https://doi.org/10.22478/ufpb.1981-1268.2023v17n1.65979

Biomarkers and health status of the crab *Ucides cordatus* to assess the impact of contaminants in an estuarine mangrove region in the Brazilian Amazon

Suelen Rosana Sampaio de Oliveira[1*], Luciana Barros Oliveira[2], Luan Jonatas da Silva Ferreira[3], Gerson dos Santos Protazio[3], Debora Martins Silva Santos[4], Lina Clara Gayoso e Almendra Ibiapina Moreno[4], Raimunda Nonata Fortes Carvalho Neta[4]

1 Postgraduate Program in Biodiversidade e Biotecnologia (BIONORTE), Universidade Estadual do Maranhão. Predio da Pós-Graduação do CCBS, Cidade Universitaria, Av. dos Portugueses, 1966, Bacanga, São Luís, MA, Brasil. 65080-805.
2 Postgraduate Program in Ecologia e Conservação da Biodiversidade (PPGECB), Universidade Estadual do Maranhão. Cidade Universitária Paulo VI – Caixa Postal 09 – São Luís/MA, Brasil.
3 Laboratório de Biomarcadores em Organismos Aquáticos, Universidade Estadual do Maranhão, Cidade Universitária Paulo VI, Avenida Lourenço Vieira da Silva 1.000 – São Luís/MA, Brasil.
4 Departamento de Biologia, Universidade Estadual do Maranhão. Sá Viana, São Luís - MA, Brasil. 65085-582 .
*Author for correspondence: suelenrsdo@gmail.com

Received 28 February 2022.
Accepted 30 April 2023.
Published 02 May 2023.

Abstract - The present work aims to analyze biomarkers and health status of the crab *Ucides cordatus* to verify the stress levels of an estuarine mangrove region in the Brazilian Amazon. For this, males *U. cordatus* were captured in two areas (P1 = industrial harbor region and P2 = region close to a fishing pier) for further analysis of biometry, biomarkers and organ damage index (gills and hepatopancreas). In addition, water samples were collected for the investigation of trace metals. The results showed higher values of trace metals and histological lesions in P1, but it was observed that the organ damage was similar between the areas. Lesions classified as irreversible were found in large numbers in P1, compromising the functioning of the organ and the health of organisms residing in this region. Therefore, crabs are under impacts showing biological responses and probably bioaccumulating harmful substances, compromising the health of the species.

Keywords: Biomonitoring. Decapoda. Harbor region. Histopathology. Impacts. Trace metals.

Biomarcadores e estado de saúde do caranguejo *Ucides cordatus* para avaliar o impacto de contaminantes em uma região de mangue estuarino na Amazônia brasileira

Resumo - O presente trabalho tem como objetivo analisar biomarcadores e estado de saúde do caranguejo *Ucides cordatus* para verificar os níveis de estresse de uma região de mangue estuarino na Amazônia brasileira. Para isso, machos de *U. cordatus* foram capturados em duas áreas (P1 = região portuária industrial e P2 = região próxima a um porto de pesca) para posterior análise de biometria, biomarcadores e índice de dano aos órgãos (brânquias e hepatopâncreas). Além disso, amostras de água foram coletadas para a investigação de metais-traço. Os resultados mostraram maiores valores de metais traço e lesões histológicas em P1, mas observou-se que o dano ao órgão foi semelhante entre as áreas. As lesões classificadas como irreversíveis foram encontradas em grande número em P1, comprometendo o funcionamento do órgão e a saúde dos organismos residentes nessa região. Portanto, os caranguejos estão sob impactos apresentando respostas biológicas e provavelmente bioacumulando substâncias nocivas, comprometendo a saúde da espécie.

Palavras-chave: Biomonitoramento. Decápodo. Histopatologia. Impactos. Metais-traço. Região Portuária.

Biomarcadores y estado de salud del cangrejo *Ucides cordatus* para evaluar el impacto de los contaminantes en una región de manglares estuarinos en la Amazonía brasileña

Resumen - El presente trabajo tiene como objetivo analizar los biomarcadores y el estado de salud del cangrejo *Ucides cordatus* para verificar los niveles de estrés de una región de manglares estuarinos en la Amazonía brasileña. Para ello, se capturaron machos de *U. cordatus* en dos zonas (P1 = región portuaria industrial y P2 = región cercana a un muelle pesquero) para su posterior análisis de biometría, biomarcadores e índice de daño de órganos. Además, se recolectaron muestras de agua para la investigación de trazas de metales. Los resultados mostraron valores más altos de metales traza y lesiones histológicas en P1, pero se observó que el daño orgánico fue similar entre las áreas. Lesiones clasificadas como irreversibles se encontraron en gran número en P1, comprometiendo el funcionamiento del órgano y la salud de los organismos que residen en esta región. Por lo tanto, los cangrejos están bajo impactos mostrando respuestas biológicas y probablemente bioacumulando sustancias nocivas, comprometiendo la salud de la especie.

Palabras clave: Biomonitoreo. Decápodo. Histopatología. Impactos. Rastrea metales. Región portuaria.

Introduction

Mangroves are high-productivity ecosystems located between terrestrial and aquatic saline environments (Liu et al. 2018). They have great socioeconomic importance, directly linked to the human subsistence, besides a fundamental ecological role, since these ecosystems participate in the protection of coastal zones and provide shelter and resources for migratory and resident species, in addition to being carbon sinks (Walker et al. 2022).

Despite being an essential environment for many living beings, the global area of mangrove forests has been significantly reduced, mainly by deforestation, with an estimated loss of one third of the original area during the 20th century, and about 1,540.22 km2 area lost between 2000 and 2017 (Polidoro et al. 2014; Jakovac et al. 2020).

In the mangroves of the Brazilian Amazon, the main sources of deforestation and contamination are aquaculture, domestic effluents and activities in the industrial and port sectors (Ferreira and Lacerda

2016; Monteiro et al. 2016; Bernardino et al. 2021; Pinheiro-Sousa et al. 2021). On the northern coastal of Brazil there is an important port complex, which includes the Ponta da Madeira terminal, the Port of Itaqui and the Alumar terminal. In 2021, this complex handled approximately 228 million tons of various products, such as iron ore, oil derivatives, fertilizers and soy (ANTAQ 2021; González-Gorbeña et al. 2015), positively impacting the economy.

On the other hand, the growth of enterprises in this region has increased the occurrence of coastal water pollution, mangrove degradation and environmental accidents (Amaral and Alfredin 2010; EMAP 2022). And, as mangroves have a great capacity to retain contaminants in the sediment, resulting from anthropogenic activities drained into tidal and river waters (Gopalakrishnan et al. 2020), there is a risk for the entire food chain. In this same region, heavy metals available in the sediment have already been found, especially Al, Fe, Mg, Zn, Cu and Pb (Pinheiro-Sousa et al. 2021), and thus available to the biota such harmful elements.

