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# ASSESSMENT OF HEAVY METAL ACCUMULATION IN THE STRAITS OF JOHOR: A COMPREHENSIVE STUDY ON WATER, SEDIMENT, AND AQUATIC ORGANISMS

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## **Graphical abstract**

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# Abstract

This study evaluated the marine water quality of the Johor Straits near Kampung Pasir Putih, revealing risks to aquatic life and aquaculture from land use changes, increased household, agricultural, and industrial waste discharge. The distribution and concentration levels of heavy metals (Zn, Cu, Cd, and Pb) in crabs, mussels, and sediment samples were particularly investigated in this study. For Zn, Cu, Cd, and Pb, the corresponding mean concentration values in the entire body of the crabs and mussels were 60.4 to 256.5 mg/g, 21.8 to 50.3 mg/g, 1.6 to 4.5 mg/g, and 8.2 to 23.7 mg/g dry weight. In and Cu consistently showed higher concentrations than other elements, with varying distribution across sampling stations. The average amounts of heavy metals in the sediment samples ranged from 17.7 to 147.1 mg/g, 17.7 to 155 mg/g, 2.4 to 5.3 mg/g, and 9.8 to 14.5 mg/g dry weight for zinc, copper, lead, and mercury, in that order. The study found that increased development leads to declining water quality and higher heavy metal accumulation in marine organisms. Dissolved oxygen levels (1.36 to 6.71 mg/L) were linked to this metal buildup. The ranges of concentrations for Zn, Cd, Cu, and Pb in marine water were 0.01 to 0.6 mg/L, 0.04 to 0.22 mg/L, and 0.03 to 0.33 mg/L, respectively. Kampung Pasir Putih was classified as Class 3 under Malaysia's Marine Water Quality Standards, indicating moderate contamination. The findings call for better management and proactive steps to protect the marine ecosystem and food resources.

Keywords: Johor Straits, marine water quality, Heavy metals (Zinc, Copper, Cadmium, Lead), Land use

# Abstrak

Kajian ini menilai kualiti air marin Selat Johor berhampiran Kampung Pasir Putih, mendedahkan risiko kepada hidupan akuatik dan akuakultur daripada perubahan guna tanah, peningkatan pembuangan sisa isi rumah, pertanian dan industri. Taburan dan tahap

# **Full Paper**

#### Article history

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\*Corresponding author shamila@utm.edu.my kepekatan logam berat (Zn, Cu, Cd, dan Pb) dalam sampel ketam, kupang dan sedimen telah disiasat secara khusus dalam kajian ini. Bagi Zn, Cu, Cd, dan Pb, nilai kepekatan min yang sepadan dalam keseluruhan badan ketam dan kerang ialah 60.4 hingga 256.5 mg/g, 21.8 hingga 50.3 mg/g, 1.6 hingga 4.5 mg/g, dan 8.2 hingga 23.7 mg/g berat kering. In dan Cu secara konsisten menunjukkan kepekatan yang lebih tinggi daripada unsur lain, dengan pengedaran yang berbeza-beza merentasi stesen pensampelan. Purata jumlah logam berat dalam sampel sedimen adalah antara 17.7 hingga 147.1 mg/g, 17.7 hingga 155 mg/g, 2.4 hingga 5.3 mg/g, dan 9.8 hingga 14.5 mg/g berat kering untuk zink, kuprum, plumbum dan merkuri, mengikut urutan itu. Kajian mendapati peningkatan pembangunan membawa kepada penurunan kualiti air dan pengumpulan logam berat yang lebih tinggi dalam organisma marin. Tahap oksigen terlarut (1.36 hingga 6.71 mg/L) dikaitkan dengan pembentukan logam ini. Julat kepekatan untuk Zn, Cd, Cu, dan Pb dalam air laut ialah 0.01 hingga 0.6 mg/L, 0.04 hingga 0.22 mg/L, dan 0.03 hingga 0.33 mg/L, masingmasing. Kampung Pasir Putih diklasifikasikan sebagai Kelas 3 di bawah Piawaian Kualiti Air Marin Malaysia, menunjukkan pencemaran sederhana. Penemuan ini memerlukan pengurusan yang lebih baik dan langkah proaktif untuk melindungi ekosistem marin dan sumber makanan.

Kata kunci: Selat Johor, kualiti air marin, Logam berat (Zink,Kuprum, Kadmium, Plumbum), Guna tanah

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## **1.0 INTRODUCTION**

The southern part of Peninsular Malaysia and Singapore are separated by the Straits of Johor, which have long been a source of environmental concern [1]. The primary source of this concern is the unique characteristics of the strait, which include a narrow and shallow waterway, significant industrial expansion, rapid urbanization, and a flurry of commercial vessel activity. Beyond its economic worth, the region is naturally diverse and supports significant ecosystems such mudflats, mangroves, seagrass beds, and coral reefs [2]. But sadly, human activities such as aquaculture practices, urbanization, industrial wastes, and agricultural attempts have created a continuous danger to the Straits of Johor's environmental health. This paper aims to comprehensively explore the heavy metal content within the Johor Straits utilizing crabs as a bio-indicator. The objectives encapsulated in this investigation are multifold. Firstly, it seeks to scrutinize the distribution and concentration levels of zinc (Zn), copper (Cu), cadmium (Cd), and lead (Pb) in both sediment and seawater across various study areas within the straits. Secondly, it endeavors to assess the extent of heavy metal accumulation within aquatic life, particularly focusing on crabs and mussels as representatives. The papers' final goal is to create models that clarify the patterns of heavy metal bioaccumulation in sand and crabs, providing insight into the complex processes of metal intake and deposition in the Johor Straits' marine ecosystem. With the help of these goals, the research hopes to make a substantial contribution to our understanding of the biological and environmental effects of heavy metal pollution in this important marine region [4].

