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Original Article

Assessment of biomonitoring potential of contamination and bioavailability of heavy metals using red blood cockle *Tegillarca granosa*: Experimental field-based transplantation study

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Abstract
This study evaluates the potential of red blood cockle *Tegillarca granosa* as a biomonitor for heavy metal contamination in coastal environments, with a focus on the differential bioaccumulation patterns across three sites: Teluk Kemang (TK), Pasir Panjang (PP), and Port Dickson (PD). Four major findings emerged from the analysis. First, *T. granosa* demonstrated significant variations in metal accumulation, with PD exhibiting the highest concentrations of Zinc (Zn), Copper (Cu), and Iron (Fe). This confirms the species' sensitivity to varying pollution levels and highlights PD as the most contaminated site among the three. Second, strong correlations between metal levels in the environment and their accumulation in the cockle's soft tissues underscored its effectiveness in reflecting the bioavailability of contaminants, particularly for Zn and Cu. This finding validates the use of *T. granosa* as an effective bioindicator of environmental metal exposure. Third, the study highlighted the role of cockle shells as indicators of long-term exposure, with consistently higher metal concentrations observed at PD. This suggests persistent environmental contamination in this area and demonstrates the value of shell analysis in assessing chronic pollution. Finally, the stepwise multiple regression analysis revealed complex interactions between environmental media and bioaccumulation processes, emphasizing the need for a comprehensive approach to biomonitoring. These interactions indicate that bioaccumulation in *T. granosa* is influenced by multiple environmental factors, necessitating an integrated monitoring strategy. These findings support the use of *T. granosa* as a reliable indicator species for monitoring heavy metal pollution and assessing ecological risks in coastal ecosystems.

Keywords: bioaccumulation; biomonitoring; coastal ecosystems; cockles; heavy metal contamination

1 | INTRODUCTION

Coastal regions are increasingly vulnerable to contamination from industrial discharges, urban runoff, and agricultural activities, which introduce significant quantities of heavy metals such as zinc (Zn), copper (Cu), cadmium (Cd), iron (Fe), and nickel (Ni) into the marine environment (Naser 2013). As heavy metals accumulate, they seriously threaten marine ecosystems, leading to detrimental effects on biodiversity and public health, particularly in regions with high anthropogenic activity (Bazzi 2014; Carolin *et al.* 2017; Aziz *et al.* 2023). The presence of heavy metals in coastal waters not only disrupts ecological balance but also poses severe health risks to populations reliant on marine resources, prompting the need for on-going monitoring and assessment of pollution levels to ensure sustainable management practices and the safety of seafood consumption (Naser 2013; Díaz-de-Alba *et al.* 2021; Wu *et al.* 2022; Dehbi *et al*. 2023).

Biomonitoring, which relies on using living organisms as indicators of environmental contamination, has emerged as a valuable tool for evaluating the extent and impact of heavy metal pollution in coastal ecosystems (Pérez-López *et al.* 2003; Chiarelli and Roccheri 2014; Corrias *et al.* 2020; Roveta *et al.* 2021). The variability in heavy metal accumulation among individual organisms can greatly influence environmental health assessments, underscoring the importance of selecting suitable bioindicator species for accurate monitoring efforts (Saavedra *et al.* 2009). In this context, incorporating bivalves like red blood *Tegillarca granosa* (Linnaeus, 1758) as bioindicators allows for more nuanced assessments of metal accumulation. It helps mitigate the risks posed by heavy metal contamination, as their physiological traits enable them to reflect the levels of pollutants in their surrounding environment (Saavedra *et al.* 2009; Roveta *et al.* 2021).

The *T. granosa* is a widely distributed bivalve species in Southeast Asian coastal waters, playing a crucial ecological and economic role in the region (Chee *et al.* 2011; Harith *et al.* 2016; Ismui *et al.* 2020; Saffian *et al.* 2020; Dinulislam *et al.* 2021; Mohamat-Yusuff *et al.* 2021). The species' ability to accumulate high concentrations of heavy metals in its tissues makes it an ideal candidate for biomonitoring, as studies have demonstrated a strong correlation between metal concentrations in *T. granosa* and the levels found in surrounding sediments (Bazzi 2014). Moreover, the dual accumulation patterns observed in the species' soft tissues and shells provide insights into both short-term and long-term exposure to heavy metals, enhancing the utility of *T. granosa* as a comprehensive biomonitor (Velez *et al.* 2016; Dinulislam *et al.* 2021).

This study aimed to evaluate the potential of *T. granosa* as a biomonitor for heavy metal contamination in coastal environments, focusing on the differential bioaccumulation patterns across three sites: Teluk Kemang (TK), Pasir Panjang (PP), and Port Dickson (PD). This study is structured first to assess the differential accumulation of heavy metals in the soft tissues and shells of *T. granosa* across the three study sites. Subsequently, the correlations between environmental metal levels and bioaccumulation in the cockles will be examined to determine the bioavailability of these metals. Finally, the study will explore the complex interactions between environmental media and bioaccumulation processes through stepwise multiple regression analysis. The findings will highlight the role of *T. granosa* in biomonitoring and the implications of metal contamination for coastal ecosystems, providing a foundation for future research and environmental management strategies.

2 | METHODOLOGY

2.1 Acclimatization and transplantation of cockle

Two preliminary studies were conducted prior to this comprehensive study. Initially, *T. granosa* was purchased from the wet market in PD, Negeri Sembilan, Malaysia. According to the supplier, the cockles were sourced from Teluk Intan, Perak– an unpolluted site (Table 1). All cockle individuals were transported to PP for a 3-day acclimatization period. PP was chosen as the acclimatization site due to its extensive tidal mudflat, which is ideal for cockle culture (Noorddin 1995). The cockles were placed in basins half-filled with sediment from the surrounding substrate. The animals were checked daily and were considered dead if they showed no response to tactile stimuli and their shell valves remained gaped.

Following acclimatization, approximately 60 'healthy-looking' cockle individuals of similar size were selected and placed into each of the three cages prepared for the exposure study. Each cage was then transplanted to a contaminated site at PD and two uncontaminated sites at PP and TK, chosen based on the anthropogenic activities observed there (Figure 1). Descriptions of the sites are provided in Table 1. Prior to the commencement of the research, 20 cockle individuals were randomly sampled to analyze the baseline levels of heavy metals in their tissues.

TABLE 1 Site descriptions of the transplanted sites for blood cockle *Tegillarca granosa*.

Study sites	Coordinates	Descriptions
Teluk Kemang TK)	2°27′N 101°51′E	Recreational area
Pasir Panjang (PP)	2°25′N 101°56′E	Fishing village with mudflat
Terminal Feri, Port Dickson (PD)	2°31′N; 101°48′E	Jetty and near Port Dickson town.

During the exposure study, cockle individuals were randomly sampled at 4-day intervals over a 16-day period, from 4 December to 20 December 2005. On each sampling day, water and surface sediments (3 – 5 cm) were collected from each site. All samples were returned to the laboratory and stored at –10°C until metal analysis. Before dissection, the cockle samples were thawed at room temperature (27°C) on clean tissue paper to drain excess water. Subsequently, the soft tissues and shells of the cockles, along with sediment samples, were dried for 72 hours at 105°C in an oven to obtain constant dry weights. The dried samples were then sieved through a 0.5-mm stainless steel sieve (63 µm) and shaken vigorously to ensure homogeneity (Yap *et al.* 2002).

