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EPT distribution pattern in relation to environmental variables of urban streams in Western Amazonia

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Abstract: Urbanization is a phenomenon that has threatened the integrity of aquatic ecosystems around the globe, affecting the quality of life and biota of these ecosystems. Streams in urban areas are subject to contamination by garbage and sewage, as well as vegetation degradation, which can affect aquatic macroinvertebrate communities such as Ephemeroptera, Plecoptera and Trichoptera (EPT). Thus, in order to verify the influence of environmental variables on the distribution patterns of these communities and thereby assist in directing public policies to outline strategies for the conservation of local biodiversity, the EPT Index, Habitat Integrity Index, PERMANOVA and Main Coordinate Analysis (PCoA) were applied using environmental data and community structuring measures (abundance and diversity) sampled in 10 streams of the city of Porto Velho, RO, Brazil. According to our results, it was observed that the number of families and the abundance of EPT have a positive relationship with habitat integrity and a negative relationship with temperature increase, which reflect the need for environmental heterogeneity for EPT conservation. Therefore, our study provides subsidies to guide public policies for biodiversity conservation.

Keywords: EPT index; bioindicator; Amazon.

Resumo: A urbanização é um fenômeno que tem ameaçado a integridade de ecossistemas aquáticos em todo o globo, afetando a qualidade de vida e a biota desses ecossistemas. Nas áreas urbanas, os igarapés estão sujeitos à contaminação por lixo e esgoto, bem como degradação da vegetação, o que pode afetar as comunidades de macroinvertebrados aquáticos como Ephemeroptera, Plecoptera e Trichoptera (EPT). Assim, com o objetivo de verificar a influência de variáveis ambientais nos padrões de distribuição dessas comunidades e com isso auxiliar no direcionamento de políticas públicas para traçar estratégias de conservação da biodiversidade local, foi realizada a aplicação do Índice EPT, Índice de Integridade de Habitat, PERMANOVA e Análise de Coordenadas Principais (PCoA), utilizando os dados ambientais e medidas de estruturação comunidade (composição, abundância e diversidade) amostrados em 10 riachos da cidade de Porto Velho - RO. Segundo nossos resultados, foi observado que o número de famílias e a abundância de EPT possuem uma relação positiva com a integridade de habitat e relação negativa com o aumento da temperatura, que refletem a necessidade da heterogeneidade ambiental para a conservação de EPT. Sendo assim, nosso estudo forneceu subsídios para guiar políticas públicas para conservação da biodiversidade.

Palavras-chave: Índice EPT; Bioindicador; Amazônia.

1. Introduction

Urbanization is a phenomenon that has threatened the integrity of ecosystems across the globe, and its disorderly expansion results in habitat and biodiversity loss (Simkin *et al.*, 2022), as well as affecting human quality of life, occurring more markedly in some regions such as North and Latin America (UN DESA, 2019). Despite the population in Brazil being concentrated in large metropolises, urbanization has also occurred rapidly in the capitals of younger states located in the Amazon biome (Richards; VanWey, 2015), as is the case of Porto Velho, the capital of Rondônia State.

Urbanization advance has also affected aquatic ecosystems due to deforestation, pollution and eutrophication (among others), which cause changes in flow speed, high concentrations of nutrients and contaminants, and altered channel morphology and stability (Bohus *et al.*, 2023). Among these ecosystems, rivers and streams are home to a great diversity of aquatic animals, such as fish, amphibians and macroinvertebrates due to their variety of resources and substrates which provide a heterogeneous environment (Salles; Ferreira-Júnior, 2014). However, these characteristics can be threatened when exposed to anthropogenic pressures (Chase *et al.*, 2020; Santiago & Beasley, 2023).

Streams in urban areas are subject to waste disposal, sewage contamination, flow interruption, deforestation, and removal of riparian vegetation (Wilson *et al.*, 2021; Junk *et al.*, 2024). This degradation causes imbalances in the ecosystem and alters habitat quality parameters, such as physical-chemical variables of the water, nutrient load, stream flow, vegetation cover, and presence of riparian vegetation reflected in the structure of the aquatic biota (Calderon *et al.*, 2023). These characteristics contribute to reduce heterogeneity and resource availability, favoring generalist organisms that have greater tolerance and food plasticity, while specialists have their populations reduced or become extinct from the site (Mendes *et al.*, 2018).

