

An Acad Bras Cienc (2024) 96(4): e20240226 DOI 10.1590/0001-3765202420240226

Anais da Academia Brasileira de Ciências | Annals of the Brazilian Academy of Sciences Printed ISSN 0001-3765 | Online ISSN 1678-2690 www.scielo.br/aabc | www.fb.com/aabcjournal

MICROBIOLOGY

Toxicological risks assessment in the Jaguaribe River watershed (Ceará, Brazil) using anthropogenic contamination reports and ecotoxicological analysis

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Abstract: The economic development of human activities contributes to the discharge of many anthropogenic pollutants. To assess the environmental risks in the Jaguaribe River, the most important river in the hydrographic region of the Eastern Northeast Atlantic, a bibliographic review of scientific articles and a series of ecotoxicological bioassays were conducted. The bioassays were conducted using sediment samples at six collection sites along the river, while the bibliographic review was used to identify the presence of anthropogenic contaminants in sediment and tissue samples of aquatic organisms within two km of each of the sediment collection sites. The bibliographic review showed the presence of thirty-eight anthropogenic pollutants in sediment samples and seven in tissue samples of aquatic organisms. The ecotoxicological bioassays showed that the sediment samples produced lethal and sublethal effects in the four tested representatives of the different trophic levels: Daphnia magna, Artemia salina, Allium cepa and Cucumis sativus. The presence of multiple anthropogenic pollutants in the Jaguaribe River and the observed lethal and sublethal effects in ecotoxicological bioassays suggest potential risks not only to the aquatic ecosystem but also to human health. Humans may be exposed to these contaminants through the consumption of water and aquatic organisms, leading to potential health issues such as increased cancer risk. The findings underscore the urgent need for regular monitoring and effective pollution control measures to mitigate these health risks and protect the well-being of local communities.

Key words: bioassays, ecotoxicity tests, environmental danger, environmental health.

INTRODUCTION

Aquatic ecosystems make up most of planet Earth and have been largely degraded by anthropic actions for economic development, especially in coastal regions. These regions are critical for flood control, soil carbon sequestration, filtering out persistent pollutants, and water supply. However, pollution of these ecosystems by anthropogenic pollutants is one of the most serious problems today due to its acute and chronic effects on human, animal, and plant health (López-Pacheco et al. 2019).

Pollutants can come from different pollution sources, such as aquaculture, agriculture, industry, livestock, shipping, tourism, and urban runoff. However, their effects are harmful to the environment and can be felt by all the economic sectors involved (Soares et al. 2020). Normally, aquatic organisms absorb pollutants during their life and transfer them through the trophic position. For this reason, many studies link the improper use of chemicals with adverse health effects and cancer (Arisekar et al. 2020, Cui et al. 2015).

The chronic and acute effects of these chemical byproducts on ecosystems can be evaluated through ecotoxicological studies with bacteria, microcrustaceans, fish, and vegetables (Wang 2018). These studies evaluate the interactions between contaminants and lethality they cause in the organisms tested after different exposure times and concentrations (Hoffman et al. 2003). However, these methods are increasingly problematic as the effects of these contaminants are exacerbated by increasing environmental degradation and accelerating climate change (Tlili & Mouneyrac 2021).

In the natural environment, anthropogenic pollutants are deposited in sediments as a function of water flow regulation and tend to have combined adverse effects on terrestrial and aquatic organisms (Santos et al. 2022). For this reason, sediment toxicity analysis is one way to understand the adverse effects that chemical residues that are not commonly measured or are unknown may have on the environment and humans (Heise et al. 2020).

In semi-arid regions, which tend to be more vulnerable to climate change, the effects of various pollutants may be even more harmful (Fernandes et al. 2020). Many scientific studies focus on the presence of anthropogenic pollutants in the Brazilian semi-arid region, and the area that most of these studies focus on is the Jaguaribe River watershed, located in the hydrographic region of the eastern Northeast Atlantic. Pollutants already studied in the region include: antibiotics (Rebouças et al. 2011), pesticides (Oliveira et al. 2016, Soares et al. 2020), herbicides (Gama et al. 2017), polycyclic aromatic hydrocarbons (Andrade et al. 2019), inorganic phosphorus compounds (Barcellos et al. 2019, Marins et al. 2011, 2020), natural and

synthetic hormones (Lima et al. 2019), crude oil (Magris & Giarrizzo 2020), microplastics (Garcia et al. 2020), and toxic metals (Costa et al. 2013, Costa & Lacerda 2014, Lacerda et al. 2013, 2009, Moura & Lacerda 2018, Rios et al. 2016). In this way, the aim of present study is evaluating the toxicological risks in the Jaguaribe River watershed (Ceará, Brazil) using anthropogenic contamination reports and ecotoxicological analyses. To do it, has been done a bibliographic review of the pollutants already studied in the watershed of the Jaguaribe River and bioassays to determine the ecotoxicological effects of the river sediment on survive of saltwater (Artemia salina) and freshwater (Daphnia magna) microcrustaceans, and on seed germination of cucumber (Cucumis sativus) and onion (Allium сера).

