

Environment

Biodiversity in context: an analysis between urban areas and a forest reserve

Biodiversidade em contexto: uma análise entre áreas urbanas e uma reserva florestal

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ABSTRACT

Biological protection areas are a crucial refuge for biodiversity, especially due to the growing populations in cities. Generally, these areas are located far from urban centers in an attempt to avoid the effects of urbanization. However, does a reserve on the outskirts of the largest urban area in the Amazon also contribute to species conservation? Our research assessed the loss of aquatic macroinvertebrate biodiversity in streams of an urban area in development, adjacent to a biological protection area—the Ducke Reserve, a tropical rainforest reserve in the municipality of Manaus, AM - Brazil. We sampled 18 streams in the Ducke Reserve and 18 streams in the urban area. The communities between the urban area and Ducke Reserve showed no similarity ($M2 = 0.917$; correlation = 0.287; $p = 0.423$). A total of 107 taxa were found exclusively in the Ducke Reserve, while seven taxa were exclusive to the urban area of Manaus, with these seven being characteristic of disturbed environments. The low richness ($w = 320$; $p = 0.001$) and abundance ($w = 242$; $p = 0.01$) of macroinvertebrates in the urban area resulted in a 91% loss of diversity. Urbanization is a major driver of environmental change, affecting both aquatic and terrestrial ecosystems. Thus, we highlight that the Ducke Reserve plays a significant role in maintaining Manaus' biodiversity, and together with biological protection areas, represents our best hope for achieving global biodiversity conservation goals.

Keywords: Aquatic environments; Aquatic macroinvertebrates; Protected areas; Urbanization

RESUMO

As áreas de proteção biológica são refúgios cruciais para a biodiversidade, especialmente diante do crescimento populacional nas cidades. Em geral, essas áreas estão localizadas longe dos centros urbanos, como forma de evitar os efeitos da urbanização. No entanto, uma reserva situada nos

arredores da maior área urbana da Amazônia também contribui para a conservação de espécies? Nossa pesquisa avaliou a perda de biodiversidade de macroinvertebrados aquáticos em igarapés de uma área urbana em expansão, adjacente a uma área de proteção biológica — a Reserva Ducke, uma reserva de floresta tropical no município de Manaus, AM - Brasil. Amostramos 18 igarapés na Reserva Ducke e 18 igarapés na área urbana. As comunidades entre a área urbana e a Reserva Ducke não apresentaram similaridade ($M^2 = 0,917$; correlação = 0,287; $p = 0,423$). Um total de 103 táxons foi registrado exclusivamente na Reserva Ducke, enquanto sete táxons foram exclusivos da área urbana de Manaus, sendo estes característicos de ambientes perturbados. A baixa riqueza ($w = 320$; $p = 0,001$) e abundância ($w = 242$; $p = 0,01$) de macroinvertebrados na área urbana resultou em uma perda de 91% da diversidade. A urbanização é um dos principais vetores de mudança ambiental, afetando tanto os ecossistemas aquáticos quanto terrestres. Assim, destacamos que a Reserva Ducke exerce um papel significativo na manutenção da biodiversidade de Manaus e, juntamente com outras áreas de proteção biológica, representa nossa melhor esperança para alcançar as metas globais de conservação da biodiversidade.

Palavras-chave: Ambientes aquáticos; Macroinvertebrados aquáticos; Áreas protegidas; Urbanização

1 INTRODUCTION

The expansion of unplanned cities leads to the discharge of domestic and industrial wastewater, which is often released directly or indirectly into nearby watercourses without treatment (Sousa et al., 2016; Ferreira et al., 2021). Additionally, urbanization increases deforestation rates, resulting in soil erosion due to wind and rain, which in turn causes soil leaching into aquatic systems and sedimentation of these systems (Couceiro et al., 2007; Martins et al., 2017; Bashir et al., 2020). These issues have already been observed in Manaus (Couceiro et al., 2007; Luo et al., 2018).

