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ORIGINAL ARTICLE – Conservation of Nature

Benthic Macroinvertebrates Associated with Riparian Habitat Structural Diversity in an Eastern Amazon Stream Urbanization Gradient

Luna Santiago[1]
Colin Robert Beasley[1]

[1]Universidade Federal do Pará (UFPA), Instituto de Estudos Costeiros, Bragança, PA, Brasil.

Abstract

Rich freshwater biodiversity is threatened by increasing deforestation and disorderly urbanization throughout the Brazilian Amazon, especially in streams and creeks, leading to loss of aquatic habitats. Biological information combined with habitat and water quality are effective tools for rapid assessment. The impact of increasing urbanization was assessed in the Cereja River, eastern Amazonia using a Riparian Habitat Diversity Index (RHDI), benthic macroinvertebrate structure and water and sediment variables in ten areas along the Cereja using multivariate analyzes. Increasing urbanization is associated with lower RHDI, abundance, diversity and equity of benthic macroinvertebrates, higher conductivity, pH, temperature, width and percentage gravel. This information is useful for rapid identification of impacts, assessment of recovery of degraded areas and maintenance of non-degraded areas along urban streams.

Keywords: River, impact, watercourse, forest, benthos.

1. INTRODUCTION AND OBJECTIVES

Brazil has rich freshwater biodiversity (Azevedo-Santos et al., 2019) increasingly threatened by unsustainable activities (Pelicice et al., 2017; Tófoli et al., 2017; Daga et al., 2020; Nobile et al., 2020), mainly deforestation and unplanned urbanization, which impact on hydrology and water quality (Piazza et al., 2017; Côrtes and Silva Júnior, 2021; Zerega et al., 2021). These impacts are progressively spreading throughout the Brazilian Amazon, especially in smaller streams and creeks (Helson and Williams, 2013; de Paiva et al., 2021). The lack of legislative protection for freshwater ecosystems in Brazil is of great concern (Frederico et al., 2018; Azevedo-Santos et al., 2019), potentially resulting in even greater vulnerability to impacts (Escobar, 2015; Brito and Magalhães, 2017), and further declines in freshwater biodiversity.

Urbanization is the second largest cause of habitat destruction worldwide, and one of the greatest threats to freshwater biodiversity (Ellis et al., 2010; Hill et al., 2015). Increased urbanization equates to deteriorating water quality, an altered hydrological regime and reduced habitat diversity (Luo et al., 2017; Piazza et al., 2017; Zerega et al., 2021; Iñiguez-Armijos et al., 2022) due to land use impacts in the surrounding catchment, pollution, urban surface impermeability and removal of riparian vegetation causing losses in aquatic biodiversity (de Paula et al., 2021), such as that of stream macroinvertebrates (Feio and Teixeira, 2019; Dala-Corte et al., 2020; Sundar et al., 2020; Zerega et al., 2021).

Effective conservation measures require knowledge of biotic distribution patterns, especially bioindicators (Poleto, 2010), through comprehensive surveys (Brito and Magalhães, 2017). Aquatic macroinvertebrates are used to assess or monitor changes in environmental conditions since many are sensitive to even minor impacts (Ilmonen et al. 2013; Faria et al., 2021) manifested as changes in taxonomic composition and abundance (Taniwaki and Smith, 2011).

The use of benthic macroinvertebrates as bioindicators associated with water quality and habitat can provide a quick, efficient (Oliveira and Callisto, 2010), accurate and low-cost diagnosis of aquatic habitats (Siqueira and Trivinho-Strixino, 2005; Maltchik et al., 2012). The Riparian Habitat Diversity Index (RHDI) protocol, proposed by Callisto et al. (2002), rapidly evaluates land use and occupation around the stream and the level of impact on instream and riparian habitat structural diversity. Lower RHDI scores are associated with greater impacts of land use and urbanization.

Our study aimed to assess the impact of a gradient in human activity on the riparian habitat and potential associations with macroinvertebrate assemblage structure (abundance and composition) along the Cereja River, Bragança, Pará, Brazil. Based on the sensitivity of some macroinvertebrate taxa to human impacts, we predicted that a less diverse macroinvertebrate fauna, especially of the orders Ephemeroptera, Plecoptera and Trichoptera (EPT), would occur in areas associated with low RHDI (Kikuchi and Uieda, 2005; Dohet et al. 2002). In contrast, Coleoptera and Chironomidae (Diptera) may be more tolerant of impacts (Goulart and Callisto, 2003) and likely to be more abundant in areas with greater urbanization.

Secondly, we expected lower sediment heterogeneity and higher fine sediment loads, such as silt and clay (Matthaei et al., 2010) in highly urbanized areas associated with riparian vegetation loss and increased siltation in the river bed and margins (Krupek and Felski, 2006). In areas with less urbanization, greater sediment heterogeneity and a lower proportion of fine sediments are expected due to the presence of riparian vegetation, which may filter material entering the river (Brito et al., 2009).

Finally, lower dissolved oxygen was expected in highly urbanized areas due to fine sediment accumulation acting as a barrier to gas exchange which, causes changes in the freshwater benthic invertebrates, either directly through burial, clogging, and associated reduction in oxygen availability, or through indirect effects of changes in habitat or food availability (Jones et al., 2011; Murphy et al., 2015). Low vegetation cover in urbanized areas raises water temperatures, reducing dissolved oxygen concentrations (Brand and Miserendino, 2015), in addition to inputs of organic matter and other pollutants, responsible for eutrophication (Esteves, 2011; Barreto et al., 2013).

2. MATERIALS AND METHODS

2.1. Study area

The Cereja River is a second order tributary of the Caeté River in northeastern Pará, eastern Amazon (Guimarães et al., 2009). Mean width is 5.5 m (range 1.2-15.0). Along its 5 km course through the city of Bragança, it receives commercial, domestic and hospital effluents and is affected by urban development and civil construction works, especially in the densely urbanized lower reaches (Guimarães et al., 2009; Monteiro et al., 2011). Urbanization is sparse in the upper course where vegetated and less urbanized areas predominate around the headwaters and increases in the middle and lower course where buildings and infrastructure predominate.

