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Detection of Chromium (Cr) and Lead (Pb) in Marine Sediments and its Bioaccumulation in *Holothuria leucospilota* (Sea Cucumber) in Jasaan, Misamis Oriental, Philippines

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ABSTRACT

Sea cucumber inhabits shallow waters of the coastal area. Consequently, these organisms are vulnerable to heavy metals through food consumption and direct exposure to sediments, considering that they are filter-feeding and bottom-dwelling organisms. *Holothuria. leucospilota*, locally known as "*balat*" is used as a bioindicator to measure environmental pollution in this study. The analysis of Cr and Pb were administered in the sediments and the body wall of *H. leucospilota* from residential and commercial areas along the coastal area of Jasaan, Misamis Oriental. Cr and Pb concentrations were determined using the Atomic Absorption Spectroscopy (AAS) method for sediments and the Graphite Furnace Atomic Absorption Spectroscopy (GFAAS) method for *H. leucospilota*. Results of the concentration of heavy metals in the sediments and body wall of *H. leucospilota* revealed that Cr>Pb while Pb>Cr in residential and commercial areas, respectively. The results of Cr and Pb concentration in the body wall of *H. leucospilota* were compared to the standard set by WHO/EPA and CODEX. Thus, *H. leucospilota* collected in the areas exceeded the permissible limit for Cr while Pb in *H. leucospilota* recorded below the permissible limit. Hence, this implies that this type of food is not safe for food intake and therefore, not recommended for human consumption.

INTRODUCTION

Heavy metals are considered a primary anthropogenic pollutant in marine and coastal environments (Yu et al., 2017). Through anthropogenic sources including industrial, municipal, and domestic waste discharges, heavy metals can directly access the coastal area (Fu and Wang, 2011), either dissolved or adsorbed onto suspended particles, which in turn, settle down into the marine sediments (Remeikaite-Nikiene et al., 2018). Aquatic organisms particularly bottom-dwelling species, may actively absorb dissolved chemicals from water through their respiratory systems, through their skin, or by consuming sediments, detritus, and prey contaminated with heavy metals (Asif et al., 2020; Siddika & Parveen, 2022). Thus, may affect bioaccumulation rates in the organisms (Katagi, 2010). According to Spanopoulos-Zarco et al. (2014), the presence of heavy metals caused by human activities including industrial and domestic waste discharges would eventually build up and become biomagnified in the food chain, harming aquatic life and possibly having an impact on human health when ingested.

One of the marine organisms considered a potential bio-indicator of heavy metal pollution is sea cucumbers. These organisms are underclass Holothuroidea and are considered a high-diversity group of the Phylum Echinodermata, which are found in almost every marine environment (Hashmi et al., 2014). They live in a vast array of habitats and depths, from the lowest floor of oceanic trenches to shallow intertidal areas. Moreover, considering sea cucumbers are bottom-dwelling, filter-feeding organisms that immediately consume sediment and break down surface debris, they are more likely to accumulate both metallic and non-metallic pollutants from their environment, particularly marine sediment (Aydın et al., 2017).

Jasaan, Misamis Oriental is situated along the coastal area which is surrounded by large industrial establishments and a large human population. Research using sea cucumbers as bioindicators of heavy metal pollution in urbanized and industrialized areas is less studied. Furthermore, *H. leucospilota* is considered one of the edible species of sea cucumbers, specifically its body wall compartment, and are considered vulnerable organisms since they are sedentary on the ground where sediments. Hence, consumed by the people as food and may pose health effects to people consuming it. Thus, this study aimed: (a) to determine the Chromium (Cr) and Lead (Pb) in *H. leucospilota* and marine sediments, as a measure of environmental pollution; (b) to compare the concentration of Cr and Pb in the body wall of *H. leucospilota* as set by World Health Organization, Environmental Protection Agency and Codex Alimentarius, respectively*;* and (c) to determine the percentage accumulation of Cr and Pb in the body wall of *H. leucospilota*.