Manaus is the largest city in the Brazilian Amazon, with a population of 2,532,226 as of 2022. It is the eleventh most populous city in Brazil and the fourth largest urban area in the country, covering 427 km², with 94% of the municipality's population residing in the urban area. Manaus serves as a center for commerce and industry (Manaus Free Trade Zone) (Viana et al., 2012; Silva et al., 2023). Disorganized urban expansion has had significant impacts on both terrestrial and aquatic environments in Manaus, contributing to the loss of local and regional biodiversity (Couceiro et al., 2007; Silvestrim et al., 2021).

To minimize biodiversity loss in the metropolitan region of Manaus, the Adolpho Ducke Forest Reserve (or simply Ducke Reserve) was established in 1962 to preserve a fragment of the Amazon rainforest (100 km² of primary tropical rainforest) and its diversity for the future (Hopkins, 2005). Studies have shown that protected areas serve as sources of species for nearby areas where these species cannot reproduce, thereby maintaining a minimum level of biodiversity (Gutiérrez-Cánovas et al., 2013; Guareschi et al., 2015; Bergamin et al., 2017; Cetra et al., 2022). It is anticipated that in the near future, the Ducke Reserve will become completely isolated from the continuous forest, surrounded by urbanization (Hopkins, 2005).

With anthropization, biodiversity is severely impacted, as it causes the homogenization of environmental characteristics, which in turn act as filters in species selection (Mori et al., 2018; Chase et al., 2020). Homogeneous environments tend to show a reduction in species composition, with a significant loss of species, while more heterogeneous areas maintain a more consistent biodiversity, where species turnover/substitution is observed (Legendre, 2014; Bergamin et al., 2017). Thus, biological protection areas play a crucial role in preserving biodiversity, serving as refuges for a wide range of species, allowing them to thrive and maintain natural populations and habitats (Azevedo-Santos et al., 2019). Biological protection areas help safeguard threatened and/or endangered species, ensuring they have a safe place to reproduce, additionally, biological protection areas provide settings for scientific research and environmental monitoring, the use of natural resources by local communities, environmental education, and raising awareness among surrounding populations about the importance of preservation (Bryan et al., 2010; Dale et al., 2020; Lukk et al., 2023).

Human activities related to urban growth cause the decline of aquatic macroinvertebrates, as they are sensitive to habitat loss and environmental changes (Couceiro et al., 2007; Martins et al., 2017; Brasil et al., 2020; Souza et al., 2023). Conversely, the loss of these organisms significantly impacts ecosystems, considering

their important roles in maintaining ecological balance and water quality (Fayomi et al., 2019; Häder et al., 2020). One of the critical roles this group plays is in nutrient cycling, helping to decompose organic matter (Vanni, 2002; Kuntz and Tyler, 2018). Additionally, the most significant impacts can be observed in the food chain, as macroinvertebrates are a vital food source for fish and other aquatic animals that depend on them for protein and other nutrients (Agouridis et al., 2015).

Despite this, it is worth noting that biomonitoring studies have used macroinvertebrates to observe the effects of urbanization on landscapes, as many groups have specific requirements for their occurrence and are highly sensitive to environmental changes (Morse et al., 2007; Sarkar et al., 2021; Nair et al., 2015; Suter and Cormier, 2015). Some species of macroinvertebrates require high levels of dissolved oxygen to survive or greater water flow, while others are more tolerant and can survive in both pristine and impacted locations (Muzón et al., 2019; Brasil et al., 2020).

Urban expansion has emerged as one of the main drivers of biodiversity loss, particularly affecting freshwater ecosystems (Couceiro et al., 2007; Ferreira et al., 2021). As cities grow, aquatic habitats are increasingly subjected to physical alterations, pollution, and habitat fragmentation, which in turn lead to declines in the diversity and abundance of aquatic organisms, especially macroinvertebrates (Rodríguez-Rodríguez et al., 2019). Macroinvertebrates are key components of freshwater ecosystems, playing fundamental roles in nutrient cycling, organic matter decomposition, and supporting higher trophic levels (Morse et al., 2007; Agouridis et al., 2015). However, their sensitivity to environmental degradation makes them particularly vulnerable to the impacts of urbanization (Agouridis et al., 2015; Ferreira et al., 2021). In this context, understanding how urban expansion affects macroinvertebrate assemblages is crucial for biodiversity conservation and ecosystem management, especially in megadiverse regions such as the Amazon (Silvestrim et al., 2021).

