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"An investment in knowledge pays the best interest"

Benjamin Franklin

Aos meus pais, Luiz Carlos e Sueli e a minha irmã, Ana Carla

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RESUMO

Mudanças ambientais associadas a conversão da vegetação nativa em agroecossistemas são consideradas um poderoso motor de homogeneização da ictiofauna no Antropoceno. Assim, é primordial entender como subsequentes alterações em ambientes já impactados podem afetar o ambiente aquático e as assembleias de peixes. Avaliar se e como subsequentes mudanças ambientais nas escalas regional (microbacia) e local (descritores do canal e ecótone) podem afetar a estrutura taxonômica e funcional da ictiofauna e a diversidade beta espaço-temporal de riachos em agroecossistemas. Trinta e oito riachos foram amostrados em dois períodos (2003 e 2013). Nós quantificamos o grau de mudança nas escalas regional e local e, posteriormente correlacionamos com o grau de mudanças taxonômicas e funcionais das assembleias e com a diversidade beta espaço-temporal. O grau de mudanças ambientais (escalas regional e local) não foi correlacionado com as mudanças biológicas ou com a diversidade beta. A diversidade beta espacial permaneceu inalterada entre os períodos, enquanto a diversidade beta temporal foi menor do que esperado ao acaso. Os resultados obtidos nesse estudo reforçam que os padrões atuais das assembleias de peixes podem estar associados com o filtros ambientais antigos decorrente do desmatamento inicial em uma região. É provável que esses filtros foram responsáveis pela seleção não randômica de espécies generalistas quanto ao uso do habitat capazes de resistir e se adaptar a uma grande diversidade de mudanças ambientais. Apesar da ausência de um inventário da fauna antes do desmatamento, a maioria das espécies de peixes amostradas nesse estudo é comum e regionalmente distribuída, enquanto poucas espécies raras estão restritas a poucos riachos. Nesse contexto, o grau de mudança ambiental observado, provavelmente, não foi suficiente para causar mudanças biológicas da ictiofauna. Além disso, há evidencias de um *time lag* na região, assim os efeitos da rápida conversão da paisagem na estrutura física do riacho e nas assembleias de peixes ainda não está evidente.

Palavras-chaves: Agroecossistemas, mudanças temporais, biocombustíveis, *time lag*, filtros ambientais antigos, *turnover*, homogeneização da ictiofauna, pastagem e cana-de-açúcar.

ABSTRACT

Environmental changes associated with conversion of native vegetation to agroecosystems are considered a powerful drive of fish fauna homogenization in the Anthropocene. Thus, it is essential to understand how subsequent alterations to already altered ecosystems may further affect assemblages. To evaluate whether and how further environmental changes at regional (watershed) and local (instream habitat and ecotone) scales can influence taxonomic and functional fish structure and spatial-temporal beta diversity in agroecosystem streams. Thirty-eight agroecosystem streams were sampled 10 years apart (2003 and 2013). We quantified the degree of regional and local changes and correlated with the degree biological changes (taxonomic and functional structure) and with the spatial-temporal beta diversity between the two periods. The degree of environmental changes at regional and local scales were not correlated with the degree of taxonomic and functional structure changes or spatial-temporal beta diversity. Spatial beta diversity remained unchanged over time and temporal beta diversity was lower than expected by chance. We believe our results indicate that current patterns of fish assemblages in agroecosystem could be widely associated with the past deforestation and the environmental filtering. Probably, past environmental filters were responsible for the selection of habitat generalist species able to adapt and resist several types of environmental changes. Despite no pre-deforestation inventory in the region, most of our fish species are common and regionally distributed, while few rare species are restricted to few streams. In this context, it is possible that the degree of environmental changes observed in this study were not strong enough to cause further changes in fish fauna aspects. Moreover, we detected some evidence for a time lag response or legacy effects. Thus, the effects of rapid conversion of the landscape on instream habitat and biodiversity are presumably not yet fully evident. Unfortunately, this is happening in numerous aquatic systems, and immediate measures that effectively restore streams assemblages should be a priority.

Keywords: Agroecosystems, temporal changes, biofuel production, time lag response, past environmental filter, turnover, fish fauna homogenization, pasture and sugarcane.

GENERAL INTRODUCTION

Earth geology and climate have been strongly affected by human activities and some authors have been arguing that we are in a new geological time unit, called Anthropocene (Steffen *et al*, 2006; Waters *et al*., 2015). The imprint of human activities in the Anthropocene has changed not only geology and climate aspects, but also energy balance across ecosystems and species extinction rates (Steffen *et al*., 2006). Freshwater streams present a strong relationship with terrestrial surroundings and they are responsible for key ecological services, such as processing of organic matter, retention of nutrients and water provision (Allan, 2004). However, streams and their communities are among the most impaired ecosystems in the Anthropocene, wherein most of them have experienced past or current human induced changes (Carpenter *et al*., 2011). They suffer not only with water quality impoverishment (Buck *et al*., 2004; Poor & McDonnell, 2007) or physical habitat degradation (Allan, 2004; Casatti *et al*., 2006; Miserendino *et al*., 2011), but also biotic changes (Dudgeon *et al*., 2006).

Agroecosystems are product of the conversion of natural to agricultural landscapes (Conway, 1987) and are considered a main drive of physical and biological above-mentioned changes in streams (Leitão *et al*., 2017). According to Foley et al. (2005), more than 40% of all Earth surface is dominated by croplands and pasture and the primary focus of these agroecosystems is food (Wood, Sebastian & Scherr, 2000) or fuel production (Lapola *et al*., 2014). Stream physical habitat homogenization commonly associated with agroecosystems can jeopardize the occurrence and maintenance of sensitive fish species in regional species pool (Scott & Helfman, 2001). Due to the intense and widespread land use changes, fish communities' homogenization and defaunation is one of the main concerns in the Anthropocene (Rahel, 2002). The effects of land use change in streams species are usually detected by comparing 'altered' versus reference

conditions (i.e. sites with none or low historical and contemporary human impact). However, in many regions, human population growth and agriculture crops development were so profound and disorganized that true reference streams are often inexistent. For example, Baattrup-Pedersen *et al*. (2008) argued that reference streams in Europe are rare due to the long-term effects that land use has had on the stream and assemblage's integrity.

In this scenario, incorporating historical records from before land use change could be interesting for agroecosystem studies. However, because of the time since science started to be done, medium and long-term data are more prone to be available in temperate regions (e.g. Europe and USA). In some tropical regions, these historical records are quite recent and worse, most of them are available only after the first human impacts have started. Thus, it is impossible to determine for sure how many or which species have existed some tropical lands. For all these reasons, working with agroecosystems streams under an intense and long historic of human impact could be tricky. Moreover, many systems are experiencing continuing or compounding environmental changes. For example, many watersheds have experienced multiple types of land use change over time (Martinelli & Filoso, 2008; Lapola *et al*., 2010; Rudorff *et al*., 2010), but little is known about how agroecosystems assemblages can respond to continuous land use changes through time.

Streams from northwest of São Paulo state, Brazil, are (unfortunately) a good ecological example of the consequences of intense and old land use change. According to Silva *et al*. (2007), deforestation in this region started in the beginning of 20th century and, nowadays, only less than 4% of the native semi-deciduous seasonal forest (Atlantic Forest) remains in small and isolated fragments (Nalon *et al*., 2008). Deforestation started with coffee crops, followed by citrus cultivation and livestock grazing (Monbeig, 1988).

In the last decade, sugarcane crops have been replacing many other cultures, especially pasture (Verdade *et al*., 2012; Lapola *et al*., 2014). Streams in this area usually present high physical habitat homogenization with channel dominated by runs, marginal grasses and sandy substrate (Casatti *et al*., 2006; Casatti *et al*., 2009; Casatti *et al*., 2012). Fish assemblages here are composed by numerous generalist species with broadly geographic distribution (Casatti *et al*., 2009; Casatti *et al*., 2015; Roa-Fuentes & Casatti, 2017). Due to high deforestation and continuous land use change, all streams in this area show some degree of human influence and true reference sites are missing. Moreover, deforestation is much more older than the first fish sample registered for this area (Garutti, 1988). Thus, historical records are also unavailable for the area.

Besides these difficulties, streams from northwest of São Paulo state could help us to elucidate the responses of biodiversity in dynamic agroecosystems, specifically those shifting towards biofuels production. Due to increasing global demand for bioenergy, land use change to biofuels should continue to be a significant dimension of environmental change for the foreseeable future (Lapola *et al*., 2014). Thus, it is primordial understand how subsequent alterations to already altered ecosystems may further affect assemblages. Based on assemblage's response, we can suggest de more effective measures to conserve, manage and restore aquatic ecosystems. This is particularly important if we consider agroecosystems prevalence on Earth surface and the recent interest for an agriculture more sustainable, which can grant not only human demand for food and water, but also preserve species and vital ecosystems functions. Considering all of this, our aims were to evaluate whether and how environmental changes at regional and local scales have influenced taxonomic and functional fish structure (Chapter 1) and spatial-temporal beta diversity of species and functional groups

(Chapter 2). The chapters were prepared according to the Freshwater Biology instructions for authors.

REFERENCES

Allan J.D. (2004) Landscape and riverscape: The influence of land use on stream ecosystems. *Annual Review of Ecology, Evolution, and Systematics*, **35**, 257–284.

Baattrup-Pederson A., Springe G., Riis T., Larsen S.E., Sand-Jensen K. & Larsen L.M.K. (2008) The search for reference conditions for stream vegetation in northern Europe. *Freshwater Biology*, **53**, 1890–1901.

Buck O., Niyogi D.K. & Townsend C.R. (2004) Scale-dependence of land use effects on water quality of streams in agricultural catchments. *Environmental Pollution*, **130**, 287–299.

Carpenter S.R., Stanley E.H. & Zanden M.J.V. (2011) State of the world's freshwater ecosystems: physical, chemical, and biological changes. *Annual Review of Environmental and Resources*, **36**, 75–99.

Casatti L., Langeani F., Silva A.M. & Castro R.M.C. (2006) Stream fish, water and habitat quality in a pasture dominated basin, southeastern Brazil. *Brazilian Journal of Biology*, **66**, 681–696.

Casatti L., Ferreira C.P. & Carvalho F.R. (2009) Grass-dominated stream sites exhibit low fish species diversity and guppies dominance: an assessment on two tropical pasture river basins. *Hydrobiologia*, **632**, 273–283.

Casatti L., Teresa, F. B., Gonçalves-Souza, T., Bessa, E., Manzotti, A. R., Gonçalves, C. S. & Zeni, J. O. (2012) From forests to cattail: how does the riparian zone influence stream fish? *Neotropical Ichthyology*, **10**, 205–214.

Casatti L., Teresa F.B., Zeni J.O., Ribeiro M.D., Brejão G.L. & Ceneviva-Bastos M. (2015) More of the same: high functional redundancy in stream fish assemblages from tropical agroecosystems. *Environmental Management*, **55**, 1300–1314.

Dudgeon D., Arthington A.H., Gessner M.O., Kawabata Z.-I., Knowler D.J., Lévêque C., Naiman R.J., Prieur-Richard A.-H., Soto D., Stiassny M.L.J. & Sullivan C.A. (2006) Freshwater biodiversity: importance, threats, status and conservation challenges. *Biological Reviews*, **81**, 163–182.

Foley J.A., DeFries R., Asner G.P., Barford C., Bonan G., Carpenter S. P., Chapin F.S., Coe M.T., Daily G.C., Gibbs H.K., Helkowski J.H., Holloway T., Howard E.A., Kucharik C.J., Monfreda C., Patz J.A., Prentice I.C., Ramankutty N. & Snyder P.K. (2005) Global consequences of land use. *Science*, **309**: 570–574.

Garutti V. (1988) Distribuição longitudinal da ictiofauna em um córrego da região Noroeste do Estado de São Paulo, bacia do rio Paraná. *Revista Brasileira de Biologia*, **48**, 747–759.

Lapola D.M., Schaldach R., Alcamo J., Bondeau A., Koch J., Koelking C. *et al*. (2010) Indirect land-use changes can overcome carbon savings from biofuels in Brazil. *Proceedings of the National Academy of Sciences*, **107**, 3388–3393.

Lapola D.M., Martinelli L.A., Peres C.A., Ometto J.P.H.B., Ferreira M.E., Nobre C.A. *et al*. (2014) Pervasive transition of the Brazilian land-use system. *Nature Climate Change*, **4**, 27–35.

Leitão, R.P., Zuanon J., Mouillot D., Leal C.G., Hughes R.M., Kaufmann P.R. *et al*. (2017) Disentangling the pathways of land use impacts on the functional structure of fish assemblages in Amazon streams. *Ecography*, **40**, 1–13.

Martinelli L.A. & Filoso S. (2008) Expansion of sugarcane ethanol production in Brazil: environmental and social challenges. *Ecological Applications*, **18**, 885–898.

Miserendino M.L., Casaux R., Archangelski M., Prinzio C.Y.D., Brand C. & Kutschker A.M. 2011. Assessing land-use effects on water-quality, in-stream, riparian ecosystems and biodiversity in Patagonian northwest streams. *Science of the Total Environmental*, **409**, 612–624.

Monbeig P. (1998) *Pioneiros e fazendeiros de São Paulo*. São Paulo, Hucitec, 392p.

Nalon M.A., Mattos I.F.A. & Franco G.A.D.C. (2008) Meio físico e aspectos da fragmentação da vegetação. In: *Diretrizes Para a Conservação e Restauração da Biodiversidade no Estado de São Paulo* (Eds R.R. Rodrigues & V.L.R. Bononi), pp. 17–21. Secretaria do Meio Ambiente e Instituto de Botânica, São Paulo.

Poor C.J. & McDonnell J.J. (2007) The effects of land use on stream nitrate dynamics. *Journal of Hydrology*, **332**, 54–68.

Pusey B.J. & Arthington A.H. (2003) Importance of riparian zone to the conservation and management of freshwater fishes: a review. *Marine and Freshwater Research*, **54**, 1–16.

Rahel F.J. (2002). Homogenization of freshwater faunas. *Annual Review of Ecology, Evolution, and Systematics*, **33**, 291–315

Roa-Fuentes C.A. & Casatti L. (2017). Influence of environmental features at multiple scales and spatial structure on stream fish communities in a tropical agricultural region. *Journal of Freshwater Ecology*, **32**, 273–287.

Rudorff B.F.T., Aguiar D.A., Silva W.F., Sugawara L.M., Adami M. & Moreira M.A. (2010) Studies on the rapid expansion of sugarcane for ethanol production in São Paulo state (Brazil) using Landsat data. *Remote Sensing*, **2**, 1057–1076.

Scott M.C. & Helfman G.S. (2001) Native invasions, homogenization, and the mismeasure of integrity of fish assemblages. *Fisheries*, **26**, 6–15.

Silva A.M., Casatti L., Alvares C.A., Leite A.M., Martinelli L.A. & Durrant S.F. (2007) Soil loss risk and habitat quality in some streams of a Meso-Scale River Basin. *Scientia Agricola*, **64**, 336–343.

Steffen W., Grinevald J., Crutzen P. & McNeill J. (2011). The Anthropocene: conceptual and historical perspectives. *Philosophical Transactions of Royal Society A*, **369**, 842–867.

Verdade L.M., Gheler-Costa C., Penteado M. & Dotta G. (2012) The impacts of sugarcane expansion on wildlife in the state of São Paulo, Brazil. *Journal of Sustainable Bioenergy Systems*, **2**, 138–144.

Waters C.N., Zalasiewicz J., Summerhayes C., Barnosky A.D., Poirier C., Gałuszka A. *et al*. (2016) The Anthropocene is functionally and stratighaphically distinct from Holocene. *Science*, **351**, 137–147.

Wood S., Sebastian K. & Scherr S. J. (2000) *Pilot Analysis of Global Ecosystems: Agroecosystems*. International Food Policy Research Institute and World Resources Institute, Washington, DC.

CHAPTER 1

**EFFECTS OF PASTURE CONVERSION TO SUGARCANE FOR BIOFUELS
PRODUCTION ON STREAM FISH ASSEMBLAGES IN TROPICAL
AGROECOSYSTEMS**

Summary

1. One of the main causes of biodiversity loss in aquatic ecosystems is instream habitat change associated with land use change in the watershed. Recently, biofuels production is an important driver of land use change in existing agricultural watershed. Conceptually, biofuels such as sugarcane (*Saccharum* spp.) can provide a sustainable alternative to fossil fuels, but little is known about the consequences of biofuels production for biodiversity. Thus, our aim was to investigate if and how the degree of environmental changes at regional and local scales can influence taxonomic and functional changes of fish assemblages in agroecosystem streams.

2. We quantified watershed land use, instream and riparian habitat, and fish assemblages in 38 agroecosystem streams with watersheds experiencing rapid expansion of sugarcane production, and calculated degree of environmental change and corresponding response in taxonomic and functional structure of stream fish assemblages. We used a Procrustes analysis to test for correlation between land use and instream habitat and redundancy analysis to test for correlation between environmental factors and fish assemblage structure. We used linear regressions to test the hypothesis that degree of environmental change is correlated with magnitude of change in taxonomic and functional structure of fish assemblages.

3. Primary gradients of environmental change included conversion of pasture to sugarcane and reduction of stream width, depth and volume. Although watershed and instream attributes were not correlated within a year, we detected a legacy effect of land use on instream habitat. Degree of environmental change was not significantly correlated with taxonomic or functional changes in stream fish assemblages, in most cases. However, abundances of resistant taxa increased whereas nektonic functional groups decreased, and the biological response also presented evidence of a legacy effect.

4. Our findings demonstrate that instream habitat and aquatic biodiversity in streams with agricultural watersheds undergoing rapid conversion to biofuels production likely will not experience the full magnitude of responses in the short term. Repeated sampling of sufficient duration to account for legacy effects or time lags is needed to test for effects of biofuels expansion on biodiversity, and similarly to assess whether management practices such as riparian zone preservation can benefit biodiversity in agroecosystems.

Key words: Anthropocene, land use change, functional traits, homogenization, legacy effect

Introduction

Environmental change is extensively studied, and consequences of environmental change have been detected for ecosystems worldwide (Brooks *et al*., 2002; Fahrig, 2003; MEA, 2005). In streams, conversion of surrounding watersheds to agriculture is associated with high levels of sedimentation, decrease of flow stability, and reduced inputs of wood debris from native forest, all of which affect structural complexity of aquatic systems (Allan, 2004; Diana, Allan & Infante, 2006; Paula *et al*., 2013). Thus, stream physical habitat is strongly influenced by watershed land use and the conversion of natural to agroecosystems often leads to habitat loss and homogenization (Allan, 2004). In this context, land use changes in the watershed have an indirect effect on aquatic communities by changing local habitat (e.g. Iwata, Nakano & Inoue, 2003; Smokorowski & Pratt, 2007; Pease *et al*., 2011; Leitão et al., 2017).

Environmental changes at watershed and local scales often lead to biodiversity loss through a non-random and gradual pattern, whereby species with narrow habitat preferences or specific requirements are more likely to be extirpated than species with

broad habitat requirements (Purvis *et al*., 2000; Pimm *et al*., 2014). In streams, environmental changes typically jeopardize the long-term persistence of specialist species by reducing the diversity and quality of suitable habitats (Scott & Helfman, 2001). The decline of specialists in local communities can disproportionately influence functional structure by replacing unique functional attributes with those that are widespread at the regional scale (Clavel, Julliard & Devictor, 2011). That being said, taxonomic and functional approaches may show different responses to the same environmental gradient or environmental change (Hoeinghaus, Winemiller & Birnbaum, 2007; Pool, Grenouillet & Villéger, 2014). For example, land use change may increase stream taxonomic diversity if the number of non-native species exceeds extirpations, but reduce functional diversity due to high functional overlap of an assemblage dominated by generalist taxa (Bojsen & Barriga, 2002; Lorion & Kennedy, 2009; Casatti *et al*., 2015).

Comparing altered versus reference conditions through taxonomic and functional community responses is a useful way to investigate effects of environmental changes on biodiversity. However, the reality of environmental dynamics in the Anthropocene (Waters *et al*., 2016) is that true reference conditions are often lacking, and many systems are experiencing continuing or compounding environmental changes. For example, many watersheds have experienced multiple different types of land use change over time, but we have a limited understanding of how subsequent alterations to already altered ecosystems may further affect communities. An influential study by Harding *et al*. (1998) described the legacy effect of agriculture on stream macroinvertebrate and fish assemblages even following subsequent reforestation. However, unlike the example from Harding *et al*. (1998), further land use change in agricultural watersheds is more likely to be to a different agricultural use rather than reforestation (e.g. Lapola *et al*., 2010; Rudorff *et al*., 2010).

Advances in 'green energy' coupled with recent changes in market valuation and/or incentives associated with biofuels production are important drivers of land use change in existing agricultural watersheds (Martinelli & Filoso, 2008; Lapola *et al*., 2010). Due to increasing global demand for bioenergy, which may supply one-third of primary energy by 2050 (IEA, 2012), land use change for biofuels production should continue to be a significant dimension of environmental change for the foreseeable future. Conceptually, biofuels can provide a sustainable alternative to fossil fuels, but the conversion of food crops or land to biofuels production has raised many questions about long-term food security and environmental costs of further land use change (Escobar *et al*., 2009; Tilman *et al*., 2009; Lapola *et al*., 2010; Koizumi, 2013). Less attention has been given to the consequences of biofuels production on biodiversity (Lu *et al*., 2012; Immerzeel *et al*., 2014; Mlambo *et al*., 2015). In this context, streams from São Paulo state, Brazil, are a good ecological model to study responses of biodiversity in dynamic agroecosystems, specifically those shifting towards biofuels production. Sugarcane (*Saccharum* spp.) is the most efficient first-generation ethanol biofuel source and Brazil, especially the state of São Paulo, is the leading producer of sugarcane worldwide (UNICA, 2016). Atlantic Forest and Cerrado biomes of southeastern Brazil have been replaced by agriculture for more than a century and recent land use change is dominated by the subsequent conversion of pasture to sugarcane (Lapola *et al*., 2014).