2.2 Analysis of heavy metals

Approximately 0.5 g of each dried cockle sample (separated into soft tissues and shells) was weighed and digested in concentrated HNO_3 (AnalaR grade, BDH 69%). The dried sample was weighed and digested for sediment samples using a combination of concentrated nitric acid (AnalaR grade, BDH 69%) and perchloric acid (AnalaR grade, BDH 60%) in a 4:1 ratio. The digestion process was conducted in a hot-block digester at a low temperature (40°C) for 1 hour, followed by complete digestion at a high temperature (140°C) for at least 3 hours (Yap *et al.* 2002, 2004). The prepared samples were analyzed for Cd, Cu, Fe, Ni, and Zn using an air-acetylene flame atomic absorption spectrophotometer Perkin Elmer AAnalyst 800 (PerkinElmer, Inc., Waltham, MA, USA). To avoid contamination, all glassware and equipment used were acid-washed (Yap *et al.* 2004). Procedural blanks were analyzed for every five samples to check for contamination. Quality control samples, prepared from standard Cd, Cu, Fe, Ni, and Zn solutions, were analyzed every five samples to assess metal recoveries (Yap *et al.* 2002).

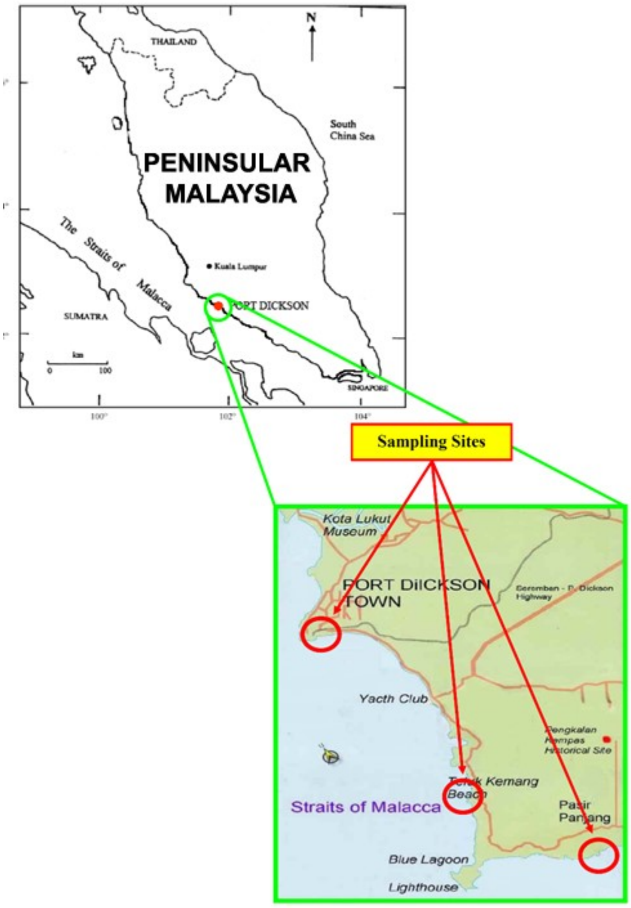

FIGURE 1 Sampling map for Port Dickson Jetty, Teluk Kemang and Pasir Panjang.

The accuracy of the analytical procedure for determining heavy metals in cockles was tested using Standard Reference Materials (SRM) for mussel tissue (SRM 2976, National Institute of Standards and Technology) and dogfish liver (DOLT-3, National Research Council Canada). The recovery ranges for the mussel tissue were 69.5% to 107.2% for all metals except Ni (unavailable), while for the dogfish liver, recoveries ranged from 78.8% to 103.5% for the five metals.

2.3 Data analysis

Statistical analyses, including overall statistics, correlation analysis, and stepwise multiple linear regression analysis of the metal levels in the soft tissues of cockles with those in the water, sediments, and cockle shells, were conducted using JASP (JASP Team 2024). Mean values for the soft tissues and shells of cockles were converted into graphs using Kaleida Graphs, version 3.08, November 1996.

3 | RESULTS

The heavy metal accumulation in *T. granosa*, focuses on the temporal trends observed at the three distinct sites (Figures 2 and 3). These sites vary significantly in their levels of environmental contamination, with PD being known for its higher pollution levels due to jetty and industrial activities. This difference is vividly captured in the data (Figures 2 and 3), which track the concentrations of several heavy metals in both the soft tissues and shells of *T. granosa* over a 16-day period from December 4 to 20 December 2005.

3.1 Heavy metal concentrations in cockles' soft tissue

At the beginning of the observation period (Day 0), the concentration of Zn in the soft tissues at PD was approximately 120 µg g^{-1} (Figure 2). This starting value was already higher than at TK and PP, where Zn concentrations begin at about 100 µg g^{-1} and 105 µg g^{-1}, respectively. Over the 16 days, Zn levels at PD increase markedly, reaching 150 µg g^{-1} by Day 16. This sharp rise indicates the high level of Zn bioavailability in the more polluted environment of PD. In contrast, the increase in Zn concentration at TK and PP was more gradual, with final values of around 115 µg g^{-1} and 110 µg g^{-1}, respectively, reflecting the lower pollution levels at these sites.

Similarly, Cu exhibits a significant accumulation pattern at PD. On Day 0, Cu concentrations in the soft tissues were around 50 µg g^{-1} at PD, compared to 40 µg g^{-1} at TK and 45 µg g^{-1} at PP (Figure 2). By Day 16, Cu levels at PD have risen to 75 µg g^{-1}, demonstrating a robust bioaccumulation in response to the higher environmental contamination. The corresponding increases at TK and PP were more modest, with Cu concentrations reaching 55 µg g^{-1} and 50 µg g^{-1}, respectively.

Cd, a metal of particular concern due to its toxicity, also shows elevated levels at PD. Starting at around 2 µg g^{-1} on Day 0, Cd concentrations at PD increase to approximately 3.5 µg g^{-1} by Day 16 (Figure 2). This rise was significantly steeper than the increases observed at TK and PP, where Cd levels start at 1.5 µg g^{-1} and 1.6 µg g^{-1} and end at around 2 µg g^{-1} and 2.1 µg g^{-1}, respectively. This pattern highlights the heightened risk of Cd exposure in more contaminated areas.

Fe follows a similar trend, with initial concentrations at PD around 500 μg g^{-1}, compared to 450 μg g^{-1} at TK and 460 μg g^{-1} at PP (Figure 2). By the end of the observation period, Fe levels at PD had increased to 650 μg g^{-1}, while at TK and PP, the increases were more moderate, reaching 500 μg g^{-1} and 510 μg g^{-1}, respectively. This substantial accumulation at PD underscores the greater exposure to Fe pollution at this site, likely due to industrial discharges.

Ni also shows a clear pattern of increased accumulation at PD. Starting at approximately 10 μg g^{-1} on Day 0, Ni concentrations at PD rise to about 15 μg g^{-1} by Day 16 (Figure 2). In contrast, TK and PP begin with lower concentrations of 8 μg g^{-1} and 8.5 μg g^{-1}, respectively, and increase to only 9.5 μg g^{-1} and 10 μg g^{-1}. This pattern further emphasizes the higher environmental burden of Ni at PD.

3.2 Heavy metal concentrations in cockles' shells

The heavy metal concentrations in the shells of *T. granosa* provide insights into the longer-term accumulation of metals in these more inert, structural components (Figure 3). The patterns observed in the shells were consistent with those in the soft tissues, though the rates of accumulation may differ slightly due to the slower incorporation of metals into the shell material.