Investigating the situation of the species that inhabit the mangrove through monitoring is important to diagnose the quality of the environment and the health of organisms. Monitoring becomes more effective when using native organisms, as they allow a realistic scenario of impacts to the environment. In particular, the crab species *Ucides cordatus* (Crustacea, Decapoda; Linnaeus, 1763) (Common name: caranguejo-uçá) is widely distributed in the mangrove ecosystem and has ecological importance, as it participates in the biogeochemical cycle, in the bioturbation of mangrove soil, etc. (Nordhaus et al. 2006; Pülmanns et al. 2014) and economic, since the traditional community survives by collecting and selling this animal. In addition to these characteristics, it is considered a key species in monitoring studies and considered an excellent biomonitor species (dos Santos et al. 2019).

An easy method to assess environmental stress in organisms is through biomarkers, which provide us with fast and effective responses at different levels of biological organization. In addition, they subsidize information about the risk to the health of organisms and the prevention of impacts. Histopathological biomarkers, for example, demonstrate the damage caused to biological tissues by exposure to contaminants (such as heavy metals) which, depending on the substance and the time of exposure, cause irreversible damage. Thus, in this study we aimed to investigate the health status of the crab *Ucides cordatus* from harbor regions through analysis of biomarkers and environmental analyses, to verify the stress levels in the estuarine environment of the Brazilian Amazon.

Material and Methods

Study area

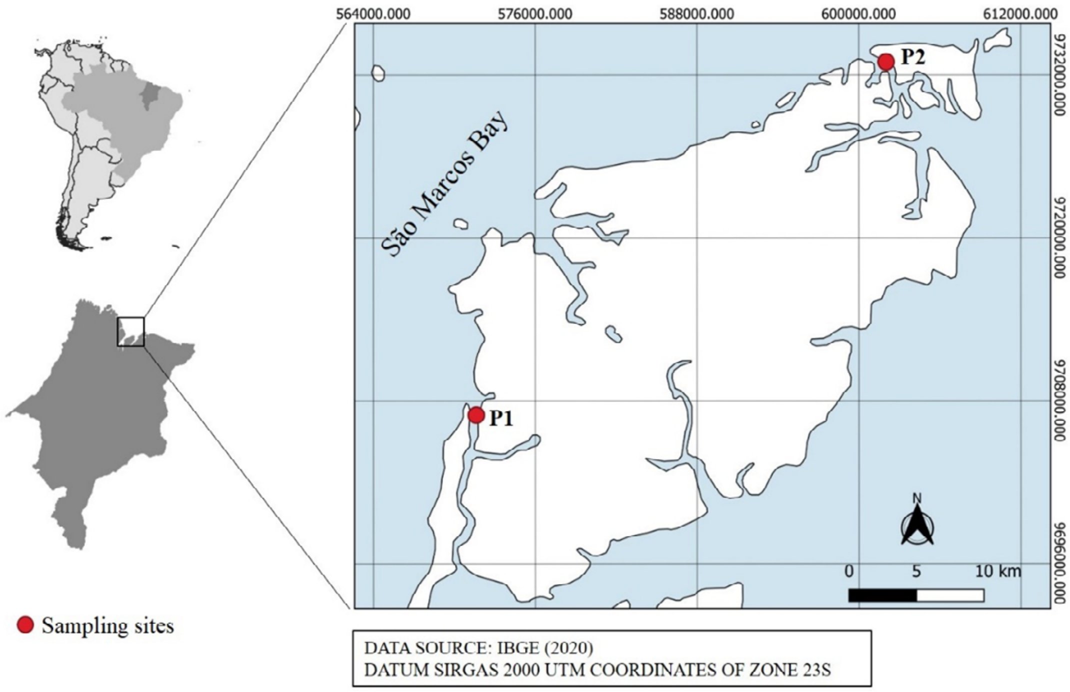
The study area comprises São Marcos Bay (Maranhão, Brazil) – an estuarine region belonging to the Brazilian Amazon, which houses part of the largest continuous band of mangroves under legal protection in the world (ICMBio 2018). It is considered an important fishing site, where fishing resources are still captured for the subsistence of riverside families (Ribeiro and Castro 2017) and notably recognized for housing the São Luís Industrial and Port Complex, which was once considered one of the largest in cargo handling in Latin America (Amaral and Alfredin, 2010).

The region is characterized by having little thermal variability during the year - from 30.3 to 32.5°C for maximum temperatures and 23.4 to 25°C for minimum temperatures (INMET 2021) and therefore, the four seasons do not are well-defined, only two are observed: the rainy season, from January to June and the dry season, from July to December (INMET 2021; Pinheiro 2015).

Sampling locations

Two sites were selected along São Marcos Bay to collect biological and environmental samples: P1 and P2 (Figure 1). Site P1 (2°39'3.70"S; 44°21'20.40"W) is located between two ports in the industrial region of the capital, where there is an intense flow of small and large vessels, as a result of port and fishing activities; site P2 (2°24'55.85"S; 44° 4'56.55"W) is close to an important fishing and tourism port in the region (approximately 1.5 km), subject to human interference (effluents, solid waste and oil from boat engine, etc.), considered in this study as a site of moderate impact.

Figure 1. Sampling sites along São Marcos Bay, northeast Brazil.



Sampling

Biological samples

In quarterly campaigns, 151 adult male specimens were captured from inside the burrows (SISBIO registration n°76372-1), randomly in the mangroves of the collection sites, during the rainy and dry seasons of 2021. The seasonal periods in the region are defined according to the series of historical meteorological data provided by the National Institute of Meteorology - INMET.

Environmental samples

During the ebb tide, samples of estuarine water were collected at 5-10 cm depth in sterilized bottles for analysis of important trace metals Aluminum, Cadmium, Lead, Copper, Chromium, Manganese, Mercury, Nickel and Zinc, analyzed according to the Standard Methods for the Examination of Water and Wastewaters and the US EPA. The water samples were filtered, preserved in nitric acid and analyzed by Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES Optima 8300, Perkin Elmer).

At the time of water collection, the following physical-chemical parameters of the water along the estuary were measured: dissolved oxygen (mg/L), temperature (°C), salinity (ppm) and pH, using an AKSO multiparameter meter (model AK88).

The collection and recording of all environmental samples always took place in the morning, during the ebb tide.

Analysis

Species biometric parameters

Biometric data for each crab (n = 151) were recorded, taking the following variables: wet weight (WW) in grams, measured on a precision scale; cephalothorax width (CW) and cephalothorax length (CL) in centimeters, measured with the aid of a digital caliper. In this research, only male crabs in intermolt state were analyzed (de Oliveira et al. 2019).

Histology

Forty gills and forty hepatopancreas samples (n = 80) of *U. cordatus* were deposited in AFA fixative solution (Alcohol Formalin Acetic acid) for 48h and subsequently dehydrated in a graded series of ethanol, diaphanized in xylene and embedded in paraffin to form blocks; finally cut transversely to 5µm thickness and staining with Hematoxylin and Eosin (HE) (Luna 1968). Two slides of each organ were analyzed per crab.

