Impacts of roads on the body condition of Amazonian stream fish

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The Neotropical ichthyofauna has been degraded by multiple anthropogenic alterations. Despite advances in understanding their negative effects on species assemblages, little is known about how such changes affect fish' nutritional status. We tested the effects of road density, road–stream crossings density, and riparian deforestation on the condition factor of Amazonian fish species. We expected a decline in the condition factor for an allochthonous insectivore, an increase for a detritivore, and no effect on omnivorous species, across 32 headwater streams in the Amazon agricultural frontier. Our predictions were partially supported: The insectivorous species was possibly negatively affected by dietary impoverishment caused by increased siltation and the formation of deeper, slower habitats associated with road expansion, and none of the variables significantly influenced the omnivorous species. Contrary to our expectations, landscape degradation and stream fragmentation did not lead to an increase in the condition factor of the detritivorous species. These results may be explained by the differential

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responses of species, mediated by their traits, to environmental changes. Long-term monitoring will be essential to enhance the predictive power of this tool in detecting responses to anthropogenic changes in aquatic ecosystems.

Keywords: Impoundments, Landscape degradation, Length-weight relationship, Nutritional status, Riverscape fragmentation.

A ictiofauna neotropical tem sido impactada por diversas alterações antrópicas, mas pouco se sabe sobre como essas mudanças afetam a condição nutricional das espécies. Neste estudo, avaliamos os efeitos da densidade de estradas, a densidade de cruzamentos e do desmatamento ripário sobre o fator de condição de peixes amazônicos, em 32 igarapés de cabeceira na fronteira agrícola da Amazônia. Esperávamos uma redução no fator de condição para uma espécie insetívora alóctone, um aumento para uma detritívora e nenhum efeito para uma onívora. As previsões foram parcialmente confirmadas: a densidade de estradas teve efeito negativo na espécie insetívora, e nenhuma variável influenciou significativamente a espécie onívora. Contrariando expectativas, a degradação da paisagem e a fragmentação dos cursos d'água não resultaram em aumento no fator de condição da espécie detritívora. Esses resultados são possivelmente explicados pelas respostas diferenciais das espécies, mediadas por seus atributos, às mudanças ambientais. O monitoramento de longo prazo será crucial para aprimorar a capacidade preditiva dessa abordagem frente às mudanças antrópicas em ecossistemas aquáticos.

Palavras-chave: Barramentos, Degradação da paisagem, Fragmentação fluvial, Relação peso-comprimento, Status nutricional.

INTRODUCTION

Anthropogenic disturbances are leading to severe levels of biodiversity loss in freshwater ecosystems worldwide (Reid *et al*., 2019; Dudgeon, Strayer, 2025; Sayer *et al*., 2025). Land-use and land-cover changes, dam construction, pollution and invasive species have been long recognized among the main stressors (Dudgeon *et al*., 2006; Reid *et al*., 2019), while new drivers are emerging to make this list (*e.g*., microplastic contamination, changing climates, fluvial fragmentation by roads). In the Neotropical region, and particularly in the Amazon Basin, the global epicenter of fish diversity (Toussaint *et al*., 2016; Albert *et al*., 2020), human-induced alterations are happening at a fast pace and leading to widespread consequences (Castello *et al*., 2013; Latrubesse *et al*., 2017; Anderson *et al*., 2018; Schiesari *et al*., 2020; Couto *et al*., 2024). However, our understanding of the impacts to small streams and the associated biota remains overlooked compared to temperate freshwater ecosystems.

Streams are strongly dependent on the channel-riparian linkages and consequently highly sensitive to the alterations in their surrounding landscapes (Pusey, Arthington, 2003; Allan, 2004). The removal of riparian forest disrupts natural shading, increasing solar incidence and water temperature, and alters streambed structure and stability by the reduced input of wood and increased runoff of fine sediments (Leal *et al*., 2016; Leitão *et al*., 2018; Montag *et al*., 2019; Cantanhêde, Montag, 2024). Changes in the riparian forest also favor the carrying of contaminants by agricultural activities into the streams (Macedo *et al*., 2013; Ilha *et al*., 2018; Mello *et al*., 2020). Finally, riparian deforestation reduces the amount of allochthonous food resources (*e.g*., terrestrial invertebrates, fruits) to the aquatic fauna. Therefore, both the physical habitat and the energy base are transformed, with the degraded system characterized by an homogenized habitat and a shift from a predominant heterotrophic to an autotrophic primary productivity (Casatti *et al*., 2012; Zeni, Casatti, 2014).

Although the role played by the riparian forest in the integrity of streams is undeniable, other drivers related to landscape alterations are relevant but often overlooked. This is the case of the expansion of roads, a growing environmental concern to freshwater ecosystems that often receive little attention (Leal *et al*., 2016; Zarri *et al*., 2022). Roads density within catchments affect hydrological connectivity, increase soil impermeability, and intensify surface runoff and can, even if indirectly, act locally through sediment and toxic components deposition into aquatic ecosystems, thereby modifying streambed structure and physical habitat conditions (Forman, Alexander, 1998; Trombulak, Frissell, 2000; Lane, Sheridan, 2002; Ramos-Scharrón, MacDonald, 2007; Roy, Sahu, 2018). Moreover, low-permeability infrastructure associated to road-stream crossings (*e.g*., undersized culverts) often create small upstream impoundments (Fuller *et al*., 2015) that disrupts sediment, organic matter, and nutrient flow (Januchowski-Hartley *et al*., 2013; Pringle, 2003; Elosegi, Sabater, 2013), limit fish migration and dispersal (Warren, Pardew, 1998; De Fries *et al*., 2023), and alter the diversity and functional structure of aquatic communities (Helms *et al*., 2011; Perkin, Gido, 2012; Leitão *et al*., 2018; Brejão *et al*., 2020).