For Zn, the initial concentration in shells at PD was around 30 μg g^{-1} on Day 0, which was already higher than the starting levels at TK (20 μg g^{-1}) and PP (25 μg g^{-1}) (Figure 3). By Day 16, Zn concentrations in PD shells increase to approximately 45 μg g^{-1}, reflecting significant metal deposition over time. In comparison, the Zn concentrations at TK and PP rise more slowly, reaching 25 μg g^{-1} and 30 μg g^{-1}, respectively. This difference underscores the cumulative impact of persistent environmental exposure to Zn at the more polluted PD site. Cu follows a similar trajectory in the shells. Starting at 5 μg g^{-1} in PD on Day 0, Cu levels rise to 8 μg g^{-1} by Day 16, again surpassing the increases observed at TK and PP, where Cu concentrations increase from 4 μg g^{-1} to 5 μg g^{-1} and from 4.5 μg g^{-1} to 5.5 μg g^{-1}, respectively (Figure 3). This suggests that the shells of *T. granosa* at PD were subjected to higher Cu exposure, likely due to industrial pollutants.

Cd concentrations in the shells at PD also show a significant rise (Figure 3). Starting at around 0.5 μg g^{-1} on Day 0, Cd levels increase to 1 μg g^{-1} by Day 16, indicating on-going environmental exposure to Cd. At TK and PP, the increases were less pronounced, with Cd levels rising from 0.3 μg g^{-1} to 0.4 μg g^{-1} and from 0.4 μg g^{-1} to 0.6 μg g^{-1}, respectively. This pattern of higher accumulation at PD was consistent with the elevated Cd levels observed in the soft tissues. Fe concentrations in the shells at PD start at 100 μg g^{-1} on Day 0 and rise to 150 μg g^{-1} by Day 16 (Figure 3), further highlighting the site's higher environmental contamination. At TK and PP, Fe concentrations increase from 90 μg g^{-1} to 110 μg g^{-1} and from 95 μg g^{-1} to 120 μg g^{-1}, respectively, again showing lower levels of accumulation compared to PD. Ni concentrations in the shells at PD also reflect a higher level of accumulation. Beginning at 2 μg g^{-1} on Day 0, Ni levels rise to 3 μg g^{-1} by Day 16, while TK and PP see more modest increases from 1 μg g^{-1} to 1.5 μg g^{-1} and from 1.5 μg g^{-1} to 2 μg g^{-1}, respectively (Figure 3).

In summary, the data depicted in Figures 2 and 3 clearly illustrate the significant impact of environmental contamination on the bioaccumulation of heavy metals in *T. granosa* at PD compared to the less polluted sites of TK and PP. The consistently higher metal concentrations observed at PD in both the soft tissues and shells of the cockles emphasize the severe pollution burden at this site, driven by industrial activities. These findings underscore the importance of continuous monitoring and remediation efforts to mitigate the environmental and ecological risks associated with heavy metal pollution in coastal regions like PD.

3.3 Overall statistics of metal levels in the water, sediments, shells and soft tissues of cockles

Among the metals analyzed in the seawater samples, Zn shows the highest concentration at PD, ranging from 0.076 mg L^{-1} to 0.088 mg L^{-1}, with a mean value of 0.083 mg L^{-1} (Table 2). This suggests that PD was more polluted with Zn compared to TK and PP, which have lower mean concentrations of 0.030 mg L^{-1} and 0.047 mgL^{-1}, respectively. Cu levels at PD also stand out, with a mean concentration of 0.104 mg L^{-1}, significantly higher than those at TK (0.041 mg L^{-1}) and PP (0.055 mg L^{-1}). The skewness and kurtosis values for Cu at PD indicate potential variability in pollution sources. Cd levels were similarly elevated at PD, with a mean of 0.089 mg L^{-1}, higher than the means at TK and PP, which were 0.067 mg L^{-1} and 0.066 mg L^{-1}, respectively. The Fe concentration at PD was particularly noteworthy, with a broad range between 0.963 mg L^{-1} and 1.004 mg L^{-1}, and a mean of 0.978 mg L^{-1}, reflecting significant industrial influence. Lastly, Ni concentrations at PD were higher than at the other sites, with a mean of 0.101 mg L^{-1}, further establishing PD as the most contaminated site among the three (Table 2).

Zn concentrations in the sediments at PD range from 109 μg g^{-1} to 111 μg g^{-1}, with a mean of 110 μg g^{-1}, indicating higher Zn deposition compared to TK and PP, where the means were 66.8 μg g^{-1} and 92.7 μg g^{-1}, respectively (Table 3). Cu contamination was markedly severe at PD, with a mean concentration of 62.3 μg g^{-1}, far exceeding the values at TK (4.87 μg g^{-1}) and PP (10.3 μg g^{-1}). Cadmium (Cd) levels in PD sediments were also higher, with a mean of 4.54 μg g^{-1}, compared to 2.82 μg g^{-1} at TK and 1.63 μg g^{-1} at PP. The Fe concentrations at PD were the highest, ranging from 1732 μg g^{-1} to 1883 μg g^{-1}, with a mean of 1807 μg g^{-1}, indicative of significant Fe deposition likely due to industrial activities. Ni concentrations follow this trend, with PD showing the highest mean concentration of 39.5 μg g^{-1}, further emphasizing the site's heavy metal burden (Table 3).

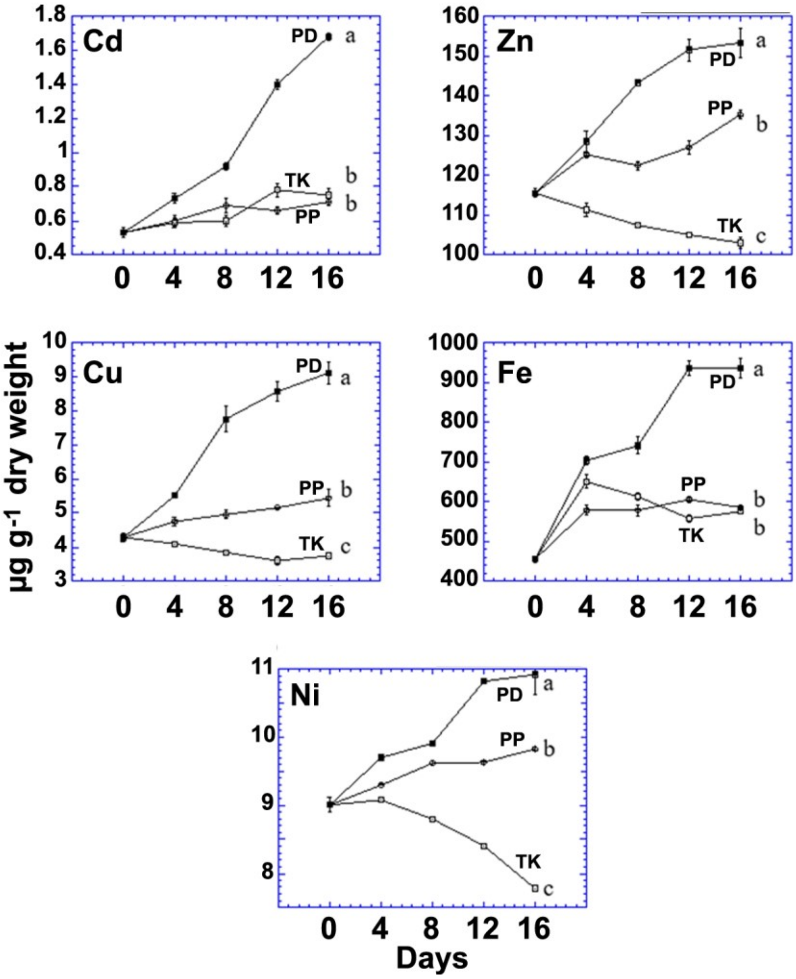

FIGURE 2 Heavy metal concentrations (mean ± SE, μg g^{-1} dry weight) in the total soft tissues of cockle *Tegillarca granosa* for 16 days at three sampling sites (TK, Teluk Kemang; PP, Pasir Panjang; PD, Port Dickson). Note: Y-axis represents metal concentrations in the total soft tissues of cockles; X-axis represent the days of transplantation. Statistical comparison at Day 16 where different alphabets (a – c) showing are significantly different at an α level of significance of 0.05.

