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CAMPUS DE SÃO JOSÉ DO RIO PRETO

# Responses of stream fish assemblages to timing and extent of deforestation in Western Amazon

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DOUTORADO

PÓS GRADUAÇÃO  
EM BIOLOGIA ANIMAL



Biologia  
Estrutural



**UNIVERSIDADE ESTADUAL PAULISTA**  
**“JÚLIO DE MESQUITA FILHO”**  
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Tese apresentada como parte dos requisitos para obtenção do título de Doutor em Biologia Animal, junto ao Programa de Pós-Graduação em Biologia Animal, do Instituto de Biociências, Letras e Ciências Exatas da Universidade Estadual Paulista “Júlio de Mesquita Filho”, Campus de São José do Rio Preto.

Financiadora: FAPESP – Proc. 2012/21916-0  
FAPESP – Proc. 2015/05827-6

Orientador: Prof<sup>a</sup>. Dr<sup>a</sup>. Lilian Casatti  
Co-orientador: Prof. Dr. David J. Hoeinghaus  
Co-orientador: Prof. Dr. Silvio F. B. Ferraz

São José do Rio Preto  
2018

Brejão, Gabriel Lourenço.

Responses of stream fish assemblages to timing and extent of deforestation in Western Amazon / Gabriel Lourenço Brejão. -- São José do Rio Preto, 2018

140 f. : il., tabs.

Orientador: Lilian Casatti

Coorientador: David J. Hoeinghaus

Coorientador: Silvio F. B. Ferraz

Tese (doutorado) – Universidade Estadual Paulista "Júlio de Mesquita Filho", Instituto de Biociências, Letras e Ciências Exatas

1. Ecologia animal. 2. Ictiologia. 3. Ictiofauna - Rondônia. 4. Desmatamento. I. Universidade Estadual Paulista "Júlio de Mesquita Filho". Instituto de Biociências, Letras e Ciências Exatas. II. Título.

CDU – 597

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Financiadora: FAPESP – Proc. 2012/21916-0  
FAPESP – Proc. 2015/05827-6

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São José do Rio Preto  
04 de Maio de 2018

À minha família, que sempre apoiou  
as minhas escolhas e me acolheu nos  
momentos duros desta caminhada

*The cave you fear to enter holds the treasure you seek*

- Joseph Campbell

## **Agradecimentos**

Gostaria de agradecer os meus pais. Minha mãe, Maria de Lourdes que como professora, e com sua breve passagem pela vida, apresentou o encantamento pelas ciências. Meu pai, Walmir, que por fazer questão de manter a natureza dentro de casa, com suas plantas e aquários, apresentou um certo fascínio por entender como a vida das “coisas” funcionava. A Avani, que aceitou o desafio de acompanhar meu pai em um momento muito difícil para ser parte fundamental dessa jornada. Agradeço também aos meus irmãos, Pedro Ivo e Guilherme, pelos raros encontros dos últimos anos regados a conversas “filosóficas”.

Agradeço à Bruna, esposa e amiga, que compreende os desafios dessa carreira e sempre esteve ao meu lado, incentivando/criticando nos momentos de desânimo, ouvindo intermináveis conversas sobre as relações entre peixes e o desmatamento, ou simplesmente estando presente.

À Lilian Casatti, que há oito anos me aceitou como membro do seu grupo de pesquisa após um e-mail enviado em dezembro de 2010 (e respondido em 3 minutos...), e que me deu a oportunidade de continuar a trabalhar em uma região que me encantou no Mestrado, e uma segunda oportunidade para seguir no doutorado, mesmo após um deslize na primeira seleção para o doutorado. Uma orientadora não só para as questões acadêmicas e científicas, mas que se preocupa demais em formar pessoas melhores. Que permite aos seus alunos a liberdade de ousar, mas deixa uma cordinha sutilmente amarrada no pé pra puxar de volta pra terra e manter o foco no que foi proposto.

Aos meus co-orientadores, Silvio Ferraz, que sempre me ajudou a descascar os abacaxis que a ecologia da paisagem apresentava e a abrir os olhos para uma visão mais regionalizada dos impactos decorrentes do desmatamento. David Hoeinghaus, que me mostrou uma outra visão da ciência (acadêmica e filosoficamente), a ecologia dos testes de hipóteses que, mesmo se tratando de peixinhos de riachos, deve sempre ter potencial para dialogar com outras áreas do conhecimento. Sou grato pela dedicação dos dois na construção desse trabalho, e pelas amizades e parcerias que se estabeleceram.

À Dana Infante, Jansen Zuanon, Danilo Boscolo and Rafael Leitão, pela disponibilidade e pela leitura crítica desta Tese.

À família Hoeinghaus – David, Ana e Brenda – que me receberam tão calorosamente e proporcionaram, no Texas, o sentimento de estar em casa.

À Angélica, que veio da Colômbia para desbravar os riachos rondonienses e construiu grande parte da base de dados que foi utilizada nesse trabalho. E ao Felipe (B-nito) pela ajuda com o desenho amostral e no trabalho de campo. Obrigado por confiar nos mapas e no GPS, mesmo quando não fazíamos ideia de onde estávamos...

Aos todos membros do Laboratório de Ictiologia, especialmente Mônica, Angélica, Camilo, Jaque, Carol, Molina, Fernandinho, Ângelo e Bruno pelos momentos compartilhados ao longo desses anos, muito aprendizado e muitas risadas com todos vocês!

À Bárbara Callegari (MCP/PUCRS), Ilana Fichberg (MZUSP), Fernanda Martins (IFPR), Flávio Lima (UNICAMP), Francisco Langeani (UNESP), Leandro Sousa (UFPA), Manoela Marinho (MZUSP), Marcelo Britto (MNRJ), Marcelo

Carvalho (IB/USP), and William Ohara (MZUSP) pela ajuda na identificação dos peixes; aos membros dos laboratórios de ictiologia da UNESP e da UNIR fpela ajuda nos trabalhos de campo; ao ICMBio (REBio Jaru) Pelo apoio logístico; à SEDAM-RO e às associações de extrativistas pela permissão para coletar nas áreas das RESEX Rio Preto-Jacundá, RESEX Castanheira and RESEX Aquariquara;

Aos amigos da graduação, sempre presentes em especial àqueles que se tornaram minha segunda família nos anos de convívio na R.R.A. Biosfera.

Agradeço imensamente ao Pedro Gerhard, Anderson Ferreira e Katia Ferraz, orientadores e amigos, por serem parte fundamental da minha formação acadêmica. Pedro e Anderson que me apresentaram a ecologia de riachos e a biologia de peixes a distantes 16 anos, e Katia aos sistemas de informação geográfica, 13 anos atrás, no extinto Laboratório de Ecologia Animal Aplicada.

A todos do CEBB Rio Preto, que é um refúgio para compreender as aflições e impermanências do cotidiano, e nos ajuda a não dar tanto peso às experiências.

À University of North Texas por me receber e fornecer a estrutura necessária para o desenvolvimento do meu estágio no exterior durante seis meses.

Ao Programa de Pós-Graduação em Biologia Animal (IBILCE/UNESP) e ao Departamento de Zoologia e Botânica pelo apoio e;

À Fundação de Amparo à Pesquisa do Estado de São Paulo (FAPESP), pelas bolsas concedidas ao longo do meu doutorado (2012/21916-0 e 2015/05827-6) e pelo Auxílio à Pesquisa que permitiu a construção do banco de dados (2010/17494-8).

Para encerrar esse espaço de agradecimento, proponho uma reflexão sobre o cenário dos últimos sete anos, tempo em que estive envolvido com esse trabalho, no que diz respeito ao panorama científico e de políticas ambientais no Brasil atual. Entre outras coisas, o código florestal foi revisado e consolidado, reduzindo a proteção dos ambientes aquáticos e, concomitantemente, a ciência sofreu sucessivos cortes de recursos financeiros. Talvez esses acontecimentos sejam reflexo do distanciamento entre cientistas e sociedade, uma vez que poucos de nós tem procurado formas de divulgar os frutos das nossas pesquisas de maneira acessível a todos e, em consequência, perdemos o apoio da sociedade. Espero que essa questão seja considerada pelos acadêmicos, que pontes sejam construídas, para que não tenhamos trabalhado em vão.

## **Acknowledgments**

I had like to acknowledge my parents. My mother, Maria de Lourdes that being a teacher, and with her brief passage through life, presented me the enchantment by the sciences. My father, Walmir for keeping nature indoors, with his plants and aquariums, showed me a particular fascination to understand how the life of "things" works. To Avani, who accepted the challenge of accompanying my father in a tough time to be a fundamental part of this journey. Also to my brothers, Pedro Ivo and Guilherme, by the rare meetings of recent years, filled with "philosophical" conversations.

To Bruna, wife and friend, who understands the challenges of this career and has always been by my side, encouraging/criticizing in the moments of deafness, listening to endless conversations about the relationships between fish and deforestation, or simply being present.

To Lilian Casatti, who eight years ago accepted me as a member of his research group after an email sent in December 2010 (and replied in 3 minutes). And offered the opportunity to continue to work in a region which enchanted me in the MSc, and a second opportunity to pursue the doctorate, even after the slide on the first selection process. An advisor not only for academic and scientific issues but who cares too much about tutoring better people. That allows your students the freedom to dare, but leaves a cord subtly tied in the foot to pull back to earth and stay focused on what was proposed.

To my co-advisors, Silvio Ferraz, who has always helped me to "peel the pineapples" presented by landscape ecology and to open my eyes to a more regionalized view of deforestation impacts. David Hoeinghaus, who showed me another view of science (academically and philosophically), the ecology of hypothesis testing which, even in the case of stream fish, must always have the potential to dialogue with other areas of knowledge. I am glad by their dedication in the construction of this work, and by the friendships and established partnerships.

To Dana Infante, Jansen Zuanon, Danilo Boscolo and Rafael Leitão, by the availability and critical reading of this Thesis.

To Hoeinghaus Family – David, Ana and Brenda – who welcomed me so warmly, and provided, in Texas, the feeling of being at home.

To Angélica, who came from Colombia to clear the rondonian streams and built much of the database that was used in this work. And Felipe (B-nito) for help with sample design and fieldwork. Thanks for relying on maps and GPS, even when we had no idea where we were ...

To all members of the of Ichthyology Lab, especially Mônica, Angélica, Camilo, Jaque, Carol, Molina, Fernandinho, Ângelo e Bruno for the moments shared over those years, much learning and many laughs with all of you!

To Bárbara Callegari (MCP/ PUCRS), Ilana Fichberg (UNIFESP), Fernanda Martins (IFPR), Fernando Carvalho (UFMS), Flávio Lima (UNICAMP), Francisco Langeani (UNESP), Leandro Sousa (UFPA), Manoela

Marinho (MZUSP), Marcelo Britto (MNRJ), Marcelo Carvalho (IB/USP), and William Ohara (MZUSP) for help with fish identifications. To members of the ichthyology laboratories from UNESP and UNIR for help with the field work. To ICMBio (REBio Jaru) for logistical support. To SEDAM and to the extractivists' associations for permission to carry out the surveys in the areas of RESEX Rio Preto-Jacundá, RESEX Castanheira and RESEX Aquariquera.

Undergraduate friends, always present, especially those who became my second family in the years living at R.R.A. Biosfera.

I am very grateful to Pedro Gerhard, Anderson Ferreira and Katia Ferraz, advisors and friends, for being fundamental to my academic education. Pedro and Anderson, who introduced me to the stream fish ecology and biology, 16 years ago, and Katia to the Geographic Information Systems, 13 years ago, at the extinct Laboratory of Applied Animal Ecology.

To all from CEBB Rio Preto, which is a refuge to understand the afflictions and impermanence of daily life, and helps us not give so much weight to experiences.

To the University of North Texas for receiving me and providing the necessary structure for the development of my internship abroad during six months.

To the Graduate Program in Animal Biology (IBILCE / UNESP) and the Department of Zoology and Botany for the support, and;

To São Paulo Research Foundation (FAPESP), for grants awarded during my doctorate (2012/21916-0 and 2015/05827-6), and for Research Support that enabled the construction of the database (2010/17494-8).

To close this session of acknowledgments, I propose a reflection about the scenario of the last seven years, which I was involved with this work, regarding the scientific panorama and environmental policies in actual Brazil. Among other things, the forest code was revised and consolidated, reducing the protection of aquatic environments and, concomitantly, science suffered successive funding cuts. Perhaps these events are a reflection of the gap between scientists and society since few of us have endeavored ways to spread the fruits of our research in a way accessible to all people and, as a result, we lose the support of society. I hope that academics consider this point and build bridges, so that we have not worked in vain.

## Resumo

Os riachos da região Neotropical abrigam uma ictiofauna de pequeno porte, que frequentemente apresentam distribuições geográficas restritas e, em geral, são altamente dependentes da vegetação ripária para alimentação, abrigo e reprodução. A remoção da vegetação nativa da bacia hidrográfica está entre as modificações ambientais mais severas que afetam os ambientes aquáticos, de uma forma ainda pouco compreendida. Conhecer o histórico de desmatamento, em conjunto com a estrutura atual da paisagem, aumenta o poder das análises para avaliar os efeitos ecológicos do desmatamento. Além disso, os rumos distintos do processo de desmatamento podem afetar a composição e o funcionamento das comunidades de peixes de riachos. O rio Machado, em Rondônia, um importante tributário da bacia Amazônica, apresenta alta diversidade e endemismo, e um histórico recente de altas taxas de desmatamento e intensificação de uso do solo, que causam profundas alterações nos ambientes aquáticos. Sendo assim, o objetivo geral desta Tese foi compreender como o processo de desmatamento afeta a ictiofauna, usando a bacia do rio Machado como modelo. Para atingir esse objetivo, analisamos a relação entre a abundância das espécies de peixes com gradientes de tempo e extensão do desmatamento, para detectar quais espécies foram fortemente afetadas – positiva ou negativamente – pelo desmatamento (Capítulo 1). Conhecendo as respostas das espécies de peixes, determinamos como a composição e a estrutura da assembleia se relacionam com o tempo e a intensidade do desmatamento (Capítulo 2). E encerramos o estudo apresentando como os componentes ambientais influenciaram as taxas de substituição de espécies/atributos funcionais, considerando a idade do processo –

desmatamento recente ou antigo (Capítulo 3). Em suma, encontramos que (1.) As populações de peixes apresentaram respostas distintas ao tempo e à extensão do desmatamento, considerando as estruturas taxonômica e funcional. As respostas negativas ocorreram em baixos níveis de desmatamento e pouco tempo após o impacto. Os limiares de resposta positivos de algumas espécies ao desmatamento extremo são tardios, não compensam a perda de taxons sensíveis e provavelmente contribuem para a homogeneização biótica. (2.) As perdas de riqueza e abundância das espécies de peixes sensíveis ocorreram sincronicamente com a perda de estrutura de hábitat, mas houve um atraso temporal no aumento dos indicadores de homogeneização de habitat e o aumento da riqueza e abundância das espécies tolerantes segue esse atraso. (3.) A substituição de espécies/atributos funcionais foi diferente do esperado pelo acaso, indicando que processos determinísticos estruturam esta assembleia de peixes. Apesar de encontrarmos alta substituição de espécies, a substituição de atributos funcionais foi menor do que a esperada pela substituição de espécies. Significa que temos comunidades taxonomicamente distintas, mas funcionalmente similares, sugerindo que a substituição de espécies ocorre principalmente entre espécies funcionalmente equivalentes. Em conclusão, ao adicionar a camada temporal para analisar os estágios iniciais das mudanças de uso do solo nesta região da Amazônia, foi possível observar a extrema sensibilidade da assembleia de peixes ao desmatamento. Ainda, é possível considerar peixes de riachos como um grupo bandeira a ser incluído em planejamentos de conservação, com o objetivo de minimizar os efeitos da perda de biodiversidade em escala regional.

Palavras-chave: Código Florestal Brasileiro, Ponto de mudança, Limiares de diversidade, Ecomorfologia, Débito de extinção, Atributos funcionais, Dinâmica da paisagem, Atraso temporal, Bacia Amazônica, Bacia do Machado.

## **Abstract**

*The streams of the Neotropical region harbor a small-sized fish fauna, frequently with limited geographical distribution and, generally, highly dependent on riparian vegetation for feeding, shelter, and reproduction. The watershed native vegetation removal is one among several modifications, which effects on aquatic environments are not entirely understood yet. The knowledge of deforestation history along with current landscape structure enhances the power of analysis to evaluate ecological deforestation effects. Moreover, distinct ways of deforestation process might affect the fish community composition and functioning. The Machado river, in Rondônia, an important tributary of Amazon basin, exhibit high diversity and endemism and presents a recent history of high deforestation and land use intensification, which can cause profound changes in aquatic environments. Thus, the general aim of this Thesis is to comprehend how deforestation process influences the ichthyofauna. To reach this aim, we analyzed the relation between fish species abundance to environmental gradients of time and extent of deforestation, to detect which species were strongly affected – positively or negatively – by deforestation (Chapter 1). Knowing the fish species responses, we determined how fish assemblage composition and structure was related to deforestation time and intensity (Chapter 2). We end the study by presenting how the environmental components influenced the taxonomic and functional turnover rates, considering the process age – recent or old deforestation (Chapter 3). In Summary, we found that (1.) Stream fish populations present distinct responses to deforestation time and extent, regarding their taxonomic and functional structures, most negative threshold responses occurred at low levels of deforestation and soon after impact, so even in minimal change is expected to affect biodiversity*

*negatively. Delayed positive threshold responses to extreme deforestation by a few species do not offset the loss of sensitive taxa and likely contribute to biotic homogenization; (2.) The sensitive fish richness and abundance lost occurred synchronically to the habitat structure loss, but there was a time-lag response for habitat homogenization indicators, and tolerant fish richness and abundance increase followed this time-lag; and (3.) Species/functional traits turnover was different than expected by chance, indicating that deterministic processes are structuring this stream fish community. Although we found a high species turnover, functional traits turnover was lower than the expected by the species turnover. It means that taxonomically dissimilar, but functionally similar, suggesting that the species turnover is occurring mainly among functionally equivalents species. In conclusion, by adding the temporal layer to analyze the initial stage of land use changes in this portion of Amazon, was possible to verify the extreme sensitivity of fish assemblages to deforestation. And there is possible to consider stream fish as a flag group to be included on conservation plannings, aiming to reduce the effects of biodiversity loss on a regional scale.*

*Key-words: Brazil Forest Code, Change point, Diversity thresholds, Ecomorphology, Extinction debt, Functional traits, Landscape dynamics, Time lag, Amazon basin, Machado basin.*

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## **General Introduction**

The most diverse fish fauna from neotropics are found in Brazilian inland waters (Azevedo et al. 2010). In a recent review, Pelicice et al. (2017) called the attention to the severe threats to Neotropical freshwater fish diversity and pointed out the agribusiness as one of the primary drivers of deforestation, habitat loss and degradation in the region. In 2012, a new Forest Code (Federal Law 12.651) started to rule in Brazil, and, despite some advances in law control and implementation systems (Brançalion et al. 2016), it has reduced the protected area around streams. Before the code alteration, landowners should maintain a minimum riparian buffer of 30 meters measured from the largest seasonal stream bed of from water bodies up to 10 m wide (Brançalion et al. 2016). After code alteration depending on property size the landowners would maintain 5, 8, 15, 20 or 30 meters of riparian buffer from the center of the permanent channel, it implicates in an average reduction in streams protection by riparian buffers of  $47.8 \pm 33.1\%$  (Brançalion et al. 2016). Rivers and streams in Brazil harbor high rates of endangered fish (Casatti 2010) and are being hardly attacked by agribusiness expansion over pristine regions that were stimulated by the relaxation of the law. This harsh scenario of fast habitat destruction brings a significant challenge to the conservation researchers and decision makers: it is crucial to detect ecological patterns while also identifying and understanding processes and presenting solutions. All these tasks together demand a stellar effort without enough information on fish assemblages and instream variables. Notwithstanding, it is possible to rescue fish dataset on museums and establish preterit baselines

that can be indirectly used to compose the original species pool from impacted watercourses. Also, historical landscape analyses can represent the land-use change dynamics including their accumulated effects (Ferraz et al. 2009). For example, actual agripasture landscapes can face distinct historical processes of deforestation, like a fast and aggressive forest clearance, in the human settlement beginning or recently, or a continuous forest removal process along the time. In all situations, the side effects of forest removal on streams – habitat loss and homogenization – does not occur immediately after the deforestation event, and the aquatic community state change will face a time lag response, which sensitive species [to deforestation] could be being lost together with allochthonous structure loss while tolerant species has favored by habitat homogenization.

Past species occurrences and land-use history together are significant, but still limited without information that reveals how the essential instream features were. Complete stream inventories (fish + local variables) that will give snapshots along deforestation gradients can, therefore, reconstruct the history of diversity loss, ecological integrity and, by modeling techniques, predict what could happen inside the water in future scenarios of deforestation.

Dias et al. (2016) have identified important gaps in Brazilian stream studies and recommended the “... *need of focusing on recently developed ecological theories and frameworks and expanding the temporal and spatial scales of studies*”. The authors also pointed out about Brazilian founding agencies policies that favor short-term projects (1-3 years) due to long-term projects, which could “... *help to produce long-term monitoring data, sound ecological results*”

*and more comprehensive conservation plans*". We agree with the authors, but considering the actual crisis surrounding the Brazilian Science, that is facing several budget cuts (Angelo 2016), this sponsoring strategy probably will continue.

Therefore, how can we work around these problems? How can we turn species inventories in ecological integrity assessments? Recently, advances in GIS tools and the availability of high-quality land-use and climatic datasets (i.e., MapBiomas and WorldClim) already allow us to recover temporal and spatial scales information. However, we need to establish standardized protocols to assess instream variables that are even respecting sampling nature (e.g., inventories or ecological diagnostics), allows researchers to expand the scale of analysis and at the same time permit them to understand accurately how the agribusiness advance is imperiling Neotropical freshwater fish.

The Rondônia State, especially the Machado river basin, presents high rates of recent deforestation (Ferraz et al. 2005). This is a consequence of the human occupation, which has begun with the Federal Highway BR-364 construction at the end of the 1960's, strengthened in the early 1980's with the expansion of adjacent roads (Numata et al. 2009). Consequently, the region presents a unique model for studies with the aim of investigating the communities' ecological attributes variation.

Considering this, the general aim of this Thesis is to comprehend how deforestation process influences the ichthyofauna. To reach this aim, we analyzed the relation between fish species abundance to environmental gradients of time

and extent of deforestation, to detect which species were strongly affected – positively or negatively – by deforestation (Chapter 1). Knowing the fish species responses, we determined how fish assemblage composition and structure was related to deforestation time and intensity (Chapter 2). We end the study by presenting how the environmental components influenced the taxonomic and functional turnover rates, considering the process age – recent or old deforestation (Chapter 3). All chapters were prepared according to Conservation Biology journal rules.

In 2015, the Division for Sustainable Development of United Nations proposed the 2030 Agenda, a new plan of action for people, planet, and prosperity, with 17 Sustainable Development Goals and 169 associated targets at its core (UN 2017). In this sense, the present Thesis fits on the SDG 15, Life on Land, which preconizes *"Protect, restore and promote sustainable use of terrestrial ecosystems, sustainably manage forests, combat desertification, and halt and reverse land degradation and halt biodiversity loss."* The first target of this SDG, expect *"... by 2020, ensure the conservation, restoration and sustainable use of terrestrial and inland freshwater ecosystems and their services, in particular forests, wetlands, mountains, and drylands, in line with obligations under international agreements."* Inside this, the proportion of important sites for terrestrial and freshwater biodiversity that are covered by protected areas, by ecosystem type, is one indicator (# 15.1.2) to evaluate this target. Therefore, we expect that this Thesis also can contribute to a framework to quantify this indicator, considering the freshwater biodiversity.

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## **Chapter 1**

### **Threshold responses of Amazonian stream fish to timing and extent of deforestation\***

\*Paper published on Conservation Biology:

Brejão GL, Hoetinghaus DJ, Pérez-Mayorga MA, Ferraz SFB, Casatti L. 2018. Threshold responses of Amazonian stream fishes to timing and extent of deforestation. Conservation Biology. doi:10.1111/cobi.13061

## **Abstract**

Deforestation is a primary driver of biodiversity change through habitat loss and fragmentation. Stream biodiversity may not respond to deforestation in a simple linear relationship. Rather, threshold responses to extent and timing of deforestation may occur. Identification of critical deforestation thresholds is needed for effective conservation and management. We tested for threshold responses of fish species and functional groups to degree of watershed and riparian zone deforestation and time since impact in 75 streams in the western Brazilian Amazon. We used remote sensing to assess deforestation from 1984 to 2011. Fish assemblages were sampled with seines and dip nets in a standardized manner. Fish species ( $n = 84$ ) were classified into 20 functional groups based on ecomorphological traits associated with habitat use, feeding, and locomotion. Threshold responses were quantified using threshold indicator taxa analysis. Negative threshold responses to deforestation were common and consistently occurred at very low levels of deforestation (<20%) and soon after impact (<10 years). Sensitive species were functionally unique and associated with complex habitats and structures of allochthonous origin found in forested watersheds. Positive threshold responses of species were less common and generally occurred at >70% deforestation and >10 years after impact. Findings were similar at the community level for both taxonomic and functional analyses. Because most negative threshold responses occurred at low levels of deforestation and soon after impact, even minimal change is expected to negatively affect biodiversity.

Delayed positive threshold responses to extreme deforestation by a few species do not offset the loss of sensitive taxa and likely contribute to biotic homogenization.

## **Introduction**

Deforestation is a primary driver of habitat loss and fragmentation affecting terrestrial and aquatic ecosystems. At the watershed level, land-use changes affect large-scale processes related to chemistry, hydrology, and sediment delivery (Allan & Johnson 1997; Allan 2004). For example, deforestation contributes to widespread biodiversity change in streams through its effects on flow complexity, depth, substrate composition, stream bank stability, and structural complexity (Gorman & Karr 1978; Cruz et al. 2013). Previous studies in the Neotropics demonstrate that high deforestation rates in rainforests, either in the past or in the present, affect many of the ecological processes occurring in streams (Wright & Flecker 2004; Paula et al. 2011). This includes the alteration of stream fish diversity and assemblage structure (Bojsen & Barriga 2002; Ferreira et al. 2012; Casatti et al. 2015). Some of the aforementioned impacts may be mediated by intact riparian zones, which provide sediment filtration; bank and flow stability; regulation of temperature and primary production; maintenance of instream habitat structure, complexity, and stability (i.e., through input of leaf litter, trunks, and roots); and food resources (e.g., Pusey & Arthington 2003; Ferreira et al. 2012; Zeni & Casatti 2014).

Stream biodiversity may not respond to deforestation (e.g., proportion of the watershed affected, degree of impact to the riparian zone, and timing of deforestation) in a linear fashion. Instead, nonlinear responses to the proportion of the watershed affected by deforestation and habitat loss may be expected (Smucker et al. 2013; Rodrigues et al. 2016), similar to well-documented

threshold responses to habitat loss and fragmentation of tropical birds and mammals (e.g., Andrén 1994; Fahrig 2003). For these taxa, the threshold response and significant declines in abundances often occurs at 60–70% of original habitat lost (e.g., Andrén 1994; Ochoa-Quintero et al. 2015; Muylaert et al. 2016). In comparison, threshold responses of fish and aquatic invertebrate diversity in temperate aquatic environments have been observed at even lower levels of watershed impact (e.g., < 50% habitat loss) (Baker & King 2010; Smucker et al. 2013).

The chronology of disturbance can also be an important determinant of current stream and watershed diversity (Harding et al. 1998; Iwata et al. 2003; Burcher et al. 2008). Effects of deforestation on stream biodiversity may exhibit significant time lags because the primary effect of deforestation (i.e., habitat simplification and homogenization) does not occur immediately after a deforestation event and populations may not respond immediately to habitat changes. Thus, patterns of stream biodiversity could be expected to vary in watersheds with similar levels of deforestation but distinct histories of land-use change (e.g., timing of first or greatest change in land cover). Simultaneously evaluating the effects of deforestation history and current landscape structure should enhance the power of analyses to reveal the ecological effects of this impact.