2.2. Sampling methodology

Riparian habitat structural diversity was characterized once using the RHDI protocol at each of ten areas along the Cereja, all within the urban district of Bragança: five in less urbanized zones in the headwaters and five in more urbanized zones in the second order channel further downstream, in June and July 2021 (See Figure 1 in Results). The survey took approximately 20 minutes to complete in each area. The first part of the protocol evaluates human impacts on the riparian zone whereas the second evaluates habitat diversity and the degree to which natural conditions are conserved. The RHDI protocol was modified from the original with 22 parameters: parameter 7, referring to water transparency, was modified, since Amazonian rivers naturally have tea color due to soil conditions and dissolved organic substances in the water (Gorayeb et al. 2010; Gorayeb et al. 2011). Thus, the color of strong or transparent tea had four points, followed by a cloudy color with two points and an opaque color with zero points. Parameters 12 and 13 (length and frequency of rapids) were removed, since the protocol was originally developed for mountainous regions (Callisto et al. 2002). A total of 20 parameters were thus evaluated and the final score is the sum of the points. The RHDI classification is based on the score of the stretch evaluated: impacted (0-40 points), altered (41-60 points), and natural (≥61 points). With the removal of two of the 22 parameters (representing a 9.1 % reduction in potential scoring), the thresholds were adjusted to 90.9% of their original range. Our modified RHDI class thresholds are: impacted (0-36 points), altered (37-54 points), and natural (≥55 points).

Habitat (water and sediment variables) and macroinvertebrate fauna sampling took place in each of the ten areas. In a 50 meter stretch in each area, three 15 meter long sampling plots the width of the stream were selected. In each plot, four randomly selected habitat and faunal replicates were taken with nearest neighbor distances of 1.5 to 2.0 m. A total of 3 plots times 4 replicates (n=12) were taken in each of the ten areas, totaling 120 replicates.

Using the above plot replicate design, water temperature (°C), electrical conductivity (μS/cm), dissolved oxygen concentration (mg/L), hydrogen potential (pH) and oxidation-reduction potential (mV) were obtained *in situ*, using a Hanna multi-parametric meter (precision 0.01 units). Stream width (m) and depth (m) were measured and surface current speed (m/s) was obtained using a digital current speed meter (precision 0.01 m/s) and discharge was estimated with Q=width.depth.speed(m^3/s). Additionally, for each replicate, water was collected in 15 ml Falcon tubes and analyzed in the laboratory for turbidity using a Hach digital colorimeter. To determine sediment composition (Suguio, 1973), sediment replicates were classified by weight (g) into percent fractions of Gravel, Very coarse sand, Coarse sand, Medium sand, Fine sand and Very fine sand.

Benthic macroinvertebrates were also collected using the plot replicate design above with the kick net procedure, gently agitating the upstream sediment for a standard duration of 3 minutes and retaining dislodged invertebrates, debris and sediment. Sampled sediment was washed in the field with a 300 μm mesh to remove coarse particles. The remaining material was packed in plastic bags and fixed in 70% alcohol, labeled, sorted and identified.

2.3. Data analysis

All analyses were performed using GNU-R 4.0.4 (R Core Team 2021). Data distributions were examined with box plots and histograms for preliminary analysis and presentation. The multivariate macroinvertebrate faunal structure (abundance and composition) was described using ordination by non-metric multidimensional scaling carried out on a Bray-Curtis dissimilarity matrix based on square root transformed faunal abundance. Correspondence with water quality variables and sediment composition was analyzed using bioenv() and envfit() functions to identify the most important habitat variables, i.e. those with the best correlation or regression fit, respectively, to the square root abundance transformed faunal structure among areas. The most important macroinvertebrate taxa in the association with multivariate differences among areas and RHDI classes were identified directly from the species display in the ordination and via smooth surface fitting of individual taxon abundance to the ordination using the ordisurf() function. Rank abundance models (Magurran, 2004) were fitted to the faunal assemblages in each area to describe variation in richness and equitability. Permutational multivariate analysis of variance (Permanova) with 1000 permutations was carried out on both square root transformed faunal abundance and untransformed water quality variables, using the Bray-Curtis dissimilarity index and Euclidean distance, respectively, in order to compare faunal structure and water quality between areas and RHDI classes. Multivariate analyses above were carried out using the vegan R package (Oksanen et al. 2020).

3. RESULTS

3.1. Riparian Habitat Diversity Index (RHDI) and fauna

Areas 1 to 5 had the highest RHDI scores (79, 83, 85, 84 and 84, respectively) and were classified as Natural. These more vegetated locations were relatively diverse and preserved (Figure 1) with riparian forest width 12 to 18 m and cover between 70% and 90% or more, stable river banks and river bed with few, if any, alterations, and widespread aquatic macrophytes and/or mosses.

Moderate RHDI scores in Areas 6, 7 and 8 (46, 49, and 42, respectively) indicated altered habitats (Figure 1) where riparian vegetation cover ranged from 0% to 50% and river margins were unstable, with 30% to 60% eroded. Riparian vegetation width varied between 6 and 12 m and there was evidence of strong human impact, such as housing, fences, litter and raw sewage discharge. Filamentous algae, mainly cyanobacteria, covered the riverbed in Areas 6 and 7. Large beds of macrophytes, especially *Cabomba* were observed in Area 8. Some modifications to the river channel were observed on both banks in Areas 6 and 8, and channelization around a bridge in Area 7.

Low RHDI scores (33 and 22, respectively) indicated impacted habitat at Areas 9 and 10 (Figure 1) with dense urban development, litter, raw sewage, severe deforestation with low riparian vegetation width (< 6-12 m) and cover (0-50%), and moderately unstable river banks (30-60% eroded) and greatly modified river margins (80%).