In light of this, the central ecological question of this study is: To what extent does urbanization contribute to the loss of macroinvertebrate diversity in streams,

and how does this compare to the diversity maintained within a protected area? Thus, the objective of this study is to assess the extent of biodiversity loss in urban areas compared to a protected area. We propose the following hypotheses: (i) There is no congruence between the composition of macroinvertebrates in the protected and urban areas, given the benefits of protected areas for maintaining biodiversity; (ii) The richness, abundance and diversity of macroinvertebrate species are greater in the protected area than in the urban area, given that the PAs have a greater quantity of microhabitat and are able to support greater richness, abundance and diversity than urban areas.

2 MATERIALS AND METHODS

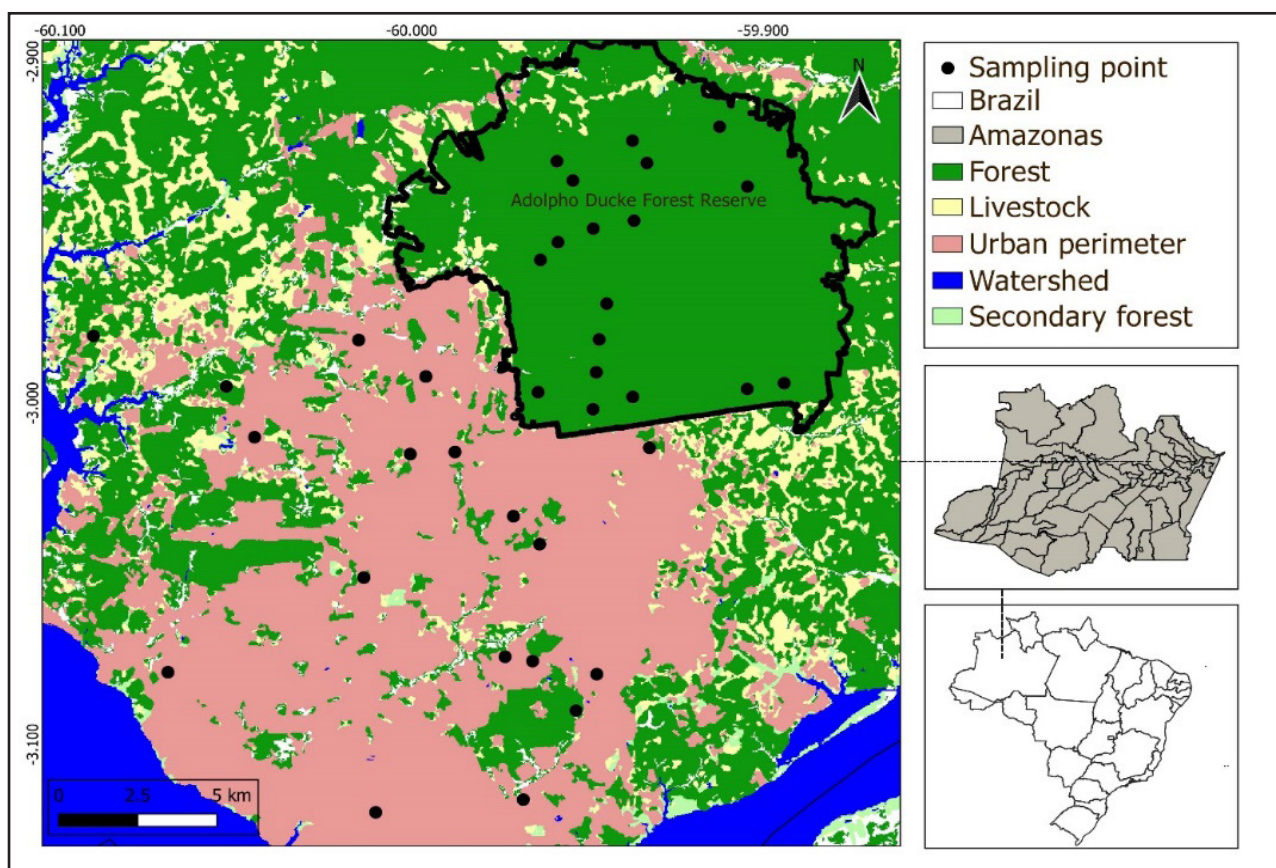
2.1 Study Area

The collections were carried out in 36 locations in the urban area (n = 18 - in August 2010) and in the Ducke Forest Reserve (n = 18 - in August 2015), located in the city of Manaus, Amazonas, Brazil (Figure 1). In each stream a 50-meter transect was delimited and subdivided into 5 segments of 10 meters each, applying the scanning methodology in areas where in each segment all the substrates present were sieved, three times each, in search of specimens. The collection was standardized with only one collector using an entomological net. After collection, the specimens were conditioned in 70% alcohol and taken to the laboratory for identification. The urban streams present great modifications of the natural conditions due to being within the urban perimeter, while the streams of the Adolpho Ducke Forest Reserve present better environmental conditions and are not accessible to the general public, being accessed only by small trails opened for this study. The region's climate is classified as Equatorial Hot and Humid according to the Köppen classification (Peel et al., 2007), with two distinct seasons: a rainy season from November to June and a dry season

from July to October. The average annual precipitation is 2286 mm, with average temperature variations ranging from 23.3° C to 31.4° C.