Detecting species and community threshold responses to spatial and temporal environmental gradients may provide important information for management and conservation (e.g., Groffman et al. 2006; Baker & King 2010;

Dodds et al. 2010), such as identification of change points that induce regime shifts or irreversible homogenization (Scheffer et al. 2001; Andersen et al. 2008). We tested for threshold responses of fish species and functional groups to deforestation and timing of impact for 75 streams along a deforestation gradient in the western Brazilian Amazon. Rondônia State, especially the Machado River basin, has experienced high rates of slash and burn deforestation (Ferraz et al. 2005) facilitated by construction of a highway in the late 1960s and by the further expansion of side roads in the early 1980s (Numata et al. 2009). This region was subjected to 2 strong deforestation waves: 19.5% of the basin was deforested from 1991 to 1995 and an additional 23.4% was cleared from 2001 to 2004 (INPE 2016). Consequently, the region provides a unique model for studying ecological responses regarding spatial and temporal dynamics of deforestation.

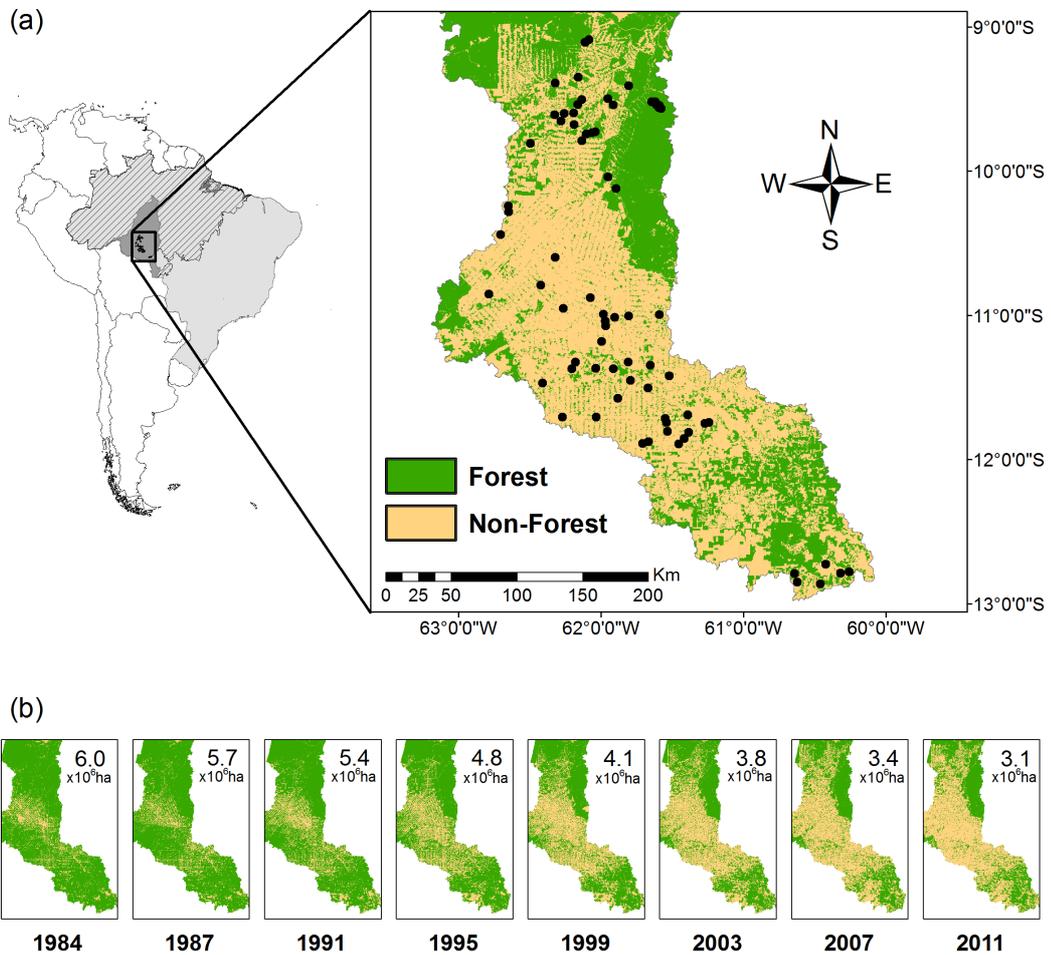
Our hypothesis is that fish populations exhibit nonlinear responses to extent and timing of deforestation; that is, a threshold response at 60% of the watershed deforested (based on previous research on vertebrates in the tropics [e.g., Andrén 1994]) and a time lag of approximately the median life expectancy for the species in these systems (estimated 2–3 years). Further, we expect threshold responses will occur at lower levels of riparian deforestation than watershed deforestation and the rate of deforestation will mediate the above responses (i.e., stronger responses to relatively more intense impacts). Finally, we expect threshold responses of functional groups will be delayed or of lower magnitude than taxonomic responses if functionally redundant species

(Winemiller 1991; Toussaint et al. 2016) compensate for changes in abundances of individual species.

## **Methods**

### *Study Area*

We surveyed 75 first- to third-order streams in the Machado River basin, Brazil (Fig. 1.1). The 1,200 km Machado River is a tributary of the Madeira River, and its 75,400 km<sup>2</sup> catchment area drains the most populated region of Rondônia, Brazil (Fernandes & Guimarães 2002). This region has many terra firme streams, which are intermittent during most of the dry season (Fernandes & Guimarães 2002). The sampled streams were shallow with low-velocity flow and warm temperatures. The streambeds were predominantly sand, litter pack, and large woody debris, and stream banks usually provided submerged microhabitat structure derived from the riparian environment such as tree roots and grasses (Supporting Information). The climate is tropical humid: temperatures range from 19 to 33 °C and annual rainfall is 2,500 mm (Krusche et al. 2005). The land cover in this region includes primary forest (open humid tropical forest), secondary forest, and pasture (Ferraz et al. 2009).



**Figure 1.1.** (a) Sampled sites (75 black dots) and land cover in the Machado River basin, Brazil. The inset map of Brazil depicts the relative location of the study area (black) within the Madeira River basin (dark gray) (diagonal stripes, Amazon biome). (b) Chronology of deforestation in the Machado River basin and approximate forest area (millions of hectares) in each year.

### *Environmental Gradients*

Land-cover data were quantified using Landsat 5TM images (30 x 30 m resolution). Land use was classified as mature forest, secondary or degraded forest, and nonforest according to the supervised classification method (Jensen 2007) in ERDAS (version 9.2) for multiple dates (1984, 1987, 1991, 1995, 1999, 2003, 2007, and 2011). Secondary and degraded forests (i.e., previously

deforested areas with regrowth or forests subject to edge effects) comprise <1% of land use; therefore, we considered only forest and nonforest categories. We used ArcGIS (version 9.3) to quantify 4 environmental gradients representing extent and chronology of landscape changes for watershed and 100-m-wide riparian zones for all 75 streams (Supporting Information). We quantified deforestation as the amount of forest lost since conditions were pristine and time since <40% forest cover as the number of years since forest cover was reduced to <40%. Because some deforestation occurred prior to the earliest available Landsat images in 1984, we quantified the amount of deforestation since 1984 as the difference in forest extent between 1984 and 2011 image dates. Time since the greatest land-use change was quantified as the number of years since the time interval with the greatest decrease in forest cover since 1984.

### *Sampling Methods*

Streams were sampled once during the dry season (August–October 2011 and June–July 2012). Sampling methods were standardized to allow for comparisons of species abundances across sites. Each stream reach was 80 m long and isolated prior to sampling with block nets (5-mm mesh). After reach delimitation, instream physicochemical and riparian ecotone (hereafter instream) habitat variables were quantified using standard methods (Supporting Information 1.1). We used principal components analysis (PCA) to reduce the dimensionality in the multivariate data set. Resulting axis scores were used in subsequent analyses to test for relationships between watershed and riparian deforestation and

instream habitat (Supporting Information 1.1). Fishes were sampled from all available microhabitats by 2 collectors using a seine (1.5 x 2 m, 2-mm mesh) and dip nets (0.5 x 0.8 m, 2-mm mesh) for 1 hour. This method accounts for 90% of expected richness for this set of streams (Casatti et al. 2013). Fishes were collected under Instituto Chico Mendes de Conservação da Biodiversidade permits 4355-1/2012. All individuals were identified with assistance from taxonomic specialists, and voucher specimens are deposited in the Coleção de Peixes do Departamento de Zoologia e Botânica at the Universidade Estadual Paulista “Júlio de Mesquita Filho,” São José do Rio Preto, São Paulo State, Brazil.

#### *Species Traits and Functional Groups*

Ten ecomorphological traits related to habitat use, resources capture, and locomotion (Table 1.1) were quantified based on 11 measurements from adult individuals of each species (1–5 individuals per species). We measured linear distances, area, and width of each individual to the nearest 0.01 mm with a stereomicroscope (Zeiss Discovery V12 SteREO) coupled with imaging software (Axio-Vision Zeiss) and a digital caliper. Subsequent threshold analyses required discrete functional groups (FGs) rather than continuous trait data across species. Therefore, we used hierarchical agglomerative clustering (UPGMA) and the ecomorphological traits to classify the 84 fish species with >3 occurrences into FGs. Clustering was performed following procedures in Borcard et al. (2011), and fit of the dendrogram to the original trait matrix was assessed using Pearson’s  $r$  correlation computed with the trait dissimilarity matrix and the cophenetic

distance matrix. Silhouette plots were used as an objective method to identify the number of meaningful FGs ( $k$ ) within the cluster. The cophenetic correlation was 88.5%, and the 84 species were assigned to 20 FGs with 1 to 45 species (Supporting Information 4.1). The abundance of FGs was calculated as the sum of individual species abundances within each group. Clustering, cophenetic correlations and silhouette plots were calculated using the R packages `stats` and `cluster`.

Table 1.1. Ecomorphological traits used to quantify functional groups.

Traits	Codes	Calculation <sup>a</sup>	Interpretation
Relative height of head	RHHd	$\frac{HdH}{MBH}$	Larger relative values of head height are found in fishes which feed on larger prey. Larger values for this index are expected for piscivores ( Winemiller 1991)
Compression index	CI	$\frac{MBH}{MBW}$	High values indicate laterally compressed fishes, typically inhabiting lentic habitats (Watson & Balon 1984)
Relative depth	RD	$\frac{MBH}{SL}$	Low values indicate fishes inhabiting fast waters. Body depth is directly related to the ability to perform vertical spins (Gatz 1979)
Index of ventral flattening	IVF	$\frac{MLH}{MBH}$	Low values indicate fishes that are able to maintain their spatial position in environments with high hydrodynamism (Hora 1930)
Relative eye position	EP	$\frac{MEH}{HdH}$	Eye position is related to vertical habitat preference (Gatz 1979). High values indicate dorsally located eyes, typical of benthic fish (Mahon 1984; Watson & Balon 1984)
Relative area of pectoral fin	RAPF	$\frac{PFA}{BA}$	High values indicate slow swimmers that use pectoral fins to perform maneuvers and breakings, or fish inhabiting fast waters, which use their pectoral fins as airfoils to deflect the water current upwards and thereby maintain themselves firmly attached to the substrate (Mahon 1984; Watson & Balon 1984)
Pectoral fin aspect ratio	PFAR	$\frac{LPF}{WPF}$	High values indicate long fins, typical of fishes that swim for long distances (Watson & Balon 1984) or pelagic fishes that swim constantly (Casatti & Castro 2006)
Fineness coefficient	FC	$\frac{SL}{\sqrt{MBH \times MBW}}$	Assesses the influence of body shape on the ability to swim. Values from 2 to 6 indicate low drag; the optimum ratio for swimming efficiency is 4.5 (Blake 1983)
Relative height of caudal peduncle	RHPd	$\frac{CPH}{MBH}$	Lower values indicate greater maneuverability potential (Winemiller 1991)
Standard length	SL		Body size affects many aspects of trophic ecology and habitat use

<sup>a</sup>Morphological measures: BA: body area; MBH: maximum body height; PFA: pectoral fin area; LPF: maximum length of pectoral fin; CPH: caudal peduncle height; MBW: maximum body width; MLH: Mid-line height; WPF: maximum width of pectoral fin; SL: standard length; HdH: head height; MEH: Mid-line eye height.

### *Data Analyses*

We used threshold indicator taxa analysis (TITAN) (Baker & King 2010) to test our hypotheses of fish population and assemblage responses to extent and chronology of watershed and riparian deforestation. We used species and functional group abundances as response variables along each environmental gradient to assess threshold responses for both the taxonomic and functional datasets. We analyzed each combination of response variable and environmental gradient for a total of 8 threshold analyses. Analysis of threshold indicator taxa is a nonparametric change-point analysis that is combined with indicator species analysis to identify a change point, the direction (i.e., increase or decrease) of the change, and the magnitude of the response (i.e., change in abundance) for each species or FG along an environmental gradient (Baker & King 2010).

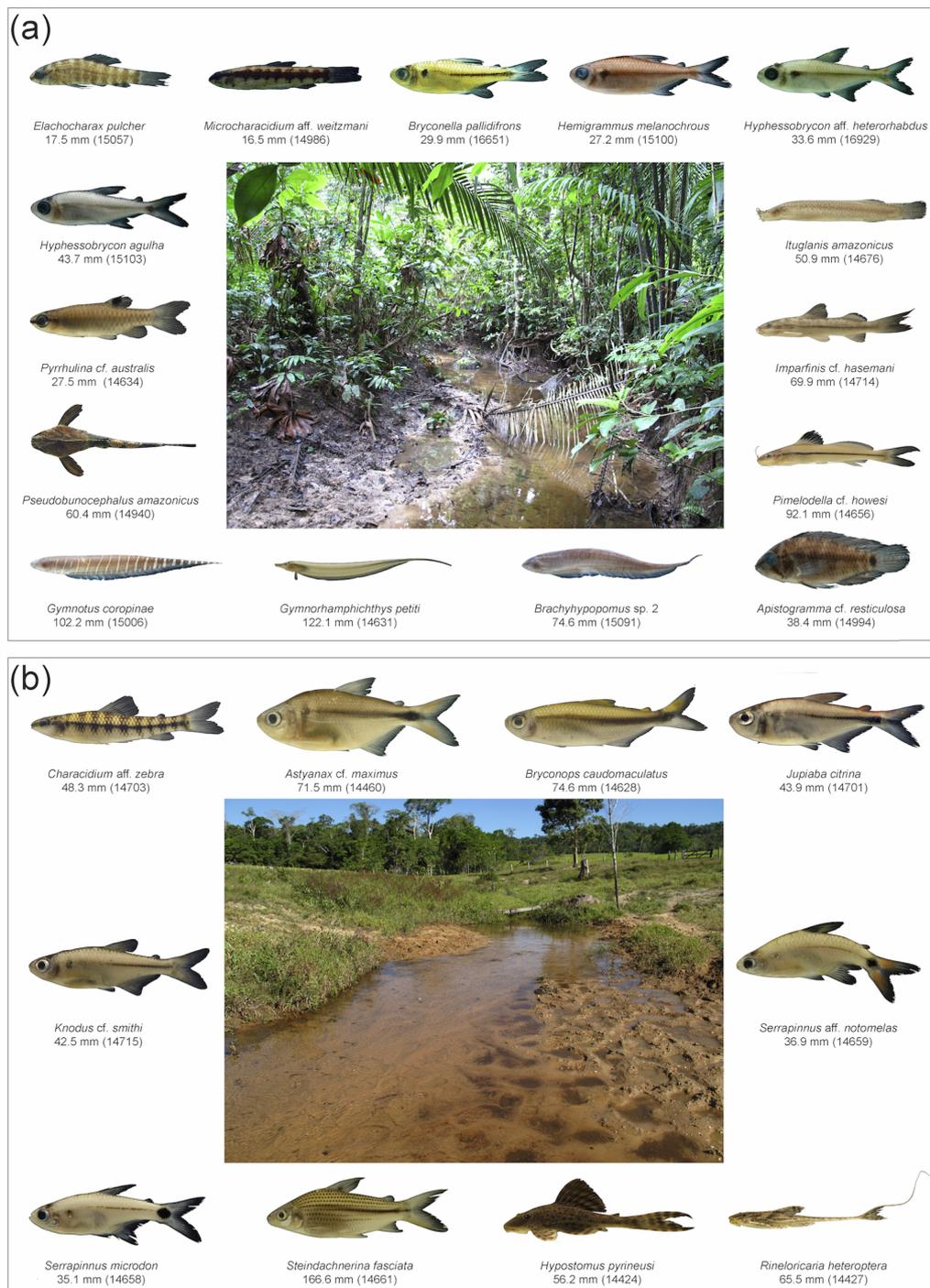
Magnitudes of threshold responses are standardized with z-scores to facilitate comparisons across taxa and FGs in a multivariate dataset. The 95th percentile range from bootstrapping provided an index of the slope of the threshold response. Community-level change points were assessed separately for positive and negative taxa and FGs as the value of the gradient with the largest cumulative  $z^+$  (i.e.,  $\text{sum}[z^+]$ ) and  $z^-$  (i.e.,  $\text{sum}[z^-]$ ), respectively. We used nonparametric change-point analysis (nCPA) calculated with Bray–Curtis distances for comparison with  $\text{sum}(z^+)$  and  $\text{sum}(z^-)$ . Abundance data were standardized using the Hellinger standardization method prior to analyses (Legendre & Gallagher 2001). Cut-off levels for TITAN were  $\alpha < 0.05$ , purity  $\geq 0.95$ , and reliability ( $\text{rel05}$  and  $\text{rel01}$ )  $\geq 0.95$ . Purity is the proportion of change-

point response directions (positive or negative) among bootstrap replicates that agree with the observed response, and reliability is the proportion of bootstrap change points with IndVal scores that consistently result in  $p$  values below the significance levels of  $\alpha < 0.05$  (rel05) and 0.01 (rel01) (Baker & King 2010). Analyses were conducted using R script provided in Baker and King (2010).

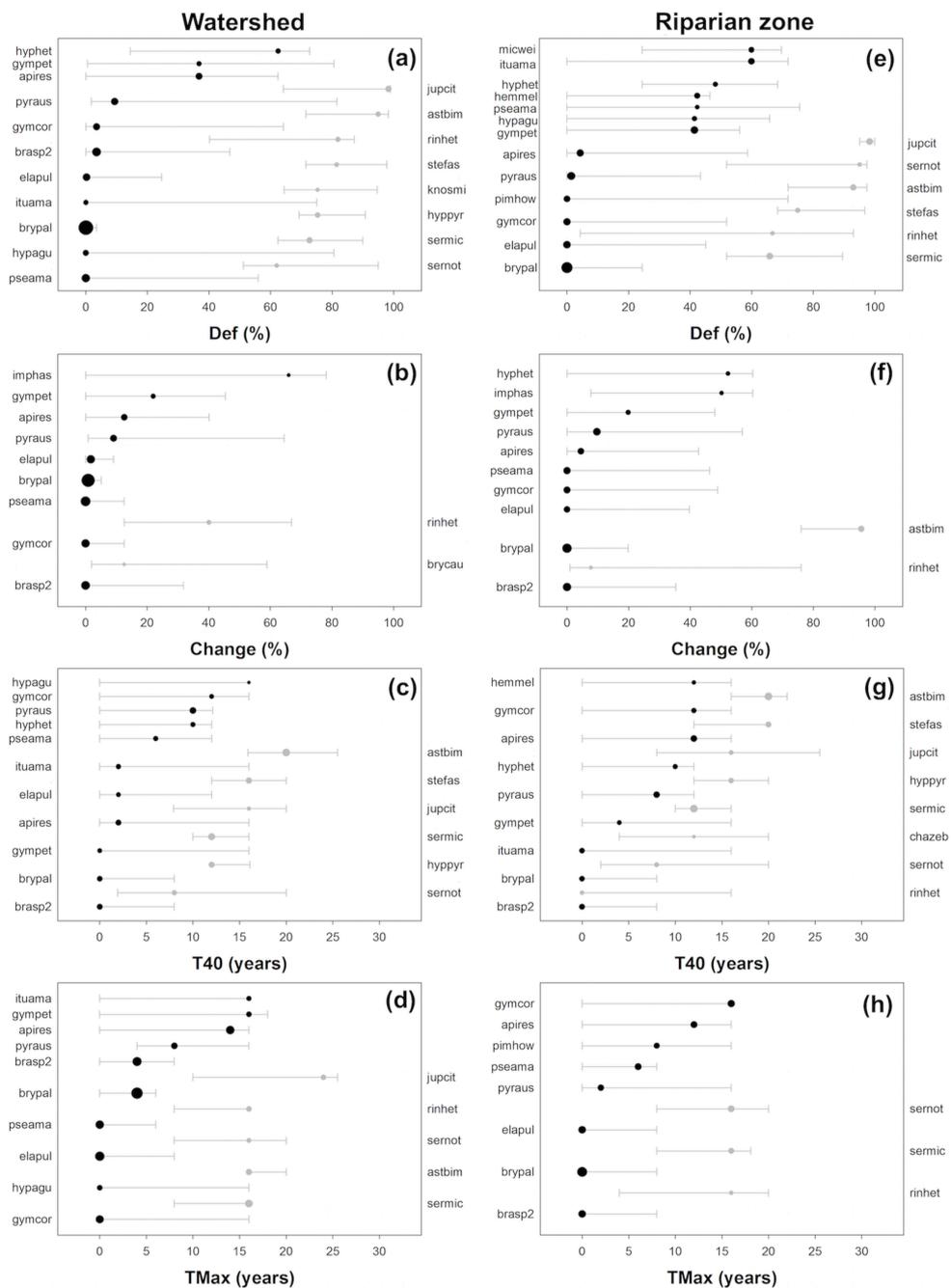
## Results

Instream habitat was significantly correlated with watershed and riparian deforestation (Supporting Information). Streams with low levels of deforestation were characterized by complex habitat and structure of allochthonous origin (e.g., litter packs, large woody debris, and submerged roots along stream banks), whereas streams with higher levels of deforestation were characterized by homogenous conditions (e.g., sandy substrate, grass along stream margins) (Supporting Information). We collected 22,355 individuals of 84 species, 22 families, 4 orders, and 20 FGs based on our clustering analysis (Supporting Information 3.1). Seventy-one species and 17 FGs had significant IndVal scores ( $p < 0.05$ ) along gradients of deforestation and time since change (Supporting Information). Of those, 25 species (29.7%) and 5 FGs (25%) attained our stringent cut-off levels for purity and reliability (Supporting Information 5.1). For comparison, relaxing the cut-off levels (purity  $\geq 0.75$ ; rel05 and rel01  $\geq 0.75$ ) yielded 49 species (58.0%) and 10 FGs (50%) dominated by negative threshold responses (i.e., 29 species and 9 FGs).

Most species had significant threshold responses to multiple gradients, and responses were largely consistent across gradients of watershed and riparian zone deforestation and time since impact. Fifteen species exhibited negative threshold responses (z<sup>-</sup>), and 10 species had positive threshold responses (z<sup>+</sup>) (Fig. 2.1). Most negative species threshold responses occurred at very low levels of deforestation (e.g., < 20%) and soon after impact (e.g., < 10 years; Fig. 3 & Supporting Information). *Brachyhypopomus* sp.2, *Bryconella pallidifrons*, *Elachocharax pulcher*, *Gymnotus coropinae*, *Hyphessobrycon agulha*, *Ituglanis amazonicus*, *Pseudobunocephalus amazonicus*, and *Microcharacidium* aff. *weitzmani* were extremely sensitive to deforestation and had negative threshold responses at or near 0% (Fig. 3 & Supporting Information). In contrast, positive species threshold responses generally occurred at > 60% deforestation and > 10 years after impact (Fig. 3 & Supporting Information). Of those species, *Jupiaba citrina* and *Astyanax* cf. *bimaculatus* had the most extreme positive threshold responses; change points were at levels of watershed deforestation exceeding 90% and 16 to 24 years following the maximum deforestation event. Four species had threshold responses to only riparian deforestation (*Hemigrammus melanochrous* [z<sup>-</sup>], *M. weitzmani* [z<sup>-</sup>], *Pimelodella howesi* [z<sup>-</sup>], and *Characidium* aff. *zebra* [z<sup>+</sup>]), and 2 species had threshold responses only to watershed deforestation (*Bryconops caudomaculatus* [z<sup>+</sup>], *Knodus* cf. *smithi* [z<sup>+</sup>]) (Supporting Information).

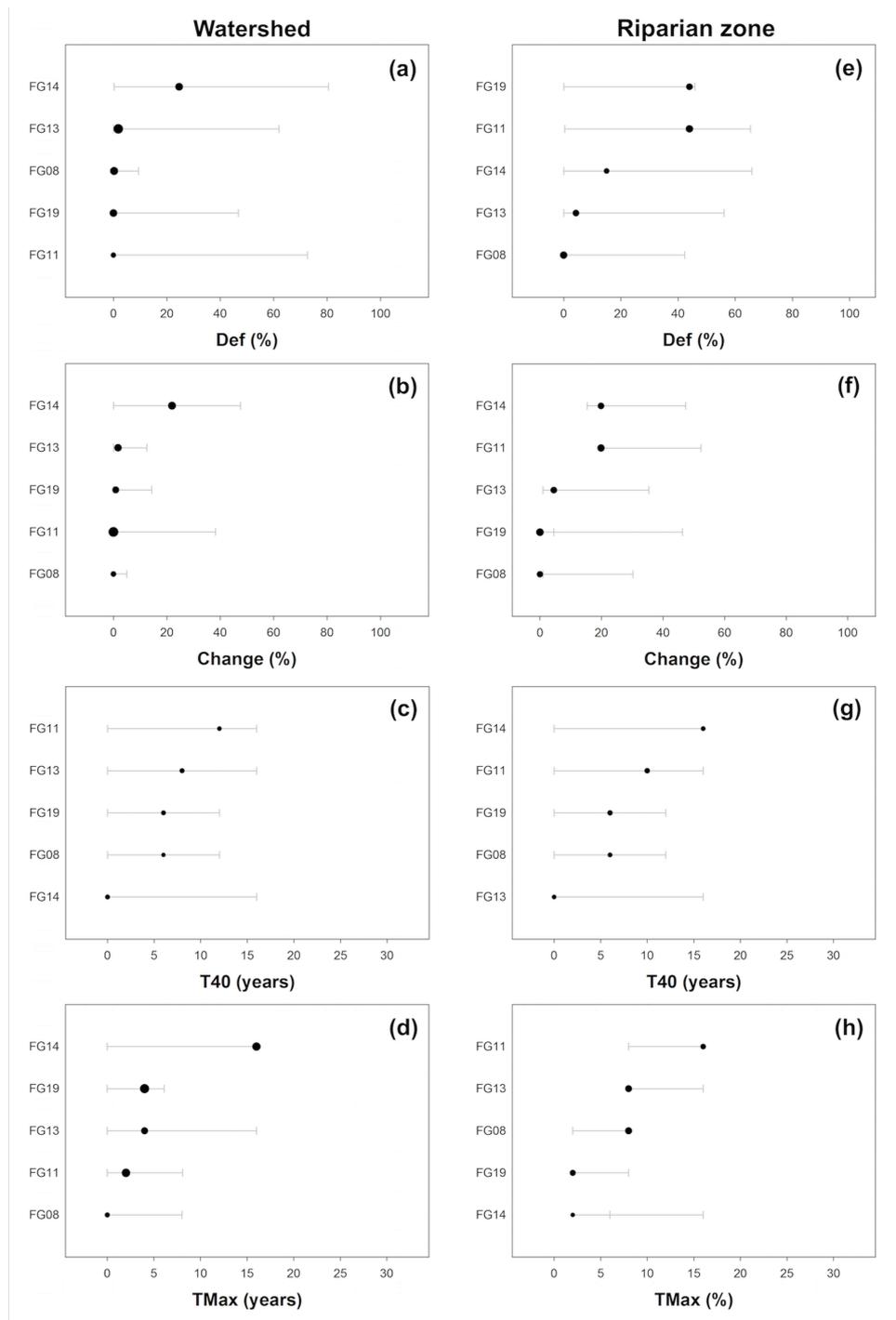


**Figure 2.1.** Fish species with (a) significant negative threshold responses to deforestation and (b) significant positive threshold responses to deforestation. See Fig. 3 and Supporting Information for details. Fish photos by F.R. Carvalho and F. Martins, and stream photos by G.L.B.



