

Effects of local land use on riparian vegetation, water quality, and *in situ* toxicity

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ABSTRACT

The conversion of riparian forests into agricultural land results in the loss of water quality and aquatic biota health. The objectives of this study were therefore to determine the proportion of land use with emphasis on the type of vegetation cover; evaluate the limnological parameters and concentration of inorganic elements in the water of the São José stream in the Dourados River Basin, Mato Grosso do Sul, Brazil; correlate land use with the concentrations of inorganic elements in water; and evaluate the risk to preservation aquatic biota and in situ toxic effect on Astvanax lacustris. We collected samples from the São José Stream in 2020 and evaluated land use with high resolution aerial images. The inorganic elements in water samples were quantified using inductively coupled plasma optical emission spectrometry. In the surroundings of the São José stream, a reduction in riparian forest was observed with a proportion of only 16.32% of the area and the predominance of agricultural areas with 75.06%. The concentrations of dissolved oxygen (1.510 mg L⁻¹) and P (> 0.235 mg L⁻¹) in the water did not comply with the national legislation. In addition, Al and P indicated risks regarding the conservation of aquatic biota (risk quotient >1). The *in situ* evaluation of *A. lacustris* also revealed toxicity in the water. The results indicate environmental imbalance in the São José stream, requiring mitigation measures for its restoration and the sustainable use of its resources.

Keywords: anthropic action, inorganic contaminants, risks for aquatic biota.



Efeitos do uso do solo na vegetação ciliar, qualidade da água e toxicidade *in situ*

RESUMO

A conversão de floresta ripária em áreas agrícolas resulta na perda da qualidade da água e da saúde da biota aquática. Assim, os objetivos deste estudo foram determinar a proporção de uso do solo com ênfase no tipo de cobertura vegetal; avaliar os parâmetros limnológicos e a concentração de elementos inorgânicos na água do córrego São José na Bacia do Rio Dourados, Mato Grosso do Sul, Brasil; correlacionar o uso do solo com as concentrações de elementos inorgânicos na água; e avaliar o risco para preservação da biota aquática e efeito tóxico in situ em Astyanax lacustris. Amostras de água do córrego São José foram coletadas em 2020 e avaliado o uso do solo com imagens aéreas de alta resolução. Os elementos inorgânicos nas amostras de água foram quantificados por espectrofotometria de emissão óptica de plasma indutivamente acoplado. No entorno do córrego São José, observou-se a redução da floresta ripária com proporção de 16,32% da área e a predominância de áreas agrícolas com 75,06%. As concentrações de oxigênio dissolvido (1,500 mg L⁻¹) e P (> 0,235 mg L⁻¹) apresentaram valores em desacordo com a legislação nacional. Além disso, Al e P indicaram riscos quanto à conservação da biota aquática (quociente de risco >1). A avaliação in situ de A. lacustris também revelou toxicidade na água. Os resultados indicaram desequilíbrio ambiental no córrego São José e, portanto, requerem medidas mitigadoras para sua recuperação e o uso sustentável de seus recursos.

Palavras-chave: ação antrópica, contaminantes inorgânicos, riscos para a biota aquática.

1. INTRODUCTION

The contamination of aquatic ecosystems can be mitigated by the presence of riparian forests, which function as filters and promote the retention of contaminants before transportation to watercourses (Cole *et al.*, 2020). Although the presence of these forests around water bodies is essential and effective at improving water quality, intensive exploitation, mainly by the expansion of agricultural and pasture areas, promote their reduction, making them increasingly degraded and consequently leaving watercourses unprotected (Collier *et al.*, 2019; Ramião *et al.*, 2020). Cole *et al.* (2020) reported that the width of the forest around water bodies is a relevant factor for the protection of water resources; however, other factors, such as the type of vegetation cover, must be considered. Law No. 12.651 of 2012 recommends riparian forest widths according to the size of the water body for the conservation of these aquatic ecosystems.