A total of 6.857 individuals among 38 benthic macroinvertebrate taxa were found overall (Table 1). A total of 36 taxa were found in natural areas, 17 in altered areas, and 10 in impacted areas. Abundance was concentrated (87.2% of total) in Chironomidae (Diptera - 3312, 48.3%), Thiaridae (Gastropoda - 1685, 24.57%), Oligochaeta (526, 7.67%) and Hydroptilidae (Trichoptera - 457, 6.66%). Chironomidae was present in all areas and RHDI classes, with high abundance in Areas 1, 2, 5 and 6 and low abundance in impacted areas. Thiaridae was present only in Areas 7 to 10, being most abundant in Area 8. Ostracoda was the most abundant taxon in altered areas and had low abundance in natural and impacted areas. Hirudinea was distinctly more abundant in impacted areas (Table 1, Figure 2). Oligochaeta was present in all areas, but was the most abundant taxon in altered and impacted areas (Table 1, Figure 2). Hydroptilidae was present only in natural areas, and abundant in areas 3-5 (Table 1, Figure 2).

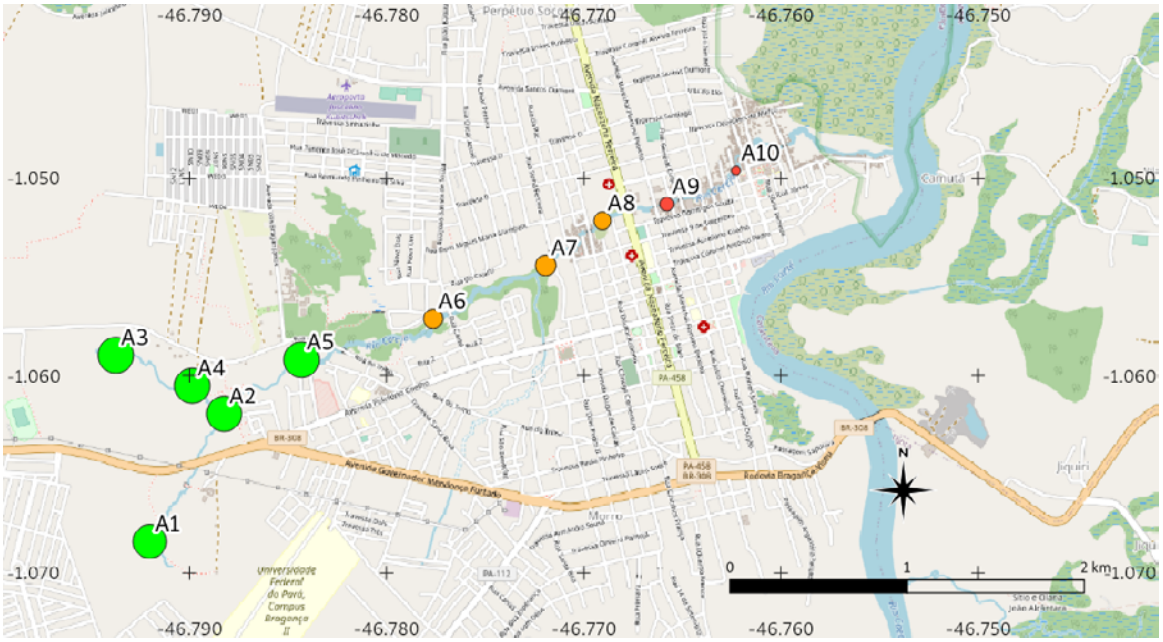

Figure 1. Map of the Cereja River, Bragança, Pará, Brazil, showing the ten areas (A1 – A10) sampled between the months of June and July 2021. Points are filled according to the RHDI class (green = Natural; orange = Altered; red = Impacted) and point diameter is proportional to the RHDI score. © OpenStreetMap contributors.

Table 1. Abundance of taxa in the ten areas classified as natural, altered and impacted, according to their RHDI score, sampled between the months of June and July 2021 along the Cereja River, Pará, Brazil, and the totals per class. N: Natural, A: Altered and I: Impacted.

Taxa		Areas and Class										Total per Class		
		1 N	2 N	3 N	4 N	5 N	6 A	7 A	8 A	9 I	10 I	1-5 N	6-8 A	9-10 I
Nematoda		0	2	4	0	2	9	0	3	0	0	8	12	0
Oligochaeta		28	2	9	2	42	8	101	119	154	61	83	228	215
Hirudinea		0	0	0	0	4	1	3	15	61	42	4	19	103
Gastropoda	Planorbidae	0	0	0	0	0	0	0	1	0	0	0	1	0
	Thiaridae	0	0	0	0	0	0	195	896	331	263	0	1091	594
Acari		0	6	6	1	8	8	3	1	0	1	21	12	1
Ostracoda		0	3	0	0	3	66	13	72	1	2	6	151	3
Cladocera		1	0	0	0	0	0	0	0	0	0	1	0	0
Decapoda	Palaemonidae	1	0	0	0	0	0	0	0	0	0	1	0	0
Isopoda		0	0	1	0	0	0	0	0	0	0	1	0	0
Collembola		0	1	1	0	0	1	3	5	0	5	2	9	5
Ephemeroptera	Baetidae	0	0	0	0	1	0	0	0	0	0	1	0	0
	Caenidae	1	0	0	0	0	0	0	0	0	0	1	0	0
	Leptohyphidae	0	1	0	0	13	2	0	0	0	0	14	2	0
	Leptophlebiidae	1	3	1	0	4	0	0	0	0	0	9	0	0
Odonata	Aeshnidae	0	0	0	0	4	0	0	0	0	0	4	0	0
	Gomphidae	0	0	0	1	1	0	0	0	0	0	2	0	0
Hemiptera	Belostomatidae	0	0	1	0	0	0	0	0	0	0	1	0	0
	Gelastocoridae	0	0	0	1	0	0	0	0	0	0	1	0	0
	Gerridae	0	0	0	0	1	0	0	0	0	0	1	0	0
	Mesoveliidae	0	0	1	0	0	0	0	0	0	0	1	0	0
Coleoptera	Elmidae	11	0	9	1	2	0	0	0	0	0	23	0	0
	Heteroceridae	0	0	1	0	0	0	0	0	0	0	1	0	0
	Staphylinidae	0	0	1	0	0	0	0	0	0	0	1	0	0
Trichoptera	Glossosomatidae	0	0	0	1	0	0	0	0	0	0	1	0	0

Table 1. Continued...