Distinct land use practices associated with pasture and sugarcane may differently affect stream habitat and biota. Compared with low-input livestock production on pasture, large areas with bare soil associated with intense sugarcane production and mechanized harvesting (annual burning prior to harvest is no longer the norm in this region) can increase superficial runoff of unconsolidated sediment and consequently lead to higher stream siltation (Martinelli & Filoso, 2008). Moreover, extensive fertilization

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of sugarcane crops and agrochemical use may modify stream productivity and water quality (e.g. pH, conductivity, contaminant load; Gunkel *et al*., 2007; Martinelli & Filoso, 2008; Christofoletti *et al*., 2013; Filoso *et al*., 2015; Schiesari & Corrêa, 2016). On the other hand, due to pressures from the international sugar trade and the need for ecologically sustainable biofuel production, sugarcane industry urges rural properties to respect Brazilian environmental legislation regarding Permanent Protection Areas (e.g. stream buffers; Bonsucro, 2015; UNICA, 2016). This, could over time contribute to riparian and stream regeneration and improved watershed biodiversity (e.g. Naiman & Décamps, 1997; Manning, Taylor & Hanley, 2015).

At present, the ecological consequences of conversion of pasture to sugarcane for biofuels production are essentially unknown (Immerzeel *et al*., 2014; but see Corbi, Trivinho-Strixino & Santos, 2008; Verdade *et al*., 2012; Schiesari & Corrêa, 2016). The few previous studies on this topic compared biodiversity among land use categories such as remnant natural biomes, pasture and sugarcane (e.g. Corbi *et al*., 2008; Schiesari & Corrêa, 2016). Our study is the first to quantify biodiversity responses to actual land use conversion for sugarcane production. For this research, thirty-eight agroecosystem streams representing different degrees of watershed land use change were sampled 10 years apart (2003 and 2013) and watershed land use, instream habitat and fish assemblages were quantified in both periods. Functional structure of fish assemblages was quantified using ecomorphological traits associated with habitat and resource use. Given that habitats and associated fish assemblages in these streams are already degraded (Casatti *et al*., 2006; Casatti, Ferreira & Carvalho, 2009; Casatti *et al*., 2015), major shifts in diversity due to loss of functionally-specialized taxa are no longer expected. Instead, our hypothesis was that degree of change in fish assemblages is correlated with the degree of change in land use and instream habitat.

Methods

Study area

Our study area is located in the São José dos Dourados and Turvo-Grande river basins of southeastern Brazil (Fig. 1). The region belongs to the Serra Geral geological formation and presents a relatively low slope and plains of quaternary fluvial sedimentary nature (IPT, 1999). Regional soils are characterized by unconsolidated sand and clay sediments with high erosive potential (Silva *et al*., 2007). This region is one of the oldest and most intensive agricultural areas in Brazil. The native semi-deciduous seasonal forest (Atlantic Forest) was replaced by coffee and citrus cultivation and livestock grazing over more than a century (Silva *et al*., 2007). Recently, due to governmental incentives for biofuel and sugar production, sugarcane has replaced other agricultural uses (notably pasture) and has become one of the most dominant land-use types in the region (Fig. 1; Verdade *et al*., 2012; Lapola *et al*., 2014). Sugarcane land cover doubled in Brazil during the study period (Lapola *et al*., 2014), and our study system represents this expansion. Cumulative land-use changes for the study area over the past century results in less than 4% of native forest remaining in small and isolated fragments (Nalon, Mattos & Franco, 2008). As a consequence, most of the streams show high physical degradation and relatively homogenous fish assemblages (Casatti *et al*., 2006; Casatti *et al*., 2015).
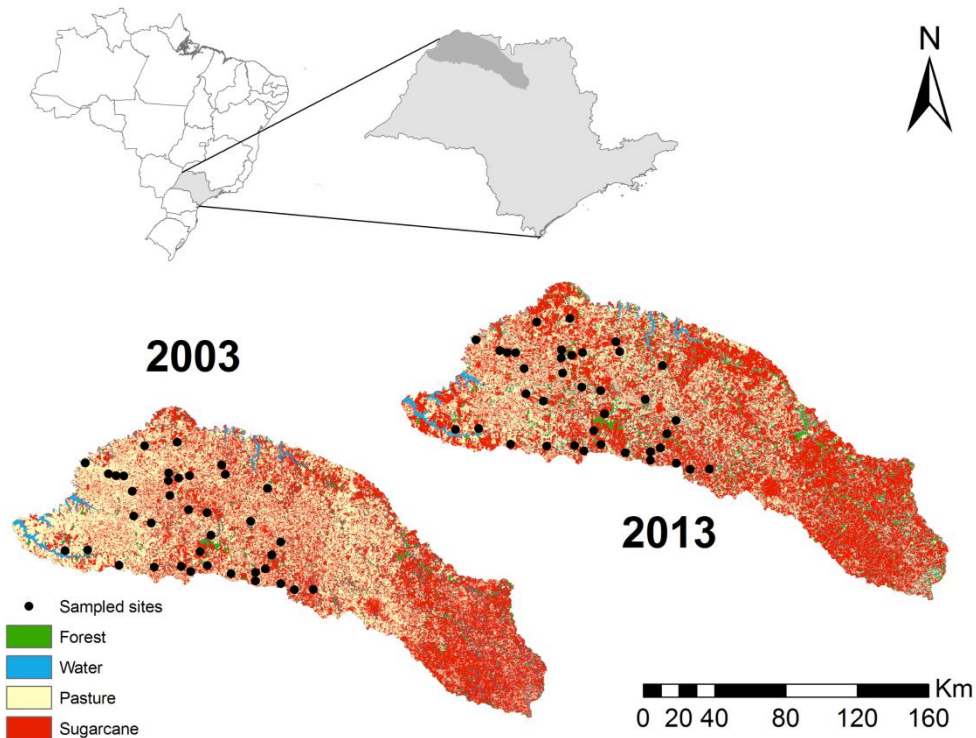
Fig. 1 Stream reaches (black circles) and land use (forest, pasture, and sugarcane) in the study area, São Paulo State, Brazil.

Land use and instream habitat

Environmental variables were quantified at regional (i.e. watershed) and local (i.e. instream and ecotone) scales. Watershed boundaries were delineated using a digital elevation model (DEM; ASTER GDEM, 2011) with the SWAT extension (Soil and Water Assessment Tool; SWAT, 2009) for ArcGIS 9, for georeferenced points taken during the sampling period (Casatti *et al.*, 2015). For each watershed, four land use classes were defined: (1) native forest - areas occupied by native vegetation (i.e. semi-deciduous seasonal forest); (2) pasture - areas used for livestock grazing; (3) sugarcane - area with sugarcane cultivation; and (4) other - includes areas with perennial and annual cultivation, rural facilities and roads. The land use classes in each watershed were obtained from analysis of Landsat 5 TM satellite images ($30 \times 30 \ m^2$ resolution), which

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were available from the National Institute for Space Research (INPE). Land use classification was conducted in the software Erdas Imagine 9.2.

For characterization of instream habitat and ecotone (i.e. presence of trees and shrubs), each 75 m stream reach was surveyed along three equidistant transects approximately located in the middle and five meters from either end of the reach. Local variables were quantified as follows: (1) Physical Habitat Index (PHI), a habitat assessment based on visual quantification of diversity of mesohabitat and substrate, channel modification, and stream bank, where higher values indicate more heterogeneous physical habitat (see Casatti *et al.*, 2006 for more details); visual assessment of the percent of riparian strip (10 m width) occupied by (2) trees and (3) shrubs; (4) substrate composition, i.e. percent of the stream bottom occupied by unconsolidated (silt, clay and sand) versus consolidated (gravel, cobble, rock and slab) material, was obtained through visual evaluation at several points along the stream reach; (5) percent of instream habitat occupied by woody debris; (6) percent of both banks occupied by grass (mostly invasive signalgrass, *Brachiaria* spp., which is widely planted for grazing); visual estimate of percent of the stream reach comprised by (7) pools, (8) riffles, and (9) runs; flow was measured at nine points (three from each transect) using a flow meter and used to calculate (10) average, (11) standard deviation and (12) coefficient of variation of velocity; depth was measured at 15 points (five from each transect) and used to calculate (13) average, (14) standard deviation, and (15) coefficient of variation of depth; stream width was measured at each transect and used to calculate (16) average, (17) standard deviation, and (18) coefficient of variation of width; (19) stream area was estimated as the product of reach length and average width; and (20) stream volume was estimated as the product of stream area and average depth.

Fish assemblages and functional groups

We sampled fish assemblages through two electrofishing passes using an AC generator (220 V, 50-60 Hz, 3.4-4.1 A, 1000 W). Prior to collections, sample reaches were isolated at the upstream and downstream limits with block nets (5 mm mesh). All collected individuals were fixed in a 10% formaldehyde solution and transferred to 70% ethanol after 48 hours. Species were identified and enumerated for each site in both time periods. Changes in taxonomic nomenclature [e.g. *Astyanax altiparanae* changed to *A. lacustris* (Lucena & Soares, 2016)] were accounted for, when necessary. Vouchers were deposited in the collection of the Department of Zoology and Botany at Universidade Estadual Paulista "Júlio de Mesquita Filho", campus of São José do Rio Preto, São Paulo (DZSJRP).

For each species, we quantified functional traits associated with habitat and resource use (Table 1). Linear distances and areas were measured from adult individuals (10 specimens per species, randomly chosen from across all sampling locations) to obtain 15 functional traits (Table 1). Species were assigned to functional groups using hierarchical agglomerative clustering (UPGMA). This method allows a species to join a group based on the mean of the functional traits distances (i.e. Euclidean). Fit of the resulting cluster to the original distance matrix was assessed using cophenetic correlation. Silhouette plots were used to define the optimal number of functional groups, and heat maps were generated to summarize the primary functional traits distinguishing each group. Clustering and cophenetic correlations were performed using *stats* package in R version 3.03.

Table 1. Ecomorphological attributes, calculations and ecological significance of functional traits related body size, vertical, horizontal habitat and resource uses. Ecomorphological attributes were calculated following Oliveira et al. (2010). Maximum body height (MBH), maximum body width (MBW), body midline height (BMH), standard length (SL), caudal peduncle length (CPdL), caudal peduncle height (CPdH), caudal peduncle width (CPdW), dorsal fin area (DA), caudal fin area (CA), pectoral fin area (PtA), pectoral fin length (PtL), head length (HdL), head height (HdH), mouth width (MW), eye height (EH).

Ecomorphological attributes	Calculation	Ecological significance
Compression index	$CI = \dfrac{MBH}{MBW}$	Higher values indicate lateral compression of the fish, which indicate fish that explore habitats with slower water velocity (Gatz, 1979; Watson and Balon, 1984).
Depression index	$DI = \dfrac{BMH}{MBH}$	Low values indicate fishes inhabiting environments with high hydrodynamism, able to maintain their position even when stationary (Hora, 1930).
Relative depth	$RD = \dfrac{MBH}{SL}$	Lower values indicate fishes inhabiting fast waters. It is directly related to the ability to perform vertical spins (Gatz, 1979).
Fineness ratio	$FC = \dfrac{SL}{\sqrt{MBH \times MBW}}$	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 length of caudal peduncle	$RLPd = \dfrac{CPdL}{SL}$	Fishes with long caudal peduncle are goods swimmers. However, fishes adapted to rapid water flow, but no necessarily nektonic as armored catfishes, also presented long caudal peduncules in function of propulsion in short distances (Watson & Balon, 1984; Winemiller, 1991)
Relative height of caudal peduncle	$RHPd = \dfrac{CPdH}{MBH}$	Lower values indicate greater maneuverability potential (Winemiller, 1991).
Relative width of caudal peduncle	$RWPd = \dfrac{CPdW}{MBW}$	Higher relative values indicate better continuous swimmers (Winemiller, 1991).
Relative area of dorsal fin	$RAD = \dfrac{DA}{SL^2}$	Dorsal fins with larger relative areas have better capacity of stabilization in deflections (Gosline, 1971).
Relative area of caudal fin	$RAC = \dfrac{CA}{SL^2}$	Caudal fins with larger relative areas are important for the acceleration (Balon, Crawford & Lelek, 1986).
Relative area of pectoral fin	$RAPt = \dfrac{PtA}{SL^2}$	The pectoral fin area is generally high for slow swimming species, which use the pectoral fin for maneuverability, as some characids. Moreover, pectoral fin area also can be high for fishes that exploit habitats with intense current, as the siluriforms (Watson & Balon, 1984; Wilga & Lauder, 1999).

Aspect ratio of pectoral fin	$ARPt = \dfrac{PtL^2}{PtA}$	Higher ratio indicates long and narrow pectoral fins, which is more expected on fish that are continuous high-speed swimmers and prefer pelagic regions (Wainwright, Bellwood & Westneat, 2002).
Relative length of head	$RLHd = \dfrac{HdL}{SL}$	Larger relative values of head length are found in fishes which feed of larger prey. This index should be larger for piscivores (Watson & Balon, 1984; Winemiller, 1991; Barrella Beaumord & Petrere, 1994; Pouilly *et al.*, 2003; Willis, Winemiller & Lopez-Fernandez, 2005).
Relative height of head	$RHHd = \dfrac{HdH}{MBH}$	Larger relative values of head height are found in fishes which feed of larger prey. Larger values for this index are expected for piscivores (Winemiller, 1991; Willis *et al.*, 2005).
Relative Width of Mouth	$RWM = \dfrac{MW}{MBW}$	Larger relative values of mouth length suggest fishes which feed of larger prey (Gatz Jr., 1979; Balon *et al.*, 1986; Winemiller, 1991; Ward-Campbell, Beamish & Kongchaiya, 2005).
Eye Position	$EP = \dfrac{EH}{HdH}$	Related with food detection and provides information about visual predation activity (Poully *et al.*, 2003). Moreover, position of eyes is related to vertical habitat preference (Gatz, 1979); high values indicate dorsally located eyes, typical of benthic fish (Mahon, 1984; Watson & Balon, 1984).

Based on the trait matrix and agglomerative clustering (cophenetic coefficient = 0.82; (see Fig. S1) we assigned fish species to six functional groups. The most diverse and abundant group was comprised mostly by nektonic species, which explore the water column and show higher compression index (CI) and relative depth (RD). A second group was comprised by benthic species that primarily forage in the stream bottom and which had lower CI, higher relative dorsal (RAD) and pectoral fin (RAPt) area, and relatively ventral mouth position and dorsal eyes. Two more groups that are associated with deeper lentic mesohabitats were distinguished by higher relative mouth width (RWM) and eye position (EP), and species with lower fineness coefficient (FC), higher RAD, RD and relative length of head (RLHd). Gymnotiform species (i.e. Neotropical knifefishes) associated with marginal grasses were in a separate group, distinguished by fusiform bodies, absence of caudal fin and an elongated anal fin. The final group was

comprised by two non-native species generally associated with slow water velocity and possessing small body size, and higher relative caudal area (RAC) and depression index (DI).

Statistical analyses

The following analyses were used to test our primary hypothesis and also quantify relationships between different components of the indirect effect that land use change is expected to have on stream diversity. We quantified the degree of change in environmental conditions between 2003 and 2013 for each stream using pairwise Euclidean distances in principal coordinates analysis (PCoA). We calculated this degree of change using regional, local, and combined (regional and local) environmental matrices. We used Procrustes and PROTEST (Jackson, 1995; Peres-Neto & Jackson, 2001) to test for correlation between regional and local environmental matrices for each year, as well as between regional land use in 2003 and local habitat in 2013 to test for a potential legacy effect of watershed land use on local habitat. The relationship between degree of change in regional and local environment was assessed using Spearman's correlation.

Changes in fish assemblage taxonomic richness, diversity (Shannon-Wiener) and evenness (Pielou) between time periods for each stream were calculated by subtracting those estimates in 2003 from the corresponding values from 2013. Thus, streams that lost or gained species, diversity or evenness over time had negative and positive values, respectively. Complementing the taxonomic diversity estimates, functional richness (FRic), evenness (FEve) and divergence (FDiv; Villéger, Mason & Mouillot, 2008) were calculated for each stream fish assemblage in 2003 and 2013 and changes in FRic, FEve and FDiv between time periods for each stream were calculated by subtracting values

from 2003 from the corresponding values in 2013. The aforementioned functional diversity indices were calculated using species scores on the first two axes from PCoA (52% of trait variation; Fig. 2 and Table S1). To provide a more direct comparison with species richness, change in functional richness was also assessed as the difference in the number of functional groups in each stream between periods. Change in fish assemblage structure between periods was assessed using pairwise assemblage similarity based on species composition and species' relative abundances (Jaccard and Bray-Curtis indices, respectively) for both the taxonomic and functional group datasets and visualized using non-metric multidimensional scaling (NMDS). T-test was used to test for differences in taxonomic and functional metrics between periods.

We used Redundancy analysis (RDA) to assess correlations between taxonomic and functional structure of fish assemblages and environmental variables in 2003 and 2013, as well as between fish assemblages in 2013 and environmental variables in 2003 (i.e. to test for a legacy effect). Significant and non-redundant variables were included through forward selection procedures (Blanchet, Legendre & Borcard, 2008). Finally, to test the hypothesis that degree of environmental change determines the degree of biological change in taxonomic and functional structure of fish assemblages, we conducted linear regressions between environmental distances (regional, local and combined) and change in taxonomic and functional attributes (i.e. all diversity metrics and similarities for assemblage structure) for each site. All of the above analyses were performed in R using the *stats*, *vegan* and *packfor* packages.
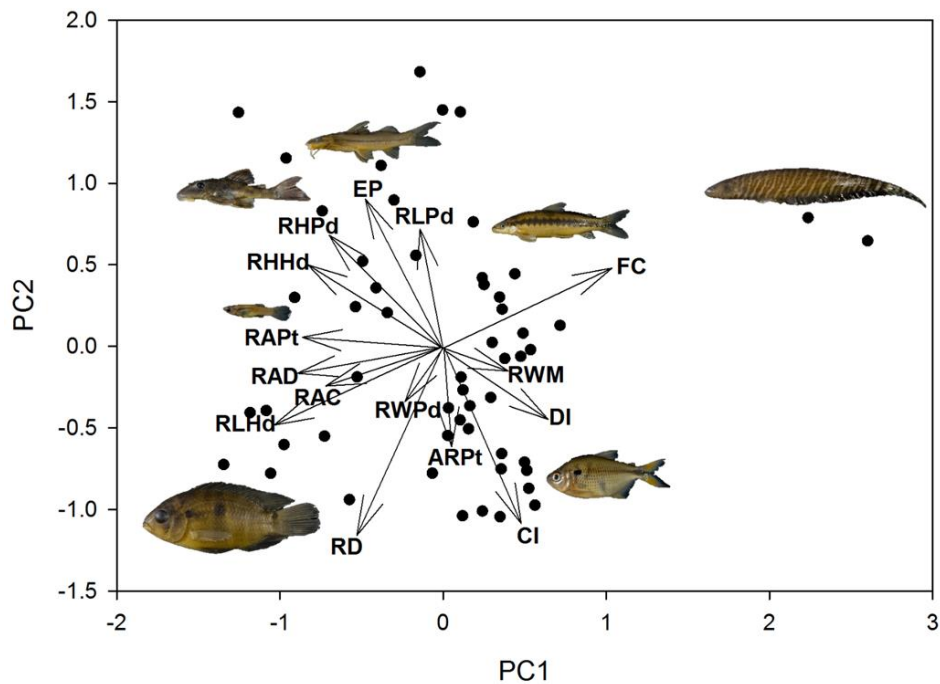
Fig. 2 Ordination of 54 species based on 15 functional attributes. Each species is represented by a black circle. See Table 1 for abbreviations of ecomorphological attributes and Table S1 for species list and loadings on each axis. Images of select species are provided to represent the body shapes most associated with functional attributes.

Results

The primary gradient of land use change between 2003 and 2013 was conversion of pasture to sugarcane, but the degree of change differed greatly among streams (Fig. 3a). A few watersheds showed increases in 'other' and 'forest' land use types, and percent of forest and sugarcane land uses were correlated. Changes in instream and riparian habitat were less consistent, but many streams showed a general decrease of width, depth, area and volume, likely as a consequence of increased siltation (Fig. 3b). PCA of combined regional and local environmental factors indicated that streams with greater habitat heterogeneity (i.e. higher proportion of wood debris, riffles, pools, consolidated substrate and PHI) were associated with watersheds that had a higher proportion of forest and to a lesser degree sugarcane (likely due to the presence of more intact riparian zones

than found in pasture; see Fig. S2). However, watershed land use and instream habitat were not correlated for either period (2003: m_{12}=0.26, P=0.14; 2013: m_{12}=0.28, P=0.11). Examination of Procrustes residuals indicated that sites with a good fit between regional and local attributes in 2003 were poorly-fit in 2013, and vice versa (see Table S2), and a marginally significant legacy effect of land use in 2003 on instream habitat in 2013 was detected (m_{12}=0.30, P=0.06). Sites with the largest Procrustes residuals in 2013 had the most extreme percent covers of sugarcane, forest or other land uses (see Table S2 and Fig. 3a). Magnitude of change in watershed land use was not correlated with magnitude of change in instream habitat (rho=0.20, P=0.21).