MATERIALS AND METHODS Study area

The Jaguaribe River is 633 km long and is used for various economic activities in the Brazilian semi-arid region, including livestock, agriculture, aquaculture, fishing, tourism, trade, and navigation. It is used for various economic activities, especially in areas closer to the sea, and has many dams along its course to combat droughts in the state of Ceará (IBGE 1999). Along the Jaguaribe River, six sampling sites with different compositions of areas with urban infrastructure, native vegetation, agriculture, pasture, aquaculture, and dams were selected for sediment toxicity analysis (MAPBIOMAS 2022), as shown in Figure 1.

Collection site P1 is closer to the sea, its surroundings are mainly forest and mangrove areas, and is used for agriculture and livestock. P2 consists of agricultural land, pasture land, and other areas that are not forest (including salt flat and rocky outcrop). P3 is the area that

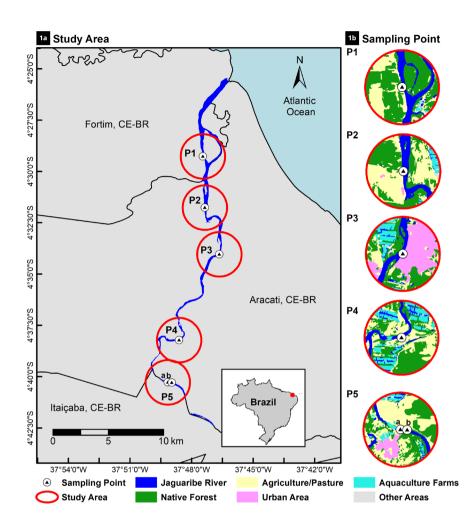


Figure 1. Geographic location of the study area with indication of sampling sites Geographic data from the Instituto de Pesquisa e Estratégia Econômica do Ceará (IPECE 2023) and Collection 7 of annual land cover and land use maps of Brazil (MAPBIOMAS 2022).

receives the greatest amount of nutrients from wastewater, as it is located within the city of Aracati. P4 is mostly composed of aquaculture farms, one of the activities that contribute the most to the destruction of mangrove ecosystems. Point P5 was divided into two points: P5a and P5b. They have a similar proportion of land with urban infrastructure, vegetation, aquaculture, agriculture, and livestock, but are separated by a dam that carries saltwater on one side of the river (P5a) and freshwater on the other (P5b). Table I summarizes key environmental information for each sediment collection site.

Bibliographic review

The bibliographic review was conducted in English based on scientific articles from Scopus to identify scientific reports of anthropogenic pollution in environmental matrices (water, sediment and aquatic organisms) along the Jaguaribe River (Ceará, Brazil). Thus, the search configuration used was as follows: (ALL(jaguaribe AND river) AND ALL(contaminant) OR ALL(pollutant) OR ALL(microplastic) OR ALL(pesticide) OR ALL(herbicide) OR ALL(hydrocarbons) OR ALL(antibiotic)) AND (LIMIT-TO (AFFILCOUNTRY,"Brazil")) AND (LIMIT-TO (DOCTYPE,"ar")) on October 16, 2022.

The selection criteria for inclusion in the study were scientific articles that used

Point	Salinity (ppm)ª	Water Temperature (°C) ^b	Air Temperature (°C) ^b	Annual Precipitation (mm) ^c	Aridity Index °	Environmental Factor
P1	42	31	30	903.2	49.55	Mangrove
P2	45	30	29	859.8	47.03	Agriculture/Pasture
P3	52	30	35	859.8	47.03	Urban Area
P4	48	31	33	859.8	47.03	Aquaculture
P5a	48	30	35	675.3	36.89	Dam
P5b	1	31	34	675.3	36.89	Dam

Table I. Environmental information of the sampling site.;

(a) observed in the field with a portable salinity meter; (b) observed in the field with a thermometer; (c) information obtained from the Fundação Cearense de Meteorologia e Recursos Hídricos.

standardized methods to detect anthropogenic contaminants in sediment or tissue of aquatic organisms within 2 km of collection sites. These articles were a source of information on: the presence of detected contaminants, concentrations (ng/g), and geographic location.