A count of the total lesions in each organ was performed and later classified according to the reaction pattern: circulatory, inflammatory, neoplastic, progressive and regressive (Bernet et al. 1999). An organ damage index (Iorg.) adapted for crabs was also calculated (Jerome et al. 2017). The index used can be calculated using the formula:

$$I_{org} = \Sigma_{alt} (a \times w)$$

"a" characterizes the intensity of the alteration (0 = absent, 1-2 = minimal occurrence, 3-4 = moderate occurrence; 5-6 = severe occurrence); "w" is the importance factor, where the values tabulated by Bernet et al. (1999) for each change it ranges from 1 to 3 (1 = reversible changes; 2 = moderate changes, reversible if the stressor is neutralized and 3 = irreversible changes).

Statistical analysis

For each analysis, the results were tabulated and tested for data normality, using the Kolmogorov-Smirnov test, accepting $p>0.05$ for normal data. The T test ($p<0.05$) was used in two stages: to compare the abiotic water variables in relation to the seasons of the year, in each area; and to compare the concentration of trace metals in water between areas. The Mann-Whitney test was used in the other analyses (biometric data and organ damage indexes). The comparative chart of the abiotic variables was constructed based on the mean and standard deviation. The median, 1st and 3rd quartiles, minimum and maximum values were used to build the table of biometric data of the species. The Statistica® Desktop program (StatSoft, version 10.0) was used in all analyses.

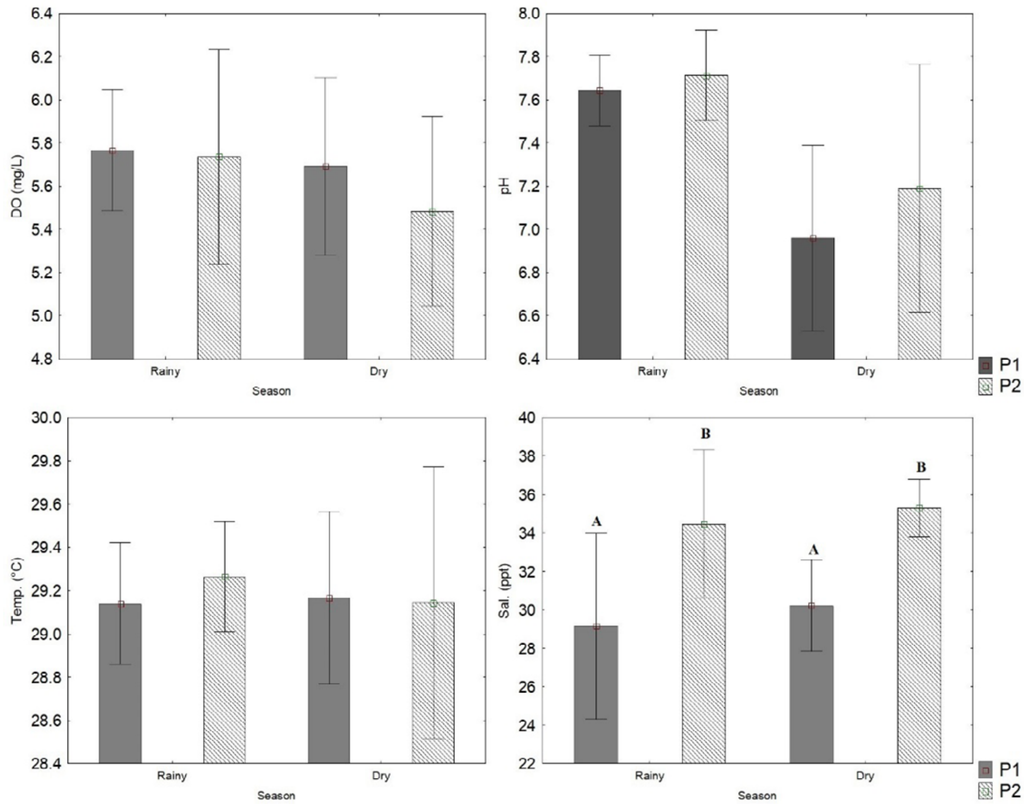
Results and Discussion

Environmental analyzes

Abiotic variables

Abiotic variables of water showed expected conditions for this estuarine region, mainly in terms of temperature, showing that there is little variation between locations; however, salinity showed a statistical difference between the sites ($p < 0.05$), highlighting the P2 area, the site closest to the Atlantic Ocean (Figure 2). These values presented are similar to research carried out in these regions (Sousa et al. 2013; Ribeiro et al. 2020) and are in accordance with acceptable local limits (CONAMA 2005).

Figure 2. Result of the T test for the abiotic water data collected during the rainy and dry seasons at the sampling sites, expressed as mean and standard deviation. Different letters indicate statistically significant difference.



The parameters already mentioned, when outside the appropriate limits, can be harmful to the biota: pH can contribute to the precipitation and potentiation of the effects of trace metals; dissolved oxygen in low concentrations causes damage to aquatic organisms, such as hypoxia; high temperatures directly interfere with the availability of oxygen in the water, mainly due to the increase in energy expenditure; the change in salinity, in addition to increasing metabolic rates, negatively interferes with the immune system of aquatic animals (Wang et al. 2018; Adeleke et al. 2021; García-Rueda et al. 2021; Wang et al. 2022). However, as the values are within acceptable limits, the environment can be considered appropriate for estuarine life, in relation to these parameters.

Trace metal analysis

The result of the analysis showed the presence of trace metals in the water at all collection sites, with the highest values occurring in area P1, the industrial region (Table 1).

Table 1. Results of trace metal analysis in water at collection sites in São Marcos Bay, Brazil.

Water (mg L^{-1})	P1	P2	Reference Values [b]	LQ
Aluminum	0.25 ± 0.01	< 0.1	1.5 mg L^{-1} Al	0.1
Cadmium	< 0.001	< 0.001	0.04 mg L^{-1} Cd	0.001
Chrome	0.04 ± 0.02	0.03 ± 0.01	1.1 mg L^{-1} Cr	0.01
Copper *	0.2 ± 0.01	0.09 ± 0.01	7.8 µg L^{-1} Cu	0.001
Lead	< 0.01	< 0.01	0.21 mg L^{-1} Pb	0.01
Manganese *	4.2 ± 0.16 [b]	1.6 ± 0.12 [b]	0.1 mg L^{-1} Mn	0.1
Mercury	< 0.0002	< 0.0002	1.8 µg L^{-1} Hg	0.0002
Nickel	< 0.005	< 0.005	74 µg L^{-1} Ni	0.005
Zinc *	0.16 ± 0.01 [b]	0.03 ± 0.01	0.12 mg L^{-1} Zn	0.01

a Threshold of reference values by Brazilian legislation (CONAMA 2005). b Indicates values above the limits defined by current federal legislation (CONAMA 2005). * Indicates significant statistical difference ($p < 0.05$). LQ: Limit of quantification.