Macroinvertebrates are represented by several groups of animals from the phyla Crustacea, Annelida, Mollusca and several orders of insects such as Ephemeroptera, Plecoptera and Trichoptera (EPT). Insects stand out as tools for environmental monitoring due to characteristics such as a shorter life cycle compared to other animals, their high abundance and habitat diversity (Hamada & Ferreira-Keppler, 2012). In addition, insects are noteworthy for their potential for rapid response (either directly or indirectly) to the type of disturbance and seasonality due to the presence of some organisms which are sensitive and others which are tolerant to changes in local environmental conditions (Martins *et al.*, 2014).

The Ephemeroptera, Plecoptera, and Trichoptera orders compose the EPT index, which is frequently used for environmental monitoring. Their abundance, richness, and composition vary according to environmental heterogeneity, temperature, and stream flow (Hamada & Ferreira-Keppler, 2012; Paprocki, 2012). As aquatic insects, they play an important ecological role in aquatic ecosystem structure, since they contribute to processing organic matter, nutrient cycling, energy flow, and maintenance of food webs, and depend on the aquatic environment for their development (Linares *et al.*, 2021).

Several studies, such as those by Martins *et al*. (2017) and Malacarne *et al*. (2024), have investigated the impacts of urbanization on EPT communities, revealing that although responses vary among taxa, environmental changes generally favor generalist groups, while resulting in a decrease in the presence of specialist species. Despite the growing recognition of the usefulness of EPT communities as bioindicators, the Amazon, especially the southwestern region of the Amazon basin, remains significantly undersampled. Few studies have explored the interactions of these communities with urban environments, which highlights an important gap in understanding the urbanization effects in this region.

In this study, we conducted a survey of the EPT community in 10 streams located in urban and periurban areas and in a Conservation Unit (CU) in the city of Porto Velho. The objective was to verify the influence of environmental variables on the distribution patterns of these communities and thus help guide public policies to outline strategies for conserving local biodiversity. To this end, we formulated the following questions: 1) Are there differences in environmental and biotic variables between urban and periurban areas? 2) Which environmental variables influence the abundance and diversity patterns of the EPT community?

2. Materials and Methods

2.1. Study area

The study area is located in the North region of Brazil in the Amazon biome, in the city of Porto Velho (Figure 1). It has an area of 34 thousand km², consisting of the largest territorial area of all Brazilian State capital cities. The city began on the banks of the Madeira River and has several microbasins distributed throughout the municipality. Its climate is Am according to the Koppen-Geiger climate classification system in a region of tropical monsoon climate, and contains Submontane Open Ombrophilous Forest, Lowland Open Ombrophilous Forest and Savanna/Ombrophilous Forest vegetation (IBAM, 2020). It is currently the most populous municipality in the state of Rondônia, with an estimated population of around 548,952 inhabitants. (IBGE, 2020).