MATERIALS AND METHODS

Research Design

The study employed a descriptive-comparative type of research in gathering and analyzing the data needed in this study. Thus, the type of environment was also observed, while different activities in the area were recorded as this can give insights into some factors affecting the area.

Study Area

The area is situated in the intertidal area of Jasaan, Misamis Oriental (8˚38''57.8'' N, 124 ˚45'17.7''E). Two 150-meter sampling sites were established along the intertidal area using a transect meter where marine sediment and sea cucumber (*H. leucospilota*) were collected. The residential area lies at geographical coordinates of 8˚ 38. 352' North Latitude and 124˚ 45. 763' East Longitude. Within the sampling area is the presence of a waterway originating from locals living in the area. Adjacent to it is a mangrove area and an existing anchorage of ships and boats, industrial establishments, and an industry for fish port purposes. This site also has a sandy muddy substrate and the presence of seagrass within the area. Measured 4.02 km away from the Commercial area.

The commercial area is specifically located at 8˚ 36. 150' North Latitude and 124˚ 45.885' East Longitude. Along the sampling site is land used in which housing predominates. Adjacent to it is also an anchorage for ships and boats, different industries, and also a power plant industry which was already inactive, five years ago. This site also has a muddy substrate and the presence of seagrass within the area.

Collection of Samples

Marine Sediment

A collection of marine sediment was conducted within the established areas during low tide. Sediment samples were collected thru grab sampling method at approximately 5cm in depth from the upper section of the sediment. A depth where the majority of sea cucumbers bury themselves in the sand to feed (Aydin et al., 2017). Collected samples were sealed and labeled in polyethylene bags (zip locks) and labeled it using markers. Foreign materials on sediments were then removed. Samples were weighed to attain 300g before it is subjected to heavy metals.

Heavy Metal Analysis for Marine Sediment

Analysis of Cr and Pb (Figure 2) was determined by obtaining 5g sample of sediment for each metal (Pb and Cr). This was analyzed using the Atomic Absorption Spectroscopy (AAS) test method (Manual on Standard Analytical Procedures of the Mines and Geosciences Bureau Laboratories).

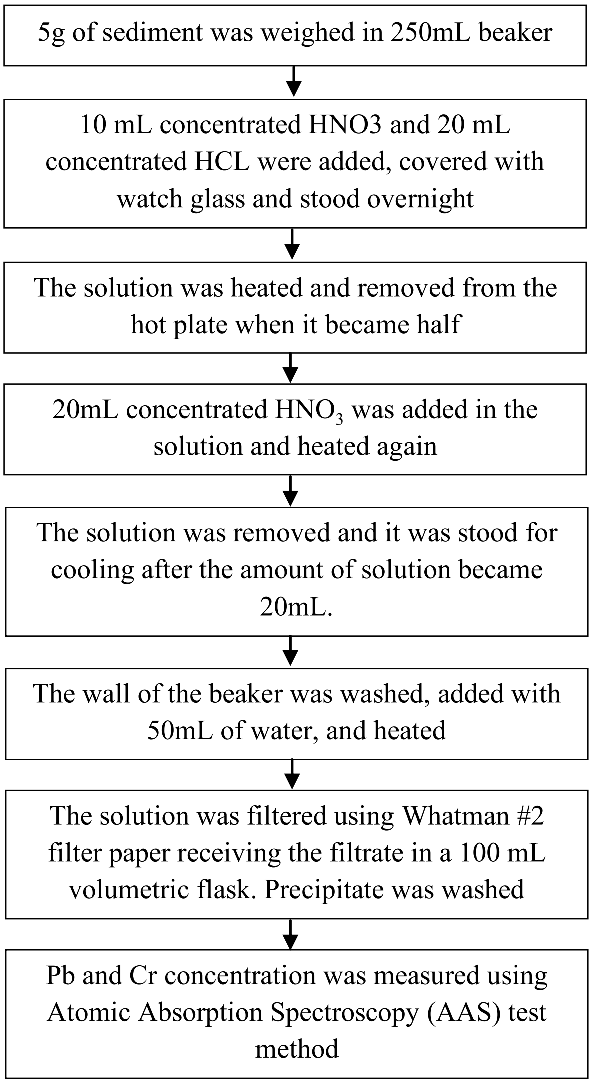

Figure 1. Determination of Cr and Pb in Marine Sediments.