Metals such as copper, manganese, nickel and zinc are considered essential for plant and animal species, provided they are in low concentrations (Rocha et al. 1985; Summer et al. 2019; Yang et al. 2021). However, the concentration of zinc and manganese in the water is above the established value by current Brazilian legislation (limit: 0.12 mg.L^{-1} Zn and 0.1 mg.L^{-1} Mn), which can cause serious impacts on the aquatic biota and on humans (Belitz et al. 2009). Studies indicate that zinc toxicity can cause diseases such as anemia (linked to copper deficiency) and damage to the respiratory and gastrointestinal tracts (Hussain et al. 2022). Manganese is linked to good development and health in animals, but chronic exposure causes disturbances in energy metabolism, neurofunctional changes: impairment of neuromotor and cognitive functions, among others (Bevan et al. 2017; Mattison et al. 2017; Vásquez-Procopio et al. 2020). The other metals investigated in this research are within acceptable local limits (CONAMA 2005).

Biometric data

Crab measurements differed statistically between areas during the rainy season, showing P1 crabs with higher measurements and weights ($p<0.05$). However, the data for the dry season did not show a significant difference in any biometric variable (Table 2). Despite showing higher values in P1, these data are lower than those recorded for reference regions in de Oliveira et al. (2019), but consistent with the records for the harbor region of São Marcos Bay (de Oliveira et al. 2019; de Jesus et al. 2021), showing that organisms are historically smaller in this area.

Table 2. Medians of the biometric variables of *Ucides cordatus* sampled at the sites P1 and P2 during the rainy and dry season of 2021.

	Rainy season		**Dry Season**	
	P1	P2	P1	P2
WW(g)	135.3*	118.0	131.0	132
Q_1-Q_3	118.7 – 156.7	91.7 - 140.5	110.0 – 153.0	116.0 – 149.0
CW (cm)	6.8*	6.0	6.6	6.7
Q_1-Q_3	6.0 – 7.0	5.0 - 7.0	6.4 - 7.1	6.1 - 7.0
CL (cm)	4.7*	4.06	4.8	4.9
Q_1-Q_3	4.0 – 5.0	4.03 – 4.09	4.6 – 5.0	4.6 – 5.2

WW: total weight; CW: cephalothorax width; CL: cephalothorax length, Q1: first quartile; Q3: third quartile. n = 151. * Indicates significant statistical difference ($p < 0.05$).

At site P1, an area under the influence of the port complex, the highest values for trace metals were recorded in the water above and within the permitted limits, so the animals are exposed to these and other substances in a complex mixture in the environment, bioaccumulating in their tissues and causing adverse effects. Thus, organisms increase their metabolic rates to defend the body from possible damage, and consequently the growth rate changes, becoming slower (Kavun and Podgurskaya 2009; Jinhui et al. 2019). In part, this may explain the reduced size and weight of the crabs in P1, as the organisms are living in the presence of harmful substances, impairing their growth rate. For site P2, far from the industrial and port sector, the biometric variables may be associated with anthropic impacts, with emphasis on predatory fishing of this resource, since it is a region where one of the main sources of income for communities is fishing. There are records of increased fishing effort, as well as biometric variables in smaller numbers in crabs from the northern coast of Brazil, showing that overfishing in the region has been occurring over the years (Legat and Legat 2009), with consequences for the species. It is worth mentioning that the growth of *U. cordatus* is slow and they are continuously harvested before their complete maturation period, impairing their reproductive cycle (de Lima et al. 2018). Thus, the species does not have enough time to recover the population number, especially in P2, which is a fishing region.

Histological biomarkers

The histological examination of the gills showed lesions in all crabs captured (Figure 3), but in greater proportion in P1. The most recurrent alterations are included in the regressive reaction pattern (processes that result in the reduction or functional loss of an organ) and are classified as irreversible (disturbances in the pilaster cells, collapsed lamellae and necrosis); the other alterations were classified as easily reversible, with the end of the exposure (deformation of the marginal canal and detachment of the cuticle). The reaction pattern was presented in the order: regressive > progressive for P1 and P2. The presence of parasites was also recorded in the two collection sites. In the hepatopancreas, the lesions are classified in the pattern of inflammatory reaction (processes belonging to several reaction patterns, being difficult to attribute to a single reaction pattern) with moderate injury (hemocyte infiltration) and regressive with reversible (abnormal lumen, damaged myoepithelial layer and epithelium delamination) and irreversible (necrosis and B vacuolated). Two patterns of reactions were found in this tissue, in the order regressive > inflammatory, for both areas (P1 and P2). As with the gills, the highest proportion of lesions in the hepatopancreas was found for the crabs from the P1 site (Figure 4).

Figure 3. Histological alterations found in the gills and hepatopancreas of *Ucides cordatus*, collected in São Marcos Bay, BR. 3-A and 3-B: P2; 3-C and 3D: P1.AL: abnormal lumen; CL: collapsed lamellae; DMC: deformation of the marginal channel; DML: damaged myoepithelial layer; NE: necrosis; VBC: vacuolated B cells. HE. 5X.