TABLE 2 Overall statistics of concentrations (mg L^{-1}) of heavy metals in the surface water of Teluk Kemang (TK), Pasir Panjang (PP) and Port Dickson (PD); *n* = 4.

	Zn TK	Cu TK	Cd TK	Fe TK	Ni TK	Zn PP	Cu PP	Cd PP	Fe PP	Ni PP	Zn PD	Cu PD	Cd PD	Fe PD	Ni PD
Min	0.022	0.033	0.054	0.354	0.088	0.044	0.042	0.059	0.482	0.070	0.076	0.097	0.081	0.963	0.095
Max	0.038	0.046	0.080	0.389	0.099	0.051	0.063	0.078	0.500	0.093	0.088	0.118	0.098	1.004	0.108
Mean	0.030	0.041	0.067	0.372	0.093	0.047	0.055	0.066	0.488	0.083	0.083	0.104	0.089	0.978	0.101
SD	0.007	0.005	0.011	0.019	0.005	0.003	0.009	0.008	0.009	0.010	0.005	0.010	0.008	0.019	0.006
SE	0.003	0.003	0.005	0.010	0.002	0.001	0.005	0.004	0.004	0.005	0.003	0.005	0.004	0.009	0.003
Skewness	-0.121	-0.600	0.000	0.000	0.000	0.298	-0.753	0.913	0.998	-0.445	-0.610	1.000	0.000	0.785	0.373
Kurtosis	-1.127	-0.938	-1.023	-1.985	-1.258	-0.980	-1.000	-0.810	-0.811	-0.955	-1.000	-0.791	-1.684	-1.017	-1.255

TABLE 3 Overall statistics of concentrations (μg g^{-1} dry weight) of heavy metals in the surface sediments of Teluk Kemang (TK), Pasir Panjang (PP) and Port Dickson (PD); *n* = 4.

	Zn TK	Cu TK	Cd TK	Fe TK	Ni TK	Zn PP	Cu PP	Cd PP	Fe PP	Ni PP	Zn PD	Cu PD	Cd PD	Fe PD	Ni PD
Min	65.6	4.70	2.53	1038	22.8	91.5	9.82	1.57	1243	29.9	109	61.1	3.95	1732	37.0
Max	67.4	5.03	2.97	1069	24.9	93.5	10.8	1.68	1363	32.8	111	63.5	5.17	1883	40.8
Mean	66.8	4.87	2.82	1056	24.0	92.7	10.3	1.63	1309	31.6	110	62.3	4.54	1807	39.5
SD	0.81	0.14	0.20	13.1	0.85	0.97	0.42	0.05	49.6	1.24	1.01	1.17	0.51	62.0	1.80
SE	0.40	0.07	0.10	6.53	0.43	0.49	0.21	0.02	24.8	0.62	0.51	0.59	0.25	30.9	0.90
Skewness	-1.08	0.00	-0.91	-0.53	-0.51	-0.39	0.45	0.00	-0.38	-0.63	-0.30	0.06	0.16	0.01	-0.67
Kurtosis	-0.72	-1.26	-0.87	-0.99	-0.96	-1.50	-1.00	-1.03	-1.01	-0.92	-1.60	-1.77	-1.09	-1.04	-1.17

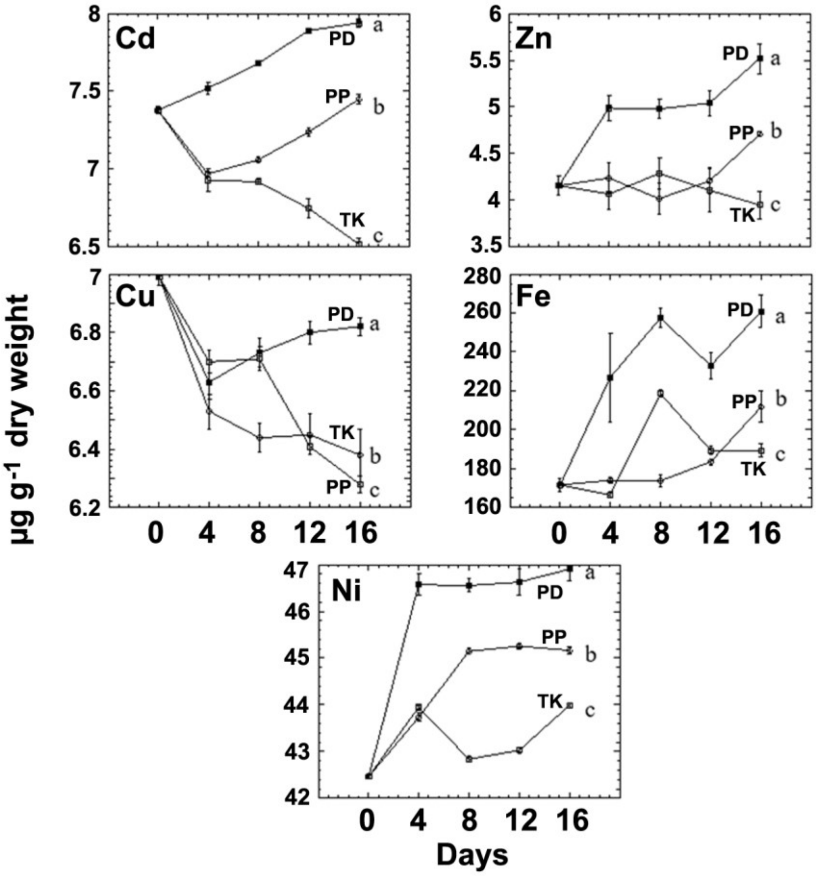

FIGURE 3 Heavy metal concentrations (mean ± SE, µg g^{-1} dry weight) in the shells of *Tegillarca granosa* for 16 days. Note: Y-axis represents metal concentrations in the shells of cockles; Y-axis represents the days of transplantation. Statistical comparison at Day 16 where different alphabets (a – c) showing are significantly different at an α level of significance of 0.05.

The results indicate that Zn accumulates more in the cockle shells at PD, with a mean concentration of 4.94 µg g^{-1}, compared to 4.12 µg g^{-1} at TK and 4.27 µg g^{-1} at PP (Table 4). Cu levels in the shells were also slightly higher at PD, with a mean of 6.79 µg g^{-1}, although the difference was less pronounced than in sediments or water. Cd concentrations in PD shells were higher as well, with a mean of 7.68 µg g^{-1}, reflecting the greater bioavailability of Cd at this site. Fe concentrations in the shells at PD were similarly elevated, with a mean of 181 µg g^{-1}, compared to 170 µg g^{-1} at TK and 172 µg g^{-1} at PP. Ni levels in the shells follow the same pattern, with a mean of 45.8 µg g^{-1} at PD, higher than the means at TK and PP, indicating greater exposure to Ni at PD (Table 4).