H. leucospilota Body wall

A collection of an adult of *H. leucospilota)* along the established sampling areas were employed during low tide. Sea cucumbers (*H. leucospilota)* were collected by burrowing in the sand with proper hand protection. The collected fresh sea cucumber samples were kept alive and placed in a container with a proper label, filled with seawater (Ahmed et al., 2017). Samples were then washed with running water. *H. leucospilota* samples were cut longitudinally the internal organs were removed and the body wall was weighed for the determination of Cr and Pb.

Heavy Metal Analysis for sea cucumber (*body wall*)

For the evaluation of Cr and Pb in the body wall of sea cucumber (Figure 1), flame and the Graphite Furnace Atomic Absorption Spectroscopy (GFAAS) technique were used to identify analytes (Manual on Standard Analytical Procedures of the Mines and Geosciences Bureau Laboratories).

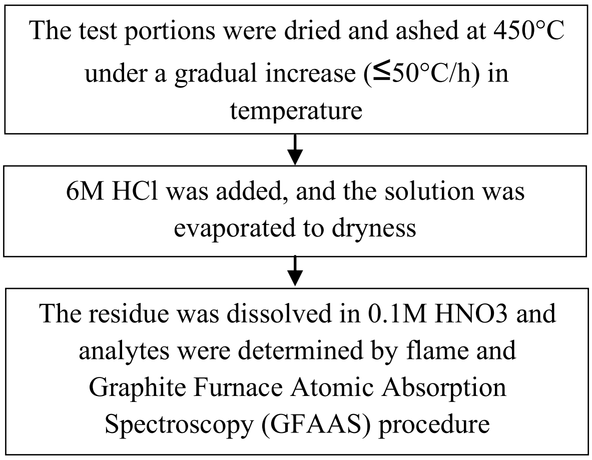

Figure 2. Determination of Cr and Pb in sea cucumber (body wall).

Statistical Analysis

The t-test statistical tool at 0.05 level of significance was employed to compare the detection of heavy metals namely, Cr and Pb in marine sediments and its bioaccumulation on sea cucumber (*H. leucospilota)* in two (2) sampling areas.

RESULTS AND DISCUSSION

Chromium (Cr) and Lead (Pb) in Marine Sediments

Heavy metals in the environment are both contributed by anthropogenic and natural sources. Significant harm has been caused to the natural geochemical cycling of trace metals into different ecosystems, principally due to an increase in man-made or anthropogenic activities in coastal cities (Mahu et al., 2015). In receiving bodies of water, heavy metals are presented fundamentally as hydroxy-complexes of low solubility associated with the suspended water-borne particles. After a sequence of natural processes, the water-borne heavy metals eventually accumulate in the sediment and the concentration of heavy metals in the sediment indicates how contaminated the marine environment is (Dong et al., 2013).

Hence, marine sediments play an important role as they serve as a habitat for some living organisms including sea cucumbers, and at the same time a place where pollutants including heavy metals will settle. Table 1 shows the mean concentration of heavy metals in mg/kg particularly Cr and Pb detected in marine sediments in two sampling sites along the coastal area of Jasaan, Misamis Oriental.

Table 1. Heavy Metal Concentration in Marine Sediments in Residential and Commercial Areas

Heavy Metals (mg/kg)	Sampling Site 1	Sampling Site 2	Standard (SQCG)
Chromium	5.67	5.0	26
Lead	4.48	5.85	31

SQCG- Sediment Quality Criteria Standard (1992)

Cr detection in marine sediments obtained the highest concentration among the two heavy metals in residential areas at 5.67 mg/kg and does not exceed the Sediment Quality Criteria Standard (SQCG). However, the presence and the contamination of Cr in marine sediments in residential areas may be due to its surrounding environment such as industrial establishments and a waterway coming from residential areas, which may have aroused the Cr concentration in sediments. According to Wang and Choi (2013), Cr is a ubiquitous contaminant that is widely used in industrial applications. This contaminant enters the aquatic environment through natural processes and human activities.

