offloaded from adult dams to pups. Offloading can occur when the dam passes on heavy metals to her pup via reproductive activities such as placental transfer, parturition, and lactation (Honda et al., 1982; 1983; 1986). Copper has been found in other young mammals in higher concentrations (Caurant et al., 1994; Julshamn et al., 1987; Wagemann et al., 1988). Wagemann et al. (1988) found Cu concentrations higher in harp seal pups than their dams. Copper is an essential element and found at higher concentrations in tissues undergoing rapid development and differentiation (Brady & Webb, 1981). Rapidly developing pups requiring more Cu than dams.

Conversely, iron concentrations in adults ( $49.5 \pm 28.2 \mu\text{g/g}$ ) and juveniles ( $64.4 \pm 16.1 \mu\text{g/g}$ ) were significantly lower than pups ( $112 \pm 31.6 \mu\text{g/g}$ ) (Table 13). Honda et al. (1982; 1983) reported Fe transfer from dam to calf in striped dolphins during pregnancy, parturition, and lactation. Iron is largely offloaded via milk in female Weddell seals to the point of decline in aerobic dive capabilities in adult females following lactation (Shero et al., 2022). The large

amount of Fe in pups can assist in breath-holding abilities at the start of independent foraging (Hadley et al., 2006; Hall et al., 2001; Proffitt et al., 2008).

Cadmium concentrations were significantly greater in adults ( $0.0955 \pm 0.0510 \mu\text{g/g}$ ) than pups ( $0.0221 \pm 0.0223 \mu\text{g/g}$ ) (Table 13). Striped dolphins do not transfer Cd across the placenta (Honda et al., 1986) so cadmium might be an element that cannot be offloaded by dams. Many studies found that Cd accumulates with age in marine mammal tissues (Hamanaka et al., 1982; Honda & Tatsukawa, 1983; Honda et al., 1983). Adults having higher Cd concentrations than pups could result from bioaccumulation or biomagnification. Bioaccumulation of heavy metals occurs when the concentration of a metal increases over the lifetime of an individual through consumption of more metal than can be offloaded; biomagnification is the increase of heavy metal concentrations with increasing trophic level within a food web (Bryan & Darracott, 1979; Yarsan & Yipel, 2013). Cephalopod-feeding cetaceans have higher Cd concentrations than teleost-feeding cetaceans due to the bioaccumulation of Cd within cephalopods (Das et al., 2002). Cephalopods have higher concentrations of Cd in their tissues than teleost fish, resulting in predators likely having higher concentrations of Cd. Cadmium concentrations in swordfish compared to common dolphinfish was related to the difference in cephalopod consumption (Kojadinovic et al., 2007). Pups are not weaned so they do not consume cephalopods like an adult seal.

### **5.5 Seal Versus Prey Heavy Metal Concentrations**

All prey taxa had significantly greater concentration than HMS for Al, As, Cd, Co, Cr, Cu, Hg, Mo, Ni, Se, and V. The tissue used for the HMS samples were only bone while all potential prey was homogenized whole samples. The tissue differences could have led to the difference in concentrations between the prey and seals. Most metal burdens within an adult seal were found in the muscle and liver when examining 15 different tissues in Weddell seals (Yamamoto et al., 1987). In marine mammals, the distribution of metals is dependent on the tissue and specific metal (Das et al., 2002; Jakimska et al., 2011b; Wagemann & Muir, 1984). Examples include Hg that is most concentrated in the liver and Cd which is usually most concentrated within the kidney (Wagemann & Muir, 1984). Yamamoto et al. (1987) found that most of the heavy metals were in highest concentrations in the liver and kidney, including Cu within Weddell seals. Northern fur seals (*Callorhinus ursinus*) had higher concentrations of Hg

and Se within liver versus kidney while the kidney had the higher concentration of Cd (Arai et al., 2004).

Tin was significantly greater within HMS bone compared to all prey taxa. One of tin's likely source within the Hawaiian Archipelago is from the use of tributyltin within antifouling agents for boats and paints in fishnets (Fent, 1996). Even with tributyltin ban, it can still leach into the environment from boating maintenance (Eklund and Eklund, 2014; Eklund et al., 2014; Turner, 2010; Turner et al., 2015; Ytreberg et al., 2016). As Sn is a nonessential element for animals, it can biomagnify from the prey to seal (Pais & Jones, 1997).

The differences in Zn concentrations between HMS bones and teleosts and crustaceans is likely the result of body tissue differences. When comparing body tissue, bone sequesters largest amount of Zn (Honda et al., 1982; Yamamoto et al., 1987). It should be expected that the bone sample would have higher Zn than a homogenized organism representing all tissues. Cephalopod Zn concentrations were only slightly less than the seal bones which may be due to the use of Zn in hemocyanin, the oxygen-carrying component in mollusks (Jakimska et al., 2011b).

Iron was significantly greater in HMS than crustaceans. Vertebrates utilize Fe for its oxygen-binding capacity. Some invertebrates, including crustaceans, have hemocyanin that utilizes Cu instead of Fe for the binding of oxygen (Jakimska et al., 2011a). Iron is also essential for the synthesis of collagen structure when bone mineralization occurs, leading to a higher concentration of Fe within bone tissue (Maciejewska et al., 2014; Propckop 1971).

Teleost were found to have significantly greater concentrations of Fe, Mn, and Sn than cephalopods and crustaceans. Iron is found in hemoglobin within teleosts for oxygen transport as compared to crustaceans and cephalopods which utilize Cu-rich hemocyanin (Jakimska et al., 2011a, b). Manganese is an essential element for vertebrates in many cellular processes such as lipid, protein, and carbohydrate metabolism as well as diverse enzyme formations (Andreini et al., 2006; Kehl-Fie & Skaar, 2010). Manganese is also utilized by vertebrates for bone mineralization and to resist bacterial infections as bacterial proteins are manganese dependent so host-mediated manganese sequestration can potentially disrupt bacterial pathogenesis (Kehl-Fie & Skaar, 2010; Pinsino et al., 2012). Within vertebrates, Sn is found to have the highest concentrations in bone compared to other organs and tissues (Kalisinska, 2019). As teleosts have bones and cephalopods and crustaceans do not, the Sn concentration found within the bone of teleost could explain the difference among the taxa.

Aluminum was the only element where the prey families' concentrations were not significantly different. As Al is the most abundant heavy metal, it might be well distributed within the marine environment (Skibniewska & Skibniewski, 2019). Within the chemical composition of lava from Hawaii volcanoes, Al is the third highest component at 11.2% (Tilling et al., 2010).