Ecotoxicological bioassays

Sampling and experimental design

At each of the sampling sites, surface sediment samples were collected from the banks of the Jaguaribe River at depths up to 10 cm. Collections took place in November 2021, the collected material was stored in sterile plastic bags, and analyzes were conducted within 24 hours of material collection. Prior to analyzes, sediment samples were sieved to remove shells, stones, and other components larger than 100 mm.

Sediment samples were tested using ecotoxicological tests with microcrustaceans from freshwater (*Daphnia magna*), saltwater (*Artemia salina*), onion (*Allium cepa*), and cucumber (*Cucumis sativus*). The experimental design was randomized with three replicates. The advantage of bioassays with organisms from different trophic levels is that they provide high accuracy in ecotoxicity testing and a holistic view of how toxic and non-toxic components of an environmental matrix affect the trophic position (Urbaniak et al. 2020).

In each test, three sediment concentrations were tested for each test organism, following the guidelines of technical standards NBR 12713 (ABNT 2022), NBR 16530 (ABNT 2021), ASTM E1706 (ASTM 2020) for aquatic ecotoxicity tests with invertebrate organisms, and technical standard EPA 712-C-012 (EPA 2012) for tests with plant seeds. In this way, five different treatments were tested: Positive Control (PC), Negative Control (NC) and three treatments (125 g/L, 250 g/L and 500 g/L), where 250 g/L is the standard concentration of grams of sediment per liter recommended by the ASTM E1706 technical standard for ecotoxicological analysis of sediments (ASTM 2020).

Toxicity tests with aquatic organisms

Toxicity tests with aquatic organisms were performed with freshwater (*Daphnia magna*) and saltwater (*Artemia salina*) microcrustaceans. These organisms were selected for this study because they are commonly used in ecotoxicological bioassays with sediment samples and because they are ecologically relevant, sensitive, and easy to handle in the laboratory (Gambardella et al. 2022). Furthermore, the microcrustaceans used in this study are also part of the aquatic biota of the Jaguaribe River watershed (Camara 2020, Diniz et al. 2020).

Freshwater microcrustaceans (Daphnia magna) were acquired as neonates from a commercial aquaculture facility. These neonates were cultured in a system with constant aeration, light and dark regime (16:8) and feeding with green algae and Saccharomyces cerevisiae. The Daphnia magna neonates used in the tests were individuals between 2 and 26 hours of age and were obtained from females between 10 and 15 days of age. Meanwhile, saltwater microcrustaceans (Artemia salina) in the form of cysts were acquired from a commercial aquarium facility. These cysts were incubated for 24 hours under aeration and constant lighting in filtered and sterilized seawater (15 ppt). Artemia salina neonates used in the tests were individuals that were at nauplius stage I and II and were between 2 and 26 hours old.

During the experiment, the microcrustaceans were kept in a static system with a 12-hour photoperiod in diffuse light, an ambient temperature of 27 ± 2°C, and no feeding or aeration for 48 hours. The sediments to be tested were diluted with the same water in which the microcrustaceans were cultured, and each experimental unit received ten live organisms after 2 hours of decantation. Prior to the toxicity tests, all microcrustaceans were visually inspected to select organisms with the same size and swimming behavior. In this case, PC was only distilled water without sediment adding and NC was only the water used during cultivation without sediment adding.

Toxicity tests with terrestrial organisms

Toxicity tests with terrestrial organisms were performed with onions (*Allium cepa*) and

cucumbers (*Cucumis sativus*). These organisms were selected based on their wide use in ecotoxicological studies with sediment samples, commercial availability, short growth time, low acquisition cost, and sensitivity to sediment cytotoxic and genotoxic effects (Wijeyaratne & Wadasinghe 2019).

Onion (Allium cepa) and cucumber (Cucumis sativus) seeds were purchased from a commercial nursery and sterilized in sodium hypochlorite solution (1%) for 10 min, then washed several times with distilled water, and then air dried. After the sterilization process, 10 seeds were aseptically placed in sterilized Petri dishes (13 × 13 cm) lined with filter paper (Whatman nº4). The filter papers were moistened with 15 mL of a solution prepared from dilutions of the sediment in drinking water. The Petri dishes were kept in a closed environment at temperatures of 27/25 °C (day/night) for germination. In this case, PC was distilled water with 0.5% NaCl and NC was drinking water.

Seeds were considered germinated only when the seed coat was broken, the radicle was visible, and measured more than 2 mm. Seed manufacturers indicated on the package label that the average germination time for onion seeds was 7 days and for cucumber seeds was 5 days. So, this was the period used for counting the germinated seeds and the size of their roots.