Taxa		Areas and Class										Total per Class		
		1 N	2 N	3 N	4 N	5 N	6 A	7 A	8 A	9 I	10 I	1-5 N	6-8 A	9-10 I
	Hydroptilidae	3	22	56	114	259	3	0	0	0	0	454	3	0
	Hydropsychidae	5	1	30	1	2	6	0	0	0	0	39	6	0
	Leptoceridae	0	2	0	26	9	0	0	0	0	0	37	0	0
	Limnephilidae	0	0	2	7	3	0	0	0	0	0	12	0	0
	Odontoceridae	0	0	2	1	0	0	0	0	0	0	3	0	0
	Polycentropodidae	0	8	2	0	40	3	0	0	0	0	50	3	0
Diptera	Ceratopogonidae	9	92	82	7	44	8	0	2	0	1	234	10	1
	Chaoboridae	0	0	1	0	0	0	0	0	0	0	1	0	0
	Chironomidae	554	599	222	62	938	545	140	114	29	109	2375	799	138
	Empididae	0	0	6	0	2	3	0	0	0	0	8	3	0
	Psychodidae	1	0	0	1	0	1	3	3	0	2	2	7	2
	Simulidae	0	1	0	1	7	0	0	0	0	0	9	0	0
	Tipulidae	4	5	7	6	0	2	0	0	0	3	22	2	3

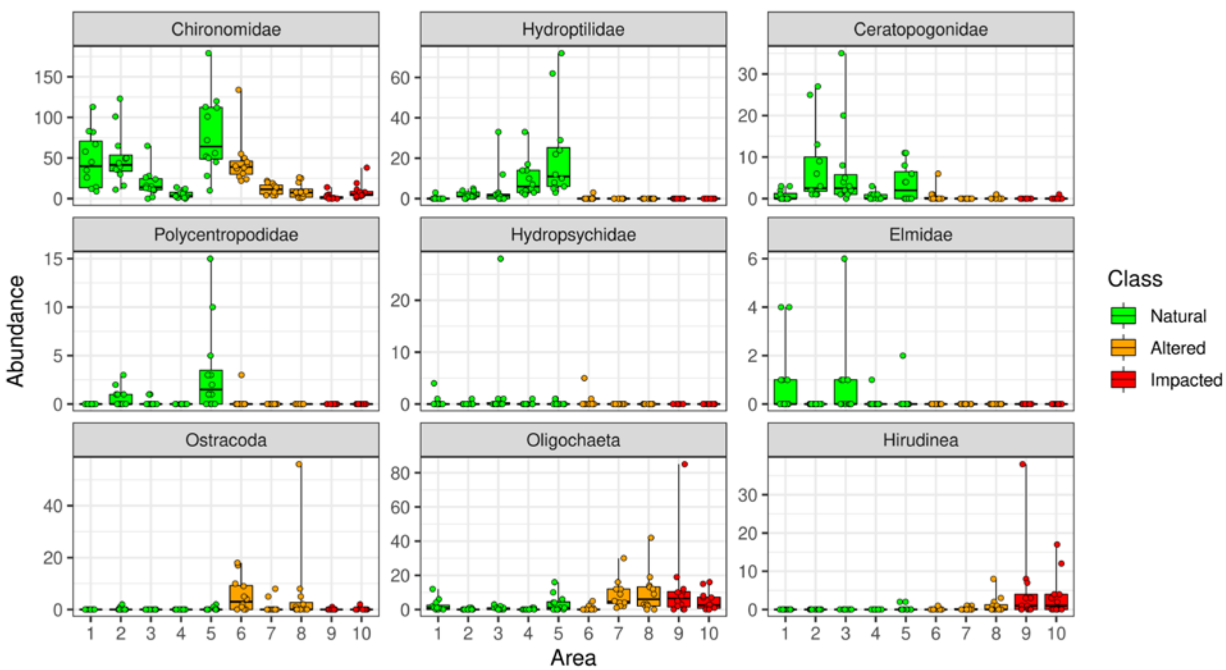

Figure 2. Abundance of selected benthic macrofauna taxa in the ten areas classified as natural, altered and impacted, according to their RHDI score, sampled between the months of June and July 2021 along the Cereja River, Pará, Brazil. Box and whisker plots of median and other quartiles and raw values.

Abundance of Ceratopogonidae (Diptera), Polycentropodidae (Trichoptera), Hydropsychidae (Trichoptera) and Elmidae (Coleoptera) was high in natural areas and lower in altered and impacted areas. Eight families with two to 36 individuals and another 13 taxa as singletons were exclusive to natural areas (Table 1). Of the 17 taxa in altered areas, only Planorbidae (Gastropoda) occurred exclusively, as a singleton. In impacted areas, none of the 10 taxa were exclusive. The rank abundance models that best fitted macroinvertebrate structure were Mandelbrot to the more diverse (16-21 taxa) and equitable faunas with high (84-85) RHDI scores, Zipf to the less diverse (12-15 taxa) and less equitable faunas, with RHDI scores from 46 to 83 and Lognormal and Preemption to the faunas with lowest diversity and equitability, with lowest RHDI scores (Figure 3).

The fauna of natural Areas 1 to 5 differed to that of impacted Areas 9 and 10, and the fauna of altered areas 6 to 8 was intermediate to these (Figure 4). Although altered, Area 6 with 15 taxa, was more similar to natural areas. Conductivity explained most of the variation between environmental variables and fauna (envfit, R^2=0.75, P=0.001, Table 2, Figure 4). Conductivity, pH and temperature were also significantly associated with macroinvertebrate structure (bioenv, r_s=0.578, P< 0.05). In natural areas, Chironomidae, Ceratopogonidae, Hydroptilidae and Polycentropodidae were associated with greater water depth, coarse sand and medium sand (Figure 4, Figure 5). In altered areas, Ostracoda was associated with the increase in percentage gravel (Figure 4, Figure 5). In impacted areas, Thiaridae, Hirudinea and Oligochaeta were associated with increased conductivity, pH and turbidity and higher water temperatures (Figure 4, Figure 5).