In this context, the contamination in aquatic environments mainly originates from various anthropic activities, especially urban waste (Bradney *et al.*, 2019; Islam *et al.*, 2020), industrial (Souza *et al.*, 2020; Islam *et al.*, 2020), agricultural and livestock effluents (Xue *et al.*, 2020). The expansion of exploitation of these activities favors the transport of sediments, fertilizers, and pesticides in these areas that infiltrate the soil and transported to the bed of water bodies (Wang *et al.*, 2019; Srinivas *et al.*, 2020; Rachels *et al.*, 2020). Contaminants that reach the water bodies may compromise water quality, ultimately affecting its viability for use and resulting in water toxicity to aquatic life and human health (Ali and Khan, 2018). Further, such contamination may cause bioaccumulation of metals in exposed organisms, exhibit genotoxic and mutagenic effects, and harm the aquatic biota, especially fish (Ali and Khan, 2018).

The *in situ* analysis with fish becomes relevant for evaluating water quality in response to the presence of riparian forest, as this type of analysis is based on more realistic environmental



conditions compared to laboratory experiments, including both complex and specific situations of the area (Chappie and Burton, 2000). *In situ* tests with fish have been used to assess surface water toxicity in aquatic environments (Mccallum *et al.*, 2017; Pei *et al.*, 2021). The species *Astyanax lacustris* has been widely used as a test organism (Bergmann *et al.*, 2020; Macêdo *et al.*, 2020; Viana *et al.*, 2020; Nascimento *et al.*, 2020; Merçon *et al.*, 2022) due to characteristics, such as high trophic plasticity (Alonso *et al.*, 2019), feeding flexibility (Alonso *et al.*, 2019), and adaptability in experiments (Viana *et al.*, 2018; Sposito *et al.*, 2019). *A. lacustris* has been demonstrated to be sensitive to the presence of a large amount of chemical contaminants in water, besides being a native species of the study basin.

Assessing the water quality of the Dourados River Hydrographic Basin (DRHB) is important for its monitoring and management, mainly owing to its use as a source of water for the municipality of Dourados and the region. The water from DRHB is used for irrigation, animal feeding, domestic and industrial use; and is of remarkable socioeconomic and environmental importance (IMASUL, 2005). Thus, the objectives of the study were to: (1) determine the proportion of land use with emphasis on the type of vegetation cover; (2) evaluate the limnological parameters and concentration of inorganic elements in the water of the São José stream belonging to the Dourados River Basin in the state of Mato Grosso do Sul; (3) correlate land use with the concentrations of inorganic elements in water; and (4) evaluate the risk for aquatic biota and *in situ* toxic effect on *A. lacustris*.

2. MATERIAL AND METHODS

2.1. Study area

The study was conducted in the São José stream, belonging to the DRHB, which is less than 10 m wide and located in a rural area of the municipality of Dourados. The surroundings of this stream present fragmented riparian vegetation cover with a predominance of agricultural areas, besides being a tributary that flows into another stream that is a tributary of the Dourados River. Three sampling sites, namely PI, PII, and PIII, located 500±100 m from one another, were employed (Figure 1). *In situ* monitoring was conducted for five days in February, 2020, characterized as a rainy period, with mean precipitation of 25.6 mm over the monitoring period.







2.2. Land use assessment

The delimitation of the microbasin was performed using Digital Elevation Model (DEM) images with a spatial resolution of 90 m downloaded from USGS Earth Explorer. The image was processed in QGis 3.18 (QGIS, 2009) with the help of Grass using the Hydrologic modeling tool for the elaboration of watersheds. The watershed of interest was converted to a polygon vector file, exported as a shapefile, and imported into the ArcGIS 10.8 trial version software (ESRI, 2015). For mapping the land use, high resolution aerial images obtained from Google Earth Pro® (2020) with a resolution of 1 m were used (Digital Globe, 2017). For this purpose, buffers with a radius of 350 m were generated around each sampling site. The types of land use were classified as follows, according to IBGE (2013) with adaptations: agricultural area, water body, road, forest fragments, planted forest, buildings, pasture, exposed soil, and fishpond. For interpretation of the images, an unsupervised classification (clustering) was conducted using the classification tools provided by the ArcGIS®. The dimensions and percentages of each type of land use were calculated based on the areas of the buffers and the delimitation of the microbasin. In addition, the size of the riparian forest was evaluated using Google Earth Pro® (2020), and it was measured from the lower to the upper bank of the stream and observed on both sides based on the average of the buffer areas (350 m) at each sampling site.