**Figure 3.1.** Species threshold responses to watershed and riparian-zone deforestation (Def, deforestation; change, land-use change since 1984; TMax, time since the greatest land-use change since 1984; T40, time since < 40% of forest cover; black, negative threshold responses [abbreviated species name on the left]; gray, positive threshold responses [abbreviated species name on the right]; circles, change point along the gradient; circle size, relative magnitude of response scaled by z score; error bars, 95th percentile range; species abbreviations are the first 3 letters of both the genus and species names [full species names are provided in Supporting Information 3.1]).

All 5 FGs that met our criteria exhibited negative threshold responses, and responses were consistent across all gradients (Supporting Information 5.1). Similar to the species analyses, negative threshold responses occurred at very low levels of deforestation and soon after impact (Fig. 4.1 & Supporting Information 5.1). In general, the traits distinguishing these FGs are associated with use of complex habitat and allochthonous materials from the riparian forest. For example, FG 08 (composed of *B. pallidifrons* and *Tytocharax madeira*, both small sized characins with compressed and relatively deep bodies and fine pectoral fins) and FG 13 (represented by *G. coropinae*, which has large body size and fine pectoral fins) are strongly associated with marginal roots and stems. Function groups 11 (*E. pulcher* and *Microcharacidium* aff. *weitzmani*) and 19 (*P. amazonicus*) are associated with banks of leaf litter. In contrast with the previous FGs associated with structure, FG 14 was composed of a single psammophilic species (*Gymnorhamphichthys petiti*) that uses its long snout and electric field to probe sandy substrates for small invertebrates during the night and buries itself in the sandy substrate during the day (Brejão et al. 2013). The threshold for FG 14 occurred at higher levels of deforestation.



**Figure 4.1.** Fish functional-group (FG) threshold responses to watershed and riparian-zone deforestation (Def, deforestation; Change, land-use change since 1984; TMax, time since the greatest land-use change since 1984; T40, time since < 40% of forest cover; circles, change point along the gradient; circle size, relative magnitude of response scaled by z score; error bars, 95th percentile range). Only negative threshold responses are presented because there were no significant positive threshold responses of FGs along any of the deforestation gradients.

At the community level for both taxonomic and functional analyses,  $\text{sum}(z^-)$  was  $< 5\%$  and  $\text{sum}(z^+)$  was  $> 40\%$  for all gradients quantifying degree of watershed deforestation (Table 2.1).  $\text{Sum}(z^-)$  was  $< 45\%$  for riparian zone deforestation and  $< 5\%$  for land-use change since 1984, and  $\text{sum}(z^+)$  was  $> 65\%$  for all riparian gradients (Table 2.1). For the taxonomic analyses and temporal gradients of both watershed and riparian-zone deforestation,  $\text{sum}(z^-)$  was  $\leq 4$  years since impact and  $\text{sum}(z^+)$  was  $\geq 14$  years since impact for all but one gradient. In contrast,  $\text{sum}(z^-)$  was 2–14 years since impact and  $\text{sum}(z^+)$  was  $\geq 16$  years since impact for the functional analyses (Table 2.1). For comparison, nCPA community change points were intermediate to the  $\text{sum}(z)$  values for each gradient (Table 2.1), typically near the 95th percentile for  $\text{sum}(z^-)$  or the 5th percentile for  $\text{sum}(z^+)$ .

**Table 2.1.** Observed (obs.) community-level thresholds along deforestation gradients and 5th and 95th percentile confidence intervals.

	Taxonomic			Functional		
	Obs.	5%	95%	Obs.	5%	95%
<i>Watershed deforestation</i>						
TITAN sum(z-)	0.0	0.0	66.3	0.3	0.0	55.9
TITAN sum(z+)	75.3	66.3	85.8	81.5	43.2	94.6
nCPA (Bray-Curtis)	62.0	0.5	80.6	0.5	0.0	97.8
<i>Watershed land use change since 1984</i>						
TITAN sum(z-)	0.0	0.0	22.0	0.0	0.0	9.1
TITAN sum(z+)	40.1	22.0	80.2	60.3	22.0	88.3
nCPA (Bray-Curtis)	22.0	0.0	38.5	0.9	0.0	91.0
<i>Time since greatest watershed land-use change</i>						
TITAN sum(z-)	4	0	8	4	0	8
TITAN sum(z+)	16	8	20	16	8	20
nCPA (Bray-Curtis)	16	2	16	2	0	24
<i>Time since &lt;40% watershed forest cover</i>						
TITAN sum(z-)	2	0	16	14	0	16
TITAN sum(z+)	16	8	16	16	4	20
nCPA (Bray-Curtis)	14	1.9	16	12	0	20
<i>Riparian zone deforestation</i>						
TITAN sum(z-)	43.4	0.0	56.1	44.0	0.0	51.8
TITAN sum(z+)	66.8	51.8	87.6	66.8	44.0	97.8
nCPA (Bray-Curtis)	43.4	8.5	68.5	0.4	0.0	98.1
<i>Riparian zone land use change since 1984</i>						
TITAN sum(z-)	0.0	0.0	39.7	0.0	0.0	36.8
TITAN sum(z+)	19.9	7.8	89.1	77.4	4.5	77.4
nCPA (Bray-Curtis)	19.9	0.0	49.0	0.0	0.0	95.6
<i>Time since greatest riparian zone land-use change</i>						
TITAN sum(z-)	0	0	16	2	0	16
TITAN sum(z+)	4	4	27	20	2	25.5
nCPA (Bray-Curtis)	16	0	16	4	0	27
<i>Time since &lt;40% riparian zone forest cover</i>						
TITAN sum(z-)	4	0	16	12	0	16
TITAN sum(z+)	14	8	16	16	4	22
nCPA (Bray-Curtis)	12	0	16	8	0	20

## **Discussion**

We found negative threshold responses at very low levels of deforestation over space and time gradients that were consistent both for taxonomic and functional analyses. In general, negative thresholds were observed for species with specific suites of functional traits associated with allochthonous structures and leaf litter. Positive threshold responses were less common and occurred at high levels and time since deforestation. Species that exhibited positive threshold responses did not have consistent traits or were not ecomorphologically unique (which precluded positive threshold responses for FGs). The observed negative threshold values were much lower than our initial hypothesis, and populations of the species that responded negatively to deforestation appeared to persist for 2 or 3 generations following impact before exhibiting sharp declines. Because negative threshold responses were more common and primarily occurred at low levels of deforestation, even minimal deforestation is expected to negatively affect stream fish biodiversity and assemblage structure, and these responses will likely occur relatively soon after impact. Delayed positive threshold responses to extreme deforestation of a few species would not offset the declines of more sensitive taxa and would likely contribute to further biotic homogenization of the faunas.

For vertebrate taxa in terrestrial environments, such as birds and mammals, it is common to find a threshold response in species richness or diversity from 50% to 70% of the area deforested (e.g., Andr n 1994; Ochoa-Quintero et al. 2015; Muylaert et al. 2016). In addition to differences in habitat use (e.g., streams nested within forest vs. forested landscapes) and life-history attributes (e.g.,

generation time, fecundity) that may affect responses among taxa, the biological scale of the response variable (i.e., species richness vs. population sizes of individual species) can affect the presence and attributes of threshold responses. In contrast with the aforementioned studies on tropical vertebrates that analyzed changes in species richness, the analytical approach we used distinguishes individual species threshold responses that are either increasing or decreasing in abundances. This is an important feature given that species can respond to environmental change differently, changes in population sizes are likely detectable before extirpation, and species loss may be offset by establishment of cosmopolitan species.

Similar to our findings, previous researchers who used TITAN to test for threshold responses of temperate stream invertebrates observed responses at lower levels of land-cover change, such as  $\text{sum}(z^-) = 20.9\%$  deforestation for diatoms (Smucker et al. 2013) and  $\text{sum}(z^-) = 1.8\%$  developed land for aquatic invertebrates (Baker & King 2010). Integrating data from terrestrial and aquatic taxa from the same system in parallel TITAN analyses would be an appealing approach to test for differences in threshold responses among taxa, scales and timing of responses, and size and location (e.g., riparian or interior watershed) of residual forest fragments. For example, in a multitaxa evaluation of an old (183 years) and severe (95%) deforestation event in Singapore, Brook et al. (2003) detected population declines and local extinction processes for several taxa. Birds and mammals could use small forest patches to recolonize forest remnants protected in reserves, whereas forest-dependent stream fish species were restricted

to a small area (5 ha) within a single reserve. Strong dependence of fish species to instream habitat complexity provided by structures from the terrestrial environment, and isolation of populations in forest islands, demonstrate a lack of connectivity between most forest remnants and the hydrological network.

Composite change points for negative threshold responses for taxonomic and functional analyses were almost identical for extent of deforestation (sum[z-] = 0.0 and 0.3, respectively), extent of land-use change since 1984 (both sum[z-] = 0.0), and time since the greatest watershed land-use change (both sum[z-] = 4). Each FG that exhibited a significant threshold response had only 1 or 2 ecomorphologically unique species, which likely contributed to the ability to distinguish a consistent response. This suggests that streams in forested watersheds harbor species with complementary ensembles of ecomorphological traits, and we detected strong negative threshold responses of functional groups associated with habitat structures derived from the terrestrial environment and that are commonly found in streams draining densely forested watersheds (Supporting Information; Bordignon et al. 2015). This result corresponds with our predictions based on a large body of research on effects of land-cover change on stream fish assemblages and recent findings of Leitão et al. (2017) from 2 different regions of the Amazon. However, not all species with negative threshold responses were represented in FGs with significant negative responses. When combined with the species exhibiting positive threshold responses (and no significant positive threshold responses for FGs), this illustrates that many of our FGs defined by ecomorphological traits incorporated species with divergent responses to

deforestation gradients. Although the FGs that exhibited negative threshold responses were composed of ecomorphologically unique species, functional redundancy may buffer the system to some changes in species occurrences or abundances.

However, other traits (e.g., additional ecomorphological traits, reproductive life-history traits, trophic guilds [Hoeinghaus et al. 2007, Pease et al. 2012, Leitão et al. 2017]) that may mediate species responses to deforestation may not have been sufficiently captured in our ecomorphological analysis. For example, in addition to significant trait–environment relationships for the same traits we used, Pease et al. (2012) also found relationships between other ecomorphological and life-history traits (e.g., mouth gape, gill raker length, gut length, clutch size) and environmental gradients in tropical streams of Mexico. Leitão et al. (2017) observed different relationships between type of dentition (i.e., viliform, spoon or comb-shaped teeth) and fish-assemblage responses to riparian zone or watershed deforestation (mediated by volume of coarse woody debris in the streams). Consideration of ecomorphological traits is a good first step in functional analyses of species-rich tropical ecosystems because these traits can be quantified for a large number of poorly studied species, but the lack of data on other types of traits (especially reproductive life history) is a significant limitation that needs urgent attention (Vitule et al. 2017).

The predominance of negative threshold responses of relatively large magnitudes soon (i.e., < 5 years) after the period of greatest deforestation seems to support our hypothesis that rate of deforestation mediates the intensity of impacts.

However, positive threshold responses of species to time since the period of greatest land use change appear to coincide with a large deforestation event that occurred in Rondônia between 1991 and 1995. A subsequent large deforestation event occurred between 2001 and 2004 and may relate to negative functional group responses to time since < 40% of forest cover remaining, for both the watershed and riparian analyses. Previous studies on aquatic (e.g., Harding et al. 1998; Burcher et al. 2008) and terrestrial (e.g., Uezu & Metzger 2016) taxa demonstrate that contemporary communities may be more closely related to habitat conditions present decades ago. The time lags reported in those studies are much longer than the temporal responses we detected. Our results indicate that erosion of native taxonomic and functional assemblage structure started soon after deforestation and may have been mostly complete within approximately 16 years. Better resolution of deforestation chronology (e.g., more observations evenly spaced along the temporal gradient vs. clustered deforestation events in time) would help resolve temporal responses of populations and assemblages to deforestation and rate of change in land cover.

Contrary to our hypothesis, threshold responses were similar along gradients of watershed and riparian deforestation. However, we were unable to assess potential for intact riparian zones to buffer responses of stream fish assemblages to watershed deforestation. Deforestation dynamics in the Machado River basin, although representing a common pattern of deforestation for the Amazon in general, do not provide a fully factorial experimental design with which to test effects of riparian zone versus watershed deforestation. Specifically,

streams with deforested watersheds but intact riparian zones are uncommon, and streams with deforested riparian zones but intact watersheds are nonexistent. Deforestation up to stream margins is a common practice even though riparian zones are considered permanent protection areas (PPAs) by the Brazilian Forest Code and landowners are required by law to keep these areas intact. Although a highly contentious recent revision to the Forest Code (Federal Law 12.651) reduced the protected areas around water bodies, it includes some advances in law enforcement and implementation systems (Brancalion et al. 2016), obligating landowners to restore deforested riparian zones. Assuming successful implementation, restoration of deforested riparian zones could provide an interesting experimental context to further assess the importance of riparian zones for stream structure and diversity. However, testing for effects of riparian-zone restoration is different from assessing the effects of native riparian buffers, and consequences likely depend on connectivity and size of forested patches along the hydrologic network as source habitats for potential colonists (Brook et al. 2003). Dispersal capability of Neotropical stream fishes is largely unknown, and an open question is whether protected forest fragments or intact riparian zones allow for sufficient instream integrity to maintain sustainable populations at the landscape scale when much of the rest of the watershed has been deforested.

Reduced impact logging (RIL) (i.e., planned harvest focused on minimizing impacts on soil and remaining forest) has been touted as a land management strategy that could contribute to stream biodiversity conservation in watersheds targeted for extractive forest activities (Prudente et al. 2017).

However, some research indicates that selective logging can cause sudden changes in fish species richness and abundance, and that effects are detectable even 8 years after the logging event (Dias et al. 2010). Given that RIL plots may be harvested on a regular schedule (e.g., every 30 years), the long-term consequences for stream fish diversity under this management strategy are unknown. The strong negative threshold responses of sensitive taxa to low levels of deforestation that we found, combined with the findings of Dias et al. (2010), provide reasons to be skeptical, but perhaps RIL can be combined with PPAs strategically distributed (and effectively enforced) to yield the greatest conservation benefit for aquatic and terrestrial taxa. For example, preserved or restored riparian zones that contribute to instream habitat complexity and integrity may also provide benefits for movement of terrestrial vertebrates (Derugin et al. 2016). In fact, because small stream fishes that depend on inputs from riparian forest, have restricted distributions, and have low lifetime dispersal rates respond strongly to watershed and riparian deforestation, they may serve as good indicators of ensuing impacts of deforestation on other taxa.

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### Supporting Information 1.1

Summary of riparian ecotone and instream variables, mean  $\pm$  standard deviation, and explanation of how each variable was obtained.

Variables	Code	Mean $\pm$ standard deviation	Explanation
Riparian ecotone variables (For this calculation, both stream sides were computed):			
Trees in stream banks (%)	TRE	13.23 $\pm$ 18.33	- Percentage of the reach bank extension that was covered by riparian trees.
Grasses in stream banks (%)	GRA	35.03 $\pm$ 38.01	- Percentage of the reach bank extension that was covered by marginal grasses derived from surrounding pasture entering the water.
Submerged roots in stream banks (%)	FRO	3.33 $\pm$ 7.63	- Percentage of the reach bank extension that presented roots derived from riparian trees entering the water.
Riparian litter (%)	FLI	10.79 $\pm$ 12.66	- Percentage of the reach bank extension that presented fallen leaves derived from riparian trees
Instream variables:			
Sand (%)	SAN	53.58 $\pm$ 29.17	- Percentage of sand on the bottom of each reach
Consolidate substrate (%)	CSU	5.08 $\pm$ 9.94	Percentage of gravel and cobbles (particles with 2-256 mm in size) on the bottom of each stream reach
Litter packs on stream bed (%)	LIT	14.34 $\pm$ 18.24	- Percentage of fallen leaves on the streambed of each reach
Large wood debris on stream bed (%)	BAT	11.35 $\pm$ 10.78	- Percentage of fallen branches and tree trunks, representing large wood debris, on the streambed of each reach.
Depth (cm)	DEP	27.54 $\pm$ 13.68	- Average value of depth
Width (m)	WID	3.00 $\pm$ 1.41	- Average value of width
Water flow (m s <sup>-1</sup> )	CUR	0.38 $\pm$ 0.23	- Average value of water flow velocity
Dissolved oxygen (mg l <sup>-1</sup> )	DOX	6.63 $\pm$ 2.24	- Dissolved oxygen value
Conductivity ( $\mu$ S)	CON	18.39 $\pm$ 21.09	- Water conductivity value
Temperature ( $^{\circ}$ C)	WTE	24.06 $\pm$ 2.26	- Water temperature value

PCA of stream riparian ecotone and instream physicochemical habitat data, color coded according to quartiles in watershed and riparian zone deforestation.

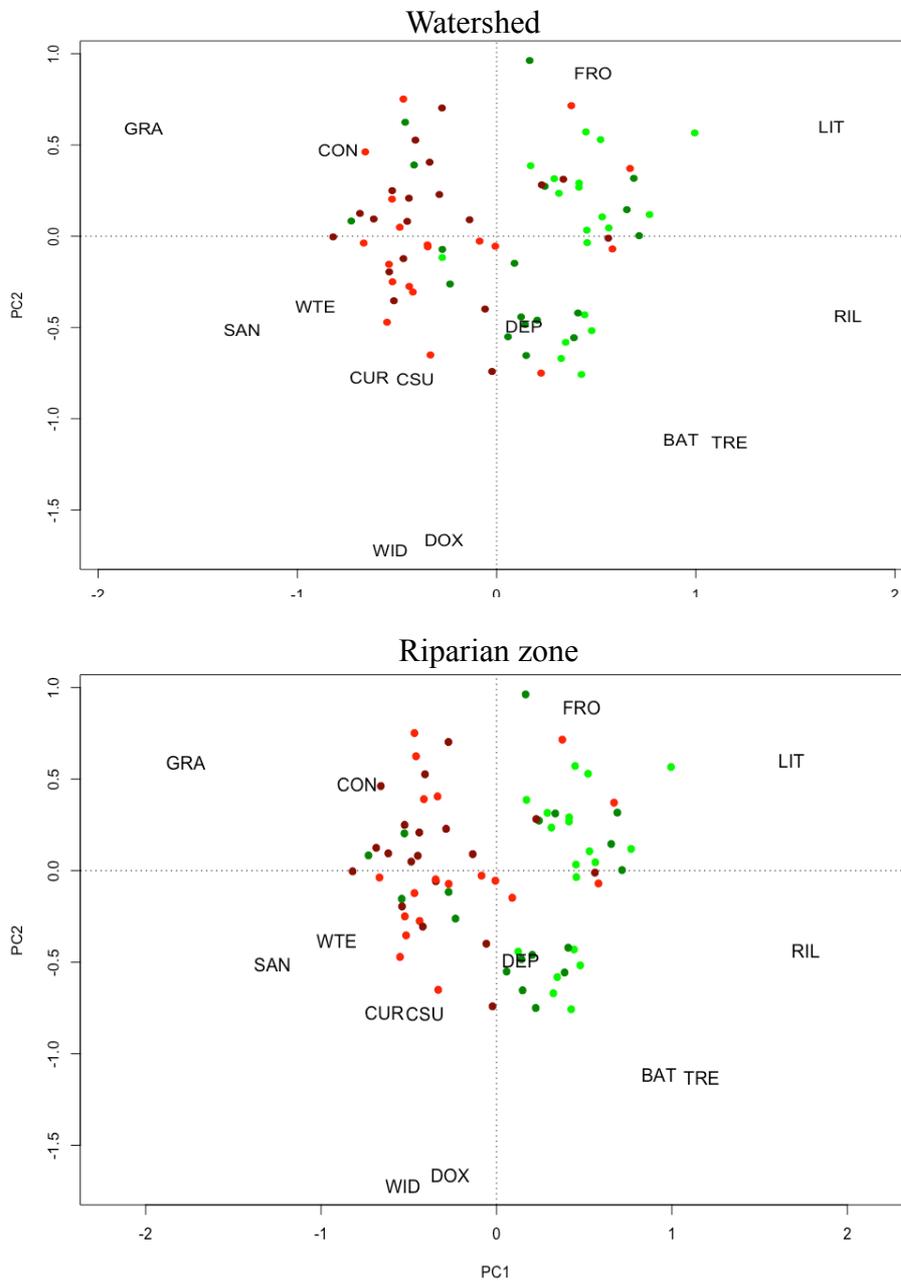


Figure Legend:

Color	DefW	DefR	Quantile
Light green	Def < 36.8%	Def < 35.0%	1 <sup>st</sup>
Dark green	36.8% ≤ Def < 75.3%	35.0% ≤ Def < 69.3%	2 <sup>nd</sup>
Light red	75.3% ≤ Def < 92.1%	69.3% ≤ Def < 93.0%	3 <sup>rd</sup>
Dark red	Def ≥ 92.1%	Def ≥ 93.0%	4 <sup>th</sup>

Axis scores for riparian ecotone and instream physicochemical habitat variables in the above PCA

	<b>PC1</b>	<b>PC2</b>
FRO	0.33816	0.5457
GRA	-1.23231	0.3624
TRE	0.81423	-0.6887
RIL	1.22693	-0.2651
SAN	-0.88815	-0.3113
LIT	1.17052	0.3686
CSU	-0.28365	-0.4772
BAT	0.64447	-0.6806
DEP	0.09617	-0.3004
WID	-0.37051	-1.0496
CUR	-0.44401	-0.4725
DOX	-0.18336	-1.0166
CON	-0.5531	0.2894
WTE	-0.63261	-0.233

## Supporting Information 2.1

Deforestation gradient values for each sampled site at watershed and 100 m width riparian zone scales. Def: Deforestation; Change: Land-use change since 1984; TMax: Time since the greatest land use change; T40: Time since <40% forest cover.

ID	Watershed				Riparian zone			
	Def (%)	Change (%)	TMax (years)	T40 (years)	Def (%)	Change (%)	TMax (years)	T40 (years)
S01	92.5	81.0	20	20	93.2	78.3	4	16
S02	84.9	79.3	20	12	89.8	75.9	20	16
S03	93.1	91.1	20	16	94.8	94.0	20	16
S04	94.6	41.5	16	24	90.7	46.0	16	16
S05	89.5	87.7	24	>27	96.8	94.8	16	16
S06	91.4	65.9	16	16	88.9	60.1	16	16
S07	83.5	49.9	16	16	84.1	62.7	16	16
S08	91.5	89.9	16	16	93.3	86.2	20	20
S09	90.5	83.6	20	16	81.3	80.0	20	16
S10	92.9	79.1	20	16	89.0	73.2	20	16
S11	75.3	47.3	27	16	67.5	29.5	27	16
S12	90.3	72.5	16	16	88.2	78.2	16	16
S13	82.1	70.5	20	20	69.3	62.3	20	16
S14	10.3	12.7	16	0	6.5	8.5	20	0
S15	57.3	61.7	16	4	42.0	45.2	4	0
S16	15.2	16.0	8	0	10.5	10.9	16	0
S17	70.6	72.5	8	12	46.9	52.2	8	0
S18	75.3	79.1	8	12	71.3	75.9	8	8
S19	81.3	83.2	8	12	72.1	77.2	16	8
S20	64.2	65.5	8	12	59.1	62.3	8	4
S21	92.2	93.0	12	16	64.9	68.6	16	12
S22	13.7	12.4	8	0	19.5	19.8	16	0
S23	0.5	0.0	0	0	0.7	0.0	0	0
S24	0.0	0.0	0	0	0.0	0.0	0	0
S25	0.0	0.0	0	0	0.0	0.0	0	0
S26	0.0	0.0	0	0	0.0	0.0	0	0
S27	0.0	0.0	0	0	0.0	0.0	0	0
S28	0.0	0.0	0	0	0.0	0.0	0	0
S29	0.0	0.0	0	0	0.0	0.0	0	0
S30	0.0	0.0	0	0	0.0	0.0	0	0
S31	0.1	0.0	0	0	0.0	0.0	0	0
S32	64.0	64.4	8	8	40.8	47.7	8	0
S33	2.9	2.2	4	0	0.0	0.0	0	0
S34	61.7	63.1	8	8	42.6	47.0	8	0
S35	47.8	50.8	8	0	29.3	30.7	16	0

<b>S36</b>	45.7	47.9	12	0	43.1	48.4	16	0
<b>S37</b>	75.9	77.1	8	8	76.0	76.2	8	8
<b>S38</b>	0.5	2.6	4	0	0.0	0.0	0	0
<b>S39</b>	0.8	4.3	8	0	0.0	0.0	0	0
<b>S40</b>	74.7	53.8	24	16	71.5	53.0	16	16
<b>S41</b>	71.3	59.6	24	16	75.1	52.4	24	20
<b>S42</b>	94.6	64.4	16	16	98.0	65.2	16	16
<b>S43</b>	4.1	1.7	4	0	2.1	2.0	4	0
<b>S44</b>	8.5	5.8	8	0	0.0	0.0	0	0
<b>S45</b>	0.0	1.7	8	0	0.0	0.0	0	0
<b>S46</b>	54.6	58.1	24	0	49.5	50.9	20	0
<b>S47</b>	86.7	79.3	16	16	92.8	76.5	16	16
<b>S48</b>	92.0	66.0	27	20	100.0	61.7	27	27
<b>S49</b>	62.6	53.5	4	4	66.0	60.6	4	4
<b>S50</b>	96.6	83.9	20	20	99.0	82.0	27	20
<b>S51</b>	73.7	67.7	16	8	74.7	74.1	24	12
<b>S52</b>	91.1	81.0	20	20	96.7	85.7	20	20
<b>S53</b>	97.4	84.9	20	20	97.1	86.2	20	20
<b>S54</b>	97.7	90.1	20	20	98.5	91.1	20	20
<b>S55</b>	95.3	87.3	20	16	95.1	87.2	20	20
<b>S56</b>	97.8	90.9	20	20	98.1	98.1	16	20
<b>S57</b>	92.4	41.4	16	27	95.0	35.4	27	>27
<b>S58</b>	71.7	52.5	16	12	65.7	54.6	16	12
<b>S59</b>	84.9	84.1	16	12	77.0	77.7	20	12
<b>S60</b>	95.4	82.5	20	16	97.7	83.6	20	20
<b>S61</b>	40.8	38.2	24	0	45.7	41.3	24	0
<b>S62</b>	39.5	38.7	16	0	86.9	86.9	8	8
<b>S63</b>	34.0	35.7	4	0	43.6	49.5	16	0
<b>S64</b>	79.8	79.2	8	4	60.7	59.1	27	4
<b>S65</b>	100.0	97.5	27	27	100.0	96.3	27	20
<b>S66</b>	99.9	87.9	20	20	99.8	98.5	16	16
<b>S67</b>	62.3	48.3	16	27	44.4	38.2	16	0
<b>S68</b>	92.4	81.9	16	20	94.5	74.1	16	16
<b>S69</b>	68.0	27.9	16	12	46.0	7.1	20	0
<b>S70</b>	99.1	38.2	16	>27	100.0	35.3	16	>27
<b>S71</b>	98.7	45.7	16	16	98.5	19.9	16	>27
<b>S72</b>	81.7	60.9	16	16	82.5	46.7	16	16
<b>S73</b>	64.5	44.8	16	12	54.1	44.1	27	0
<b>S74</b>	87.7	88.6	20	16	58.1	60.5	20	12
<b>S75</b>	93.9	79.4	16	16	70.1	54.0	16	12

### Supporting Information 3.1

Fish species included in this study, functional group assignment, frequency of occurrence, and maximum site and total abundances. Classification follows Reis et al. (2003); except Serrasalminae follows Calcagnotto et al. (2005). **FG**: Functional Group; **Max**: Maximum abundance at a single location.