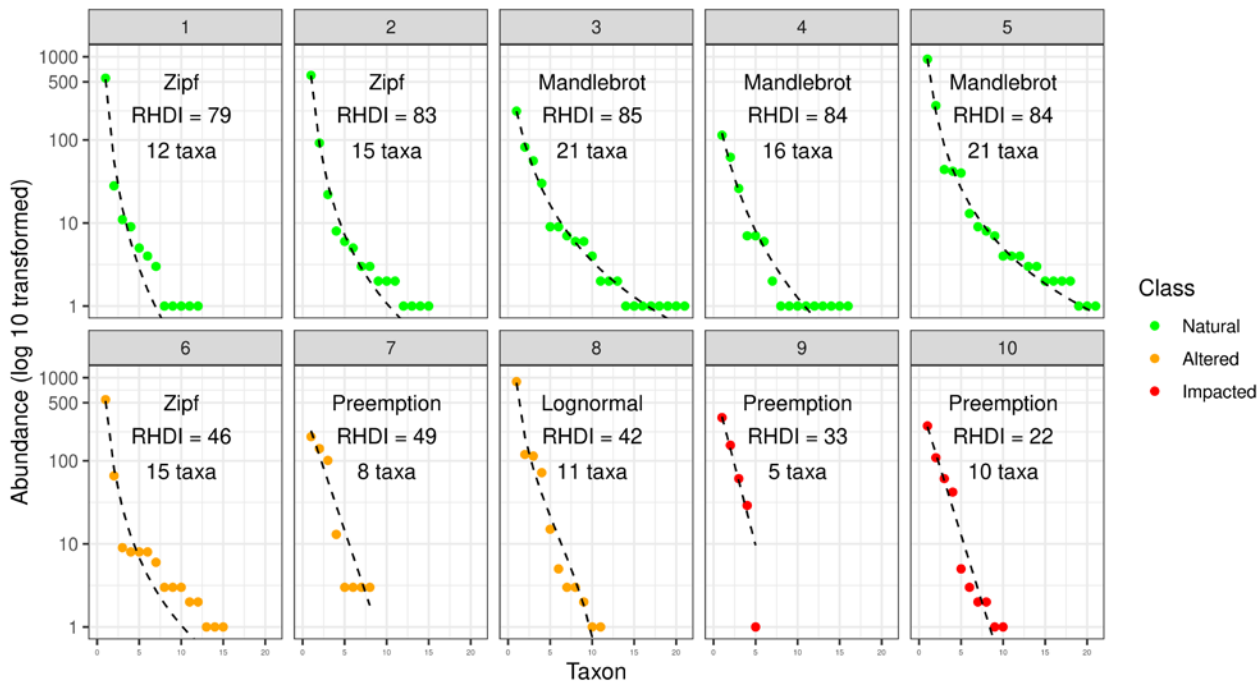

Figure 3. Rank abundance plots and the distribution models with the best fit to the structure of benthic macrofauna in the ten areas classified as natural, altered and impacted, according to their RHDI score, sampled between the months of June and July 2021 along the Cereja River, Pará, Brazil.

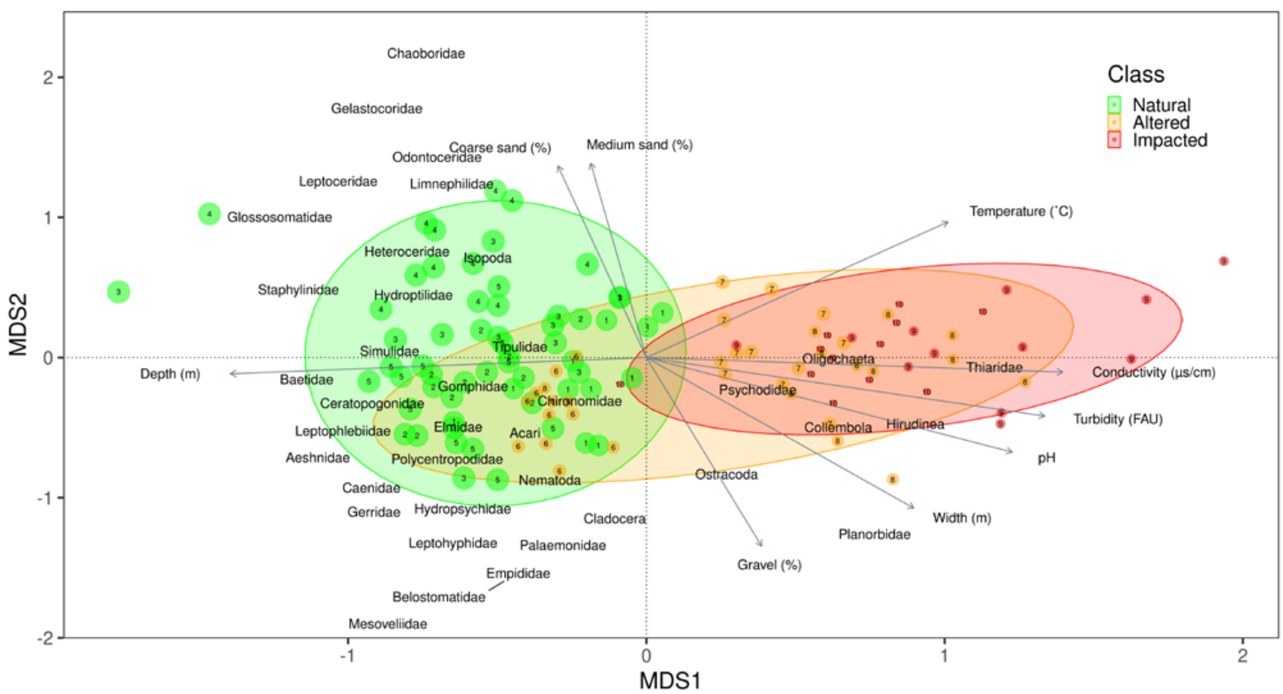

Figure 4. Non-metric multidimensional scaling (nMDS) with square root transformation of faunal abundance that minimized stress to 15.7%, showing differences in the structure of the benthic macrofauna among the ten areas classified as natural, altered and impacted, according to their RHDI score, sampled between the months of June and July 2021 along the Cereja River, Pará, Brazil. The diameter of the points is proportional to the RHDI score of the sampled Area. The direction and length of the envfit vector indicate an increase in values of a variable in the direction of the vector and effect size, respectively.