Similar numbers of individuals and species were collected in both years (i.e. 7,439 individuals of 47 species in 2003 and 7,535 individuals of 44 species in 2013), but species composition and relative abundances differed between years (Table S3). Abundance of non-native species, almost entirely *Poecilia reticulata*, accounted for approximately 16% and 37% of total abundance in 2003 in 2013, respectively. In 2003, the most abundant species were *A. lacustris* (n=1,926), *Knodus moenkhausii* (n=1,347) and *P. reticulata* (n=1,199). Two other non-native species were sampled in 2003: *Tilapia rendalli* (n=8) and *Oreochromis niloticus* (n=1). In 2013, the most abundant species was *P. reticulata* (n=2,651), followed by *A. lacustris* (n=1,113) and *K. moenkhausii* (n=886). *T. rendalli* and *O. niloticus* were not collected in 2013, but two other non-native species were registered in the region for the first time: *Xiphophus maculatus* (n=108) and *Crenicichla semifasciata* (n=22). In general, few differences in assemblage taxonomic and functional diversity were observed between years; species richness and FDiv decreased whereas FEve increased between periods (Table 2).
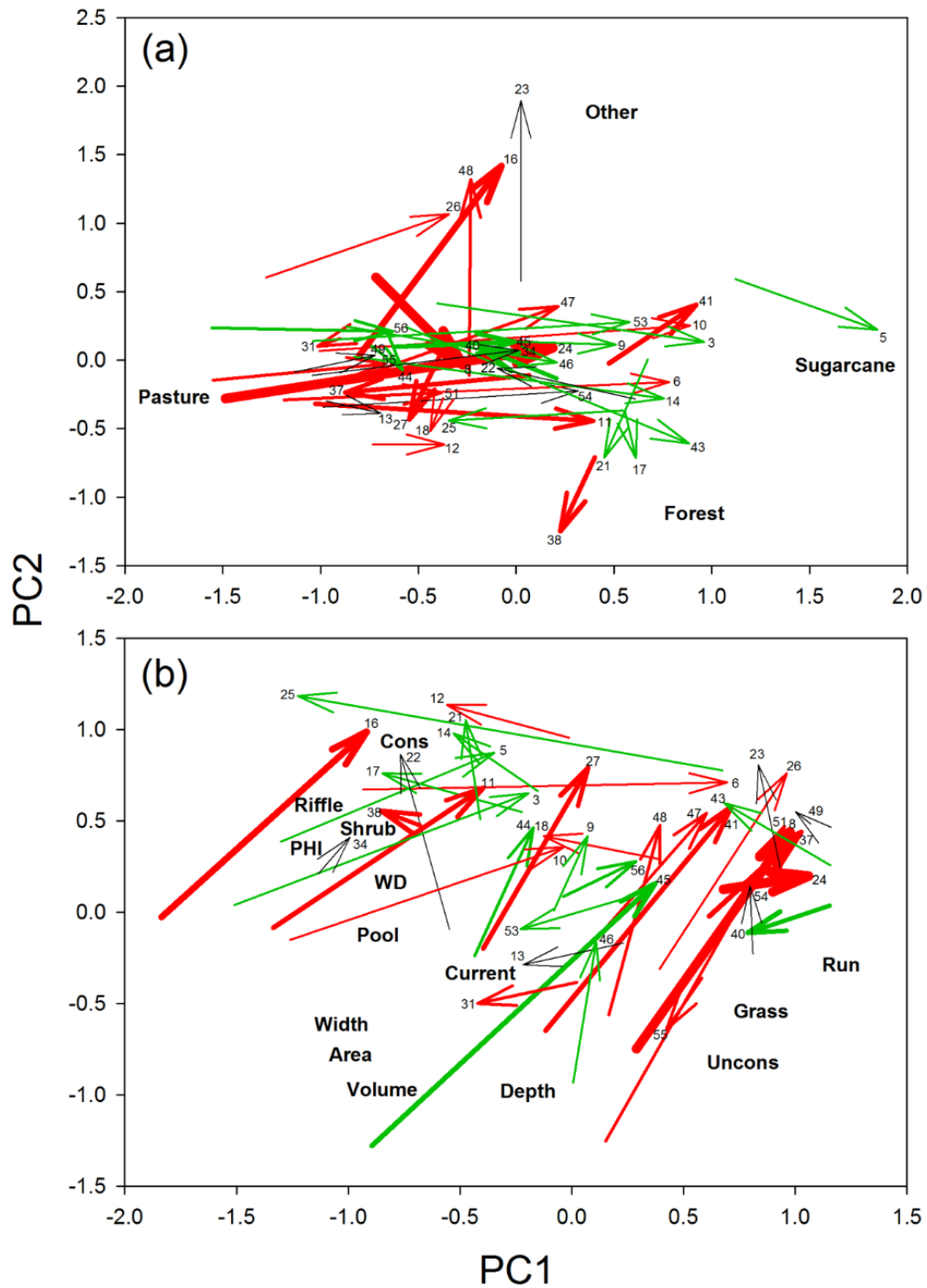
Fig. 3 PCoA bi-plots calculated using a) watershed land use and b) instream and ecotone attributes for 38 stream reaches in 2003 and 2013. Arrows link site scores for each stream between periods, starting at the score in 2003 and with the arrow head at the score for 2013. Arrow color indicates increasing (green) or decreasing (red) species richness over time, and arrow thickness represents the relative magnitude of change in species richness. Labels for environmental factors are centered on their respective axis loadings. Site numbers are consistent between plots for comparison of degree of regional and local change. See Table S2 for Procrustes residuals.

Table 2. Maximum, minimum, mean and standard deviation of species richness, diversity, and eveness, and richness of functional groups (FR), functional richness (FRic), functional evenness (FEve) and functional divergence (FDiv) in 2003 and 2013. * represents significant differences (p value < 0.05) in mean values between periods.

	Richness*		Diversity		Eveness		FR		FRic		FEve*		FDiv*	
	2003	2013	2003	2013	2003	2013	2003	2013	2003	2013	2003	2013	2003	2013
Max.	20	20	2.50	2.40	0.90	1.00	6	6	11.00	10.80	0.80	0.85	0.90	0.80
Min.	3	3	0.05	0.40	0.05	0.25	2	2	0.10	0.20	0.30	0.40	0.50	0.45
Mean	10.50	9.30	1.50	1.40	0.70	0.70	4.50	4.30	6.50	5.00	0.60	0.65	0.75	0.70
SD	3.90	3.70	0.50	0.50	0.20	0.20	1.20	1.00	3.20	3.00	0.10	0.10	0.10	0.08

Taxonomic and functional assemblage structure (both composition and abundance) were also similar between periods (see Fig. S3). However, the environmental variables correlated with fish assemblage structure differed between years (Table 3). Most notably, percent sugarcane in the watershed was a significant variable in the species and functional composition models in 2003, but was not included in any models for 2013. Also of interest was that consolidated substrate and PHI were the only significant variables in the species and functional abundance models for 2003, respectively, but percent of stream bank occupied by grasses was the only factor for both of those models in 2013 (associated with increased abundances of knifefishes, cichlids and invasive poecillids in shallower streams). Variation in species and functional composition was explained primarily by aspects of local habitat (e.g. percent riffle and pool, depth and volume, current velocity, riparian trees and shrubs). Contrary to expectations, degree of environmental change was not significantly correlated with degree of change in stream fish assemblages. Only a single significant, though weak, positive relationship was observed between degree of change in watershed land use and change in functional fish assemblage structure (Table 4).

Table 3. Relationship between taxonomic and functional structure (composition and abundance) and environment in both periods (2003 and 2013) as well as the relationship between biological variables in 2013 and environment in 2003. All significant variables include in each model are listed.

Period	RDA	Ajusted-R^2	p-value	Variables
2003	Composition vs Environment	0.08	0.005	Riffles, Depth_SD, Current_M and Sugarcane
	Abundance vs Environmnet	0.13	0.140	Consolidated substrate
	Functional Composition vs Environment	0.05	0.015	Sugarcane
	Functional Abundance vs Environment	0.10	0.015	PHI
2013	Composition vs Environment	0.13	0.005	Current_M; Tree; Volume
	Abundance vs Environmnet	0.03	0.025	Grasses
	Functional Composition vs Environment	0.14	0.005	Tree; Pools; Width_CV
	Functional Abundance vs Environment	0.09	0.005	Grasses
2013 vs 2003	Composition vs Environment	0.05	0.005	Riffles
	Abundance vs Environmnet	0.02	0.030	PHI
	Functional Composition vs Environment	0.14	0.005	Shrubs
	Functional Abundance vs Environment	0.11	0.005	Grasses, Depth_SD

Table 4. Linear regressions between environmental changes (regional, local and combined distances) and taxonomic and functional changes of fish assemblages between 2003 and 2013.

		Regional		Local		Combined	
		R^2	p-value	R^2	p-value	R^2	p-value
Taxonomic	Δ Richness	0.07	0.10	0.01	0.46	0.02	0.38
	Δ Diversity	0.00	0.74	0.00	0.85	0.00	0.89
	Δ Evenness	0.03	0.27	0.00	0.59	0.02	0.31
	Composition	0.03	0.25	0.08	0.06	0.05	0.15
	Abundance	0.00	0.66	0.05	0.15	0.05	0.14
Functional	Δ Functional Richness	0.01	0.46	0.04	0.21	0.03	0.27
	ΔFRic	0.00	0.94	0.01	0.27	0.00	0.73
	ΔFDiv	0.00	0.87	0.00	0.85	0.00	0.86
	ΔFEve	0.00	0.61	0.00	0.70	0.00	0.98
	Composition	0.04	0.22	0.00	0.94	0.00	0.85
	Abundance	**0.13**	**0.02**	0.00	0.85	0.01	0.53

Discussion

Despite associations between environmental variables and fish assemblages in both periods, the magnitude of change in taxonomic and functional attributes of fish assemblages was not significantly correlated with magnitude of land use changes in most cases. The single exception was the weak correlation between degree of watershed change (i.e. replacement of pasture by sugarcane) and change in functional fish assemblage structure. The general lack of relationships is quite surprising given the commonly observed effects of land use change on instream habitat and biological diversity of freshwater ecosystems (Allan, 2004; Smokorowski & Pratt, 2007). For an indirect effect of land use change on stream assemblages to occur, watershed land use must affect instream habitat, which in turn affects biological communities. We found that for these agroecosystem streams, instream habitat was not associated with watershed land use, and degree of change in land use between periods was not associated with degree of change in instream habitat. Importantly, we detected evidence of a time lag or legacy effect, thus the effects of rapid conversion of the landscape for biofuels production on instream habitat and biodiversity are presumably not yet fully evident.

Our interpretation of a significant time lag in the instream habitat and biodiversity responses to pasture conversion for sugarcane production is supported by multiple lines of evidence. Streams with the most extreme land uses in 2013 had the largest residuals in the Procrustes analysis (i.e. worst fit between watershed land use and instream habitat), and streams with the highest Procrustes residuals in 2003 were comparatively well fit in 2013 (i.e. good fit between watershed land use and instream habitat). This is further supported by the marginally significant relationship between watershed land use in 2003 and instream habitat in 2013. Furthermore, taxonomic and functional assemblage structure in 2013 was significantly correlated with percent of the stream margin occupied

by grasses, which in this case is primarily invasive signalgrass planted for grazing in pastures (i.e. a remnant of pasture land use even following conversion to sugarcane). What is less clear is whether magnitude of change in the watershed will eventually be correlated with magnitude of change in instream habitats and biodiversity (i.e. catch-up with the time lag). Should we expect continued changes to instream habitats and biodiversity over time (e.g. extinction debt or return of extirpated species), or are instream habitats and diversity somehow buffered from further change due to their already degraded nature? The answers to such questions have important implications for basic understanding of community dynamics as well as when considering the environmental consequences of biofuels production.

Biodiversity patterns are determined by the interaction among four primary processes: selection (e.g. competition, predation, habitat filtering), drift (i.e. chance events), dispersal (i.e. movement of individuals) and speciation (Vellend, 2010). In streams, habitat filtering mediated by deforestation can increase taxonomic and functional similarity between communities (i.e. decrease of beta diversity; Bojsen & Barriga, 2002). Generalist species with broad ranges of habitat use, resource use and physiologic tolerance tend to be favored and increase in abundance and frequency of occurrence (Scott, 2006; Pardini *et al*., 2009). Despite no detailed faunistic surveys in our study area before massive deforestation in the beginning of the 20th century, we can infer that the assemblages observed in our surveys represent homogeneous and generalist subsets of the pre-deforestation species pool (Casatti *et al*., 2006; Casatti *et al*., 2009; Casatti *et al*., 2015). In addition to filtering sensitive taxa, non-native species common in degraded habitats invaded the system. Abundance of non-native *P. reticulata* more than doubled from 2003 to 2013, dominating assemblages. The other most common species in our assemblages are widespread in different types of environments (Fialho *et al*., 2007; Daga

et al., 2012; Cunico *et al*., 2012). These generalist species were likely 'filtered' from the regional pool by previous land use change and instream habitat change, and thus these generalist communities may exhibit some resistance to further changes through modifications of diet, behavior, etc. in response to changes in the abiotic environment. Similarly, Jackson & Sax (2009) found that plant species in highly fragmented habitat seemed to be more persistent over time than what is expected by ecological theories. In this context, the degree of environmental change required to induce changes in such assemblages may be much higher than in the initial stages of alteration.

That being said, associations between some taxa or functional groups and environmental factors such as percent of stream margin occupied by grass, including a legacy effect of previous land use, indicates that further selection is possible even in assemblages dominated by generalists. Such changes are more likely to be in species relative abundances rather that species composition (Smokorowski & Pratt, 2007). From a functional standpoint, we observed reduced relative abundances of nektonic species (functional group A) and increased relative abundances of Cichlids (functional group D) and Poeciliids (functional group F, including the aforementioned *P. reticulata*). Functional groups D and F are composed by species tolerant to hypoxia, organic pollution and siltation. Similarly, Santos, Ferreira & Esteves (2015) found that sugarcane in the riparian zone modified stream physical habitat, which corresponded with increased abundance of tolerant species and decreased abundance of nektonic species.

On the other hand, strong relationships between degree of environmental change and community attributes may be precluded by drift. Simplification of the instream habitat and loss of sensitive species over time may result in assemblages that are more strongly affected by stochastic factors than niche processes (Püttker *et al*., 2015). For example, Baselga, Bonthoux & Balent (2015) interpreted a weak relationship between

patterns of bird temporal beta diversity and land use change as the result of historical human disturbance that filtered species such that contemporary assemblages of generalist taxa use the landscape in a random way. Thus, we may expect that habitat filtering acted at initial stages of deforestation, wherein rare and sensitive species were extirpated from the regional pool, and contemporary fish assemblages are composed mainly by species able to persist across a broad range of conditions with assemblage dynamics driven primarily by stochastic processes. However, the relationship between diversity and stability has been debated for decades (e.g. McCann, 2000), as has the relative importance of stochastic and deterministic processes on stream communities (Gido & Jackson, 2010). Further research is needed to understand the relative importance of deterministic and stochastic processes on agroecosystem streams.

As eloquently stated by Schiesari & Corrêa (2016), "the environmental benefits of biofuels are critically contingent on which, where, and how biofuel feedstocks are produced". Although sugarcane is the most efficient first-generation biofuel feedstock, there is a massive deficiency in our understanding of how agroecosystem dynamics for the production of sugarcane affect aquatic and terrestrial biodiversity (Immerzeel *et al*., 2014). In their review of effects of sugarcane expansion on wildlife in São Paulo state, Verdade *et al*. (2012) report reduced diversity but increased abundances of small rodents (mostly commonly *Calomys tener* and *Necromys lasiurus*) and their mesopredators on sugarcane plantations, as well as dramatic population increases of capybaras (*Hydrochoerus hydrochaeris*) in some regions. Increased densities of rodent species may contribute to the emergence or resurgence of infectious diseases such as Hantavirus, leptospirosis, typhus and spotted fever (Verdade *et al*., 2012). Bird diversity is also generally reduced in agricultural landscapes (Verdade *et al*., 2012) and land conversion may increase contaminant loads in aquatic ecosystems and species (Corbi *et al*., 2008;

Verdade *et al*., 2012). Using a landscape comparative approach across a land-use intensity gradient (i.e. native habitats < pasture < sugarcane), Schiesari & Corrêa (2016) found that land-use intensity affected aquatic ecosystem productivity, with increased conductivity, turbidity and phytoplankton biomass. Biodiversity and community organization of the lentic ecosystems differed in agricultural vs. native landscapes, with ponds imbedded in sugarcane fields supporting only a fraction of the potential aquatic diversity. However, when present, riparian buffers surrounding larger wetlands in sugarcane fields appeared to reduce the impact on aquatic diversity (Schiesari & Corrêa, 2016).

Our findings demonstrate that instream habitat and aquatic biodiversity in streams with agricultural watersheds undergoing rapid conversion to biofuels production likely will not experience the full magnitude of responses in the short term (i.e. < 10 yrs.). Whether or not this is due to the already degraded nature of the system is worth considering, but a key limitation is the general lack of long-term studies (e.g. Matthews *et al*., 2013) on these types of ecosystems. Previous studies compared biodiversity responses among land use categories. Although this is clearly a useful approach, it does not incorporate the complex history of land use change that we know can influence biodiversity (e.g. Harding *et al*., 1998). For example, Dupouey *et al*. (2002) found that farming practices influenced gradients of soil nutrients and species composition and diversity more than 2000 years later. In addition, for our study, not all watersheds experienced the exact same changes in land use (i.e. directionality of arrows in Fig. 3a, b). General trends were present (i.e. pasture to sugarcane, decrease of width, depth, area and volume), but variation in direction of change among watersheds likely combined with time lags to limit our ability to detect a relationship between degree of change and biodiversity responses. Thus, effects of biofuels production on biodiversity should

ideally be tested using repeated sampling of sufficient duration to account for time lags. This approach would also be appropriate to assess sustainable management practices and certifications (e.g. Bonsucro) on biodiversity. For example, we are interested in the recovery of riparian zones and instream habitat associated with sugarcane production on former pasture lands (e.g. increased PHI and woody debris associated with intact riparian zones) – will recovery of riparian zones and increased instream habitat complexity over time be followed by return of certain sensitive taxa that were previously excluded from those systems? That would be significant for consideration of sugarcane as an ecologically sustainable biofuel feedstock.

Besides the uncertain consequences of conversion of pasture to sugarcane production on biodiversity within the same watersheds, there is a growing concern about how this expansion can indirectly affect other regions and ecosystems in Brazil. For example, the area utilized for sugarcane production increased by more than 200% near our study area, primarily through occupation of areas previously used for direct (i.e. pasture) or indirect (i.e. soybean, corn) food production (Ferreira, Alves & Shimabukuro, 2015). This scenario could potentially increase deforestation in other Brazilian biomes, like the Amazon and Cerrado, as compensation for food production (Martinelli & Filoso, 2008; Lapola *et al*., 2010; Immerzeel *et al*., 2014; but see Verdade *et al*., 2012). Food security is one of the main claims of the agribusiness lobby to loosen environmental regulations, such as the new Brazilian Forest Code (Código Florestal) approved in 2012. Previous authors have argued that the new Forest Code represents an environmental setback by providing amnesty for illegal deforestation and reducing the protection of sensitive areas (Sparovek *et al*., 2012; Soares-Filho *et al*., 2014), with critical consequences for provisioning of global ecosystem services (Metzger *et al*., 2010). However, compliance with regulations such as permanent preservation areas is

historically very low (Sparovek *et al*., 2012), thus certification schemes (e.g. Bonsucro) associated with sugarcane production that incentivize participation in protection of riparian corridors is an important contribution to environmental sustainability. The potential for biofuels production in environmental sustainability is complex (Tilman *et al*., 2009). Clearly, the scientific community, industry and public need better data on biological responses to agroecosystem dynamics to be able to make decisions about the relative benefits of biofuels production and where, when and how that production takes place. Can certain management practices (e.g. buffer regeneration, local community engagement in biofuels production) contribute to a win-win scenario for agriculture and biodiversity (Martinelli *et al*., 2010; Manning *et al*., 2015)? This is an important topic to address and our data indicate that it requires long-term studies of community dynamics in order to offset legacy effects of previous land uses.

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References

Allan J.D. (2004) Landscape and riverscape: The influence of land use on stream ecosystems. *Annual Review of Ecology, Evolution, and Systematics*, **35**, 257–284.

Balon E.K., Crawford S.S. & Lelek A. (1986) Fish communities of the Upper Danube River (Germany, Austria) prior to the new Rhein-Main-Donau connection. *Environmental Biology of Fishes*, **15**, 243–271.

Barrella W., Beaumord A.C. & Petrere Jr M. (1994) Comparacion de la comunidad de peces de los rios Manso (MT) y Jacare Pepira (SP), Brasil. *Acta Biologica Venezuelica*, **15**, 11–20.

Baselga A., Bonthoux S. & Balent G. (2015) Temporal beta diversity of bird assemblages in agricultural landscapes: Land cover change vs. stochastic processes. *PLoS ONE*, **10**, e0127913.

Blake R.W. (1983) Functional design and burst-and-coast swimming in fishes. *Canadian Journal of Zoology*, **61**, 2491–2494.

Blanchet F.G., Legendre P. & Borcard D. (2008) Forward selection of explanatory variables. *Ecology*, **89**, 2623–2632.

Bojsen B.H. & Barriga R. (2002) Effects of deforestation on fish community structure in Ecuadorian Amazon streams. *Freshwater Biology*, **47**, 2246–2260.

Bonsucro (2015) Bonsucro Production Standard, version 4.1.1. Available at: http://www.bonsucro.com/site/production-standard

Brooks T.M., Mittermeier R.A., Mittermeier C.G., Fonseca G.A.B., Rylands A.B., Konstant, W.R. *et al*. (2002) Habitat loss and extinction in the hotspots of biodiversity. *Conservation Biology*, **16**, 909–923.