These human-induced changes ultimately affect fish assemblages, mediated by the species traits related to habitat use and foraging (Brejão *et al*., 2018; Leitão *et al*., 2018). Flow-regulated streams lacking riparian forests and having homogeneous streambeds may experience a reduction in the availability of terrestrial food sources, thereby negatively affecting species that feed small terrestrial invertebrates (Zeni, Casatti, 2014; Lobón-Cerviá *et al*., 2016; Montag *et al*., 2019). On the other hand, these conditions may favor algivorous and detritivores, due to increased primary production and greater availability and quality of organic detritus (Bojsen, Barriga, 2002; Zeni, Casatti, 2014; Lobón-Cerviá *et al*., 2016). Finally, omnivorous species tend to exhibit high dietary plasticity, adjusting their diet composition according to environmental conditions (Ferreira *et al*., 2012; De Carvalho *et al*., 2019).

Despite recent advances in our understanding of the effects of roads and land use change on fish assemblages at multiple facets of diversity (*e.g*., taxonomic, functional), a major knowledge gap remains regarding their effects on fish at the population scale. Some indicators related to individual nutritional status can be used to predict early biological responses to anthropogenic stressors (Hook *et al*., 2014; López-López, Sedeño-Díaz, 2015; Dalzochio *et al*., 2016). For instance, the condition factor stands out as an intuitive and easily measurable metric, calculated based on the weight-length relationship of individuals (Le Cren, 1951; Froese, 2006; Camara *et al*., 2011). It reflects the assimilation and storage capacity of energy reserves in somatic tissues, often associated with the availability and quality of food resources in the environment (Vila-Gispert, Moreno-Amich, 2001; Pereira et al., 2016; Cavraro et al., 2019). Low condition factor in a population may indicate elevated stress affecting individual nutritional status and, ultimately, decreased fitness and long-term species viability (Jakob *et al*., 1996). The assessment of condition factor represents a promising cost-efficient biomonitoring tool for fish populations, which may exhibit differential responses to environmental stressors based on their life-history attributes (Bojsen, 2005; Abujanra *et al*., 2009; Luz-Agostinho e*t al*., 2009).

Using fish populations from streams located in the Amazonian agriculture frontier, we assess the effects of landscape alterations on the condition factor of species belonging to three different trophic guilds: allochthonous insectivore, detritivore, and omnivore. Our general hypothesis is that road density, road crossings and riparian deforestation will affect, directly or indirectly, stream fish condition according to each trophic guild. Following these human-induced stressors, we expect a decrease in the condition factor for the allochthonous insectivorous species as a result of the limitation of terrestrial food sources and of the general changes in the stream channel physical habitats promoted by forest removal and roads. On the other hand, due to the greater availability of detritus and enhanced primary production instream, we predict an increase in the condition factor of the detritivorous species. Finally, the omnivorous species would not express differences in the condition factor, as they exhibit high dietary plasticity. Considering that road expansion and deforestation is a overlooked reality in the Amazon (Pocewicz, Garcia, 2016; Botelho *et al*., 2022), understanding the response of different fish species to these disturbances becomes essential for predicting future impacts on the most species-rich freshwater biodiversity on Earth.

MATERIAL AND METHODS

Study area. This study was carried out in 32 small streams (first- to third-order Strahler), locally known as "*igarapés*", located southeast of the confluence between the Tapajós and Amazon rivers (Fig. 1). The streams belong to the Curuá-Una (26 streams), Tapajós (4), and Amazon River (2) basins and are located in the municipalities of Santarém, Belterra, and Mojuí dos Campos, Pará state, Brazil (henceforth called 'Santarém Region'). The study area comprises a heterogeneous mosaic of land use and land cover, including mechanized agriculture, pasturelands, urban settlements, second and degraded forest, and large expanses of continuous forest such as the Tapajós National Forest (FLONA Tapajós) conservation unit (Gardner *et al*., 2013; Almeida *et al*., 2016). Due to its proximity to two major federal highways, the Transamazônica (BR-230) and Cuiabá-Santarém (BR-163), this region is relevant for grain exports (Fearnside, 2007). The streams and their associated drainage basins are distributed along a broad gradient of environmental conditions, ranging from sites with very little anthropogenic influence (located in FLONA tributaries, Fig. 2A) to highly fragmented streams affected by secondary unpaved roads and surrounded by extensive pasturelands and monoculture areas (Figs. 2B–D). All streams belong to different catchments and are spatially independent. Stream waters are warm (25.98 ± 1.01 °C), moderately acidic (5.33 ± 0.46), well-oxygenated (6.65 ± 1.17mg.L^{-1}), and of low conductivity (13.41 ± 3.03). The physical habitat features vary widely but generally consists of a sandy substrate with a major presence of leaf packs, roots, and wood debris. These streams are shallow (49.2 cm,11.6–115 cm), narrow (5.01 m, 1.08–13.8), and with slow flowing glides and riffles. The native vegetation consists of dense ombrophilous forest, characterized by a tropical monsoon climate, with an average annual precipitation of 1,950 mm and a mean annual air temperature of 27.5 °C (Vasconcelos *et al.*, 2006). The sampling design follows the first sampling carried out by the Sustainable Amazon Network (*Rede Amazônia Sustentável* - RAS) (Gardner *et al.*, 2013).