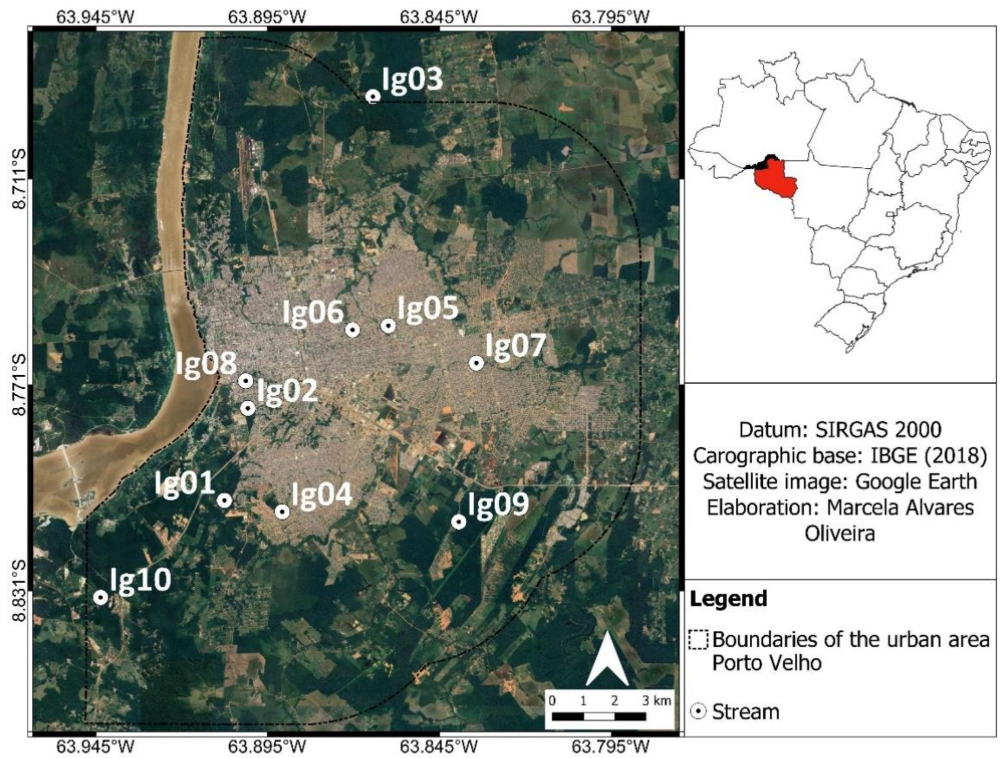


Figure 1. Geographical distribution of collection points in the city of Porto Velho, RO, Brazil.

There are five urban micro-basins whose courses are tributaries of the Madeira River and are the responsibility of the municipality, namely: the Riacho Grande basin with 1,327.84 ha, Riacho dos Tanques 2,196.84 ha, Riacho Tancredo Neves with 2,153.80 ha, Belmont with 2,456.93 ha and Bate-Estaca with 2,456.93 ha, located within the urban perimeter, as demonstrated in Silva (2020).

In turn, 10 streams were selected for this study (Figure 1, Table 1), classified as low-order streams (first, second and third order) by the methodology of Strahler (1952). Seven of the ten streams are located in urban areas and share similar characteristics in their surroundings, such as the presence of little or no vegetation with tree species in their surroundings, a large number of residences, the presence of garbage inside or outside the stream, and evidence of domestic and/or industrial effluent dumping. Two other streams are located in the periurban area of Porto Velho and one is located within the Raimundo Paraguassu de Oliveira Natural Park Conservation Unit (CU). The periurban streams and the one located within the CU are far from urban centers and have intact or little altered riparian forest with tree species, in addition to the absence of garbage inside or outside the stream and no evidence of domestic and/or industrial effluent dumping.

Table 1. Geographic coordinates of the collection points.

Igarapé	Area	Geographic coordinates	
		Latitude	Longitude
Ig_01	Urban	-8.804731	-63.907608
Ig_02	Urban	-8.777872	-63.900783
Ig_03	Unidade de Conservação	-8.687056	-63.864361
Ig_04	Urban	-8.808041	-63.890761
Ig_05	Urban	-8.755018	-63.870291
Ig_06	Urban	-8.756044	-63.859711
Ig_07	Urban	-8.764679	-63.834249
Ig_08	Urban	-8.769907	-63.901383
Ig_09	Periurban	-8.810971	-63.839371
Ig_10	Periurban	-8.833011	-63.943667

2.2. Sampling design

The study was conducted in 2022, with sampling during the rainy and dry seasons following an adaptation of the Chico Mendes Institute for Biodiversity Conservation's basic stream protocol of the Monitoring Program (Dantas *et al*., 2022).