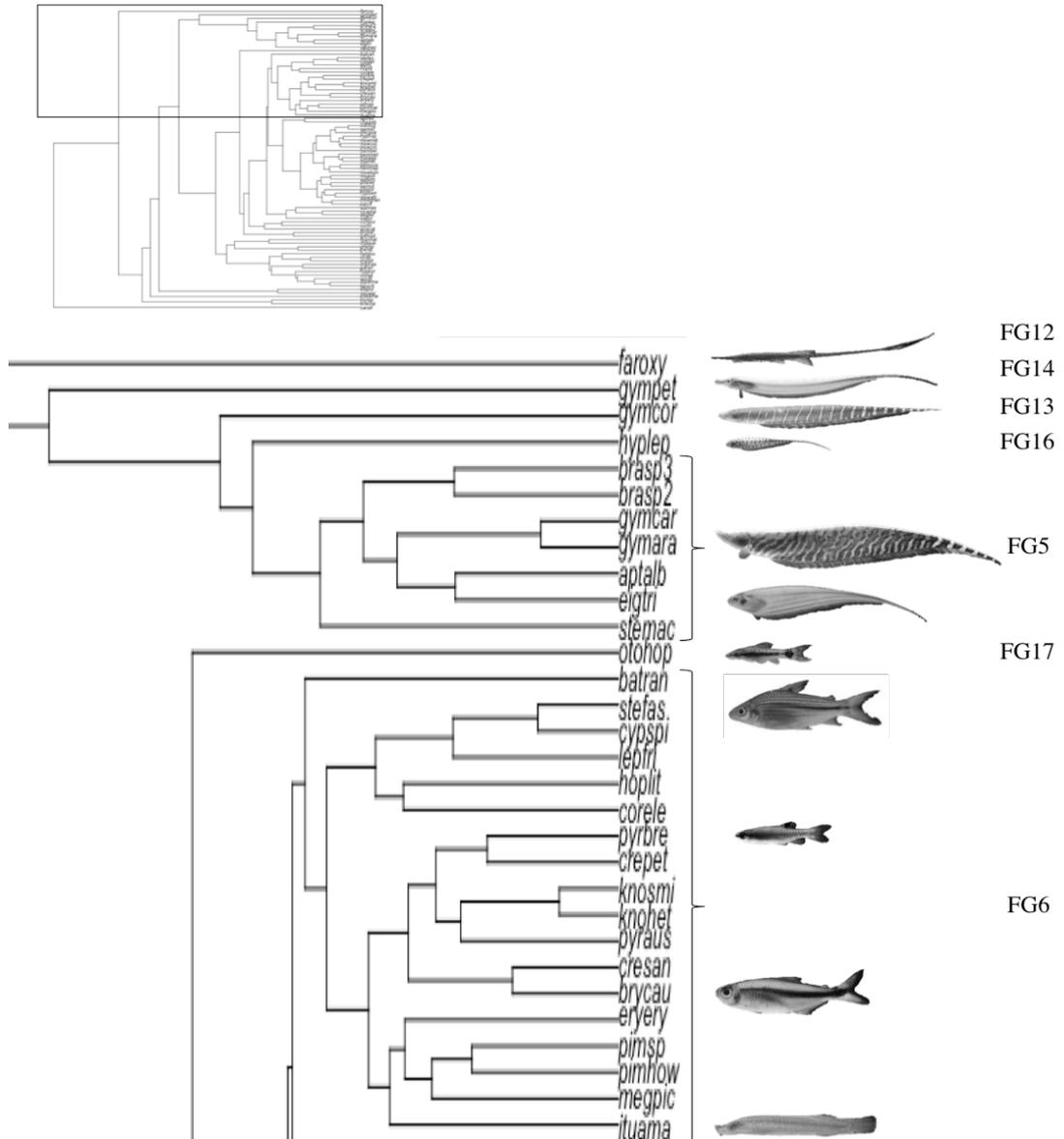
TAXON	Code	FG	Occurrence	Abundance	
				Total	Max
<b>CHARACIFORMES</b>					
<b>Curimatidae</b>					
<i>Cyphocharax spiluroopsis</i> (Eigenmann & Eigenmann, 1889)	cypspi	6	14	40	7
<i>Steindachnerina fasciata</i> (Vari & Géry, 1985)	stefas	6	14	57	8
<b>Anostomidae</b>					
<i>Leporinus friderici</i> (Block, 1794)	lepfri	6	18	36	11
<b>Crenuchidae</b>					
<i>Characidium</i> aff. <i>zebra</i> Eigenmann, 1909	chazeb	4	54	762	94
<i>Elachocharax pulcher</i> Myers, 1927	elapul	11	9	79	29
<i>Microcharacidium</i> aff. <i>weitzmani</i> (Buckup, 1993)	micwei	11	9	38	20
<b>Gasteropelecidae</b>					
<i>Carnegiella strigata</i> (Günther, 1864)	carstr	9	7	40	13
<b>Characidae</b>					
<i>Astyanax</i> cf. <i>bimaculatus</i> (Linnaeus, 1758)	astbim	4	19	108	29
<i>Astyanax</i> cf. <i>maximus</i> (Steindachner, 1876)	astmax	2	7	18	8
<i>Brachyhalcinus copei</i> (Steindachner, 1822)	bracop	7	34	147	18
<i>Bryconella pallidifrons</i> (Fowler, 1946)	brypall	8	8	695	339
<i>Bryconops caudomaculatus</i> (Günther, 1864)	brycau	6	50	912	243
<i>Creagrutus petilus</i> Vari & Harold, 2001	creapet	6	48	1021	208
<i>Hemigrammus</i> aff. <i>ocellifer</i> (Steindachner, 1882)	hemoce	4	11	62	25
<i>Hemigrammus bellotti</i> (Steindachner, 1882)	hembel	4	5	152	84
<i>Hemigrammus melanochrous</i> Fowler, 1913	hemmel	4	11	1418	364
<i>Hemigrammus neptunus</i> Zarske & Géry, 2002	hemnep	4	4	60	54
<i>Hyphessobrycon</i> aff. <i>heterorhabdus</i> (Ulrey, 1894)	hyphet	4	11	144	42
<i>Hyphessobrycon agulha</i> Fowler, 1913	hypagu	4	15	1131	402
<i>Hyphessobrycon bentosi</i> Durbin, 1908	hypben	4	8	178	51
<i>Hyphessobrycon copelandi</i> Durbin, 1908	hycop	4	7	151	98
<i>Jupiaba citrina</i> Zanata & Ohara, 2009	jupcit	4	19	273	92
<i>Jupiaba poranga</i> Zanata, 1997	juppor	4	4	9	3
<i>Knodus</i> cf. <i>smithi</i> Fowler, 1913	knosmi	6	35	827	150
<i>Knodus heteresthes</i> Eigenmann, 1908	knohet	6	12	736	582
<i>Microchemobrycon guaporensis</i> Eigenmann, 1915	micgua	4	14	166	74
<i>Moenkhausia</i> cf. <i>bonita</i> Benine, Castro & Sabino, 2004	moebon	4	7	339	201
<i>Moenkhausia collettii</i> (Steindachner, 1882)	moecol	4	42	1924	322
<i>Moenkhausia cotinho</i> Eigenmann, 1908	moecot	4	17	259	93

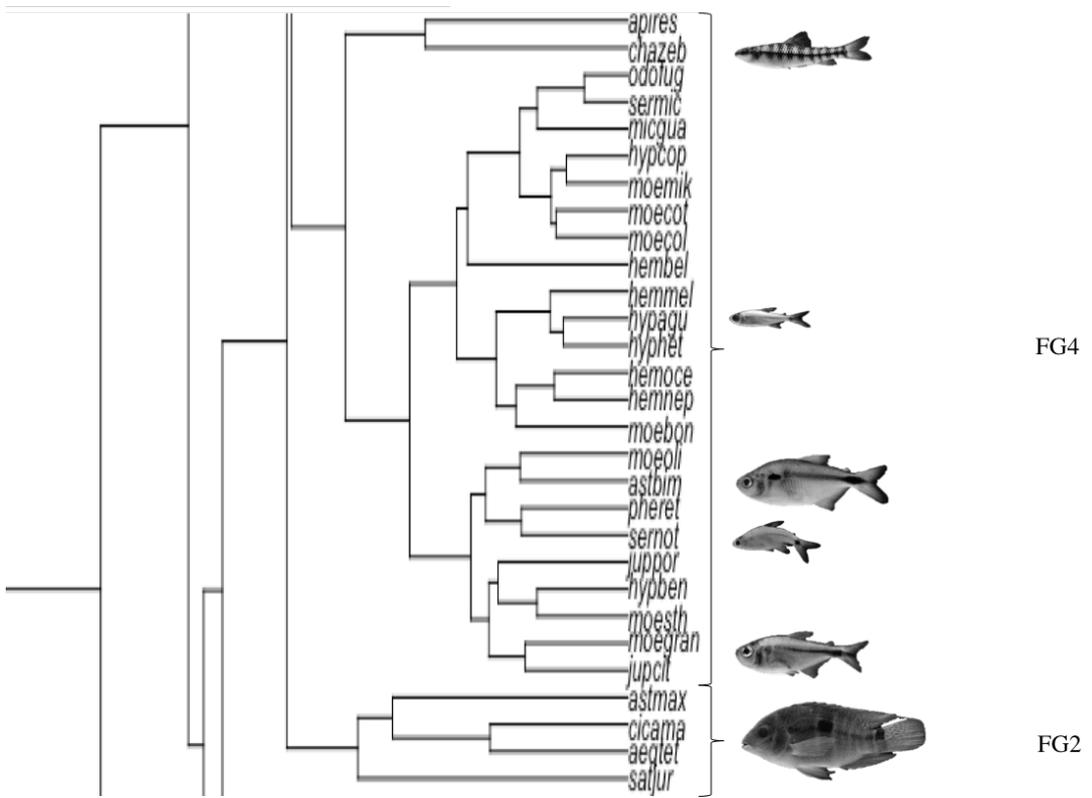
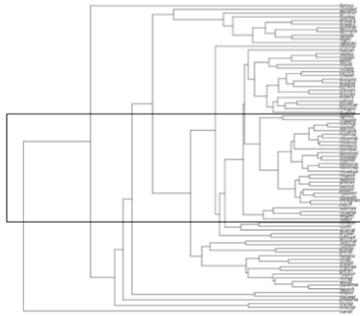
<i>Moenkhausia grandisquamis</i> Müller & Troschel, 1845	moegrn	4	7	11	3
<i>Moenkhausia mikia</i> Marinho & Langeani, 2010	moemik	4	11	105	44
<i>Moenkhausia oligolepis</i> (Günther, 1864)	moeoli	4	49	330	73
<i>Moenkhausia sthenostoma</i> Petrolli & Bertaco, 2015	moesth	4	7	41	23
<i>Odontostilbe fugitiva</i> Cope, 1870	odofug	4	6	307	136
<i>Phenacogaster retropinnus</i> Lucena & Malabarba, 2010	pheret	4	42	386	41
<i>Serrapinnus microdon</i> (Eigenmann, 1915)	sermic	4	30	1901	231
<i>Serrapinnus</i> aff. <i>notomelas</i> (Eigenmann, 1915)	sernot	4	34	3642	1742
<i>Tyttocharax madeirae</i> Fowler, 1913	tytmad	8	4	32	16
<b>Serrasalminidae</b>					
<i>Myleus</i> sp.	mylsp	7	5	12	5
<b>Erythrinidae</b>					
<i>Erythrinus erythrinus</i> (Bloch & Schneider, 1801)	eryery	6	8	11	3
<i>Hoplias malabaricus</i> (Bloch, 1794)	hopmal	15	36	88	15
<b>Lebiasinidae</b>					
<i>Pyrhulina</i> cf. <i>australis</i> Eigenmann & Kennedy, 1903	pyraus	6	24	193	26
<i>Pyrhulina</i> cf. <i>brevis</i> Steindachner, 1876	pyrbre	6	9	65	33
<b>SILURIFORMES</b>					
<b>Cetopsidae</b>					
<i>Helogenes gouldingi</i> Vari & Ortega, 1986	helgou	3	9	22	6
<b>Aspredinidae</b>					
<i>Pseudobunocephalus amazonicus</i> (Mees, 1989)	pseama	19	8	37	10
<b>Trichomycteridae</b>					
<i>Ituglanis amazonicus</i> (Steindachner, 1882)	ituama	6	20	108	24
<i>Paracanthopoma</i> sp. 1	parsp	18	6	19	12
<b>Callichthyidae</b>					
<i>Corydoras acutus</i> Cope, 1872	coracu	10	4	5	2
<i>Corydoras elegans</i> Steindachner, 1876	corele	6	5	7	2
<i>Corydoras trilineatus</i> Cope, 1872	cortri	10	22	82	10
<i>Hoplosternum littorale</i> (Hancock, 1828)	hoplit	6	5	7	3
<i>Megalechis picta</i> (Müller & Troschel, 1849)	megpic	6	6	49	32
<b>Loricariidae</b>					
<i>Ancistrus lithurgicus</i> Eigenmann, 1912	anclit	3	26	290	73
<i>Farlowella</i> cf. <i>oxyrryncha</i> (Kner, 1853)	faroxy	12	22	120	15
<i>Hypostomus pyrineusi</i> (Miranda Ribeiro, 1920)	hyppyr	3	15	28	3
<i>Lasiancistrus schomburgkii</i> (Günther, 1864)	lassch	3	18	61	18
<i>Otocinclus hoppei</i> Miranda Ribeiro, 1939	otohop	17	10	119	32
<i>Parotocinclus</i> aff. <i>aripuanensis</i> Garavello, 1988	parari	3	6	24	14
<i>Rineloricaria heteroptera</i> Isbrücker & Nijssen, 1976	rinhet	3	40	164	18
<i>Rineloricaria</i> sp.	rinsp	3	4	6	3
<i>Squaliforma emarginata</i> (Valenciennes, 1840)	squema	3	7	22	10
<b>Pseudopimelodidae</b>					

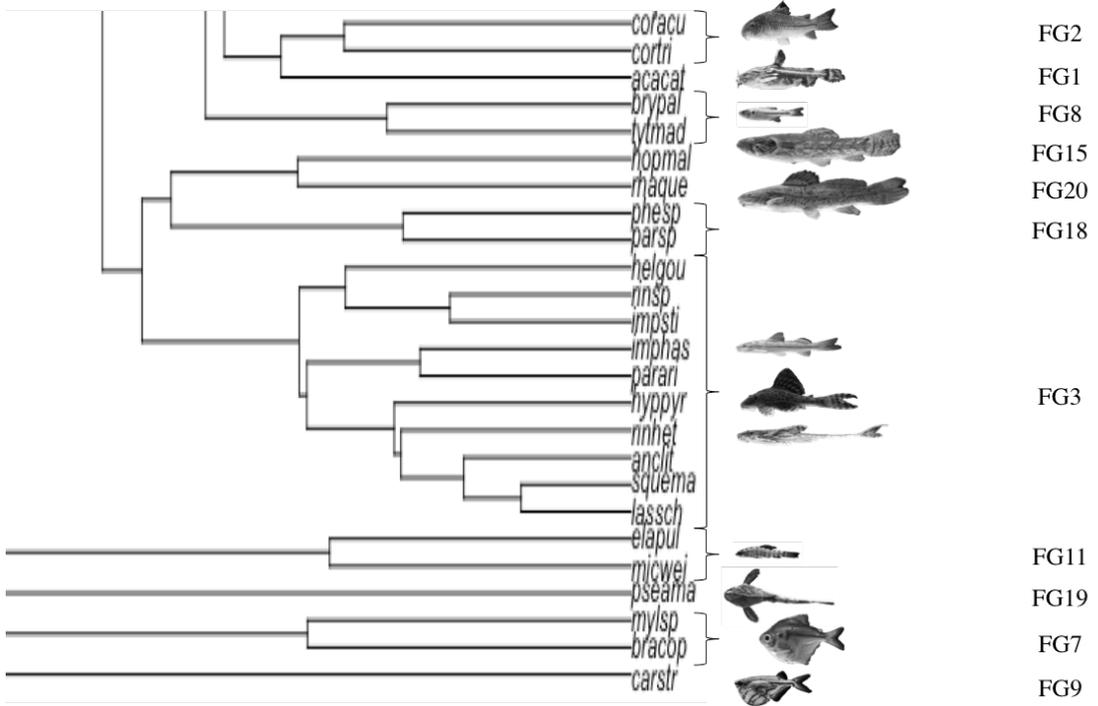
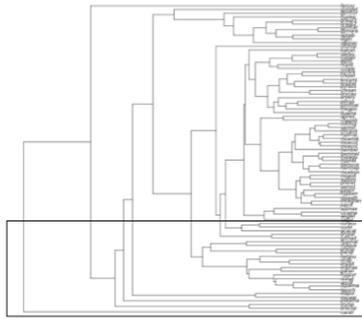
<i>Batrochoglanis cf. raninus</i> (Valenciennes, 1840)	batran	6	5	16	5
<b>Heptapteridae</b>					
<i>Imparfinis cf. hasemani</i> Steindachner, 1917	imphas	3	19	124	27
<i>Imparfinis stictonotus</i> (Fowler, 1940)	imsti	3	13	49	15
<i>Phenacorhamdia</i> sp.	phesp	18	8	70	22
<i>Pimelodella cf. howesi</i> Fowler, 1940	pimhow	6	16	55	17
<i>Pimelodella</i> sp.	pimsp	6	8	11	2
<i>Rhamdia quelen</i> (Quoy & Gaimard, 1824)	rhaque	20	4	6	2
<b>Doradidae</b>					
<i>Acanthodoras cataphractus</i> (Linnaeus, 1758)	acacat	1	4	19	16
<b>GYMNOTIFORMES</b>					
<b>Gymnotidae</b>					
<i>Gymnotus aff. arapaima</i> Albert & Crampton, 2001	gymara	5	9	26	9
<i>Gymnotus carapo</i> Linnaeus, 1758	gymcar	5	10	36	17
<i>Gymnotus coropinae</i> Hoederman, 1962	gymcor	13	15	81	19
<b>Sternopygidae</b>					
<i>Eigenmannia trilineata</i> López & Castello, 1966	eigtri	5	19	196	64
<i>Sternopygus macrurus</i> (Bloch & Schneider, 1801)	stemac	5	24	97	31
<b>Rhamphichthyidae</b>					
<i>Gymnorhamphichthys petiti</i> Géry & Vu-Tân-Tuê, 1964	gympet	14	31	287	65
<b>Hypopomidae</b>					
<i>Brachyhypopomus</i> sp. 2	brasp2	5	8	15	4
<i>Brachyhypopomus</i> sp. 3	brasp3	5	5	26	14
<i>Hypopygus lepturus</i> Hoedeman, 1962	hyplep	16	17	128	28
<b>Apteronotidae</b>					
<i>Apteronotus albifrons</i> (Linnaeus, 1766)	aptalb	5	4	6	2
<b>PERCIFORMES</b>					
<b>Cichlidae</b>					
<i>Aequidens tetramerus</i> (Heckel, 1840)	aeqtet	2	39	199	31
<i>Apistogramma cf. resticulusa</i> Kullander, 1980	apires	4	23	563	136
<i>Cichlasoma amazonarum</i> Kullander, 1983	cicama	2	9	46	18
<i>Crenicichla santosi</i> Ploeg, 1991	cresan	6	40	163	61
<i>Satanoperca jurupari</i> (Heckel, 1840)	satjur	2	13	60	26

### Supporting Information 4.1

Result from cluster analysis indicating species assignment into functional groups. Images of select taxa provided as representations of traits most associated with each functional group. Taxa codes and species names are provided in the Supporting Information (Supporting Information 3.1).







### Supporting Information 5.1

Environmental change points detected for each species and functional group along gradients of watershed and riparian zone deforestation. Only species with  $\alpha < 0.05$ , purity  $\geq 0.95$  and reliability (rel05 and rel01)  $\geq 0.95$  are shown. Def: Deforestation; Change: Land use change since 1984; TMax: Time since the greatest land use change; T40: Time since <40% of forest cover. Taxa codes and species names are provided in the Supporting Information (Supporting Information 3.1).

Taxa	+/-	Watershed				Riparian zone			
		Def (%)	Change (%)	TMax (years)	T40 (years)	Def (%)	Change (%)	TMax (years)	T40 (years)
apires	z-	36.8	12.6	14	2	41.4	4.5	12	12
brasp2	z-	3.5	0.0	4	0	1.4	0.0	0	0
brypal	z-	0.0	0.9	4	0	0.0	0.0	0	0
elapul	z-	0.3	1.7	0	2	0.0	0.0	0	-
gymcor	z-	3.5	0.0	0	12	0.0	0.0	16	12
gympet	z-	36.8	22.0	16	0	41.4	19.9	-	4
hemmel	z-	-	-	-	-	48.2	-	-	12
hypagu	z-	0.0	-	0	16	42.3	-	-	-
hyphet	z-	62.5	-	-	10	51.8	-	-	10
imphas	z-	-	66.0	-	-	-	50.2	-	-
ituama	z-	0.0	-	16	2	59.9	-	-	0
micwei	z-	-	-	-	-	59.9	-	-	-
pimhow	z-	-	-	-	-	0.0	-	8	-
pseama	z-	0.0	0.0	0	6	42.3	-	6	12
pyraus	z-	9.4	9.1	8	10	4.3	9.7	2	0
astbim	z+	94.9	-	16	20	93.0	95.6	-	-
brycau	z+	-	12.6	-	-	-	-	-	-
chazeb	z+	-	-	-	-	-	-	-	12
hyppyr	z+	75.3	-	-	12	-	-	-	16
jupcit	z+	98.3	-	24	16	98.3	-	-	16
knosmi	z+	75.3	-	-	-	-	-	-	-
rinhet	z+	81.9	40.1	16	-	66.8	7.8	-	0
sermic	z+	72.7	-	16	12	65.9	-	16	12
sernot	z+	62.0	-	16	8	95.1	-	16	8
stefas	z+	81.5	-	-	16	74.9	-	16	20
FG08	z-	0.3	0.0	0	6	0.0	0.0	8	6
FG11	z-	0.0	0.0	2	12	44.0	19.9	16	10
FG13	z-	1.8	1.7	4	8	4.3	4.5	8	0
FG14	z-	24.6	22.0	16	0	15.0	19.9	2	16
FG19	z-	0.0	0.9	4	6	44.0	0.0	2	6

## **Chapter 2**

**Using deforestation chronology to understand changes in stream  
habitat structure and fish diversity in the Amazon**

## **Abstract**

Deforestation is a primary driver of habitat loss and fragmentation affecting terrestrial and aquatic ecosystems. The primary effects by which deforestation influences stream features (i.e., habitat simplification or homogenization) do not occur immediately after logging and populations of many stream species may also not respond directly to changes in instream habitat. Adopting a chronosequence approach may allow for detecting temporal responses to deforestation or extended periods of time. The chronosequence approach has frequently been used to study biodiversity structure and functioning (mostly of plant and soil communities) during succession or after restoration. In this study, we used a deforestation chronosequence to investigate how time since deforestation and the land use intensification affected instream habitat structure and species richness of sensitive and tolerant fish species, defined by TITAN analysis, from 75 streams in the western Brazilian Amazon. The degree of deforestation at watershed and riparian zone scales derived from remote sensing techniques for multiple periods (1984 to 2011). Differently than expected, alteration of the instream habitat structure and sensitive fish richness and abundance lost occurred synchronically with the habitat structure loss (litter packs, riparian trees, and litter). By contrast, we observed a time-lag response for habitat homogenization indicators (grass, sand, and conductivity), and tolerant fish richness and abundance increase followed this time-lag. To achieve indirect improvements in the aquatic environment, due to forest restoration, is important to halt or reverse deforestation before reaching the critical threshold affecting instream habitat structure and associated biodiversity.

Restoration of instream structure and functioning through forest regeneration will likely take substantially more time once the threshold has passed.

## Introduction

High diversity tropical forests are often deforested for conversion into agriculture (Aide et al. 2000). This deforestation process is so drastic that the adventitious ecosystem associated (e.g., streams, rivers, mangroves) are dramatically altered. However, deforestation often carries new stressors, such as pollution, species invasion and diseases (Rahel 2002; Munns 2006; Niyogi et al. 2007; Couillard et al. 2008). Taking streams as an example, the multiple stressors affecting them has led to sensitive species loss (Palmer et al. 2010), diversity loss and biotic homogenization (Rahel 2002; Olden 2006). The primary effects by which deforestation influences stream features, i.e., habitat homogenization (Allan 2004), do not occur immediately after logging and populations of many stream species may also not respond immediately to changes in instream habitat.

The chronology of disturbance is an important determinant of current stream and watershed diversity. For example, the forest to agriculture conversion may result in long-term modifications in aquatic assemblage structure (Harding et al. 1998), and the instream habitat alteration were still evident regardless the reforestation of riparian zones, lowering the richness and abundance of some assemblage components (*i.e.*, benthic species) while favoring others (*i.e.*, nektonic species) species (Harding et al. 1998; Iwata et al. 2003; Burcher et al. 2008). In this sense, adopting a chronosequence approach may allow for detecting temporal responses to deforestation without monitoring for extended periods of time. The chronosequence approach has frequently been used to study biodiversity structure and functioning, mostly of plant and soil communities, during succession or after

restoration (Aide et al. 2000; Johnson & Miyanishi 2008; Alfaro et al. 2017). It is also essential to consider landscape dynamics and historical changes to misleading changes in diversity in highly diverse forests (Metzger et al. 2009; Tabarelli et al. 2010; Uezu & Metzger 2016). Equally important is the identification of change points that induce regime shifts or irreversible assemblage homogenization (Scheffer et al. 2001; Andersen et al. 2008).

Despite the use of landscape variables in neotropical stream ecology studies is becoming more common in the last decades (see Nessimian et al. 2008; Casatti et al. 2015; Leitão et al. 2017; Roa-Fuentes & Casatti 2017; Zeni et al. 2017), the use of landscape dynamics variables still is extremely rare (but see Leal et al. 2016; Roa-Fuentes 2016; Brejão et al. 2018; and this work). However, this historical information is essential to understand the influence of land use changes in aquatic assemblages, because nonlinear responses to the surrounding impacts are expected to occur (Dodds et al. 2010; Smucker et al. 2013; Rodrigues et al. 2016). The use of deforestation history along with current landscape structure enhances the power of analysis to evaluate ecological effects in different regions, which appear to be similar in the present time, although their path until the current state has not been the same (Ferraz et al. 2009).

The Rondônia State, especially the Machado river basin, presents high rates of recent deforestation (Ferraz et al. 2005). As a consequence of the colonization process, which began with the Federal Highway BR-364 construction at the end of the 1960's and strengthened in the early 1980's, with the expansion of adjacent roads (Numata et al. 2009). Because a range from entirely forested to

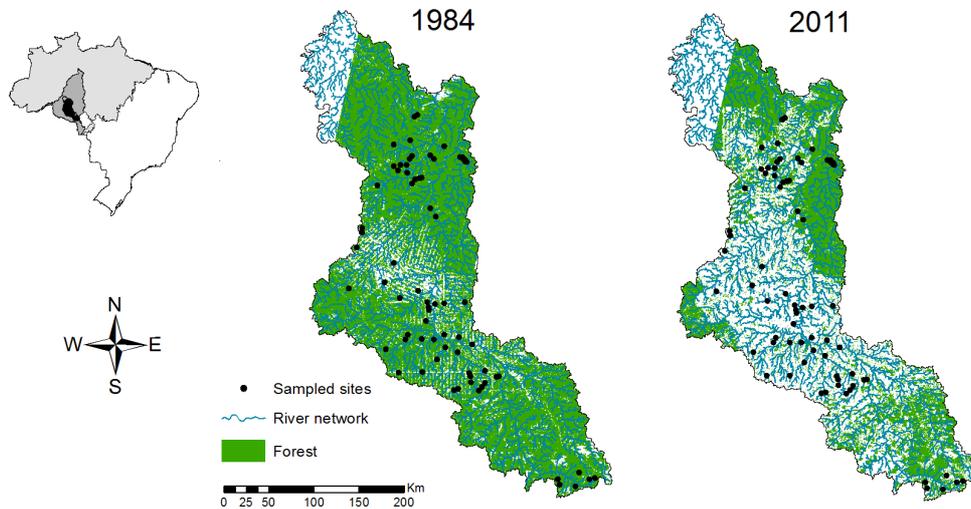
wholly deforested basins is available, with a good number of replicates, the region represents a unique model for studies interested in exploring chronology and intensity of deforestation.

Our aim was to investigate how time since deforestation and land use intensification affected instream habitat structure and sensitive and tolerant fish richness and abundance by using a deforestation chronosequence. Considering that nonlinear responses are expected between the proportion of watershed affected by deforestation and habitat loss (Smucker et al. 2013; Rodrigues et al. 2016), and that stream biodiversity responses to deforestation are also nonlinear (Brejão et al. 2018, Chapter 1), we expect that the loss of sensitive species diversity will show a time-lag response after the beginning of habitat structure indicators loss. On the other hand, tolerant species diversity will be benefited by deforestation since its beginning.