Table 2. R^2 values and *P* values of the envfit analysis of environmental variables identified as significantly associated with the structure of the benthic macrofauna in the ten areas classified as natural, altered and impacted, according to their RHDI score, sampled between the months of June and July 2021 along the Cereja River, Pará, Brazil. P values in bold are significant.

Environmental variables	R^2	P
Conductivity (μs/cm)	0.75	**0.001**
pH	0.65	**0.001**
Temperature (˚C)	0.34	**0.001**
Turbidity (FAU)	0.21	**0.001**
Width (m)	0.20	**0.001**
Gravel (%)	0.11	**0.001**
Coarse sand (%)	0.08	**0.017**
Depth (m)	0.07	**0.022**
Medium sand (%)	0.06	**0.036**

Multivariate differences in macroinvertebrate abundance and composition were significant (Table 3), both among Areas 1 to 10, and among RHDI classes (Natural: Areas 1-5, Altered: Areas 6-8 and Impacted: Areas 9-10). Differences among areas explained almost twice as much variation in the data (Permanova R^2=56.2%) than among RHDI classes (Permanova R^2=30.8%).

Natural areas had greater proportions of fine sediments and were more heterogeneous in sediment composition than altered or impacted areas, which tended to be dominated by gravel and coarse sand (Figure 6). The percentages of gravel and coarse sand varied between areas, and in Areas 6 to 8 varied greatly among replicates (Figure 6). The highest percentage of gravel occurred in altered to impacted Areas 6 to 10. Lower percentages of coarse sand were found in altered Areas 6 to 8, whereas these were higher and similar among both natural Areas 1 to 5 and impacted Areas 9 to 10 (Figure 6). Medium sand was similar in all areas, but varied considerably, especially in altered and impacted areas (Figure 5, Figure 6).

Multivariate patterns in water quality differed significantly (Table 4) both among areas and RHDI classes. Similar to faunal structure, area explained more variation in the data (Permanova R^2=65.3%) than class (Permanova R^2=53.5%), although class had a larger measure of effect (pseudo-F) than area (Table 4).

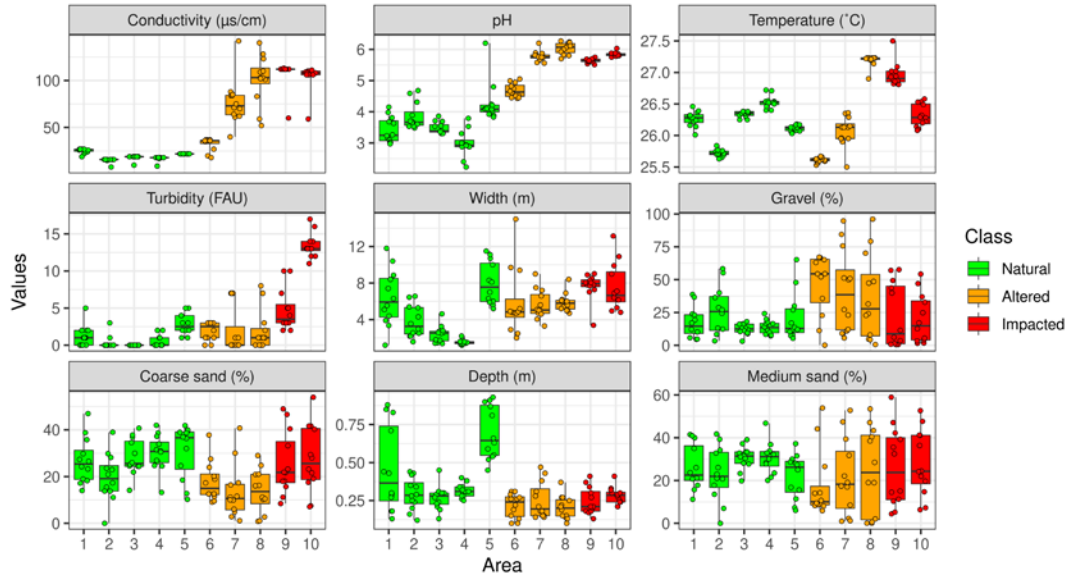

Figure 5. Environmental variables: conductivity (µS/cm), pH, temperature (ºC), turbidity (FAU), width (m), gravel (%), coarse sand (%), depth (m) and medium sand (%) in the ten areas classified as natural, altered and impacted, according to their RHDI scores, measured in June and July 2021 along the Cereja River, Pará, Brazil.

Table 3. Summary of the Permanova analysis for significant multivariate differences in benthic macrofauna structure in the ten areas classified as natural, altered and impacted, according to their RHDI score, sampled between the months of June and July 2021 along the Cereja River, Pará, Brazil. df: degrees of freedom, SS: Sum of Squares, MS: Mean Square. P values in bold are significant.