In the Commercial area, Pb obtained the highest concentration among the two heavy metals detected in marine sediments at 5.85 mg/kg and does not exceed the Sediment Quality Criteria Standard (SQCG). The presence of Pb in the commercial area may be due to municipal and domestic sewage discharges and an existing anchorage of ships and boats adjacent to the area. According to Walker et al. (2018), petroleum spills including gasoline from ships and boats due to accidental discharges caused by technological failure or malfunction and human error. Eventually, wave action incorporates oil containing Pb into the water column which persists in the environment and sinks through the water column into marine sediments.

In the study of Elvira et al. (2016), anthropogenic activities such as waste discharges from point and non-point sources cause the increasing content of heavy metals including Cr and Pb in marine environments. Heavy metals are released into the marine ecosystem in coastal zones by anthropogenic sources such as surface runoff, domestic and industrial sewage discharges (Pan & Wang, 2012; Ra et al., 2014) and with the largest release coming from industrial establishments (Tchnounwou et al., 2012). Consequently, heavy metals released in the coastal area will incorporate into suspended particles, which in turn, will settle down to the marine sediments (Remeikaite-Nikiene et al., 2018) and hence, remain in solution for a long time (Ahmed et al., 2017).

Heavy metals in aquatic systems are either carried in a dissolved or particulate form as a suspended load or bedload, and they eventually integrate with abiotic elements including the atmosphere, water, and marine sediments; they can also be carried over great distances (Boxberg, 2017). This may be the reason for the close concentration values obtained in two sampling areas for Cr and Pb in marine sediments. According to Green and Coco (2014), the transportation of heavy metals to other areas is due to strong hydrodynamic processes and sediment bioturbation. Marine sediments will eventually resuspended and sent back to the overlying seawater by wind-wave-generated bottom stress (van Maren et al., 2015) and consequently, larger-scale redistribution of the heavy metal contaminants transported by the sediments through physical processes including wind-driven waves and tidal currents, thus, causing heavy metal pollution to disperse to other areas

However, Cr and Pb concentrations do not exceed the Sediment Quality Criteria Standard (SQCG). This may be due to the distance from the source of pollutants. Additionally, based on the findings of Boxberg (2017), the transport of heavy metal pollutants in water and marine sediments caused by physical processes including waves and tidal currents, the concentration of heavy metals tends to decrease and become more diluted with increasing distance away from the input source. Wave resuspension has also been shown to cleanse marine sediments of heavy metals (Green & Coco 2014).

Furthermore, According to Li et al. (2017), low concentration of heavy metals in marine sediments may be due to the characteristics of marine sediments and the presence of organic matter in two sampling sites, for it is well known that grain size and organic matter are two primary factors influencing the heavy metal mechanisms in marine sediments. Two sampling areas may have low levels of organic matter; consequently, high concentrations of organic matter have been indicated to have a high affinity for heavy metals in marine sediments through adsorption or complexation. (Chakraborty et al., 2014). Thus, heavy metal concentrations, particularly, Cr and Pb in marine sediment are within the permissible limit set by SQCG, however, the low concentration of Cr and Pb in sediments may still pose a great threat to some benthic organisms including sea cucumbers.