Casatti L., Langeani F., Silva A.M. & Castro R.M.C. (2006) Stream fish, water and habitat quality in a pasture dominated basin, southeastern Brazil. *Brazilian Journal of Biology*, **66**, 681–696.

Casatti L., Ferreira C.P. & Carvalho F.R. (2009) Grass-dominated stream sites exhibit low fish species diversity and guppies dominance: an assessment on two tropical pasture river basins. *Hydrobiologia*, **632**, 273–283.

Casatti L., Teresa F.B., Zeni J.O., Ribeiro M.D., Brejão G.L. & Ceneviva-Bastos M. (2015) More of the same: high functional redundancy in stream fish assemblages from tropical agroecosystems. *Environmental Management*, **55**, 1300–1314.

Clavel J., Julliard R. & Devictor V. (2011) Worldwide decline of specialist species toward a global functional homogenization? *Frontiers in Ecology and the Environment*, **9**, 222–228.

Corbi J.J., Trivinho-Strixino S. & Santos A. (2008) Environmental evaluation of metals in sediments and dragonflies due to sugar cane cultivation in Neotropical streams. *Water Air Soil Pollution*, **195**, 325–333.

Christofoletti C.A., Escher J.P., Correia J.E., Marinho J.F.U. & Fontanetti C.S. (2013) Sugarcane vinasse: environmental implications of its use. *Waste Management*, **33**, 2752–2761.

Cunico A.M., Ferreira E.A., Agostinho A.A., Beaumord A.C. & Fernandes R. (2012) The effects of local and regional environmental factors on the structure of fish assemblages in the Pirapó Basin, Southern Brazil. *Landscape and Urban Planning*, **105**, 336–344.

Daga V.S., Gubiani E.A., Cunico A.M. & Baumgartner G. (2012) Effects of abiotic variables on the distribution of fish assemblages in streams with different anthropogenic activities in southern Brazil. *Neotropical Ichthyology*, **10**, 643–652.

Diana M., Allan J.D. & Infante D. (2006) The influence of physical habitat and land use on stream fish assemblages in southeastern Michigan. *American Fisheries Society Symposium*, **48**, 359–374.

Dupouey J.L., Dambrine E., Laffite J.D. & Moares C. (2002) Irreversible impact of past land use on forest soils and biodiversity. *Ecology*, **83**, 2978–2984.

Escobar J.C., Lora E.S., Venturini O.J., Yáñez E.E., Castillo E.F. & Almazan O. (2009) Biofuels: environment, technology and food security. *Renewable and Sustainable Energy Reviews*, **13**, 1275–1287.

Fahrig L. (2003) Effects of habitat fragmentation on biodiversity. *Annual Review of Ecology, Evolution, and Systematics*, **34**, 487–515.

Ferreira M.P., Alves D.S. & Shimabukuro Y.E. (2015) Forest dynamics and land-use transitions in the Brazilian Atlantic Forest: the case of sugarcane expansion. *Regional Environmental Change*, **15**, 365–377.

Fialho A.P., Oliveira L.G., Tejerina-Garro F.L. & Gomes L.C. (2007) Fish assemblage structure in tributaries of the Meia Ponte River, Goiás, Brazil. *Neotropical Ichthyology*, **5**, 53–60.

Filoso S., do Carmo J.B., Mardegan S.F., Lins S.R.M., Gomes T.F. & Martinelli L.A. (2015) Reassessing the environmental impacts of sugarcane ethanol production in Brazil to help meet sustainability goals. *Renewable and Sustainable Energy Reviews*, **52**, 1847-1856.

Gatz A.J. (1979) Ecological morphology of freshwater stream fishes. *Tulane Studies on Zoology and Botany*, **21**, 91–124.

Gido K.B. & Jackson D.A. (2010) *Community Ecology of Stream Fishes: Concepts, Approaches, and Techniques*. American Fisheries Society, New York.

Gosline W.A. (1971) *Functional morphology and classification of teleostean fishes*. Honolulu, University Press of Hawaii.

Gunkel G., Kosmol J., Sobral M., Rohn H., Montenegro S. & Aureliano J. (2007) Sugar cane industry as a source of water pollution – case study on the situation in Ipojuca River, Pernambuco, Brazil. *Water Air and Soil Pollution*, **180**, 261–269.

Harding J.S., Benfield E.F., Bolstad P.V., Helfman G.S. & Jones III E.B.D. (1998) Stream biodiversity: the ghost of land use past. *Proceedings of the National Academy of Sciences of United States of America*, **95**, 14843–14847.

Hoeinghaus D.J., Winemiller K.O. & Birnbaum J.S. (2007) Local and regional determinants of stream fish assemblage structure: inferences based on taxonomic versus functional groups. *Journal of Biogeography*, **34**, 324–338.

Hora S.L. (1930) Ecology, bionomics and evolution of the torrential fauna, with special reference to the organs of attachment. *Philosophical Transactions of the Royal Society of London, series B*, **218**, 172–282.

IEA (2012) *Bioenegy – A Sustainable and Reliable Energy Source – Main Report*. International Energy Agency, Paris.

Immerzeel D.J., Verweij P.A., van der Hilst F. & Faaij A.P.C. (2014) Biodiversity impacts of bioenergy crop production: a state-of-the-art review. *Global Change Biology Bioenergy*, **6**, 183–209.

IPT – Instituto de Pesquisas Tecnológica do Estado se São Paulo (1999) *Diagnóstico da situação atual dos Recursos Hídricos e estabelecimento de diretrizes técnicas para a elaboração do Plano da Bacia Hidrográfica do Turvo/Grande* – Relatório 40.515.

Iwata T., Nakano S. & Inoue M. (2003) Impacts of past riparian deforestation on stream communities in a tropical rain forest in Borneo. *Ecological Applications*, **13**, 461–473.

Jackson D.A. (1995) PROTEST: A PROcrustean randomization TEST of community environment concordance. *Ecoscience*, **2**, 297–303.

Jackson S.T. & Sax D.F. (2009) Balancing biodiversity in a changing environment: extinction debt, immigration credit and species turnover. *Trends in Ecology and Evolution*, **25**, 153–160.

Koizumi T. (2013) Biofuel and food security in China and Japan. *Renewable and Sustainable Energy Reviews*, **21**, 102–109.

Lapola D.M., Schaldach R., Alcamo J., Bondeau A., Koch J., Koelking C. *et al*. (2010) Indirect land-use changes can overcome carbon savings from biofuels in Brazil. *Proceedings of the National Academy of Sciences*, **107**, 3388–3393.

Lapola D.M., Martinelli L.A., Peres C.A., Ometto J.P.H.B., Ferreira M.E., Nobre C.A. *et al*. (2014) Pervasive transition of the Brazilian land-use system. *Nature Climate Change*, **4**, 27–35.

Leitão, R.P., Zuanon J., Mouillot D., Leal C.G., Hughes R.M., Kaufmann P.R. *et al*. (2017) Disentangling the pathways of land use impacts on the functional structure of fish assemblages in Amazon streams. *Ecography*, **40**, 1–13.Lorion C.M. & Kennedy B.P. (2009) Riparian forest buffers mitigate the effects of deforestation on fish assemblages in tropical headwater streams. *Ecological Applications*, **19**, 468–479.

Lu H., Lin B., Campbell D.E., Sagisaka M. & Ren H. (2012) Biofuel vs. biodiversity? Integrated emergy and economic cost-benefit evaluation of rice-ethanol production in Japan. *Energy*, **46**, 442–450.

Lucena C.A.S. & Soares H.G. (2016) Review of species of the *Astyanax bimaculatus* "caudal peduncle spot" subgroup sensu Garutti & Langeani (Characiformes, Characidae) from the rio La Plata and rio São Francisco drainages and coastal systems of southern Brazil and Uruguay. *Zootaxa*, **4072**, 101–125.

Mahon R. (1984) Divergent structure in fish taxocenes of north temperate streams. *Canadian Journal of Fisheries and Aquatic Sciences*, **41**, 330–350.

Manning P., Taylor G. & Hanley M.E. (2015) Bioenergy, food production and biodiversity – an unlikely alliance? *Global Change Biology Bioenergy*, **7**, 570–576.

Martinelli L.A. & Filoso S. (2008) Expansion of sugarcane ethanol production in Brazil: environmental and social challenges. *Ecological Applications*, **18**, 885–898.

Martinelli L.A., Naylor R., Vitousek P.M. & Moutinho P. (2010) Agriculture in Brazil: impacts, costs and opportunities for a sustainable future. *Current Opinion in Environmental Sustainability*, **2**, 431-438.

Matthews W.J., Marsh-Matthews E., Cashner R.C. & Gelwick F. (2013) Disturbance and trajectory of change in a fish community over four decades. *Oecologia*, **173**, 955–969.

McCann K.S. (2000) The diversity–stability debate. *Nature*, **405**, 228–233.

MEA (2005) *Ecosystems and Human Well-Being, Synthesis of the Millennium Ecosystem Assessment*. Island Press, Washington, D.C., USA.

Metzger J.L., Lewinsohn T.M., Joly C.A., Verdade L.M., Martinelli L.A. & Rodrigues R.R. (2010) Brazilian law: full speed in reverse? *Science*, **329**, 276–277.

Mlambo M.C., Paavola R., Louhi P., Soininen J., Virtanen R. & Muotka T. (2015) Bioenergy vs biodiversity: effects of intensive forest biomass removal on stream and riparian communities. *Forestry*, **88**, 368–375.

Naiman R.J. & Décamps H. (1997) The ecology of interfaces: riparian zones. *Annual Review of Ecology and Systematics*, **28**, 621–658.

Nalon M.A., Mattos I.F.A. & Franco G.A.D.C. (2008) Meio físico e aspectos da fragmentação da vegetação. In: *Diretrizes Para a Conservação e Restauração da Biodiversidade no Estado de São Paulo* (Eds R.R. Rodrigues & V.L.R. Bononi), pp. 17–21. Secretaria do Meio Ambiente e Instituto de Botânica, São Paulo.

Oliveira E.F., Goulart E., Breda L., Minte-Vera C.V., Paiva L.R.S. & Vismara M.R. (2010) Ecomorphological patterns of the fish assemblage in a tropical floodplain: effects of trophic, spatial and phylogenetic structures. *Neotropical Ichthyology*, **8**, 569–586.

Pardini R., Faria D., Accacio G.M., Laps R.R., Mariano-Neto E., Paciencia M.L.B. *et al.* (2009) The challenge of maintaining Atlantic forest biodiversity: a multi-taxa conservation assessment of specialist and generalist species in an agro-forestry mosaic in southern Bahia. *Conservation Biology*, **142**, 1178–1190.

Paula F.R., Gerhard P., Wenger S.J., Ferreira A., Vettorazzi C.A. & Ferraz S.F.B. (2013) Influence of forest cover on in-stream large wood in an agricultural landscape of southeastern Brazil: a multi-scale analysis. *Landscape Ecology*, **28**, 13–27.

Pease A.A., Taylor J.M., Winemiller K.O. & King R.S. (2011) Multiscale environmental influences on fish assemblage structure in Central Texas streams. *Transactions of the American Fisheries Society*, **140**, 1409–1427.

Peres-Neto P.R. & Jackson D.A. (2001) How well do multivariate datasets match? The advantages of a Procrustean superimposition approach over the Mantel test. *Oecologia*, **129**, 169–178.

Pimm S.L., Jenkins C.N., Abell R., Brooks T.M., Gittleman J.L., Joppa L.N. *et al.* (2014) The biodiversity of species and their rates of extinction, distribution, and protection. *Science*, **344**, 987–996.

Pool T.K., Grenouillet G. & Villéger, S. (2014) Species contribute differently to the taxonomic, functional, and phylogenetic alpa and beta diversity of freshwater fish communities. *Diversity and Distributions*, **20**, 1235–1244.

Pouilly M., Lino F., Bretenoux J.G. & Rosales C. (2003) Dietary-morphological relationships in a fish assemblage of the Bolivian Amazonian floodplain. *Journal of Fish Biology*, **62**, 1137–1158.

Purvis A., Agapow P.M., Gittleman J.C. & Mace G.M. (2000) Non-random extinction and the loss of evolutionary history. *Science*, **288**, 328–330.

Püttker T., Bueno A.A., Prado P.I. & Pardini R. (2015) Ecological filtering or random extinction? Beta-diversity patterns and the importance of niche-based and neutral processes following habitat loss. *Oikos*, **124**, 206–215.

Rudorff B.F.T., Aguiar D.A., Silva W.F., Sugawara L.M., Adami M. & Moreira M.A. (2010) Studies on the rapid expansion of sugarcane for ethanol production in São Paulo state (Brazil) using Landsat data. *Remote Sensing*, **2**, 1057–1076.

Santos F.B., Ferreira F.C. & Esteves K.E. (2015) Assessing the importance of the riparian zone for stream fish communities in a sugarcane dominated landscape (Piracicaba River Basin, Southeast Brazil). *Environmental Biology of Fishes*, **98**, 1895–1912.

Schiesari L. & Corrêa D.T. (2016) Consequences of agroindustrial sugarcane production to freshwater biodiversity. *Global Change Biology Bioenergy*, **8**, 644–657.

Scott M.C. (2006) Winners and losers among stream fishes in relation to land use legacies and urban development in the southeastern US. *Biological Conservation*, **127**, 301–309.

Scott M.C. & Helfman G.S. (2001) Native invasions, homogenization, and the mismeasure of integrity of fish assemblages. *Fisheries*, **26**, 6–15.

Silva A.M., Casatti L., Alvares C.A., Leite A.M., Martinelli L.A. & Durrant S.F. (2007) Soil loss risk and habitat quality in some streams of a Meso-Scale River Basin. *Scientia Agricola*, **64**, 336–343.

Smokorowski K.E. & Pratt T.C. (2007) Effect of a change in physical structure and cover on fish and fish habitat in freshwater ecosystems – a review and meta-analysis. *Environmental Reviews*, **15**, 15–41.

Soares-Filho B., Rajão R., Macedo M., Carneiro A., Costa W., Coe M. *et al*. (2014) Cracking Brazil's forest code. *Science*, **344**, 363–364.

Sparovek G., Berndes G., Barretto A.G.O.P. & Klug I.L.F. (2012) The revision of the Brazilian Forest Act: increased deforestation or a historic step towards balancing agricultural development and nature conservation? *Environmental Science and Policy*, **16**, 65–72.

Tilman D., Socolow R., Foley J.A., Hill J., Larson E., Lynd L. *et al*. (2009) Beneficial biofuels – the food, energy, and environment trilemma. *Science*, **325**, 270–271.

UNICA – Sugar cane industry union. (2016) *Production and use of fuel ethanol in Brazil: answers to the most frequently asked questions.* Available at: http://www.globalbioenergy.org/bioenergyinfo/background/detail/fi/c/2801/

Vellend M. (2010) Conceptual synthesis in community ecology. *Quarterly Review of Biology*, **85**, 183–206.

Verdade L.M., Gheler-Costa C., Penteado M. & Dotta G. (2012) The impacts of sugarcane expansion on wildlife in the state of São Paulo, Brazil. *Journal of Sustainable Bioenergy Systems*, **2**, 138–144.

Villéger S., Mason N.W.H. & Mouillot D. (2008) New multidimensional functional diversity indices for a multifaceted framework in functional ecology. *Ecology*, **89**, 2290–2301.

Wainwright P.C., Bellwood D.R. & Westneat M.W. (2002) Ecomorphology of locomotion in labrid fishes. *Environmental Biology of Fishes*, **65**, 47–62.

Ward-Campbell B.M.S., Beamish F.W.H. & Kongchaiya C. (2005) Morphological characteristics in relation to diet in five coexisting Thai fish species. J*ournal of Fish Biology*, **67**, 1266–1279.

Waters C.N., Zalasiewicz J., Summerhayes C., Barnosky A.D., Poirier C., Gałuszka A. *et al*. (2016) The Anthropocene is functionally and stratighaphically distinct from Holocene. *Science*, **351**, 137–147.

Watson D.J. & Balon E.K. (1984) Ecomorphological analysis of fish taxocenes in rainforest streams of northern Borneo. *Journal of Fish Biology*, **25**, 371–384.

Wilga C.D. & Lauder G.V. (1999) Locomotion in sturgeon: function of the pectoral fins. *Journal of Experimental Biology*, **202**, 2413–2432.

Willis S.C., Winemiller K.O. & Lopez-Fernandez H. (2005) Habitat structural complexity and morphological diversity of fish assemblages in a Neotropical floodplain river. *Oecologia*, **142**, 284–295.

Winemiller K.O. (1991) Ecomorphological diversification in lowland freshwater fish assemblages from five biotic regions. *Ecological Monographs*, **61**, 343–365.

Fig. S1. UPGMA cluster of 54 fish species based on 15 functional attributes yielding six functional groups. Group A includes all Characiformes, except *Hoplias malabaribus*, and *Melanorivulus pictus* (Cyprinodontiformes: Rivulidae), and *Hoploternum littorale* (Siluriformes: Callichthyidae). Group B includes two species of *Hypostomus* (Siluriformes: Loricariidae) and *Aspidoras fuscoguttatus* and *Corydoras aeneus* (Siluriformes: Callichthyidae). Group C includes Heptapteridae family (Siluriformes),

Callichthys callichthys (Siluriformes: Callichthyidae), *Hoplias malabaricus* (Characiformes: Erythrinidae), *Pseudopimelodus pulcher* (Siluriformes: Pseudopimelodidae) and the two species of *Crenicichla* (Perciformes: Cichlidae). Group D includes the others Cichlidae species (Perciformes). Group E includes *Gymnotus sylvius* (Gymnotiformes: Gymnotidae) and *Eigenmannia trilineata* (Gymnotiformes: Sternopygidae). Group F includes two non-native Poeciliidae, *Poecilia reticulata* and *Xiphophorus maculatus* (Cyprinodontiformes). The pictures represent the body shapes most associated with each functional group. Two groups (A and C) have more than one picture because of the comparatively higher diversity of body shape within each group.

Fig. S2. PCoA bi-plot calculated using combined attributes (watershed land use and instream/ecotone) for 38 stream reaches in 2003 and 2013. Arrows link site scores for each stream between periods, starting at the score in 2003 and with the arrow head at the score for 2013. Arrow color indicates increasing (green) or decreasing (red) species richness over time, and arrow thickness represents the relative magnitude of change in species richness. Labels for environmental factors are centered on their respective axis loadings.
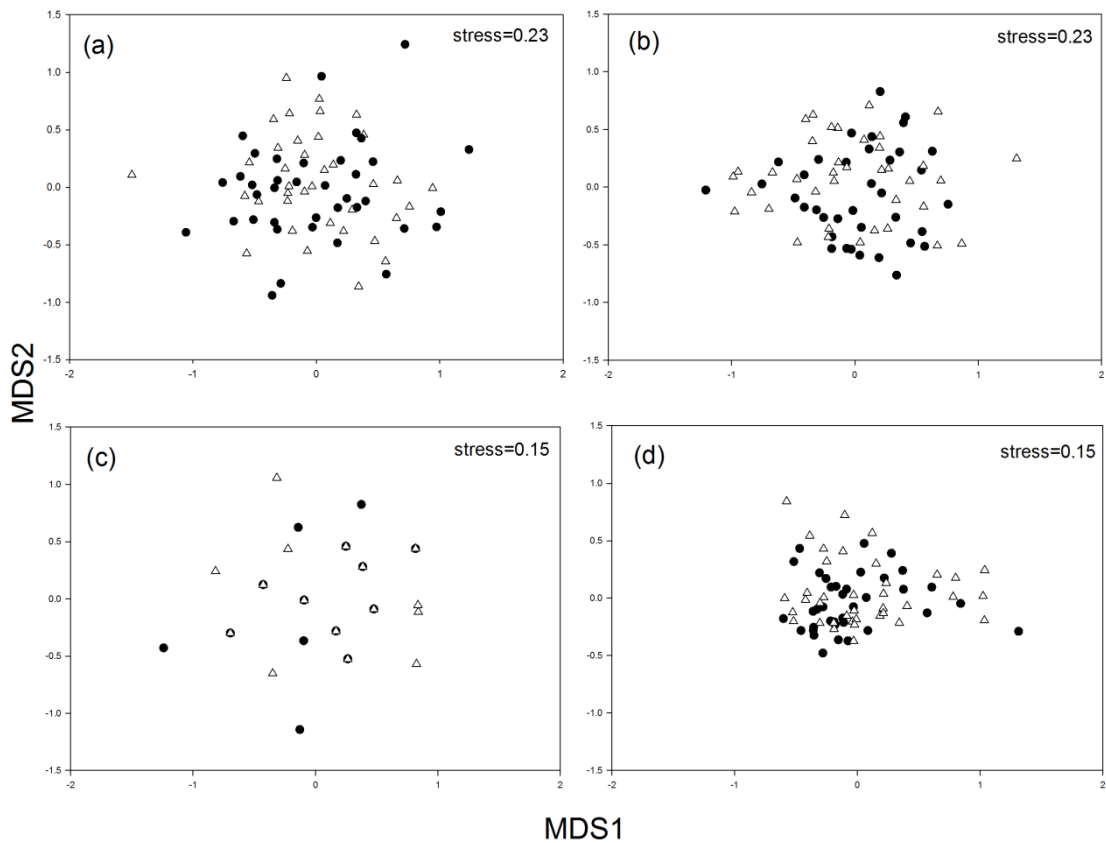
Fig. S3. NMDS ordination of taxonomic and functional fish assemblage structure in 2003 (black circle) and 2013 (open triangle). (a) taxonomic structure based on presence and absence of species; (b) taxonomic structure based on relative abundances; (c) functional structure based on presence and absence of functional groups and (d) functional structure based on relative abundances of functional groups.