As stated, the rising degree of heavy metal contamination in the Straits of Johor is one important consequence of human activities. Approximately 62% of all sources of heavy metals, including lead (Pb), zinc (Zn), copper (Cu), nickel (Ni), cadmium (Cd), chromium (Cr), and manganese (Mn), continuous industrial effluent discharge is a persistent source of concern [3]. These metals not only degrade the water's quality but also pose a serious threat to the range of aquatic organisms that inhabit the area. In addition, consuming contaminated seafood exposes human populations to health risks [2].

Assessing the health and sustainability of aquaticecosystems requires а thorough understanding of water quality characteristics, such as dissolved oxygen (DO), temperature, pH, salinity, and conductivity. These metrics are basic markers of ecosystem health, and variations frequently indicate possible effects on aquatic flora and fauna. The dynamics of assessing water quality are further complicated by the interaction of tidal hydrodynamics, atmospheric factors, and water characteristics [4] Observable patterns in air and surface ocean temperatures reflect diurnal variations as well as inter-decadal and centenary scale variability, indicating the interaction between atmospheric and oceanic processes across multiple time scales [5] The health of aquatic ecosystems depends heavily on dissolved oxygen (DO) measurement, which quantifies the quantity of gaseous oxygen  $(O_2)$  dissolved in water. Temperature, salinity, turbulence, and air pressure are some of the

variables that affect DO concentration and cause variations in its levels [6]. In addition, the presence of nutrients, total solids, and living things in water bodies influences the concentration of DO, and fluctuations in DO are seen because of respiration, photosynthesis, and bio depletion [7]. The distribution and behavior of aquatic species are influenced by several characteristics such as pH, temperature, and salinity, which are strongly related to DO levels and further affect aquatic ecosystems. Specifically, pH is a crucial measure of the chemistry of water, impacting not only the well-being of marine life but also the toxicity of certain substances [8]. Conversely, temperature has a major impact on the dynamics of aquatic ecosystems and the composition of aquatic species because it regulates biological activities such as growth, metabolism, and reproduction [23]. The quantity of salts dissolved in water is expressed as salinity, which is measured in parts per thousand (ppt) and has a major impact on the distribution and behavior of marine species [9][24].

Apart from the criteria related to water quality, the composition of sediments becomes a crucial factor in determining the overall health of the ecosystem, especially in terms of the build-up and movement of contaminants like heavy metals. According to Sarkar et al. (2004), sediments are essential for the fate and movement of contaminants in aquatic ecosystems because they serve as reservoirs for pollutants. According to Maringo et al. (2006), the mobilization capability and bioavailability of heavy metals in sediments are determined by their geochemical forms, which include complexation, absorption, and precipitation. These forms also have an impact on the potential effects of the metals on aquatic ecosystems. Furthermore, the reactivity and environmental destiny of heavy metals are further modulated by their interaction with different fractions of sediment, including exchangeable, carbonate-bound, and organic-bound fractions. To protect the health of aquatic ecosystems and lessen the effects of human activity on marine biodiversity, management strategies must consider the complex relationships that exist between various aspects of water quality, sediment dynamics, and pollutant fate [25].

Further investigation into crab species in the Straits of Johor is crucial due to their significance in human nutrition and the surrounding ecological balance. It is particularly common in the area where crabs are an essential source of protein for human consumption and make up a sizeable portion of the diet [4]. But the crabs are more than just food to our ecosystem; they are essential to the complex coastal ecosystems around the Straits of Johor. These crustaceans are significant to the ecosystem; however, it has been shown that they bioaccumulate toxic compounds, particularly heavy metals, which might be dangerous for aquatic life and human health [10].

Comprehending the patterns and processes of heavy metal bioaccumulation in crab species is crucial for evaluating the potential health hazards linked to their intake and for putting into practice efficient management techniques that protect human health and the integrity of the coastal ecosystem [12]. Furthermore, examining crab species as bioindicators of heavy metal pollution sheds light on the state of the Straits of Johor's environment and emphasizes the need for more study to fully understand their function in coastal ecosystems and the health consequences for humans.

According to Wager and Boman [7], Yap *et al.* [16], the area's heavy metal contamination problem is becoming worse, which highlights the necessity for additional in-depth research to ascertain its ecological and health impacts on crucial marine species, including crabs. Considering the natural richness of the Straits of Johor and its critical role as a shipping waterway, efficient monitoring and mitigation strategies are needed [1]. The next sections will look at specific studies conducted in the Straits of Johor to understand the intricate dynamics of heavy metal pollution, with a focus on using transplanting experiments to measure the quantities of heavy metals in green mussels (Perna viridis) [5].

#### 2.0 METHODOLOGY

#### **Study Area**

The study area is situated by an estuary in Kampung Pasir Putih, which is in the Malaysian state of Johor. Figure 1 shows the sampling site at the Straits of Johor, Malaysia. Table 1 displays the coordinates of sampling stations in the study area.



Figure 1 The sampling site at the straits of Johor, Malaysia

These stations are strategically located to gather data in the estuary and surrounding regions. The coordinates provide a precise geographical reference for the research conducted in this significant economic zone. Its exact coordinates are 1°26'30" North, 103°55'50" East. As the nexus for the economies of Singapore, Malaysia, and Indonesia, Kampung Pasir Putih is a major place in terms of economic significance. Because of its location on the Johor Strait, this prominence is particularly noticeable. The decision to relocate the green mussel aquaculture operations from Sungai Melayu to this area was influenced by this. Near the Straits of Johor, several economic subregions have grown in this area, making use of current infrastructure, and encouraging plans for expansion.