Chromium (Cr) and Lead (Pb) Concentration in H. *leucospilota*

Heavy metals can directly access the coastal area (Fu and Wang, 2011), either dissolved or adsorbed onto suspended particles, which in turn, will settle down and incorporate into marine sediments (Remeikaite-Nikiene et al., 2018). Aquatic organisms including bottom-dwelling species, may directly absorb dissolved chemicals from water through the respiratory system, through the skin, or may consume chemicals through the ingestion of detritus, heavy metal-contaminated sediments, and prey. Thus, may affect bioaccumulation rates in the organisms (Katagi, 2010) it would unavoidably collect and become biomagnified in the food chain, harming aquatic life and perhaps impacting human health when ingested. (Spanopoulos-Zarco et al., 2014). Table 2 shows the mean concentration of heavy metals in mg/kg particularly Cr and Pb, which were detected in *H. leucospilota* collected from the residential and commercial areas of Jasaan, Misamis Oriental, Philippines.

Table 2. Heavy Metal Concentration in H. *leucospilota* (Body Wall) in Two Sampling Sites

Heavy Metals	Site 1	Site 2	Standard
Chromium	0.64	0.49	0.15 mg/kg (WHO/EPA)
Lead	0.43	0.85	1 mg/kg (Codex Alimentarius)

Cr>Pb is detected in the body wall of *H. leucospilota* in the residential are at 0.64 mg/kg. However, Cr concentration detected in *H. leucospilota* in two sampling areas exceeded the maximum allowable limit set by WHO/EPA which is 0.15 mg/kg. Conversely, Pb>Cr was detected in *H. leucospilota* at 0.85 mg/kg in the commercial area, while Pb<Cr was detected in *H. leucospilota* in both sampling areas recorded below the permissible limit. However, this may pose a risk since anthropogenic activities with improper management will continue to exacerbate the concentration of heavy metals in a marine environment, thus contaminating sea cucumber specifically *H. leucospilota*, which could possibly reach beyond the permissible limit sooner.

Thus, Cr and Pb accumulation in aquatic organisms including sea cucumber may be due to its surrounding environment, in which both sampling areas are adjacent to anthropogenic interventions such as industrial establishments and residential areas. In due course, industrial effluents and domestic sewage from these sources are directly discharged into the receiving water body, where heavy metals are introduced. These heavy metals will eventually settle down and accumulate in marine sediments (Remeikaite-Nikiene et al., 2018), and get rapidly integrated into marine benthic organisms including sea cucumbers. Additionally, an organism's overabundance of both necessary and non-essential heavy metals can harm its cells and tissues, which can result in diseases (Boxberg 2017), Moreover, heavy metals can accumulate in the human body through the amplification effect of the food chain (Li et al., 2018; Talabi et al, 2023). Thus, exposure to heavy metals through food consumption may cause a great risk to human health.

Damage to the blood composition, lungs, kidneys, liver, and other essential organs, as well as diminished or impaired mental and central nervous system function, can all be consequences of heavy metal toxicity. Prolonged exposure to heavy metals can cause progressively developing neurological, musculoskeletal, and physical degenerative processes that resemble multiple sclerosis, Alzheimer's disease, Parkinson's disease, and muscular dystrophy. (Tripathi and Tiwari, 2012). Exposure of humans to high enough concentrations of Cr could be harmful due to its carcinogenic, genotoxic, and toxic properties (Zohdi et al., 2012; De Flora 2016; Zhang et al., 2011). Cr effects frequently appear as rashes or irritation brought on by allergies. Dryness, erythema, fissured skin, papules, tiny vesicles, and swelling are some of the signs of dermatitis. Other symptoms could worsen if the dosage is higher than the threshold. For mild symptoms, Cr may cause dizziness, general weakness and eye irritation. In terms of severe symptoms, it may include development issues, nasal perforation, ocular injury, renal, liver, gastrointestinal, cardiac, hematologic, or reproductive diseases (Zhang *et al*., 2011). However, the aforementioned cases were not yet evident in the area, and thus, should conduct health monitoring within the community.

Furthermore, Pb toxicity poses major risks to one's health, including long-term brain damage that results in learning difficulties, hearing loss, odd behavior, hypertension, blood pressure issues, and heart disease (Tiwari and Tripathi, 2012). Once in the bloodstream, Pb is mostly found in soft tissue, mineralizing tissue, and blood (Yu et al., 2016). Chronic Pb intoxication in adults causes anemia, some cancers, problems with male reproduction, and hormonal imbalances related to vitamin D metabolites (Tandon et al., 2001; Tiwari and Tripathi, 2012). Thus, exposure to heavy metals through food consumption may cause a great risk to human health and may cause serious health hazards.