Table S1. Species scores on the first two axes of functional PCoA based on 15 ecomorphological attributes.

Order	Species	PC1	PC2
Characiformes	*Acestrorhynchus lacustris*	0.536	-0.021
	Leporinus friderici	0.163	-0.365
	Leporinus lacustris	0.103	-0.452
	Leporinus paranaensis	0.032	-0.378
	Leporinus striatus	0.377	-0.075
	Astyanax lacustris	0.348	-1.045
	Astyanax fasciatus	0.498	-0.710
	Astyanax schubarti	0.155	-0.506
	Astyanax paranae	0.240	-1.010
	Bryconamericus stramineus	0.489	0.080
	Hemigrammus marginatus	0.525	-0.871
	Hyphessobrycon anisitsi	0.355	-0.752
	Hyphessobrycon eques	0.118	-1.039
	Knodus moenkhausii	0.475	-0.062
	Moenkhausia sanctaefilomenae	-0.066	-0.778
	Oligosarcus pintoi	0.358	-0.658
	Piabina argentea	0.361	0.227
	Planaltina britskii	0.717	0.128
	Roeboides descalvadensis	0.563	-0.974
	Serrapinnus notomelas	0.513	-0.761
	Cyphocharax modestus	0.027	-0.547
	Cyphocharax vanderi	0.109	-0.189
	Steindachnerina insculpta	0.291	-0.314
	Characidium zebra	0.184	0.762
	Characidium lagosantense	0.121	-0.268
	Hoplias malabaricus	-0.167	0.557
	Pyrrhulina australis	0.300	0.023
	Apareiodon affinis	0.439	0.444
	Apareiodon piracicabae	0.345	0.301
	Parodon nasus	0.239	0.420
Cyprinodontiformes	*Poecilia reticulata*	-0.537	0.242
	Xiphophorus maculatus	-0.975	-0.603
	Melanorivulus pictus	0.250	0.378
Gymnotiformes	*Gymnotus sylvius*	2.602	0.646
	Eigenmannia trilineata	2.237	0.788
Perciformes	*Cichlasoma paranaense*	-1.345	-0.725
	Crenicichla britskii	-0.342	0.206
	Crenicichla cf. *semifasciata*	-0.411	0.358
	Geophagus brasiliensis	-1.084	-0.394
	Laetacara araguaiae	-1.183	-0.407
	Oreochromis niloticus	-0.727	-0.551
	Satanoperca pappaterra	- 1.057	-0.779

	Tilapia rendalli	-0.574	-0.940
Siluriformes	*Aspidoras fuscoguttatus*	-0.741	0.829
	Callichthys callichthys	-0.301	0.896
	Corydoras aeneus	-0.910	0.299
	Hoplosternum littorale	-0.527	-0.188
	Hypostomus ancistroides	-0.962	1.153
	Hypostomus cf. *nigromaculatus*	-1.254	1.433
	Cetopsorhamdia iheringi	-0.142	1.682
	Imparfinis schubarti	-0.003	1.448
	Pimelodella avanhandavae	0.105	1.436
	Rhamdia quelen	-0.380	1.108
	Pseudopimelodus pulcher	-0.493	0.521

Table S2. Residuals of the relationship between watershed land use and instream variables for 2003 and 2013. High values indicate poor fit between watershed land use and instream habitat.

Residuals	2003	2013
R5	1.32	1.60
R26	1.28	0.89
R8	1.23	0.44
R22	1.12	0.26
R56	1.10	0.56
R23	1.09	1.27
R11	0.99	0.47
R41	0.97	0.78
R17	0.95	0.68
R24	0.93	0.10
R27	0.92	0.73
R25	0.91	0.66
R21	0.87	0.73
R55	0.82	0.30
R40	0.80	0.15
R9	0.76	0.34
R38	0.75	1.66
R16	0.73	1.43
R37	0.69	0.91
R47	0.68	0.21
R43	0.64	1.03
R51	0.64	0.37
R13	0.61	0.29
R34	0.58	0.28
R14	0.52	0.56
R49	0.52	0.65
R53	0.50	0.95
R10	0.49	0.85
R3	0.48	1.03
R48	0.47	0.94
R45	0.38	0.09
R54	0.37	0.47
R6	0.36	0.75
R31	0.34	0.71
R12	0.33	0.95
R46	0.24	0.46
R44	0.23	0.45
R18	0.20	0.58

Table S3. Taxonomic classification and total abundances (i.e. combined across all 38 streams) of the 54 species collected in 2003 and 2013. * non-native species.

Order	Family	Species and author	Abundance 2003	2013
Characiformes	Acestrorhynchidae	*Acestrorhynchus lacustris* (Lütken, 1875)	1	1
	Anostomidae	*Leporinus friderici* (Bloch, 1794)	7	2
		Leporinus lacustris Amaral Campos, 1946	0	1
		Leporinus paranaensis Garavello & Britski, 1987	1	0
		Leporinus striatus Kner, 1858	1	0
	Characidae	*Astyanax lacustris* (Lütken 1875)	1,926	1,113
		Astyanax fasciatus (Cuvier, 1819)	179	122
		Astyanax schubarti Britski, 1964	0	4
		Astyanax paranae Eigenmann, 1914	2	0
		Bryconamericus stramineus Eigenmann, 1908	13	10
		Hemigrammus marginatus Ellis, 1911	152	130
		Hyphessobrycon anisitsi (Eigenmann, 1907)	0	18
		Hyphessobrycon eques (Steindachner, 1882)	4	4
		Knodus moenkhausii (Eigenmann & Kennedy, 1903)	1,347	886
		Moenkhausia sanctaefilomenae (Steindachner, 1907)	31	32
		Oligosarcus pintoi Campos, 1945	332	251
		Piabina argentea Reinhardt, 1867	121	160
		Planaltina britskii Menezes,Weitzman & Burns, 2003	3	0
		Roeboides descalvadensis Fowler, 1932	0	2
		Serrapinnus notomelas (Eigenmann, 1915)	329	321
	Curimatidae	*Cyphocharax modestus* (Fernández-Yépez, 1948)	8	0
		Cyphocharax vanderi (Britski, 1980)	71	134
		Steindachnerina insculpta (Fernández-Yépez, 1948)	18	1
	Crenuchidae	*Characidium zebra* Eigenmann, 1909	141	50
		Characidium lagosantense Travassos, 1947	5	0
	Erythrinidae	*Hoplias malabaricus* (Bloch, 1794)	50	43
	Lebiasinidae	*Pyrrhulina australis* Eigenmann & Kennedy, 1903	84	27
	Parodontidae	*Apareiodon affinis* (Steindachner, 1879)	0	1
		Apareiodon piracicabae (Eigenmann, 1907)	3	0
		Parodon nasus Kner, 1858	100	30
Cyprinodontiformes	Poeciliidae	*Poecilia reticulata* Peters, 1859*	1,199	2,651
		Xiphophorus maculatus (Günther, 1866)*	0	108

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	Rivulidae	*Melanorivulus pictus* (Costa, 1989)	5	8
Gymnotiformes	Gymnotidae	*Gymnotus sylvius* Albert & Fernandes-Matioli, 1999	173	162
	Sternopygidae	*Eigenmannia trilineata* López & Castello, 1966	9	2
Perciformes	Cichlidae	*Cichlasoma paranaense* Kullander, 1983	55	68
		Crenicichla britskii Kullander, 1982	54	29
		Crenicichla semifasciata (Heckel, 1840)	0	22
		Geophagus brasiliensis (Quoy & Gaimard, 1824)	27	42
		Laetacara araguaiae Ottoni & Costa, 2009	120	73
		Oreochromis niloticus (Linnaeus, 1758)*	1	0
		Satanoperca pappaterra (Heckel, 1840)	24	41
		Tilapia rendalli (Boulenger, 1897)*	8	0
Siluriformes	Callichthyidae	*Aspidoras fuscoguttatus* Nijssen & Isbrücker, 1976	256	264
		Callichthys callichthys (Linnaeus, 1758)	4	1
		Corydoras aeneus (Gill, 1858)	84	273
		Hoplosternum littorale (Hancock, 1828)	2	12
	Heptapteridae	*Cetopsorhamdia iheringi* Schubart & Gomes, 1959	2	7
		Imparfinis schubarti (Gomes, 1956)	112	129
		Pimelodella avanhandavae Eigenmann, 1917	11	0
		Rhamdia quelen (Quoy & Gaimard, 1824)	78	72
	Loricariidae	*Hypostomus ancistroides* (Ihering, 1911)	182	136
		Hypostomus cf. *nigromaculatus* (Schubart, 1964)	98	80
	Pseudopimelodidae	*Pseudopimelodus pulcher* (Boulenger, 1887)	6	12
		TOTAL	7,439	7,535

CHAPTER 2

LAND USE CHANGES AND SPATIAL-TEMPORAL BETA DIVERSITY OF FRESHWATER FISH FROM AN AGRICULTURAL REGION

Summary

1. Environmental changes and stochasticity can influence regional patterns of beta diversity over space and time. Our aim was to investigate how spatial and temporal beta diversity (turnover and nestedness) of fish assemblages in agroecosystem streams can respond to regional (watershed land use) and local (instream features) changes. Moreover, we investigated whether the observed pattern of temporal beta diversity was different from that one expected by chance.

2. We sampled 38 agroecosystem streams ten years apart - in 2003 and 2013 - and quantified regional and local changes. After that, we obtained spatial and temporal turnover and nestedness of species and functional groups. Based on null assemblages (null model = independent swap), we calculated the null temporal beta diversity and compare with our observed values using a T-test.

3. Taxonomic and functional spatial beta diversity in both periods were mostly due to turnover. Regarding temporal beta dissimilarity, turnover was the main responsible for species dissimilarity, while nestedness was more important to functional groups dissimilarity. Contrary to our hypothesis, beta diversity and its components were not associated with the degree of environmental changes. Only the functional turnover was positively correlated to regional changes, though weak relationship. However, while temporal nestedness did not differ from the null mean distribution, turnover was lower than expected by chance.

4. Probably, deforestation and the development of agriculture led to regional (landscape) and local (stream physical habitat) homogenization, and consequently to homogenized fish assemblages by selecting habitat-generalist species, which are also highly resistant to further environmental changes. Moreover, environment and assemblages could show a time lag response to the environmental changes greater than our time scale (ten years).

This study helps addressing this knowledge gap by assessing how further environmental changes in established agroecosystems affect spatial and temporal beta diversity of stream fish assemblages.

Keywords: turnover, nestedness, agroecosystems, temporal dissimilarity, null beta diversity.

Introduction

Birth, death, immigration and emigration rates can naturally vary over space and time (Mittelback, 2010). For this reason, assemblage composition is often dynamic. In addition to natural fluctuations, anthropogenic environmental changes have caused a widespread pattern of species extinction (Dirzo *et al*., 2014; Waters *et al*., 2016) and biological homogenization (McKinney & Lockwood, 1999; Rahel, 2002). Consequently, human-induced processes have drastically changed previous patterns of species and functional group distributions (Young *et al*., 2016). In this context, beta diversity represents an effective tool to identify how these environmental changes produce different patterns of species dissimilarity (Socolar *et al*., 2016). First proposed by Whittaker (1960), beta diversity quantifies changes in species composition among sites along an environmental gradient or in a given region. Recently, Baselga (2010) proposed a method of beta diversity partitioning into two additive components: turnover and nestedness. According to the authors, differentiation between turnover and nestedness can improve our understanding of how different processes (biogeographic, historic and ecological) act to produce similar and/or distinct species composition through space and time. Turnover is the replacement of species among sites and can be result of niche and dispersal processes, while nestedness represents a non-random gain or loss of species

among sites due to selective colonization/extinction or habitat nestedness. By identifying the underlying processes, beta diversity partitioning can be useful for conservation and management of wildlife, since high turnover across an area may indicate that effective conservation measures should consider a larger number of sites, while high nestedness suggests conservation can target the richest site in the region (Socolar *et al*., 2016).

Agroecosystems are the product of the conversion of natural into agricultural ecosystems mediated by human activities (Conway, 1987). According to Newbold *et al*. (2016), assemblages in these human-altered systems show remarkable differences in species composition when compared to native environments. Such differences in species composition are likely due to environmental filtering associated with habitat loss/change, which jeopardizes the occurrence of sensitive species or functional groups while favoring taxa suited for the new environmental conditions (Scott & Helfman, 2001). Moreover, since most agroecosystems are comprised by monocultures, a large region can experience similar environmental filtering that leads to similar species and functional composition across multiple sites. Over time, this process contributes to decreased beta diversity, which is a phenomenon known as biological homogenization (Flynn *et al*., 2009; Karp *et al*., 2012; Segre *et al*., 2014). For example, watersheds dominated by pasture show similar instream features, such as marginal grasses, sandy substrate and runs (Casatti *et al*., 2006). Consequently, taxonomic and functional composition of aquatic organisms can be very similar among sites within watersheds experiencing this same land use (Casatti, Ferreira & Casatti, 2009; Casatti *et al*., 2015).

Thus, deterministic changes in species composition caused by land-use change can homogenize assemblages (McKinney & Lockwood, 1999). However, random birth and death rates and unpredictable environmental fluctuations (i.e. stochasticity) can influence the patterns of spatial and temporal assemblage dynamics (Engen, Bakke &

Islam, 1998, Kalyuzhny *et al*., 2014). Recently Baselga, Bonthoux & Balent (2015) demonstrated stochastic processes could drive temporal beta diversity of bird assemblages in France. In that case, observed changes in species composition over time were likely due to appearance and disappearance of populations from specific localities in a random way. In plant assemblages, random environmental disturbance increased beta diversity when dispersal was limited, but it was not a key driver when dispersal was high (Catano, Dickson & Myers, 2017). Therefore, not only deterministic (e.g. species-specific responses to environmental change), but also stochastic processes can shape the current beta diversity patterns in a region. Identification of the relative importance of deterministic vs. stochastic processes on beta diversity can inform ecosystem management (Kalyuzhny *et al*., 2014).

More than 50% of the Earth's surface has already been transformed into agricultural and urban lands (Ellis & Ramankutty, 2008), accompanied by widespread homogenization of local communities (Rahel, 2002). Therefore, it is crucial to understand whether and how further environmental changes in agroecosystems can affect assemblages (Kessler *et al*., 2009). In this context, streams from São Paulo state, Brazil, represent a useful system to understand the influence of regional and local changes in spatial and temporal beta diversity and its components (turnover and nestedness). In this region, native semi-deciduous seasonal forests (a subtype of Atlantic Rainforest) and savannah (Cerrado) biomes were replaced by agriculture more than a century ago (Monbeig, 1988) and recent land use change is due to the conversion of pasture to sugarcane (Lapola *et al*., 2014).

Thus, our aim was to investigate how spatial and temporal beta diversity of fish assemblages from agroecosystems streams changed in response to recent regional and local land-use changes. Furthermore, we investigated whether the observed pattern of

temporal beta diversity was different from the one expected by chance. To achieve our aims, we sampled 38 agroecosystem streams ten years apart (2003 and 2013) and quantified spatial (within years) and temporal (between years) beta diversity and its components (turnover and nestedness). Working with the same dataset, Zeni, Hoeinghaus & Casatti (first chapter in this dissertation and currently in review), demonstrated that taxonomic and functional changes were not correlated to the degree of regional and local changes. However, the underlying mechanism shaping local (alpha) and regional (beta) biodiversity patterns can be different (Whittaker, Willis & Field, 2001). Our hypothesis was that environmental changes at regional and local scales associated with the conversion of pasture to sugarcane would impose a different environmental filter (than pasture) and spatial-temporal beta diversity patterns will be driven by deterministic factors.

Material and Methods

Study area

The study area is located in the São José dos Dourados and Turvo-Grande river basins of Northwest of São Paulo state, southeastern Brazil. Soils are characterized by unconsolidated sand and clay, with high erosive potential (Silva *et al*., 2007). Since the early 20th century, native Atlantic Forest and Cerrado have been replaced by agricultural crops, such as coffee, citrus and livestock grazing (Silva *et al*., 2007). Due to this old and intense anthropogenic disturbance, the region is one of the most deforested areas in Brazil with less than 4% of native forest concentrated in small, disconnected and highly degraded fragments (Nalon, Mattos & Franco, 2008). Similar to the terrestrial environment, agriculture also left strong imprints in aquatic systems, since most of them show massive changes to instream habitat and fish assemblages (Casatti *et al*., 2006;

Casatti *et al.*, 2009; Casatti *et al.*, 2015; Roa-Fuentes & Casatti, 2017). Further land use changes in this region are mainly caused by the replacement of one agricultural crop to another currently more profitable crop. In the past decade, sugarcane (*Saccharum* spp.) for biofuel production has been replacing pasture as the dominant matrix (Lapola *et al.*, 2014) (Fig. 1).
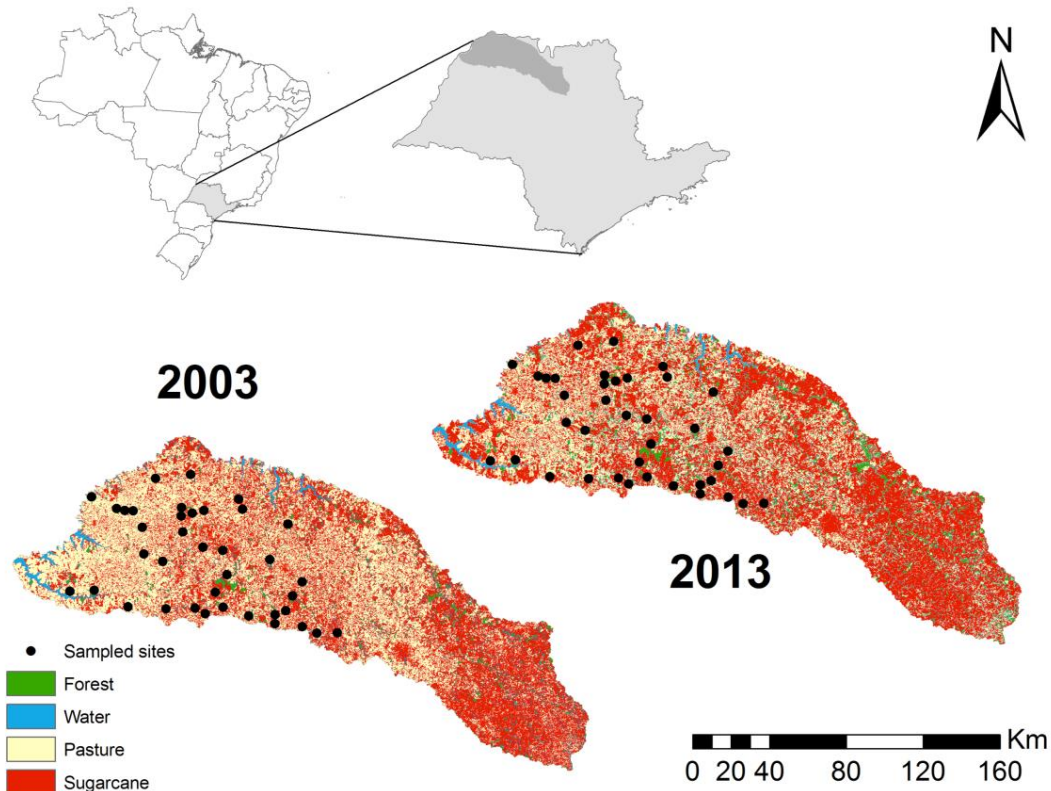
Fig. 1 Stream reaches (black circles) and land use (forest, pasture, and sugarcane) in the study area, São Paulo State, Brazil (Figure from Zeni *et al.* in review).

Environmental variables

To obtain environmental changes between the periods, we quantified regional (watershed land use) and local (instream) variables in 2003 and 2013. For each watershed (regional variables), we defined four land use classes: (1) native forest - areas occupied by semi-deciduous seasonal forest and Cerrado; (2) pasture - area used for livestock

grazing; (3) sugarcane - area occupied by sugarcane crops mainly used for biofuel production; and (4) other - includes areas with perennial and annual cultivation, rural facilities and roads. Land use classes were obtained from analysis of Landsat 5 TM satellite images (30x30 m^2 resolution), which were available from the National Institute for Space Research (INPE). To quantify the percentage of each land use class, we used images from 2003 and 2011 (for our 2013 samples), primarily due to the availability of images. However, changes in the area occupied by sugarcane in the region between 2011 and 2013 were minimal.

We characterized instream habitat and ecotone attributes (local variables) at three transects located upper, middle and downstream of each 75 m stream reach. Local variables in 2013 were quantified using the same sampling protocol applied in 2003 (Table 1). The degree of regional and local environmental changes between 2003 and 2013 were obtained for each stream using pairwise Euclidean distances in Principal Coordinates Analysis (PCoA).

Table 1. Instream variables, description of how they were obtained and their ecological significance.

Variables	Obtention	Ecological significance
Physical Habitat Index (PHI)	Habitat assessment based on visual quantification of mesohabitat, substrate, channel modification, and stream bank (Casatti *et al*., 2006).	Higher values indicate more heterogeneous and structured physical habitat and it can reflect more diversified fish assemblages (Casatti *et al*., 2006).
Tree	Visual assessment of the percent of trees in the 10 m width riparian strip.	Trees can reflect the amount and quality of riparian forest, which is widely associated with several aspects physical habitat and fish assemblages (Pusey & Arthington, 2003; Allan, 2004).
Shrubs	Visual assessment of the percent of shrubs in the 10 m width riparian strip.	Shrubs can indicate the amount and quality of riparian forest, which is also associated with several aspects physical habitat and fish assemblages (Pusey & Arthington, 2003; Allan, 2004).