Statistical analysis and multivariate approach

At the end of the period during which the organisms were exposed to the different sediment concentrations (PC, 125 g/L, 250 g/L, 500 g/L and NC), the percentage of microcrustacean mortality (MR = number of dead organisms/10 × 100), the percentage of seed germination (GR = number of germinated seeds/10 × 100), and the average root size of germinated seeds were determined. The values obtained were subjected to the Shapiro-Wilk normality test and analysis of variance, according to the normality of the samples, to determine a significant difference between the 3 treatments (N = 30 seeds/ treatment; N = 30 microcrustaceans/treatment), between the 3 treatments and the 2 controls (positive and negative with the same N as the treatments) and between the 6 collection sites (P1, P2, P3, P4, P5a and P5b).

Sediment toxicity was evaluated using the toxicity index calculated according to the formula ST = $[(CT) / C] \times 100$, where: C is the analyzed parameter (microcrustacean mortality, seed germination, or root/shoot length) in the control and T is the analyzed parameter in the treatment (Nikolaeva et al. 2019). Regression analysis was used to estimate the median effective concentration (EC 50) or median lethal concentration (LC 50) that can cause 50% mortality of microcrustaceans and the median inhibitory concentration (IC 50) that can inhibit 50% germination of plant seeds (lethal effect) or affect root size of germinated seeds (sublethal effect).

The sensitivity of the organisms to the toxicity of the sediment was compared using toxic units (TU = 100/ EC 50), where the sensitivity of the organism and the toxicity of the sediment are proportional to the number of toxic units. The toxic units approach (TU) is based on a model that estimates the cumulative effect of toxicity for the test organisms and can be used for analyzes of sediment, chemicals, and metals (Castro-Català et al. 2016).

RESULTS

Bibliographic review

As a result of the bibliographic review, 139 scientific articles published between 2009 and 2022 were reviewed and analyzed. Seventeen of them were eligible for this study because they reported anthropogenic contamination within 2 km of each of the sampling sites. According to these articles, the main sources of anthropogenic contamination in sediments and biological matrices were agriculture, aquaculture, livestock, and untreated urban wastewater. Site P1 had more data on anthropogenic contaminants, while P4 had the lowest number of reports of contaminants.

Table II summarizes the anthropogenic pollutants found in the tissues of aquatic organisms (ng/g), of two types: pesticide metabolites (Santana et al. 2020) and toxic metals (Costa & Lacerda 2014, Lacerda et al. 2009, Moura & Lacerda 2018, 2022, Rios et al. 2016). Other authors detected the presence of resistance genes to ampicillin (10 mg), aztreonam (30 mg) and oxytetracycline (30 mg) in bacteria of the genus Vibrio isolated from shrimp samples in the study region (Rebouças et al. 2011) and the presence of 0.31 pieces/m3 of microplastic particles in plankton samples collected with a 120 μ m trawl (Garcia et al. 2020).

Table III summarizes all contaminants (ng/g) detected in the sediment, of which there are five types: herbicides (Gama et al. 2017), organochlorine pesticides (Oliveira et al. 2016), polycyclic aromatic hydrocarbons (Andrade et al. 2019), toxic metals (Costa et al. 2013, Dias et al. 2013, Lacerda et al. 2013), and inorganic phosphorus components (Barcellos et al. 2019, Marins et al. 2011, 2020). These chemicals can be classified in the following order of total concentration in sediment samples: Polycyclic aromatic hydrocarbons > Herbicides > Organochlorine pesticides > Inorganic phosphorus components > Toxic metals.

Two of the articles studied did not indicate the exact geographic location of their sediment collection sites. These authors detected the presence of mercury (7.8 - 93 ng/g) (Lacerda et al. 2013), copper (1.7 - 21 μ g/g), zinc (0.4 - 8.9 mg/g), iron (4.6 - 51.4 mg/g), and aluminum

Туре	Contaminant	Organism	P1	P2	P3	P4	P5a-b
Pesticide metabolite ^ª	CarbPhenol	Omnivorous Fish	3.06	-	-	-	-
	Malaox	Omnivorous Fish	6.04	-	-	-	-
	3-PBA	Omnivorous Fish	20.67	-	-	-	-
	Copper ^b	Shrimp	-	2460	_	6360	-
	Mercury ^c	Oyster	75.05	-	-	-	-
	Mercury ^d	Detritivorous Fish	-	-	4	4	-
		Omnivorous Fish	49.2	49.2	12.03	12.03	_
cals		Carnivorous Fish	25.09	25.09	32.85	32.85	_
Toxic metals	Mercury ^e	Detritivorous Fish	18.00	18.00	-	19.83	19.83
Toxi		Omnivorous Fish	45.97	45.97	-	44.00	44.00
		Carnivorous Fish	39.00	39.00	-	88.00	88.00
		Crab	32.33	32.33	-	110.36	110.36
		Shrimp	11.00	11.00	-	16.00	16.00
-		Shellfish	-	-	-	51.45	51.45
	Mercury ^f	Omnivorous Fish	51	57	33		30
		Carnivorous Fish	101.3				49.2
		Shrimp	13.3				10.7