Bioaccumulation Percentage in the body wall of H. *leucospilota*

Heavy metals become quickly assimilated, especially into marine benthic organisms like sea cucumbers, which lead sedentary lives by eating, burrowing, and engaging in social behaviors in marine sediments. These are activities that cause heavy metal accumulation in their bodies from feed consumption and direct exposure to the sedimentary environment (Jinadasa et al., 2014). Table 3 shows the percent level of bioaccumulation of Cr and Pb in the body wall of H. *leucospilota* from the concentration of heavy metals in marine sediments.

Table 3. Percent Level of Heavy Metal Bioaccumulation in H. *leucospilota* (body wall).

Heavy Metals	Sampling Site 1	Sampling Site 2
Chromium	11.29%	9.8%
Lead	10%	14.53%

In sampling site 1, the body wall of *H. leucospilota* species accumulated 11.29% of the total concentration of Cr detected in marine sediment and an equivalent of 10% bioaccumulation of Pb in *H. leucospilota* from the marine sediments. In sampling site 2, the body wall of *H. leucospilota* species accumulated 9.8% of the total concentration of Cr detected in marine sediment and an equivalent of 14.53% bioaccumulation of Pb in *H. leucospilota* from the marine sediments. According to Givianrad et al. (2014), since the body wall makes up 85% to 90% of the sea cucumber's total body mass, it is typically characterized by lower metal concentrations. On the other hand, the digestive tract is a very tiny volume of material, and the metals that have accumulated there are highly concentrated (Warnau et al., 2006; Jinadasa et al., 2014). In addition, the accumulation of intestinal tissue and sediment has a very strong association, while it remains low in the body wall compartment. Moreover, the low percentage bioaccumulation in *H. leucospilota* may be due to the characteristics of marine sediments and the presence of organic matter in the sampling site, for it is known that grain size and organic matter are the two primary determinants of the heavy metal mechanism in marine sediments (Li et al., 2017). Organic matter content in two sampling sites may be low thus, high organic matter content was demonstrated to have a high affinity through adsorption or complexation for heavy metals in sediments (Chakraborty et al., 2014).

Furthermore, sea cucumber is inversely proportional in terms of the organism's variation such as the species of different sizes, the lengths and weights of the organisms to the concentration of heavy metals such as Cr and Pb accumulated in their body (Ahmed et al., 2017). Hence, other significant factors affecting the accumulation of elements in living organisms are characteristics of the elements accumulated including essential or non-essential elements, their solubility in water and structure, the concentrations of heavy metals, duration of exposure of pollutants in the organisms, the existence of additional substances or elements in the surroundings, the physical and chemical characteristics of the medium, the habitat in which an organism lives, feeding habits or behaviors, the organism's condition, sex, and, in certain situations, its metabolic rate (Jinadasa et al., 2014). Thus, these features have a great influence in terms of heavy metal accumulation.

CONCLUSION

Based on the findings along Jasaan, Misamis Oriental, Philippines, the concentration of marine sediments is Cr>Pb in residential areas while Pb>Cr in commercial areas. This is due to anthropogenic activities such as waste discharges from point and non-point sources which greatly affect the concentration of Cr and Pb. Furthermore, low concentrations in the percentage of Cr and Pb were detected in the body wall of H. *leucospilota* due to its physiological characteristics and varying size may have influenced the concentration uptake. However, Cr concentration accumulated in H. *leucospilota* exceeded the maximum allowable limit set by WHO/EPA, while Pb concentration was recorded below the permissible limit for food consumption. This indicates that consuming this type of food may cause adverse effects on human health, particularly H. *leucospilota* species in the coastal area of Jasaan, Misamis Oriental, Philippines.

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