The General Standard for Contaminants and Toxins in Food and Feed, developed by the Codex Alimentarius Commission (CAC), has established toxicological guidance values or maximum level values for As, Cd, Pb, Hg, and Sn for human consumption. Arsenic's toxicological guidance value is 0.003  $\mu\text{g/g}$  body weight per day (bw/day) for inorganic As (Codex Alimentarius Commission (CAC), 2019). When considering a person that weighs 75 kg, the inorganic As intake per day would be 0.225  $\mu\text{g/g}$ . All prey families, most also consumed by humans, were one to three orders of magnitude greater in As concentration than the daily intake of inorganic As. Arsenic from this study measured total As concentration, As speciation would need to be completed to determine the concentration of inorganic and organic As species within the prey to determine human consumption safety as a proxy to HMS consumption. The Cd maximum safe level within cephalopods for human consumption is 2  $\mu\text{g/g}$ . The geometric mean concentration of Octopodidae in this study was 19.0  $\mu\text{g/g}$ . This value is of concern if humans are also consuming the same octopuses as the HMS. Lead's maximum safe level of contaminants within fish for human consumption is 0.3  $\mu\text{g/g}$ . Acanthuridae, Balistidae, Labridae, and Scaridae all have geometric mean concentrations that are greater than the maximum safe level. Mercury's toxicological guidance value is 0.004  $\mu\text{g/g}$  body weight of inorganic mercury for provisional tolerable weekly intake. When considering a person that weighs 75 kg, the Hg intake for a week is 0.3  $\mu\text{g/g}$ . Congridae and Muraenidae have geometric means higher than 0.3  $\mu\text{g/g}$ . Tin's toxicological guidance value is 14  $\mu\text{g/g}$  body weight for provisional tolerable weekly intake (CAC, 2019). With geometric mean concentrations of the 10 prey families ranging from 1.92 (Holocentridae) to 0.0459 (Octopodidae)  $\mu\text{g/g}$ , the prey concentrations are below the weekly tolerable intake. The heavy metal concentrations reported for the prey represent the whole body; prey individuals were homogenized for sampling. When comparing heavy metal distribution across the body of a fish, each heavy metal has different target organs where they are more likely to accumulate. Arsenic is more often found in the blood, kidneys, central nervous, digestive, and skin systems while Hg is more often found in the brain and kidneys and Cd in the liver, placenta,

kidneys, lungs, brain, and bones (Roberts, 1999). This is important to note that fish, octopuses, and lobsters are mainly consumed for their meat/muscle. Additionally, these prey samples were collected within the NWHI Southern region where fishing is currently banned (DiNardo & Marshall, 2001; Parrish et al., 2012).

## 5.6 Se to Hg Ratio

Examining the molar ratio of Se to Hg provides information on the detoxification of Hg by Se by the sequestration of Hg in the toxicologically inert mercury selenide compounds (McCormack et al. 2020). The detoxification of Hg by Se has been observed in several marine mammals (Correa et al., 2015; Koeman et al., 1973; McCormack et al., 2020). All but one HMS in this study had a Se:Hg molar ratio of 1 or greater, suggesting that the detoxification of Hg by Se is occurring within the seals. The lone exception was a seal pup with a Hg concentration that was three orders of magnitude greater than all other seals. The high Hg could have come from the dam during fetal development or through milk. Heavy metals can also be obtained from water, meaning this pup could be living near a Hg source. Mercury is found within antifouling paint meaning a shipwreck nearby the seal's haul out location could be leaching Hg within the water (Raine et al., 1995). Selenium is also potentially detoxifying mercury within the prey species as the molar ratio of Se:Hg was greater than 1 for all prey individuals. Mercury detoxification was found in other bony fish studies. Kehrig et al. (2009) found a tropical marine food web in Guanabara Bay, Brazil where *Centropomus undecimalis* (snook), *Micropogonias furnieri* (croaker), *Bagre spp.* (catfish), and *Mugil lisa* (mullet) all had Se:Hg greater than 1:1. Also off the coast of Brazil in Ilha Grande Bay, *Farfantepenaeus brasiliensis* (shrimp) was found with a Se:Hg greater than 1:1 (Seixas et al., 2014). Evidence of demethylation by Se off the coast of Portugal was found in *Octopus vulgaris* in three different locations (Raimundo et al., 2010). The availability of Se within the Hawaiian environment may be due to the volcanic-sourced geology. Selenium concentrations have been found to be higher within particulates emitted by the volcano at Mt. Etna compared to anthropogenic sources within the remaining Mediterranean area (Buat-Menard & Arnold, 1978).

## 5.7 Conclusion

Heavy metal concentrations within an organism are based on concentrations available within the environment in which they live. These metals can come from natural sources, such as

volcanic activity, or anthropogenic sources, such as military activity, agricultural practices, and urbanization. HMS live near all of these sources within the MHI but the majority of the population lives within the more pristine NWHI. The seals can uptake heavy metals from their diet, the water in which they reside, what is offloaded to them from their mother, and from the air they breathe. These metals can be incorporated into the bone of the seal, using the blood stream as transport, where they are deposited until osteocyte turnover. Bone is a useful tissue, especially with an endangered organism, as it can be collected postmortem.

Iron and Zn had the highest concentrations within the bone of the HMS which are also essential elements for bone formation. The higher concentrations of As, Cd, Cr, Cu, and Se in the NWHI compared to the MHI were not expected to be from anthropogenic sources. Instead, monk seal concentrations indicate preferential diet throughout the archipelago (NWHI vs MHI) may be a cause. Other contaminant studies have found that heavy metals tend to bind and sink near their sources instead of dispersing into the ocean water, leaving the metals bound in locations where the seals may not reside. Copper and Fe concentrations indicate maternal offloading may be occurring while Cd shows evidence of bioaccumulation and biomagnification. Selenium may be detoxifying the Hg within all of the prey and almost every seal. Volcanic eruption is a source of Se within the environment, leading to a high availability of Se within the hotspot-formed Hawaiian archipelago.

Based on the toxicological guidance values, established by the CAC (2019), As, Cd, Pb, and Hg concentrations found within some of the prey were higher than the maximum safe level for human consumption. As these prey were all collected within the NWHI Southern region, another study should be completed within the MHI to examine the heavy metal concentrations within fish, lobster, and octopus where active fishing is occurring. In addition to analyzing the total heavy metal concentrations of these organisms, element speciation, specifically As, should be completed. Arsenic can be found in inorganic or organic forms within the ecosystem but only inorganic As is a concern for toxicity. With the concerning concentration of As found within this study's prey, it's important to know what type of As is within the prey sample.

Bone sequester heavy metals within the HMS and concentrations differences based on seal colony, age class, and sex, but the toxicity levels in HMS remain unknown. Each heavy metal has a binding affinity to different body tissues. To understand total body load and potential

toxicity, more research needs to be completed using multiple tissues from HMS. These soft tissues could include skin, blubber, liver, brain, muscle, and kidney tissue.

Knowing that HMS, the 2<sup>nd</sup> most endangered pinniped, can uptake heavy metals, there will always be a concern that the seals could have or reach toxic concentrations. The NWHI lie within a preserved national monument, protecting the HMS from direct human disturbance. In contrast, the MHI seal population is small but growing. Those seals are not within a protected area and can more readily be influenced by human activity. If a seal established itself within an active harbor or near a deposit source from industrial, agricultural, or military activity, those seals may be at risk for toxic levels of heavy metals. This potential will always be there in the MHI.