2.3. Assessment of the limnological parameters

The limnological parameters in this study were temperature (°C), dissolved oxygen $(mg L^{-1})$, electrical conductivity (μ S cm⁻¹), pH, and total dissolved solids (TDS) measured using the multiparameter probe, YSI Professional Plus. The alkalinity, hardness, ammonia, and nitrite content of the water were determined using the Alfakit ® colorimetric method, while its transparency was determined with the Secchi disk.

2.4. Identification of inorganic elements in water

Water samples (1000 mL) were collected daily (five days) from each sampling site using previously washed and sterilized amber glass bottles. After collection, the bottles were stored under refrigerated conditions and transported to the laboratory. The water samples were acidified with nitric acid until pH < 2, and then stored at 4°C until analysis. The water samples were filtered to remove suspended particles using a 0.45 μ m membrane filter (Millipore filtration assembly). To determine the inorganic elements Al, As, Ba, Ca, Cd, Co, Cr, Cu, Fe, K, Mg, Mn, Mo, P, Pb, Se, and Zn, the samples were prepared according to the method proposed by Mermet and Poussuel (1995) and tested using Inductively Coupled Plasma Optical Emission Spectrophotometry (ICP-OES) (Thermo Scientific, USA, model iCAP 6000®).

2.5. Risk assessment for aquatic biota

The water of the São José stream was classified as Class III, which comprises waters that can be used for human consumption, after conventional or advanced treatment; irrigation of tree, cereal, and forage crops; amateur fishing; secondary contact recreation; and as water supply for animals as per Resolution 357 of March 17, 2005 by the National Environment Council (CONAMA, 2005). Preliminary risk assessment for the protection of aquatic biota was performed according to the maximum limits allowed by the Brazilian legislation CONAMA 357/2005 for Class II freshwater. As Class II water is associated with the conservation of aquatic biota, the calculation of the risk quotient (RQ) was based on the criteria for Class II water quality by the formula:

$$RQ = \frac{WATER}{WQC}$$



Where WATER is the concentrations of each element detected in the water samples, WQC = water quality criterion for the preservation of aquatic life (mg L^{-1}), and RQ = risk quotient.

An RQ \geq 1 indicates a probable ecological risk for aquatic life (Godoy *et al.*, 2015). The risk index (RI) for the preservation of aquatic life was determined using the sum of the RQ values obtained for each chemical element individually. High RI values indicate potential risk and harmful effects to aquatic biota (Evans *et al.*, 2015; Gustavsson *et al.*, 2017).

2.6. Assembly and installation of monitoring chambers

The chambers were derived using plastic bottles with 14 cm diameter, 22 cm height, and 2.000 mL water capacity, and comprised lateral holes and 500 mm nylon mesh at the top and bottom, fixed with non toxic glue. Wooden stakes, nylon rope, and bricks were used to maintain the depth and fixation of the chambers in the *in situ* sampling sites, ensuring the chamber had no contact with the bottom of the water body. The retention chambers were installed in triplicate with a distance of 20 ± 5 m between replicates.

2.7. In situ toxicity analysis with A. lacustris

A. lacustris belongs to the family, Characidae (Characiformes), which includes small fishes called lambari (Lucena and Soares, 2016). This species has an oval-shaped body and yellow caudal fin (Botelho et al., 2019), which is responsible for its popular name, "lambari do rabo amarelo" ("yellow tail lambari"). Fish were collected from individual breeding lots on a fish farm at the State University of Mato Grosso do Sul (UEMS) of Aquidauana, Mato Grosso do Sul, Brazil. Thereafter, fish were acclimated to the ideal conditions of temperature (26±2°C), dissolved oxygen (7 \pm 1 mg L⁻¹,) pH (7.0 to 7.6), and photoperiod (12:12 h light/dark) in the laboratory aquarium tanks for 30 days (ABNT, 2003). The fish were fed once per day with pelleted feed containing 28% crude protein (Laguna®, batch No. 05EX180067109). After the pre-acclimatization period, fish were kept in experimental tanks containing five individuals per tank for seven days. Thereafter, the fish were transferred to plastic bags containing water with oxygenation and transported to the three *in situ* sampling sites in the study stream. Before the fish were inserted into the retention chambers, plastic bags containing the fish were immersed in the river water for 10 min. Five fish were placed in each chamber, totaling 15 fish per sampling site. Toxicity evaluation of the sampling sites was based on fish mortality in situ after 96 h.