 Table 1 Coordinates of sampling stations

No.	Parameters	Capability Values
\$1	Sungai Kampung Pasir Putih	01°25'56.2 N, 103°56'49.2 E
S2	Green mussels aquaculture area I	01°25'48.6 N, 103°56'34.1 E
\$3	Near Villagers house and restaurants	01°25'59.4 N, 103°55'50.9 E
S4	Sungai Laloh	01°26'13.2 N, 103°55'27.3 E
S5	Near Johor Port Terminal	01°26'06.3 N, 103°55'15.9 E
S6	Green mussels aquaculture area II	01°26'00.4 N, 103°55'34.5 E

#### **Sample Collection and Preparation**

S1 to S6, six well-chosen sample sites, were positioned along the estuary with the purpose of concentrating on places with residential zones, the influence of Sungai Laloh, and areas with aquaculture of fish and green mussels. The Perna viridis, or green mussels, were collected from three different stations and came in a variety of sizes, representing both juvenile and adult stages. The study spanned one year, encompassing sampling during both the wet and dry seasons at six selected locations along Sungai Laloh. The time frame of the study was from January 2018-2019. From this analysis, three separate size groups were identified by methodically classifying the measured shell lengths: small, medium, and giant. Exact categorization was achieved by establishing certain length requirements. Small shells were described as having a length of 0 to 5 centimeters, signifying the early stages of mussel growth. Shells that were in the middle of the development phases were classified as medium-sized and measured between 5 and 10 centimeters. Large shells, or mature individuals, were defined as those longer than 10 centimeters. This strict classification method made sure that sizes were categorized consistently and made it easier to accurately analyze size-dependent factors in the study, which strengthened the validity of the research findings. The sampling process carefully followed a digestion protocol that was modified from Llagostera and Romero's (2011) work.

To study the biological dynamics of crab populations in the area, intertidal zones along the Straits of Johor shoreline were the site of systematic crab sample collection. The goal of the sample technique was to minimize environmental disturbance while guaranteeing thorough coverage of a variety of intertidal environments. To make it easier to access intertidal regions, the sampling missions were carefully scheduled for low tide periods. To enable systematic sampling across various ecosystems, transect lines were built perpendicular to the coastline, and sample sites were assigned at regular intervals along each transect. Quadrat sampling was used at each sample site, with standardized 1-square-meter quadrats randomly positioned across the intertidal zone. Trained field researchers visually surveyed each quadrat to determine the presence and number of crab specimens [16]. To adequately represent the variety of crab populations, special emphasis was paid to capturing the abundance and size distribution of several crab species. Gentle handling procedures were employed to collect crab specimens manually, with the aim of minimizing stress and disruption to the surrounding ecosystem. After each item was carefully moved to its own container, it was guickly preserved in preparation for further study. To give context for evaluating crab distribution patterns and habitat preferences, environmental variables, such as water temperature, salinity, substrate type, and habitat parameters, were collected at each sample site. Specimens of collected crabs were brought to the lab for additional examination, where they underwent measurements of morphological features and taxonomic identification. To evaluate crab abundance, species composition, and habitat relationships, data from the sampling operations were analyzed. The analysis yielded important insights into the ecological dynamics of crab populations in the intertidal zones of the Straits of Johor [7,17]

Decomposition was one of the first steps in the process, followed by thorough washing and drying until the desired weight was reached. The dried samples were then carefully combined with HNO<sub>3</sub>. A comprehensive metal analysis was performed on the resultant specimen using a Perkin Elmer Atomic Absorption Spectrometer [5,6]. Making sure the specimens were consistent and clean for further examination was one of the first procedures in sample preparation. As part of this procedure, any organic waste or dirt that had adhered was gently washed away using distilled water. The samples then went through a drying process to reach a constant weight. Depending on the conditions, this procedure might be done in an oven set at a certain temperature of 70°C for around 24 hours or by exposing the samples to sunshine. After the samples had sufficiently dried, they were combined with HNO<sub>3</sub> to undergo digestion. This was an important step since it released the bound metals for examination. To ensure accuracy and precision, the digestion process was carefully carried out in a controlled laboratory setting. After digestion, the resultant specimens were subjected to a thorough metal examination with a Perkin Elmer Atomic Absorption Spectrometer. The accuracy of this equipment in determining the amounts of metals in environmental samples is well known [5, 6]. The reliability and integrity of the data collected are guaranteed by this methodical approach to sample preparation and analysis, which raises the validity of the study's conclusions.

To be safe, precautions were taken while gathering water samples from crab habitats near the estuary during low tide. As soon as the samples were collected, they were immediately put on ice and treated with hydrochloric acid (5ml) to maintain the water quality. The materials were then meticulously kept within a lab refrigerator [6,7]. The crab samples underwent digestion using 5 milliliters of concentrated nitric acid (HNO<sub>3</sub>) each sample, evaluated on a dry weight basis as well as a wet/fresh weight basis. After that, six different stations' worth of water samples were combined, filtered, and kept in storage at 4°C. These samples were designated for further analysis using Flame Atomic Absorption Spectroscopy (FAAS) for heavy metals.