## **Methods**

### *Study Area*

We surveyed 75 first- to third-order streams in the Machado River basin, Brazil (Fig. 1.2). The 1,200 km Machado River is a tributary of the Madeira River, and its 75,400 km<sup>2</sup> catchment area drains the most populated region of Rondônia, Brazil (Fernandes & Guimarães 2002). This region has many terra firme streams, which are intermittent during most of the dry season (Fernandes & Guimarães 2002). The sampled streams were shallow with low-velocity flow and warm temperatures. The streambeds were predominantly sand, litter pack, and large woody debris, and stream banks usually provided submerged microhabitat structure derived from the riparian environment such as tree roots and grasses (Supporting Information). The climate is tropical humid: temperatures range from 19 to 33° C, and annual rainfall is 2,500 mm (Krusche et al. 2005). The land cover in this region includes primary forest (open humid tropical forest), secondary forest, and pasture (Ferraz et al. 2009).



**Figure 1.2.** Sampled sites (75 black dots) and forest cover in the Machado River basin, Brazil in 1984 and 2011. The inset map of Brazil depicts the relative location of the study area (black) within the Madeira River basin (dark gray), inside the Amazon biome (light gray).

### *Predictor variables*

Land-cover data were quantified using Landsat 5TM images (30 x 30 m resolution). Land use was classified as mature forest, secondary or degraded forest, and nonforest according to the supervised classification method (Jensen 2007) in ERDAS (version 9.2) for multiple dates (1984, 1987, 1991, 1995, 1999, 2003, 2007, and 2011). Land use change was analyzed from 1984 to 2011 at 4-year intervals and calculated using the Land-use Change Analysis Tool - LUCAT (Ferraz et al. 2011, 2012), which calculates the area and proportion of the area occupied by each type of land use present in a unit of analysis.

The time since the greatest land-use change (TMax), quantified as the number of years since the time interval with the greatest decrease in forest cover since 1984 (Brejão et al. 2018). Land Use Intensification (LUI) represents the average time since the deforestation and can be considered as an accumulated effect metric over the time (Ferraz et al. 2009). Both metrics were used to verify the temporal effects of deforestation on local environmental variables (Table 1.2) and fish species richness and abundance at for watershed and 100-m-wide riparian zones for all 75 streams (Supporting Information S1.2).

#### *Response variables*

Fish was collected in 75 streams reaches (80 m) were sampled once during the dry season (August–October 2011 and June–July 2012). Sampling methods were standardized to allow for comparisons of species abundances across sites. Instream physicochemical and riparian ecotone (hereafter instream) habitat variables were quantified using standard methods (Table 1.2) (for details see the Table 1 on Pérez-Mayorga et al. 2017). Fishes were sampled from all available microhabitats by two collectors using a seine (1.5 x 2 m, 2-mm mesh) and dip nets (0.5 x 0.8 m, 2-mm mesh) for 1 hour. This method accounted for 90% of expected richness for this set of streams (Casatti et al. 2013).

**Table 1.2.** Summary of riparian ecotone and instream variables, mean  $\pm$  standard deviation, and explanation of how each variable was obtained.

<b>Variables</b>	<b>Code</b>	<b>Mean <math>\pm</math> standard deviation</b>	<b>Explanation</b>
Riparian ecotone variables (For this calculation, both stream sides were computed):			
Trees in stream banks (%)	TRE	13.23 $\pm$ 18.33	- Percentage of the reach bank extension that was covered by riparian trees.
Grasses in stream banks (%)	GRA	35.03 $\pm$ 38.01	- Percentage of the reach bank extension that was covered by marginal grasses derived from surrounding pasture entering the water.
Submerged roots in stream banks (%)	FRO	3.33 $\pm$ 7.63	- Percentage of the reach bank extension that presented roots derived from riparian trees entering the water.
Riparian litter (%)	RLI	10.79 $\pm$ 12.66	- Percentage of the reach bank extension that presented fallen leaves derived from riparian trees
Instream variables:			
Sand (%)	SAN	53.58 $\pm$ 29.17	- Percentage of sand on the bottom of each reach
Consolidate substrate (%)	CSU	5.08 $\pm$ 9.94	Percentage of gravel and cobbles (particles with 2-256 mm in size) on the bottom of each stream reach
Litter packs on stream bed (%)	LIT	14.34 $\pm$ 18.24	- Percentage of fallen leaves on the streambed of each reach
Large wood debris on stream bed (%)	BAT	11.35 $\pm$ 10.78	- Percentage of fallen branches and tree trunks, representing large wood debris, on the streambed of each reach.
Depth (cm)	DEP	27.54 $\pm$ 13.68	- Average value of depth
Width (m)	WID	3.00 $\pm$ 1.41	- Average value of width
Water flow (m s <sup>-1</sup> )	CUR	0.38 $\pm$ 0.23	- Average value of water flow velocity
Dissolved oxygen (mg l <sup>-1</sup> )	DOX	6.63 $\pm$ 2.24	- Average value of Dissolved oxygen
Conductivity ( $\mu$ S)	CON	18.39 $\pm$ 21.09	- Average value of Water conductivity
Temperature ( $^{\circ}$ C)	WTE	24.06 $\pm$ 2.26	- Average value of Water temperature

Fishes were collected under “Instituto Chico Mendes de Conservação da Biodiversidade” permits 4355-1/2012. All individuals were identified with assistance from taxonomic specialists, and voucher specimens are deposited in the “Coleção de Peixes do Departamento de Zoologia e Botânica (DZSJRP)” at the “Universidade Estadual Paulista Júlio de Mesquita Filho”, São José do Rio Preto, São Paulo State, Brazil.

To define which species were sensitive or tolerant to deforestation, we used threshold indicator taxa analysis (TITAN) (Baker & King 2010), and the defined cut-off levels for TITAN were  $\alpha < 0.05$ , purity  $\geq 0.75$ , and reliability (rel05 and rel01)  $\geq 0.75$ .

#### *Data analysis*

Generalized linear models (GLMs) were employed to assess the effects of the predictor variables (TMax and LUI) at watershed and riparian zone scales on fish assemblage richness and Abundance (log transformed) and local habitat structure. These statistical models assume errors from the exponential family in which the predicted values are determined by discrete and continuous predictor variables and by the link function (e.g., logistic regression, Poisson regression) (Bolker et al. 2009). All analysis was conducted in R software (R Development Core Team 2014).

## Results

In total, we collected 22,851 individuals belonging to 138 species, 30 families and six orders in 75 stream reaches. TITAN analysis cut-off levels (purity  $\geq 0.75$ ; rel05 and rel01  $\geq 0.75$ ) yielded 16,963 individuals, belonging to 49 species, of which 28 species are sensitive, and 21 species are tolerant to deforestation (Supporting Information S1.2).

According to the GLM results, LUI and TMax, at watershed and riparian scales, were significantly correlated with richness and abundance of sensitive and tolerant to deforestation species, and also with the grasses and trees on stream banks, riparian litter, and conductivity (Table 2.2). Litter packs on stream bed were related significantly with LUI, at watershed and riparian scales, and with TMax at the watershed scale. Sensitive species richness and abundance, trees on stream banks, riparian litter and litter packs on stream bed presented negative relationship with intensity and time of deforestation, while positive relations were found between tolerant species richness and abundance, grasses on stream banks and conductivity, and intensity and time of deforestation.

**Table 2.2.** GLM results of relations between the predictor variables and fish assemblage and habitat structure. Significant relations in bold.

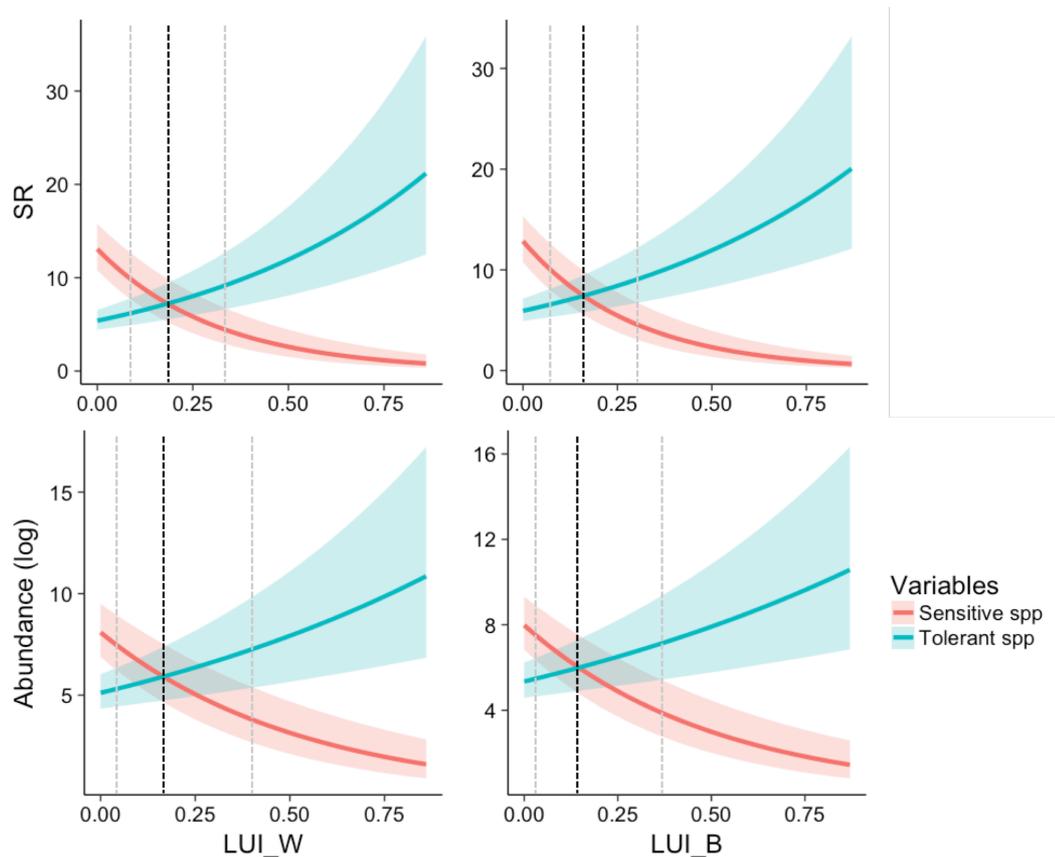
	LUIW		LUIB		TMaxW		TMaxB	
	<i>P</i>	Pseudo R <sup>2</sup>						
Sensi.SR	<b>&lt;0.001</b>	<b>57.3</b>	<b>&lt;0.001</b>	<b>60.3</b>	<b>&lt;0.001</b>	<b>51.2</b>	<b>&lt;0.001</b>	<b>44.9</b>
Tol.SR	<b>&lt;0.001</b>	<b>45.8</b>	<b>&lt;0.001</b>	<b>41.0</b>	<b>&lt;0.001</b>	<b>39.1</b>	<b>&lt;0.001</b>	<b>26.6</b>
Sensi.Ab	<b>&lt;0.001</b>	<b>40.8</b>	<b>&lt;0.001</b>	<b>43.7</b>	<b>&lt;0.001</b>	<b>38.4</b>	<b>&lt;0.001</b>	<b>31.9</b>
Tol.Ab	<b>&lt;0.001</b>	<b>37.3</b>	<b>&lt;0.001</b>	<b>33.5</b>	<b>&lt;0.001</b>	<b>31.4</b>	<b>&lt;0.001</b>	<b>23.4</b>
FRO	0.681	2.1	0.735	1.4	0.217	1.7	0.402	0.7
GRA	<b>&lt;0.001</b>	<b>40.4</b>	<b>&lt;0.001</b>	<b>38.2</b>	<b>&lt;0.001</b>	<b>37.3</b>	<b>&lt;0.001</b>	<b>21.3</b>
TRE	<b>0.021</b>	<b>38.3</b>	<b>0.022</b>	<b>42.2</b>	<b>&lt;0.001</b>	<b>24.8</b>	<b>&lt;0.001</b>	<b>19.6</b>
RIL	<b>0.033</b>	<b>31.7</b>	<b>0.028</b>	<b>32.5</b>	<b>&lt;0.001</b>	<b>30.2</b>	<b>0.001</b>	<b>14.6</b>
SAN	0.509	2.6	0.497	2.7	0.070	3.3	0.736	0.1
LIT	<b>0.028</b>	<b>35.4</b>	<b>0.026</b>	<b>40.2</b>	<b>0.036</b>	<b>28.6</b>	0.105	16.4
CSU	0.578	3.1	0.554	3.4	0.395	7.7	0.370	8.8
BAT	0.616	3.2	0.673	2.3	0.829	0.6	0.994	0.0
DEP	0.851	0.2	0.867	0.1	0.580	0.3	0.177	1.7
WID	0.292	2.4	0.387	1.6	0.110	5.5	0.149	4.5
CUR	0.887	0.2	0.992	0.0	0.755	1.1	0.568	3.6
DOX	0.598	0.5	0.488	0.8	0.374	1.3	0.392	1.2
CON	<b>&lt;0.001</b>	<b>14.7</b>	<b>0.001</b>	<b>14.4</b>	<b>&lt;0.001</b>	<b>10.6</b>	<b>&lt;0.001</b>	<b>9.0</b>
WTE	0.705	0.9	0.631	1.5	0.098	2.8	0.458	0.5

**LUI\_W**: Land use intensification on watershed; **LUI\_B**: Land use intensification on riparian zone; **TMax\_W**: Time since the high land use change since 1984 on watershed; **TMax\_B**: Time since the high land use change since 1984 on riparian buffer; **Sensi.SR**: Sensitive species richness; **Tol.SR**: Tolerant species richness; **Sensi.Ab**: Sensitive species abundance; **Tol.Ab**: Tolerant species abundance. Codes for environmental variables on Table 1.2.

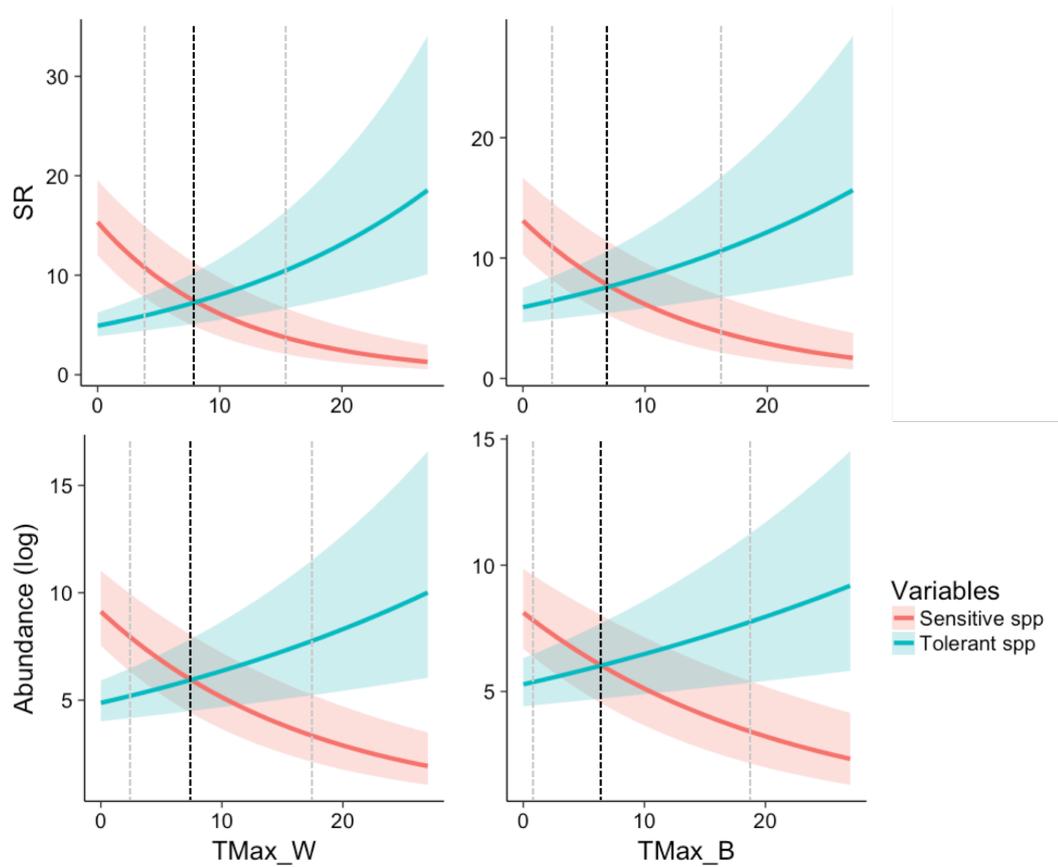
The alteration of instream habitat structure and sensitive fish richness and abundance lost (Fig. 2.2, 3.2) occurred synchronically with the habitat structure loss (litter packs, marginal trees, and riparian litter; Supporting Information S3.2). By contrast, we observed a time-lag response for habitat homogenization indicators (marginal grasses and conductivity; Supporting Information S4.2), and tolerant fish richness and abundance increase followed this time-lag (Fig. 2.2, 3.2). Marginal grasses and conductivity were indirect indicators of habitat homogenization. The marginal grasses amount is indicating an extreme

simplification of riparian vegetation and allochthonous material source, and the conductivity increase is related to the ionic input from stream banks, usually associated with stream siltation.

The sensitive and tolerant species curves, given by the GLM, intersect occurs at values of land use intensification lower than 0.25 (Fig. 2.2) and in less than ten years after a considerable land use change (Fig. 3.2), whether for basin watershed or riparian zone.



**Figure 2.2.** Sensitive and Tolerant fish species richness (SR) and abundance responses to the proportion of Land Use Intensification at the evaluated period – 1984 to 2011 – on watershed (LUI\_W) and riparian zone (LUI\_B). Black dashed line indicates the crossing point between sensitive and tolerant species. Gray dashed lines indicates the crossing of confidence intervals given by GLM.



**Figure 3.2.** Sensitive and Tolerant fish species richness (SR) and abundance responses to the time since the high land use change since 1984 on watershed (TMax\_W) and riparian zone (TMax\_B). Black dashed line indicates the crossing point between sensitive and tolerant species. Gray dashed lines indicates the crossing of confidence intervals given by GLM.

## Discussion

Although deforestation causes forest loss immediately in the terrestrial environment, our results show that its effects on aquatic ecosystems (e.g., habitat loss and homogenization) take time to occur. We expected that the sensitive fish diversity loss would follow the same pattern, exhibiting a lag response to habitat loss. However, we observed that the decrease of sensitive species diversity occurs synchronously to the loss of habitat structure indicators, while the increase of the tolerant species diversity occurs synchronously to the increase of habitat homogenization indicators. Furthermore, the tipping point between sensitive species loss and tolerant species increase occurs at low levels of land use intensification (less than 0.25) and in a short time since a considerable land use change (less than ten years), being consistent to species richness and abundance at watershed and riparian zone scales.

For amphibians, birds and mammals from the Atlantic Forest, an important biodiversity hotspot, the tipping points for community state change is around 15-40% of forest cover, and indicate that forest specialists species decrease and disturbance-adapted species increase (Banks-Leite et al. 2014). Since LUI index is measuring the accumulated deforestation over the evaluated period (1984-2011), varying from 0 to 1, the tipping point for fish community state change is 2-3 times lower than that detected for terrestrial vertebrates.

Previous studies on aquatic (Harding et al. 1998; Burcher et al. 2008) and terrestrial (Uezu & Metzger 2016) taxa demonstrate that contemporary communities may be more closely related to habitat conditions from decades ago.

However, the time lags reported in those studies are much longer than the temporal responses detected here. Moreover, even minimal impact activities, as selective logging, can cause sudden changes in fish species richness and abundance, and those effects are detectable even eight years after the logging event (Dias et al. 2010). Differently from this minimal impact activity, our assemblage was submitted to an intense land use change, where usually large areas were opened by forest clear-cutting. Consequently, we observed sudden changes on fish communities richness and abundance, as already detected for fish populations that respond to low levels of deforestation over space and time (Brejão et al. 2018). The extirpation of sensitive species will undermine the integrity of critical ecological processes occurring in streams (Leitão et al. 2016). Nevertheless, the streams studied by Dias et al. (2010) were buffered by a forest matrix, and probably even though detecting a state change in the fish community, it is more presumable that these assemblages will not suffer species extirpations along time.

At the Machado river basin scale, approximately 50% of the total amount of forest remains, but they are located mainly in the upper and lower reaches of the basin, with the most deforested area concentrated in the middle of the basin and along the left bank of the Machado river. For example, Ochoa-Quintero et al. (2015) modeled deforestation area of the lower Machado for the next 20 years, during which a critical threshold of <43% remaining forest cover is surpassed, and their species richness distribution model suggests the loss of up to 44% of studied terrestrial species. For stream fishes in this region, our analyses indicated that

thresholds for most sensitive taxa will be crossed far sooner (see Brejão et al. 2018, Chapter 1) than the critical threshold for SR used by Ochoa-Quintero et al. (2015), but we are unable to infer about how the spatial arrangement of deforestation (e.g., protecting intact riparian zones as deforestation continues) may mediate this process. The 100 m width riparian zone along all streams is highly fragmented and represents only 12% of the total area of Machado basin. This clustering of deforestation and lack of the deforestation treatments in our dataset (i.e., streams with deforested watersheds but intact riparian zones, and streams with deforested riparian zones but intact watersheds are uncommon; Brejão et al. 2018) limits our ability to predict if species responses under a scenario where riparian zones are protected, as determined by law, should be able to influence our results.

The instream habitat structure can be recovered through forest restoration. Notwithstanding, it may take some years until the restoration reaches the goal and this process is heavily dependent on the interactions between site-specific factors and land use (Guariguata & Ostertag 2001). It is unknown how long the restored forest begins to act as a facilitator the restructuring of aquatic habitat, and how long it will take. To an effective restructuring of the fish assemblage, the restoration must be planned on the landscape scale, reconnecting areas that have passed through the disturbance to the remnants of diversity, allowing the dispersal of individuals and the establishment of new populations. Detect the peak in time to reverse the disturbance by ecological restoration without the community reaches a critical state of diversity loss, would optimize the community's return to

a stable state close to the original, minimizing diversity loss. On the other hand, riparian forests maintenance become an essential strategy to the channel and aquatic communities' conservation, since it is the most efficient way to promote stream protection in managed areas (Quinn et al. 2004; Moore & Richardson 2012; Richardson et al. 2012).

In summary, according to our results, the process of colonization and establishment of tolerant species in degraded streams takes longer time than the loss of sensitive species, which occurs little after deforestation, so the stream fish assemblage from Machado river basin is responding to a temporal landscape dynamic mediated habitat suitability-unsuitability gradient.

These results also reinforce the conclusion on the first chapter: “the small stream fishes dependent on inputs from riparian forest have restricted distributions, and have low lifetime dispersal rates respond strongly to watershed and riparian deforestation, they may serve as good indicators of ensuing impacts of deforestation on other taxa” (Brejão et al. 2018).

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## Supporting Information 1.2

Fish species included in this study, TITAN positioning (z+/z-), occurrence, and total abundances. Classification follows Reis et al. (2003).

TAXON	+/-	Occurrence	Abundance
<b>CHARACIFORMES</b>			
<b>Curimatidae</b>			
<i>Steindachnerina fasciata</i> (Vari & Géry, 1985)	z+	14	57
<b>Crenuchidae</b>			
<i>Characidium</i> aff. <i>zebra</i> Eigenmann, 1909	z+	54	762
<i>Elachocharax pulcher</i> Myers, 1927	z-	9	79
<i>Microcharacidium</i> aff. <i>weitzmani</i> (Buckup, 1993)	z-	9	38
<b>Gasteropelecidae</b>			
<i>Carnegiella strigata</i> (Günther, 1864)	z-	7	40
<b>Characidae</b>			
<i>Astyanax</i> cf. <i>bimaculatus</i> (Linnaeus, 1758)	z+	19	108
<i>Bryconella pallidifrons</i> (Fowler, 1946)	z-	8	695
<i>Bryconops caudomaculatus</i> (Günther, 1864)	z+	50	912
<i>Creagrutus petilus</i> Vari & Harold, 2001	z+	48	1021
<i>Hemigrammus melanochrous</i> Fowler, 1913	z-	11	1418
<i>Hyphessobrycon</i> aff. <i>heterorhabdus</i> (Ulrey, 1894)	z-	11	144
<i>Hyphessobrycon agulha</i> Fowler, 1913	z-	15	1131
<i>Jupiaba citrina</i> Zanata & Ohara, 2009	z+	19	273
<i>Knodus</i> cf. <i>smithi</i> Fowler, 1913	z+	35	827
<i>Moenkhausia cotinho</i> Eigenmann, 1908	z+	17	259
<i>Moenkhausia grandisquamis</i> Müller & Troschel, 1845	z-	7	11
<i>Phenacogaster retropinnus</i> Lucena & Malabarba, 2010	z+	42	386
<i>Serrapinnus microdon</i> (Eigenmann, 1915)	z+	30	1901
<i>Serrapinnus</i> aff. <i>notomelas</i> (Eigenmann, 1915)	z+	34	3642
<b>Erythrinidae</b>			
<i>Erythrinus erythrinus</i> (Bloch & Schneider, 1801)	z-	8	11
<i>Hoplias malabaricus</i> (Bloch, 1794)	z-	36	88
<b>Lebiasinidae</b>			
<i>Pyrrhulina</i> cf. <i>australis</i> Eigenmann & Kennedy, 1903	z-	24	193
<i>Pyrrhulina</i> cf. <i>brevis</i> Steindachner, 1876	z-	9	65
<b>SILURIFORMES</b>			
<b>Cetopsidae</b>			
<i>Helogenes gouldingi</i> Vari & Ortega, 1986	z-	9	22
<b>Aspredinidae</b>			
<i>Pseudobunocephalus amazonicus</i> (Mees, 1989)	z-	8	37
<b>Trichomycteridae</b>			
<i>Ituglanis amazonicus</i> (Steindachner, 1882)	z-	20	108
<b>Callichthyidae</b>			

<b>TAXON</b>	<b>+/-</b>	<b>Occurrence</b>	<b>Abundance</b>
<i>Corydoras trilineatus</i> Cope, 1872	z-	22	82
<i>Megalechis picta</i> (Müller & Troschel, 1849)	z-	6	49
<b>Loricariidae</b>			
<i>Ancistrus lithurgicus</i> Eigenmann, 1912	z+	26	290
<i>Hypostomus pyrineusi</i> (Miranda Ribeiro, 1920)	z+	15	28
<i>Lasiancistrus schomburgkii</i> (Günther, 1864)	z-	18	61
<i>Rineloricaria heteroptera</i> Isbrücker & Nijssen, 1976	z+	40	164
<b>Pseudopimelodidae</b>			
<i>Batrochoglanis</i> cf. <i>raninus</i> (Valenciennes, 1840)	z-	5	16
<b>Heptapteridae</b>			
<i>Imparfinis</i> cf. <i>hasemani</i> Steindachner, 1917	z-	19	124
<i>Pimelodella</i> cf. <i>howesi</i> Fowler, 1940	z-	16	55
<i>Rhamdia quelen</i> (Quoy & Gaimard, 1824)	z+	4	6
<b>Doradidae</b>			
<i>Acanthodoras cataphractus</i> (Linnaeus, 1758)	z-	4	19
<b>GYMNOTIFORMES</b>			
<b>Gymnotidae</b>			
<i>Gymnotus</i> aff. <i>arapaima</i> Albert & Crampton, 2001	z+	9	26
<i>Gymnotus coropinae</i> Hoederman, 1962	z-	15	81
<b>Sternopygidae</b>			
<i>Eigenmannia trilineata</i> López & Castello, 1966	z+	19	196
<i>Sternopygus macrurus</i> (Bloch & Schneider, 1801)	z+	24	97
<b>Rhamphichthyidae</b>			
<i>Gymnorhamphichthys petiti</i> Géry & Vu-Tân-Tuê, 1964	z-	31	287
<b>Hypopomidae</b>			
<i>Brachyhypopomus</i> sp. 2	z-	8	15
<i>Brachyhypopomus</i> sp. 3	z-	5	26
<i>Hypopygus lepturus</i> Hoedeman, 1962	z-	17	128
<b>PERCIFORMES</b>			
<b>Cichlidae</b>			
<i>Aequidens tetramerus</i> (Heckel, 1840)	z+	39	199
<i>Apistogramma</i> cf. <i>resticulosa</i> Kullander, 1980	z-	23	563
<i>Crenicichla santosi</i> Ploeg, 1991	z+	40	163
<i>Satanoperca jurupari</i> (Heckel, 1840)	z+	13	60

## Supporting Information S2.2

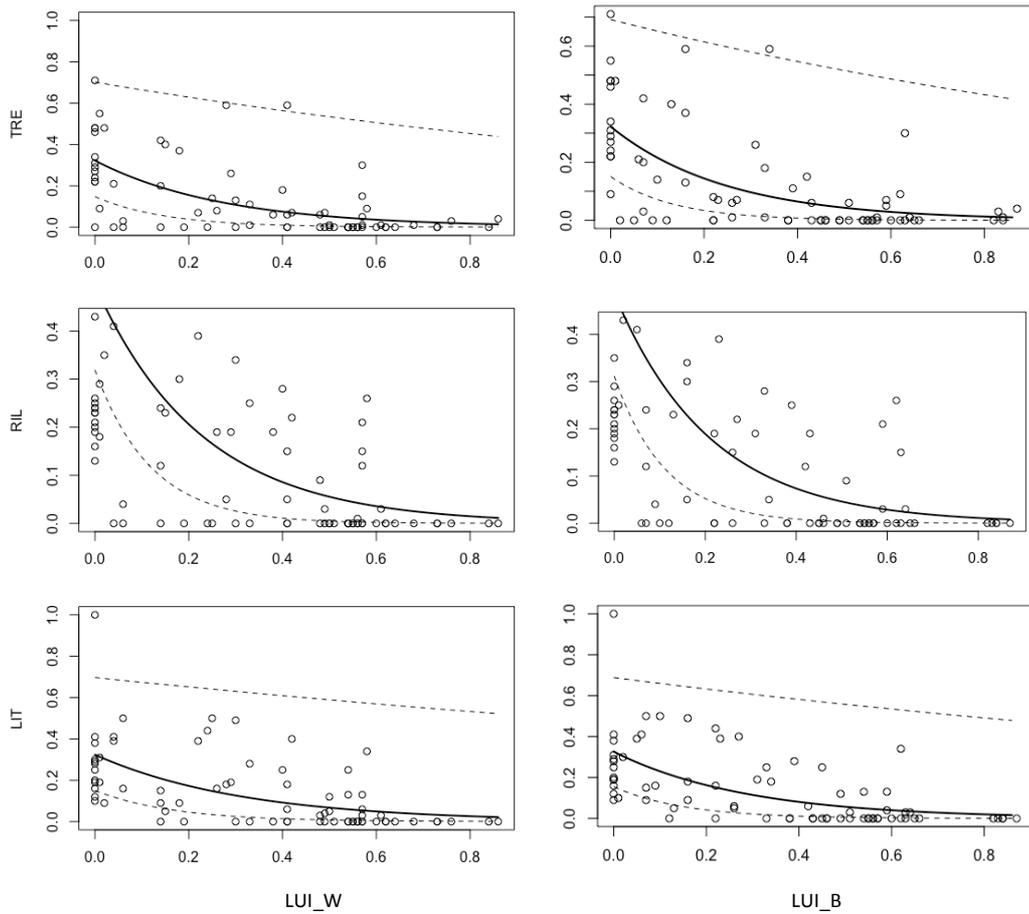
Environmental gradients, sensitive and tolerant species richness, and abundance for each of 75 sampled streams. **LUI**: Land Use Intensification at the evaluated period – 1984 to 2011 – on watershed (**LUI\_W**) and riparian zone (**LUI\_B**); **TMax**: time since the high land use change since 1984 on watershed (**TMax\_W**) and riparian zone (**TMax\_B**).