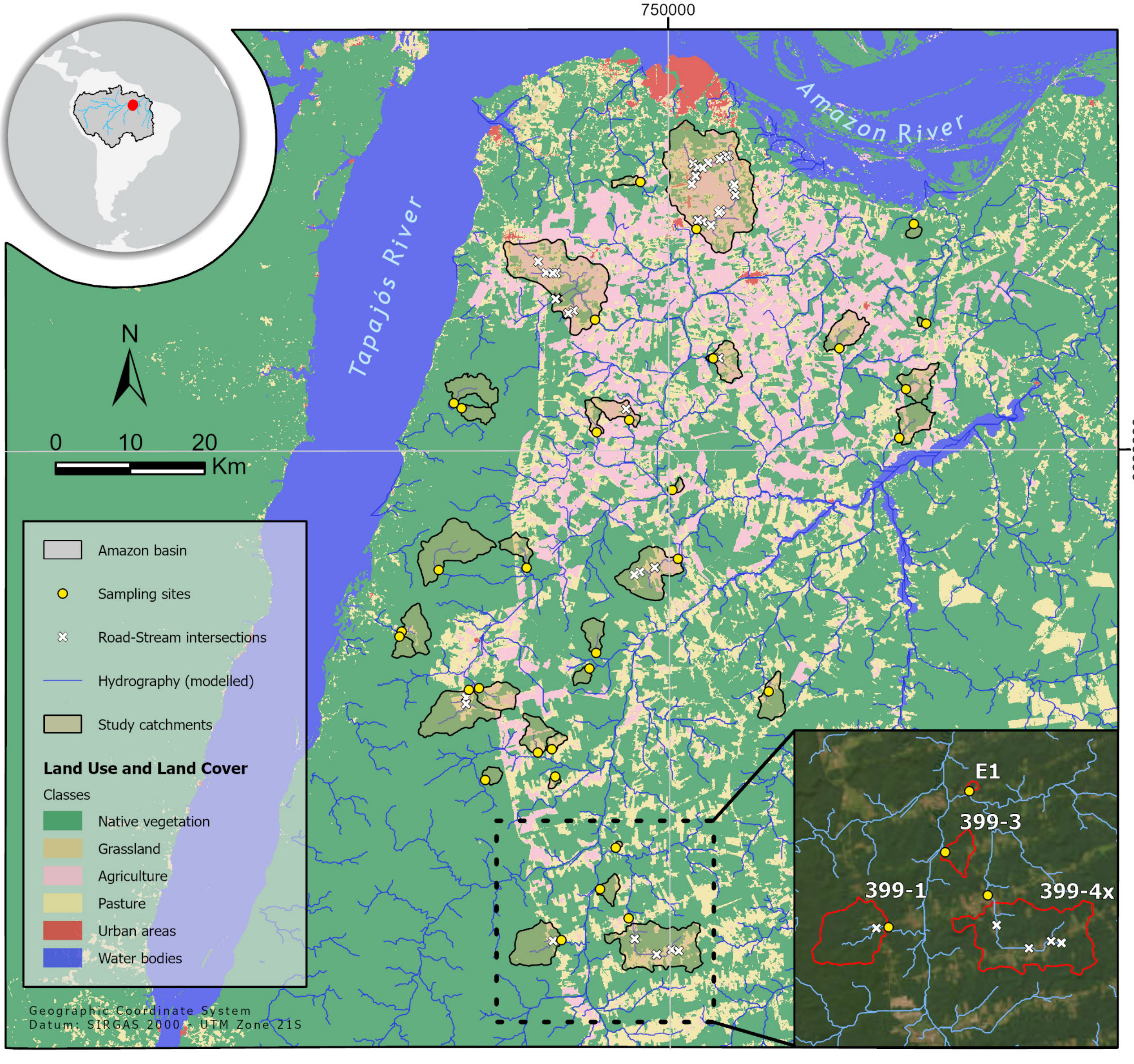

FIGURE 1 | Study area in the Santarém Region, Pará State, Brazil, Amazon basin. Sampled streams sites (yellow dots) are located at the outlet of each catchment, and belong to the Curuá-Una (26 streams), Tapajós (4), and Amazon River (2) catchments. Satellite imagery of some sampling sites (E1, 399–1, 399–3, and 399–4x) was zoomed in to highlight differences in the size, land cover and road-stream intersections within catchments.