The rapid urban expansion has caused significant degradation of the local vegetation, leading to an increase in temperature (Viana et al., 2012). Deforestation for infrastructure construction and the unplanned growth of neighborhoods result in the loss of vegetation cover, threatening biodiversity and reducing ecosystem services (Silva et al., 2023). In contrast, the Adolpho Ducke Reserve is a well-preserved area of 10 km², with ombrophilous vegetation and a uniform canopy averaging 32 meters in height. The topography consists of plateaus intersected by streams, which originate within the reserve (Sousa et al., 2016).

Figure 1 – Collection points in the urban perimeter and in the Adolpho Ducke Forest Reserve in the municipality of Manaus, Amazonas, Brazil



Source: Authors (2025)

2.2 Data Analysis

To measure the collection efficiency in both the Adolpho Ducke Forest Reserve and the urban zone of Manaus, we conducted an online interpolation and extrapolation analysis using iNext (iNterpolation and EXTrapolation: [<https://chao.shinyapps.io/iNEXTOnline/>]) (<https://chao.shinyapps.io/iNEXTOnline/>) for each of the matrices.

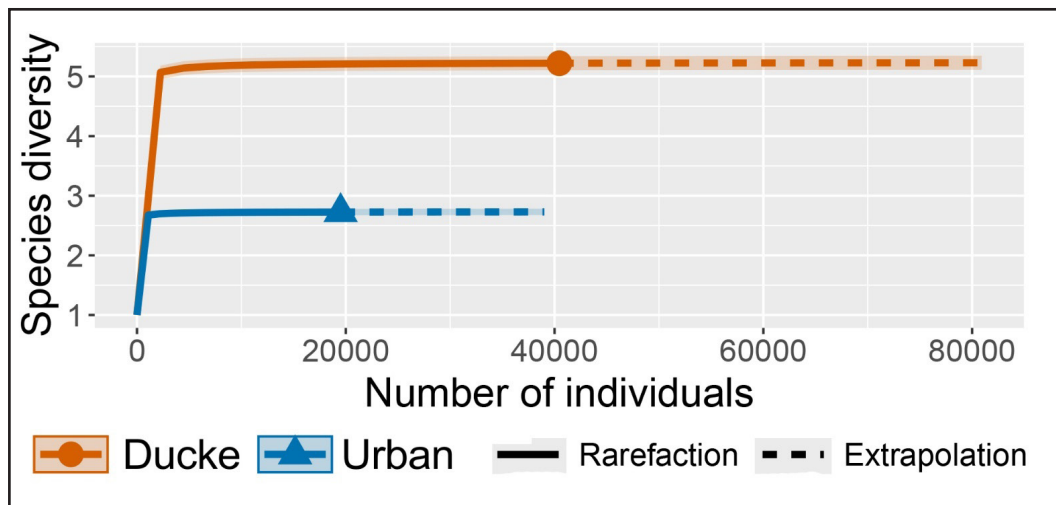
To test the first hypothesis, we performed Principal Coordinate Analysis (PCoA) for both abundance matrices (data from the Ducke Reserve and the urban area of Manaus). The abundance data were log-transformed, then using the Bray-Curtis distance matrix to obtain the PCoA axes (Ricotta and Podani, 2017). After obtaining the ordination results, the first axes of the PCoAs were used to test the congruence of the composition through the Procrustes (Procrustean Randomization Test) – using 10,000 permutations – which measures the significance of the m^2 statistic and the level of similarity between the compared ordinations (Juen et al., 2013; Shimano et al., 2018). To test the second hypothesis, even after transforming the data using log+1, the assumptions of normality and homogeneity were not met. Therefore, we conducted non-parametric Wilcoxon/Mann-Whitney tests to compare the richness and abundance between the areas (protected area vs. urban zone). To assess the diversity of the two matrices (Ducke and Urban), we used the Shannon index and Hill numbers (Hill, 1973; Buckland et al., 2005). The Shannon index (H') and Hill numbers are tools commonly employed to measure species diversity and evenness. H' quantifies diversity by accounting for both the number of species and their relative abundance, while Hill numbers provide a more comprehensive measure of diversity. All statistical analyses were performed in the R environment, version 4.2.2 (R Core Team, 2022) using the vegan package.