Streams	TMax_W	TMax_B	LUI_W	LUI_B	Richness		Abundance	
					Sensitive	Tolerant	Sensitive	Tolerant
S01	20	4	0.57	0.51	0	13	0	91
S02	20	20	0.41	0.49	1	14	1	192
S03	20	20	0.5	0.49	1	12	1	180
S04	16	16	0.73	0.65	3	18	3	131
S05	24	16	0.41	0.46	2	19	2	429
S06	16	16	0.54	0.54	2	11	2	80
S07	16	16	0.64	0.62	4	19	7	348
S08	16	20	0.51	0.66	3	7	28	79
S09	20	20	0.49	0.43	2	17	5	494
S10	20	20	0.48	0.45	3	17	8	185
S11	27	27	0.55	0.56	2	21	5	707
S12	16	16	0.62	0.55	1	13	2	404
S13	20	20	0.54	0.45	2	17	3	364
S14	16	20	0.06	0.07	13	6	266	25
S15	16	4	0.25	0.1	18	10	283	65
S16	8	16	0.04	0.06	9	4	368	62
S17	8	8	0.3	0.16	16	5	499	262
S18	8	8	0.22	0.23	12	9	286	49
S19	8	16	0.24	0.22	13	9	61	72
S20	8	8	0.19	0.22	16	10	65	134
S21	12	16	0.41	0.26	3	7	27	81
S22	8	16	0.04	0.05	16	5	353	58
S23	0	0	0	0.02	11	9	188	108
S24	0	0	0	0	17	7	303	24
S25	0	0	0	0	16	2	435	4
S26	0	0	0	0	21	2	670	17
S27	0	0	0	0	12	2	216	3
S28	0	0	0	0	16	7	307	32
S29	0	0	0	0	19	5	419	18
S30	0	0	0	0	11	1	453	1
S31	0	0	0	0	15	2	139	2
S32	8	8	0.28	0.16	11	10	149	53
S33	4	0	0	0	7	2	46	5
S34	8	8	0.14	0.07	5	7	72	25
S35	8	16	0.14	0.07	4	6	16	15

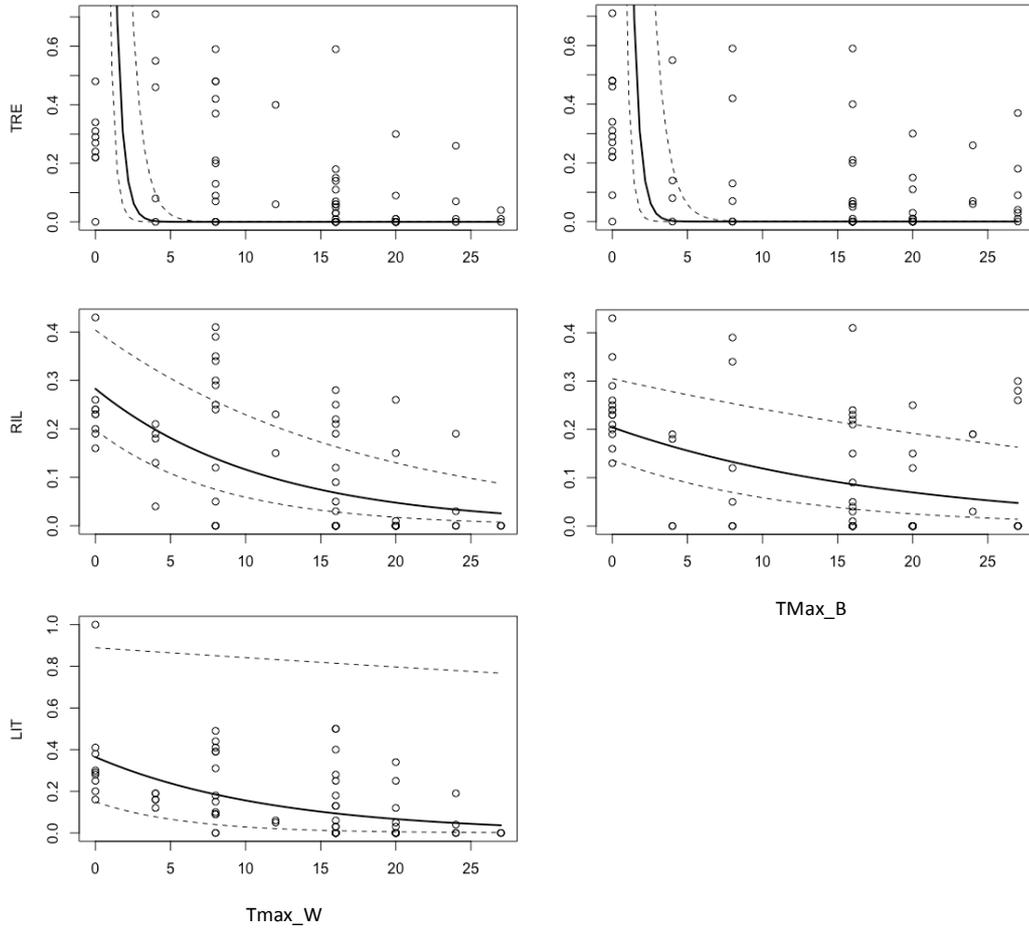
S36	12	16	0.15	0.13	12	8	83	52
S37	8	8	0.14	0.12	7	15	19	112
S38	4	0	0	0	11	10	61	55
S39	8	0	0.01	0	15	7	55	52
S40	24	16	0.55	0.54	1	7	12	87
S41	24	24	0.49	0.59	1	12	2	244
S42	16	16	0.61	0.64	0	15	0	148
S43	4	4	0.01	0	6	9	30	36
S44	8	0	0.02	0	6	5	38	13
S45	8	0	0	0.01	6	0	17	0
S46	24	20	0.33	0.33	5	10	12	84
S47	16	16	0.48	0.51	1	13	1	46
S48	27	27	0.68	0.84	1	20	1	474
S49	4	4	0.26	0.22	3	7	4	32
S50	20	27	0.58	0.62	3	17	7	222
S51	16	24	0.38	0.43	0	10	0	98
S52	20	20	0.57	0.57	2	18	6	266
S53	20	20	0.62	0.56	2	15	3	300
S54	20	20	0.6	0.6	0	16	0	209
S55	20	20	0.54	0.57	2	14	7	144
S56	20	16	0.56	0.55	2	18	11	2042
S57	16	27	0.76	0.83	2	17	2	439
S58	16	16	0.41	0.34	2	10	3	84
S59	16	20	0.33	0.39	6	13	34	115
S60	20	20	0.57	0.63	1	11	2	331
S61	24	24	0.29	0.31	3	3	4	4
S62	16	8	0.3	0.38	1	2	1	50
S63	4	16	0.06	0.09	0	3	0	39
S64	8	27	0.18	0.16	0	1	0	3
S65	27	27	0.86	0.87	0	10	0	53
S66	20	16	0.56	0.46	1	8	1	121
S67	16	16	0.42	0.27	3	9	3	100
S68	16	16	0.57	0.59	1	6	4	14
S69	16	20	0.57	0.42	4	9	28	118
S70	16	16	0.84	0.82	1	11	8	201
S71	16	16	0.73	0.84	4	15	21	180
S72	16	16	0.56	0.63	1	17	1	344
S73	16	27	0.4	0.33	4	13	10	62
S74	20	20	0.5	0.26	2	15	6	118
S75	16	16	0.54	0.38	2	20	5	527

### Supporting Information S3.2

GLM results between Land Use Intensification at the evaluated period – 1984 to 2011 – on watershed (LUI\_W) and riparian zone (LUI\_B) and habitat structure indicators. **TRE**: Proportion of trees stream banks; **RIL**: Proportion of riparian litter; **LIT**: Proportion of litter in stream bed.

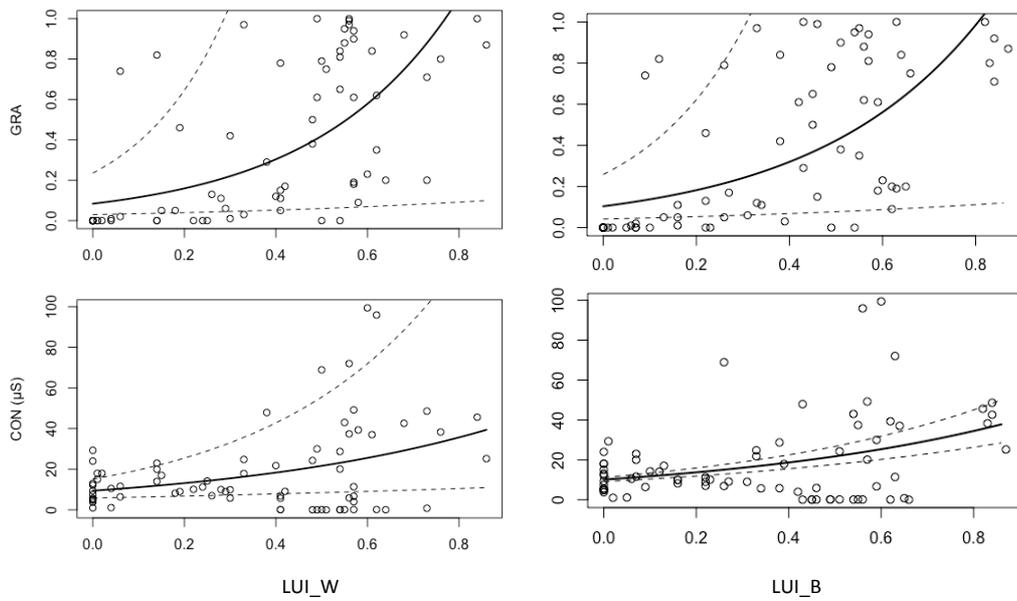


GLM results between time since the high land use change since 1984 on watershed (TMax\_W) and riparian zone (TMax\_B) and habitat homogenization indicators. **TRE**: Proportion of trees stream banks; **RIL**: Proportion of riparian litter; **LIT**: Proportion of litter in stream bed.

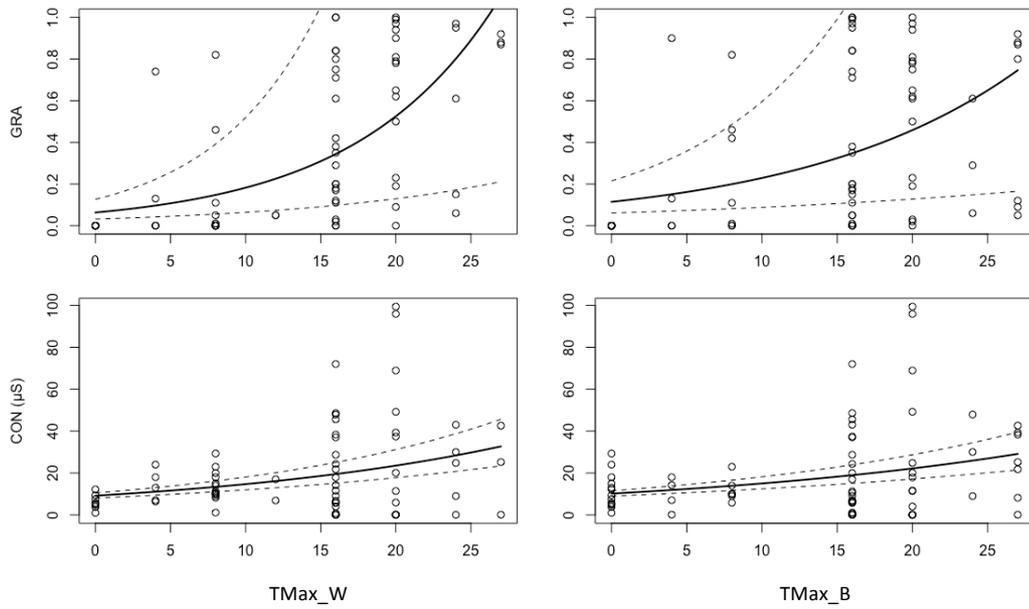


## Supporting Information S4.2

GLM results between Land Use Intensification at the evaluated period – 1984 to 2011 – on watershed (LUI\_W) and riparian zone (LUI\_B) and habitat homogenization indicators. **GRA**: Proportion of grasses stream banks; **CON**: Conductivity ( $\mu\text{S}$ ).



GLM results between time since the high land use change since 1984 on watershed (TMax\_W) and riparian zone (TMax\_B) and habitat homogenization indicators. **GRA**: Proportion of grasses stream banks; **CON**: Conductivity ( $\mu\text{S}$ ).



## **Chapter 3**

**Current stream fish assemblage composition is determined by  
deforestation history and different scale environmental variables  
in Amazon**

## **Abstract**

High rates of deforestation in rainforests, either in the past or in the present, affect many of the ecological processes occurring in the streams and there are evidences about the importance of the past land use to determine stream biodiversity in the present. The deforestation history and current landscape structure enhance the analytical power to evaluate ecological effects of this impact, which appear to be similar in the present time, although their path until the current state has not been the same. Complementarily, the use of functional diversity arises as an important tool to evaluate the magnitude of the impact on the aquatic environment, regarding these distinct paths of deforestation process, and how it might affect the fish communities' composition and functioning. In matter to understand how deforestation dynamics affected the species/traits turnover in a recently deforested watershed, we applied a deconstructive approach, splitting 75 streams from Machado river basin into three groups according to their deforestation dynamics. We observed that the turnover was different than expected by chance, indicating that deterministic processes are structuring this stream fish assemblage, presenting high species turnover, but the functional traits turnover was lower than the expected by the species turnover. We also observed differences among the three stream groups, verifying that the recently deforested streams presented higher species turnover rates than the forested and oldened deforested streams, but lower functional than the forested and olden deforested streams. Turnover among forested streams was driven by instream environmental variables, while in deforested streams the turnover was driven by local and watershed environmental

variables. In summary, this deconstructive approach, grouping streams by landscape dynamic allow us to better detect the effects of deforestation on fish assemblage taxonomic and functional turnover, suggesting that, besides being scale-dependent, turnover patterns could be dynamic-dependent.

## **Introduction**

Rivers and streams are characterized by its linear form, unidirectional flow and unstable bed. The elongated shape of its channels entails an intimate connection with the surrounding terrestrial environment (Townsend et al. 2003). In these specific conditions, the freshwater ecosystems are likely to accumulate impacts related to the landscape modification, since rivers and streams connect and concentrate the land use activities effects in its surroundings (Hynes 1975; Wear et al. 1998).

High rates of deforestation in rainforests, either in the past or the present, affect many of the ecological processes occurring in the streams (Wright & Flecker 2004; Paula et al. 2011). Forest ecosystems have critical ecological functions in mitigating impacts caused by land use and cover alterations (e.g., forest management and agriculture) in aquatic ecosystems and water resources (Dwire & Lowrance 2006). Moreover, native vegetation removal from watersheds is one of these modifications, whose effects are not fully understood (e.g., time since deforestation, deforestation dynamics, spatial configuration of the deforestation, forest recover) mainly in tropical ecosystems (Bojsen & Barriga 2002; Lorion & Kennedy 2009; Leal et al 2016).

There is evidence about the land use in the past as a determiner of the streams species diversity in the present (Harding et al. 1998; Iwata et al. 2003). The use of deforestation history along with current landscape structure enhances the analytical power to evaluate ecological effects in different regions, which appear to be similar in the present time, although their path until the current state

has not been the same (Ferraz et al. 2009). Complementarily, the use of functional diversity arises as an important tool to evaluate the magnitude of the impact on the aquatic environment, regarding these distinct paths of deforestation process, and how it might affect the fish communities' composition and functioning.

According to Wittaker (1960), species diversity can be decomposed into three components: gamma, alpha, and beta. The  $\beta$ -diversity is the spatial or temporal variation on species composition (Anderson et al. 2006). Habitat heterogeneity is an important driver to  $\beta$ -diversity (Anderson et al. 2011), and the landscape modification also is a key factor in determining the  $\beta$ -diversity in a region (Siqueira et al. 2015).

When comparing communities along an environmental gradient, is expected to found a positive relationship between species  $\beta$ -diversity and functional  $\beta$ -diversity. However, whether the functional turnover between communities is higher or lower than that expected by the species turnover remains as a challenge question (Swenson et al. 2011). Moreover, different species, which presents similar sets of traits, tend to perform the same ecological function and to occupy the same environments (Winemiller 1991), so a high rate of species turnover not necessarily will determine a high rate of functional traits turnover. Thus, including multiple facets of biodiversity, besides the taxonomic, permits the improvement of the ecological mechanisms underlying biodiversity patterns and community assembly (Petchey & Gaston 2006; Swenson et al. 2012).

According to the intermediate disturbance hypothesis (Connell 1978), biological communities are subjected to disturbances that occur with different

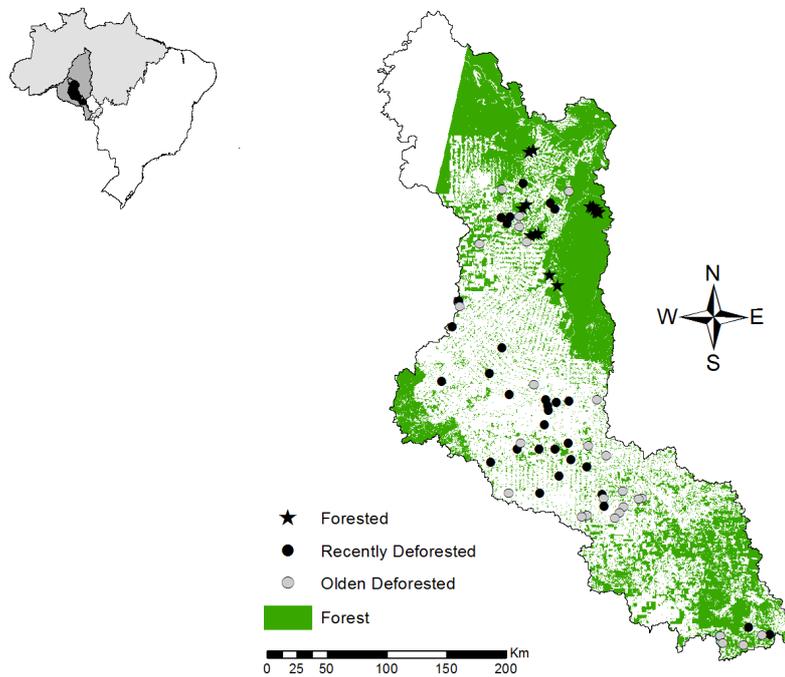
frequencies and intensities. Considering the deforestation process, we can classify the landscape in pristine areas as homogeneous with rare, disturbing events; when they occur, stochastic events drive them, with the consequences buffered by the forested condition in the surroundings. By contrast, in altered areas, the landscape is more heterogeneous, and two disturbance states can be found (1.) recent deforestation, with high disturbance frequency and intensity, and (2.) old deforestation, presenting low disturbance frequency and intensity. Based on this scenario, we tested whether:

- (i) Turnover will be higher than expected by chance in the deforested pool of streams when compared to the forested ones, and the recently deforested streams will present higher turnover than the older ones.
- (ii) Local (instream) variables will drive the turnover in forested streams, while both watershed (landscape) and local variables driving the turnover in deforested streams.

## **Methods**

### *Study area*

We surveyed 75 first- to third-order streams in the Machado River basin, Brazil (Fig. 1.3). Machado River is a tributary of the Madeira River, this region has many terra-firme streams, which are intermittent during most of the dry season and drains the most populated region of Rondônia, Brazil (Fernandes & Guimarães 2002). In general, sampled streams were shallow with low-velocity flow and warm temperatures. The streambeds were predominantly sand, litter pack, and large woody debris, and stream banks usually provided submerged microhabitat structure derived from the riparian environment such as tree roots and grasses (Table 1.3). The climate is tropical humid: temperatures range from 19 to 33° C, and annual rainfall is 2,500 mm (Krusche et al. 2005). The land cover in this region includes primary forest (open humid tropical forest), secondary forest, and pasture (Ferraz et al. 2009).



**Figure 1.3.** Sampled sites and forest fragments in the Machado River basin, Brazil. The inset map of Brazil depicts the relative location of the study area (black) within the Madeira River basin (dark gray), inside the Amazon biome (light gray).

### *Sampling Methods*

Streams were sampled once in the dry season (August–October 2011 and June–July 2012). Sampling methods were standardized to allow for comparisons of species abundances across sites. Each stream reach was 80 m long and isolated before sampling with block nets (5-mm mesh). Fishes were sampled from all available microhabitats by two collectors using a seine (1.5 x 2 m, 2-mm mesh) and dip nets (0.5 x 0.8 m, 2-mm mesh) for 1 hour. Fishes were collected under “Instituto Chico Mendes de Conservação da Biodiversidade” permits 4355-1/2012. All individuals were identified with assistance from taxonomic specialists, and voucher specimens deposited in the “Coleção de Peixes do

Departamento de Zoologia e Botânica (DZSJRP)” at the “Universidade Estadual Paulista Júlio de Mesquita Filho”, São José do Rio Preto, São Paulo State, Brazil.

*Environmental variables*

After reach delimitation, local physicochemical and riparian ecotone habitat variables were quantified using standard methods (Table 1.3). Land use classification was made using Landsat 5 TM satellite images (30 x 30 m resolution) obtained and released by the National Institute for Space Research (INPE) for watershed and riparian buffer scales. The land use map consists of three categories: primary forest, secondary forest, and pasture. The classification was made using the method supervised by Maximum Likelihood Classification (Jensen 2007) and the software Erdas 9.2.

**Table 1.3.** Summary of local variables, mean  $\pm$  standard deviation, and explanation of how each variable was obtained.

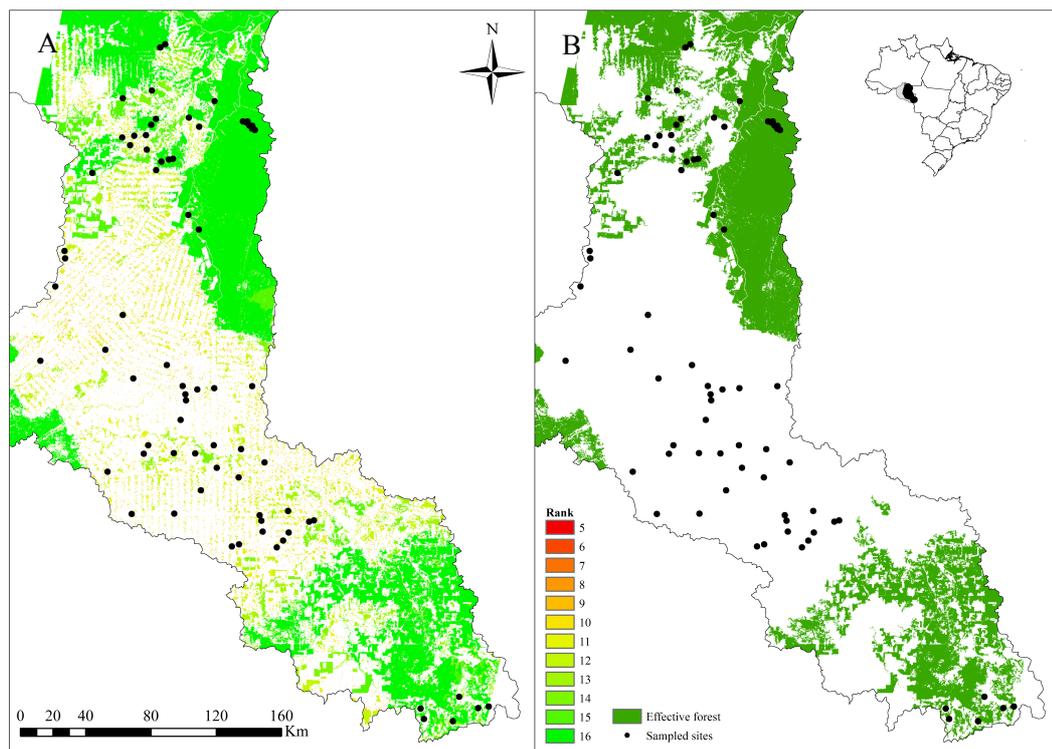
<b>Variables</b>	<b>Code</b>	<b>Mean <math>\pm</math> standard deviation</b>	<b>Explanation</b>
Riparian ecotone variables (For this calculation, both stream sides were computed):			
Trees in stream banks (%)	TRE	13.23 $\pm$ 18.33	- Percentage of the reach bank extension that was covered by riparian trees.
Grasses in stream banks (%)	GRA	35.03 $\pm$ 38.01	- Percentage of the reach bank extension that was covered by marginal grasses derived from surrounding pasture entering the water.
Submerged roots in stream banks (%)	FRO	3.33 $\pm$ 7.63	- Percentage of the reach bank extension that presented roots derived from riparian trees entering the water.
Riparian litter (%)	RLI	10.79 $\pm$ 12.66	- Percentage of the reach bank extension that presented fallen leaves derived from riparian trees
Instream variables:			
Sand (%)	SAN	53.58 $\pm$ 29.17	- Percentage of sand on the bottom of each reach
Consolidate substrate (%)	CSU	5.08 $\pm$ 9.94	Percentage of gravel and cobbles (particles with 2-256 mm in size) on the bottom of each stream reach
Litter packs on stream bed (%)	LIT	14.34 $\pm$ 18.24	- Percentage of fallen leaves on the streambed of each reach
Large wood debris on stream bed (%)	BAT	11.35 $\pm$ 10.78	- Percentage of fallen branches and tree trunks, representing large wood debris, on the streambed of each reach.
Depth (cm)	DEP	27.54 $\pm$ 13.68	- Average value of depth
Width (m)	WID	3.00 $\pm$ 1.41	- Average value of width
Water flow (m s <sup>-1</sup> )	CUR	0.38 $\pm$ 0.23	- Average value of water flow velocity
Dissolved oxygen (mg l <sup>-1</sup> )	DOX	6.63 $\pm$ 2.24	- Average value of Dissolved oxygen
Conductivity ( $\mu$ S)	CON	18.39 $\pm$ 21.09	- Average value of Water conductivity
Temperature ( $^{\circ}$ C)	WTE	24.06 $\pm$ 2.26	- Average value of Water temperature

Following the methodology proposed by Ferraz et al. (2014), we calculated a multimetric landscape index, which scores each forest pixel according to the metrics shown in table 2.3. These metrics take into account the landscape configuration throughout the Machado River basin and assign each sample unit (pixels) a value based on the effect of the deforestation process on a regional scale.