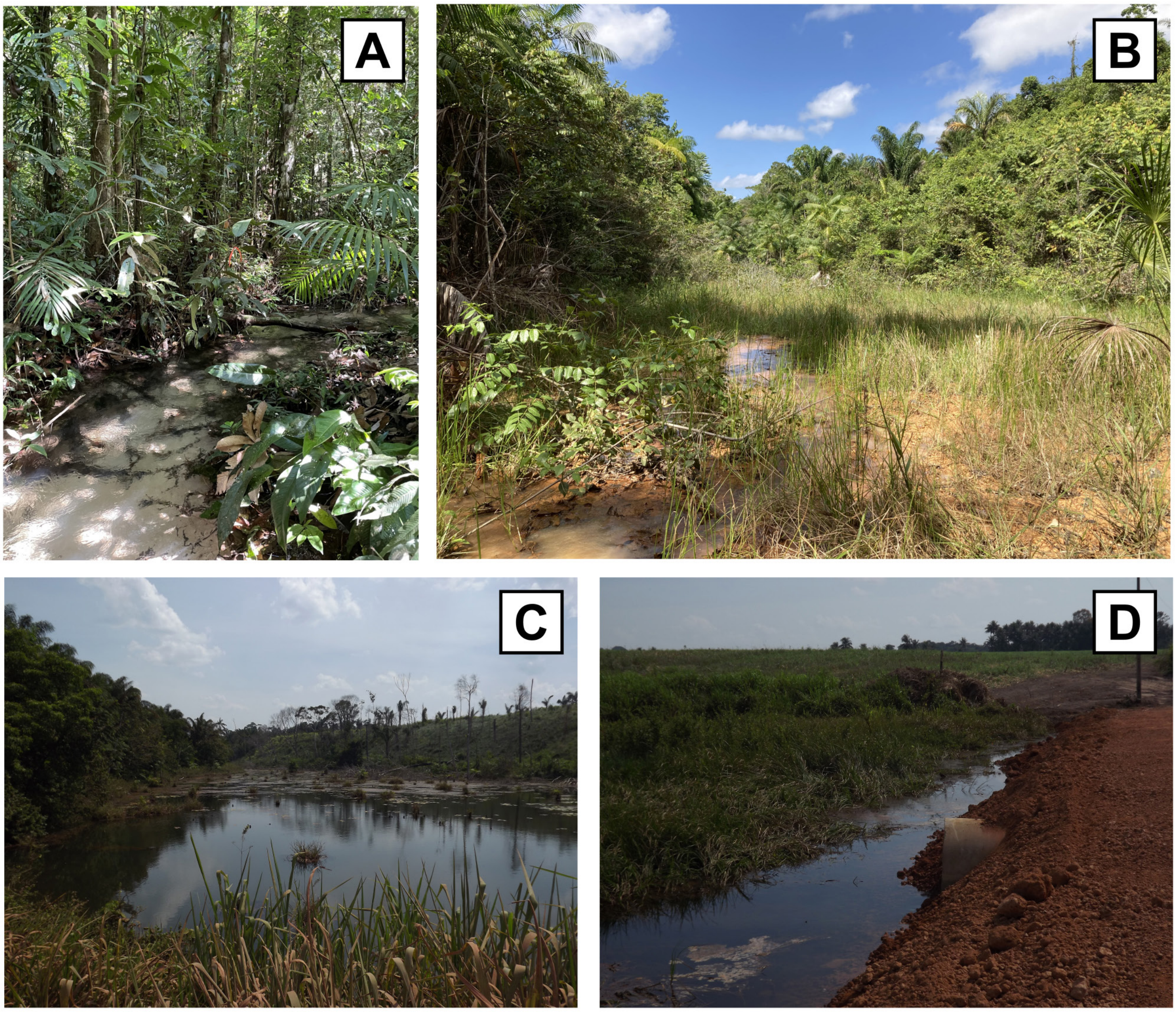

FIGURE 2 | Different environmental conditions observed in streams from the Santarém Region, Pará State, Brazil, Amazon basin: **A.** Well-preserved stream, without human impact at the local or catchment scale; **B.** Degraded stream, with an homogeneous substrate and a clearing caused by the removal of riparian forest; **C.** Formation of a small reservoir upstream of a watercourse interrupted by a road crossing; **D.** Downstream of a road crossing, where the stream is buried due to earthworks and the installation of low-permeability culverts. An extensive area cleared for cattle pasture is visible in the background of the image. Photographs were taken by Débora R. de Carvalho (**A**), Gabriel L. Brejão (**B**), and Gabriel O. Ferraz (**C**–**D**).

Land use. We used the plug-in SWAT tool (Soil and Water Assessment Tool) (Neitsch *et al*., 2011) based on the NASADEM 30 m resolution digital terrain model (NASA JPL, 2020) to model the hydrography considering a minimum contribution area of 500 hectares. Based on the modelled hydrography, catchments were delimited for each site considering the sampling point as the outlet. We calculated land use and land cover for a riparian network buffer (*i.e*., 100 m buffer in each margin along the entire drainage network upstream from stream site) of each stream site using 30 m resolution data from MapBiomas Collection 9 (MapBiomas, 2024).

We focused on three categories of land use most related to roads: pasture, agriculture (soybean plantations and temporary crops), and urban areas, this latter with very low representation across the studied catchments (see Tab. **S1**). We calculated a Riparian Disturbance Index (RDI) considering the sum of the weighted proportion of urban areas (weight 4), agriculture (weight 2) and pasture (weight 1) in the riparian network upstream from each stream site. The RDI is an adaptation from the Catchment Disturbance Index (CDI) (Ligeiro *et al*., 2013), including the weight for each land use categories, with the difference that RDI consider land use at the riparian network while CDI consider the entire catchment upstream from the focal site. The RDI ranges from 0 (absence of the three land use types) to 4 (riparian buffer fully covered by urban land use); the higher the total score, the greater the level of riparian network degradation upstream from each sample site. All landscape analyses were carried out using ArcGIS Pro software (version 3.3).

Road variables. We built a road database by manually mapping the study area using Planet satellite images (Planet Team, 2017), 4.77 m resolution, with the scenes between May and September 2023 to avoid the presence of clouds as much as possible. We calculated road density by dividing the total length of roads inside a catchment by the catchment area (km/km^2). Road-stream crossings were identified using the Intersect function with the modelled hydrography and road database. Given the large variation in catchment areas (Tab. **S1**), we calculated the stream crossing density by dividing the number of road-stream crossings by the catchment area (n/km^2). Road calculations were performed in the ArcGIS Pro Python Notebook environment (Environmental Systems Research Institute, 2024).