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## Appendix

Table 21. Heavy metal concentrations ( $\mu\text{g/g}$ ) in all seal bone samples with the seal's collection data (region, decade, sex, and age class).

Lab ID	Region	Decade	Sex	Age Class	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
ARC1174	Central	2000	F	A	5.47	0.0324	0.370	0.0165	0.231	0.310	54.2	0.0829	0.887	0.0875	0.175	0.154	0.133	0.860	0.0209	103
ARC466/ARC855	Central	2000	F	A	2.71	0.0196	0.0573	0.0043	0.188	0.196	20.4	0.0108	0.740	0.0271	0.0412	0.309	0.0647	1.40	0.0990	90.6
ARC541	Central	2000	F	A	2.89	0.0256	0.428	0.0189	0.119	0.294	20.6	0.0852	0.928	0.0151	0.0488	0.662	0.105	0.936	0.0271	122
ARC452/ARC813	Central	2000	M	A	1.62	0.104	0.0588	0.0035	0.135	0.387	49.1	0.0434	0.511	0.0168	0.0721	0.0500	0.250	1.53	0.0465	100
ARC449/ARC868	Central	2000	U	A	2.13	0.0331	0.0695	0.0053	0.278	0.246	37.6	0.0861	0.510	0.0372	0.0923	0.0522	0.151	1.23	0.0685	97.7
ARC460/ARC863	Central	2000	U	A	2.52	0.0303	0.108	0.0029	0.0990	0.348	39.8	0.0321	0.551	0.0216	0.0285	0.0831	0.100	1.66	0.0638	99.0
ARC1049	Central	2000	F	J	1.51	0.279	0.0548	0.0040	0.295	0.789	158	0.0081	0.694	0.0185	0.120	0.101	0.306	1.50	0.228	107
ARC1069	Central	2000	F	J	2.20	0.0258	0.120	0.0111	0.254	0.310	41.9	0.0165	0.435	0.0220	0.347	0.139	0.174	2.35	0.0615	92.8
ARC1148	Central	2010	F	J	3.03	0.167	0.149	0.0069	0.355	0.368	36.4	0.0413	0.862	0.0244	0.0453	0.100	0.127	1.38	0.0921	92.9
ARC444/829	Central	2000	F	J	1.64	0.0078	0.0465	0.0090	0.199	0.217	64.7	0.0168	0.526	0.0265	0.111	0.164	0.126	1.49	0.0662	98.8
ARC445/ARC825	Central	2000	F	J	50.5	0.163	0.0672	0.0112	0.478	1.33	56.9	0.0217	1.63	0.0580	0.441	0.411	0.249	1.74	0.130	176
ARC464/ARC821	Central	2000	F	J	1.70	0.0360	0.0585	0.0018	0.151	0.279	56.2	0.0409	0.670	0.0253	0.0237	0.0849	0.127	1.07	0.203	105
ARC536/ARC857	Central	2000	F	J	1.24	0.0104	0.0886	0.0038	0.194	0.242	57.0	0.0181	0.638	0.0157	0.0790	0.151	0.315	1.19	0.0182	97.3
ARC1060	Central	2000	M	J	3.02	0.0695	0.420	0.0047	0.618	1.56	253	0.0982	1.06	0.0548	0.210	0.159	0.292	1.55	0.127	152
ARC1097	Central	2000	M	J	1.93	0.149	0.0266	0.0038	0.316	0.901	63.4	0.121	0.653	0.0210	0.0488	0.0851	0.309	1.36	0.0873	98.7
ARC1209	Central	2000	M	J	20.4	0.159	0.0327	0.0189	0.504	0.842	112	0.0129	1.41	0.0402	0.423	0.148	0.258	1.53	0.0762	135
ARC434/ARC807	Central	2000	M	J	3.07	0.130	0.0789	0.0049	0.286	1.14	89.4	0.274	0.815	0.0360	0.138	0.161	0.458	4.40	0.0762	102
ARC447/ARC806	Central	2000	M	J	1.44	0.0126	0.0142	0.0094	0.191	0.225	32.3	0.0077	0.649	0.0219	0.119	0.0833	0.106	1.26	0.0666	85.7
ARC451/ARC808	Central	2000	M	J	2.10	0.433	0.0554	0.0123	0.390	0.476	66.8	0.0105	0.596	0.0736	0.100	0.0797	0.285	2.74	0.429	127
ARC453/ARC882	Central	2000	M	J	1.33	0.0726	0.0288	0.0059	0.123	0.470	94.9	0.0399	0.646	0.0170	0.0972	0.0500	0.193	1.66	0.119	103
ARC465/ARC805	Central	2000	M	J	1.24	0.0776	0.0297	0.0105	0.200	0.434	49.3	0.0121	0.752	0.0284	0.0561	0.0894	0.199	1.00	0.110	80.3
ARC468	Central	2000	M	J	2.53	0.0151	0.0423	0.0023	0.0870	0.101	83.1	0.0193	0.774	0.0142	0.0136	0.0476	0.175	1.38	0.0371	99.1
ARC1057	Central	2000	U	J	2.82	0.0338	0.128	0.0055	0.236	0.355	61.9	0.0225	0.577	0.0251	0.539	0.124	0.167	1.59	0.0992	143
ARC450/ARC877	Central	2000	U	J	2.26	0.0280	0.0510	0.0039	0.257	0.389	138	0.0288	0.925	0.0185	0.0578	0.0745	0.278	1.34	0.0518	108
ARC455/ARC887	Central	2000	U	J	1.91	0.0920	0.447	0.0039	0.141	0.815	90.6	0.0810	0.997	0.0262	0.0515	0.236	0.332	1.26	0.0685	129
ARC461/ARC858	Central	2000	U	J	1.37	0.0151	0.0795	0.0039	0.115	0.199	36.7	0.0184	0.608	0.0215	0.0311	0.0660	0.224	1.34	0.0304	100