To this end, a 100m stretch was established in each stream, in which the 0m, 50m, and 100m points were marked in the mouth-to-headwater direction. Data collection began after delimiting the stretch to be sampled. Macroinvertebrates were collected using a D-net for 1 minute at each point and stored in 80% alcohol. After collection, they were sorted at the Insect Biology and Diversity Laboratory of the Federal University of Rondônia and identified at the order level (for macroinvertebrates in general), and EPT individuals were identified at the family level using the identification key of Hamada *et al*. (2018). After identification, all EPT specimens were stored in glass vials with 70% alcohol.

2.3. Environmental variables

Measurements of water limnological variables such as temperature (°C), conductivity ($\mu S.cm^{-1}$), dissolved oxygen (DO) ($mg.L^{-1}$), pH, turbidity (NTU), and flow rate (m^3/s) were obtained at all points (0m, 50m and 100m). These variables were measured for both environmental characterization of the collection points and to verify possible relationships with the aquatic macroinvertebrate community structure. Physical variables such as canopy cover and Habitat Integrity Index were also obtained.

Canopy cover was obtained by analyzing photographs taken by a person positioned in the middle of the stream channel. The photographs were subsequently converted to monochromatic images using ImageJ software, coloring the vegetation black and the absence of vegetation indicated by the incidence of white light. Canopy cover was obtained by the ratio between the number of pixels in the black area and the total number of pixels in the image, expressed as a percentage.

Finally, the physical characteristics of the streams were assessed using a version of the Habitat Integrity Index (HII), adapted by Monteiro-Júnior *et al*. (2014), for urban environments. The index is composed of 12 metrics which provide a final score ranging from 0 to 1. Thus, the streams were classified into three integrity categories according to their score: scores below 0.33 were considered degraded; scores between 0.34 and 0.66 were considered intermediate; and scores above 0.67 were considered preserved.

2.4 Data analysis

Richness was calculated by adding the number of EPT families in each stream, and absolute abundance was represented by the total number of individuals in each order and family, while relative abundance is the ratio of the number of individuals in each order and family to the total number of individuals captured. Diversity was calculated using the Shannon-Wiener index (H') and dominance was calculated using the Simpson index.

The number of individuals in each order was used to apply the EPT index, which assesses the relative abundance of insects in the Ephemeroptera, Plecoptera and Trichoptera orders (which are the groups of organisms most sensitive to environmental changes) in relation to the total number of macroinvertebrates collected. The higher the frequency of EPT organisms, the more preserved the environment. The reverse is true for more degraded environments (Reyes & Peralbo, 2001).

Then, a PERMANOVA was performed to verify whether there is a significant difference in the environmental variables (limnological and physical) and composition matrix between the areas (urban and periurban), for which the stream located in the CU was considered to be in a periurban area. The canopy values were disregarded for the environmental variables during the analysis due to the collinearity between this variable and the HII. Next, a Principal Coordinates Analysis (PCoA) was conducted to verify the EPT community structure and its relationship with environmental variables between urban and periurban areas. The analysis was performed using the R software (version .4.4.0; R Core Team, 2024), with the vegan and ape packages for calculating and visualizing the ordination. The data were transformed using the Hellinger function, and then a dissimilarity matrix was constructed based on the Gower index to compare the EPT community between the different areas.

Finally, a simple linear regression was performed to verify the relationship between the variables analyzed and the diversity and abundance of EPT, in which the environmental variables represented the predictor variable and the abundance and diversity of EPT the response variables. The Habitat Integrity Index was considered as a predictor variable for temperature (response variable). All analyses were performed using the R 4.4.0 software considering each of the sampled streams as the sampling unit.

3. Results

3.1. EPT community structure

A total of 1,071 EPT individuals were counted from the total of 10,953 macroinvertebrates collected, of which 48.5% were from the order Ephemeroptera, 2.6% from Plecoptera and 48.8% from Trichoptera. The Trichoptera order presented the greatest taxonomic richness, with individuals from eight families being recorded, followed by Ephemeroptera with seven families and Plecoptera with one (Table 2).

Table 2. Taxonomic composition of EPT in urban streams of Porto Velho, RO, Brazil, identified at the family level. The values correspond to the number of individuals recorded in each sampled stream (Ig_01, Ig_03, Ig_05, Ig_08, Ig_09, Ig_10).