**Table 2.3.** Metrics used to estimate forest quality ranking based on landscape and fragments structure.

<b>Metrics</b>	<b>Weight</b>	
Forest age (years)		Forestage is defined by temporal overlaying of land cover maps, using the difference between the most recent date and the first year of forest occurrence in the past. A zonal statistical analysis can be performed to calculate area-weighted mean values of forest age for each forest cell (Ferraz et al. 2014).
0 – 7	1	
7.1 – 19	2	
19.1 – 23	3	
> 23	4	
Local forest neighborhood dominance (%)		Considering that interior forest can perform better ecosystem services than forest edge, to assess local forest neighborhood dominance by examining the eight surrounding cells around each forest focal cell, in order to calculate the proportion covered by forest. A moving window analysis can be performed at this step (Ferraz et al. 2014).
0	1	
0.1 – 0.33	2	
0.34 – 0.66	3	
0.67 - 1	4	
Forest proximity (no units)		Proximity was used as surrogate of local habitat connectivity, considering that more connected patches provided higher levels of some regulating ecosystem services than more isolated ones. This index was calculated as the mean proximity index of forest present in a 2 km buffer around forest cells (Ferraz et al. 2014).
0 – 76	1	
76.1 – 1200	2	
1200.1 – 5000	3	
> 5000	4	
Forest contiguity (%)		This metric brings to forest cells the relative size of their forest patch in relation to focal landscape. We considered that bigger forest patches were able to provide higher levels of ecosystem services provisioning. In order to capture the forest contiguity of each forest cell, we propose to use the proportion of focal landscape occupied by forest patches where forest cells are inserted (Ferraz et al. 2014).
0	1	
0.1 – 0.8	2	
0.9 – 2.4	3	
> 2.4	4	

For each metric, we produced a thematic map, ranking each 1 ha pixel (100x100m) according to the sum operation among all thematic maps; the sum values varied between 5 and 16 (Fig. 2.3A). From this maps operation, we took the highest valued pixels (15 and 16) to determine forest areas with high quality, and thus, ancient forests, with high contiguity and proximity from the relevant forest patches in the basin located far from the patch edge. This high-quality forest has been called Effective Forest, that is that forest patches that would be adequately performing their ecosystem functions (Fig. 2.3B).



**Figure 2.3.** Forest patches distribution at Machado River Basin. 2A – Forest patches ranked according to the forest quality multimetric index. 2B – Effective forest cover.

Land use change was analyzed from 1984 to 2011 at 4-year intervals and calculated using the Land-use Change Analysis Tool - LUCAT (Ferraz et al.

2011, 2012) for watershed and riparian zone, which calculates the area and proportion of the area occupied by each type of land use present in a unit of analysis. This tool also provides indexes such as the profile of the Forest Change Curve Profile (FCCP) and the Land Use Intensification (LUI) (Ferraz et al. 2009). The FCCP index represents the historical deforestation patterns, indicating when the vegetation change process has occurred, where positive and negative values represent old or recent changes. For deforestation, positive FCCP means old changes and FCCP negative recent change. This index varies from -1 to 1, where values close to the extremes represent more significant transformations in the landscape and values close to zero represent gradual changes in land use. LUI index represents the average time since the deforestation and can be considered as an accumulated effect metric over the time.

To verify the effect of deforestation processes (old or recent) clustering the watersheds in three groups, according to their FCCP classification: (1.) Reference - watersheds not deforested (ref, n = 18); (2.) Recently deforested - watersheds that underwent recent deforestation (new, n = 31); (3.) Older deforested - and watersheds that went through the process of old deforestation (old, n = 26).

Fluvial distance was assessed by using Network Analyst extension in the ArcGIS, that produces a triangular matrix containing the watercourse distance among each sampled site with the other 74. Landscape variables were summarized in table 3.3.

**Table 3.3.** Summary of Landscape variables, mean  $\pm$  standard deviation.

<b>Landscape variables</b>	<b>Code</b>	<b>Mean <math>\pm</math> standard deviation</b>
Watershed scale:		
Mature forest proportion	MF_WSHD	0.36 $\pm$ 0.36
Effective forest proportion	EF_WSHD	0.27 $\pm$ 0.40
Land use intensity	LUI_WSHD	0.35 $\pm$ 0.25
Riparian zone scale:		
Mature forest proportion	MF_BUFF	0.41 $\pm$ 0.38
Effective forest proportion	EF_BUFF	0.28 $\pm$ 0.42
Land use intensity	LUI_BUFF	0.34 $\pm$ 0.26

### *Functional traits*

Ten ecomorphological traits related to habitat use, foraging, and locomotion (see Table 1.1, Chapter 1) were quantified based on 11 measurements from adult individuals of each species (1–5 individuals per species). We measured linear distances, body and fins areas, and width of each individual to the nearest 0.01 mm with a stereomicroscope (Zeiss Discovery V12 SteREO) coupled with imaging software (Axio-Vision Zeiss) and a digital caliper.

### *Quantifying turnover rates*

The taxonomic turnover was estimated by using a null-modeling approach and a modified Raup-Crick metric (Raup & Crick 1979) for abundance data (Püttker et al. 2015). The advantage of this metric combined with the null-modeling approach is its independence from local species richness and permits to discriminate the compositional variation among local communities independently from differences in the local species richness (Chase et al. 2011). The null-

modeling approach measures the deviation of the expectation that the dissimilarity of the communities is stochastic (Raup & Crick 1979; Chase et al. 2011; Püttker et al. 2015), allowing to evaluate the importance of deterministic (niche-based) and stochastic (neutral) in the assembly of community (Chase et al. 2011; Püttker et al. 2015).

Functional turnover was estimated by using distance metrics based on the pairwise and nearest neighbor distances associated with a null-modeling approach (Roa-Fuentes 2016), using the functional traits matrix and species abundance data. For the pairwise metric, the  $\beta$ MPPD index was used, while for the measurements of the nearest neighbor metric the  $\beta$ MNTD index was used, both extensions of the  $\alpha$ MPPD and  $\alpha$ MNTD diversity metrics.  $\beta$ MPPD evaluates the total similarity or dissimilarity between two samples or communities, and it is sometimes considered as a basal diversity metric (Swenson 2014), whereas  $\beta$ MNTD is regarded as a terminal relatedness measure, being more sensitive to variations towards the tips of the dendrogram (Webb 2000). For these reasons, these measures may be considered complementary and be employed to discriminate patterns in turnover (Swenson 2011). For each metric (i.e.,  $\beta$ MPPD and  $\beta$ MNTD) the standardized effect size was calculated.

In this approach, positive values of functional turnover indicate a higher traits turnover than expected by species turnover, meaning that each community, in general, contains distantly functional related species (Swenson et al. 2011; Swenson 2014). Negative values of functional turnover indicate a lower traits turnover than expected by species turnover, meaning that variation between the

two communities occurs between close functional species (Swenson et al. 2011; Swenson 2014).

### *Data analysis*

To verify if the mean value of each turnover metric between sites was significantly different from the expected value for random data (zero), we performed a series of one sample t-test (Wang et al. 2013). To evaluate the variation of turnover, we applied a distance-based approach (Tuomisto & Ruokolainen 2006), where the turnover metrics was related to the environmental and spatial distances among the sampled communities. Mann-Whitney U test was applied to verify if the differences of turnover rates between stream groups were significant.

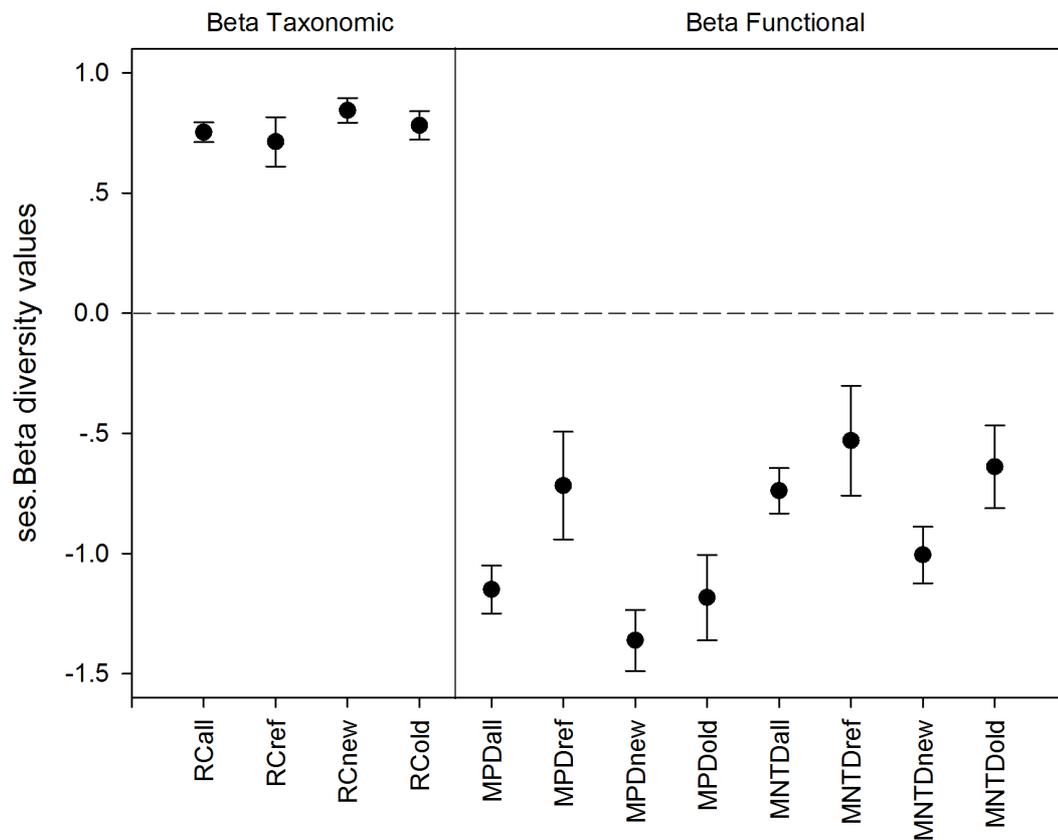
For each stream group, we applied the 'bioenv' function, from 'vegan' package (Oksanen et al. 2018) function to obtain the subsets of environmental variables best correlated with community turnover (Clarke & Ainsworth 1993) (Supplementary Information S3.3). To determine the significance of variation in turnover related to environmental, and spatial distances, we used Mantel and partial Mantel tests with 10,000 permutations (Legendre & Legendre 2012).

To verify the relative contributions of local, watershed and network distance on fish community turnover, we applied a multiple regression on matrices (MRM) approach (Legendre et al. 1994). We also performed a commonality analysis, that allows the partition of the coefficient of determination

( $R^2$ ) into effects explained uniquely by each predictor or predictor subset (*Unique*), and effects explained commonly by all possible combinations of predictors or predictor subsets (*Common*) (Sorice & Conner 2010; Ray-Mukherjee et al. 2014). For MRM analysis and estimations of beta coefficients we used ‘MRM’ function in ‘ecodist’ package (Goslee & Urban 2007), and for commonality analysis and structure coefficients we used ‘regr’ function of ‘yhat’ package (Nimon et al. 2017). All the analyses were performed in the R software (R Development Core Team 2014).

## **Results**

In total, we collected 22,851 individuals belonging to 138 species, 30 families and six orders in 75 stream reaches. From this total, forested streams (n=18) harbor 5,292 individuals from 84 species, recently deforested streams (n=31) 10,497 individuals from 110 species, and olden deforested streams (n=28) 7,062 individuals from 91 species (Supporting Information S1.3 and S2.3). In all of 12 evaluated metrics, mean turnover differs significantly from the expected zero value ( $P < 0.05$ , Fig. 3.3), indicating that each fish community is different from another than expected by chance. Taxonomic turnover has shown mean values greater than zero, indicating higher species turnover expected by chance. Functional turnover has demonstrated mean values lower than zero, suggesting that functional turnover was lower than the expected, given the observed species turnover (Supporting Information S4.3).



**Figure 3.3.** Mean values among sites of standardized effect sizes of each taxonomic and functional turnover metrics and their 95% confidence intervals. Significant differences from the expected value of zero for random data were assessed through a series of one sample t-test ( $P < 0.05$ ; Table 2). RC = Raup-Crick; MPD = mean pairwise distance; MNTD = mean nearest taxon distance; all = All sampled sites; ref = Forested sampled sites; new = Recently deforested sampled sites; old = Olden deforested sampled sites.

Turnover rates increased with environmental distance, the relationship between turnover and environmental distance showed a significant distance – decay on functional similarity for all standardized effect size for the tested metrics (Table 4.3). After controlling for stream network distance, all turnover standardized effect size remained significantly correlated with environmental

distance (Table 4.3). On the other hand, distance-decay relationships between pairwise turnover and stream network distance were significant for all taxonomic metrics and functional ses.β.MNTD.new (Table 4.3). After controlling for environmental distance, stream network distance was correlated significantly with ses.β.RC.all, ses.β.RC.new and ses.β.RC.old (Table 4.3).

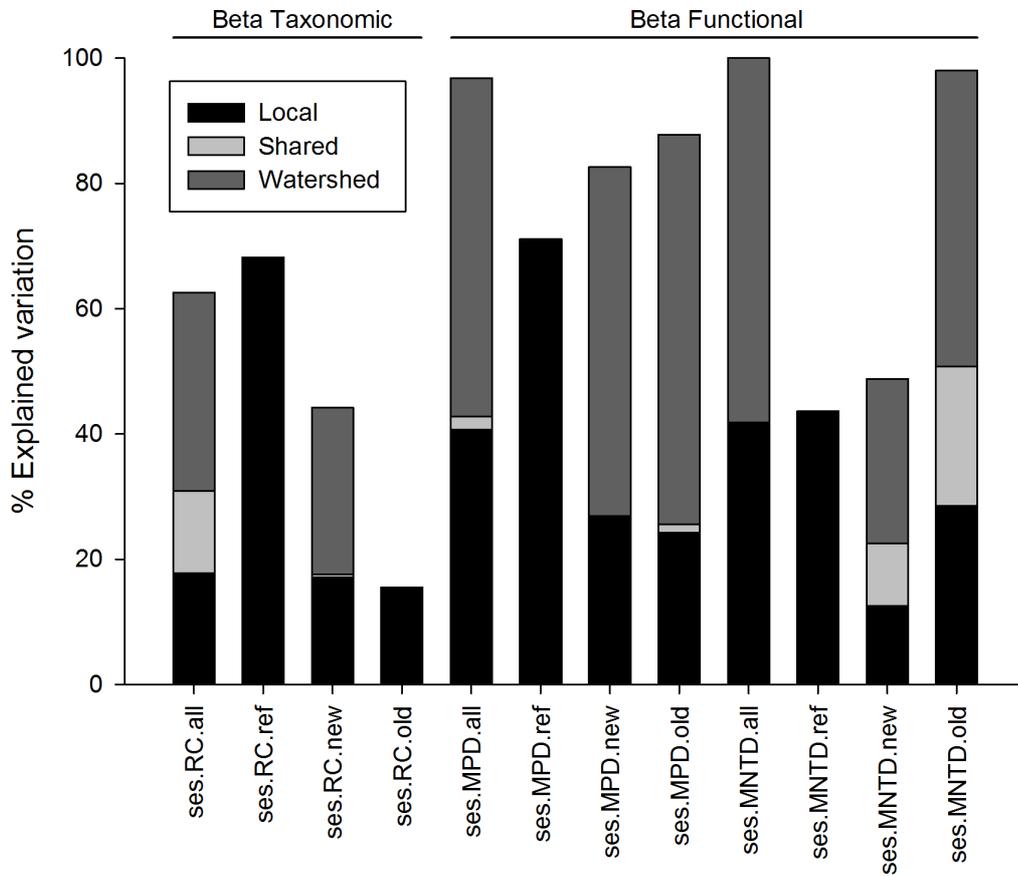
**Table 4.3.** Mantel and partial Mantel tests for the correlation between standardized effect size (SES) of turnover metrics and predictor distances (environmental and network) using Pearson's correlation ( $\rho$ ), with 10,000 permutations. Significant values in bold: \*\*\*  $P < 0.001$ ; \*\*  $P < 0.01$ ; \*  $P < 0.05$ .

Turnover facet	Effect of environment	Effect of network	Effect of environment controlling for network	Effect of network controlling for environment
Taxonomic				
ses.β.RC.all	<b>0.3584***</b>	<b>0.2297***</b>	<b>0.3132***</b>	<b>0.1407***</b>
ses.β.RC.ref	<b>0.6503***</b>	<b>0.372***</b>	<b>0.5867***</b>	0.1445
ses.β.RC.new	<b>0.2596***</b>	<b>0.2175**</b>	<b>0.1956**</b>	<b>0.1324*</b>
ses.β.RC.old	<b>0.2349**</b>	<b>0.2944***</b>	<b>0.1423*</b>	<b>0.23***</b>
Functional				
ses.β.MPD.all	<b>0.2874***</b>	0.05406	<b>0.2831***</b>	-0.0167
ses.β.MPD.ref	<b>0.4095***</b>	-0.2845	<b>0.4649***</b>	-0.3668
ses.β.MPD.new	<b>0.4257***</b>	0.1889	<b>0.389***</b>	0.02191
ses.β.MPD.old	<b>0.5171**</b>	0.1785	<b>0.4956**</b>	0.05594
ses.β.MNTD.all	<b>0.2949***</b>	0.04031	<b>0.2932***</b>	-0.02301
ses.β.MNTD.ref	<b>0.3418*</b>	-0.4219	<b>0.4087**</b>	-0.474
ses.β.MNTD.new	<b>0.3277***</b>	<b>0.2362***</b>	<b>0.2386***</b>	0.0497
ses.β.MNTD.old	<b>0.4277**</b>	0.07247	<b>0.4226**</b>	-0.0001

Mann-Whitney pairwise results indicate significant differences in βRC between forested and recently deforested streams ( $P=0.047$ ), no differences were detected between forested and olden deforested streams, and between recent and olden deforested streams. Regarding, we detected functional turnover, significant differences in βMPD between forested and recent deforested streams ( $P < 0.001$ )

and between forested and olden deforested streams ( $P=0.008$ ), in  $\beta$ MNTD, significant differences were detected between forested and recent deforested streams ( $P=0.003$ ) and recent and olden deforested streams ( $P=0.02$ ). For  $\beta$ MPPD, no significant differences were detected between recent and olden deforested streams, and between forested and olden deforested streams, considering  $\beta$ MNTD.

The results of MRM and commonality analysis indicate that both local- and watershed- scale variables explained the variation in the environmental-turnover relationship for impacted streams, and only local scale variables explained the variation in the environmental-turnover relationship for forested streams. However, environmental-turnover relationship was mostly weak for all 75 streams ( $R^2 \leq 0.14$ ;  $P < 0.001$ ; Fig. 4.3; Supporting Information S5.3) and for recently deforested streams ( $R^2 \leq 0.20$ ;  $P < 0.001$ ; Fig. 4.3; Supporting Information S5.3). On the other hand, forested streams have presented a stronger environmental-turnover relationship than the set of all and recently deforested streams ( $R^2 \geq 0.20$ ;  $P < 0.001$ ; Fig. 4.3; Supporting Information S5.3). The olden deforested streams presented a variation on environmental-turnover relationship; this set of streams presented a weak relationship for the taxonomic facet ( $R^2 = 0.11$ ;  $P < 0.001$ ; Fig. 4.3; Supporting Information S5.3), contrasting to the stronger relationship observed for the functional facet ( $R^2 \geq 0.26$ ;  $P < 0.001$ ; Fig. 4.3; Supporting Information S5.3).



**Figure 4.3.** Explained variation in turnover metrics partitioned by MRM and associated commonality analysis into pure local, shared and pure catchment components. RC = Raup-Crick; MPD = mean pairwise distance; MNTD = mean nearest taxon distance; all = All sampled sites; ref = Forested sampled sites; new = Recently deforested sampled sites; old = Oldly deforested sampled sites.

## Discussion

According to our results, the turnover was different than expected by chance, indicating that deterministic processes are structuring this stream fish community. Although we found a high species turnover, functional traits turnover was lower than the expected by the species turnover. Taxonomically, the communities are dissimilar from each other, but they are functionally similar, suggesting that the species turnover is occurring mainly among functionally equivalent species. We also observed meaningful differences among the three stream groups, verifying that the recently deforested streams presented higher mean values for species turnover rates than the forested and olden deforested streams, but regarding the functional turnover metrics, recently deforested presented lower mean values than the forested and olden deforested streams, partially confirming our first hypothesis. In turn, we confirmed our second hypothesis; the turnover among forested streams was driven only by local-scale environmental variables, while in deforested streams the turnover was driven both by local- and watershed-scale environmental variables (see Supporting Information S3.3).

The information given by abundance data for taxonomic turnover (communities more different than expected by chance) is indicating that the dominant species differs between sites (Püttker et al. 2015; Siqueira et al. 2015). If we consider that the resource gradients or patch types generate differences in the local demography of species (Leibold et al. 2004), it is expected that each site

had unique levels of species abundances (Janzen & Schoener 1968; Tucker et al. 2016).

The higher species turnover found in recently deforested sites may be associated to the emerging of novel niches, due to the side effects of deforestation beginning (i.e., changes in flow complexity, depth, substrate composition, stream banks stability, emerging of marginal grasses), but which still retains relict structures and microhabitats found in pristine forest streams (i.e., fine roots, litter packs, branches, and trunks derived from the riparian forest) (Pérez-Mayorga et al. 2017). This novel environmental scenario found in this group of streams would be allowing the beginning of colonization and establishment process of tolerant species while the sensitive species still are occupying the relictual habitat structure from the previous condition, and probably this is one reason to find the highest species richness in this group. Applying a deconstructive approach based on site occupancy by fish species to study the same stream system, Pérez-Mayorga et al. (2017) identified two processes structuring this metacommunity: while intermediate species are explained only by dispersal-based processes, the satellite species are explained mainly by niche-based but also by dispersal-based processes. It is interesting to observe that inside these two groups we found both deforestation sensitive and tolerant species, so the presence of a fish species in a deforested site is independent of its dispersal or colonization ability, but these findings reinforce this possibility of a colonization-establishment process occurring in the deforested streams.

In general, land use intensification reduces the communities' dissimilarity, leading to the biotic homogenization process in which sensitive species will be gradually lost when tolerant species became more abundant (see Chapter 2; Brejão et al. 2018, Chapter 1). Surprisingly, the taxonomic turnover did not differ between forested and olden deforested streams, and both were lower than that found in recently deforested streams. According to landscape divergence hypothesis (Laurance et al. 2007), disturbed areas are likely to diverge in species composition because of differences in the effects of disturbance, or in how disturbances processes interact with underlying differences in environmental heterogeneity (Arroyo-Rodríguez et al. 2013).

Usually, forested sites have higher changes in species abundance ( $\beta$ -diversity) than agricultural landscapes (Karp et al. 2012; Siqueira et al. 2015; Solar et al. 2015). Agricultural intensification may lead to a higher  $\beta$ -diversity in higher  $\beta$ -diversity in high-intensity agriculture patches than that found on forest and low-intensity agriculture patches (Karp et al., 2012). Nevertheless, since each fish species response to the extent and time since deforestation is distinct, where sensitive species abundance decreases and tolerant species abundance increases over the time since the impact (Brejão et al. 2018, Chapter 1). The abundance structure (see the abrupt abundance increase of *Serrapinnus microdon* and *S. notomelas* on recently deforested streams, Supporting Information S1.3) on recently deforested streams could be facing a non-stable state, driven by the dynamic filtering of environmental conditions (Connell 1978; Leibold et al. 2004). It may be affecting the species turnover on this transitional situation

between different stable states (forest and old deforestation), which also could explain the weak relationship between species/traits and the environment detected in this stream group. On the other hand, in forested and olden deforested streams either by disturbance absence or by disturbance consolidation, the abundance structure (i.e., for forested streams see *Hyphessobrycon agulha* and *Gymnorhamphichthys petiti*, and for olden deforested streams see *Bryconops caudomaculatus* and *Creagrutus petilus*, Supporting Information S1.3). Consequently, the species turnover may be more stable when compared with recently deforested streams.

Functional traits turnover was lower than the expected by the species turnover, indicating that variation between two communities occurs mainly between closely functional species (Swenson et al. 2011; Swenson 2014) and determined by the environment. Large overlaps between fish assemblages functional space could explain the low level of functional turnover, with frequent species replacements occurring mostly between functionally redundant species (Villéger et al. 2013). In our assemblage, two species embracing singular sets of traits (*Farlowella* cf. *oxyryncha* and *Gymnorhamphichthys petiti*) are shared by the three groups of streams and are located in the extremes of the functional space (see Supporting Information S6.3), having a high contribution to functional diversity, especially in deforested streams (Bordignon 2017). The morphological singularities of these species allow them to explore specific niches and structures closely related to the riparian forest.

Due to its skinny and elongated body, together with a brown coloration, *Farlowella* species resemble narrow tree branches (Covain & Fisch-Muller 2007), where usually they were foraging, grazing the periphyton (Brejão et al. 2013). *Gymnorhamphichthys petiti* is a species with psammophilic habit, using its long snout and electric field to probe sandy and litter substrates, looking for small invertebrates during the night and buries itself in the sandy substrate during the day (Zuanon et al. 2006; Brejão et al. 2013). The presence of these species in all groups of streams is probably buffering the overlap of functional space between fish communities, explaining low rates of functional turnover when we have high rates of taxonomic turnover.

Regarding the environmental variables related to the turnover, it is interesting that turnover metrics are mainly related to instream habitat complexity indicators (i.e., litter packs, fine roots, marginal trees) and forest quality in the watershed and riparian buffer scales. Landscape modifications can influence streams environmental characteristics (Gorman & Karr 1978; Cruz et al. 2013; Siqueira et al. 2015), affecting many of the ecological processes occurring in streams (Wright & Flecker 2004; Paula et al. 2011). Maintaining pristine forest remnants associated with landscape complexity and connectivity could extend the prevalence of sensitive species with singular sets of traits in this fish community keeping the integrity of fish communities' diversity and functionality.

The studied fish assemblage still presents a diverse regional pool with a large proportion of rare species (see Bordignon 2017) but, apparently, the rare species turnover has a minor contribution to the functional turnover. However, in

assemblages with a simplified regional pool (Casatti et al. 2015; Zeni et al. 2017), composed mostly by tolerant species with a common set of traits playing very similar functional roles (Flynn et al. 2009; Mouillot et al. 2013; Casatti et al. 2015), rare species are, coincidentally, those species with specific sets of traits, and that has a high weight in the functional turnover of these communities (Roa-Fuentes 2016).

In summary, this deconstructive approach, grouping streams by landscape dynamic allow us to better detect the effects of deforestation on fish assemblage taxonomic and functional turnover, suggesting that, besides being scale-dependent (Karp et al. 2012), turnover patterns could be dynamic-dependent.