ARC872	Central	2000	U	J	5.19	0.0321	0.0942	0.0073	0.179	0.277	44.7	0.0399	0.742	0.0223	0.0459	0.0889	0.164	1.19	0.160	92.4
ARC1076	Central	2000	F	P	7.61	0.785	4.00	0.0298	1.18	1.83	255	0.134	1.55	0.120	2.45	0.151	0.315	2.77	0.123	148
ARC1145	Central	2000	F	P	4.70	0.0049	0.0185	0.0085	0.717	1.05	86.6	0.0647	1.15	0.0249	0.346	0.117	0.112	1.90	0.0338	132
ARC1230	Central	2000	F	P	11.0	0.0255	0.0119	0.0399	0.787	2.02	286	0.0305	0.713	0.287	0.410	0.203	0.212	1.73	0.0457	159
ARC435/ARC834	Central	2000	F	P	4.28	0.207	0.126	0.0129	0.405	2.17	123	0.0079	0.857	0.0407	0.324	0.465	0.199	1.79	0.0527	96.9
ARC1103	Central	2000	M	P	5.13	0.214	0.0930	0.0073	0.553	0.670	77.3	0.0104	0.781	0.0208	1.27	0.0838	0.0796	1.50	0.0389	83.0
ARC1164	Central	2000	M	P	1.90	0.195	0.0373	0.0044	0.251	0.702	86.8	0.0135	0.685	0.0316	0.0821	0.108	0.561	1.64	0.0355	88.8
ARC1165	Central	2000	M	P	1.44	0.0333	0.0155	0.0037	0.131	1.01	142	0.0154	0.653	0.0164	0.0451	0.0534	0.148	1.60	0.0223	105
ARC1216	Central	2000	M	P	1.58	0.0393	0.0273	0.0034	0.115	1.71	166	7.70	0.790	0.0322	0.0582	0.0345	0.224	1.44	0.0365	123
ARC1217	Central	2000	M	P	1.80	0.0595	0.0371	0.0065	0.163	0.701	92.5	0.0413	0.622	0.0187	1.08	0.0533	0.267	1.73	0.0236	105
ARC1168	Central	2000	U	P	1.04	0.107	0.0173	0.0045	0.194	1.76	186	0.0270	0.849	0.117	0.196	0.0478	0.209	1.82	0.0726	116
ARC474	Central	2000	F	U	1.78	0.0396	0.0110	0.0025	0.276	0.231	20.1	0.0065	0.611	0.0159	0.0694	0.0644	0.132	1.18	0.0443	97.7
ARC1059	Central	2000	U	U	88.3	0.301	0.151	0.0142	0.750	1.84	225	0.0123	2.22	0.108	0.719	0.271	0.445	1.90	0.149	244
ARC1067	Central	2000	U	U	4.59	0.416	0.0551	0.0261	0.910	1.48	175	0.0145	1.31	0.0187	0.151	0.0875	0.145	2.77	0.0829	118
ARC1195	Main	2000	M	A	2.84	0.0138	0.0221	0.0109	0.127	0.158	34.3	0.0088	0.690	0.0151	0.305	0.0860	0.148	1.17	0.0727	85.4
ARC1162	Main	2010	U	A	3.99	0.0091	0.0456	0.0093	0.0842	0.385	109	0.0268	1.00	0.0162	0.835	0.0925	0.0818	1.39	0.0470	112
ARC1138	Main	2010	F	J	2.32	0.0692	0.0074	0.0153	0.0677	0.180	60.1	0.0066	1.19	0.0106	0.896	0.0926	0.104	1.10	0.178	67.8
ARC1210	Main	2000	F	J	3.81	0.0392	0.0131	0.0235	0.0744	1.50	66.7	0.0062	0.422	0.0310	0.217	0.122	0.0832	1.17	0.173	82.7
ARC1055	Main	2010	U	J	2.91	0.0122	0.0407	0.0039	0.111	0.0675	66.6	0.0276	0.434	0.0111	0.0233	0.0365	0.176	2.02	0.0205	77.5
ARC1088	Main	2010	U	J	2.13	0.0018	0.0083	0.0057	0.109	0.633	153	0.0271	0.538	0.0224	0.402	0.149	0.0930	1.58	0.0128	110
ARC1124	Main	2000	F	P	1.53	0.0056	0.0035	0.0056	0.119	0.240	113	0.0067	0.776	0.0239	0.173	0.0813	0.0470	1.41	0.255	114
ARC1082	Main	2010	U	P	1.90	0.0049	0.0021	0.0287	0.109	0.325	85.8	0.0167	0.707	0.162	0.379	0.0233	0.0451	1.39	0.0366	111
ARC1128	Main	2000	U	U	1.95	0.0119	0.0500	0.0029	0.115	0.187	63.2	0.353	0.427	0.0175	0.219	0.100	0.226	1.33	0.0481	129
ARC1192	Main	2000	U	U	2.09	0.0029	0.0057	0.0199	0.248	0.195	60.4	0.0521	0.751	0.0504	0.0166	0.0506	0.0923	1.43	0.235	84.0
ARC1213	Main	2010	U	U	2.40	0.0123	0.0122	0.0271	0.155	0.213	61.2	0.0061	0.535	0.0199	0.585	0.0687	0.0728	2.02	0.232	82.1
ARC1052	Northern	2010	F	A	5.35	0.0526	0.0665	0.0463	0.554	0.278	49.4	0.0071	1.03	0.0993	0.169	0.192	0.100	0.794	0.0909	77.2
ARC1101	Northern	2000	F	A	2.33	0.0172	0.0448	0.0079	0.110	0.122	25.7	0.171	0.707	0.0164	0.0435	0.0890	0.270	1.03	0.0308	94.6
ARC446/ARC847	Northern	2000	F	A	3.00	0.0063	0.0839	0.0103	0.184	0.181	26.5	0.0384	0.623	0.0231	0.0976	0.663	0.135	1.56	0.0632	94.2
ARC442/ARC811	Northern	2000	M	A	3.23	0.200	0.157	0.0035	0.164	0.397	110	0.0488	0.595	0.0261	0.158	0.249	0.218	1.76	0.104	86.8
ARC1092	Northern	2010	U	A	2.82	0.101	0.0251	0.0059	0.687	0.112	17.3	0.0079	0.370	0.0403	0.0785	0.0303	0.0602	1.23	0.0717	3.46
ARC1105	Northern	2000	U	A	1.60	0.0337	0.132	0.0036	0.286	0.189	59.7	0.0335	0.702	0.0387	0.0649	0.0754	0.0933	0.980	0.0670	78.7
ARC439/ARC818	Northern	2000	F	J	1.39	0.0890	0.0629	0.0222	0.193	0.113	24.2	0.0164	0.501	0.0171	0.0657	0.0964	0.142	1.58	0.100	91.4