Substrate composition	Visual evaluation at several points along the stream reach of the percent of the stream bottom occupied by unconsolidated (silt, clay and sand) versus consolidated (gravel, cobble, rock and slab) material.	The presence and amount of unconsolidated substrate are usually associated with siltation. And, siltation has been linked to biological homogenization, since can act as an environmental filter extirpating species that are substrate dependent (Burdon, McIntosh & Harding, 2013).
Wood debris (WD)	Visual estimation of the percent of instream habitat occupied by wood debris	The presence of wood debris can create different mesohabitats and feeding sites, increasing niche availability and providing new species establishment opportunities (Teresa & Casatti, 2012)
Grasses	Visual estimation of the percent of both banks occupied by grass (mostly invasive signalgrass, *Brachiaria* spp.), which is widely related to pasture.	Grasses dominance is associated with physical habitat and biological homogenization of stream (Casatti *et al.*, 2009).
Mesohabitat	Visual estimation of percent of the stream reach comprised pools, riffles and runs.	Higher diversity of mesohabitats can support a richer and more diversified fish assemblages (Teresa & Casatti, 2012), because several fish species are associated with different water column depth and water velocity.
Water velocity	Water velocity was measure with a flowmeter at nine points (three from each transect). After that, we used these values to calculate average and standard deviation of velocity in each stream reach.	Water velocity average can indicate the main mesohabitat in the stream, while standard deviation can indicate the variation of the velocity along stream reach. Both can influence fish assemblages (Teresa & Casatti, 2012).
Depth	It was measured at 15 points (five from each transect) and the values were used to calculate depth average and standard deviation of each stream reach.	Deeper streams can have more habitats and increase niche diversity and availability (Connor & McCoy, 2001).
Width	It was measured at each transect and the values were used to calculate width average and standard deviation of each stream reach.	Wider streams can have more habitats, probably increasing niche diversity and availability (Connor & McCoy, 2001).
Stream area	It was estimated as the product of reach length (75 meters) and average width	High stream area can provide high habitat availability and diversity and, consequently it can increase niche availability (Connor & McCoy, 2001)
Stream volume	It was estimated as the product of stream area and average depth	High water volume may support more habitat types and increase the niche availability and diversity (Connor & McCoy, 2001)

Fish assemblages and functional groups

To sample fish assemblages, we isolated upstream and downstream reach limits with block nets (5 mm mesh) and used two electrofishing passes (generator 220 V, 50-60 Hz, 3.4-4.1 A, 1000 W). Fish specimens were fixed in a 10% formaldehyde solution in the field and transferred to 70% ethanol after 48 hours. The same methodology was employed in both sampling periods. We identified a total of 54 species across all samples from the two periods (Table S1). Vouchers were deposited in the collection of the Department of Zoology and Botany at the Universidade Estadual Paulista "Júlio de Mesquita Filho" campus of São José do Rio Preto, São Paulo (DZSJRP).

To assess functional aspects of fish assemblages, we took linear distances and areas from 10 adult individuals for each species (randomly chosen from across all sampling locations) to obtain 15 ecomorphological traits (Table S2). Functional traits were associated with habitat and resource use. Based on the mean of the functional traits distances (i.e. Euclidean), we used hierarchical agglomerative clustering (UPGMA) (cophenetic coefficient = 0.82; Fig.S1) and assigned species to one of six functional groups (see Chapter 1). The most diverse and abundant group (Group A) represented water column explorers (nektonic species) with higher compression index (CI) and relative depth (RD). A second group (Group B) was comprised by species that primarily forage in the stream bottom with lower CI, higher relative dorsal (RAD) and pectoral fin (RAPt) area, and relatively ventral mouth position and dorsal eyes (benthic species). Two more groups (Group C and D) associated with deeper lentic mesohabitats were distinguished by higher relative mouth width (RWM) and eye position (EP), and species with lower fineness coefficient (FC), higher RAD, RD and relative length of head (RLHd). Gymnotiform species (Group E) (i.e. Neotropical knifefishes) associated with marginal grasses were in a separate group, distinguished by fusiform bodies, absence of

caudal fin and an elongated anal fin. Final group (Group F) was comprised by two non-native species generally associated with slow water velocity with small body size, higher relative caudal area (RAC) and depression index (DI) (Fig. S1). Clustering and cophenetic correlations were performed using *stats* in R version 3.03.

Spatial, temporal and null beta diversity

We calculated spatial and temporal beta diversity using Sorensen's dissimilarity index based on species and functional groups composition (presence-absence data) (Baselga, 2010). We partitioned global beta diversity (β_{SOR}) into turnover (β_{SIM}) and nestedness (β_{SNE}) components. To assess beta diversity spatial patterns, we obtained the multiple-site dissimilarity for 2003 and 2013 using *beta.multi* (Baselga *et al*., 2015). This procedure gave us an average value for global beta diversity as well as the turnover and nestedness components in both periods. To test for differences in multiple-site dissimilarity distribution over time, we obtained 1000 multiple-site dissimilarity values randomly generated using *beta.sample* (Baselga *et al*., 2015).

Even if multiple-site dissimilarity does not change, pairwise dissimilarity can dramatically change if the same pair of streams show different patterns in turnover and nestedness over time. Mantel test (one for turnover and another for nestedness) was used to test the concordance between pairwise dissimilarity patterns (Anderson *et al*., 2011). We investigated the role of environmental changes in the observed changes of pairwise patterns. First, we calculated pairwise Euclidian distances (using regional and instream variables) in 2003 and 2013. Second, to obtain the degree of change in environmental dissimilarity (ΔEnv) for each pair of streams, we subtracted the values of dissimilarity in 2013 from those in 2003. Based on this procedure, we identified a gradient in pairwise similarity over time. Next, we obtained the degree of change in turnover ($\Delta\beta_{SIM}$) and

nestedness ($\Delta\beta_{SNE}$) components for each pair of streams, following the same procedure applied to the degree of change in environmental dissimilarity. Finally, we used linear regression to test for a relationship between degree of change in environmental dissimilarity (ΔEnv) and degree of change in turnover and nestedness ($\Delta\beta_{SIM}$ and $\Delta\beta_{SNE}$).

For temporal beta diversity, we quantified pairwise dissimilarity in species and functional groups composition between streams in 2003 and 2013 using *beta.temp* (Baselga *et al*., 2015). To evaluate the role of environmental changes (regional and local) in fish species and functional groups dissimilarity, we conducted a linear regression between the degree of regional and local changes (obtained through PCoA) and temporal beta diversity and its components (turnover and nestedness). Based on this approach, we can identify if temporal beta diversity components were correlated to regional and/or local changes. To compare if the observed temporal beta diversity was different from the null pattern, we generated null assemblages for 2003 and 2013 using 999 replicates (null model = independent swap). Based on these null assemblages, we obtained the null temporal beta diversity and its components. To test for differences between observed values (based on our sampled assemblages) and null estimate, we used a t-test. The same protocol was used for fish species and functional groups. This procedure identified if our observed values for temporal beta diversity, turnover, and nestedness were greater, equal or smaller than expected by chance (Baselga *et al*., 2015).

Results

Spatial multiple-site dissimilarity patterns in both periods were mostly due to turnover of species. In 2003, the average of taxonomic turnover was 0.67 ($\sigma = 0.02$), while nestedness was 0.09 ($\sigma = 0.02$). For 2013, the values were almost identical: the average of turnover was 0.68 ($\sigma = 0.02$) and nestedness was 0.09 ($\sigma = 0.03$) (Fig. 2). For

functional groups, dissimilarity patterns were somewhat different between periods. In 2003, functional groups dissimilarity was due to approximately equal parts of turnover (average = 0.25; σ = 0.09) and nestedness (average = 0.27; σ = 0.05). In 2013, turnover (average = 0.37; σ = 0.09) accounted for twice as much of the total beta diversity as nestedness (average = 0.17; σ = 0.06) (Fig. 3). However, there were no significant differences between spatial turnover and nestedness patterns in taxonomic or functional dissimilarity between years (p value > 0.05) (Fig. 2 and 3).

Even though regional dissimilarity patterns remained unchanged, spatial pairwise dissimilarity structure was different between 2003 and 2013. We identified low correlation between taxonomic and functional dissimilarity matrices (mantel statistic r), indicating that stream pairs changed in turnover and nestedness patterns over time (Table 2). For most of cases, observed changes were not correlated with the degree of changes in regional and local environmental dissimilarity (Table 3).
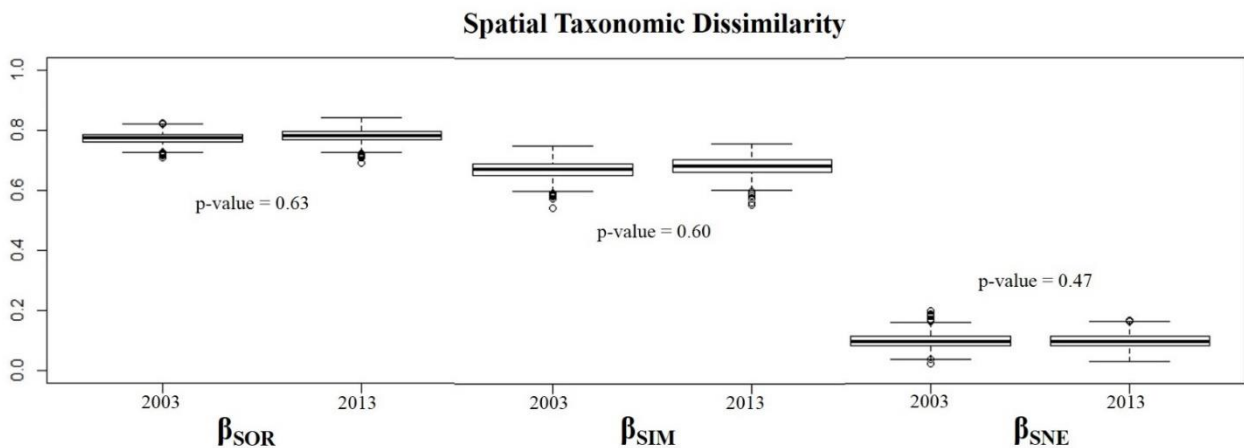
Fig. 2 Spatial beta diversity (β_{SOR}), turnover (β_{SIM}) and nestedness (β_{SNE}) of species in 2003 and 2013.
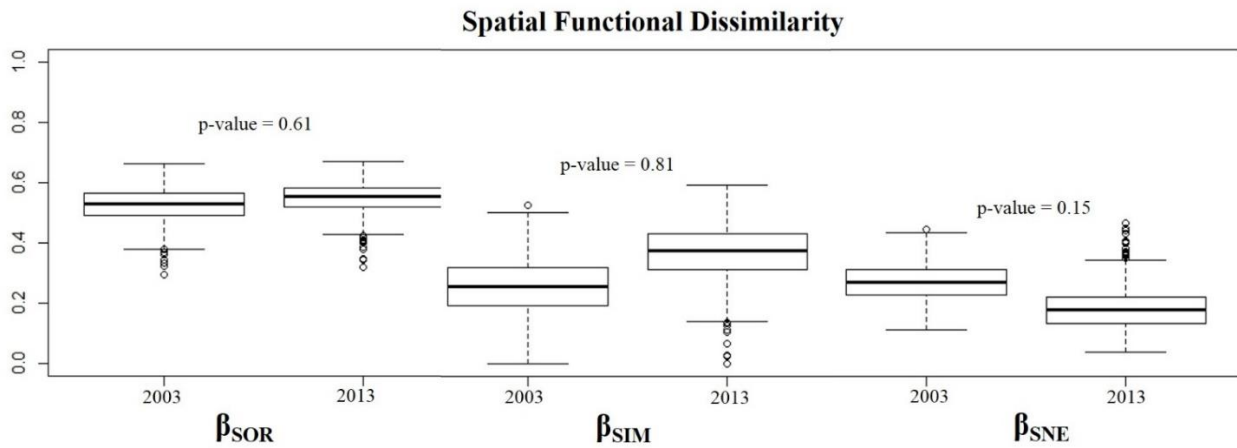
Fig. 3 Spatial beta diversity (β_{SOR}), turnover (β_{SIM}) and nestedness (β_{SNE}) of functional groups in 2003 and 2013.

Table 2. Mantel statistic r and p-value for taxonomic and functional turnover (β_{SIM}) and nestedness (β_{SNE}) components.

	Mantel statistic r	p-value
Taxonomic β_{SIM}	**0.33**	**0.001**
Taxonomic β_{SNE}	**0.22**	**0.001**
Functional β_{SIM}	**0.23**	**0.001**
Functional β_{SNE}	0.05	0.210

Table 3. Regression between degree of environmental changes at regional and local scales and degree of changes in pairwise turnover ($\Delta\beta_{SIM}$) and nestedness ($\Delta\beta_{SNE}$) for taxonomic (Taxo) and functional (Func) data.

	ΔEnv	$\Delta\beta_{SIM}$		$\Delta\beta_{SNE}$	
		p-value	R-square	p-value	R-square
Taxo	Regional	**0.02**	**0.00**	**0.00**	**0.02**
	Instream	0.11	0.00	0.25	0.00
Func	Regional	0.15	0.00	0.61	0.00
	Instream	0.11	0.00	0.27	0.00

Temporal beta diversity (β_{SOR}) for species (mean = 0.33; σ = 0.16) and functional groups (mean = 0.14; σ = 0.12) were relatively low and the mechanisms behind the

patterns were different for taxonomic and functional structure. Turnover was the main component responsible (65%) for temporal species dissimilarity between 2003 and 2013 (β_{SIM} average = 0.22 and σ = 0.16; β_{SNE} average = 0.11 and σ = 0.12), whereas nestedness was responsible for 67% of functional groups dissimilarity (β_{SNE} average = 0.10 and σ = 0.11; β_{SIM} average = 0.05 and σ = 0.11). Thus, species replacement coincided with loss or gain of functional groups over time. Temporal beta diversity, turnover and nestedness were not associated with the degree of regional or local environmental changes. Only functional turnover was positively correlated with regional changes, but the relationship was very weak (Table 4).

Table 4. Regression between degree of regional and local changes and temporal beta diversity (β_{SOR}), turnover (β_{SIM}) and nestedness (β_{SNE}) for taxonomic (Taxo) and functional (Func) data.

		βSOR		βSIM		βSNE	
		p-value	R-square	p-value	R-square	p-value	R-square
Taxo	Regional	0.28	0.03	0.07	0.08	0.49	0.01
	Local	0.12	0.06	0.31	0.02	0.50	0.01
Func	Regional	0.22	0.04	**0.02**	**0.12**	0.51	0.01
	Local	0.94	0.00	0.44	0.01	0.50	0.01

In comparison to the null distributions, the same general pattern was observed for taxonomic and functional beta diversity and their components. Overall, temporal beta diversity and turnover were lower than expected by chance, whereas the observed nestedness did not differ from the null distribution (Table 5). Thus, the lower than expected beta diversity was due almost entirely to lower than expected turnover.

Table 5. Mean of the observed and null temporal beta diversity (β_{SOR}), turnover (β_{SIM}) and nestedness (β_{SNE}) for species composition (Taxonomic) and functional groups (Functional).

		Observed	Null	p-value
Taxonomic	β_{SOR}	**0.33**	**0.54**	**0.00**
	β_{SIM}	**0.21**	**0.46**	**0.00**
	β_{SNE}	0.11	0.08	0.17
Functional	β_{SOR}	**0.14**	**0.21**	**0.00**
	β_{SIM}	**0.04**	**0.12**	**0.00**
	β_{SNE}	0.09	0.09	0.74

Discussion

Most of the spatial beta diversity of fish species and functional groups composition was due to turnover. Despite environmental changes observed between 2003 and 2013, regional patterns of spatial turnover and nestedness were not different between the years. By contrast, while regional beta diversity, turnover and nestedness remained unchanged, the pairwise pattern changed over time. However, that change was not correlated with the degree of changes in environmental dissimilarity at regional and local scales. Temporal turnover was responsible for the most part of the species beta diversity, whereas nestedness was relatively more important to functional assemblages. Contrary to our hypothesis, temporal beta diversity, turnover and nestedness were not associated with the degree of regional (watershed land use) and local (instream) changes. Temporal beta diversity and turnover were lower than expected by chance whereas temporal nestedness did not differ from the null distribution. According these results, the environmental changes observed in this study (i.e. our deterministic factors) did not influence in the different pattern from the one expected by chance.

The dominance of turnover over nestedness in spatial beta diversity was already demonstrated for freshwater fish fauna in tropical areas and it was correlated to past glacial history and climate stability (Leprieur *et al*., 2011). Turnover was negatively correlated to the percentage of glacier coverage in the basins and the amplitude between present and quaternary temperature. A similar pattern was also observed in amphibians (Baselga, Gómez-Rodrigues & Lobo, 2012). According to these authors, tropical area has experienced milder climate changes and consequently, species assemblages would be older and with more speciation events than temperate assemblages. Over time, turnover would be higher in areas with older assemblages because these new species would present smaller ranges than ancestral species. Despite the role of climate stability on current turnover, we believe that historical human activities may also influence spatial turnover by increasing even more the turnover rates. Specifically, we suggest that regionally widespread habitat homogenization and previous biotic filtering makes further spatial and temporal loss or gain of species from the regional pool relatively unlikely, which limits the relative importance of nestedness. Thus, beta diversity in agroecosystems would be mostly due to the replacement of relatively tolerant species (i.e. those already filtered from the regional pool) over space and time. Similar to our results, Baselga *et al*. (2015) found dominance of turnover in their study of bird assemblages in agroecosystems in France. We expect that conversion of native systems to agriculture probably led to similar environmental and biotic homogenization in both their study and ours.

Current environmental conditions can play an important role in beta diversity patterns (Costa & Melo, 2008). Landscape and local heterogeneity promote higher regional and site-to-site beta diversity by diversifying niche availability and increasing the chance of different species colonization and establishment (Astorga *et al*., 2014). For these reasons, we expected recent environmental changes in our region caused by pasture

conversion to sugarcane crops would change beta diversity patterns. Surprisingly, spatial beta diversity remained unchanged over time. Furthermore, watershed (regional) and instream (local) changes observed in our study area were not correlated with pairwise changes and temporal beta diversity. According to Vellend *et al*. (2007), past environmental filters are responsible for weakened current species composition-environment relationships. Indeed, a weak relationship between environmental condition at different scales (spatial, watershed and local) and fish assemblages has already been detected for our region (Roa-Fuentes & Casatti, 2017). Regarding beta diversity, Baselga *et al*. (2015) also found temporal dissimilarity of bird assemblages in long-term agroecosystem were not related to environmental changes, but rather to stochastic processes. The authors proposed habitat generalist species would use the agroecosystem environment in a random manner. Considering the current fish assemblages from our agroecosystem probably are a generalist sub-set of a more diverse regional pool that existed before deforestation (Roa-Fuentes & Casatti, 2017), beta diversity patterns could be driven by stochasticity. Thus, even though Baselga *et al*. (2015) and this study explored diversity patterns with taxa with different dispersal dynamics (i.e. birds dispersing across connected terrestrial landscapes vs. fishes in a dendritic network, respectively), the importance of stochasticity in the current patterns of beta diversity for both could be due to the homogenization and filtering processes common to agroecosystems.

Despite temporal beta diversity were different from the one expected by change, the environmental change quantified in our study did not influenced the observed pattern. This result is very intriguing, and we proposed some hypotheses that could explain it. First, the environmental variables quantified in our study may not be the primary factors driving patters of beta diversity. Thus, our data would have underestimate the role of

deterministic factors in beta diversity patterns. Notwithstanding, this first hypothesis seems very unlikely, since the same regional and local variables have been associated with stream fish assemblages not only in our region (Casatti *et al*., 2006; Casatti *et al*., 2009; Casatti *et al*., 2015), but in several aquatic systems around the world (Smokorowski & Pratt, 2007). The magnitude of environmental variation can be strongly correlated with beta diversity (Hatosy *et al*., 2013). Considering species from long-term agroecosystems are mainly composed by generalists able to survive in multiples types of habitats, it is possible that the degree of further environmental changes required to change species composition is larger than the one in this study. We believe regional and instream changes observed in these ten years did not reach the threshold that would lead to further fish assemblage's changes. However, even if this threshold has already been reached, assemblage's patterns could be more associated with past than current land use or environmental condition (Harding *et al*., 1998; Burcher *et al*., 2008; Uezu & Metzger, 2016). In fact, using the same dataset Zeni *et al* (first chapter in this dissertation and currently in review) found that 2013 assemblages were still correlated to 2003 environment. Possibly, the interaction between low degree of environmental change and a time lag response longer than 10 years influenced in the nonexistent correlation between temporal beta diversity and environmental changes in our study.

However, if we consider that the primary driver of environmental changes in our study area is the conversion of pasture to sugarcane crops, physicochemical variables are more likely to change drastically over time than instream physical habitat. Extensive fertilization and agrochemical use, not common in pasture management but widely employed in sugarcane production, may modify water quality (e.g. productivity, pH, conductivity, Gunkel *et al*., 2007; Martinelli & Filoso, 2008; Christofoletti *et al*., 2013; Filoso *et al*. 2015; Schiesari & Corrêa, 2016) and select for different species.

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Unfortunately, physicochemical variables were sampled using different methods in 2003 and 2013 and we decided not to include them in our analyses to maintain standardized structure.

Our observed temporal beta diversity lower than expected by chance could be a direct result of the factors mentioned above (low degree of instream physical habitat change and time lag response), but it could also be a product of the effects of long-term land use change (Flynn *et al*., 2009; Karp *et al*., 2012). Environmental filters due to deforestation in this region were mainly associated with stream habitat homogenization, which led to a non-random extinction of habitat-sensitive species. According to McKinney & Lockwood (1999), in the next mass extinction, few "winner" species regionally widespread and broadly adapted to human-induced changes will replace many "loser" species unable to tolerate new conditions. Consequently, this process will lead to taxonomic and functional homogenization (Villéger *et al*., 2015). Pre-deforestation fish inventories are lacking for this region. However, most of the fish species collected in this study are common and regionally spread, while only a few rare species were collected in just a few streams. This differs greatly from the pattern in other species-rich environments where most of the diversity is comprised by rare species (McGill *et al*., 2007). Regrettably, it is very plausible that streams in our region already represent the homogenized faunas as proposed by McKinney & Lockwood (1999).