Family	Ig_01	Ig_03	Ig_05	Ig_08	Ig_09	Ig_10

Ephemeroptera						
Caenidae	21	1	48	0	5	0
Euthyplociidae	0	55	0	0	0	0
Leptophlebiidae	0	144	0	0	22	0
Leptohyphidae	0	59	0	0	2	0
Polymitarcyidae	0	2	0	0	1	0
Coryphoridae	0	6	0	0	0	0
Baetidae	3	95	4	18	32	2
Plecoptera						
Perlidae	0	28	0	0	0	0
Trichoptera						
Calamoceratidae	0	70	0	0	2	0
Helicopsychidae	0	91	0	0	0	0
Hydropsychidae	0	110	0	0	3	11
Polycentropodidae	0	32	0	0	4	38
Glossosomatidae	0	118	0	0	0	0
Odontoceridae	0	7	0	0	2	0
Leptoceridae	0	14	0	0	2	0
Hydroptilidae	0	7	0	0	0	0

EPT individuals were recorded in six of the 10 streams sampled (Ig01, Ig03, Ig05, Ig08, Ig09 and Ig10). Only the Baetidae (Ephemeroptera) family was recorded in stream Ig08, and the Perlidae (Plecoptera) family was restricted to stream Ig03. The Hydropsychidae family was the most abundant for Trichoptera, with 88.7% of its individuals recorded in Ig03 (Natural Park), followed by Glossosomatidae (118) and Helicopsychidae (91), all restricted to the Conservation Unit.

The Ephemeroptera order was most widely distributed among the EPTs, since ephemeropterans were present in six streams, three of which are urban. According to the results in Table 2, it is possible to observe that the abundance of the Caenidae (Ephemeroptera) family was concentrated in streams Ig01 and Ig05.

The Shannon and Simpson indices showed that stream Ig03 was the most diverse and abundant, followed by Ig09, with 16 and 10 families respectively (Table 3).

Table 3. EPT diversity indices in urban streams of Porto Velho, RO, Brazil.

	Ig01	**Ig03**	**Ig05**	**Ig08**	**Ig09**	**Ig10**
No. of individuals	24	839	52	18	75	51
Richness	2	16	2	1	10	3
Shannon	0.38	2.35	0.27	0	1.63	0.68

Only one stream was classified as Regular (Ig03) regarding the EPT Index, while all the other nine streams were classified as Poor, and four of these nine streams did not have any records of EPT individuals.

Table 4. Water quality of streams by EPT index.

	∑EPT	**EPT%**	**Water quality**
Ig_01	24	1.69	Poor
Ig_02	0	0	Poor
Ig_03	839	39.74	Regular
Ig_04	0	0	Poor
Ig_05	64	7.4	Poor
Ig_06	0	0	Poor
Ig_07	0	0	Poor
Ig_08	18	2.29	Poor
Ig_09	75	6.93	Poor
Ig_10	51	11.49	Poor

[1] ∑EPT is the sum of Ephemeroptera, Plecoptera and Trichoptera; EPT% is the proportion of Ephemeroptera, Plecoptera and Trichoptera in relation to the total sample.

According to PERMANOVA, there was a significant difference in the EPT composition between urban and periurban areas (P=0.031 and R^2=0.1425).

3.2. Environmental variables

Among the streams with EPT, the average results obtained through the limnological variables had a minimum of 6.42 (Ig03) and a maximum of 7.96 (Ig04) for pH; 0.02 μS.cm^{-1} (Ig03 and Ig09) and 0.47 μS.cm^{-1} (Ig04) for conductivity; temperature with a maximum of 30.4°C (Ig04) and a minimum of 23.5°C (Ig09); DO with a minimum of 5.05 mg/L (Ig06) and a maximum of 7.3 mg/L (Ig02) (Table 5).