ARC454/ARC827	Northern	2000	F	J	25.1	0.592	0.0672	0.0151	0.552	1.17	68.1	0.0051	1.30	0.0386	1.27	0.578	0.0816	2.02	0.163	131
ARC458/ARC894	Northern	2000	F	J	2.40	0.300	0.711	0.0059	0.166	0.543	48.4	0.0175	0.710	0.0409	0.0797	0.128	0.190	1.43	0.131	79.4
ARC1041	Northern	2000	F	P	1.00	0.214	0.0039	0.0149	0.131	0.447	57.5	0.0237	0.592	0.0278	0.173	0.0802	0.192	1.10	0.111	85.0
ARC1113/ARC887	Northern	2000	F	P	6.62	0.0073	0.311	0.0144	1.54	0.911	126	0.0412	1.27	0.0405	0.352	0.578	0.129	2.04	0.0424	89.4
ARC1119	Northern	2000	F	P	2.99	0.0063	0.0987	0.0198	0.602	0.817	79.6	0.0004	0.564	0.0472	0.306	0.222	0.0901	1.78	0.0283	117
ARC1080	Northern	2010	M	P	2.70	0.0790	0.0939	0.0098	0.128	0.413	44.5	0.0426	1.25	0.0158	0.136	0.0786	0.115	1.24	0.0240	106
ARC1142	Northern	2010	M	P	1.76	0.0081	0.0024	0.0020	0.133	0.500	62.3	0.0080	0.808	0.0172	0.117	0.0581	0.0991	1.28	0.0133	112
ARC1123	Northern	2000	U	P	0.842	0.0063	0.0033	0.0032	0.190	0.185	33.3	0.0228	0.615	0.0214	0.0531	0.0498	0.0582	1.02	0.0061	113
ARC1133	Northern	2000	U	P	1.63	0.0879	0.0468	0.0173	0.286	0.908	170	0.0300	1.10	0.0242	0.0597	0.0516	0.155	1.30	0.118	104
ARC1077	Northern	2000	U	U	8.53	0.0246	0.0147	0.0098	0.145	6.242	177	0.0318	1.31	0.0197	0.107	0.262	0.112	1.50	0.219	132
ARC1081	Northern	2010	U	U	2.88	0.0845	0.102	0.0071	0.426	0.310	124	0.0167	1.26	0.0234	0.0827	0.256	0.159	1.62	0.142	141
ARC1100	Northern	2000	U	U	3.69	0.0126	0.123	0.0145	0.746	0.851	108	0.0179	1.41	0.0331	0.296	0.331	0.158	1.32	0.0427	98.5
ARC1121	Northern	2000	U	U	53.1	0.407	0.133	0.0555	4.18	1.43	163	0.0298	3.36	0.0370	0.785	14.9	0.191	2.22	0.274	114
ARC1042	Northern	2000	U	U	2.09	0.0219	0.109	0.0049	0.130	0.806	226	0.0236	0.903	0.0308	0.0831	0.0878	0.262	1.65	0.147	99.0
ARC1056	Southern	2000	F	A	16.1	0.0279	0.183	0.0845	0.363	0.387	191	0.0232	1.38	0.0179	1.32	0.133	0.174	1.20	0.109	107
ARC545	Southern	2000	F	A	2.24	0.0044	0.230	0.0058	0.107	0.291	67.5	0.298	0.481	0.0064	0.0869	0.0875	0.537	1.62	0.0215	116
ARC1116	Southern	2000	M	A	13.1	0.251	0.0512	0.0082	0.162	0.892	239	0.462	0.599	0.0356	1.32	0.201	0.650	1.34	0.166	115
ARC436/ARC814	Southern	2000	M	A	2.31	0.0057	0.202	0.0070	0.192	0.347	14.7	0.0052	1.13	0.0204	0.0639	0.131	0.128	1.73	0.0097	270
ARC1054	Southern	2010	U	A	11.2	0.0621	0.182	0.0157	0.377	5.15	146	0.0222	1.25	0.0248	0.193	0.844	0.120	1.44	0.111	125
ARC1141	Southern	2000	F	J	1.49	0.0290	0.0148	0.0020	0.0634	0.236	55.9	0.0608	0.543	0.0525	0.0477	0.0476	0.288	0.917	0.170	77.9
ARC1187	Southern	2000	F	J	1.97	0.0090	0.0096	0.0087	0.139	0.185	56.1	0.0039	0.519	0.0180	0.0621	0.0582	0.122	1.24	0.0560	99.0
ARC540	Southern	2000	F	J	1.72	0.0149	0.0126	0.0029	0.131	0.248	42.6	0.0137	0.436	0.0128	0.0757	0.124	0.237	0.938	0.0594	94.9
ARC1115	Southern	2000	U	J	1.56	0.0435	0.0201	0.0086	0.166	0.194	37.1	0.0102	0.415	0.0127	0.0940	0.0580	0.165	1.26	0.0800	93.5
ARC1045	Southern	2000	F	P	1.99	0.0141	0.0061	0.0067	0.361	0.397	152	0.0065	0.455	0.0280	0.0869	0.146	0.0910	1.42	0.0059	73.4
ARC1093	Southern	2010	F	P	1.46	0.0068	0.0087	0.0019	0.0771	0.813	148	0.0472	0.628	0.0302	0.137	0.0575	0.124	1.67	0.0208	79.1
ARC1196	Southern	2000	F	P	12.3	0.109	0.0948	0.0254	1.17	3.94	320	0.0010	1.34	0.0436	1.41	0.660	0.232	1.75	0.114	133
ARC1220	Southern	2000	F	P	9.63	0.0691	0.0522	0.0237	0.844	2.00	191	0.0040	0.958	0.0946	0.736	0.253	0.140	1.57	0.0699	131
ARC1234	Southern	2000	F	P	1.31	0.0055	0.0100	0.0032	0.255	0.426	103	0.0069	0.654	0.0199	0.116	0.127	0.943	1.51	0.0090	98.8
ARC1066	Southern	2000	M	P	2.25	0.0039	0.0055	0.0031	0.0762	265	45.5	0.0209	0.383	0.0133	0.0775	0.0617	0.0778	1.95	0.0011	132
ARC1070	Southern	2000	M	P	7.46	0.0171	0.0180	0.0077	0.548	4.83	130	0.0100	1.05	0.0306	0.307	0.210	0.162	1.46	0.0474	98.9
ARC1075	Southern	2000	M	P	1.83	0.0077	0.0011	0.0102	0.245	0.668	35.7	0.0328	0.603	0.0332	0.243	0.0192	0.153	1.49	0.0068	72.2
ARC1130	Southern	2000	M	P	2.57	0.0413	0.0136	0.0114	0.212	0.732	206	0.0559	1.10	0.0310	0.108	0.168	0.172	1.67	0.0365	99.3