Table II. Anthropogenic pollutants (ng/g)	detected in tissues of	f aquatic organisms within 2 km of collection sites.
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References: (a) Santana et al. 2020; (b) Lacerda et al. 2009; (c) Rios et al. 2016; (d) Costa & Lacerda, 2014; (e) Moura & Lacerda, 2018; (f) Moura & Lacerda 2022.

(6.7 - 47.3 mg/g) (Dias et al. 2013) in the area corresponding to sites P1, P2, and P3 when they took sediment samples.

Ecotoxicological bioassays

Ecotoxicological bioassays showed that sediment from site P5a had the highest toxicity (< EC_{50}) to aquatic organisms (*Artemia salina* -A and *Daphnia magna* -B), while P5b had the lowest toxicity (Figure 2). The EC_{50} for freshwater microcrustaceans (*Daphnia magna*) ranged from 125 to 181.7 g of sediment per liter, while saltwater (*Artemia salina*) values ranged from 211.25 to 286.48 g /L. In the ecotoxicological analyzes using seeds, the sediment showed lethal and sublethal effects for onion (*Allium cepa*), as shown in Figure 3. Sediment from collection site P5a had the highest toxicity to seed germination (IC 50 = 39.33 g/L), while P3 had the lowest toxicity (IC 50 = 200.28 g/L). For sublethal effects, sediment on P3 showed the highest inhibition of root size (IC 50 = 59.38 g/L), while P2 was the lowest (IC 50 = 323.11 g/L).

In the case of cucumber (*Cucumis sativus*), some concentrations of sediment resulted in higher seed germination than the control, as shown in Figure 4. Although sediment has no

Туре	Contaminant	P1	P2	P3	P4	P5a-b
	Alachlor	17.42	-	-	8.27	3.65
e a	Bromacil	33.84	-	-	31.49	38.54
cide	Ethalfluralin	407.55	-	-	302.96	371.97
Herbicide ^a	Fluridone	0.80	-	-	0.14	0.21
Ť	Norflurazon	0.15	-	-	0.04	0.05
	Tebuthiuron	7.76	-	-	2.53	5.32
	Σ Total	467.52			345.45	419.73
<u>م</u>	НСВ	1.31	1.82	4.25	-	2.35
cide	Heptachlor	16.69	51.89	51.40	-	24.29
esti	Methoxychlor	-	2.95	8.51	-	-
Organochlorine Pesticide	p,p-DDD	2.42	1.61	3.68	-	1.35
lorir	p,p-DDE	3.11	2.83	5.15	-	1.96
ochl	p,p-DDT	3.45	2.98	6.68	-	2.45
gano	α-Endosulfan	69.30	87.20	136.70	-	45.67
Ő	ү-НСН	0.58	3.15	2.79	-	1.38
	ΣTotal	96.86	154.43	219.16		79.45
	Acenaphthene	1.90	3.30	6.30	-	3.90
	Acenaphthylene	0.20	1.60	0.80	-	0.60
	Anthracene	9.60	11.20	14.65	-	12.50
	Benz[a]anthracene	0.20	6.20	7.50	-	0.10
S	Benzo[a]pyrene	4.50	12.20	11.00	-	3.40
pon	Benzo[b]fluoranthene	14.60	4.10	21.70	-	0.02
car	Benzo[e]pyrene	4.70	12.20	15.40	-	1.20
ydro	Benzo[ghi]perylene	67.20	0.80	2.60	-	6.00
Polycyclic Aromatic Hydrocarbons ^c	Benzo[k]fluoranthene	1.50	0.20	6.80	-	1.30
nati	Chrysene	19.70	52.30	61.10	-	21.50
Aror	Dibenz[ah]anthracene	0.06	3.50	5.21	-	0.30
clic	Fluoranthene	1614.80	1801.20	2254.00	-	25.30
ycyd	Fluorene	0.03	13.70	16.50	-	9.00
Pol	Indeno[1,2,3-cd]pyrene	20.00	6.50	4.50	-	3.30
	Naphtalene	0.04	0.04	0.13	-	0.10
	Perylene	1.40	1.70	10.40	-	0.30
	Phenanthrene	549.60	733.10	1160.50	-	425.20
	Pyrene	246.90	649.00	1075.00	-	536.20
	ΣTotal	2556.93	3312.84	4674.09		1050.22
	Mercury ^d	13.1 ^d	-	-	-	-
Other	,	6.91 ^e	7.65 ^e	8.72 ^e	-	7.45 ^e
Ò	Inorganic Phosphorus	213.6 ^f	-	-	-	-
	<u> </u>	6.9E+07 ^g	1.01E+08 ^g	-	-	_

Table III. Anthropogenic contaminants (ng/g) detected in sediment within 2 km of sampling sites.