2.8. Statistical analysis

The normality of the data was verified using Shapiro-Wilk tests. Confirmation of the nonnormality (p < 0.05) allowed the use of non-parametric tests, in which Spearman's correlation coefficients were determined between the types of soil in the buffers relative to the concentration of inorganic elements quantified in the water of the stream. The R platform (R Development Core Team, 2020) was used to conduct these analyses.

3. RESULTS AND DISCUSSION

3.1. Land use

The highest proportions of the land use in the São José stream microbasin were agricultural areas (75.06%), followed by forest fragments (16.32%), pasture (5.86%), water bodies (1.50%), fishponds (0.37%), buildings (0.31%), planted forest (0.13%), and exposed soil (0.12%) (Figure 2).







Therefore, agricultural land was predominant, which led to the reduction of riparian forest. The analysis of land use around the sampling sites revealed that PI had higher proportions of forest fragments (52%), followed by agricultural area (36%) and pastures (12%). PII had a higher proportion of agricultural area (56%), followed by forest fragments (44%). Agricultural activities occupied 66% of the area of PIII and forest fragments accounted for the remaining 34% (Figure 2). Of the study areas, greater proportions of agricultural areas were found around PII and PIII, while a greater proportion of forest fragments was found at PI, with evident expansion of the agricultural area. The riverbed becomes vulnerable with the reduction of riparian forest cover as the vegetation seeks to retain contaminants and maintain the quality of water bodies, especially in the conservation of limnological parameters (Fierro *et al.*, 2017). Another relevant aspect is the density of the riparian forest, which is directly proportional to its efficiency in protecting the slopes, thereby preventing the emergence of erosive processes and siltation in riverbeds (Cole *et al.*, 2020). In this study, such protection is undermined as the riparian forest is mainly fragmented due to anthropogenic activities, prevailing riparian undergrowth vegetation.

The riparian forest size varied from 3.35 to 24.46 m wide when measured on both sides of PI, presenting a mean of 12.52 m. In PII, the forest extent varied from 21.48 to 77.53 m, with a mean of 40.28 m. Finally, PIII had size variations from 16 to 35.49 m with a mean of 25.23 m. In the surroundings of all sampling sites, we observed that some stretches of riparian forest had

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strips of vegetation less than 10 m wide, which were smaller than the size determined by the Brazilian Forest Code (Law No. 12.651 of 2012). This code states that the minimum width of the riparian forest is 30 m on both sides of the water body. The irregularity of deforestation in this study area is not a local occurrence, but can be extended to other Brazilian agricultural areas that are in the continuous process of expansion (IBGE, 2020). Thus, many water bodies have their riparian vegetation degraded or reduced, which can cause serious risks for the conservation of water sources, resulting in damage to the structure and functioning of the aquatic environment. Such reduction can also impact the maintenance of aquatic and terrestrial species as the riparian forest helps regulate the limnological parameters, contributes to the feeding of many species of fish, serves as a food source for numerous organisms (terrestrial or aquatic), and serves as microhabitats for breeding and escape from predators (Lo et al., 2020; Selwood and Zimmer, 2020). Hilary et al. (2021) suggest that the length of the riparian forest is a key factor for the preservation of river water quality. Thus, to enable better efficiency, a fixed width throughout the water body covering the entire length of the stream is recommended, which was not observed in this study. In summary, the existence of riparian forests is of paramount importance for maintaining hydrological and ecological processes.

3.2. Limnological parameters

All sites sampled in the stream presented dissolved oxygen content in the water below the limit established by the Brazilian legislation CONAMA (357/2005) for Class III freshwater (Table 1), being inadequate for the maintenance of aquatic life, with values below 4 mg L⁻¹.