ARC1131	Southern	2000	M	P	4.85	0.0892	0.0306	0.0078	0.263	0.880	240	0.0265	1.18	0.0278	0.0717	0.197	0.183	1.77	0.0380	103
ARC1155	Southern	2000	M	P	29.0	0.157	0.0328	0.0126	0.668	1.10	174	0.0124	1.86	0.0427	0.817	0.299	0.172	1.89	0.0527	146
ARC1191	Southern	2000	M	P	21.0	0.252	0.399	0.0302	0.815	5.42	494	0.0294	1.67	0.0598	1.94	1.01	0.349	0.985	0.186	196
ARC1197	Southern	2000	M	P	0.964	0.0063	0.0119	0.0131	0.0794	0.272	73.1	0.0138	0.462	0.0176	0.0493	0.0378	0.314	1.59	0.0069	95.5
ARC538	Southern	2000	M	P	2.80	0.0046	0.0117	0.0051	0.147	0.831	42.0	0.0354	0.687	0.0206	0.216	0.104	0.201	1.28	0.0080	91.1
ARC1084	Southern	2010	U	P	3.12	0.0128	0.0034	0.0140	0.300	0.549	126	0.0214	1.15	0.0680	0.205	0.0883	0.0900	1.32	0.0758	105
ARC1182	Southern	2000	U	P	1.26	0.0615	0.0470	0.0034	0.117	0.727	56.1	0.0336	0.603	0.0300	0.0791	0.505	0.182	1.41	0.0178	107
ARC1118	Southern	2000	U	U	6.89	0.0284	0.0328	0.0266	0.328	0.818	81.4	0.0501	0.869	0.0768	0.623	0.289	0.245	1.56	0.130	111
ARC1153	Southern	2000	U	U	5.10	0.456	0.125	0.0051	0.206	1.09	202	0.0561	1.19	0.0266	0.0663	1.39	0.298	1.40	0.310	93.7
ARC1178	Southern	2000	U	U	16.5	0.352	0.0880	0.0105	0.319	1.47	226	0.0011	1.00	0.0249	0.357	0.393	0.186	1.64	0.110	152
ARC1186	Southern	2000	U	U	1.57	0.0134	0.0452	0.0061	0.0981	2.59	362	0.0532	1.69	0.0135	1.18	0.0815	0.212	1.74	0.0156	111
ARC1200	Southern	2000	U	U	1.71	0.129	0.0269	0.0395	0.106	0.518	78.3	0.0118	0.562	0.0124	0.103	0.544	0.219	1.64	0.112	85.5
	Minimum				0.842	0.00180	0.00110	0.00180	0.0634	0.0675	14.7	0.000400	0.370	0.00640	0.0136	0.0192	0.0451	0.794	0.00110	3.46
	Maximum				88.3	0.785	4.00	0.0845	4.18	265	494	7.70	3.36	0.287	2.45	14.9	0.943	4.40	0.429	270
	Arithmetic Mean				6.03	0.0931	0.121	0.0119	0.342	3.48	106	0.117	0.871	0.0360	0.310	0.333	0.197	1.54	0.0871	109
	Standard Deviation				11.7	0.136	0.404	0.0123	0.468	26.2	82.5	0.761	0.438	0.0362	0.432	1.47	0.131	0.469	0.0747	32.5
	Geometric Mean				3.188	0.0377	0.0417	0.00833	0.234	0.580	81.7	0.0230	0.792	0.0282	0.155	0.131	0.167	1.48	0.0580	103
	95% Confidence Interval				2.27	0.0263	0.0784	0.00239	0.0908	5.08	16.0	0.148	0.0850	0.00702	0.0837	0.286	0.0254	0.0910	0.0145	6.30

Table 22. Heavy metal concentrations ( $\mu\text{g/g}$ ) in all whole potential prey samples with the prey's family and species.

Lab ID	Family	Species Name	Al	As	Cd	Co	Cr	Cu	Fe	Hg	Mn	Mo	Ni	Pb	Se	Sn	V	Zn
MSP 542	Acanthuridae	Acanthurus achilles	7.80	1.96	0.397	0.0321	3.27	2.13	110	0.0556	2.29	0.386	0.584	0.321	0.291	2.03	1.54	48.3
MSP 2977	Acanthuridae	Acanthurus dussumieri	166	15.4	0.363	0.0781	4.14	1.03	64.1	0.0140	3.05	0.137	0.900	1.11	0.768	0.396	1.17	12.1
MSP 2450	Acanthuridae	Acanthurus leucopareius	113	4.46	0.119	0.0851	2.18	0.955	35.4	0.0138	3.82	0.0924	0.509	0.436	0.418	0.976	0.795	14.3
MSP 2686	Acanthuridae	Acanthurus leucopareius	142	2.26	0.744	0.0413	0.472	1.02	31.8	0.0217	2.16	0.104	0.454	0.530	0.475	1.23	0.635	15.6
MSP 635	Acanthuridae	Acanthurus nigroris	5.19	4.41	4.62	0.0757	2.10	2.33	66.5	0.116	1.40	0.184	0.457	0.284	0.640	0.832	1.18	33.8
MSP 44	Acanthuridae	Acanthurus triostegus	10.2	1.57	0.382	0.0404	2.16	2.40	90.2	0.0188	2.73	0.165	0.601	0.420	0.276	0.659	0.756	19.6
MSP 1827	Acanthuridae	Ctenochaetus strigosus	197	3.62	0.0538	0.0534	2.65	2.90	953	0.0541	4.13	0.227	0.373	8.58	0.423	2.67	0.941	25.5
MSP 2444	Acanthuridae	Naso unicornis	48.8	5.77	1.04	0.127	2.81	2.23	100	0.0463	1.24	0.194	1.64	0.168	0.537	1.97	2.52	65.3
MSP 2384	Acanthuridae	Zebrasoma flavescens	17.3	1.32	0.118	0.0305	4.01	1.72	113	0.0124	4.10	0.174	0.747	0.266	0.173	1.18	0.712	32.6
MSP 104	Balistidae	Melichthys niger	4.11	24.5	0.212	0.0503	2.11	3.76	124	0.0123	6.95	0.155	0.740	0.106	1.26	1.01	5.08	89.8
MSP 2433	Balistidae	Melichthys niger	108	12.4	1.10	0.0407	3.40	1.49	90.0	0.0526	10.7	0.135	0.913	0.574	0.634	0.980	0.931	48.6
MSP 2976	Balistidae	Melichthys niger	237	15.9	1.07	0.0362	3.14	1.32	86.1	0.0196	13.2	0.153	0.939	0.494	0.608	0.966	2.32	105
MSP 638	Balistidae	Melichthys niger	4.72	19.0	1.02	0.0408	3.25	2.95	94.1	0.0368	18.9	0.286	0.877	0.185	0.570	1.26	1.81	257
MSP 642	Balistidae	Melichthys niger	4.57	12.4	0.345	0.0887	7.08	39.1	120	0.0313	10.1	0.287	30.2	1.61	0.806	1.76	0.901	269
MSP 2779	Balistidae	Melichthys vidua	5.83	7.94	0.712	0.0231	1.61	1.91	90.6	0.0334	15.2	0.255	1.36	0.228	0.400	1.95	2.82	85.8
MSP 2783	Balistidae	Melichthys vidua	5.31	13.7	0.338	0.0801	0.990	2.86	55.5	0.0268	6.69	0.118	1.86	0.173	0.959	0.356	1.18	75.8
MSP 1314	Balistidae	Sufflamen bursa	4.55	25.7	0.134	0.0235	4.47	1.53	87.1	0.0594	7.27	0.141	0.215	0.251	1.18	1.24	3.04	167
MSP 1315	Balistidae	Sufflamen bursa	8.54	37.0	0.260	0.0238	1.12	1.48	116	0.0966	19.4	0.0823	0.271	9.60	1.23	0.922	1.10	355
MSP	Congridae	Conger cinereus	6.54	27.9	0.107	0.0221	1.82	1.86	97.9	0.345	2.83	0.0918	0.305	0.187	1.31	0.274	0.0502	39.7