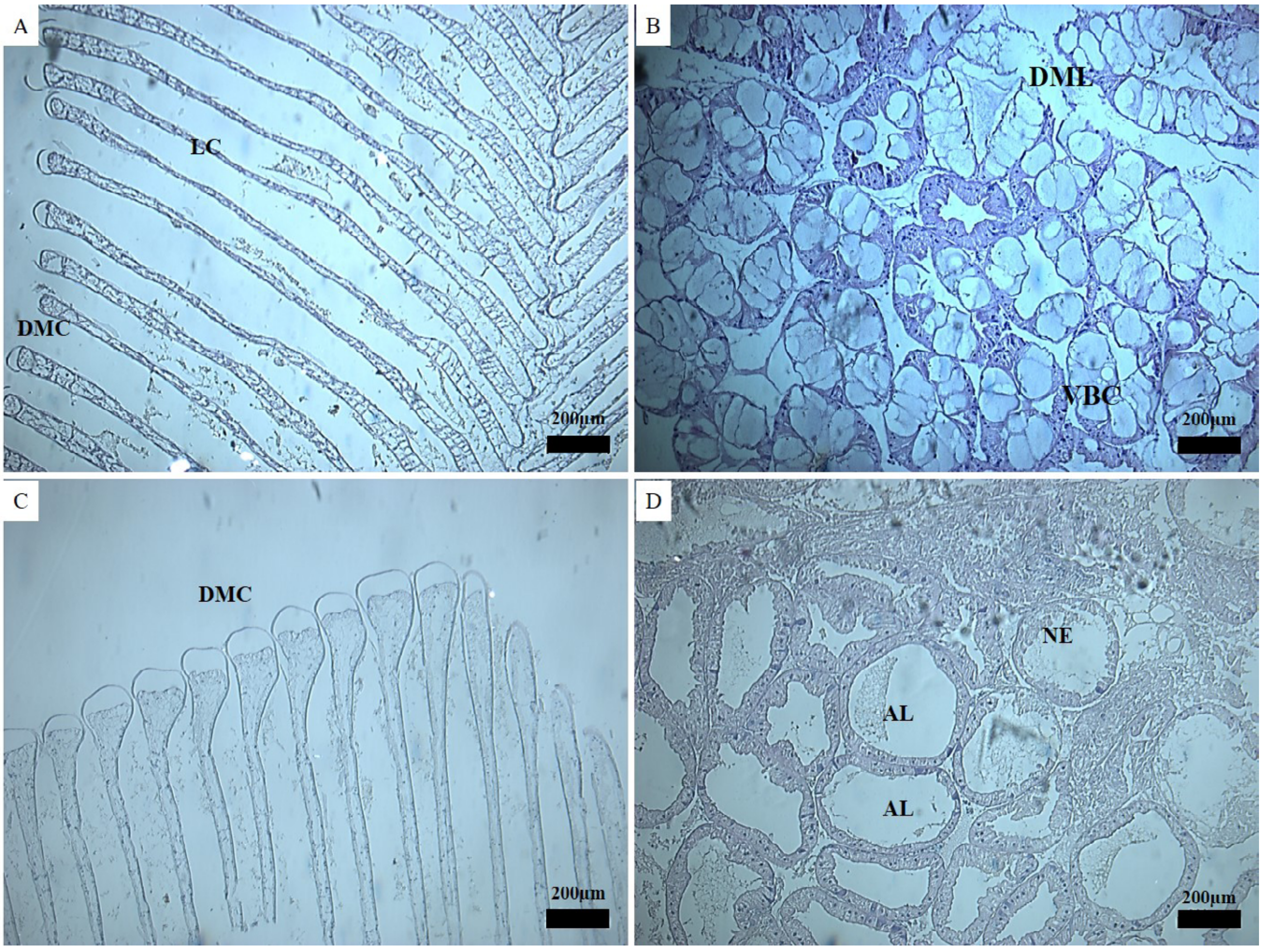
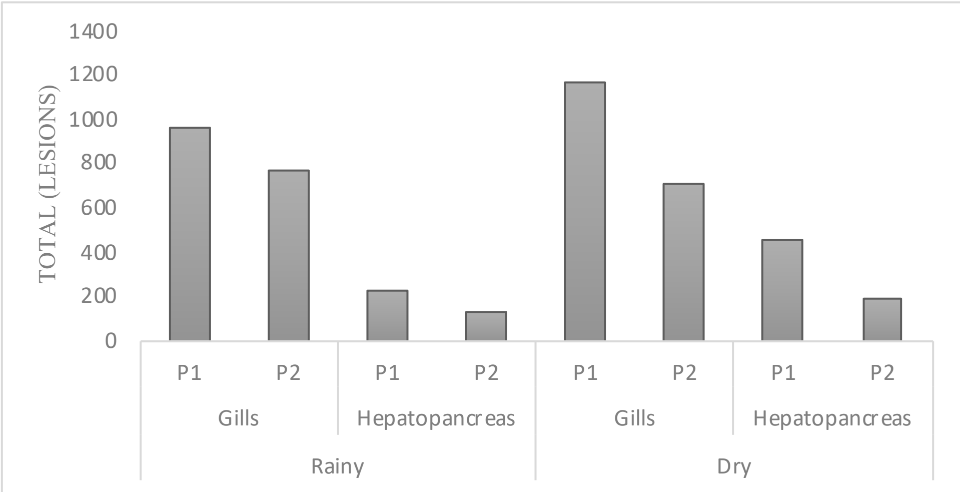


Figure 4. Frequency of lesions in *Ucides cordatus* collected during the 2021 seasonal periods at two collection sites.

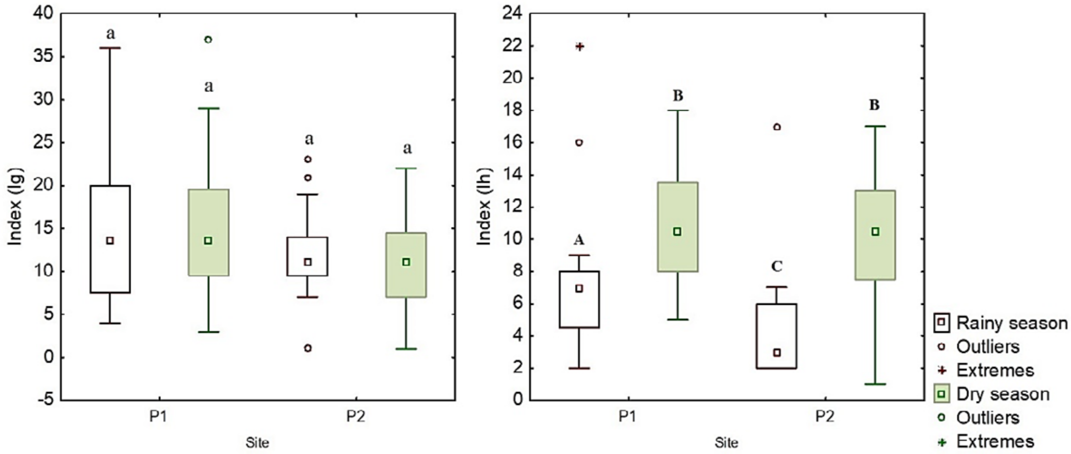


The organ index showed no statistically significant differences for the gills. For the hepatopancreas only the rainy season showed statistical difference between areas ($p<0.05$) (Figure 5). It is estimated that this difference in I_h during the rainy season is because a greater volume of xenobiotics is leached into the estuary in this season. The index reflects how severely the organ is affected by the histopathologies. In this way, the values for the sites represent a high degree of organ damage, such as disturbance in the pilaster cells and necrosis, irreversible damage. As a consequence, we have a high probability of total loss in the functioning of the organ, compromising the health of the organism or even leading to death.

The gills are key organs for osmoregulation, respiration and ion exchange, and because it has a high permeability, it tends to incorporate harmful substances into its tissues (Rainbow 2018). The hepatopancreas (midgut gland) performs absorption, secretion and detoxification functions in crustaceans, minimizing the effects of contaminants (Corrêa et al. 2002; Kang et al. 2012). As in this study trace metals were found in the water, crabs can easily absorb these components passively, being potential bioaccumulators.