Despite the dominance of agroecosystems and a growing interest that beta diversity has aroused in ecologists, there are few studies with beta diversity in agroecosystems. Thus, we cannot speak broadly about general patterns of beta diversity in agroecosystems, and we can only speculate about what ecological mechanisms might lead to consistent patterns in such systems (if any). This study helps address this knowledge gap by assessing how further environmental changes in established

agroecosystems affect spatial and temporal beta diversity of stream fish assemblages. We hypothesize that the current patterns of fish assemblages in agroecosystem streams are primarily the result of past processes of deforestation and environmental filtering during the initial development of agriculture in this region. Deforestation and the development of agriculture led to regional (landscape) and local (stream physical habitat) homogenization, and consequently to homogenized fish assemblages by selecting habitat-generalist species, which are also highly resistant to further environmental changes (Fig. 4). Agroecosystems are the dominant landscape of the Anthropocene, and future studies should further develop our ecological understanding of assemblage structure and dynamics in these ecosystems. In addition to testing the hypotheses presented above, such studies could also assess whether efforts to mitigate environmental impacts in agroecosystems (e.g. riparian corridors, protected patches, changes in harvesting and processing of sugarcane, etc.) can elicit ecological improvements at the landscape scale given the already degraded state of the regional species pool in most agroecosystems. This information would be very important to biodiversity conservation and management as our agroecosystems are further stressed to feed an ever-growing human population.
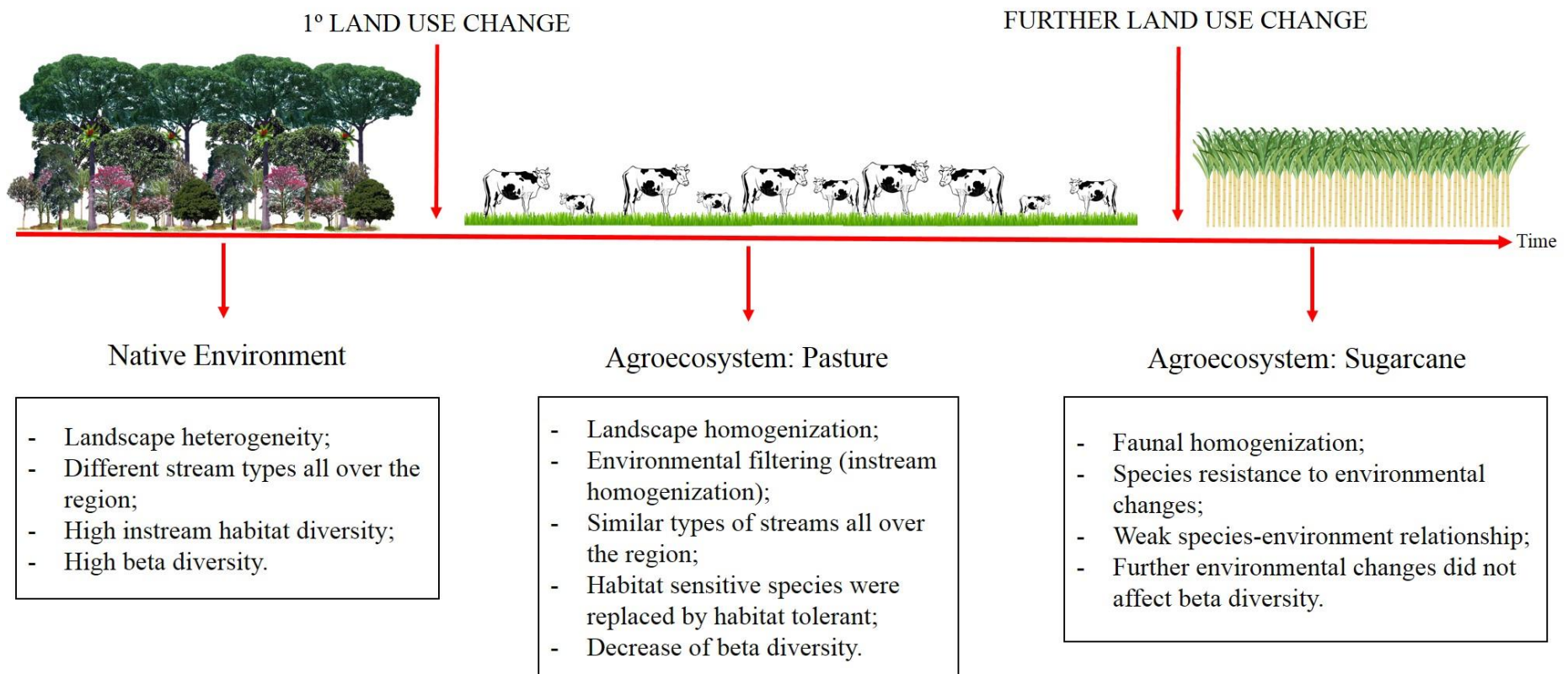
Fig. 4. The processes that probably happened in the region after the initial deforestation in the beginning of 20th century and led to landscape, stream physical habitat and fish fauna homogenization.

References

Allan J.D. (2004) Landscape and riverscape: The influence of land use on stream ecosystems. *Annual Review of Ecology, Evolution, and Systematics*, **35**, 257–284.

Al-Shami S.A., Heino J., Salmah M.R.C., Hassan A.A., Suaila A.H. & Madrus M.R. (2013) Drivers of beta diversity of macroinvertebrate communities in tropical forest streams. *Freshwater Biology*, **58**, 1126–1137.

Anderson M.J., Crist T.O., Chase J.M., Vellend M., Inouye B.D., Freestone A.L. *et al.* (2011) Navigating the multiple meanings of b diversity: a roadmap for the practicing ecologist. *Ecology Letters*, **14**, 19–28.

Astorga A., Death R., Death F., Paavola R., Chakraborty M. & Muotka T. (2014) Habitat heterogeneity drives the geographical distribution of beta diversity: the case of New Zealand stream invertebrates. *Ecology and Evolution*, **4**, 2693–2702.

Balon E.K., Crawford S.S. & Lelek A. (1986) Fish communities of the Upper Danube River (Germany, Austria) prior to the new Rhein-Main-Donau connection. *Environmental Biology of Fishes*, **15**, 243–271.

Barrella W., Beaumord A.C. & Petrere Jr M. (1994) Comparacion de la comunidad de peces de los rios Manso (MT) y Jacare Pepira (SP), Brasil. *Acta Biologica Venezuelica*, **15**, 11–20.

Baselga A. (2010) Partitioning the turnover and nestedness components of beta diversity. *Global Ecology and Biogeography*, **19**, 134–143.

Baselga A., Gómez-Rodrigues C. & Lobo J.M. (2012) Historical legacies in world amphibian diversity revealed by the turnover and nestedness components of beta diversity. *PLoS ONE*, **7**, e32341.

Baselga A., Orme D., Villeger S., Bortoli J. & Leprieur F. (2014) *Package betapart: Partitioning beta diversity into turnover and nestedness components*.

Baselga A., Bonthoux S. & Balent G. (2015) Temporal beta diversity of bird assemblages in agricultural landscapes: Land cover change vs. stochastic processes. *PLoS ONE*, **10**, e0127913.

Blake R.W. (1983) Functional design and burst-and-coast swimming in fishes. *Canadian Journal of Zoology*, **61**, 2491–2494.

Burcher C.L., McTammany M.E., Benfield E.F. & Helfman G.S. (2008) Fish assemblage responses to forest cover. *Environmental Management*, **41**, 336–346.

Burden F.J., McIntosh A.R. & Harding J.S. (2013) Habitat loss drives threshold response of benthic invertebrate communities to deposited sediment in agricultural streams. *Ecological Applications*, **23**, 1036–1047.

Casatti L., Langeani F., Silva A.M. & Castro R.M.C. (2006) Stream fish, water and habitat quality in a pasture dominated basin, southeastern Brazil. *Brazilian Journal of Biology*, **66**, 681–696.

Casatti L., Ferreira C.P. & Carvalho F.R. (2009) Grass-dominated stream sites exhibit low fish species diversity and guppies dominance: an assessment on two tropical pasture river basins. *Hydrobiologia*, **632**, 273–283.

Casatti L., Teresa F.B., Zeni J.O., Ribeiro M.D., Brejão G.L. & Ceneviva-Bastos M. (2015) More of the same: high functional redundancy in stream fish assemblages from tropical agroecosystems. *Environmental Management*, **55**, 1300–1314.

Catano C.P., Dickson T.L. & Myers J.A. (2017) Dispersal and neutral sampling mediate contingent effects of disturbance on plant beta-diversity: a meta-analysis. *Ecology Letters*, **20**, 347-356.

Christofoletti C.A., Escher J.P., Correia J.E., Marinho J.F.U. & Fontanetti C.S. (2013) Sugarcane vinasse: environmental implications of its use. *Waste Management*, **33**, 2752–2761.

Connor E.F. & McCoy E.D. (2001) Species area relationships. *Encyclopedia of Biodiversity*, **5**, 397–411.

Conway G.R. (1987) The properties of agroecosystems. *Agricultural systems*, **24**, 95–117.

Costa S.S. & Melo A.S. (2008) Beta diversity in stream macroinvertebrate assemblages: among-site and among-microhabitat components. *Hydrobiologia*, **598**, 131–138.

Dirzo R., Young H.S., Galetti M., Ceballos G., Isaac N.J.B. & Collen B. (2014) Defaunation in the Anthropocene. *Science*, **345**, 401–406.

Ellis E.C. & Ramankutty N. (2008) Putting people in the map: anthropogenic biomes of the world. *Frontiers in Ecology and the Environment*, **6**, 439–447.

Engen S., Bakke O. & Islam A. (1998) Demographic and environmental stochasticity concepts and definitions. *Biometrics*, **54**, 840–846.

Filoso S., do Carmo J.B., Mardegan S.F., Lins S.R.M., Gomes T.F. & Martinelli L.A. (2015) Reassessing the environmental impacts of sugarcane ethanol production in Brazil to help meet sustainability goals. *Renewable and Sustainable Energy Reviews*, **52**, 1847-1856.

Flynn D.F.B.,Gogol-Prokurat M., Nogeire T., Molinari N., Richers B.T., Lin B.B. *et al*. (2009). Loss of functional diversity under land use intensification across multiple taxa. *Ecology Letters*, **12**, 22–33.

Gatz A.J. (1979) Ecological morphology of freshwater stream fishes. *Tulane Studies on Zoology and Botany*, **21**, 91–124.

Gosline W.A. (1971) *Functional morphology and classification of teleostean fishes*. Honolulu, University Press of Hawaii.

Gunkel G., Kosmol J., Sobral M., Rohn H., Montenegro S. & Aureliano J. (2007) Sugar cane industry as a source of water pollution – case study on the situation in Ipojuca River, Pernambuco, Brazil. *Water Air and Soil Pollution*, **180**, 261–269.

Harding J.S., Benfield E.F., Bolstad P.V., Helfman G.S. & Jones III E.B.D. (1998) Stream biodiversity: the ghost of land use past. *Proceedings of the National Academy of Sciences of United States of America*, **95**, 14843–14847.

Hatosy S.M., Martiny J.B.H., Sachdeva R., Steele J., Fuhrman J.H. & Martiny A.C. (2013) Beta diversity of marine bacteria depends on temporal scale. *Ecology*, **94**, 1898–1904.

Hora S.L. (1930) Ecology, bionomics and evolution of the torrential fauna, with special reference to the organs of attachment. *Philosophical Transactions of the Royal Society of London, series B*, **218**, 172–282.

Kalyuzhny M., Schreiber Y., Chocron R., Flather C.H., Kadmon R., Kessler D.A. *et al.*
(2014) Temporal fluctuation scaling in populations and communities. *Ecology*,
95, 1701–1709.

Karp D.S., Rominger A.J., Zook J., Ranganathan J., Ehrilich P.R. & Daily G.C. (2012)
Intensive agriculture erodes b-diversity at large scales. *Ecology Letters*, **15**, 963–
970.

Kessler M., Abrahamczyk S., Bos M., Buchori D., Putra D.D., Gradstein S.R. *et al.* (2009)
Alpha and beta diversity of plants and animals along a tropical land-use gradient.
Ecological Applications, **19**, 2142–2156.

Lapola D.M., Martinelli L.A., Peres C.A., Ometto J.P.H.B., Ferreira M.E., Nobre C.A. *et
al.* (2014) Pervasive transition of the Brazilian land-use system. *Nature Climate
Change*, **4**, 27–35.

Legendre P. (2007) Studying beta diversity: ecological variation partitioning by multiple
regression and canonical analysis. *Journal of Plant Ecology*, **1**, 3–8.

Leprieur F., Tedesco P.A., Hugueny B., Beauchard O., Dürr H.H., Brosse S. & Oberdorff
T. Partitioning global patterns of freshwater fish beta diversity reveals contrasting
signatures of past climate changes. *Ecology Letters*, **14**, 325–334.

Mahon R. (1984) Divergent structure in fish taxocenes of north temperate streams.
Canadian Journal of Fisheries and Aquatic Sciences, **41**, 330–350.

Martinelli L.A. & Filoso S. (2008) Expansion of sugarcane ethanol production in Brazil:
environmental and social challenges. *Ecological Applications*, **18**, 885–898.

McGill B.J., Etienne R.S., Gray J.S., Alonso D., Anderson M.J., Benecha, H.K. *et al.*
(2007) Species abundance distributions: moving beyond single prediction theories
to integration within an ecological framework. *Ecology Letters*, **10**, 995–1015.

Mittelback G.G. (2010) *Community Ecology*. Sinauer Associates, 400p.

Monbeig P. (1998) *Pioneiros e fazendeiros de São Paulo*. São Paulo, Hucitec, 392p.

Nalon M.A., Mattos I.F.A. & Franco G.A.D.C. (2008) Meio físico e aspectos da fragmentação da vegetação. In: *Diretrizes Para a Conservação e Restauração da Biodiversidade no Estado de São Paulo* (Eds R.R. Rodrigues & V.L.R. Bononi), pp. 17–21. Secretaria do Meio Ambiente e Instituto de Botânica, São Paulo.

Newbold T., Hudson L.N., Hill S.L.L., Contu S., Gray C.L., Scharlemann J.P.W. *et al.* (2016) Global patterns of terrestrial assemblage turnover within and among land uses. *Ecography*, **39**, 1151–1163.

Oliveira E.F., Goulart E., Breda L., Minte-Vera C.V., Paiva L.R.S. & Vismara M.R. (2010) Ecomorphological patterns of the fish assemblage in a tropical floodplain: effects of trophic, spatial and phylogenetic structures. *Neotropical Ichthyology*, **8**, 569–586.

Pouilly M., Lino F., Bretenoux J.G. & Rosales C. (2003) Dietary-morphological relationships in a fish assemblage of the Bolivian Amazonian floodplain. *Journal of Fish Biology*, **62**, 1137–1158.

Rahel F.J. (2002). Homogenization of freshwater faunas. *Annual Review of Ecology, Evolution, and Systematics*, **33**, 291–315

Roa-Fuentes C.A. & Casatti L. (2017). Influence of environmental features at multiple scales and spatial structure on stream fish communities in a tropical agricultural region. *Journal of Freshwater Ecology*, **32**, 273–287.

Santos F.B., Ferreira F.C. & Esteves K.E. (2015) Assessing the importance of the riparian zone for stream fish communities in a sugarcane dominated landscape (Piracicaba River Basin, Southeast Brazil). *Environmental Biology of Fishes*, **98**, 1895–1912.

Schiesari L. & Corrêa D.T. (2016) Consequences of agroindustrial sugarcane production to freshwater biodiversity. *Global Change Biology Bioenergy*, **8**, 644–657.

Scott M.C. & Helfman G.S. (2001) Native invasions, homogenization, and the mismeasure of integrity of fish assemblages. *Fisheries*, **26**, 6–15.

Segre H., Ron R.,1 Malach N., Henkin Z., Mandel M. & Kadmon R. (2014) Competitive exclusion, beta diversity, and deterministic vs. stochastic drivers of community assembly. *Ecology Letters*, **17**, 1400–1408.

Silva A.M., Casatti L., Alvares C.A., Leite A.M., Martinelli L.A. & Durrant S.F. (2007) Soil loss risk and habitat quality in some streams of a Meso-Scale river basin. *Scientia Agricola*, **64**, 336–343.

Smokorowski K.E. & Pratt T.C. (2007) Effect of a change in physical structure and cover on fish and fish habitat in freshwater ecosystems – a review and meta-analysis. *Environmental Reviews*, **15**, 15–41.

Socolar J.B., Gilroy J.J., Kunin W.E. & Edwards D.P. (2016) How should beta-diversity inform biodiversity conservation? *Trends in Ecology & Evolution*, **31**, 67–80.

Teresa F.B. & Casatti L. (2012) Influence of forest cover and mesohabitats types on functional and taxonomic diversity of fish communities in Neotropical lowland streams. *Ecology of Freshwater Fish*, **21**, 433–442.

Uezu A. & Metzger J.P. (2016) Time-lag in responses of birds to Atlantic forest fragmentation: Restoration opportunity and urgency. *PLoS ONE*, **11**, e0147909

Villéger S., Blanchet S., Beauchard O., Oberdorff T. & Brosse S. (2015) From current distinctiveness to futurehomogenization of the world'sfreshwater fish faunas. *Diversity and Distributions*, **21**, 223–235.

Vellend M., Verheyen K., Flinn K.M., Jacquemyn H., Kolb A., Calster H.V. *et al*. (2007) Homogenization of forest plant communities and weakening of species-environment relationship via agricultural land use. *Journal of Ecology*, **95**, 565–573.

Wainwright P.C., Bellwood D.R. & Westneat M.W. (2002) Ecomorphology of locomotion in labrid fishes. *Environmental Biology of Fishes*, **65**, 47–62.

Ward-Campbell B.M.S., Beamish F.W.H. & Kongchaiya C. (2005) Morphological characteristics in relation to diet in five coexisting Thai fish species. J*ournal of Fish Biology*, **67**, 1266–1279.

Waters C.N., Zalasiewicz J., Summerhayes C., Barnosky A.D., Poirier C., Gałuszka A. *et al*. (2016) The Anthropocene is functionally and stratighaphically distinct from Holocene. *Science*, **351**, 137–147.

Watson D.J. & Balon E.K. (1984) Ecomorphological analysis of fish taxocenes in rainforest streams of northern Borneo. *Journal of Fish Biology*, **25**, 371–384.

Whittaker, R.H. (1960) Vegetation of the Siskiyou Mountains, Oregon and California. *Ecological Monographs*, **30**, 279–338.

Whittaker R.J., Willis K.J & Field R. (2001) Scale and species richness: towards a general, hierarchical theory of species diversity. *Journal of Biogeography*, **28**, 453–470.

Wilga C.D. & Lauder G.V. (1999) Locomotion in sturgeon: function of the pectoral fins. *Journal of Experimental Biology*, **202**, 2413–2432.

Willis S.C., Winemiller K.O. & Lopez-Fernandez H. (2005) Habitat structural complexity and morphological diversity of fish assemblages in a Neotropical floodplain river. *Oecologia*, **142**, 284–295.

Winemiller K.O. (1991) Ecomorphological diversification in lowland freshwater fish assemblages from five biotic regions. *Ecological Monographs*, **61**, 343–365

Young H.S., McCauley D.J., Galetti M. & Dirzo R. (2016) Patterns, causes, and consequences of Anthropocene defaunation. *Annual Review of Ecology, Evolution, and Systematics*, **47**, 333–358.

Zeni J.O., Hoeinghaus D.J. & Casatti L. Effects of pasture conversion to sugarcane for biofuels production on stream fish assemblages in tropical agroecosystems. In review.

Supporting material: Chapter 2

Table S1. Taxonomic classification and total abundances (i.e. combined across all 38 streams) of the 54 species collected in 2003 and 2013. * non-native species.