649																		
MSP 650	Congridae	Conger cinereus	6.63	23.6	0.212	0.0249	2.38	7.60	79.5	0.390	2.13	0.0980	0.145	0.0460	1.09	1.31	0.0351	41.8
MSP 1290	Holocentridae	Myripristis berndti	17.5	47.9	2.81	0.0767	3.02	6.21	144	0.684	1.82	0.142	0.349	0.171	3.51	2.29	0.626	108
MSP 451	Holocentridae	Myripristis berndti	5.19	51.1	0.873	0.0469	2.37	4.24	101	0.0798	0.883	0.120	0.456	0.255	2.67	2.95	0.558	69.1
MSP 1902	Holocentridae	Myripristis chryseres	2.92	38.9	0.579	0.0224	0.727	1.54	101	0.316	0.671	0.0343	0.192	0.323	3.45	0.479	0.200	28.4
MSP 1903	Holocentridae	Myripristis chryseres	2.91	34.5	0.400	0.0513	1.01	1.64	98.1	0.0818	1.37	0.0752	0.215	0.520	2.56	1.51	0.391	44.9
MSP 1291	Holocentridae	Neoniphon sammara	9.42	4.10	0.905	0.0125	1.75	1.63	185	0.405	1.26	0.102	0.108	0.343	1.34	2.22	0.307	61.5
MSP 22	Holocentridae	Neoniphon sammara	3.56	5.81	0.344	0.0091	0.771	1.64	88.6	0.124	1.37	0.0795	0.103	0.103	0.904	2.49	0.243	46.2
MSP 2466	Holocentridae	Neoniphon sammara	194	5.52	0.335	0.0164	1.76	1.56	85.4	0.0448	1.45	0.109	0.175	0.0742	1.09	2.16	0.130	43.1
MSP 23	Holocentridae	Sargocentron diadema	4.07	5.13	0.809	0.0146	1.28	1.56	106	0.155	2.27	0.0727	0.149	0.151	1.72	2.31	0.978	42.7
MSP 242	Holocentridae	Sargocentron xantherythrum	4.37	2.03	0.248	0.0325	6.03	1.40	100	0.0427	2.39	0.195	0.280	0.197	1.56	2.00	0.539	29.5
MSP 245	Holocentridae	Sargocentron xantherythrum	4.53	2.70	0.177	0.0291	9.70	0.873	114	0.0828	2.90	0.277	0.349	0.293	1.88	2.56	0.438	35.1
MSP 728	Labridae	Coris flavovittata	29.3	15.5	0.216	0.0576	3.44	1.90	74.2	0.0276	9.23	0.119	0.412	7.65	0.916	0.337	0.236	60.8
MSP 663	Labridae	Coris venusta	8.64	9.03	0.228	0.0302	2.70	1.10	74.1	0.0549	12.2	0.114	0.269	0.230	1.31	1.13	2.32	46.7
MSP 1995	Labridae	Cymolutes lecluse	98.5	20.5	0.504	0.0342	1.43	1.61	57.1	0.0242	4.15	0.0776	0.262	5.26	2.83	0.366	0.362	43.4
MSP 1937	Labridae	Inistius pavo	32.3	27.3	0.240	0.0228	2.43	2.02	103	0.0530	3.60	0.0870	0.309	0.446	1.11	0.121	0.114	46.8
MSP 1938	Labridae	Inistius umbrilatus	39.8	39.6	0.128	0.0362	4.30	1.23	102	0.0569	4.09	0.121	0.230	0.0838	1.43	0.0701	0.167	44.2
MSP 2344	Labridae	Macropharyngodon geoffroy	29.9	13.0	0.0698	0.0320	2.77	1.58	91.6	0.0324	5.97	0.121	0.426	0.429	0.782	1.07	0.240	27.8
MSP 2860	Labridae	Oxycheilinus unifasciatus	16.9	2.98	0.0729	0.0148	3.99	0.778	58.9	0.0511	2.45	0.111	0.190	0.0427	0.594	2.42	0.195	41.7
MSP 36	Labridae	Thalassoma ballieui	4.26	7.93	0.144	0.0290	6.86	1.32	129	0.0806	4.73	0.189	0.267	0.152	0.692	0.881	0.590	63.3
MSP	Labridae	Thalassoma	13.1	25.5	0.374	0.0173	0.723	1.37	44.6	0.0777	4.86	0.0615	0.113	0.0546	2.05	0.540	0.232	54.4

778		duperry																
MSP 651	Muraenidae	Gymnothorax albimarginatus	7.33	1.58	0.917	0.0155	0.840	5.23	53.2	1.43	13.1	0.0559	0.145	0.0672	2.10	1.17	0.147	60.7
MSP 1880	Muraenidae	Gymnothorax berndti	3.77	52.0	1.03	0.0136	0.978	2.23	71.6	0.715	9.27	0.0626	0.0940	0.0694	1.11	1.60	0.111	63.0
MSP 1789	Muraenidae	Gymnothorax eurostus	3.56	5.66	1.54	0.0193	1.40	3.68	46.6	0.573	9.80	0.0549	0.215	0.0887	2.44	0.515	0.931	49.1
MSP 1810	Muraenidae	Gymnothorax flavimarginatus	57.3	6.11	2.58	0.0127	1.54	5.07	23.1	0.149	8.46	0.0767	0.209	0.0376	1.58	0.156	0.310	39.2
MSP 1794	Muraenidae	Gymnothorax melatremus	8.52	6.28	1.30	0.0530	0.678	2.94	52.9	2.03	2.12	0.0884	0.202	0.449	2.55	0.416	3.22	51.9
MSP 511	Muraenidae	Gymnothorax meleagris	13.5	9.90	0.867	0.0250	1.71	1.64	98.3	0.810	38.8	0.0866	0.175	0.188	2.04	2.44	0.294	61.4
MSP 512	Muraenidae	Gymnothorax meleagris	8.79	7.65	0.827	0.0119	1.65	2.25	35.9	0.470	36.2	0.0587	0.144	0.533	1.05	0.839	0.263	54.8
MSP 296	Muraenidae	Gymnothorax steindachneri	5.20	6.55	0.106	0.0183	0.746	1.30	30.6	0.479	0.520	0.0430	0.106	0.140	2.33	0.140	0.0242	13.9
MSP 257	Muraenidae	Gymnothorax undulatus	24.2	4.59	0.330	0.0206	1.27	13.0	73.7	0.253	8.50	0.0974	0.170	0.212	1.49	1.43	0.685	49.0
MSP 317	Muraenidae	Gymnothorax undulatus	10.0	62.5	0.0441	0.0233	1.15	1.77	101	0.309	73.8	0.0619	0.143	1.20	0.886	1.09	0.134	155
MSP 1783	Octopodidae	Octopus cyanea	76.5	62.1	13.4	0.0602	1.04	69.9	25.1	0.0477	1.70	0.688	0.998	0.109	1.29	0.0276	0.570	61.6
MSP 1784	Octopodidae	Octopus cyanea	6.98	83.5	11.1	0.102	0.802	73.3	25.5	0.0674	2.02	0.629	1.50	0.237	1.50	0.0312	0.808	69.3
MSP 1904	Octopodidae	Octopus cyanea	4.29	130	40.4	0.269	1.73	164	46.3	0.238	2.33	1.55	3.42	1.10	1.71	0.0527	0.982	147
MSP 2798	Octopodidae	Octopus cyanea	27.7	96.1	16.7	0.0868	2.87	108	54.7	0.0654	1.59	0.771	2.76	0.144	1.41	0.0066	0.676	70.7
MSP 2799	Octopodidae	Octopus ornatus	126	329	20.6	0.478	1.30	60.5	90.7	0.238	1.55	0.878	4.11	0.667	17.5	0.0715	1.09	197
MSP 515	Octopodidae	Octopus ornatus	280	165	21.1	0.128	0.586	116	33.7	0.128	2.03	0.589	1.79	0.139	2.02	1.08	1.09	75.1
MSP 135	Octopodidae	Octopus sp.	5.89	84.0	24.2	0.0881	1.82	125	40.5	0.0725	1.79	0.986	1.53	0.387	1.10	0.0245	0.811	76.0
MSP 137	Octopodidae	Octopus sp.	6.85	53.9	15.8	0.0469	0.675	72.7	67.5	0.0584	1.65	0.583	1.90	0.136	1.56	0.0346	0.598	68.2
MSP 326	Palinuridae	Panulirus marginatus	16.0	163	3.26	0.0461	1.20	131	17.9	0.136	0.858	0.133	0.950	0.182	1.24	0.173	0.331	60.7
MSP	Palinuridae	Panulirus	74.3	88.6	2.65	0.0563	1.79	124	29.3	0.0437	1.33	0.171	1.31	0.978	1.13	0.0777	1.06	44.5