Certain metals are indisputably harmful to organisms and their effects are visible at different biological levels, which have already been discussed in many studies (Chiodi Boudet et al. 2015; Frías-Espericueta et al. 2022). In general, the damage depends on the concentration, exposure time and its properties. Frequent exposure to zinc, for example, causes histological and molecular damage (Gestin et al. 2022), which may explain a higher number of lesions in the regressive reaction pattern in both organs. Manganese, which was found in high concentrations in water, is described as harmful to the immune system of crustaceans, and these organisms are highly sensitive to this metal (Dey et al. 2023). In the environment it is difficult to specify which contaminant causes major disturbances in organisms, however some studies that have documented the presence of one or more metals bring some type of tissue impairment. Examples include lamellar fusion and disorganization, abnormal tubules and lumen, necrosis and impairment of the epithelial layer (Chiodi Boudet et al. 2015; Maisano et al. 2017; Rezaei Tavabe et al. 2019). Therefore, we can link the highest histological values to environmental contamination, as well as other anthropic stressors for the organism.

Figure 5. Index values for the gills (Ig) and hepatopancreas (Ih) of *Ucides cordatus* in P1 and P2. Different letters indicate statistically significant difference.



Monitoring studies of estuaries impacted by human activities, employing aquatic invertebrates, found significant responses using histological biomarkers (Maisano et al. 2017; Carvalho Neta et al. 2019; Joshy et al. 2022). Thus, the high values of biomarkers found in crabs from impacted sites in São Marcos Bay confirm that it is an excellent methodology for assessing contamination in estuarine environments.

Conclusion

Changes in biometry and histological damage seen in the gills and hepatopancreas of *U. cordatus* show that these organisms are under anthropic impacts, regardless of the seasonal period. As metals are available in the environment, it is concluded that biological damage may be linked to the harmful effects of these substances, which are in a complex mixture and can trigger antagonistic, additive or synergistic effects.

The native species *U. cordatus* was able to confirm that the sites are impacted and with high levels of stress for the organism, especially the P1 site. Thus, this methodology added to environmental analyzes is efficient to evidence the environmental degradation of estuarine environments.

Author's contributions: SRSO: Writing – original draft, Investigation, Methodology, Data curation, Project administration, Funding acquisition; LBO, LJSF, GSP: Conceptualization, Methodology; DMSS: Writing – review & editing, Supervision, Funding acquisition; LCGAIM: Writing – review & editing, Data curation, Supervision, Funding acquisition; RNFCN: Writing – review & editing, Conceptualization, Data curation, Supervision, Funding acquisition.

Ethical approval and Research authorization: The research has authorization for the collection of *Ucides cordatus* issued by SISBIO (registration n°76372-1).

Data availability: The data is not available in databases or repositories.

Funding: This work was supported by the Foundation for Research Support and Scientific and Technological Development of Maranhão - FAPEMA, through a research grant.

Conflict of interest: The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

References

Adeleke B, Robertson-Andersson D, Moodley G. 2021. Osmotic response of *Dotilla fenestrata* (sand bubbler crab) exposed to combined water acidity and varying metal (Cd and Pb). Heliyon. 7(4):e06763. doi:10.1016/j.heliyon.2021.e06763.

Amaral R, Alfredin P. 2010. Modelação Hidrossedimentológica no Canal de Acesso do Complexo Portuário do Maranhão. Revista Brasileira De Recursos Hídricos. 15(2):5–14. doi:10.21168/rbrh.v15n2.p5-14.

ANTAQ - Agencia Nacional de Transportes Aquaviários. 2021. Painel Estatístico Aquaviário 2.1.4. Brasília - DF. http://ea.antaq.gov.br/QvAJAXZfc/opendoc.htm?document=painel%5Cantaq - anuário 2014 - v0.9.3.qvw&lang=pt-BR&host=QVS%40graneleiro&anonymous=true.

Belitz HD, Grosch W, Schieberle P. 2009. Food Chemistry. 4th ed. Berlin, Heidelberg: Springer Berlin Heidelberg. doi: 10.1007/978-3-540-69934-7

Bernardino AF, Nóbrega GN, Ferreira TO. 2021. Consequences of terminating mangrove's protection in Brazil. Marine Policy. 125:104389. doi:10.1016/j.marpol.2020.104389.

Bernet D, Schmidt H, Meier W, Burkhardt-Holm P, Wahli T. 1999. Histopathology in fish: Proposal for a protocol to assess aquatic pollution. Journal of Fish Diseases. 22(1):25–34. doi:10.1046/j.1365-2761.1999.00134.x.

Bevan R, Ashdown L, McGough D, Huici-Montagud A, Levy L. 2017. Setting evidence-based occupational exposure limits for manganese. Neurotoxicology. 58:238–248. doi:10.1016/J.NEURO.2016.08.005.

Carvalho Neta RNF, Mota Andrade T de S de O, de Oliveira SRS, Torres Junior AR, da Silva Cardoso W, Santos DMS, dos Santos Batista W, de Sousa Serra IMR, Brito NM. 2019. Biochemical and morphological responses in *Ucides cordatus* (Crustacea, Decapoda) as indicators of contamination status in mangroves and port areas from northern Brazil. Environmental Science and Pollution Research. 2616. 26(16):15884–15893. doi:10.1007/S11356-019-04849-0.

Chiodi Boudet LN, Polizzi P, Romero MB, Robles A, Marcovecchio JE, Gerpe MS. 2015. Histopathological and biochemical evidence of hepatopancreatic toxicity caused by cadmium in white shrimp, *Palaemonetes argentinus*. Ecotoxicology and Environmental Safety. 113:231–240. doi:10.1016/j.ecoenv.2014.11.019.

CONAMA - CONSELHO NACIONAL DO MEIO AMBIENTE. 2005. Resolução CONAMA no 357, de 17 de março de 2005. http://www.siam.mg.gov.br/sla/download.pdf?idNorma=2747.

Corrêa JD, Farina M, Allodi S. 2002. Cytoarchitectural features of *Ucides cordatus* (Crustacea Decapoda) hepatopancreas: structure and elemental composition of electron-dense granules. Tissue and Cell. 34(5):315–325. doi:10.1016/S0040816602000101.

de Jesus WB, Mota Andrade T de S de O, Soares SH, Pinheiro-Sousa DB, de Oliveira SRS, Torres HS, Protazio G dos S, da Silva DS, Santos DMS, de Carvalho Neta AV, et al. 2021. Biomarkers and occurrences of heavy metals in sediment and the bioaccumulation of metals in crabs (*Ucides cordatus*) in impacted mangroves on the Amazon coast, Brazil. Chemosphere. 271:129444. doi:10.1016/j.chemosphere.2020.129444.

De Lima CDM, Da Silva HRC, Bernard E. 2018. Efetividade do defeso do caranguejo-uçá (*Ucides cordatus* L.): análise de percepção de consumidores e vendedores. Ambiente & Sociedade. 21:463. doi:10.1590/1809-4422ASOC0046R3VU18L1AO.

de Oliveira SRS, Batista W dos S, Sousa JBM, Noleto KS, Arouche Lima IM, Andrade TSOM, Cardoso W da S, Carvalho Neta RNF. 2019. Enzymatic and Histological Biomarkers in *Ucides cordatus* (Crustacea, Decapoda) in an Industrial Port on the North Coast of Brazil. Bulletin of Environmental Contamination and Toxicology. 102(6):802–810. doi:10.1007/s00128-019-02594-1.