Zn concentrations in the soft tissues at PD were significantly higher, with a mean of 138 µg g^{-1}, compared to 108 µg g^{-1} at TK and 125 µg g^{-1} at PP (Table 5), indicating substantial Zn accumulation. Cu levels in the soft tissues at PD were also higher, with a mean of 7.04 µg g^{-1}, compared to 3.91 µg g^{-1} at TK and 4.92 µg g^{-1} at PP, suggesting more significant exposure to Cu. Cd concentrations in the soft tissues at PD were elevated as well, with a mean of 1.05 µg g^{-1}, higher than the means at TK and PP, which were 0.65 µg g^{-1} and 0.64 µg g^{-1}, respectively. Fe concentrations in the soft tissues at PD were notably higher, ranging from 456 µg g^{-1} to 936 µg g^{-1}, with a mean of 754 µg g^{-1}, reflecting the higher contamination levels at PD. Ni concentrations in the soft tissues at PD were also higher, with a mean of 10.03 µg g^{-1}, compared to 8.62 µg g^{-1} at TK and 9.48 µg g^{-1} at PP, indicating more substantial Ni exposure at PD (Table 5). Overall, these results confirm that the soft tissues of *T. granosa* at PD accumulate higher concentrations of heavy metals than those at TK and PP, mirroring the patterns observed in the water and sediments.

3.4 Correlation analysis and stepwise multiple linear regression analysis

The concentration of Cd in soft tissues shows a moderate correlation with Cd levels in water (r = 0.55) (Table 6), indicating that water may be a primary source of Cd bioaccumulation in the soft tissues, particularly in PD. However, the correlation was lower with Cd in sediments (r = 0.20) and shells (r = 0.23), suggesting that these sources contribute less to Cd bioaccumulation in the cockles. Cu concentrations in soft tissues exhibit a strong correlation with Cu levels in both water (r = 0.74) and sediments (r = 0.85), indicating that both these environmental media play significant roles in Cu accumulation in the cockles' soft tissues. In contrast, the correlation with Cu in shells was weaker (r = 0.06), suggesting a lesser contribution from shells to Cu bioaccumulation.

TABLE 4 Overall statistics of concentrations (μg g^{-1} dry weight) of heavy metals in the shells of blood cockle *Tegillarca granosa* transplanted to Teluk Kemang (TK), Pasir Panjang (PP) and Port Dickson (PD); *n* = 5.

	Zn TK	Cu TK	Cd TK	Fe TK	Ni TK	Zn PP	Cu PP	Cd PP	Fe PP	Ni PP	Zn PD	Cu PD	Cd PD	Fe PD	Ni PD
Min	3.95	6.28	6.52	167	42.5	4.02	6.38	6.97	170	42.5	4.16	6.63	7.38	170	42.5
Max	4.29	6.99	7.38	172	43.9	4.71	6.99	7.45	181	45.3	5.52	6.99	7.94	192	46.9
Mean	4.12	6.62	6.90	170	43.3	4.27	6.56	7.22	172	44.4	4.94	6.79	7.68	181	45.8
SD	0.12	0.28	0.32	2.06	0.68	0.26	0.25	0.20	4.81	1.23	0.49	0.13	0.24	8.27	1.88
SE	0.06	0.12	0.14	0.92	0.31	0.12	0.11	0.09	2.15	0.55	0.22	0.06	0.11	3.70	0.84
Skewness	0.10	0.07	0.47	-1.01	0.14	1.09	1.33	-0.11	1.47	-0.78	-0.65	0.35	-0.12	-0.18	-1.48
Kurtosis	-0.80	-1.23	-0.66	-0.41	-1.65	-0.17	0.02	-1.56	0.21	-1.05	-0.38	-0.75	-1.52	-0.90	0.22

TABLE 5 Overall statistics of concentrations (μg g^{-1} dry weight) of heavy metals in the total soft tissues of blood cockle *Tegillarca granosa* transplanted to Teluk Kemang (TK), Pasir Panjang (PP) and Port Dickson (PD); *n* = 5.

	Zn TK	Cu TK	Cd TK	Fe TK	Ni TK	Zn PP	Cu PP	Cd PP	Fe PP	Ni PP	Zn PD	Cu PD	Cd PD	Fe PD	Ni PD
Min	103	3.60	0.53	456	7.78	116	4.28	0.53	456	9.01	116	4.28	0.53	456	9.01
Max	116	4.28	0.78	650	9.08	135	5.44	0.71	605	9.83	153	9.11	1.68	936	10.8
Mean	108	3.91	0.65	571	8.62	125	4.92	0.64	561	9.48	138	7.04	1.05	754	10.03
SD	5.03	0.28	0.11	73.3	0.54	7.16	0.44	0.07	59.7	0.32	16.1	2.07	0.48	198	0.75
SE	2.25	0.12	0.05	32.8	0.24	3.20	0.20	0.03	26.7	0.14	7.20	0.92	0.21	88.7	0.33
Skewness	0.38	0.28	0.24	-0.66	-0.78	0.12	-0.35	-0.56	-1.37	-0.48	-0.49	-0.38	0.29	-0.53	-0.20
Kurtosis	-1.23	-1.37	-1.64	-0.65	-0.86	-0.73	-0.92	-1.13	0.10	-1.13	-1.31	-1.50	-1.45	-0.96	-1.31

TABLE 6 Correlation coefficients between the metal levels in the soft tissues of cockles with those in the water, sediments and the blood cockle *Tegillarca granosa* shells.

Samples (*n* = 12)	CdST	CuST	FeST	NiST	ZnST
Cd Water	0.55	0.32	0.45	0.32	0.30
Cu Water	0.25	**0.74**	0.29	**0.64**	**0.75**
Fe Water	0.33	**0.70**	0.36	**0.63**	**0.68**
Ni Water	**0.62**	0.19	0.50	0.19	0.16
Zn Water	0.47	**0.88**	0.41	**0.77**	**0.86**
Cd Sediment	0.20	0.20	0.25	0.15	0.18
Cu Sediment	0.40	**0.85**	0.50	**0.80**	**0.83**
Fe Sediment	0.44	**0.87**	0.37	**0.78**	**0.89**
Ni Sediment	0.40	**0.86**	0.38	**0.78**	**0.84**
Zn Sediment	0.54	**0.81**	0.48	**0.75**	**0.80**
Cd Shell	0.23	**0.81**	0.32	**0.70**	**0.80**
Cu Shell	-0.49	0.06	-0.20	-0.05	0.07
Fe Shell	**0.84**	**0.60**	**0.72**	0.56	**0.61**
Ni Shell	**0.84**	**0.77**	**0.89**	**0.87**	**0.75**
Zn Shell	0.46	**0.61**	**0.60**	0.55	**0.66**
Cd Soft tissue	—	0.54	**0.74**	**0.59**	**0.53**
Cu Soft tissue		—	0.57	**0.96**	**0.99**
Fe Soft tissue			—	**0.72**	0.57
Ni Soft tissue				—	**0.95**
Zn Soft tissue					—

Values in bold are statistically significant at $p < 0.05$.

Fe in soft tissues was moderately to strongly correlated with Fe levels in both water ($r = 0.70$) and sediments ($r = 0.87$), indicating these sources' significant influence on Fe bioaccumulation in cockle tissues. The moderate correlation with Fe in shells ($r = 0.60$) further suggests that shells also contribute to the Fe levels in soft tissues, albeit to a lesser extent. Ni in soft tissues shows a moderate correlation with Ni in water ($r = 0.19$) but a strong correlation with Ni in sediments ($r = 0.86$) and shells ($r = 0.77$). This indicates that sediments and shells are key contributors to Ni bioaccumulation in the soft tissues, with water playing a less significant role. Zn concentrations in soft tissues were strongly correlated with Zn levels in both water ($r = 0.88$) and sediments ($r = 0.81$), reflecting the multi-source nature of Zn bioaccumulation in cockle tissues. The moderate correlation with Zn in shells ($r = 0.61$) further supports the idea that Zn bioaccumulation was influenced by multiple environmental media (Table 6).