329		marginatus																
MSP 334	Palinuridae	Panulirus marginatus	33.3	142	2.81	0.0304	1.95	113	24.9	0.124	0.779	0.169	0.522	0.104	1.25	0.296	0.309	55.0
MSP 335	Palinuridae	Panulirus marginatus	12.6	86.1	3.31	0.0269	0.904	82.6	13.9	0.106	1.08	0.210	0.977	0.453	1.07	0.0472	1.01	50.0
MSP 344	Palinuridae	Panulirus marginatus	37.3	151	4.80	0.0484	2.33	90.0	28.2	0.114	1.34	0.249	1.75	0.109	1.65	0.0596	0.683	63.6
MSP 345	Palinuridae	Panulirus marginatus	17.0	102	2.59	0.0414	3.69	94.2	36.9	0.125	0.802	0.188	0.795	0.132	0.999	0.0630	0.498	48.6
MSP 546	Palinuridae	Panulirus marginatus	18.2	115	1.93	0.0536	1.01	93.6	11.7	0.149	0.963	0.212	1.37	0.185	1.49	0.241	0.656	61.4
MSP 1747	Palinuridae	Panulirus pencillatus	23.4	39.1	15.4	0.0522	0.830	59.0	32.2	0.418	1.15	0.137	1.34	0.202	1.38	2.47	0.487	80.0
MSP 621	Palinuridae	Panulirus pencillatus	8.92	106	5.40	0.0418	2.10	96.0	25.4	0.0978	0.780	0.231	0.531	0.237	1.07	0.111	0.196	80.7
MSP 626	Palinuridae	Panulirus pencillatus	123	85.6	2.16	0.0267	1.07	118	16.0	0.0796	0.896	0.305	0.523	0.214	1.36	0.0431	0.426	80.4
MSP 48	Scaridae	Chlorurus perspicillatus	5.06	3.81	0.237	0.0288	1.07	4.14	42.7	0.0106	4.27	0.105	0.485	0.130	0.805	0.583	1.74	15.5
MSP 495	Scaridae	Chlorurus perspicillatus	10.7	2.24	0.326	0.0432	3.06	2.36	87.7	0.0213	5.75	0.144	0.732	0.205	0.463	2.03	1.15	26.3
MSP 497	Scaridae	Chlorurus perspicillatus	19.5	2.40	0.302	0.0418	3.13	1.83	65.7	0.0137	9.63	0.137	0.558	0.462	0.580	0.892	1.54	17.7
MSP 1620	Scaridae	Chlorurus sordidus	7.98	3.08	0.390	0.110	1.36	2.39	82.2	0.0135	4.00	0.105	0.571	2.49	0.939	1.55	0.573	19.7
MSP 1621	Scaridae	Chlorurus sordidus	5.62	2.59	0.364	0.0448	1.46	2.02	100	0.0205	5.32	0.133	0.354	0.398	0.963	0.931	0.526	19.0
MSP 1355	Scaridae	Scarus dubius	8.67	3.08	0.784	0.0704	2.38	1.67	99.2	0.0224	3.46	0.136	0.296	0.156	1.97	1.64	1.09	15.7
MSP 1440	Scaridae	Scarus dubius	36.3	3.06	0.566	0.0394	0.947	1.83	56.7	0.0226	4.95	0.127	0.466	0.143	1.41	1.82	1.42	25.9
MSP 2501	Scaridae	Scarus dubius	54.5	3.68	0.100	0.0362	4.24	2.07	872	0.0208	4.28	0.288	0.478	1.75	0.838	2.00	0.940	22.8
MSP 2861	Scaridae	Scarus psittacus	19.2	1.63	0.643	0.0306	2.94	1.07	48.2	0.0188	13.6	0.155	0.319	0.202	0.717	0.840	3.68	14.3
MSP 2862	Scaridae	Scarus psittacus	16.0	1.67	0.647	0.0393	2.66	1.57	48.9	0.0361	4.84	0.143	0.325	0.193	1.15	1.32	1.58	13.6
MSP 2966	Serranidae	Epinephelus quernus	3.48	3.40	0.0139	0.0141	0.337	0.683	18.3	0.154	0.359	0.0226	0.0511	0.0323	1.30	1.43	0.0064	19.7
MSP	Serranidae	Epinephelus	6.11	8.99	0.0570	0.0069	0.367	0.822	50.9	0.777	0.362	0.0249	0.0560	0.0260	1.52	0.692	0.0169	19.5

2968		quernus																	
MSP 260	Serranidae	Pseudanthias thompsoni	2.67	4.20	0.178	0.0223	0.746	0.772	38.1	0.0234	5.17	0.0358	0.0952	0.0448	1.22	0.938	0.187	37.2	
MSP 754	Serranidae	Pseudanthias thompsoni	50.3	3.15	0.214	0.0542	0.648	1.59	58.9	0.0424	3.30	0.0507	0.155	0.198	1.96	0.894	0.188	30.8	