Dey S, Tripathy B, Kumar MS, Das AP. 2023. Ecotoxicological consequences of manganese mining pollutants and their biological remediation. Environmental Chemistry and Ecotoxicology. 5:55–61. doi:10.1016/j.enceco.2023.01.001.

EMAP - Empresa Maranhense de Administração Portuária. 2022. Porto do Itaqui cresce 23% em 2021. https://www.portodoitaqui.com/imprensa/noticia/porto-do-itaqui-cresce-23-em-2021.

Ferreira AC, Lacerda LD. 2016. Degradation and conservation of Brazilian mangroves, status and perspectives. Ocean & Coastal Management. 125:38–46. doi:10.1016/j.ocecoaman.2016.03.011.

Frías-Espericueta MG, Bautista-Covarrubias JC, Osuna-Martínez CC, Delgado-Alvarez C, Bojórquez C, Aguilar-Juárez M, Roos-Muñoz S, Osuna-López I, Páez-Osuna F. 2022. Metals and oxidative stress in aquatic decapod crustaceans: A review with special reference to shrimp and crabs. Aquatic Toxicology. 242:106024. doi:10.1016/j.aquatox.2021.106024.

García-Rueda A, Tremblay N, Mascaró M, Díaz F, Paschke K, Caamal-Monsreal C, Rosas C. 2021. The thermal tolerance of a tropical population of blue crab (*Callinectes sapidus*) modulates aerobic metabolism during hypoxia. Journal of Thermal Biology. 102:103078. doi:10.1016/J.JTHERBIO.2021.103078.

Gestin O, Lopes C, Delorme N, Garnero L, Geffard O, Lacoue-Labarthe T. 2022. Organ-specific accumulation of cadmium and zinc in *Gammarus fossarum* exposed to environmentally relevant metal concentrations. Environmental Pollution. 308:119625. doi:10.1016/j.envpol.2022.119625.

González-Gorbeña E, Rosman PCC, Qassim RY. 2015. Assessment of the tidal current energy resource in São Marcos Bay, Brazil. Journal of Ocean Engineering and Marine Energy. 1(4):421–433. doi:10.1007/s40722-015-0031-5.

Gopalakrishnan G, Wang S, Mo L, Zou J, Zhou Y. 2020. Distribution determination, risk assessment, and source identification of heavy metals in mangrove wetland sediments from Qi'ao Island, South China. Regional Studies in Marine Science. 33:100961. doi:10.1016/J.RSMA.2019.100961.

Hussain S, Khan M, Sheikh TMM, Mumtaz MZ, Chohan TA, Shamim S, Liu Y. 2022. Zinc Essentiality, Toxicity, and Its Bacterial Bioremediation: A Comprehensive Insight. Frontiers in Microbiology. 0:1912. doi:10.3389/FMICB.2022.900740.

ICMBio - Instituto Chico Mendes de Conservação da Biodiversidade. 2018. Atlas dos Manguezais do Brasil. Brasília. http://www.icmbio.gov.br/portal/images/stories/manguezais/atlas_dos_manguezais_do_brasil.pdf.

INMET - Instituto Nacional de Meteorologia. 2021. Normais Climatológicas do Brasil (1991-2020). https://portal.inmet.gov.br/normais.

Jakovac CC, Latawiec AE, Lacerda E, Leite Lucas I, Korys KA, Iribarrem A, Malaguti GA, Turner RK, Luisetti T, Baeta Neves Strassburg B. 2020. Costs and Carbon Benefits of Mangrove Conservation and Restoration: A Global Analysis. Ecological Economics. 176:106758. doi:10.1016/J.ECOLECON.2020.106758.

Jerome FC, Hassan A, Omoniyi-Esan GO, Odujoko OO, Chukwuka AV. 2017. Metal uptake, oxidative stress and histopathological alterations in gills and hepatopancreas of *Callinectes amnicola* exposed to industrial effluent. Ecotoxicology and Environmental Safety. 139:179–193. doi:10.1016/j.ecoenv.2017.01.032.

Jinhui S, Sudong X, Yan N, Xia P, Jiahao Q, Yongjian X. 2019. Effects of microplastics and attached heavy metals on growth, immunity, and heavy metal accumulation in the yellow seahorse, *Hippocampus kuda* Bleeker. Marine Pollution Bulletin. 149. doi:10.1016/j.marpolbul.2019.110510.

Joshy A, Sharma SRK, Mini KG, Gangadharan S, Pranav P. 2022. Histopathological evaluation of bivalves from the southwest coast of India as an indicator of environmental quality. Aquatic Toxicology. 243:106076. doi:10.1016/J.AQUATOX.2022.106076.

Kang X, Mu S, Li W, Zhao N. 2012. Toxic Effects of Cadmium on Crabs and Shrimps. In: Acree B, editor. Toxicity and Drug Testing. InTech. p. 528. http://www.intechopen.com/books/toxicity-and-drug-testing/toxic-effects-of-cadmium-on-crabs-and-shrimps.

Kavun VY, Podgurskaya O V. 2009. Adaptation strategy of bivalve *Modiolus modiolus* from upwelling regions of the Kuril Islands shelf (Sea of Okhotsk) to heavy metal effects. Continental Shelf Research. 29(13):1597–1604. doi:10.1016/J.CSR.2009.05.001.

Legat JFA, Legat AP. 2009. Metodologia Para O Transporte De Caranguejo Vivo Com Baixos Índices De Desperdícios. Boletim Tecnico-Cientifico do Cepene. 17(1):115–121.

Liu M, Zhang H, Lin G, Lin H, Tang D. 2018. Zonation and Directional Dynamics of Mangrove Forests Derived from Time-Series Satellite Imagery in Mai Po, Hong Kong. Sustainability. 10(6):1913. doi:10.3390/SU10061913.

Luna LG. 1968. Manual of histologic staining methods of the Armed Forces Institute of Pathology. 3. ed. New York: McGraw-Hill.

Maisano M, Cappello T, Natalotto A, Vitale V, Parrino V, Giannetto A, Oliva S, Mancini G, Cappello S, Mauceri A, et al. 2017. Effects of petrochemical contamination on caged marine mussels using a multi-biomarker approach: Histological changes, neurotoxicity and hypoxic stress. Marine Environmental Research. 128:114–123. doi:10.1016/J.MARENVRES.2016.03.008.

Mattison DR, Milton B, Krewski D, Levy L, Dorman DC, Aggett PJ, Roels HA, Andersen ME, Karyakina NA, Shilnikova N, Ramoju S, McGough D. 2017. Severity scoring of manganese health effects for categorical regression. Neurotoxicology. 58:203–216. doi:10.1016/J.NEURO.2016.09.001.