Overall, the results (Table 6) underscore the significant correlations between heavy metal concentrations in the soft tissues of *T. granosa* and the metal levels in both water and sediments, particularly at PD. This highlights the importance of these environmental media in the process of metal bioaccumulation in cockles, with variations depending on the specific metal in question.

The regression model for Cd in soft tissues identifies Cu in shells ($\beta = -0.75$), Fe in shells ($\beta = 0.04$), and Ni in shells ($\beta = -0.08$) as significant predictors (Table 7). This model suggests that the interactions between these metals in shells significantly influence the accumulation of Cd in soft tissues, particularly at the Port Dickson site. The negative β value for Cu in shells implies an inverse relationship, indicating that Cd accumulation in soft tissues may decrease as Cu in shells increases, and vice versa. The regression model for Cu in soft tissues includes Cu in shells ($\beta = -4.20$) and Cd in shells ($\beta = 4.68$) as significant predictors. This finding highlights a complex relationship between Cu and Cd concentrations in shells and their combined effect on Cu accumulation in soft tissues. The negative β value for Cu in shells suggests that higher Cu levels in shells might reduce Cu accumulation in soft tissues. In contrast, the positive β for Cd in shells indicates that Cd presence in shells could enhance Cu bioaccumulation (Table 7).

The regression model for Fe in soft tissues includes Zn in shells (β = 174) and Cu in shells (β = –224) as significant predictors. This suggests that the presence of Zn and Cu in shells strongly influences Fe accumulation in soft tissues, with Zn contributing positively to Fe bioaccumulation and Cu exerting a negative effect. The large magnitude of these β values indicates a substantial impact of these shell metals on Fe levels in soft tissues. The model for Ni in soft tissues identifies Ni in shells (β = 0.12) and Fe in shells (β = 8.03) as significant predictors. This underscores the importance of shell composition, particularly the levels of Ni and Fe, in determining the extent of Ni bioaccumulation in the soft tissues of *T. granosa*. The regression model for Zn in soft tissues includes Zn in shells (β = 28.5) and Cd in shells (β = 0.33) as significant predictors. This model indicates that Zn and Cd concentrations in shells play crucial roles in Zn bioaccumulation in soft tissues, with both metals positively influencing Zn levels in these tissues (Table 7).

Overall, the results (Table 7) reveals the complex interplay between various metals in the shells and their influence on metal accumulation in the soft tissues of *T. granosa*, especially at the polluted site of PD. The regression models emphasize the significant impact of metal concentrations in shells on the bioaccumulation processes in soft tissues, highlighting shell composition as a key factor in determining the overall metal burden in these organisms. These findings suggest monitoring metal levels in shells could be crucial for understanding and managing heavy metal pollution in coastal environments.

TABLE 7 The most influential variables selected for metal accumulation (dependent variable) in the soft tissues (ST) of cockle *Tegillarca granosa* using stepwise multiple linear regression analysis.

Cd ST	β	Cu ST	β	Fe ST	B	Ni ST	β	Zn ST	β
Intercept	-0.05	Intercept	-0.76	Intercept	-34.9	Intercept	-0.058	Intercept	-2.15
Cu Shell	-0.75	Cu Shell	-4.20	Zn Shell	174	Ni Shell	0.12	Zn Shell	28.5
Fe Shell	0.04	Cd Shell	4.68	Cu Shell	-224	Cd Shell	1.33		
Ni Shell	-0.08			Fe Shell	8.03	Cu Shell	-0.83		
Cd Shell	0.33								

Note. The following independent variables were considered in the regression analysis, including Cd Water, Cu Water, Fe Water, Ni Water, Zn Water, Cd Sediment, Cu Sediment, Fe Sediment, Ni Sediment, Zn Sediment, Cd Shell, Cu Shell, Fe Shell, Ni Shell and Zn Shell.

4 | DISCUSSION

4.1 Differential bioaccumulation patterns across sites indicate *Tegillarca granosa* as an effective biomonitor of heavy metal pollution

The present findings reveal significant variations in heavy metal accumulation in the edible cockle *T. granosa* across the three study sites (Saavedra *et al.* 2009; Velez *et al.* 2016). The data indicate that the distribution of metals, specifically Zn, Cu, and Fe, varied significantly among these sites, with PD exhibiting the highest concentrations. This elevated level of contamination was likely due to its proximity to industrial activities known to release pollutants into the marine environment (Ismui *et al.* 2020). These findings were consistent with other studies that have documented a correlation between urban and industrial runoff and increased metal accumulation in marine organisms, underscoring the role of environmental factors in influencing bioaccumulation trends in species such as *T. granosa* (Saavedra *et al.* 2009; Velez *et al.* 2016).

The differential accumulation patterns observed in this study underscore the potential of *T. granosa* as a reliable biomonitor for assessing heavy metal pollution in coastal ecosystems (Lebrun *et al.* 2015; Villagrán *et al.* 2019; Pereira *et al.* 2023). The species' ability to accumulate metals provides critical insights into environmental health; however, it is essential to further investigate the biological responses of this species to increased metal exposure to fully understand the implications for both marine life and human consumption (Halit *et al.* 2018). Moreover, understanding how environmental stressors like heavy metal contamination affect the physiological and histological aspects of *T. granosa* will enhance the effectiveness of this species as a bioindicator, leading to more comprehensive ecological assessments and informing sustainable management practices (Samsi *et al.* 2017; Ruíz-Jarabo *et al.* 2020). Previous studies have shown that the health condition of *T. granosa* can be significantly impacted by heavy metal accumulation, leading to deformities and tissue structure alterations, further reinforcing its role as a bioindicator of environmental quality (Ismui *et al.* 2020).

Using *T. granosa* as a biomonitor is advantageous due to its widespread distribution and ability to accumulate metals in both soft tissues and shells. This dual accumulation provides a comprehensive picture of the environmental metal burden: soft tissues reflect more immediate exposure, while shells represent longer-term accumulation. This dual approach is crucial for understanding the health status of marine organisms in polluted ecosystems and supports the notion that effective monitoring requires a multifaceted approach (Bazzi 2014; Díaz-de-Alba *et al.* 2021). This multifaceted approach enhances our understanding of heavy metal dynamics within marine ecosystems and serves as a critical tool for predicting potential ecological risks associated with metal accumulation in marine food webs. Ultimately, it contributes to safeguarding human health and promoting sustainable coastal management practices.

4.2 Correlation between environmental metal levels and bioaccumulation in soft tissues supports the use of *Tegillarca granosa* in assessing bioavailability

The significant correlations between the concentrations of Cu and Zn, in the soft tissues of *T. granosa* and their levels in the surrounding water and sediments (Bazzi 2014; Rouane–Hacene *et al.* 2015) underscore the utility of *T. granosa* as a bioindicator species in environmental monitoring, effectively reflecting the bioavailability of metals within its habitat. Similar trends have been observed in other marine organisms, where bioaccumulation was linked to surrounding metal concentrations (Szefer *et al.* 1998; Lebrun *et al.* 2015; Rouane–Hacene *et al.* 2015).