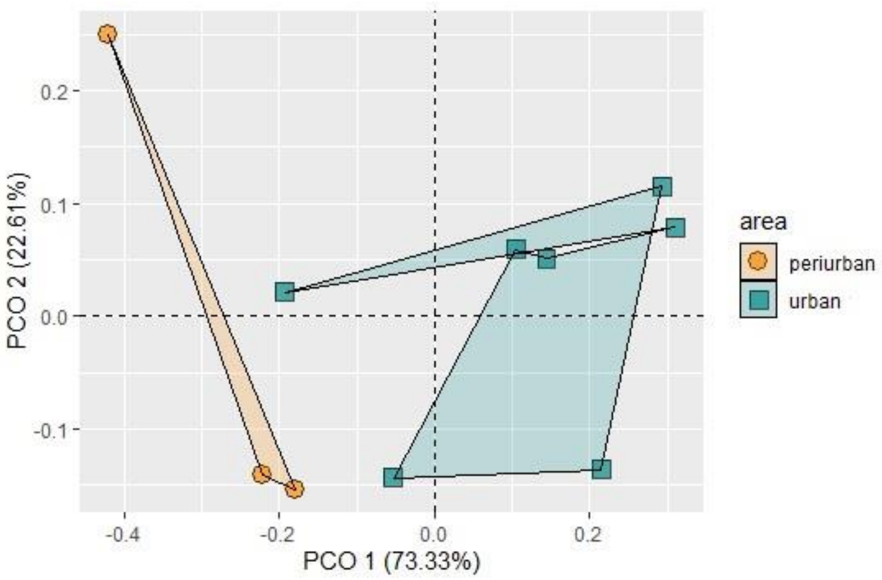
A significant difference was obtained among the environmental variables during the comparison of the environmental variables between the areas (urban and periurban) (P=0.013, R^2=0.4858).

Table 5. Environmental variables in the streams of Porto Velho, RO, Brazil.

Full variables	**Ig01**	**Ig02**	**Ig03**	**Ig04**	**Ig05**	**Ig06**	**Ig07**	**Ig08**	**Ig09**	**Ig10**
pH	7.07	7.64	6.42	7.96	6.89	7.17	7.76	6.76	7.09	7.26
Cond	0.25	0.23	0.02	0.47	0.39	0.36	0.44	0.21	0.02	0.26
Temp	27.55	27.6	24.6	30.4	26.95	29.55	30.35	28.05	24.5	25.7
OD	5.5	7.3	6	6.45	7.05	5.05	5.8	6.1	6.45	6.6
Turb	8.72	38.25	0	32.55	9.75	17.05	49.35	34.85	10.46	2.92
Flow	0.18	0.44	0.02	0.06	0.01	0.06	0.14	0.08	0.15	0.14
HII	0.364	0.364	0.854	0.159	0.267	0.139	0.153	0.223	0.678	0.688
Canopy	33.23	26.73	88.09	0	60.85	0	23.15	8.33	73.65	59.06

[1] Cond - conductivity (μS.cm^{-1}); Temp - temperature (°C); DO – dissolved oxygen (mg.L^{-1}); Turb - turbidity (NTU); Flow rate (m^3/s); Canopy cover (%).