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### Supporting Information S1.3

Fish species included in this study, frequency of occurrence (Occur.), and total abundances (Abund.) for each stream group. Classification follows Reis et al. (2003); except Serrasalminae follows Calcagnotto et al. (2005).

TAXON	codes	Forest		Recent deforestation		Old deforestation	
		Occur.	Abund.	Occur.	Abund.	Occur.	Abund.
<b>CHARACIFORMES</b>							
<b>Parodontidae</b>							
<i>Parodon nasus</i>	parnas	-	-	2	3	1	1
<b>Curimatidae</b>							
<i>Curimatopsis macrolepis</i>	curmac	1	6	-	-	-	-
<i>Cyphocharax plumbeus</i>	cypplu	-	-	1	1	-	-
<i>Cyphocharax spiluropsis</i>	cypspi	-	-	11	37	3	3
<i>Steindachnerina cf. dobula</i>	stedob	-	-	1	4	-	-
<i>Steindachnerina fasciata</i>	stefas	-	-	7	38	7	19
<i>Steindachnerina guentheri</i>	stegue	-	-	1	3	-	-
<b>Prochilodontidae</b>							
<i>Prochilodus nigricans</i>	pronig	-	-	1	1	-	-
<b>Anostomidae</b>							
<i>Anostomus ternetzi</i>	anoter	-	-	2	5	-	-
<i>Leporinus friderici</i>	lepfri	3	3	9	27	6	16
<b>Crenuchidae</b>							
<i>Characidium aff. gomesi</i>	chagom	-	-	-	-	2	7
<i>Characidium aff. zebra</i>	chazeb	10	100	25	432	19	230
<i>Characidium sp.</i>	chasp	-	-	3	8	-	-
<i>Elachocharax pulcher</i>	elapul	7	77	2	2	-	-
<i>Microcharacidium aff. weitzmani</i>	micwei	3	9	2	2	4	27
<i>Melanocharacidium dispilomma</i>	meldis	-	-	1	1	-	-
<i>Melanocharacidium pectorale</i>	melpec	1	1	-	-	-	-
<i>Microcharacidium sp.</i>	micsp	3	50	-	-	-	-
<b>Hemiodontidae</b>							
<i>Hemiodus unimaculatus</i>	hemuni	-	-	2	2	-	-
<b>Gasteropelecidae</b>							
<i>Carnegiella strigata</i>	carstr	4	27	2	11	1	2
<b>Characidae</b>							
<i>Amazonspinther dalmata</i>	amadal	-	-	-	-	1	7
<i>Astyanax cf. bimaculatus</i>	astbim	1	1	8	62	10	45
<i>Astyanax cf. maximus</i>	astmax	3	11	-	-	4	7
<i>Astyanax maculisquamis</i>	astmac	-	-	1	1	2	42
<i>Bario steindachneri</i>	barste	2	3	-	-	-	-
<i>Brachychalcinus copei</i>	bracop	4	10	17	105	13	32
<i>Bryconella pallidifrons</i>	brypall	8	695	-	-	-	-
<i>Bryconops caudomaculatus</i>	brycau	6	20	23	429	21	463
<i>Bryconops piracolina</i>	brypir	-	-	1	23	-	-
<i>'Cheirodon' troemneri</i>	chetro	-	-	2	62	-	-
<i>Creagrutus petilus</i>	crepet	8	39	21	314	19	668
<i>Hemigrammus aff. ocellifer</i>	hemoce	4	20	4	14	3	28
<i>Hemigrammus bellotti</i>	hembel	3	143	2	9	-	-
<i>Hemigrammus melanochrourus</i>	hemmel	5	364	2	187	4	867
<i>Hemigrammus neptunus</i>	hemnep	1	4	2	55	1	1
<i>Hemigrammus sp.</i>	hemsp	1	14	-	-	-	-
<i>Hyphessobrycon aff. heterorhabdus</i>	hyphet	8	103	1	31	2	10
<i>Hyphessobrycon agulha</i>	hypagu	7	823	4	209	4	99

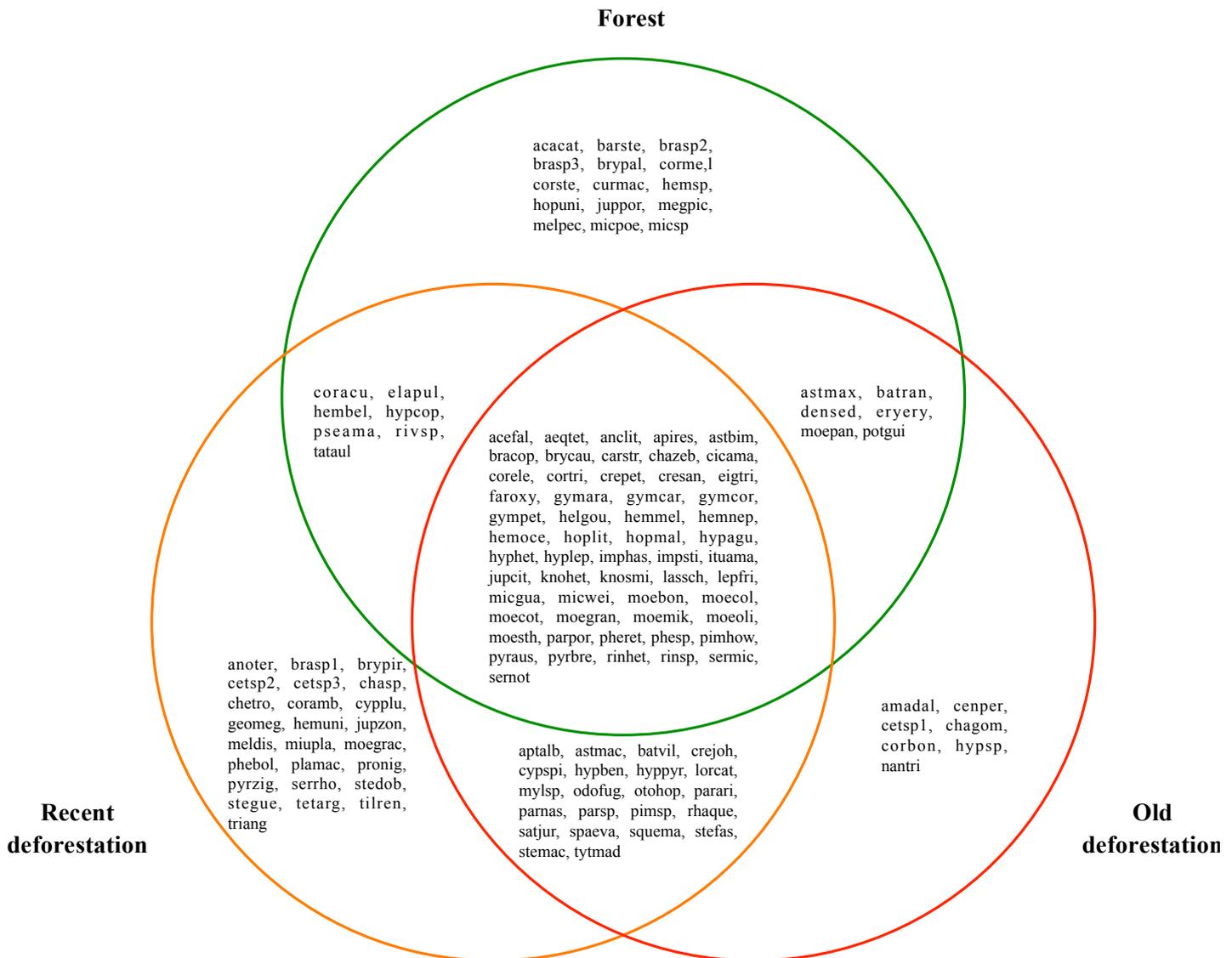
TAXON	codes	Forest		Recent deforestation		Old deforestation	
		Occur.	Abund.	Occur.	Abund.	Occur.	Abund.
<i>Hyphessobrycon bentosi</i>	hypben	-	-	5	69	3	109
<i>Hyphessobrycon copelandi</i>	hycop	4	144	3	7	-	-
<i>Jupiaba citrina</i>	jupcit	1	1	7	66	11	206
<i>Jupiaba poranga</i>	juppor	4	9	-	-	-	-
<i>Jupiaba zonata</i>	jupzon	-	-	3	55	-	-
<i>Knodus cf. smithi</i>	knosmi	4	61	18	340	13	426
<i>Knodus heteresthes</i>	knohet	2	583	4	99	6	54
<i>Microchemobrycon guaporensis</i>	micgua	4	34	8	127	2	5
<i>Moenkhausia aff. gracilima</i>	moegra	-	-	1	1	-	-
<i>Moenkhausia cf. bonita</i>	moebon	1	2	4	230	2	107
<i>Moenkhausia collettii</i>	moecl	7	176	21	1489	14	259
<i>Moenkhausia cotinho</i>	moecl	3	26	11	187	2	46
<i>Moenkhausia grandisquamis</i>	moegran	4	7	1	2	2	2
<i>Moenkhausia mikia</i>	moecl	3	31	6	67	2	7
<i>Moenkhausia oligolepis</i>	moeoli	8	100	25	144	16	86
<i>Moenkhausia pankilopteryx</i>	moecl	1	1	-	-	2	18
<i>Moenkhausia sthenosthoma</i>	moecl	1	4	5	14	1	23
<i>Odontostilbe fugitiva</i>	odofug	-	-	5	288	1	19
<i>Phenacogaster retropinnus</i>	pheret	7	32	21	242	14	112
<i>Serrapinnus microdon</i>	sermic	2	9	17	1114	11	778
<i>Serrapinnus aff. notomelas</i>	sernot	1	1	20	2470	13	1171
<i>Tetragonopterus argenteus</i>	tetarg	-	-	1	2	-	-
<i>Triportheus angulatus</i>	triang	-	-	2	2	-	-
<i>Tytocharax madeirae</i>	tytmad	-	-	2	22	2	10
<b>Serrasalminae</b>							
<i>Myleus sp.</i>	mylsp	-	-	2	5	3	7
<i>Serrasalmus rhombeus</i>	serrho	-	-	1	1	-	-
<b>Acestrorhynchidae</b>							
<i>Acestrorhynchus falcatus</i>	acefal	1	1	1	1	1	1
<b>Erythrinidae</b>							
<i>Erythrinus erythrinus</i>	eryery	6	9	-	-	2	2
<i>Hoplerythrinus unitaeniatus</i>	hopuni	3	3	-	-	-	-
<i>Hoplias malabaricus</i>	hopmal	12	46	14	27	10	15
<b>Lebiasinidae</b>							
<i>Nannostomus trifasciatus</i>	nantri	-	-	-	-	1	1
<i>Pyrrhulina cf. australis</i>	pyraus	15	156	5	27	4	10
<i>Pyrrhulina cf. brevis</i>	pyrbre	2	4	3	17	4	44
<i>Pyrrhulina cf. zigzag</i>	pyrzig	-	-	1	9	-	-
<b>SILURIFORMES</b>							
<b>Cetopsidae</b>							
<i>Denticetopsis seducta</i>	densed	1	1	-	-	2	3
<i>Helogenes gouldingi</i>	helgou	2	8	3	4	4	10
<b>Aspredinidae</b>							
<i>Pseudobunocephalus amazonicus</i>	pseama	7	33	1	4	-	-
<b>Trichomycteridae</b>							
<i>Ituglanis amazonicus</i>	ituama	10	92	6	10	4	10
<i>Miuroglanis platycephalus</i>	miupla	-	-	1	1	-	-
<i>Paracanthopoma sp.</i>	parsp	-	-	2	3	4	16
<b>Callichthyidae</b>							
<i>Corydoras acutus</i>	coracu	2	3	2	2	-	-
<i>Corydoras aff. ambiacus</i>	coramb	-	-	1	3	-	-
<i>Corydoras cf. bondi</i>	corbon	-	-	-	-	1	1

TAXON	codes	Forest		Recent deforestation		Old deforestation	
		Occur.	Abund.	Occur.	Abund.	Occur.	Abund.
<i>Corydoras cf. melanistius</i>	cormel	1	55	-	-	-	-
<i>Corydoras elegans</i>	corele	1	1	3	4	1	2
<i>Corydoras stenocephalus</i>	corste	2	5	-	-	-	-
<i>Corydoras trilineatus</i>	cortri	8	52	11	24	3	6
<i>Hoplosternum littorale</i>	hoplit	1	1	2	4	2	2
<i>Megalechis picta</i>	megpic	6	49	-	-	-	-
<b>Loricariidae</b>							
<i>Ancistrus lithurgicus</i>	anclit	6	26	12	117	8	147
<i>Farlowella cf. oxyrryncha</i>	faroxy	2	3	12	64	8	53
<i>Hypostomus pyrineusi</i>	hyppir	-	-	9	21	7	13
<i>Hypostomus</i> sp.	hypsp	-	-	-	-	1	1
<i>Lasiancistrus schomburgkii</i>	lassch	4	8	11	45	3	8
<i>Loricaria cataphracta</i>	lorcat	-	-	1	2	2	2
<i>Otocinclus hoppei</i>	otohop	-	-	7	91	3	28
<i>Parotocinclus aff. aripuanensis</i>	parari	-	-	2	15	4	9
<i>Rineloricaria heteroptera</i>	rinhet	4	5	23	99	13	60
<i>Rineloricaria</i> sp.	rinsp	1	1	2	4	1	1
<i>Spatuloricaria evansii</i>	spaeva	-	-	1	1	2	3
<i>Squaliforma emarginata</i>	squema	-	-	3	12	4	10
<b>Pseudopimelodidae</b>							
<i>Batrochoglanis cf. raninus</i>	batran	4	14	-	-	1	2
<i>Batrochoglanis villosus</i>	batvil	-	-	2	3	1	2
<i>Microglanis poecilus</i>	micpoe	1	1	-	-	-	-
<b>Heptapteridae</b>							
<i>Cetopsorhamdia</i> sp. 1	cetsp1	-	-	-	-	2	24
<i>Cetopsorhamdia</i> sp. 2	cetsp2	-	-	2	8	-	-
<i>Cetopsorhamdia</i> sp. 3	cetsp3	-	-	1	6	-	-
<i>Imparfinis cf. hasemani</i>	impphas	6	76	6	16	7	32
<i>Imparfinis stictonotus</i>	impsti	5	20	5	10	3	19
<i>Phenacorhamdia cf. boliviana</i>	phebol	-	-	3	4	-	-
<i>Phenacorhamdia</i> sp.	phesp	2	10	4	43	2	17
<i>Pimelodella cf. howesi</i>	pimhow	8	36	5	15	3	4
<i>Pimelodella</i> sp.	pimsp	-	-	4	6	4	5
<i>Rhamdia quelen</i>	rhaque	-	-	3	5	1	1
<b>Doradidae</b>							
<i>Acanthodoras cataphractus</i>	acacat	4	19	-	-	-	-
<b>Auchenipteridae</b>							
<i>Centromochlus cf. perugiae</i>	cenper	-	-	-	-	1	1
<i>Parauchenipterus porosus</i>	parpor	1	1	1	1	1	3
<i>Tatia aulopygia</i>	tataul	1	1	1	1	-	-
<b>GYMNOTIFORMES</b>							
<b>Gymnotidae</b>							
<i>Gymnotus aff. arapaima</i>	gymara	1	1	3	11	5	14
<i>Gymnotus carapo</i>	gymcar	1	1	5	28	4	7
<i>Gymnotus coropinae</i>	gymcor	10	54	3	10	2	17
<b>Sternopygidae</b>							
<i>Eigenmannia trilineata</i>	eigtri	1	1	11	136	7	59
<i>Sternopygus macrurus</i>	stemac	-	-	12	37	12	60
<b>Rhamphichthyidae</b>							
<i>Gymnorhamphichthys petiti</i>	gympet	12	180	12	62	7	45
<b>Hypopomidae</b>							
<i>Brachyhypopomus</i> sp. 1	braspl	-	-	2	2	-	-

TAXON	codes	Forest		Recent deforestation		Old deforestation	
		Occur.	Abund.	Occur.	Abund.	Occur.	Abund.
<i>Brachyhypopomus</i> sp. 2	brasp2	8	15	-	-	-	-
<i>Brachyhypopomus</i> sp. 3	brasp3	5	26	-	-	-	-
<i>Hypopygus lepturus</i>	hyplep	7	47	8	50	2	31
<b>Apteronotidae</b>							
<i>Apteronotus albifrons</i>	aptalb	-	-	2	2	2	4
<i>Platyurosternarchus macrostomus</i>	plamac	-	-	1	2	-	-
<b>CYPRINODONTIFORMES</b>							
<b>Rivulidae</b>							
<i>Rivulus</i> sp.	rivsp	1	3	1	1	-	-
<b>BELONIFORMES</b>							
<b>Belonidae</b>							
<i>Potamorrhaphis guianensis</i>	potgui	1	1	-	-	1	1
<b>PERCIFORMES</b>							
<b>Cichlidae</b>							
<i>Aequidens tetramerus</i>	aeqtet	8	17	19	66	12	116
<i>Apistogramma</i> cf. <i>resticulosa</i>	apires	12	453	6	66	5	44
<i>Cichlasoma amazonarum</i>	cicama	1	1	4	18	4	27
<i>Crenicichla johanna</i>	crejoh	-	-	1	1	1	1
<i>Crenicichla santosi</i>	cresan	6	8	18	110	16	45
<i>Geophagus megasema</i>	geomeg	-	-	1	1	-	-
<i>Satanoperca jurupari</i>	satjur	-	-	9	52	4	8
<i>Coptodon rendalli</i> *	tilren	-	-	2	2	-	-

### Supporting Information S2.3

Venn diagram indicating exclusive and shared species among each stream group. The codes for each species were depicted on Supporting Information S1.3.



### Supporting Information S3.3

Subset of environmental variables correlated (Pearson (r)) with the standard effect size (SES) of turnover for each stream gorup

Turnover metrics	Selected variables	r
Taxonomic		
ses.β.RC.all	TRE, DOX, DEP, MF_WSHD, EF_WSHD, LUI_WSHD	0.358
ses.β.RC.ref	FRO, SAN, DOX, CON, WID	0.650
ses.β.RC.new	CUR, DOX, CON, LUI_WSHD, LUI_BUFF	0.260
ses.β.RC.old	TRE, CUR, DOX, EF_WSHD, MF_BUFF	0.235
Functional		
ses.β.MPD.all	FRO, DOX, DEP, MF_WSHD, EF_WSHD, EF_BUFF	0.287
ses.β.MPD.ref	FRO, DOX, DEP	0.409
ses.β.MPD.new	LIT, EF_BUFF	0.426
ses.β.MPD.old	FRO, TRE, EF_WSHD, EF_BUFF	0.517
ses.β.MNTD.all	FRO, DEP, MF_WSHD, EF_WSHD	0.295
ses.β.MNTD.ref	FRO, DEP	0.342
ses.β.MNTD.new	FRO, LIT, DOX, DEP, EF_WSHD, EF_BUFF	0.328
ses.β.MNTD.old	FRO, LIT, EF_WSHD, EF_BUFF	0.501

**TRE:** Trees in stream banks; **FRO:** Submerged roots in stream banks; **LIT:** Litter packs on stream bed; **SAN:** Sand; **CUR:** Velocidade da água; **DOX:** Oxigênio dissolvido; **CON:** Conductivity; **DEP:** Depth; **WID:** Width; **MF\_WSHD:** Mature Forest proportion on watershed; **MF\_BUFF:** Mature Forest proportion on riparian buffer; **EF\_WSHD:** Effective Forest proportion on watershed; **EF\_BUFF:** Effective Forest proportion on riparian buffer; **LUI\_WSHD:** Land Use Intensity on watershed; **LUI\_BUFF:** Land Use Intensity on riparian buffer.

### Supporting Information S4.3

T-test results and mean values of taxonomic and functional turnover among sampled sites.

	Mean	95% confidence interval		<i>t</i>	df	<i>P</i>
		Lower	Upper			
Taxonomic						
ses.β.RC.all	0.75	0.71	0.79	36.49	74	< 0.001
ses.β.RC.ref	0.71	0.61	0.82	14.66	17	< 0.001
ses.β.RC.new	0.84	0.79	0.90	33.55	30	< 0.001
ses.β.RC.old	0.78	0.72	0.84	27.10	25	< 0.001
Functional						
ses.β.MPD.all	-1.15	-1.25	-1.05	-19.24	74	< 0.001
ses.β.MPD.ref	-0.72	-0.94	-0.49	-5.57	17	< 0.001
ses.β.MPD.new	-1.36	-1.49	-1.23	-18.10	30	< 0.001
ses.β.MPD.old	-1.18	-1.36	-1.01	-11.35	25	< 0.001
ses.β.MNTD.all	-0.74	-0.83	-0.64	-13.07	74	< 0.001
ses.β.MNTD.ref	-0.53	-0.76	-0.30	-4.05	17	< 0.001
ses.β.MNTD.new	-1.01	-1.12	-0.89	-14.50	30	< 0.001
ses.β.MNTD.old	-0.64	-0.81	-0.47	-6.34	25	< 0.001

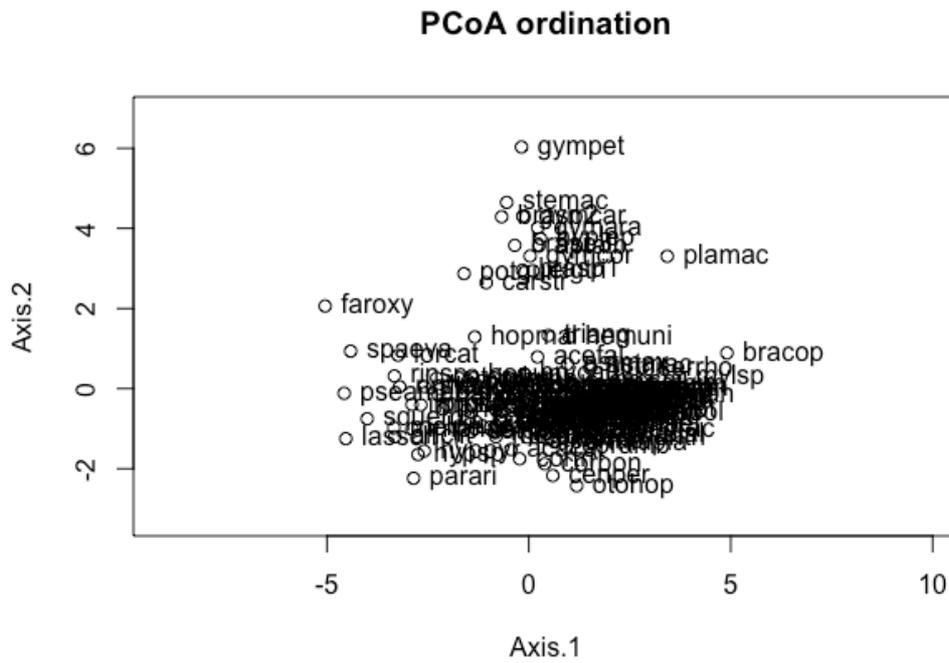
### Supporting information S5.3

Relative importance of environmental factors, grouped on local and watershed scale on turnover rates. This table includes multiple  $R^2$ , P-value ( $P$ ), beta coefficients ( $\beta$ ), structure coefficients ( $r_s$ ), each predictor's total unique (U), total common (C), and total variance (Total) in the regression equation. When the number of regressors was  $< 2$  commonality analysis was not conducted.

Turnover facet	Scale	$\beta$	$P$	$r_s$	Commonality analysis		Total
					U	C	
Taxonomic							
ses. $\beta$ .RC.all ( $R^2=0.14$ ; $P<0.001$ )	Local	0.165	$<0.001$	0.659	0.025	0.036	0.061
	Watershed	0.226	$<0.001$	0.815	0.045	0.049	0.094
ses. $\beta$ .RC.ref ( $R^2=0.44$ ; $P<0.001$ )	Local	0.600	$<0.001$	-	-	-	0.435
ses. $\beta$ .RC.new ( $R^2=0.08$ ; $P<0.001$ )	Local	0.123	0.007	0.563	0.015	0.012	0.027
	Watershed	0.162	$<0.001$	0.762	0.023	0.027	0.049
ses. $\beta$ .RC.old ( $R^2=0.11$ ; $P<0.001$ )	Local	0.132	0.015	-	-	-	0.107
Functional							
ses. $\beta$ .MPD.all ( $R^2=0.09$ ; $P<0.001$ )	Local	0.193	$<0.001$	0.664	0.037	0.003	0.040
	Watershed	0.229	$<0.001$	0.770	0.049	0.005	0.054
ses. $\beta$ .MPD.ref ( $R^2=0.28$ ; $P<0.001$ )	Local	0.449	$<0.001$	-	-	-	0.280
ses. $\beta$ .MPD.new ( $R^2=0.20$ ; $P<0.001$ )	Local	0.235	$<0.001$	0.563	0.055	0.01	0.064
	Watershed	0.374	$<0.001$	0.854	0.113	0.035	0.148
ses. $\beta$ .MPD.old ( $R^2=0.31$ ; $P<0.001$ )	Local	0.273	$<0.001$	0.537	0.074	0.014	0.088
	Watershed	0.465	$<0.001$	0.87	0.19	0.041	0.232
ses. $\beta$ .MNTD.all ( $R^2=0.09$ ; $P<0.001$ )	Local	0.198	$<0.001$	0.639	0.039	-0.0009	0.038
	Watershed	0.24	$<0.001$	0.762	0.055	-0.0002	0.054
ses. $\beta$ .MNTD.ref ( $R^2=0.32$ ; $P<0.001$ )	Local	0.371	$<0.001$	-	-	-	0.315
ses. $\beta$ .MNTD.new ( $R^2=0.11$ ; $P<0.001$ )	Local	0.141	$<0.01$	0.781	0.014	0.053	0.067
	Watershed	0.194	$<0.001$	0.858	0.029	0.052	0.080
ses. $\beta$ .MNTD.old ( $R^2=0.26$ ; $P<0.001$ )	Local	0.282	$<0.001$	0.725	0.074	0.063	0.137
	Watershed	0.383	$<0.001$	0.832	0.123	0.057	0.180

### Supporting information S6.3

Principal coordinates analysis (PCoA) representing functional distance among species. The codes for each species were depicted on Supporting Information S1.3.



## Synthesis

In Amazon, the deforestation is the primary environmental filter driving habitat loss and fragmentation affecting terrestrial and aquatic ecosystems. However, the effects of this impact on neotropics are commonly assessed through community and landscape snapshots (a single measure in the present time), missing the historical legacy-effects of land use changes on stream fish communities.

The effects of deforestation accumulate over the time since its beginning, affecting fish populations in distinct ways. The time and the direction of each species response to this impact will define the community integrity evaluated on the snapshot. Therefore, the information given only by these snapshots offers a narrow contribution for conservation plannings. By being able to measure the legacy-effects of impact, it may be possible to project if the environment has already reached a new stable state, or if it still is in the adjustment of richness and abundance process. To drive the communities back to a stable state, resembling the pristine conditions, is necessary much time and energy. However, if the disturbance is in progress and the community is unstable, facing the adjustment processes, it may be possible to decelerate the process of state change (i.e., which can be from a diverse and equitable to a homogeneous and dominant community) through ecological restoration.

However, the effects of deforestation, even occurring in low intensity (i.e., reduced impact logging projects), can promote immediate changes in fish communities richness and abundance, and these changes remain by years after the

impact. The main deforestation mode in Amazon is the typical slash-and-burn in extensive areas. Therefore, the effects of such deforestation on the fish communities structure are very intense and interfere in the way which deforestation dynamics is structuring the diversity patterns of the fish assemblages.

Including the time scale as a variable in stream ecology research is essential to better comprehend the legacy effects of land use changes on fish assemblages and instream habitat structure. Notwithstanding, it is indispensable to maintain, on a regional scale, forest remnants that may reflect this history. Even if large areas are converted into production systems, these remnants can make it possible to recover the past scenarios. Without this minimum, what is remaining allows us only tell a very poorly understood story.

By adding the temporal layer to analyze the initial stage of land use changes in this portion of Amazon, it was possible to verify the extreme sensitivity of fish assemblage to deforestation. With our results, it is reasonable to consider stream fish as a flag group to be included on conservation plannings aiming to reduce the effects of biodiversity loss on a regional scale.