The strong positive correlations observed for Cu and Zn suggest that these metals were highly bioavailable in the study environment (Szefer *et al.* 1998; Bazzi 2014). This is particularly concerning as studies have indicated that metal bioaccumulation in marine organisms can amplify associated risks for higher trophic levels within the food web, highlighting the potential for biomagnification in contaminated ecosystems (Bazzi 2014; Mustafa *et al.* 2024). Recognizing *T. granosa* as a reliable bioindicator reflects its significance in studying the dynamics of metal exposure and accumulation across different ecosystems. In various environments, organisms have been shown to bioconcentrate heavy metals at rates that raise concerns for their own health and that of consumers (Szefer *et al.* 1998; Bazzi 2014).

Findings of this study emphasize the potential of *T. granosa* to serve as a proxy for assessing the bioavailability of heavy metals in marine environments. Bioaccumulation is a critical factor that can lead to elevated concentrations of heavy metals in higher trophic levels, thereby contributing to ecological and human health risks. This has been observed in various marine organisms, such as oysters and mussels, which also exhibit significant metal accumulation (Szefer *et al.* 1998; Bazzi 2014; Lebrun *et al.* 2015; Rouane–Hacene *et al.* 2015). The strong correlations observed for Cu and Zn in this study indicate that *T. granosa* can effectively reflect the bioavailability of these metals, providing valuable insights into environmental exposure that could potentially affect other marine species, including those higher in the food chain (Szefer *et al.* 1998; Saavedra *et al.* 2009; Bazzi 2014; Rouane–Hacene *et al.* 2015).

Therefore, the correlations between environmental metal levels and bioaccumulation in *T. granosa* validate its use in assessing metal bioavailability. The species' ability to mirror the metal concentrations in water and sediments in its soft tissues suggests that it is a reliable indicator of the bioavailable fraction of contaminants in marine ecosystems. This makes *T. granosa* an essential tool for environmental assessments, particularly in areas impacted by heavy metal pollution.

4.3 Shell accumulation as an indicator of long-term exposure and environmental persistence of metals

The accumulation of heavy metals in the shells of the cockle *T. granosa* serves as a valuable indicator of long-term environmental exposure (Cardellicchio *et al.* 2008; Miedico *et al.* 2013; Velez *et al.* 2016). This phenomenon highlights the potential of using bivalve shells as a reliable bioindicator for assessing historical pollution levels, particularly in coastal regions where industrial activity may influence metal concentrations in marine organisms. The capacity for bioaccumulation in shells underscores the importance of monitoring these structures, as they provide a tangible record of environmental changes and pollution trends over time. Such records offer critical insights for ecological assessments and conservation efforts in affected areas (Roveta *et al.* 2021). Moreover, the inter-individual variability in metal concentrations among *T. granosa* from different sites suggests that localized conditions play a significant role in shaping the exposure profiles of these organisms. This finding emphasizes the need for comprehensive monitoring strategies for such variability (Saavedra *et al.* 2009; Piras *et al.* 2013).

The higher levels of metals, such as Cu and Zn, observed in the shells of *T. granosa* collected from PD compared to those from TK and PP indicate sustained exposure to metal pollution in the PD region (Saavedra *et al.* 2009). This observation was consistent with findings from various studies that stress the importance of regional monitoring, as different sites exhibit variability in metal accumulation due to both anthropogenic and natural factors. Such variability necessitates targeted assessments for effective environmental management (Miedico *et al.* 2013; Piras *et al.* 2013). The significant differences in metal concentrations across sampling locations highlight not only the local impacts of human activities but also the broader implications for biodiversity and public health. Given that these species are often part of the food web consumed by both marine fauna and humans, there are growing concerns about the potential for biomagnification through trophic levels (Velez *et al.* 2016; Ruíz-Fernández *et al.* 2018).

The integration of metals into the shells of *T.*

granosa over extended periods provides a valuable historical record of environmental exposure, complementing the insights gained from analyzing soft tissues. Additionally, assessing metal concentrations in bivalve tissues, particularly concerning environmental pollution, highlights the critical need for on-going monitoring to ensure public health and marine ecosystem sustainability, as these organisms represent broader ecological risks in coastal areas (Cardellicchio *et al.* 2008). The accumulation of heavy metals, such as Lead, and Cd, in molluscs has been well-documented, underscoring the importance of comprehensive monitoring programs to detect and mitigate the impacts of these persistent contaminants (Saavedra *et al.* 2009; Piras *et al.* 2013).

Hence, the accumulation of heavy metals in the shells of *T. granosa* at PD underscores the utility of shell analysis in biomonitoring programs. The shells serve as reliable indicators of long-term exposure and the persistence of metals in the environment. This capability made *T. granosa* an important species for monitoring current and historical contamination, contributing to a more comprehensive understanding of the ecological risks of heavy metals.

4. 4 Stepwise multiple regression analysis highlights the complex interactions between environmental media and bioaccumulation processes

The stepwise multiple regression analysis reveals the complex interactions between environmental media (water, sediments, and shells) and the bioaccumulation of heavy metals in *T. granosa* (Lebrun *et al.* 2015). This complexity is further highlighted by the observation that heavy metal concentrations in marine organisms can significantly exceed those found in surrounding water and sediments, indicating a pronounced tendency for bioconcentration from environmental sources. Risk assessments must account for this phenomenon (Bazzi 2014; Bamanga *et al.* 2019; Corrias *et al.* 2020; Al-Mutairi and Yap 2021). Moreover, similar bioaccumulation patterns have been observed in various marine invertebrates, suggesting a widespread phenomenon driven by biotic and abiotic factors. These include the influence of seasonal variations and the geochemical characteristics of the environment, which ultimately determine metal bioavailability (Chiarelli and Roccheri 2014; Lebrun *et al.* 2015).

The regression models suggest that metal levels in shells significantly predict metal accumulation in soft tissues, reflecting the interaction between different environmental compartments. Previous studies have demonstrated that bioaccumulation of metals in tissues can be influenced not only by the concentrations in surrounding water but also by sediment compositions and the organisms' life history traits, indicating the multifaceted relationships that govern these processes (Lovejoy 1999). This finding underscores the importance of considering multiple environmental media when assessing the bioaccumulation potential of species like *T. granosa*, as the metal content in shells appears to play a crucial role in determining the overall metal burden in the organism (Lovejoy 1999; Bazzi 2014; Chiarelli and Roccheri 2014). Moreover, the observed interactions between metals suggest that bioavailability and uptake mechanisms can vary across different habitats and environmental conditions, emphasizing the need for a comprehensive understanding of these dynamics in ecological risk assessments of marine organisms (Bazzi 2014; Chiarelli and Roccheri 2014).

Therefore, the findings from the stepwise multiple regression analysis highlight the complexity of bioaccumulation processes in *T. granosa*. The significant influence of shell metal concentrations on accumulation in soft tissues underscores the interconnectedness of environmental compartments in determining metal bioavailability and uptake. This complexity necessitates a comprehensive approach to biomonitoring, one that considers the interactions between various environmental media to better predict and manage the risks associated with heavy metal contamination.

4.5 Comparison of present findings with reported studies on heavy metal accumulation in marine bivalves

The findings of this study on heavy metal accumulation in *T. granosa* are consistent with those reported in previous studies on marine bivalves, which have extensively documented the capacity of these organisms to accumulate metals in their tissues and shells. These results reinforce the critical role of marine bivalves as bioindicators for monitoring environmental pollution. Studies such as those by Lü *et al.* (2017), Liu *et al.* (2019), and Pérez-López *et al.* (2003) have highlighted similar patterns of metal accumulation, demonstrating the widespread ability of bivalves to reflect the metal concentrations in their surrounding environments. The implications of these findings extend beyond the organisms themselves, as elevated heavy metal concentrations can lead to bioaccumulation in higher trophic levels, ultimately affecting food safety and public health through consuming contaminated seafood (Bazzi 2014).