Fish sampling and trophic guilds. We sampled fish during daytime in the dry season (July-August of 2023) by two people over two hours in each 150 m stream site. All sampled stream stretches were entirely lotic systems and located at least 140 m far from any road crossing upstream (*i.e*., the sampled sites were not characterized by impoundment dammed by the road). We used semicircular sieves (80 cm in diameter and 1 mm mesh size) and seine nets (4 m in length, 2 m in height, and 5 mm mesh size) to reach different habitats used by fish for shelter and feeding (*e.g*., pools, leaf banks, wood debris, and macrophytes). Fish were anesthetized using a lethal dose of Eugenol and fixed in 10% Formalin. In the laboratory, the specimens were identified using identification keys and with the assistance of taxonomists.

Although we sampled the entire assemblage in each stream site, we selected three species that were abundant and widely distributed in the study region and that represent distinct trophic guilds: *Aequidens epae* (Cichliformes: Cichlidae), a small- to medium-sized omnivore; *Apistogramma taeniata* (Cichliformes: Cichlidae), a small-sized detritivore; and *Hyphessobrycon ericae* (Characiformes: Acestrorhamphidae), a small-sized allochthonous insectivore. The classification of each species into trophic guilds was based on stomach content analyses of individuals previously sampled in the same streams, representing different conditions across the gradient of landscape degradation (Leal *et al*., 2018). We measured standard length (SL, at nearest 0.01 mm) and weight (W, at nearest 0.01 g) using a digital caliper and a precision scale, respectively. Fish were preserved in 70% alcohol and will be deposited in the Coleção de Peixes do

Departamento de Ciências Biológicas (DZSJRP) at the Universidade Estadual Paulista Júlio de Mesquita Filho, São José do Rio Preto, São Paulo State, Brazil.

Statistical analysis. Before conducting the weight-length relationship analyses, outliers were removed through graphical inspection (Froese, Binohlan, 2000). Then, we determined the coefficients of individual fattening (*a*, constant of proportionality) and growth (*b*, coefficient of allometry) for each species ($_i$) through a linear regression between both natural logarithms of standard length as the independent variable and total weight as the dependent variable (logSL ~ LogW). Relative body condition (K_n) was estimated for every individual through the ratio of observed weight and the predicted weight for a given standard length ($K_n = W/a.SL^b$). We chose not to use the Fulton Condition Factor, as it assumes a fixed isometric coefficient (b = 3) in the weight-length relationship, which was not observed for the species *Aequidens epae*, that exhibited negative allometric growth (b = 2.95).

Before assessing the collinearity of the predictors (road density, road-stream crossings density, and RDI), we standardized their values using the z-score (mean = 0, standard deviation = 1). We then applied The Pearson's Correlation among predictors and none exceeded r > 0.7. We performed Generalized Linear Mixed Models (GLMMs) with a Gaussian distribution to test the relationships between the individual condition factor and the predictors for each species, with the sampling site as a random factor. We then built eight regression models: three testing the relationship of each predictor individually, two with the interactions of the road predictors with the RDI, one additive model between the road predictors, one with full interactions, and a null model (Tab. 1). We performed an Akaike's Information Criterion adjusted for small sample size (AICc) to assess the strength of candidate models, using the "aictab" function from "AICcmodavg" package (Mazerolle, 2020). Models with ΔAICc value less than 2 compared to the top-ranked model were considered to have substantial support and potentially explain the variance of individual condition factor across degrees of disturbance. We checked for normality of residuals and model assumptions by drawing histograms of models' residuals and plotting them against each predictor. All statistical analysis was performed using the R software (R Development Core Team, 2023).

TABLE 1 | Simplified R arguments of the Generalized Linear Mixed Models built to test the influence of road density (RoadDensity), road-stream crossing density (CrossDensity), Riparian Disturbance Index (RDI), and their random factor [(1|SITE)], on the individual condition factor (CF) for each fish species ($_i$).

Models	Argument
Model 1 (simple)	CF_i ~ RoadDensity + (1 | SITE)
Model 2 (simple)	CF_i ~ CrossDensity + (1 | SITE)
Model 3 (simple)	CF_i ~ RDI + (1 | SITE)
Model 4 (multiple)	CF_i ~ RoadDensity + CrossDensity + (1 | SITE)
Model 5 (multiple)	CF_i ~ RoadDensity:RDI + (1 | SITE)
Model 6 (multiple)	CF_i ~ CrossDensity:RDI + (1 | SITE)
Model 7 (multiple)	CF_i ~ RoadDensity:RDI + CrossDensity:RDI + (1 | SITE)
Model 8 (null)	CF_i ~ (1 | SITE)

RESULTS

Measurements of standard length (SL) and total weight (W) were obtained for a total of 1,751 fish specimens: 243 *Aequidens epae*, 365 *Apistogramma taeniata*, and 1,143 *Hyphessobrycon ericae*. The individual SL and W respectively ranged from 7.42–81.37 mm (27.67 ± 13.83) and 24.248 g (1.83 ± 3.24) in *A. epae*; 9.2–49.74 mm (20.92 ± 6.45) and 0.01–4.52 g (0.45 ± 0.46) in *A. taeniata*; and 9.07–33.66 mm (20.53 ± 4.52) and 0.012–0.846 g (0.21 ± 0.14) in *H. ericae*. *A. taeniata* and *H. ericae* exhibited an isometric growth pattern, with the 95% confidence limits of the allometric coefficient (*b*) ranging from 2.90 to 3.04 and 2.95 to 3.02, respectively. In contrast, *A. epae* displayed a negative allometric growth pattern, ranging from 2.93 to 2.99.