Order	Family	Species and author	Abundance 2003	2013
Characiformes	Acestrorhynchidae	*Acestrorhynchus lacustris* (Lütken, 1875)	1	1
	Anostomidae	*Leporinus friderici* (Bloch, 1794)	7	2
		Leporinus lacustris Amaral Campos, 1946	0	1
		Leporinus paranaensis Garavello & Britski, 1987	1	0
		Leporinus striatus Kner, 1858	1	0
	Characidae	*Astyanax lacustris* (Lütken 1875)	1,926	1,113
		Astyanax fasciatus (Cuvier, 1819)	179	122
		Astyanax schubarti Britski, 1964	0	4
		Astyanax paranae Eigenmann, 1914	2	0
		Bryconamericus stramineus Eigenmann, 1908	13	10
		Hemigrammus marginatus Ellis, 1911	152	130
		Hyphessobrycon anisitsi (Eigenmann, 1907)	0	18
		Hyphessobrycon eques (Steindachner, 1882)	4	4
		Knodus moenkhausii (Eigenmann & Kennedy, 1903)	1,347	886
		Moenkhausia sanctaefilomenae (Steindachner, 1907)	31	32
		Oligosarcus pintoi Campos, 1945	332	251
		Piabina argentea Reinhardt, 1867	121	160
		Planaltina britskii Menezes,Weitzman & Burns, 2003	3	0
		Roeboides descalvadensis Fowler, 1932	0	2
		Serrapinnus notomelas (Eigenmann, 1915)	329	321
	Curimatidae	*Cyphocharax modestus* (Fernández-Yépez, 1948)	8	0
		Cyphocharax vanderi (Britski, 1980)	71	134
		Steindachnerina insculpta (Fernández-Yépez, 1948)	18	1
	Crenuchidae	*Characidium zebra* Eigenmann, 1909	141	50
		Characidium lagosantense Travassos, 1947	5	0
	Erythrinidae	*Hoplias malabaricus* (Bloch, 1794)	50	43
	Lebiasinidae	*Pyrrhulina australis* Eigenmann & Kennedy, 1903	84	27
	Parodontidae	*Apareiodon affinis* (Steindachner, 1879)	0	1
		Apareiodon piracicabae (Eigenmann, 1907)	3	0
		Parodon nasus Kner, 1858	100	30

Cyprinodontiformes	Poeciliidae	*Poecilia reticulata* Peters, 1859*	1,199	2,651
		Xiphophorus maculatus (Günther, 1866)*	0	108
	Rivulidae	*Melanorivulus pictus* (Costa, 1989)	5	8
Gymnotiformes	Gymnotidae	*Gymnotus sylvius* Albert & Fernandes-Matioli, 1999	173	162
	Sternopygidae	*Eigenmannia trilineata* López & Castello, 1966	9	2
Perciformes	Cichlidae	*Cichlasoma paranaense* Kullander, 1983	55	68
		Crenicichla britskii Kullander, 1982	54	29
		Crenicichla semifasciata (Heckel, 1840)	0	22
		Geophagus brasiliensis (Quoy & Gaimard, 1824)	27	42
		Laetacara araguaiae Ottoni & Costa, 2009	120	73
		Oreochromis niloticus (Linnaeus, 1758)*	1	0
		Satanoperca pappaterra (Heckel, 1840)	24	41
		Tilapia rendalli (Boulenger, 1897)*	8	0
Siluriformes	Callichthyidae	*Aspidoras fuscoguttatus* Nijssen & Isbrücker, 1976	256	264
		Callichthys callichthys (Linnaeus, 1758)	4	1
		Corydoras aeneus (Gill, 1858)	84	273
		Hoplosternum littorale (Hancock, 1828)	2	12
	Heptapteridae	*Cetopsorhamdia iheringi* Schubart & Gomes, 1959	2	7
		Imparfinis schubarti (Gomes, 1956)	112	129
		Pimelodella avanhandavae Eigenmann, 1917	11	0
		Rhamdia quelen (Quoy & Gaimard, 1824)	78	72
	Loricariidae	*Hypostomus ancistroides* (Ihering, 1911)	182	136
		Hypostomus cf. *nigromaculatus* (Schubart, 1964)	98	80
	Pseudopimelodidae	*Pseudopimelodus pulcher* (Boulenger, 1887)	6	12
		TOTAL	7,439	7,535

Table S2. Ecomorphological attributes, calculations and ecological significance of functional traits related body size, vertical, horizontal habitat and resource uses. Ecomorphological attributes were calculated following Oliveira et al. (2010). Maximum body height (MBH), maximum body width (MBW), body midline height (BMH), standard length (SL), caudal peduncle length (CPdL), caudal peduncle height (CPdH), caudal peduncle width (CPdW), dorsal fin area (DA), caudal fin area (CA), pectoral fin area (PtA), pectoral fin length (PtL), head length (HdL), head height (HdH), mouth width (MW), eye height (EH).
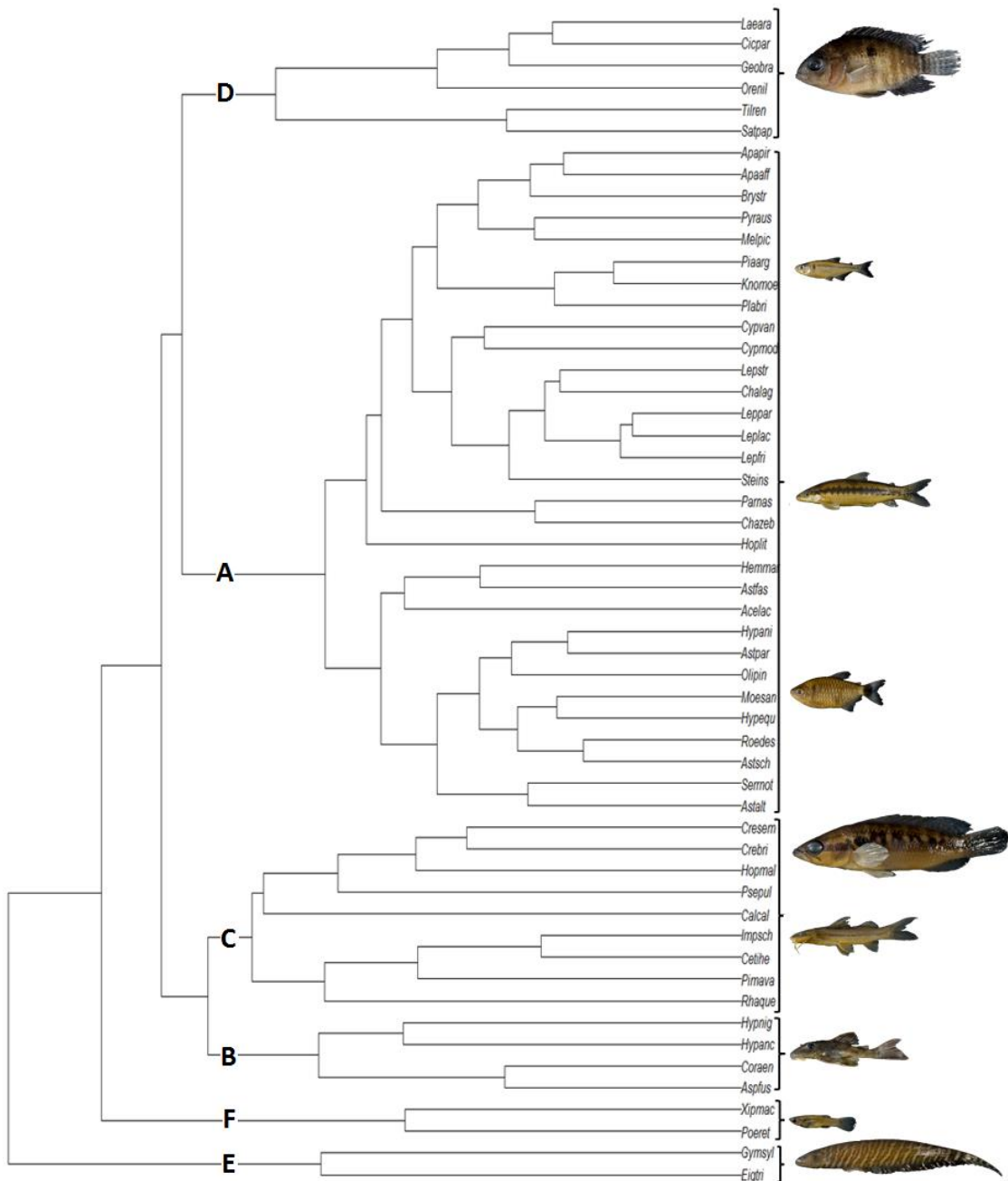
Ecomorphological attributes	Calculation	Ecological significance
Compression index	$CI = \dfrac{MBH}{MBW}$	Higher values indicate lateral compression of the fish, which indicate fish that explore habitats with slower water velocity (Gatz, 1979; Watson and Balon, 1984).
Depression index	$DI = \dfrac{BMH}{MBH}$	Low values indicate fishes inhabiting environments with high hydrodynamism, able to maintain their position even when stationary (Hora, 1930).
Relative depth	$RD = \dfrac{MBH}{SL}$	Lower values indicate fishes inhabiting fast waters. It is directly related to the ability to perform vertical spins (Gatz, 1979).
Fineness ratio	$FC = \dfrac{SL}{\sqrt{MBH \times MBW}}$	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 length of caudal peduncle	$RLPd = \dfrac{CPdL}{SL}$	Fishes with long caudal peduncle are goods swimmers. However, fishes adapted to rapid water flow, but no necessarily nektonic as armored catfishes, also presented long caudal peduncules in function of propulsion in short distances (Watson & Balon, 1984; Winemiller, 1991)
Relative height of caudal peduncle	$RHPd = \dfrac{CPdH}{MBH}$	Lower values indicate greater maneuverability potential (Winemiller, 1991).
Relative width of caudal peduncle	$RWPd = \dfrac{CPdW}{MBW}$	Higher relative values indicate better continuous swimmers (Winemiller, 1991).
Relative area of dorsal fin	$RAD = \dfrac{DA}{SL^2}$	Dorsal fins with larger relative areas have better capacity of stabilization in deflections (Gosline, 1971).
Relative area of caudal fin	$RAC = \dfrac{CA}{SL^2}$	Caudal fins with larger relative areas are important for the acceleration (Balon, Crawford & Lelek, 1986).
Relative area of pectoral fin	$RAPt = \dfrac{PtA}{SL^2}$	The pectoral fin area is generally high for slow swimming species, which use the pectoral fin for maneuverability, as some characids. Moreover, pectoral fin area also can be high for fishes that exploit habitats with intense current, as the siluriforms (Watson & Balon, 1984; Wilga & Lauder, 1999).

Aspect ratio of pectoral fin	$ARPt = \dfrac{PtL^2}{PtA}$	Higher ratio indicates long and narrow pectoral fins, which is more expected on fish that are continuous high-speed swimmers and prefer pelagic regions (Wainwright, Bellwood & Westneat, 2002).
Relative length of head	$RLHd = \dfrac{HdL}{SL}$	Larger relative values of head length are found in fishes which feed of larger prey. This index should be larger for piscivores (Watson & Balon, 1984; Winemiller, 1991; Barrella Beaumord & Petrere, 1994; Pouilly *et al.*, 2003; Willis, Winemiller & Lopez-Fernandez, 2005).
Relative height of head	$RHHd = \dfrac{HdH}{MBH}$	Larger relative values of head height are found in fishes which feed of larger prey. Larger values for this index are expected for piscivores (Winemiller, 1991; Willis *et al.*, 2005).
Relative Width of Mouth	$RWM = \dfrac{MW}{MBW}$	Larger relative values of mouth length suggest fishes which feed of larger prey (Gatz Jr., 1979; Balon *et al.*, 1986; Winemiller, 1991; Ward-Campbell, Beamish & Kongchaiya, 2005).
Eye Position	$EP = \dfrac{EH}{HdH}$	Related with food detection and provides information about visual predation activity (Poully *et al.*, 2003). Moreover, position of eyes is related to vertical habitat preference (Gatz, 1979); high values indicate dorsally located eyes, typical of benthic fish (Mahon, 1984; Watson & Balon, 1984).

* References from this table were included in the references section in the main document (Chapter 2).

Fig. S1. UPGMA cluster of 54 fish species based on 15 functional attributes yielding six functional groups. Group A includes all Characiformes, except *Hoplias malabaribus*, and *Melanorivulus pictus* (Cyprinodontiformes: Rivulidae), and *Hoploternum littorale* (Siluriformes: Callichthyidae). Group B includes two species of *Hypostomus* (Siluriformes: Loricariidae) and *Aspidoras fuscoguttatus* and *Corydoras aeneus* (Siluriformes: Callichthyidae). Group C includes Heptapteridae family (Siluriformes), *Callichthys callichthys* (Siluriformes: Callichthyidae), *Hoplias malabaricus* (Characiformes: Erythrinidae), *Pseudopimelodus pulcher* (Siluriformes: Pseudopimelodidae) and the two species of *Crenicichla* (Perciformes: Cichlidae). Group D includes the others Cichlidae species (Perciformes). Group E includes *Gymnotus sylvius* (Gymnotiformes: Gymnotidae) and *Eigenmannia trilineata* (Gymnotiformes: Sternopygidae). Group F includes two non-native Poeciliidae, *Poecilia reticulata* and

Xiphophorus maculatus (Cyprinodontiformes). The pictures represent the body shapes most associated with each functional group. Two groups (A and C) have more than one picture because of the comparatively higher diversity of body shape within each group.

ADDITIONAL INFORMATION

In this section, we are going to present some extra data obtained during fieldwork sampling. Due to our methodological approach, which compared biological (assemblage's structure and beta diversity) and environmental (regional and instream variables) changes in two periods 10 years apart, these data were not included in our two chapters above. However, we strongly believe that they are examples of how inappropriate conservation politics can compromise the quality of aquatic systems.

During the fieldwork in 2013, we revisited 54 stream reaches previously sampled in 2003. However, we were unable to sampled fish or any environmental variables in ten of those streams. In these reaches, we were unable to identify the stream channel, because of the grasses or *Typha* sp. invasion (Fig. 1). According to Dala-Corte *et al*. (2016), agriculture in the riparian area was positively associated with the increase of macrophytes abundance and siltation. We believe that the "disappearance" of these stream reaches is deeply connected to the consequences of the long-term native riparian forest removal and siltation progression. Besides many other functions (Pusey & Arthington, 2003), riparian forests are responsible for mitigate the input of inorganic sediment into stream channel. Historically, riparian forests in our region were removed and replaced by several agriculture crops (Monbeig, 1988). In fact, siltation in 2003 had already reached a worrying point with most of the stream bottoms homogenized and composed by sandy substrate (Casatti *et al*., 2006). In the streams sampled (included in the two chapters above), we also observed a progression of the siltation over 10 years with a decrease of water volume and depth (Fig 3b, Chapter 1). However, the process was much more severe in these 10 streams and created a favorable environment to establishment of macrophytes.
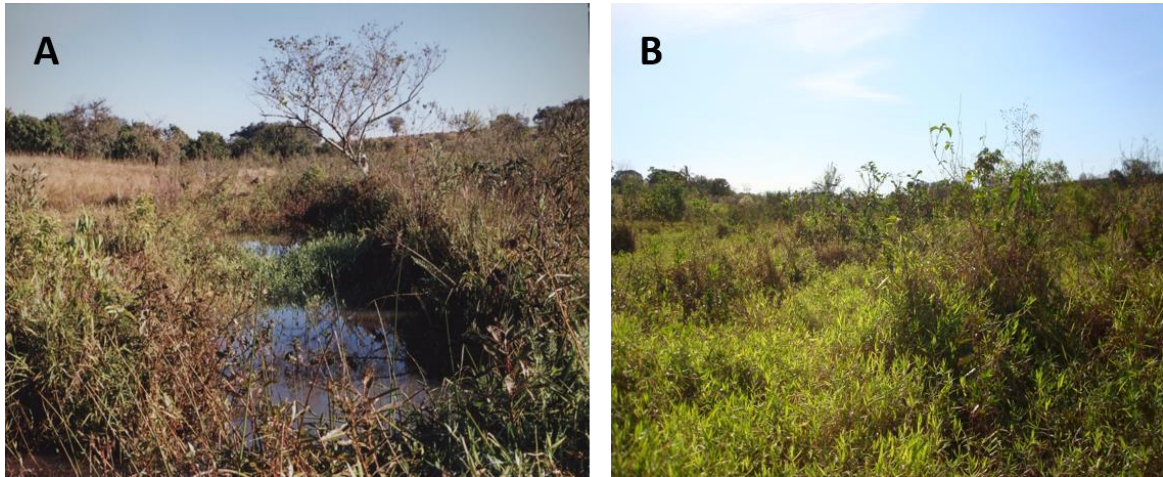
Figure 1. Example of macrophytes invasion in the stream reaches during the 10 years period. General view of a stream 2003 (A) and the same view in 2013 (B). The process observed in this figure was very similar to the other nine stream reaches in our study.

The presence of macrophytes and high levels of siltation are responsible for the increase taxonomic and functional fish assemblage's similarity (Dala-Corte *et al*., 2016). We are not able to say precisely what happened with the species from these streams, but the massive environmental change associated with the reduction of habitat availability by siltation and macrophytes dominance (Fig. 1), probably change fish assemblages aspects and led to local extinction of species. It is possible that without restoration efforts, the consequences of these observed alterations are irreversible in a short time. It takes years to riparian forests grow and play an effective role buffering the input of inorganic sediments and reducing macrophytes abundance through stream channel shading (Ceneviva-Bastos & Casatti, 2014). In meantime, aquatic assemblages are being changed or, even worse, extinct. This is particularly worrying for agroecosystems under an old and intense land use change, because these areas usually do not have any source of later colonization.

Besides biotic consequences, water availability and quality can be tremendous influenced by these observed alterations. In reservoirs, sedimentation is one main responsible for water quality degradation and reduction (Ahmed & Sanchez, 2011).

Despite the ability to mitigate the loads from agriculture crops (Connelly *et al*., 2015), few efforts were done towards restoration riparian forests in the Northwest of São Paulo state. Moreover, we were used to hear from farmers that water was an infinite resource and that had much more water now than years ago. It represents a clear misunderstood concept on more accessibility to water *versus* higher availability. This wrong thought allied to the public politic negligence (e.g. bland environmental law and no control) hamper riparian conservation and restoration. Unfortunately, the scenario observed here is happening in numerous aquatic systems, since the establishment of agriculture in the area predestined to riparian forests is widespread all around the world. For this reason, understanding the consequences of these long-term impacts are crucial to propose measures that effectively restore assemblages and some ecological services associated with streams, such as the water supply.

CONCLUSION

The conversion of natural to agroecosystems is one of the main drives of freshwater changes. Nevertheless, the effects of further land use change in agroecosystems are poorly known. Our study is the first one to quantify the direct effects of environmental changes at regional (mainly the conversion of pasture to sugarcane) and local scales (instream habitat) on aspects of fish assemblages, such as taxonomic and functional structure, besides spatial-temporal beta diversity. Surprisingly, further environmental changes are weakly related to changes in taxonomic and functional fish assemblages. Moreover, instream habitat and fish assemblage structure showed a time lag response, since they were also related to past land use. Similar results were found for beta diversity aspects, where spatial and temporal beta diversity were not correlated to further environmental changes. However, temporal beta diversity was lower than expected by chance. Taken together, these results might indicate that fish assemblages

from agroecosystems like this - drastically and disorderly deforested since a long time ago - can be profoundly influenced by land use change history. As ecological theory predicts assemblages are composed by few dominant species with high abundance and many rare species with low abundance. Due to narrow requirements, rare fish species are more prone to be locally and regionally extinct because of the environmental filter inflicted by human induced changes. The current species are those ones able to survive to the massive deforestation process in the beginning of 20th. Most of them have broad habitat requirements, wide regional distribution and probably they are environmentally resistant and resilient. Therefore, the degree of environmental changes and time required to cause fish assemblage response may be greater than observed in the present study.

Despite no correlation between environmental changes and fish assemblages, it is worth to mention that even resistant assemblages (i.e. composed by generalist species) may have a threshold for further environmental change (additional information). This could be particularly important if we consider the recent land use change in the area. Until now, the main impact in our streams were physical habitat modification (channel homogenization). However, extensive fertilization and agrochemical use in sugarcane crops can drastically affect water quality (Filoso *et al.*, 2015). The environmental filter due to water quality change is still unknown for most of our species. Thus, consequences of sugarcane culture intensification on these fish assemblages should be monitored, especially because riparian forests that could mitigate the input from terrestrial surroundings are extremely rare in the region. In this context, riparian reforestation must be mandatory to conserve the remaining species and the ecosystems services associated with these streams.

Lastly, we believe the difficulties associated with the study of agroecosystems assemblages should not be a barrier for scientists and future questions. We are facing a

growing demand for agriculture products associated with an environmental and political concern about deforestation in pristine areas. It would be rational to expect major agriculture crop intensification and changes in systems already under human modification. Thus, understanding how species from agroecosystem respond to further environmental changes may be a key factor to conserve not only agroecosystems assemblages, but also native and pristine environments and their species.

REFERENCES

Ahmed K.B. & Sanchez M. (2011) A study of the factors and processes involved in the sedimentation of Tarbela reservoir Pakistan. *Environmental Earth Sciences*, **62**, 927–933.

Casatti L., Langeani F., Silva A.M. & Castro R.M.C. (2006) Stream fish, water and habitat quality in a pasture dominated basin, southeastern Brazil. *Brazilian Journal of Biology*, **66**, 681–696.

Ceneviva-Bastos M. & Casatti C. (2014) Shading effects on community composition and food web structure of a deforested pasture stream: Evidences from a field experiment in Brazil. *Limnologica*, **46**, 9–21.

Chase J.W., Benoy G.A., Hann S.W.R. & Culp J.M. (2016) Small differences in riparian vegetation significantly reduce land use impacts on stream flow and water quality in small agricultural watersheds. *Journal of Soil and Water Conservation*, **71**, 194–205.

Connelly N.M., Pearson R.G., Loong D., Maughan M. & Brodie, J. (2015) Water quality variation along streams with similar agricultural development but contrasting riparian vegetation. *Agriculture, Ecosystems & Environment*, **213**, 11–20.

Dala-Corte R.B., Giam X., Olden J.D., Becker F.G., Guimarães T.F. & Melo A.S. (2016) Revealing the pathways by which agricultural land-use affects stream fish communities in South Brazilian grasslands. *Freshwater Biology*, **61**, 1921–1934.

Filoso S., do Carmo J.B., Mardegan S.F., Lins S.R.M., Gomes T.F. & Martinelli L.A. (2015) Reassessing the environmental impacts of sugarcane ethanol production in Brazil to help meet sustainability goals. *Renewable and Sustainable Energy Reviews*, **52**, 1847-1856.

Monbeig P. (1998) *Pioneiros e fazendeiros de São Paulo*. São Paulo, Hucitec, 392p.

Pusey B.J. & Arthington A.H. (2003) Importance of riparian zone to the conservation and management of freshwater fishes: a review. *Marine and Freshwater Research*, **54**, 1–16.