In transplantation studies of bivalves, the relationship between heavy metal concentrations in bivalves and their surrounding environment has been emphasized, underscoring the potential of these organisms as effective bioindicators for monitoring marine pollution levels. Understanding the mechanisms of metal accumulation in *T. granosa* provides valuable insights into the bioavailability of heavy metals in coastal ecosystems. This reinforces the importance of utilizing such species in biomonitoring efforts to provide early warning signals of environmental contamination (Chiarelli and Roccheri 2014). The distinct patterns of metal bioaccumulation observed in *T. granosa* further indicate the necessity for routine monitoring strategies to better assess the ecological impacts of heavy metal pollution, particularly in regions experiencing significant industrial activity (Saavedra *et al.* 2009; Bazzi 2014; Chiarelli and Roccheri 2014; Nanda 2014).

The accumulation patterns identified in this study align with those reported in previous research, underscoring the need for on-going monitoring and assessment. Routine monitoring not only aids in protecting marine biodiversity but also informs regulatory frameworks aimed at safeguarding public health through enhanced seafood safety protocols (Bazzi 2014). By comparing the current data with reported studies, this study highlighted the relevance of *T. granosa* in biomonitoring programs. It emphasizes the importance of continued research on species-specific responses to environmental contamination. This comparison also reinforces the need to use multiple species and tissue types in environmental assessments to obtain a comprehensive view of the ecological impacts of heavy metal pollution in coastal ecosystems.

The present findings corroborate previous studies on heavy metal accumulation in marine bivalves and expand the understanding of bioaccumulation processes in *T. granosa*. The consistency of these findings with past research emphasizes the significance of *T. granosa* as a reliable biomonitor species while highlighting the broader ecological and public health implications of heavy metal pollution in marine environments.

4.6 Influence of environmental factors on heavy metal bioaccumulation in *Tegillarca granosa* during the transplantation period

While the primary focus of this study was on the bioaccumulation of heavy metals in *T. granosa* across different sites, it is essential to consider the potential influence of environmental factors such as salinity, temperature, and sediment composition. Variations in temperature across the three sites may have led to differential metabolic rates in the cockles, affecting their metal uptake and storage capacity. This is consistent with findings that highlight the significant role of climatic factors in influencing bioaccumulation patterns (Cheggour *et al.* 2001; Jung *et al.* 2006; Mubiana and Blust 2007; Lebrun *et al.* 2015; Velez *et al.* 2016; Samsi *et al.* 2017; Marques *et al.* 2018; Pereira *et al.* 2023).

Salinity is another crucial factor that can alter metal bioavailability and accumulation in marine organisms. Changes in salinity can influence the chemical speciation of metals, impacting their solubility and subsequent availability for organism uptake. This is supported by research that emphasizes the dependence of bioaccumulation on various environmental factors beyond just the concentration of metals in the water (Lovejoy 1999; Chiarelli and Roccheri 2014). Additionally, the interaction between temperature and salinity may produce synergistic effects on metal uptake, further complicating the interpretation of bioaccumulation results. Comprehensive analyses that account for these interrelated environmental parameters are necessary (Lovejoy 1999; Chiarelli and Roccheri 2014; Lebrun *et al.* 2015).

Furthermore, the potential influence of sediment composition on metal bioavailability and transfer to benthic organisms like *T. granosa* cannot be overlooked. Sedimentary metal concentrations vary significantly between different environments, directly affecting the metal concentrations found within the tissues of marine organisms residing in those sediments. This highlights the importance of considering sediment characteristics in bioaccumulation studies (Bazzi 2014; Chiarelli and Roccheri 2014; Fonti *et al.* 2015; Lebrun *et al.* 2015; Chen *et al.* 2016). Variations in sediment type, particularly in grain size and organic matter content, can further modulate the retention and release of heavy metals and their availability for uptake by bivalves. This emphasizes the necessity for a holistic assessment of the environmental factors that may influence the observed bioaccumulation patterns in this study (Chiarelli and Roccheri 2014; Lebrun *et al.* 2015; Velez *et al.* 2016; Birch *et al.* 2019).

Therefore, while the primary focus of this study was on the differences in heavy metal accumulation across the three sites, it is important to acknowledge that environmental factors such as salinity, temperature, and sediment composition likely played a role in influencing these patterns. These factors can significantly affect metal bioavailability and the capacity of *T. granosa* to accumulate metals, contributing to the site-specific variations observed in this study. Future research should consider these environmental variables more explicitly to better understand their interactions with heavy metal pollution and their effects on bioaccumulation processes in marine bivalves.

4.7 Study limitations

Several limitations of this study should be acknowledged to provide a more balanced interpretation of the findings. One of the primary limitations is the lack of replication across different seasons and broader geographical areas, which could influence the generalizability of the results. The study was conducted at a single time point, which may not fully capture the temporal variations in heavy metal concentrations and bioaccumulation patterns in *T. granosa*. Seasonal changes in factors such as temperature, salinity, and pollution levels could significantly affect metal uptake, and without replication, the study's findings may not represent the full range of environmental conditions. It is often difficult to find a suitable replication field site for the study (Fraser *et al.* 2020; Galib et al., 2022) which was also the case for us.

Additionally, the study focused on three specific sites, which, while providing valuable insights into localized contamination, may not reflect broader regional trends. The absence of spatial replication across different coastal regions limits the ability to generalize the results to other areas with potentially different environmental conditions and pollution sources. Furthermore, the study did not incorporate many individual samples within each site, which could impact the statistical robustness of the conclusions. A larger sample size would allow for more precise estimates of metal concentrations and a better understanding of the variability within and between populations.

5 | CONCLUSIONS

This study has demonstrated the utility of *T. granosa* as a highly effective biomonitor for assessing heavy metal contamination in coastal environments. By analyzing metal concentrations in both the soft tissues and shells across three sites, we observed significant variations in metal accumulation, with PD consistently exhibiting the highest levels due to its proximity to industrial activities. The strong correlations between environmental metal levels and bioaccumulation in soft tissues further validate the use of *T. granosa* in assessing the bioavailability of contaminants, providing a reliable measure of environmental exposure.

The study also underscored the importance of shells as indicators of long-term metal exposure. The persistent accumulation of metals in the shells at PD reflects the chronic contamination in this area and highlights the ongoing environmental burden of industrial pollutants. Additionally, the stepwise multiple regression analysis findings revealed the complex interactions between environmental media and the bioaccumulation processes in *T. granosa*. These interactions emphasize the need for a comprehensive approach to biomonitoring that accounts for the dynamics between water, sediments, and shells. These findings support using *T. granosa* as a reliable indicator species for monitoring heavy metal pollution and assessing ecological risks in coastal ecosystems. However, to strengthen the conclusions drawn from this study, future research should incorporate multiple replicates across different seasons and geographical areas to verify these findings and better understand the temporal and spatial variability of metal bioaccumulation. This approach would enhance the robustness of biomonitoring guidelines and provide a more comprehensive environmental health assessment.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

AUTHORS' CONTRIBUTION

Conceptualization, CKY and KAA-M; methodology and validation, CKY and KAA-M; formal analysis, CKY; investigation, CKY; resources, KAA-M; data curation, CKY; writing—original draft preparation, CKY; writing—review and editing, CKY and KAA-M. Both authors have read and agreed to the published version of the manuscript.

DATA AVAILABILITY STATEMENT

The data that support the findings of the study will be made available on a reasonable request from the corresponding author.

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