Limnological parameters		Study area	CONAMA (357/2005)	
	PI	PII	PIII	WMP
Temp. (°C)	23.42±0.56	22.64±0.31	22.30±0.20	*
Transp. (cm)	28.80 ± 6.76	31.00±11.40	26.40 ± 5.72	*
OD (mg L ⁻¹)	2.25 ± 0.57	2.57±0.65	3.52±0.94	>4
Cond. (µs cm ⁻¹)	38.00 ± 0.00	38.00±0.00	38.00±0.00	*
pН	7.30 ± 0.02	7.29 ± 0.03	7.31±0.11	6.0 - 9.0
TDS (mg L ⁻¹)	$20.54{\pm}11.48$	15.99±14.59	20.55±11.52	<500
Alkalinity	26.00 ± 5.47	24.00 ± 5.47	22.00 ± 4.47	*
Hardness	22.00 ± 4.47	24.00 ± 5.47	24.00 ± 5.47	*
Ammonia (mg L ⁻¹)	0.09 ± 0.05	0.12 ± 0.00	0.12 ± 0.00	<2.0
Nitrite (mg L ⁻¹)	0.00 ± 0.00	0.00 ± 0.00	0.00 ± 0.00	<1.0

Table 1. Limnological parameters of the water, presented as mean and standard deviation, at the sampling sites compared with the maximum values allowed by the resolution CONAMA (357/2005).

Temp: Temperature; Transp: Transparency; DO: Dissolved oxygen; Cond: Conductivity; TDS: Total Dissolved Solids; MPV: Maximum permitted value by the Brazilian legislation for Class III based on the National Environment Council CONAMA (357/2005); Bold: values above the limits established by legislation CONAMA (357/2005).

The scarcity of dissolved oxygen in water may be a reflection of the reduction in riparian forest as larger areas of vegetation improve water oxygenation (Iñiguez-armijos et al., 2018) and the process of self-purification performed by the roots of plants. The increase in nutrients from agricultural activities can also trigger eutrophication (Carpenter *et al.*, 1998; Van Beusekom, 2017), which is also a consequence of the reduction in dissolved oxygen in the water. Thus, the prolongation of a hypoxic aquatic environment can induce the extinction of species, causing mass mortality of aquatic biota (Galic *et al.*, 2019). As oxygen is reduced in the water, the most sensitive species are eliminated, leaving those that are the most resilient, and subsequently causing an imbalance in the structure of aquatic trophic levels. Franklin (2014) demonstrated that juvenile fish of different species had a mortality of 50% to 100% in



less than 48 h of exposure when the dissolved oxygen levels were below 3 mg L^{-1} . For other parameters, such as pH, TDS, ammonia, and nitrite, the values were within the limits allowed by the legislation. The national legislation does not have regulations for temperature, electrical conductivity of the water, alkalinity, and hardness.

3.3. Inorganic elements in water

The concentrations of Al and P were above the limits considered safe for aquatic life, according to the Brazilian legislation, CONAMA 357/2005, for Class III freshwater (Table 2). The values for Ba, Fe, and Mn were below the limits stipulated by the Brazilian legislation cited above (Table 2). No limits have been established by the national legislation for Ca, K, and Mg. Further, Cu level was found to be below the limit of quantification at all sampling sites. The elements As, Cd, Co, Cr, Mo, Pb, Se, Cu, and Zn were not detected.

Table 2. Concentration of inorganic elements in the water (mg L-1) at the sampling sites (PI, PII, and PIII) in the São José stream and values established by the CONAMA legislation.

Chemical elements	LOD	LOQ	PI	PII	PIII	CONAMA (357/2005)
				mg L ⁻¹		
Al	0.0180	0.0599	1.510±0.046	1.721±0.086	2.130 ± 0.032	0.200
Ba	0.0005	0.0016	0.025 ± 0.000	0.027 ± 0.000	0.026 ± 0.000	1.000
Ca	0.0070	0.0233	3.004 ± 0.089	3.206±0.025	3.024±0.105	*
Cu	0.0024	0.0080	<loq< th=""><th><loq< th=""><th><loq< th=""><th>0.013</th></loq<></th></loq<></th></loq<>	<loq< th=""><th><loq< th=""><th>0.013</th></loq<></th></loq<>	<loq< th=""><th>0.013</th></loq<>	0.013
Fe	0.0058	0.0193	3.319±0.072	3.130±0.063	3.303±0.060	5.000
K	0.0154	0.0515	0.405 ± 0.004	0.396 ± 0.003	0.396±0.021	*
Mg	0.0010	0.0033	1.812 ± 0.007	1.874 ± 0.011	1.801 ± 0.078	*
Mn	0.0003	0.0011	0.007 ± 0.000	0.006 ± 0.000	0.007 ± 0.000	0.500
Р	0.0135	0.0450	0.235±0.004	0.243±0.017	0.293±0.007	0.150

Bold: values above the limits established by legislation CONAMA 357/2005.