Sediment samples were carefully collected from approved sites identified as S2, S3, S4, and S5, following the guidelines in US EPA 3050 procedure B [7] to achieve accurate metal analysis. Through careful grinding, the sediment experienced a transformation, going from silt to a fine powder. The powdered samples were then heated to enable additional analysis; however, the description that was supplied did not specify the precise heating technique or how long it was heated for. However, it is vital to remember that throughout this critical stage, strict temperature controls were maintained to preserve sample integrity. Samples are usually cooked for a certain amount of time (10 to 30 minutes), utilizing apparatus such a hot plate or digesting block, at temperatures between 95°C and 105°C. After heating, a carefully mixed solution of hydrochloric acid, deionized water, nitric acid (HNO3), and hydrogen peroxide was carefully added to the sediment samples in 1:1 ratio. By breaking down the silt, this solution released bound metals for further examination. After the digesting process was finished, the mixture that was left behind was carefully filtered to remove any insoluble residues. To ensure consistency and accuracy in the filtration results, special attention was paid to keeping the temperature at 60 degrees Celsius throughout the filtering process [15]

#### **Analytic Procedures**

To achieve the highest level of precision in the field of metal analysis, specific and laborious protocols were used for both sediment and green mussel samples. Defrosting was the first step in the green mussel digestion process, which was then followed by a thorough washing and drying stage. The materials then went through an HNO<sub>3</sub>-assisted digestion procedure before being carefully diluted with deionized water [8]. A thorough metal analysis was performed on the resultant specimens utilizing a Pin AAcle 900T that was acquired from the Perkin Elmer Atomic Absorption Spectrometer.

The goal in this research was to better understand the ecological implications of heavy metal bioaccumulation in aquatic animals, namely in mussels and crabs. The approach used entails a thorough analysis of the body of research to comprehend the causes and effects of bioaccumulation. Foundational insights into the process of bioaccumulation are provided by references to Wager and Boman [7], Yap et al. [16], and Yunus and Nakagoshi [17], highlighting the significance of this process in both terrestrial and aquatic habitats. Furthermore, research mentioned in the paragraph emphasizes the harmful consequences of metal exposure, especially copper (Cu), on mussels and crabs, including changes to metabolic pathways, disruptions to vital metabolic processes, and mutagenic or genotoxic effects [1, 9]

Additionally, the technique takes into account variables that affect the rate of bioaccumulation, including the aquatic organisms' capacity for digestion, the concentrations of metals in their aquatic environment, their eating habits, and their exposure pathways [6,13]. The functions of mussels and crabs as top predators in the aquatic food chain were examined, as well as the locations where heavy metals accumulate in their liver and aills. Remarkably, studies have indicated a relationship between these species' sizes and the amounts of heavy metals in their tissues, with larger animals showing higher rates of bioaccumulation [19]. This scientific approach offers a thorough knowledge of heavy metal bioaccumulation in aquatic animals by synthesizing ideas from numerous research. This understanding guides our inquiry into the possible dangers to ecosystem health and the ecological consequences of heavy metal bioaccumulation.

A sediment digestion process that was in line with US EPA 3050 method B was followed to the letter, with a temperature restriction of less than 60 degrees Celsius. This procedure included grinding, heating, and adding chemical reagents, all of which were planned to enable a precise and trustworthy metal analysis [3]. All these thorough protocols from the methodical sample collection to the intricate analytical techniques combined to form the essential basis for assessing the quantities of heavy metals in the research region.

The Statistical Package for the Social Sciences (SPSS) software, version 13.0 for Windows, was used to do the statistical analysis [2]. The data were screened before analysis to make sure that the fundamental assumptions of homogeneity of variance and normality were followed, which are necessary for trustworthy statistical inference. Outliers were recognized and dealt with appropriately since they are highly abnormal numbers that have the potential to dramatically affect the datasets statistical behavior. When there was a non-normal distribution of the data, both the log(mean) and log (mean + 1) transformations were used to stabilize variance before parametric statistical analysis. Analysis of variance (ANOVA) and post hoc multiple comparisons testing using the Student Newman-Keuls method were used to evaluate differences in mean physicochemical parameter measurements and mean heavy metal concentrations across different sample sites. Additionally, separate T-tests were performed to examine any correlations between the amounts of heavy metals in the sediments of crab and mussel microhabitats and the body weight and carapace width of these species. A 95% significance threshold was used for all statistical comparisons [22].

## 3.0 RESULTS AND DISCUSSION

The results of laboratory tests and in-situ analyses along Kampung Pasir Putih, Pasir Gudang, are interpreted and explained in this chapter. The main objective is to describe how land use affects water quality and how that affects the build-up of metals in green mussels (*Perna viridis*) and crabs (*Pelagicus portunus*). This discourse delves into the levels of heavy metals present in green mussels, crabs, and water, as well as the relationships among these species throughout the Johor Straits.

#### In situ Analysis

Table 2 presents the average in-situ physicochemical characteristics of the water samples that are superimposed, with an emphasis on the habitats of crabs and mussels at various sampling locations. Temperature, pH, dissolved oxygen (DO), salinity, and total dissolved solids (TDS) are measured during low tides, which is an important time for the social and feeding behaviors of mussels and crabs.

The study's conclusions clarify significant differences in important water quality metrics among the various sample locations in the aquatic environment under investigation. The average temperature recorded was 30.32°C, with a range of 27.70 to 32.4°C, suggesting somewhat warm surrounding air. The research area's pH values demonstrated a wide range, ranging from 6.7 to 8.54, indicating the presence of both acidic and alkaline conditions. The quantities of dissolved oxygen (DO) varied, ranging from 1.36 to 6.71 mg/L, indicating variations in the availability of oxygen that are essential for the survival of aquatic life. Variations in the amounts of dissolved salt were reflected in the salinity levels, which varied from 19.43 to 31.89 ppt. In addition, fluctuations in the quantities of dissolved substances were highlighted by the shifting patterns displayed by total dissolved solids (TDS). The observed differences between sample sites highlighted how diverse the aquatic environment was, which was probably impacted by a wide range of elements such natural processes, human activity, as and hydrological dynamics. Numerous reasons might be responsible for the observed variances, including tidal oscillations, sediment composition, urban pollution, agricultural runoff, and industrial effluents. An understanding of the overall ecological health of the aquatic environment under investigation s provided by the thorough examination of these water quality metrics, which is an essential first step towards developing well-informed conservation and management plans. To guarantee the studied ecosystem's continued vitality and resilience in the face of changing environmental stresses, ongoing monitoring and evaluation are essential [17].