Catchments exhibited considerable variation in landscape use, as represented by the Riparian Zone Disturbance Index (RDI) (0.39 ± 0.37; ranging from 0 to 1.25), road density (0.60 ± 0.48; 0 to 1.88 km/km²) and road–stream crossings density (0.02 ± 0.03; 0 to 0.07) (Tab. **S1**). Road crossings were the least variable among the other predictors, with only 31% of the catchments presenting at least one road–stream intersection (Fig. 3). Riparian land use ranged from zero to 0.03 (0.001 ± 0.005) for urban, from zero to 0.50 (0.09 ± 0.13) for agriculture, and from zero to 0.79 (0.21 ± 0.20) for pasture.

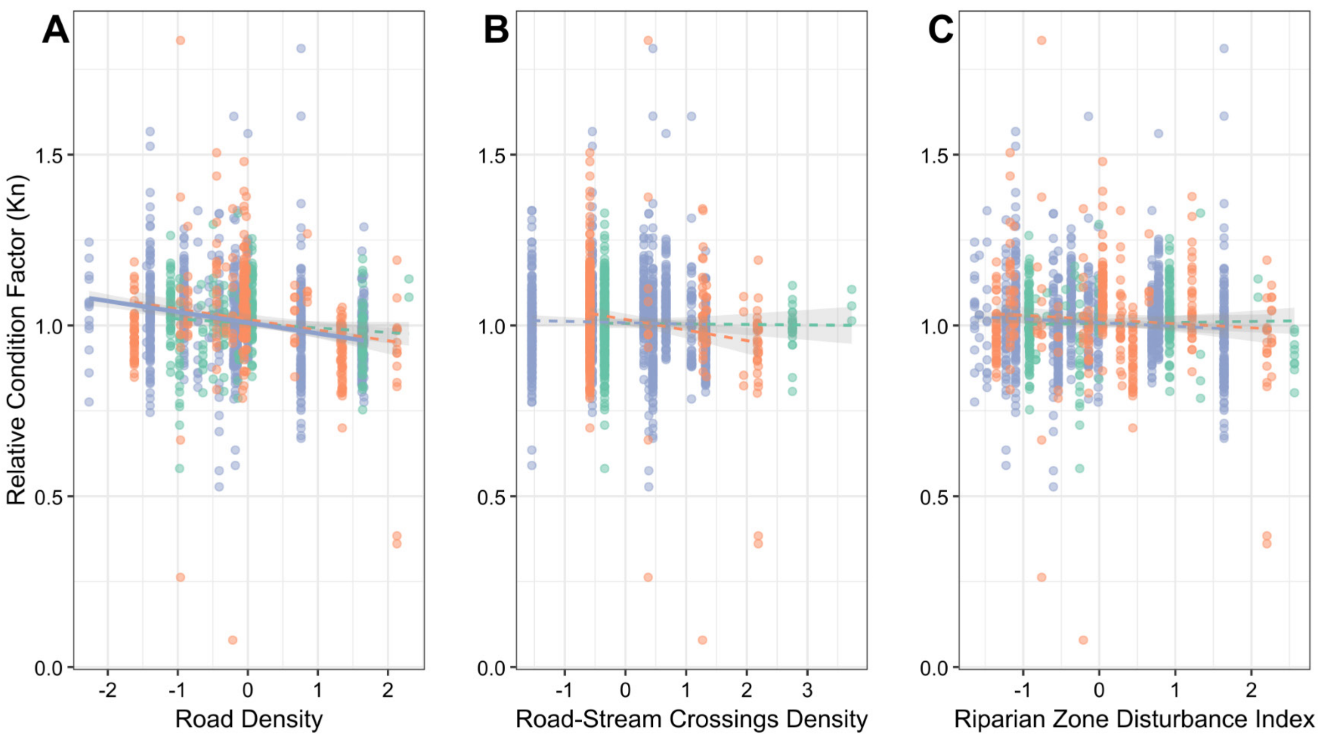

FIGURE 3 | Relationship of Road Density (**A**), Road-Stream Crossings Density (**B**) and Riparian Zone Disturbance Index (RDI) (**C**) on the individual body condition factor (Kn) of Amazonian stream fish species of different trophic guilds: *Aequidens epae* – omnivorous (green dots); *Apistogramma taeniata* – detritivorous (red dots); and *Hyphessobrycon ericae* – allochthonous insectivore (blue dots). Significant relationships are indicated by a solid line, and non-significant relationships by a dashed line.

The individual condition factor showed variable ranges among the three species: 0.58 to 1.33 (1.00 ± 0.11) in *A. epae*; 0.08 to 1.83 (1.02 ± 0.16) in *A. taeniata*; and 0.53 to 1.81 (1.01 ± 0.13) in *H. ericae* (Fig. 3). According to the Akaike Information Criterion for small samples (AICc) ranking, the null model was the most important for all species (Akaike weights ≥ 0.69), and the relationships between predictors and species condition factor were overall weak (Tab. 2). However, the effect of road density on the condition factor in *H. ericae* (allochthonous insectivore) was also supported (Model 1, ΔAICc = 1.81), showing a significantly negative relationship (x^2 = 8.63, df = 1, p = 0.003), with an average decrease of 0.035 in the condition factor per unit of road density (km/km^2) (Tab. 2; Fig. 3A). The road-stream crossing density and the RDI did not exhibit a significant effect on the condition factor of any of the species (Figs. 3B–C).

TABLE 2 | Akaike Information Criterion (AIC), conditional r^2 and slope results for the regression models testing the influence of predictors (road density, road-stream crossing density, and RDI) on the individual condition factor of the species *Aequidens epae*, *Apistogramma taeniata*, and *Hyphessobrycon ericae*. Asterisks indicate the most parsimonious regression models. Variables included in each model are indicated in Tab. 1.