Source of variation	df	SS	MS	pseudo-F	R^2	P	Source of variation	df	SS	MS	pseudo-F	R^2	P
Area	9	14.95	1.661	15.17	0.562	**0.001**	Class	2	8.21	4.10	25.24	0.308	**0.001**
Plot	3	0.21	0.069	0.63	0.008	0.833	Plot	3	0.21	0.07	0.43	0.008	0.981
Area:Plot	27	2.69	0.100	0.91	0.101	0.717	Class:Plot	6	0.63	0.11	0.65	0.024	0.925
Residuals	80	8.76	0.109		0.329		Residuals	108	17.55	0.16		0.660	
Total	119	26.60			1.000		Total	119	26.60			1.000	

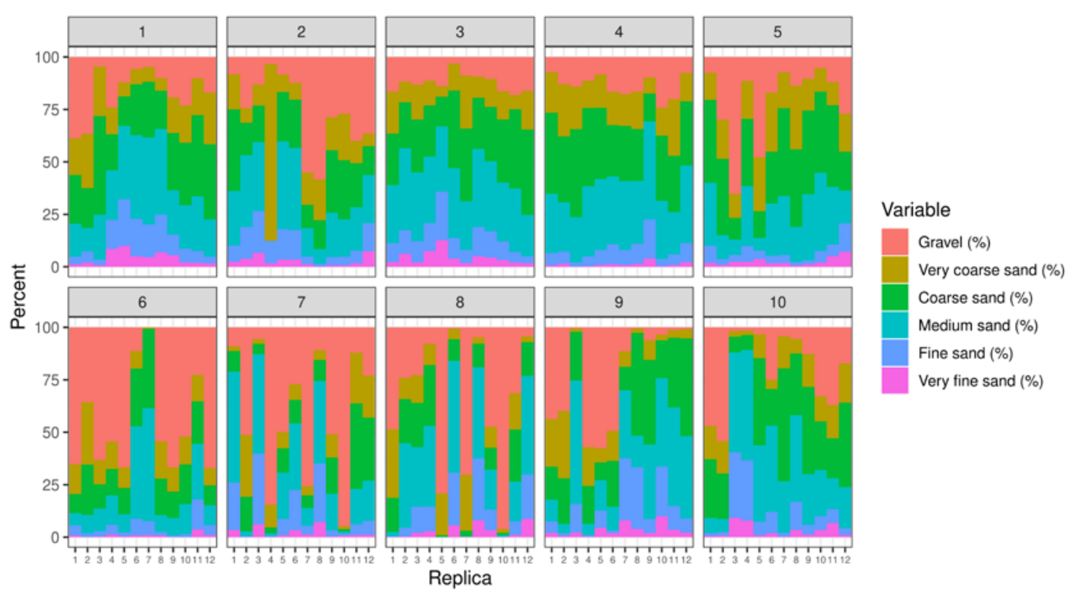

Figure 6. Composition of the river bed sediment in the ten areas classified as natural, altered, and impacted, according to their RHDI score, sampled between the months of June and July 2021 along the Cereja River, Pará, Brazil.

Table 4. Summary of the Permanova analysis for significant multivariate differences in water variables in the ten areas classified as natural, altered and impacted, according to their RHDI score, sampled between the months of June and July 2021 along the Cereja River, Pará, Brazil. df: degrees of freedom, SS: Sum of Squares, MS: Mean Square. P values in bold are significant.

Source of variation	df	SS	MS	pseudo-F	R^2	P	Source of variation	df	SS	MS	pseudo-F	R^2	P
Area	9	193798	21533	22.84	0.653	**0.001**	Class	2	158639	79319	66.80	0.535	**0.001**
Plot	3	2917	972	1.03	0.010	0.404	Plot	3	2914	972	0.8	0.010	0.540
Area:Plot	27	24417	904	0.96	0.082	0.542	Class:Plot	6	6847	1141	1.0	0.023	0.464
Residuals	80	75426	943		0.254		Residuals	108	128155	1187		0.432	
Total	119	296559			1.000		Total	119	296559			1.000	

4. DISCUSSION

The RHDI protocol provided a rapid and robust assessment of human impact on riparian habitat associated with macroinvertebrate structure in the Cereja River. However, the RHDI protocol was originally designed and applied in Cerrado headwater streams (Callisto et al., 2002) and there may be potential, as yet unknown, limitations to its use in streams in other biomes. Our results suggest it may be useful in Amazonian streams. Macroinvertebrates clearly responded to changes in structural diversity of the stream habitat, similar to other urban streams with aquatic faunas impacted by humans (Docile et al., 2016; Brito et al., 2021; de Paiva et al., 2021), thus functioning as robust bioindicators (de Faria et al., 2017; Giehl et al., 2020; Firmiano et al., 2021). Although the RHDI protocol is easy to carry out, evaluation of macroinvertebrate structure demands more time and skill.

Although the less urbanized areas (1 to 5) were all classified as natural, with a much more diverse benthic macroinvertebrate fauna, the RHDI score of Area 1 was lower than the others, despite being in a headwater. Thus, even in relatively more remote, less urbanized and apparently natural areas, stream habitat may be affected by human encroachment (Brito et al., 2021). This includes reductions in or removal of part of the riparian vegetation cover (Marmontel et al., 2018), trail opening along riparian forest, bathing of both humans and domestic animals, and washing bicycles, motorbikes, clothes and tableware. The RHDI score appears to be sensitive enough to identify such alterations at an early stage, which if left unchecked, may intensify and degrade even further the habitat in these areas. Although characteristics of first order stretches (Areas 1 to 4) may differ from those of second order stretches (Areas 5 to 10) and which affect fauna and habitat (Miserendino and Masi, 2010; de Paiva et al., 2021), RHDI score and classification decrease along the second order stretch associated with increasing urbanization and changes in fauna and habitat.