LOQ: limit of quantification.

* Absence of maximum permitted values.

The presence of these elements may be related to the rainfall conditions in the collection period, which may have favored the carriage of inorganic contaminants to the riverbed, which is associated with low riparian forest protection on the banks of the aquatic environment. Importantly, Al is an element with potential toxicity to aquatic life, which can induce serious effects to organisms, such as genotoxicity (Capriello *et al.*, 2021). The occurrence of Al in the aquatic environment may be related to typical characteristics and composition of the soil, which is the most abundant in the Earth's crust (CETESB, 2012), and its intensification in the aquatic environment by anthropic sources arising from the discharge of untreated domestic sewage and agricultural waste (Dalzochio *et al.*, 2018). Of note, P is a component present in chemical fertilizers used to stimulate the growth of monocultures, which mainly comprise corn, soybean, and sugarcane crops, which are common in the study region (Bojórquez-Quintal *et al.*, 2017; Mekonnen and Hoekstra, 2018; Mundim *et al.*, 2018; Waller *et al.*, 2021). The presence of P can also be related to pasture areas, as most of the P ingested by animals during grazing is returned to the soil via manure, with a return proportion of up to 60% (Assmann *et al.*, 2017).

The concentrations of Al and P in the watercourse of the study stream corroborate the results of land use that indicated intense occupation of the area by agricultural activities. Notably, different fertilizers containing chemical elements are used for land preparation for planting, which can help increase Al and P in the watercourse.



3.4. Correlation analysis

The present study revealed a significant positive correlation between pasture areas and K concentrations (p<0.001) (Figure 3). These results highlight the current scenario of the expansion of pasture areas at the expense of forest fragments in Brazil, ultimately indicating the use of chemical components to accelerate the growth of predominant crops. Another relevant aspect is the release of cattle manure that may be related to the increase in P and K in the water body (Assmann *et al.*, 2017).



Figure 3. Spearman correlation coefficients (r) between land use in relation to the concentration of inorganic elements quantified in the water of the stream belonging to the DRHB in Mato Grosso do Sul, Brazil. *p<0.05; ***p<0.001.

A negative correlation was found between Al and areas of forest fragments (p<0.05), suggesting that the reduction in riparian forests can determine greater Al runoff in the aquatic environment. Among the inorganic elements, a negative correlation was found between Ca and K. Of note, the imbalance and lack of Ca in the aquatic environment can harm vertebrate organisms via their skeletal structures, and several mollusks that require Ca carbonate for maintenance and production of their shells (Yarra *et al.*, 2021). Ca with K displayed a significant negative correlation (p<0.05), while Fe with Mn displayed a positive correlation, indicating possible synergy between these elements, which may be the result of intense agricultural activities around the water body. In addition to the sum and aggravation of the use of different chemicals, such as fertilizers and pesticides, for the maintenance of crops, later runoff of agrochemical waste in the riverbed may occur (Covert *et al.*, 2020).

3.5. Risk assessment for the preservation of aquatic biota

RQ was derived for the elements Al, P, and Fe, which had high concentrations in water and the maximum permitted limit according to the CONAMA legislation 357/2005. Water samples from PI, PII, and PIII had concentrations of Al, Fe, and P that indicated risks for the preservation of aquatic biota, with a value > 1 (Figure 4a).

These risks can result in deleterious and irreversible effects that compromise the health of aquatic biota, especially fish. Notably, even low concentrations of Al can cause embryological damage, teratogenic, neurotoxic, and oxidative stress in fish (Monaco *et al.*, 2017). Correia *et al.* (2021) observed that Al functioned as an endocrine disruptor, thereby interfering with the hormonal system of *Astyanax altiparanae*, and resulting in failure of ovulation and deleterious effects on reproduction. Excess Fe in the aquatic environment can cause obstruction in the gills of fishes and respiratory disorders, ultimately interfering in gas exchange and causing suffocation (Bury *et al.*, 2011), which causes toxic aquatic environments (Alipour and Banagar, 2018). Moreover, cases of acute concentrations of Fe can result in necrosis of gill tissue and