# The Concentration of Heavy Metals in the Overlying Water

The mean amounts of Zn, Cu, Cd, and Pb in overlying water samples from crab, mussels, and sediment at six distinct locations were examined in Table 3. For instance, there were differences in dissolved zinc concentrations between sites, ranging from 0.01 to 0.14 mg/L. The overall average concentrations of Zn, Cu, Cd, and Pb were determined to be 0.02 mg/L, 0.06mg/L, 0.15 mg/L, and 0.24 mg/L, respectively. In a similar vein, the concentrations of Cd, Pb, and Cu show fluctuation. The study's conclusions showed that different heavy metal concentrations varied widely among the sites that were investigated. The average content of zinc (Zn) was 0.02 mg/L, with a range of 0.01 to 0.14 mg/L. The amounts of copper (Cu) ranged from 0.01 to 0.14 mg/L, with an average of 0.06 mg/L overall. The concentration of cadmium (Cd) varied, ranging from 0.01 to 0.14 mg/L, with an average of 0.15 mg/L. The average content of lead (Pb) was 0.24 mg/L, with a wide range of values from 0.01 to 0.14 mg/L.

Moreover, variations in the amounts of Cd, Pb, and Cu were observed among the sample locations, suggesting that there is geographical heterogeneity in the contamination of heavy metals. These results highlight the intricate dynamics of heavy metal distribution in aquatic environments, which are probably impacted by a variety of elements including natural processes, urban runoff, and industrial activity. The thorough examination of heavy metal concentrations highlights the significance of ongoing monitoring and management initiatives to protect water quality and ecosystem health and offers vital insights into possible environmental hazards.

#### **Heavy Metal Concentration in Sediments**

The mean Zn, Cu, Cd, and Pb concentrations in soil samples from four different locations are shown in Table 4. Zn, Cu, Cd, and Pb were found to have average amounts of 86.12, 88.25, 3.9, and 12.47 mg/g in sediments, respectively. It is important to remember that the sediment measurements were not subjected to any statistical analysis. Across all sampling stations, the mean amounts of heavy metals in the sediment were determined to be Cu > Zn > Pb > Cd.

#### Heavy Metal Concentrations in Crab and Mussels

The average levels of heavy metals (Zn, Cu, Cd, and Pb) in crab and mussels that were collected from different sites are shown in Table 6. The average Zn,

Cu, Cd, and Pb concentrations in crab varied from 136.01 to 151.24, 35.12 to 39.81, and 2.92 to 3.21 mg/g dry weight, in that order. According to Table 6, the average concentrations of Zn, Cu, Cd, and Pb in mussels ranged from 19.21 to 23.3, 11.27 to 11.75, 0.24 to 0.27, and 4.41 to 4.67 mg/g dry weight. Inconsistent patterns for Zn and Cu were shown by the order of mean heavy metal concentrations in crab across sites, with Cu>Zn or Zn>CU>Pb>Cd (Table 6). It's crucial to remember that statistical analysis was not used to determine these metal ordering. When Zn, Cu, Cd,

and Pb contents in crab and mussels were compared across several locations, it was found that the mean concentrations of mussels were often greater. But when compared to crab, where concentrations were significantly lower (p<0.05), only Cu, Cd, and Pb showed statistically significant higher values (p<0.05) in mussels (Table 5a,5b). This study highlights the importance of comprehending the varied metal uptake patterns in mussels and crabs and throws light on the different ways in which heavy metals accumulate in these two species.

Table 2 In-situ physicochemical parameter measurement in the overlaying water samples (mean ± standard error) tak	en from crab
and mussels habitat surroundings of the different sampling stations. (N=13)	

Station	N	Temperature(°C)	рН	DO (mg/L)	Salinity (ppt)	Conductivity (\$/m)
1	18	30.26±0.21 (28.9-32.4)	7.78±0.06 (7.35-8.35)	3.31±0.15 (2.34-4.59)	29.43±0.33 (26.44-31.89)	509.43±9.07 (455.6-629.53)
2	18	30.33±0.126 (29.1-31.2)	7.94±0.05 (7.58-8.24)	4.38±0.26 (2.73-6.71)	29.59±0.33 (26.75-31.82)	511.99±8.79 (457.36-628.6)
3	18	30.36±0.144 (29.1-31.2)	7.94±0.06 (7.27-8.49)	3.99±0.27 (2.25-5.93)	29.22±0.34 (26.45-31.57)	507.85±8.64 (452.93-610.87)
4	18	30.34±0.227 (28.9-32.10)	7.94±0.06 (7.65-8.54)	3.66±0.29 (1.97-6.39)	28.52±0.72 (19.43-31.48)	483.38±14.56 (341.45-543.53)
5	18	30.22±0.222 (27.70-31.40)	7.85±0.08 (6.97-8.36)	3.06±0.22 (1.56-5.5)	28.66±0.44 (25.1-31.16)	478.04±18.63 (184.47-529.35)
6	18	30.42±0.158 (29.10-31.30)	7.86±0.11 (6.7-8.54)	2.76±0.33 (1.36-5.7)	28.68±0.57 (21.3-31.08)	487.04±11.5 (369.56-529.68)
Overall Average	9	30.32±0.18	7.88±0.07	3.52±0.25	29.01±0.45	496.28±11.86