Better conserved riparian vegetation along the Cereja River, measured by the RHDI, is associated with greater macroinvertebrate diversity, especially of Ephemeroptera and Trichoptera, which were greatly reduced in abundance and diversity in altered areas, and absent from impacted areas, partly supporting our first hypothesis. Plecoptera were not found and have low diversity in tropical streams (de Paiva et al., 2017; Luiza-Andrade et al., 2020). In contrast to our predictions, higher Chironomidae and Coleoptera abundance were associated with high RHDI scores in natural areas. Coleoptera were exclusive to natural areas, especially Elmidae with higher abundance, and are sensitive to habitat changes (Segura et al., 2011). Despite some tolerance to impacts, Coleoptera appears to prefer more conserved habitat (Goulart and Callisto, 2003; Docile et al., 2016). High abundance of Chironomidae in the Cereja may be due to their diverse habitat preferences and feeding modes (Porinchu and Macdonald, 2003; Ferreira et al., 2021). Chironomidae is usually an abundant family with a variety of sensitive species, as well as several groups of species tolerant of environmental gradients, ranging from undisturbed to human impacted ecosystems (Heino and Paasivirta, 2008; Roque et al., 2010; Tang et al., 2010; Cortelezzi et al. 2020; Martins et al., 2021a) and having diverse ecological functions in streams (Biasi et al., 2010; Nicacio and Juen, 2015; Zequi et al., 2019; Camargo et al., 2019). Chironomid larvae are relatively difficult to identify to species (Milošević et al., 2014) and are usually identified to family, masking the sensitivity of genera and species to impacts (Cordeiro et al., 2016; Serra et al., 2017; Camargo et al., 2019; Zequi et al., 2019). Our results suggest potentially sensitive Chironomidae species in the Cereja River.

Conductivity in the Cereja River was higher than 100 µS/cm in downstream areas with lowest RHDI and dense urban settlements, reflecting impacted environments (Araújo and Oliveira, 2013; Menezes et al., 2016). Values of pH and turbidity were also highest in altered and impacted areas, with lowest RHDI scores, associated with increasing discharge of domestic effluents, since these variables are influenced by the transport and leaching of allochthonous materials (Gholizadeh et al., 2016), especially with urbanization (Menezes et al., 2016). Dumping of solid waste, sewage runoff and storm water exfiltration is common along the Cereja River (Guimarães et al., 2009; Monteiro et al., 2011; Sousa et al., 2016) and much of this enters the stream, especially in the rainy season, elevating dissolved salts and nutrient concentrations (Daniel et al., 2002; Nascimento et al., 2015).

In Amazon streams, conductivity, pH and turbidity are normally low (Batalha et al., 2014; Bertaso et al., 2015; de Paiva et al., 2017), due to rapid leaching of organic matter and acids and rapid absorption and recycling of nutrients in the riparian forest (Oliveira et al., 2009; Lopes and Magalhães, 2010; Brejão et al., 2021). The increase in conductivity, pH and turbidity from natural areas to impacted areas in the Cereja River is considered harmful to the macroinvertebrate community and may modify biological processes and interfere in aquatic photosynthetic processes (Nascimento et al., 2015).

Loss of riparian vegetation cover is associated with increased temperature, lower amounts of organic matter, decreased oxygen and consequent mortality of sensitive macroinvertebrates (Mesa 2014; Kusch, 2015; Lima et al., 2019; Dala-Corte et al., 2020). However, differently to what we predicted, despite decreasing riparian vegetation cover and increasing water temperature, even in areas with lower RHDI scores, dissolved oxygen concentrations did not decrease. Both buffering effects of forest cover on temperature in natural areas and physical turbulence, especially in altered areas, may help maintain and distribute oxygenated water along the entire stream. Oxygen concentrations along the Cereja were relatively low, median values between 4 and 5 mg/L, but variable in all areas, reaching 8 mg/L or more in natural areas. Reductions in dissolved oxygen in freshwater are generally due to organic matter decomposition, losses to the atmosphere by heating, respiration of aquatic organisms and oxidation of metal ions (Esteves, 2011; Lima et al., 2015; Jane et al., 2021) and in urban streams with organic pollution, they have serious consequences for taxa of sensitive macroinvertebrates (Batista et al., 2010; Lima et al., 2019).

Urban stream benthic macroinvertebrates are significantly impacted by sedimentation and siltation (Harding and Jellyman, 2015), but faunal diversity may not change between moderate levels of disturbance, as some fine sediment benefits certain taxa (Buendia et al., 2013). In our study, differently to what we predicted, the large decrease in macroinvertebrate diversity from natural areas to impacted areas was not associated with increasing fine sediments. However, our second hypothesis was partly supported since natural areas had, as predicted, higher sediment heterogeneity. Catchment modification, land use and local or stream reach habitat conditions and river flow runoff influence hydrodynamics and the particle size of sediments and their transport or deposition (Silva et al., 2007; Zerega et al., 2021). The highest percentages of gravel were found in altered and impacted areas, where accelerated erosive processes occur, associated with the removal of riparian vegetation (Martins et al., 2021b), lower RHDI scores and as a likely result of rapid washout, fine sand and very fine sand were consistently lower and highly variable in Altered and Impacted areas, respectively.

5. CONCLUSIONS

Natural areas with higher RHDI scores in the Cereja River have a higher diversity of macroinvertebrates, 12-21 taxa, especially Ephemeroptera and Trichoptera, which are sensitive to degraded environments. The degree of impact and urbanization increased downstream, associated with greater loss of riparian vegetation, verified by lower RHDI scores, lower macroinvertebrate diversity, and higher values of conductivity, pH, temperature, and turbidity. Chironomidae was the most abundant taxon in the study, especially in the natural areas, suggesting diversity in the group. The abundance of tolerant taxa in altered and impacted areas was associated with the above physicochemical conditions. Dissolved oxygen concentrations were relatively low and variable in the Cereja River, but did not decrease with urbanization and low RHDI. The sediment was more heterogeneous in upstream natural areas, more homogeneous with less sand and more gravel in central altered areas, and more heterogeneous in downstream impacted areas.

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CORRESPONDENCE TO

Luna Santiago

Alameda Leandro Ribeiro, s/n, 68600-000, Bragança, PA, Brasil.
e-mail: lunasantiagobio@gmail.com

AUTHORS' CONTRIBUTIONS

Luna Santiago: conceptualization (supporting), data curation (lead), formal analysis (supporting), investigation (lead), methodology (supporting), project administration (supporting), writing – original draft (lead), writing – review & editing (supporting).

Colin Robert Beasley: conceptualization (lead), data curation (supporting), formal analysis (lead), funding acquisition (lead), investigation (supporting), methodology (lead), project administration (lead), supervision (lead), writing – review & editing (lead).

DATA AVAILABILITY

All data used in this study are available on Figshare at https://doi.org/10.6084/m9.figshare.22677598.v1

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