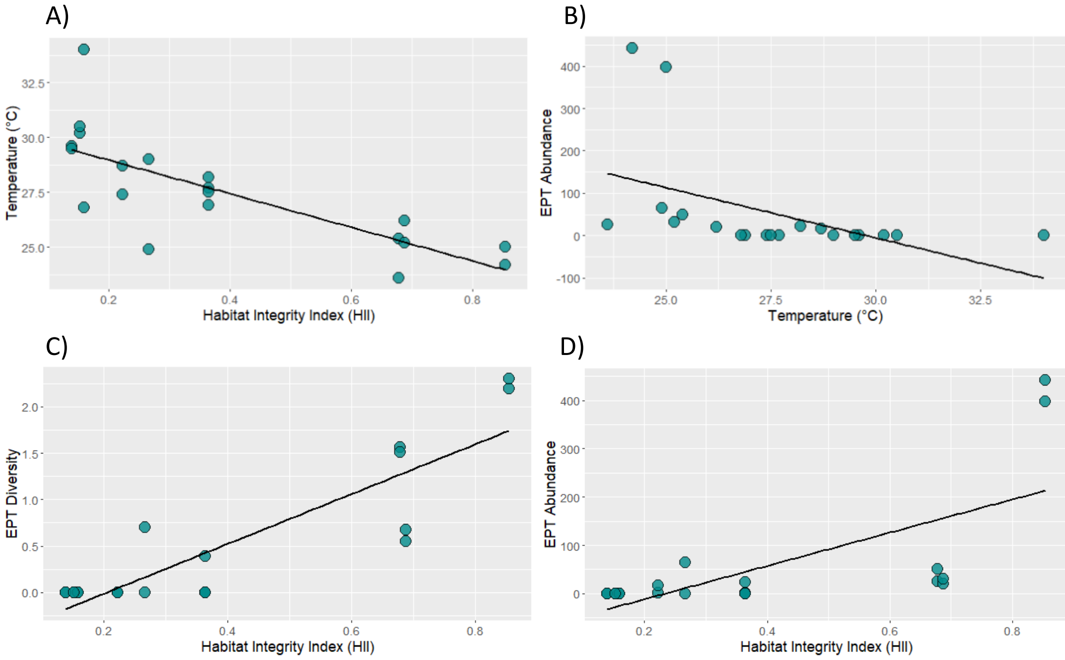
Principal Coordinate Analysis (PCoA) performed on community abundance data revealed that the first two axes explained 95.94% of the total variation in the data (Axis 1: 73.33%, Axis 2: 22.61%), indicating that these two components are sufficient to accurately represent the community structure between urban and periurban areas. This high explanation of variation suggests that the observed separation between areas can be well understood from these two dimensions. The samples were clearly grouped along these axes, reflecting the differences in the variables studied, as can be seen in graph 1 below.



Graph 1. Principal Coordinates Analysis (PCoA) applied to the EPT community between urban and periurban areas of Porto Velho, RO, Brazil.

3.3. *Relationship between environmental variables and the EPT community*

Linear regression showed a positive relationship between HII and abundance (p=0.000859, R^2=0.469) and EPT diversity (p=3.67e-07, R^2=0.77) (Graph 2). In addition, HII showed a significant negative relationship with temperature values (p=9.5e-05, R^2=0.557). Considering temperature as an independent variable, there was a significant and negative relationship between temperature and EPT abundance (p=0.0340, R^2=0.226).



Graph 2. Linear regression. A) Habitat Integrity Index (HII) as a predictor variable for Temperature (°C) with a significant and negative relationship (p=9.5e-05, R^2=0.557). B) Temperature (°C) as a predictor variable for EPT Abundance with a significant and negative relationship (p=0.0340, R^2=0.226). C) HII as a predictor variable for EPT Diversity with a significant and positive relationship (p=3.67e-07, R^2=0.77). D) HII as a predictor variable for EPT Abundance (p=0.000859, R^2=0.469).

4. Discussion

The results showed that habitat quality (HII) and environmental characteristics such as temperature and canopy cover are important for maintaining EPT diversity and abundance. It was observed that EPT richness and abundance were higher in streams with high environmental integrity levels, especially in periurban areas (Table 3). The relationship between canopy and diversity maintenance is known, since the presence of riparian forest and riparian vegetation promotes maintenance of microhabitats (Marques *et al*., 2021) and temperature (Santiago & Beasley, 2023). This pattern is corroborated by the strong positive relationship between HII and EPT abundance and diversity, evident in stream Ig03 because it has the highest coverage, lowest temperature and high diversity and abundance of these organisms (see Figure 3). This reinforces the importance of maintaining riparian forests, even in urban and periurban areas, as a focus for minimum biodiversity conservation.

In addition to the significant influence of variables related to the canopy and temperature in streams in urban areas, reduced EPT abundance, even in areas with high conservation levels, may also be influenced by exposure to contamination sources such as sewage and solid waste, which threaten the sampled points (Catâneo *et al*., 2024). This may justify the results obtained by the EPT Index, which categorized preserved streams as having poor and regular quality (Ig03, Ig09, and Ig10) in terms of water quality (Table 4). However, the relevance of this interference needs to be further investigated.