loss of the ability to excrete ammonia concentrated in the blood of fish (Slaninova *et al.*, 2014). Marins *et al.* (2019) observed genotoxic effects in adult *Danio rerio* after Fe exposure over a period of 30 days. High concentrations of P in water bodies near agricultural areas are related to the runoff of fertilizer containing P in its composition, which are used to improve nutrients during land preparation. However, high P concentration can lead to eutrophication of the aquatic environment as P is a dominant element in the acceleration of nutrients (Mardamootoo *et al.*, 2021). High concentrations of P in water samples from the study stream coincide with the conditions of land use in soil preparation, as the presence of agriculture in these areas is intense and located close to the water body, favoring the runoff of these elements.



Figure 4. (a) Risk quotient (RQ) for the preservation of aquatic biota based on the levels of Fe, Al, and P at PI, PII, and PIII; and (b) Risk index (RI) for the preservation of aquatic biota through the sum of individual RQs of inorganic elements at PI, PII, and PIII in the São José stream. RQ and RI above the dotted line represent values > 1, which indicate risks for aquatic biota.

Depending on the concentrations of certain elements isolated in the aquatic environment, the health of aquatic biota can be harmed, and when evaluated in mixtures, they increase the risks (Figure 4b) and damage the preservation of aquatic biota, especially fish. When the inorganic elements were evaluated in mixtures in the aquatic environment, the RI values depicted risks to aquatic life based on the water samples collected from the study stream (Figure 4b). The inorganic elements that presented RQ > 1 had the greatest contribution to the increase in risks to biota in the RI, thereby highlighting Al at PI, PII, and PIII (RQs ~15.08, ~17.21, and ~21.30, respectively). Mixtures of chemical elements in water bodies can potentiate their effects (Martin *et al.*, 2021) and result in various damages to aquatic organisms, from genotoxic effects to mortality, which can compromise the proper functioning of the organism or even lead to death (Jijie *et al.*, 2020; Carvalho *et al.*, 2020).

3.6. In situ toxicity analysis

Based on water toxicity with *A. lacustris in situ* in the study stream, of the 15 individuals exposed at each of the sites, 5, 11, and 9 died, indicating a percentage of death of 33%, 73%, and 60%, respectively, at PI, PII, and PIII. The low oxygenation ($< 4 \text{ mg L}^{-1}$) in the water of the stream may have contributed to the mortality of fish, as only 44.44% of the exposed fish survived during the *in situ* analysis. The lower mortality in PI relative to the other sites may be related to the lower proportions of agricultural areas in its surroundings, thereby mitigating the risk of contamination of this area. Sampling sites PII and PIII were areas with larger extensions



of agricultural areas and high concentrations of Al and P. These results corroborate the literature data, where toxicity in aquatic ecosystems was reported with changes in the landscapes of their surroundings, in addition to elevated levels of Al or xenobiotic mixtures in the course of this aquatic environment (Dos Reis Oliveira *et al.*, 2018; De Souza *et al.*, 2019; De Castilhos Ghisi *et al.*, 2020; Riveros *et al.*, 2021). Therefore, in view of the results, restrictive measures are not only needed for riparian forest degradation, but also to mitigate the increase in unsustainable land use around water bodies, especially in agricultural areas, in order to minimize the impacts of these activities and preserve the quality of water and aquatic life.

4. CONCLUSION

The proportion of land use around the São José stream revealed a predominance of agricultural areas and consequently a low proportion of riparian forest, indicating inadequacy in relation to the Brazilian Forest Code. This reduction in riparian area may be reflected in the decreased water oxygenation and the increased Al and P concentrations, which might be due to the use of pesticides and agricultural fertilizers. The stream water led to the mortality of *A. lacustris* and was associated with risks that affect the health of aquatic life. Al was identified as the chemical element with the most contribution to this risk. Thus, to avoid negative impacts from anthropogenic activities on water bodies, a more effective monitoring program is necessary to protect the degradation of these areas and control the increase in agricultural areas around water bodies to minimize contamination. In relation to the study area, reforestation is proposed to ensure that the proportion of riparian forest is reestablished and efficient for retaining contaminants in the watercourse to ensure ecological balance for aquatic life.

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