Monteiro MC, Jiménez JA, Pereira LCC. 2016. Natural and human controls of water quality of an Amazon estuary (Caeté-PA, Brazil). Ocean & Coastal Management. 124:42–52. doi:10.1016/J.OCECOAMAN.2016.01.014.

Nordhaus I, Wolff M, Diele K. 2006. Litter processing and population food intake of the mangrove crab *Ucides cordatus* in a high intertidal forest in northern Brazil. Estuarine, Coastal and Shelf Science. 67(1–2):239–250. doi:10.1016/J.ECSS.2005.11.022.

de Oliveira SRS, Batista W dos S, Sousa JBM, Noleto KS, Arouche Lima IM, Andrade TSOM, Cardoso W da S, Carvalho Neta RNF. 2019. Enzymatic and Histological Biomarkers in *Ucides cordatus* (Crustacea, Decapoda) in an Industrial Port on the North Coast of Brazil. Bulletin of Environmental Contamination and Toxicology. 102(6):802–810. doi:10.1007/s00128-019-02594-1.

Pinheiro-Sousa DB, da Costa Soares SH, Torres HS, de Jesus WB, de Oliveira SRS, Bastos WR, de Oliveira Ribeiro CA, Carvalho-Neta RNF. 2021. Sediment contaminant levels and multibiomarker approach to assess the health of catfish *Sciades herzbergii* in a harbor from the northern Brazilian Amazon. Ecotoxicology and Environmental Safety. 208:111540. doi:10.1016/J.ECOENV.2020.111540.

Pinheiro JM. 2015. Dinâmica climática da ilha do maranhão. In: Farias Filho MS, Celeri MJ (Eds). Geografia da Ilha do Maranhão. 1st ed. São Luís: EDUFMA. p. 29–37. https://docero.com.br/doc/ne818s1.

Polidoro BA, Carpenter KE, Dahdouh-Guebas F, Ellison JC, Koedam NE, Yong JWH. 2014. Global patterns of mangrove extinction risk: implications for ecosystem services and biodiversity loss. In: Maslo B, Lockwood JL (Eds). Coastal Conservation. Cambridge University Press. p. 15–36.

Pülmanns N, Diele K, Mehlig U, Nordhaus I. 2014. Burrows of the Semi-Terrestrial Crab *Ucides cordatus* Enhance CO2 Release in a North Brazilian Mangrove Forest. PLoS One. 9(10):e109532. doi:10.1371/JOURNAL.PONE.0109532.

Rainbow P. S. 2018. Heavy Metal Levels in Marine Invertebrates. In: Furness RW, Rainbow Philip S. (Eds). Heavy Metals in the Marine Environment. 1st ed. CRC Press. p. 67–79. https://www.taylorfrancis.com/chapters/edit/10.1201/9781351073158-5/heavy-metal-levels-marine-invertebrates-rainbow.

Rezaei Tavabe K, Pouryounes Abkenar B, Rafiee G, Frinsko M. 2019. Effects of chronic lead and cadmium exposure on the oriental river prawn (*Macrobrachium nipponense*) in laboratory conditions. Comparative Biochemistry and Physiology Part C: Toxicology. 221:21–28. doi:10.1016/j.cbpc.2019.03.009.

Ribeiro EB, Noleto KS, de Oliveira SRS, Batista de Jesus W, de Sousa Serra IMR, da Silva de Almeida Z, de Sousa de Oliveira Mota Andrade T, de Araújo Soares R, Antonio ÍG, Santos DMS, et al. 2020. Biomarkers (glutathione S-transferase and catalase) and microorganisms in soft tissues of *Crassostrea rhizophorae* to assess contamination of seafood in Brazil. Marine Pollution Bulletin. 158:111348. doi:10.1016/J.MARPOLBUL.2020.111348.

Ribeiro I, Castro ACL de. 2017. Pescadores artesanais e a expansão portuária na praia do Boqueirão, Ilha de São Luís-MA. Revista de Políticas Públicas. 20(2):863. doi:10.18764/2178-2865.v20n2p863-884.

Rocha AA, Pereira DN, De Padua HB. 1985. Fishing yield and chemical contamination of the water of the Billings Reservoir, S. Paulo (Brazil). Revista de Saude Publica. 19(5):401–410. doi:10.1590/s0034-89101985000500003.

dos Santos CCM, da Costa JFM, dos Santos CRM, Amado LL. 2019. Influence of seasonality on the natural modulation of oxidative stress biomarkers in mangrove crab *Ucides cordatus* (Brachyura, Ucididae). Comparative Biochemistry and Physiology Part A: Molecular & Integrative Physiology. 227:146–153. doi:10.1016/J.CBPA.2018.10.001.

Sousa DBP, Almeida ZS, Carvalho-Neta RNF. 2013. Biomarcadores histológicos em duas espécies de bagres estuarinos da Costa Maranhense, Brasil. Arquivo Brasileiro de Medicina Veterinária e Zootecnia. 65(2):369–376. doi:10.1590/S0102-09352013000200011.

Summer K, Reichelt-Brushett A, Howe P. 2019. Toxicity of manganese to various life stages of selected marine cnidarian species. Ecotoxicology and Environmental Safety. 167:83–94. doi:10.1016/j.ecoenv.2018.09.116.

Vásquez-Procopio J, Osorio B, Cortés-Martínez L, Hernández-Hernández F, Medina-Contreras O, Riós-Castro E, Comjean A, Li F, Hu Y, Mohr S, et al. 2020. Intestinal response to dietary manganese depletion in Drosophila. Metallomics. 12(2):218–240. doi:10.1039/C9MT00218A.

Walker JE, Ankersen T, Barchiesi S, Meyer CK, Altieri AH, Osborne TZ, Angelini C. 2022. Governance and the mangrove commons: Advancing the cross-scale, nested framework for the global conservation and wise use of mangroves. Journal of Environmental Management. 312:114823. doi:10.1016/J.JENVMAN.2022.114823.

Wang H, Wei H, Tang L, Lu J, Mu C, Wang C. 2018. A proteomics of gills approach to understanding salinity adaptation of *Scylla paramamosain*. Gene. 677:119–131. doi:10.1016/J.GENE.2018.07.059.

Wang X, Yao Q, Zhang D ming, Lei X yu, Wang S, Wan J wu, Liu H jian, Chen Y ke, Zhao Y long, Wang G qin, et al. 2022. Effects of acute salinity stress on osmoregulation, antioxidant capacity and physiological metabolism of female Chinese mitten crabs (*Eriocheir sinensis*). Aquaculture. 552:737989. doi:10.1016/J.AQUACULTURE.2022.737989.

Yang L, Wang D, Xin C, Wang L, Ren X, Guo M, Liu Y. 2021. An analysis of the heavy element distribution in edible tissues of the swimming crab (*Portunus trituberculatus*) from Shandong Province, China and its human consumption risk. Marine Pollution Bulletin. 169:112473. doi:10.1016/J.MARPOLBUL.2021.112473.