Species	Model	AICc	ΔAICc	AICcWt	K	Likelihood	Conditional r²	Slope
Aequidens epae	Model 8 *	-408.39	0	0.85	3	207.25	0.281	-
	Model 5	-403.81	4.58	0.09	4	205.99	0.282	0.029
	Model 1	-400.53	7.86	0.02	4	204.35	0.326	0.009
	Model 6	-400.48	7.91	0.02	4	204.33	0.321	-0.005
	Model 3	-400.47	7.93	0.02	4	204.32	0.313	-0.012
	Model 2	-399.86	8.53	0.01	4	204.02	0.318	0.002
	Model 7	-395.58	12.82	0	5	202.92	0.256	0.028, -0.002
	Model 4	-392.05	16.34	0	5	201.16	0.345	0.009, 0.002
Apistogramma taeniata	Model 8 *	-309.55	0	0.77	3	157.81	0.178	-
	Model 2	-304.82	4.73	0.07	4	156.47	0.185	-0.032
	Model 1	-304.72	4.83	0.07	4	156.43	0.188	-0.035
	Model 5	-303.82	5.73	0.04	4	155.97	0.181	-0.026
	Model 6	-302.93	6.61	0.03	4	155.53	0.183	-0.020
	Model 3	-302.18	7.37	0.02	4	155.15	0.187	-0.017
	Model 4	-297.97	11.58	0	5	154.08	0.194	-0.022, -0.022
	Model 7	-296.31	13.24	0	5	153.25	0.192	-0.024, -0.002
Hyphessobrycon ericae	Model 8 *	-1039.2	0	0.69	3	522.62	0.068	-
	Model 1 *	-1037.39	1.81	0.28	4	522.72	0.077	-0.026
	Model 5	-1030.86	8.34	0.01	4	519.45	0.069	0.010
	Model 3	-1030.54	8.66	0.01	4	519.3	0.070	-0.007
	Model 6	-1030.08	9.13	0.01	4	519.06	0.069	0.005
	Model 2	-1029.82	9.38	0.01	4	518.93	0.068	-0.002
	Model 4	-1028.53	10.67	0	5	519.3	0.071	-0.029, 0.008
	Model 7	-1022.25	16.95	0	5	516.16	0.070	0.013, -0.004

DISCUSSION

We examined how the nutritional status of three stream fish species, belonging to distinct trophic guilds, is influenced by roads and riparian degradation in the Amazon basin. Our hypothesis that the body condition would decline with increasing disturbance for the allochthonous insectivore (*Hyphessobrycon ericae*), increase for the detritivorous (*Apistogramma taeniata*), and remain unaffected for the omnivorous (*Aequidens epae*) species, was partially supported. Increased road density in the catchment negatively affected the condition factor of *H. ericae*, whereas the number of road crossings and land use changes in the riparian network showed no effects on this fish species. Furthermore, as expected, none of the three predictors affected the condition factor of the omnivore *A. epae*. However, contrary to our expectations, none of the predictors have positively influenced the condition factor of *A. taeniata.*

The decline in the condition factor of *H. ericae* suggests that this species appears to be sensitive to environmental disturbances associated with increased road density in catchment. The impacts related to roads can be indirectly associated to the loss of riparian forest, which reduces the vertical and longitudinal transport of allochthonous resources downstream (Casatti *et al*., 2012; Zeni, Casatti, 2014; Lobón-Cerviá *et al*., 2016). Although the riparian degradation index (RDI) did not appear to directly influence the fish body condition, land conversion to agricultural systems can drive dietary shifts (Bojsen, 2005; Ferreira *et al*., 2012), leading to lower selectivity for preferred food resources (De Carvalho *et al*., 2017). Studies have already reported changes in diet composition and a subsequent decline in the condition factor in some characid species because of anthropogenic disturbances associated with deforestation and road-induced impoundments (Bojsen, 2005; Barros *et al*., 2024; de Souza *et al*., 2025).

More than potentially reduce the amount of allochthonous resources to streams, roads contribute to greater surface impermeability, leading to enhanced runoff and erosion rates, which, in turn, elevate sediment deposition and cause structural changes in streambeds (Forman, Alexander, 1998; Trombulak, Frissell, 2000). The presence of roads may lead to an elevated concentration of suspended inorganic sediment in the streams (Couceiro *et al*., 2020), which not only poses a risk of mechanical damage to fish gills, but also reduces foraging efficiency by requiring increased energetic costs for food detection and acquisition (Kemp *et al*., 2011). Species of *Hyphessobrycon* exhibits a good visual acuity, swimming capacity and are commonly associated to pool mesohabitats (Rezende *et al*., 2010; Brejão *et al*., 2013; Manna *et al*., 2014), and such alterations may be inducing higher stress levels in the individuals. Deforestation driven and associated road expansion may be decreasing the input of large wood into streambeds, thereby reducing the amount and the volume of residual pools (*i.e*., deeper and slow water current mesohabitats that, in Amazonian streams, are primarily formed by the obstruction of the stream flow due to large wood debris), the average depth of the streams and, consequently, the availability of suitable habitat for the species. *Hyphessobrycon ericae* seemed thus to have responded to the complex set of processes involved in cumulative landscape disturbances. The decline in nutritional conditions in response to road density is likely associated with reduced availability of essential habitats, as well as the impoverishment of the diet caused by the increase in inorganic suspended sediment, and not only by limitations in the species' dietary food availability.