References: (a) Gama et al. 2017; (b) Oliveira et al. 2016; (c) Andrade et al. 2019; (d) Costa et al. 2013; (e) Moura & Lacerda 2022; (f) Barcellos et al. 2019; (g) Marins et al. 2011.

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lethal effects, there are sublethal effects in terms of root growth. Sediment from collection site P5a showed the highest toxicity to root growth (IC 50 = 36.91 g/L), while P3 showed the lowest (IC 50 = 258.88 g/L).

Analysis of the relative sensitivity of organisms tested in toxic units showed that onion seeds and saltwater crustaceans were more sensitive to the toxicity of Jaguaribe River sediment than cucumber seeds and freshwater microcrustaceans (Table IV).

Analysis of variance showed no significant difference between sites, but there was a significant difference between concentrations and the negative control and between organisms (Figure 5). Among aquatic organisms, the mortality rate of freshwater microcrustaceans was significantly higher than that of saltwater organisms. For terrestrial organisms, there was a significant difference between seed germination rates, but no difference between root size inhibition rates. Detailed results of the statistical tests can be found in Table SI (Supplementary Material – Table SI).

DISCUSSIONS

In this study, we assessed toxicological risks in the Jaguaribe River watershed (Ceará, Brazil) using reports of anthropogenic contamination and ecotoxicological bioassays. In this way, we performed a bibliographic review of anthropogenic contaminants detected in the Jaguaribe River and evaluated the acute toxicity of the river sediment to aquatic and terrestrial organisms, including *Daphnia magna* (freshwater microcrustacean), *Artemia salina* (saltwater microcrustacean), *Allium cepa* (onion seed) and *Cucumis sativus* (cucumber seed).

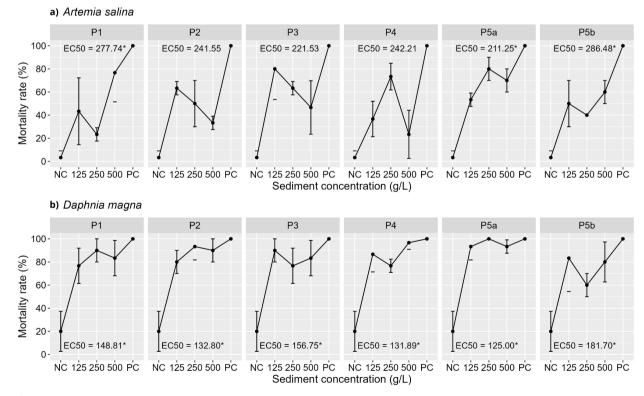


Figure 2. Mortality rate of aquatic organisms per sampling site at different sediment concentrations *** Indicates** that the EC50 regression is significant (p value < 0.05).

Polycyclic aromatic hydrocarbons were the most detected anthropogenic pollutants in the Jaguaribe River sediment samples, with emphasis on fluoranthene, phenanthrene, and pyrene, which had the highest concentrations compared to other pollutants (Andrade et al. 2019). These pollutants are present in the atmosphere, aquatic and terrestrial systems and have been described as genotoxic, mutagenic, carcinogenic and/or teratogenic (Adeniji et al. 2019).

Meanwhile, pesticide residues and toxic metals were the most common contaminants found in the tissues of aquatic organisms, especially mercury, which was the most abundant (Costa & Lacerda 2014, Moura & Lacerda 2018, Rios et al. 2016, Santana et al. 2020). Like other toxic metals, mercury is toxic, has carcinogenic potential, and can accumulate (Kadim & Risjani 2022). The combined effect of these pollutants has already been observed in aquatic organisms and on the life cycle of *Daphnia magna* they have several negative effects, including the time to production of first brood, brood size and total number of live offspring per female (Caixeta et al. 2022). In this way, the present study also indicates that the combined effect of these pollutants is also lethal to *Daphnia magna* and *Artemia salina*.