**Table 3** Water physicochemical parameter measurement in the overlaying water samples (mean ± standard error) taken from crab and mussels habitat surroundings of the different sampling stations. (N=18)

Station	Ν	Cadmium (mg/L)	Zinc (mg/L)	Copper (mg/L)	Lead (mg/L)
1	18	0.15±0.01 (0.05-0.22)	0.07±0.007(0.01-0.14)	0.07±0.005(0.01-0.08)	0.26±0.03(0.16-0.31)
2	18	0.12±0.01(0.05-0.19)	0.06±0.006(0.01-0.1)	0.07±0.004(0.02-0.08)	0.26±0.04(0.15-0.32)
3	18	0.12±0.01 (0.05-0.18)	0.06±0.006(0.01-0.08)	0.06±0.004(0.01-0.08)	0.26±0.04(0.15-0.32)
4	18	0.3±0.01 (0.04-0.22)	0.06±0.007(0.01-0.1)	0.06±0.004(0.02-0.08)	0.26±0.04(0.14-0.33)
5	18	0.10±0.01 (0.05-0.15)	0.11±0.03(0.01-0.6)	0.06±0.004(0.01-0.08)	0.20±0.06(0.08-0.33)
6	18	0.11±0.01(0.04-0.17)	0.07±0.008(0.01-0.12)	0.06±0.004(0.02-0.07)	0.24±0.04(0.13-0.33)
Overall average		0.15±0.01	0.02±0.01	0.06±0.04	0.24±0.04

 Table 4
 Sediment Physiochemical parameter measurement in the overlaying water samples (mean± standard error) taken from crab and mussels habitat surrounding of the different sampling stations. (N=18)

Station	Ν	Copper (mg/g)	Zinc (mg/g)	Cadmium (mg/g)	Pb (mg/g)
2	18	26.62±1.32(17.7-33.1)	26.08±1.23(17.7-32.6)	3.50±0.14(2.5-4.2)	12.44±0.17(11.5-14.3)
3	18	62.56±1.77(56.1-77.7)	63.37±1.92(56.1-77.7)	4.08±0.12(3.4-4.9)	12.39 ±0.33(9.8-14.5)
4	18	137.76±5.30(89.3-155.2)	129.43±4.96(89.3-147.1)	4.01±0.24(2.4-5.3)	12.58±0.28(9.8-14.3)
5	18	126.07±2.24(112-145.4)	125.63±2.21(112-145.4	4.03±0.14(2.4-5.3)	12.5±0.22(11.2-14.1)
Overall average		88.25 ± 2.65	86.12 ± 2.58	3.9 ± 0.16	12.47 ± 0.25

Table 5a Crab physicochemical parameter measurement in the overlaying water samples (mean ± standard error) taken from crab and mussels habitat surroundings of the different sampling stations. (N=13)

Station	Ν	Copper (mg/g)	Zinc (mg/g)	Cadmium (mg/g)	Pb (mg/g)
1	18	31.89±24.41(60.4-148)	97.77±5.75(60.4-148)	2.19±0.09(1.6-2.9)	10.14±0.29(8.2-12.5)
2	18	37.30±36.84(60.4-172)	140.31±18.68(60.4-172)	2.38±0.08(2-3.4)	11.9 ±0.37(8.7-13.7)
3	18	39.36±58.73(71.1-256.5)	151.0±13.84(71.1-256.5)	3.08±0.18(2-4.5)	15.43±0.95(12-23.7)
Overall average		36.18±39.99	129.69±12.75	2.55±0.11	12.49±0.53

**Table 5b** Mussels physicochemical parameter measurement in the overlaying water samples (mean ± standard error) taken from crab and mussels habitat surroundings of the different sampling stations. (N=13)

Station	Ν	Copper (mg/g)	Zinc (mg/g)	Cadmium (mg/g)	Lead (mg/g)
2	18	12.12±0.52(8.8-16.6)	21.97±1.31 (10.40-30.30)	0.26±0.01(0.19-0.34)	5.50±0.097(4.71-6.20)
3	18	12.21±0.46 (8.7-15.4)	27.25±1.75(14.5-38.7)	23±0.007(0.17-0.28)	5.10±0.13(4.32-6.11)
Overall		20 21+0 09	7 01+0 02	2 2140 15	28.05+0.25
average		30.2110.08	7.71±0.03	5.2110.15	28.75±0.25

 Table 6
 Level of heavy metals concentration in the sediment, crab species (mean ± standard error) collected from the different sampling stations and their heavy metals occurrence pattern. The heavy metals occurrence is not based on statistical analysis

Station	Sample	Cu (mg/g)	Zn (mg/g)	Cd (mg/g)	Pb (mg/g)	Metal occurrence pattern
2	Sediment	26.62±1.32	26.08±1.23	$3.50\pm0.14$	12.44±0.17	Cu>Zn>Pb>Cd
	Crab	31.89±24.41 (60.4-148)	97.77±5.75 (60.4-148)	2.19±00.09 (1.6-2.9)	(11.3 14.3) 10.14±0.29 (8.2-12.5)	Zn>Cu>Pb>Cd
	Mussels	12.12±0.52 (8.8-16.6)	21.97±1.31 (10.40-30.30)	0.26±0.01 (0.19-0.34)	(4.71-6.20)	Zn>Cu>Pb>Cd
3	Sediment	62.56±1.77 (56.1-77.7)	63.37±1.92 (56.1-77.7)	4.08±0.12 (3.4-4.9)	12.39±0.33 (9.8-14.5)	In>Cu>Pb>Cd
	Crab	37.30±36.84 (60.4-172)	140.31±18.68 (60.4-172)	2.38±0.08 (2-3.4)	11.9 ±0.37 (8.7-13.7)	Zn>Cu>Pb>Cd
	Mussels	12.21±0.46 (8.7-15.4)	27.25±1.75 (14.5-38.7)	0.23±0.007 (0.17-0.28)	(4.32-6.11)	Zn>Cu>Pb>Cd
4	Sediment	137.76±5.30 (89.3-155.2)	129.43±4.96 (89.3-147.1)	4.01±0.24 (2.4-5.3)	12.58±0.28 (9.8-14.3)	Zn>Cu>Pb>Cd
	Crab	39.36±58.73 (71.1-256.5)	151.0±13.84 (71.1-256.5)	3.08±0.18 (2-4.5)	15.43±0.95 (12-23.7)	Zn>Cu>Pb>Cd
	Mussels	-	-	-	-	-
5	Sediment	126.07±2.24 (112-145.4)	125.63±2.21 (112-145.4	4.03±0.14 (2.4-5.3)	12.5±0.22 (11.2-14.1)	Cu>Zn>Pb>Cd
	Crab Mussels	-	-	-	-	-