Environmental quality influenced the taxonomic composition, since more preserved environments presented greater diversity, as shown by the regression results (Graph 2), while low diversity or absence of EPT in degraded streams in urban areas is associated with factors such as lower integrity and high temperatures. These results corroborate the findings of De Paiva *et al*. (2021) and Malacarne *et al*. (2024), who reported a reduction in EPT diversity and abundance in streams with greater degradation and land use impact. This highlights the vulnerability of EPT to environmental degradation and their use as bioindicators.

Ephemeroptera was more widely distributed among the sampled streams, including in urban areas, indicating a relative tolerance of some families (such as Caenidae) to impacted conditions. The occurrence of the Caenidae family (Ephemeroptera) concentrated in the Ig01 and Ig05 streams can be attributed to the preferences of this family (belonging to the scavenger functional group) for lentic environments with fine sediment, which provides both shelter and food (Merritt, Cummins and Campbell, 2014; Santos *et al*., 2024).

The Plecoptera and Trichoptera orders showed similar responses, since both had their distribution restricted to preserved environments with greater canopy coverage and reduced temperatures. However, Plecoptera was more sensitive, with its occurrence restricted to the stream located in the CU. This may be related to the fact that the order is recognized as "anti-tropical" (Lecci and Duarte, 2024), requiring lotic environments with high dissolved oxygen concentrations and substrate diversity (Azmi, Hussin and Amin, 2018).

All families for Trichoptera were recorded in the three streams categorized as preserved. This highlights the importance of substrate diversity for the group, which is related to the presence of allochthonous material offered by the presence of preserved riparian forest, favoring construction of different types of shelters using leaves, branches and silk for the order, in addition to providing food diversity for the organisms (Santos *et al*., 2024). The grouping of Hydropsychidae, Glossosomatidae, Calamoceratidae, Odontoceridae (Trichoptera) families, as well as Euthyplociidae and Leptophlebiidae families (Ephemeroptera) in streams with greater environmental integrity suggests that these groups are sensitive to environmental changes.

Therefore, the presence of riparian vegetation and riparian forests is essential to measure EPT diversity and distribution in streams, as they act as a filter for pollutants and as a source of allochthonous matter (Leal *et al*., 2016; Luiza-Andrade *et al*. 2020; Lima *et al*., 2022). Changes in land use with the removal of riparian forests consequently cause greater sunlight incidence in water bodies, increasing the temperature and negatively affecting EPT communities. This suggestion is confirmed by the results of the linear regression, which indicated a significant and positive relationship between integrity and abundance and diversity, as well as a significant and negative relationship between temperature and abundance of EPT.

Our results are corroborated by those of Lima *et al*. (2022), in which the increase in temperature and deforestation also negatively influenced EPT communities, reducing the most sensitive groups. However, streams such as Ig04 and Ig06, which did not present any vegetation cover and consequently high temperatures, in addition to the large discharge of effluents observed *in loco* and described by Catâneo *et al*. (2024), made conditions unfavorable even for the most tolerant groups, explaining the absence of EPT in these locations.

5. Conclusion

Our study demonstrated that the EPT group responds negatively to environmental impacts, such as reduced environmental integrity and increased temperature. This indicates that streams can maintain relative EPT diversity and abundance if their integrity is preserved, with emphasis on maintaining the riparian forests of urban and periurban streams.

From a conservationist perspective, this information is important to maintain the health of streams and positively influence their overall health through integrated management with current legislation such as Complementary Law No. 97 of 1999, which regulates land use in Porto Velho, and Law 12.651 of 2012, which establishes guidelines for permanent preservation areas. This guides stakeholders towards restorative actions for urban streams, in which adopting the structure of periurban streams can be a first step.

However, the influence of the discharge of domestic effluents and solid waste needs to be better evaluated, since chemical factors can alter abundance and diversity without necessarily altering the structural integrity analyzed herein.

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