According to the studies that have evaluated the pollution of the Jaguaribe River by polycyclic aromatic hydrocarbons, pesticides and herbicides, the pollutants found come from aquaculture, agriculture, livestock, navigation and urban runoff, making it impossible to identify a single source responsible for each type of pollution (Andrade et al. 2019, Barcellos et al. 2019, Costa et al. 2013, Gama et al. 2017,

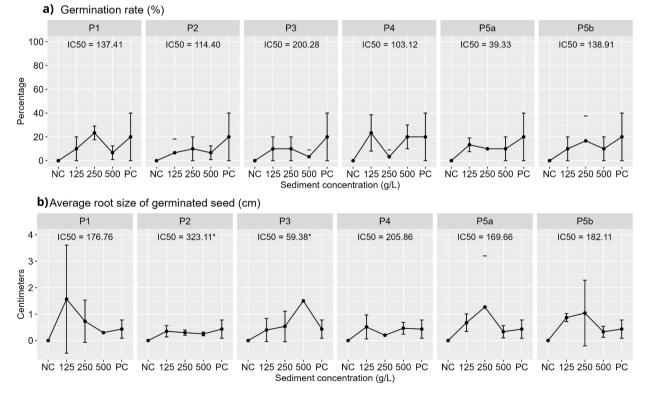


Figure 3. Germination rate of onion (*Allium cepa*) seeds and average root size of germinated seeds per sampling site at different sediment concentrations.

* Indicates that the EC50 regression is significant (p value < 0.05).

Oliveira et al. 2016). According to studies that estimated the contribution of human activities to the disposal of toxic metals in the Jaguaribe River, aquaculture, wastewater, and solid waste contribute the most to mercury emissions, emitting 200, 400, and 175 mg of mercury per hectare/year, respectively (Lacerda et al. 2011).

Compared to other river watershed in the state of Ceará, the Jaguaribe River watershed is the one with the most dams and is the most

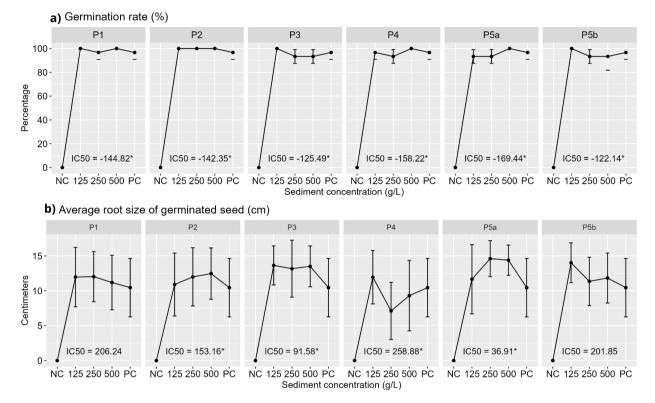


Figure 4. Germination rate of cucumber (*Cucumis sativus*) seeds and average root size of germinated seeds per sampling site at different sediment concentrations.

* Indicates that the EC50 regression is significant (p value < 0.05).

Table IV. Relative sensitivity of organisms tested in toxic units (TU).

	Mortality		Germination		Root size	
Sample Point	Artemia	Daphnia	Cucumis	Allium	Cucumis	Allium
P1	0.488	0.264	-0.691	0.728	0.485	0.566
P2	0.625	0.302	-0.703	0.874	0.653	0.309
P3	0.548	0.305	-0.797	0.499	1.092	1.684
P4	0.476	0.296	-0.632	0.970	0.386	0.486
P5a	0.552	0.332	-0.590	2.543	2.709	0.589
P5b	0.370	0.254	-0.819	0.720	0.495	0.549
Average	0.510	0.292	-0.705	1.056	0.970	0.697
Std. Deviation	0.09	0.03	0.09	0.75	0.89	0.49

affected by drought due to its lower rainfall (Freire et al. 2021). In this way, anthropogenic pollutants tend to deposit more easily in the sediment because inorganic substances are more retained (Cavalcante et al. 2021) and the transport of substances to the ocean decreases significantly during periods of drought (Dias et al. 2016). As a result, pollutants are detected more frequently and in higher concentrations in the tissues of aquatic organisms over the years (Santos et al. 2022), as shown by the extensive list of pollutants detected near the collection sites.

The water flow of rivers provides the ecosystem services of control, transport, and biotransformation of chemical residues. Therefore, it is only natural that the concentration and toxicity of anthropogenic pollutants are higher in the river delta than upstream (Sun et al. 2023). In the case of the Jaguaribe River, disturbance of the natural river flow contributes to the opposite. The results of ecotoxicological tests have shown that sediments are more toxic upstream than in the delta of the Jaguaribe River. This effect is even more evident when comparing sites P5a and P5b, because although they are close to each other, the dam constructed between them can alter the toxicity of the sediment.