3 RESULTS

A total of 58,710 specimens were collected, distributed across 160 families and/or genera. In the Ducke Reserve, 39,268 individuals were collected, while in the urban zone, 19,442 individuals were collected. Of the 160 identified groups, 153 occurred in the Ducke Reserve and 56 in the urban zone, with Chironomidae being the most abundant group in both the Ducke Reserve ($n = 29,278$) and the urban zone ($n = 13,209$). Among these, 103 groups were exclusive to the Ducke Reserve, while seven occurred only in the urban zone (Amanahyphes, Dolichopodidae, Ephydriidae, Libellulidae sp., Naididae, Stratiomyidae, and Syrphidae). iNext showed a collection efficiency of 98% in the Ducke Reserve and 99% in the urban zone, with an estimated richness of 169.9 species for Ducke and 70.03 for the urban area (Figure 2). Where we can observe almost three times more wealth and twice more abundance in the Ducke Forest Reserve compared to the urban area.

Procrustes analysis revealed no similarity between the macroinvertebrate communities in the sampled streams ($M^2 = 0.917$; correlation = 0.287; $p = 0.423$). The Procrustes analysis indicated a poor fit between the two datasets, and the relationship was not statistically significant, indicating a lack of congruence between them. The Wilcoxon/Mann-Whitney tests showed a significant difference in richness ($W = 320$, p -value < 0.001) and abundance ($W = 242$, p -value = 0.01) between the studied areas. The Shannon index and Hill numbers for the Ducke Reserve were 1.652 and 5.222, respectively, whereas for the urban zone they were 1.001 and 2.725, respectively, indicating that the reserve exhibits greater diversity.

Figure 2 – Rarefaction and extrapolation curve corrected by Hill numbers for specimens collected in the Ducke Reserve and urban area of Manaus



Source: Authors (2025)

4 DISCUSSION

Our analyses demonstrated that there was no similarity in macroinvertebrate species composition between the urban zone and the Ducke Reserve, thereby supporting our first hypothesis. We observed a low correlation value between the species matrices and a loss of more than 90% of the matrix information, meaning that 91% of the biological information is lost when comparing the urban zone with the Ducke Reserve. This indicates that the Ducke Reserve plays a crucial role in maintaining local biodiversity.

Our second hypothesis was also confirmed, as there was greater richness, abundance and diversity of macroinvertebrates in the Ducke Reserve compared to the urban area. The Ducke Reserve has more than double the number of individuals and identified groups compared to those found in the urban zone of Manaus. Taxa such as *Corydalus* and *Protosialis*, belonging to the order Megaloptera, and *Anacroneuria* and *Macrogynoplax*, from the order Plecoptera, which are indicators of good environmental conditions and require high levels of dissolved oxygen (Savic et al., 2017; Tubić et al., 2024), were only collected in the Ducke Reserve.