Table 7 Guidelines of maximum permissible limits of heavy metals ( $\mu g/g$ ) in seafood from different countries (Yap et al., 2004a)

Guideline	Cd	Cu	Pb	Zn	Ni	Fe
Malaysian Food Regulations (1985)	1.0	30.0	2.00	100.00	-	-

#### Heavy Metal Concentration in Water, Crab, and Green Mussels Samples

The levels of lead (Pb), zinc (Zn), copper (Cu), and cadmium (Cd) in water samples, green mussels (Perna viridis), and crabs (Pelagicus portunus) are thoroughly

discussed in this section. The water's Cd concentration was found to be between 0.05 and 0.22 mg/L, which is higher than the Malaysia Marine Water Quality Criteria and Standard (MMWQS) allowable limit of 10  $\mu$ g/L. This puts the water in Class 3. Nonetheless, the levels of Cd in green mussels and crab were below than the 1  $\mu$ g/g allowable limit as stated in the Malaysian Food Regulation (1985). Analogous conversations concerning lead, copper, and zinc concentrations are also included, offering perspectives on the possible consequences for the environment and human health [9] exposures are countered by homeostatic mechanisms, which results in a persistent disturbance of metabolic processes. Both green mussels and crab have cadmium amounts below the permitted limits specified by the Malaysian Food Regulation (1985), which is  $1 \mu g/g$ .

# Concentrations of Cadmium in Water, Mussels, and Crab

The water near Kampung Pasir Putih had concentrations of cadmium ranging from 0.05 to 0.22 mg/L. Higher levels were probably caused by surface runoff caused by rain, which carried cadmium from the ground into the sea [9,10]. It has been determined that one of the main causes of the cadmium buildup in green mussels is untreated industrial waste. If the cadmium concentration is more than 10  $\mu$ g/L, it is categorized as Class 3 by the Malaysia Marine Water Quality Criteria and Standard (MMWQS).

Figure 2 shows the varying quantities of cadmium in green mussels and crabs. Concentrations of green mussels ranged from 0.17 to 0.34 mg/g, with Station S2 showing the highest amount. The greatest amounts of crab, ranging from 1.6 to 4.5 mg/g, were recorded at Station S3. Green mussels may contain higher amounts of cadmium due to untreated trash from industry. The observed closure of valves in response to exposure to harmful metals stopped further accumulation in the soft tissue of green mussels, even at low concentrations [1,4,7].



Figure 2 Concentration of cadmium in water, green mussels and crab

#### **Zinc Concentration Analysis**

At Kampung Pasir Putih, the zinc content of the water ranges from 0.01 to 0.12 mg/L, as shown in Figure 3 The range of zinc contents in crab and mussels, respectively, is 60.4 to 256.5 mg/g and 10.40 to 38.7 mg/g. Zn levels are classified as Class 3 below 100 µg/L by the Malaysia Marine Water Quality Criteria and Standard [11]. The U.S. EPA suggests that marine and estuarine waters have 0.09 mg/L of zinc.

The zinc concentrations in green mussels and crab are shown in Figure 3, which follows the 100 µg/g allowable level established by the Malaysian Food Regulation (1985). In concentrations in water, crab, and mussels are, respectively, 0.02, 129.69, and 7.91 mg/g on average. In concentrations in crab are higher than those in water quality, which is a significant divergence from other heavy metals. In's critical function in *Perna viridis* metabolism and its regulation may be the cause of the significant variation seen in the amounts of crab, green mussels, and water samples [10,12].

Zinc is an essential heavy metal for marine life that frequently outperforms other metal concentrations in enhancing enzymatic function. Temperature and salinity affect *Perna viridis*'s ability to absorb zinc. Zinc is mostly absorbed by aquatic animals from the water; zinc that has been dissolved is bioavailable. Bioavailability is dependent on biological processes and environmental factors [13].



Figure 3 Concentration of zinc in water, green mussels and crab

#### Lead Concentration Analysis

The amount of lead present in the water of Kampung Pasir Putih is shown in Figure 4. Pb was found in water, crab, and mussels at mean amounts of 0.24, 12.49, and 28.95 mg/g, respectively. The greatest concentration was found in mussels. Pb levels surpassing 8.5 µg/L placed it in Class 3 of the Malaysia Marine Water Quality Criteria and Standard. The health of humans and aquatic life may be adversely affected by elevated Pb levels [3,13]. The highest Pb amounts were found in 8.2 to 23.7 mg/g of crab and 4.32 to 6.20 mg/g of mussels, respectively. Given the popularity of green mussels and crab as seafood in Malaysia, these amounts exceed the allowed level stipulated by the Malaysian Food Regulation (1985), which is 2.00  $\mu$ g/g, creating health hazards. There were no discernible patterns in the Pb concentrations in mussels, crab, and water quality, despite the fact that the amounts in mussels and crab were significantly greater than those in water.