In a study using sediment toxicity bioassays with flax seeds (*Linum usitatissimum*) in the Rovinj watershed (Croatia), inhibition of seed germination and root growth was 5.36 and 1.9 greater, respectively, in the delta than at the upstream sampling site (Pelikan et al. 2022). Similar results were found in the Llobregat watershed (Spain) with *Daphnia magna* bioassays, where sediment toxicity was up to 1.6 times greater in the river delta than upstream (Castro-Català et al. 2016). Therefore, the results found in our study differ from what would be expected naturally, as microcrustacean mortality, inhibition of germination, and seed root size were greater at sites P2, P3, P4, and P5a

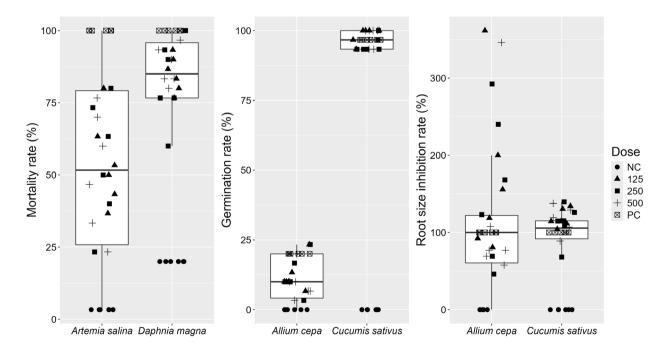


Figure 5. Analysis of variance of ecotoxicological bioassay results. **NC: Negative** Control; PC: Positive Control.

(upstream of the Jaguaribe) than at site P1 (in the delta).

The lethal and sublethal effects observed in this study indicate the risks of synergistic effects of contaminants. Studies suggest that the combined effects of pesticides, toxic metals, and polycyclic aromatic hydrocarbons, even at low concentrations, can poison people and make them more resistant to the effects of antibiotics (Cui et al. 2015), promote genetic changes in plants (Gallego & Olivero-Verbel 2021) and humans (Costa et al. 2021), and alter the physiological behavior of aquatic organisms (Tenorio et al. 2017).

The environmental composition surrounding the sampling sites also plays a critical role in the results of sediment toxicity bioassays. Bacteria present in mangrove roots are widely known as phytoremediators of polycyclic aromatic hydrocarbons (Verâne et al. 2020), toxic metals (Meng et al. 2021), and pesticides (Ivorra et al. 2021). Thus, the lower toxicity of sediments in the delta than upstream is also related to the fact that all other sites are surrounded by various economic activities that have degraded native vegetation, while the delta is the only collection site that has riparian forests and much of its surrounding area is occupied by mangroves.

The quality of natural resources is critical to sustaining aquatic life. Therefore, the degradation of water bodies is directly related to agricultural production and the quality of life of the surrounding population (Chimwamurombe & Mataranyika 2021). Studies show that there is a direct relationship between the quality of ecosystem services supported by freshwater and the water and food security of the population in large river basins in arid and semi-arid regions of the world (Sun et al. 2023). In the case of the Jaguaribe River watershed, there is already evidence that anthropogenic pollution is affecting the water's potability and bath (Freire et al. 2021).

Finally, we conclude that agricultural expansion is partly responsible for the presence of many chemical contaminants in the Jaguaribe River watershed, and that deforestation of native forests and construction of dams along the river are affecting important ecosystem services for the control, transport, and biotransformation of chemical wastes. Given the interaction of the results found with other economic, environmental, and social variables, it is critical that the ecotoxicological assessment of sediments in the Jaguaribe River watershed be continuous and aimed at equitable management of water resources without compromising ecosystems and the quality of life of local populations.

Acknowledgments

This study was funded in part by the Fundação Cearense de Apoio ao Desenvolvimento Científico e Tecnológico -Brazil (FUNCAP) and Coordenação de Aperfeiçoamento de Pessoal de Nível Superior - Brazil (CAPES) - Finance Code 001. The authors thank Prof. Dr. André Henrique Barbosa de Oliveira for their suggestions during the conceptualization of the manuscript.

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SUPPLEMENTARY MATERIAL

Table SI.

How to cite

VIEIRA JL, DANTAS ICD, OLIVEIRA AVS, RODRÍGUEZ MTT, DE MENEZES FGR & DE MENDONÇA KW. 2024. Toxicological risks assessment in the Jaguaribe River watershed (Ceará, Brazil) using anthropogenic contamination reports and ecotoxicological analysis. An Acad Bras Cienc 96: e20240226. DOI 10.1590/0001-3765202420240226.

Manuscript received on March 6, 2024; accepted for publication on May 26, 2024

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