Some studies suggest that richness and abundance may not always be the best metrics for monitoring studies (Calvão et al., 2016; Pereira-Moura et al., 2021); however, when the degree of alteration is so substantial, it becomes nearly impossible not to observe significant differences in these metrics, which in this case are also associated with bioindicator species of preserved environments.

Urbanization is a major driver of environmental change, impacting both aquatic and terrestrial ecosystems (Martins et al., 2017; Fayomi et al., 2019; Bashir et al., 2020; Häder et al., 2020). Couceiro et al. (2007) and Martins (2013) have highlighted significant degradation in the aquatic environments of the urban zone of Manaus, caused by deforestation and domestic sewage, as well as greater richness in preserved environments.

Freshwater ecosystems rely heavily on macroinvertebrates for their maintenance and functioning (Kuntz and Tyler, 2018; Häder et al., 2020). Human activities pose a significant threat to aquatic biodiversity (Martins et al., 2017; Bashir et al., 2020), and current socioeconomic development shows little indication of reducing these impacts in the near future (Bernhardt et al., 2008; McGrane, 2016; Häder et al., 2020), especially in the urban area of Manaus. Impacts on macroinvertebrates have been documented since 2007, and re-sampling ten years later indicates that no changes have occurred.

Biological protection areas help address part of this problem by playing a critical role in preservation and ensuring local biodiversity (Azevedo-Santos et al., 2019). Among the many benefits these areas provide to macroinvertebrates, one of the most significant is the protection of natural habitat (Lukk et al., 2023). By providing a safe refuge for threatened species, they help prevent declines in biodiversity (Agouridis et al., 2015).

Biological protection areas act as a buffer against the impacts of climate change, such as rising temperatures and flood pulses that increase water levels (Malhi et al., 2020). They contribute to ecosystem resilience and also serve as a source of individuals for other habitat patches (Wintle et al., 2019). To enhance their effectiveness, ecological corridors, buffer zones, or other means should be used to connect biological protection areas,

allowing species to move freely between areas, providing larger habitats, and maintaining genetic diversity (Ćurčić and Đurđić, 2013; Stewart et al., 2019; Isola et al., 2022). Therefore, investing in the connectivity of protected areas is recommended to ensure their long-term sustainability (Newmark, 1993; Stewart et al., 2019; Kietzka et al., 2021).

Local management plans often emphasize the protection of charismatic species, whose conservation also benefits a wide range of other species (Albert et al., 2018). However, it is important to recognize that management goals set for specific species do not always benefit other species and, in some cases, may even threaten the survival of others (Courchamp et al., 2003; Le Saout et al., 2013).

It is important to consider that these areas provide significant economic benefits to local communities through the sustainable use of resources and responsible tourism, which, when combined, ensure food security and promote the long-term conservation of natural resources (Forje et al., 2021; Xu et al., 2021). However, optimizing the management of protected areas is vital to improve the overall performance of the global network (Cuthbert et al., 2021, 2022). To effectively maintain the biodiversity they host, it is essential to manage and fund these areas appropriately (McCarthy et al., 2012; Xu et al., 2021), strategically considering the biodiversity of each location (Bryan et al., 2018; Wu et al., 2019).

5 CONCLUSION

Protected areas are vital for conserving both the richness and abundance of biodiversity, encompassing a wide array of species and ecological functions. The Ducke Forest Reserve has effectively fulfilled this role for aquatic macroinvertebrates. On average, the reserve maintained more than twice the abundance and nearly three times the species richness observed in the urban zone. Moreover, 65% of the identified taxa were exclusive to the reserve, including several sensitive groups known to indicate good environmental quality. These results underscore not only the ecological value of

protected areas in preserving biodiversity under intense anthropogenic pressure but also the magnitude of their effect, thus justifying the continued creation, maintenance, and connectivity of such areas as a key conservation policy in the Amazon region.

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