This complex scenario illustrates the need to future studies also consider other niche dimensions rather than single diet-related ones. For instance, species functional traits related to habitat use and life history can be extremely elucidative to understand the multiple landscape-change effects on the fish condition.

The lack of significant effects of landscape alterations on *A. epae* and *A. taeniata* may be associated with their greater tolerance to adverse environmental conditions (Burress, 2015; Ferreira *et al*., 2018), while maintaining stable the body condition levels (Ilha *et al*., 2018). Most cichlids are sedentary species with parental care and often exhibit considerable trophic plasticity, consuming a wide range of resources despite seasonal and environmental variations (Costa, Soares, 2015). Interestingly, given that these two species were predominantly found in streams without road crossings (see Fig. 3B), we suggest that road crossings may influence species occurrence and abundance rather than their condition factor. Infrastructure associate with road stream crossings are particularly precarious in the Amazon, often consisting of undersized and perched culverts that create small impoundments upstream of the road (Leal *et al*., 2016), and prone to disrupt hydrological connectivity and habitat structure, acting as environmental filters that shape local fish assemblages and species distributions (Nislow *et al*., 2011; Leitão *et al*., 2018; Brejão *et al*., 2020). While condition factor is one of the indicators of fish nutritional status, additional traits including those associated with life-history could also be useful for assessing how cichlids respond to environmental conditions in the context of river fragmentation (Mendes *et al*., 2021).

The effects of anthropogenic disturbances on fish body condition in streams remain largely understudied. In larger riverine systems, condition factor can vary at the trophic level due to seasonal hydrological fluctuations and dam presence (Abujanra *et al*., 2009; Luz-Agostinho *et al*., 2009; Pereira *et al*., 2016; Tribuzy-Neto *et al*., 2017). However, the mechanisms by which these disturbances influence energy allocation in fish are still not well understood. A decline in resource availability can both alter species trophic niche breadth and position (De Carvalho *et al*., 2017; Urbano *et al*., 2024) and drive an increase in intraspecific trophic specialization (Barros *et al*., 2024). The negative effect of road density on the condition factor of *H. ericae* may indicate lower tolerance to environmental degradation and suggest early warning responses to population declines (Camara *et al*., 2011). Although this finding was limited here to one species, we believe similar effects would be found among other taxa with similar ecological requirements and tolerances. As an indirect indicator of energy reserves in body tissues, poor body condition is generally linked to lower fecundity and delayed sexual maturation in populations (Mion *et al*., 2018; Rodgveller, 2019), potentially reducing the recruitment of new individuals. Integrating approaches of population dynamics with temporal patterns in nutritional condition can enhance our understanding of the lagged responses of fish assemblages to anthropogenic disturbances.

Fish may exhibit delayed responses to anthropogenic changes (Brejão *et al*., 2018; Dala-Corte *et al*., 2020; Camana *et al*., 2022), and considering the relatively recent history of environmental degradation in the Amazon, stream communities may currently be experiencing an intermediate stage of disturbance. In this context, the implementation of conservation actions will be essential to mitigate the ongoing impacts on ichthyofauna and ecosystem processes in Amazonian streams, particularly concerning road expansion (Couto *et al*., 2024). Improving land management within hydrological basins can not only increase the availability of allochthonous resource but also help to reduce erosion and the input of fine sediment into streambeds, thereby enhancing the quality of water and physical habitat conditions (Castello, Macedo, 2016; Leal e*t al*., 2018; Mello *et al*., 2020). These improvements would ultimately support the health of species directly dependent on such conditions, such as *Hyphessobrycon ericae* and ecologically similar species. We recognize that assessments based on only few species limit broader inferences to understand fish health status at the level the trophic guild or the community. In this context, future studies should scale up condition factor assessments to entire trophic and functional groups. This approach will allow the detection of more consistent patterns in the relationship between landscape alterations, particularly those associated with roads, and the nutritional condition of stream fish assemblages in the Amazon.

Our findings suggest that increasing road density can negatively impact the body condition of an allochthonous insectivorous species, while no significant effects of anthropogenic disturbance were observed on the body condition of an omnivorous or a detritivorous species. These results may be partially associated with fish strategies related to food acquisition, but may also be influenced by other ecological aspects of each species. Long-term studies taking into account multiple dimensions of species traits are essential to understanding the cumulative impacts of anthropogenic disturbances on stream fish communities. We also emphasize the need to integrate body condition assessments with additional methodological approaches, such as stomach content and stable isotope analyses, to better understand the effects of road crossings and other landscape alterations on the main energy sources and on the mechanisms of fish energy allocation. These integrative approaches will be crucial in determining species response thresholds and forecasting future trends in community dynamics in the face of increasing human-induced degradation of freshwater ecosystems.

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Dennys Heilbuth Cachapuz Drager: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Writing-original draft, Writing-review and editing.
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Marcos Angelo Alves Filho: Resources, Writing-review and editing.
Rafael Pereira Leitão: Conceptualization, Data curation, Funding acquisition, Investigation, Methodology, Project Administration, Supervision, Validation, Writing-review and editing.

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ETHICAL STATEMENT

This research was approved by the Ethics Committee for Animal Use of the Federal University of Pará (CEUA number 8293020418/2021). Fish sampling was authorized under SISBIO license number 87389–1/2023.

DATA AVAILABILITY STATEMENT

The data supporting the findings of this study are available from the project coordinator, Cecília G. Leal, upon reasonable request.

AI STATEMENT

The authors used the Gemini AI for grammatical revision.

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COMPETING INTERESTS

The authors declare no competing interests.

SUPPLEMENTARY MATERIAL

Supplementary material **S1**

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