Figure 4 Concentration of Pb in water, green mussels and crab

#### **Copper Concentration**

The amount of copper present in the water of Kampung Pasir Putih is shown in Figure 5. Cu can be found at concentrations ranging from 0.01 to 0.08 mg/L. Cu values in green mussels range from 8.7 to 16.6 mg/g and 21.8 to 50.3 mg/g, respectively, in crab. Cu is categorized under Class 3 in the Malaysia Marine Water Quality Criteria and Standard, with a threshold of less than 100  $\mu$ g/L [19]. The U.S. EPA has established a recommended limit of 30.0 mg/g for Cu in marine and estuarine waters [20,21]. The amounts of Cu in crab and green mussels are shown in Figure 5. The Malaysian Food Regulation (1985) sets an acceptable level of 30 mg/g for Cu. As a result, the measured concentrations stay within the permissible range and don't go over the 30 mg/g limit.



Figure 5 Concentration of Cu in water, green mussels and crab

The investigation of heavy metal concentrations in the water, green mussels, and crabs in Kampung Pasir Putih offered important new information about the processes governing metal accumulation in the environment. Water samples had cadmium (Cd) values ranging from 0.05 to 0.22 mg/L. This is because surface runoff carries Cd from the land into the sea, possibly via untreated industrial waste. Cd concentrations in green mussels ranged from 0.17 to 0.34 mg/g, but those in crabs were greater, ranging from 1.6 to 4.5 mg/g. This discrepancy implies that crabs could collect more Cd than green mussels, presumably as a result of different habitat choices and feeding schedules. The amounts of zinc (Zn) in water were found to be very low, ranging from 0.01 to 0.12 mg/L. However, larger levels of zinc were found in green mussels (10.40 to 38.7 mg/g) and crabs (60.4 to 256.5 mg/g). Because of their metabolic and regulatory systems, which may promote greater absorption of this vital heavy metal, crabs may have higher Zn concentrations [14]. Lead (Pb) levels were higher than allowed in water, crabs, and green mussels, with mussels having the highest levels (4.32 to 6.20 mg/g). Pb accumulates significantly in mussels and crabs as opposed to water, which implies bioaccumulation and draws attention to possible health hazards connected to seafood consumption. Water samples had copper (Cu) contents ranging from 0.01 to 0.08 mg/L, but crabs and green mussels had greater quantities (21.8 to 50.3 mg/g and 8.7 to 16.6 mg/g, respectively) [18]. The buildup of Cu in crabs and mussels, even at levels within allowable limits, emphasizes their significance as bioindicators of metal contamination. All things considered, our results highlight the necessity of ongoing management and monitoring programs to reduce heavy metal pollution in coastal ecosystems and protect public health.

# 4.0 CONCLUSION

In the Straits of Johore in Kampung Putih, Peninsular Malaysia, the current study carried out a thorough analysis of Insite physicochemical parameters, heavy metal concentrations in water samples, and microhabitat sediments of crabs and mussels. The results showed that there was no irregularity in the physicochemical parameters in the overlaying water samples near crab habitats, suggesting a stable environment at the time of the measurements. However, differences in characteristics between sampling stations indicated that distinct factors affecting water quality were caused by hydrologic processes, tidal impacts, and runoff from nearby sources. A baseline for comparison was established by the collected values' overall agreement with past data from the same and neighboring places.

Zn, Cu, Cd, and Pb heavy metal concentrations in the overlapping water samples ranged from 0.05 to 1.39 mg/L. All metal concentrations met Malaysian Department of Environment regulations, with the exception of Cd. Zn, Cd, and Pb levels did not present acute toxicity to the studied crab and mussel species, according to comparison with the US EPA's Criterion Maximum Concentration criteria. Cu concentrations, however, greatly exceeded suggested levels, raising the possibility of acute toxicity to mussels and crabs.

Sediment study revealed that the concentrations of Zn, Cu, Cd, and Pb in the sediments of crab microhabitats were either lower than or equal to those of the non-contaminated sediments, suggesting a low level of contamination. There were no discernible biological impacts, according to comparisons with the impacts Range-Low (ERL) and Effects Range-High

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(ERM) standards. Due to elements including detrital content and sediment composition, sediments from crab microhabitats showed greater metal concentrations than those from mussel microhabitats.

The accumulation pattern of (Cu > Zn) or (Zn > Cu)> Pb > Cd was seen in comparisons between crab and mussel species; Zn and Cu were consistently greater, most likely because of their crucial functions in the growth and metabolism of crustaceans. In crabs, the reduced quantities of non-essential Cd and Pb may be explained by detoxification and excretion activities. Both species of mussels and crabs showed biomonitoring promise as animals, satisfying requirements including simple identification, yearround availability, and restricted migration. However, the study indicates that because of their greater rate of heavy metal uptake from sediments, crabs, and mussels in particular, crabs are more efficient biomonitoring agents. Potential biases in sample techniques include the investigation's constrained temporal scope, and the need for more research to completely comprehend the long-term effects of heavy metal poisoning on the ecosystems under study are some of the study's weaknesses or shortcomings. Subsequent studies must concentrate on continuous observation initiatives aimed at defining safety criteria for heavy metals in sediment and tracking metal contamination levels in Malaysia's coastal areas. It is advised to conduct ongoing monitoring to determine safety standards for sediment heavy metal concentrations and to monitor the levels of metal pollution in Malaysia's coastal regions.

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#### **Conflicts of Interest**

The author(s) declare(s) that there is no conflict of interest regarding the publication of this paper.

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