

**University of São Paulo
“Luiz de Queiroz” College of Agriculture
Center for Nuclear Energy in Agriculture**

**Effects of intensive agriculture in the structure and functioning of
tropical headwater streams**

Ricardo Hideo Taniwaki

Thesis presented to obtain the degree of Doctor in
Science. Area: Applied Ecology

**Piracicaba
2016**

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Bachelor degree in Biology

Effects of intensive agriculture in the structure and functioning of tropical headwater streams

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DEDICATORY

To my parents, sister and all of my past professors.

Gratitude

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EPIGRAPH

“Somewhere, something incredible is waiting to be known.”

Carl Sagan

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RESUMO

Efeitos da agricultura intensiva na estrutura e funcionamento de riachos de cabeceira tropicais

As regiões tropicais possuem os ecossistemas mais biodiversos do planeta. Apesar da sua importância na manutenção da biodiversidade, as atividades antrópicas estão degradando esses ecossistemas, gerando consequências negativas para sua estrutura e funcionamento. Entre os diversos ecossistemas tropicais, as águas doces chamam a atenção por ocupar uma pequena área em comparação com ecossistemas terrestres, porém, representam grande importância para a sobrevivência e desenvolvimento humano. Os riachos de cabeceira representam a maior parte dos corpos aquáticos em uma microbacia e, portanto, sua conservação é essencial para a saúde de toda a rede de drenagem de água doce. Diversos riachos de cabeceira estão inseridos em ecossistemas agrícolas, sofrendo as consequências da agricultura intensiva. Nesse sentido, esta tese foi desenvolvida com o intuito de entender quais são os principais impactos que os riachos de cabeceira tropicais vêm sofrendo, globalmente e localmente na bacia do rio Corumbataí (SP, Brasil). O primeiro capítulo contribui para entender quais os principais problemas que riachos tropicais estão sofrendo, focando nos múltiplos estressores advindos da agricultura e efeitos de mudanças climáticas. O segundo capítulo analisa como a conversão de pastos para cultivo de cana de açúcar modifica a qualidade da água em riachos da bacia do rio Corumbataí e também analisa o quanto fundamentais são as florestas nas áreas de nascente para manter a qualidade da água em plantios de cana de açúcar. O terceiro capítulo analisa como a qualidade da água e características climáticas influenciam biofilmes bentônicos em riachos de cabeceira na bacia do rio Corumbataí. Os resultados demonstraram a existência de diversos tópicos que necessitam de maior entendimento, principalmente relacionados às mudanças climáticas e estressores múltiplos e a falta de políticas e estratégias de mitigação para os efeitos de mudanças climáticas. Em relação à qualidade da água, demonstrou-se que a conversão de pastos para cultivo de cana de açúcar reduz a qualidade da água. A presença de florestas nas nascentes demonstrou ser essencial na manutenção da qualidade da água em plantios de cana. Os biofilmes bentônicos demonstraram ser controlados principalmente por características sazonais e não pela disponibilidade de nutrientes como observado em riachos temperados. Portanto, essa comunidade será severamente afetada diante das mudanças climáticas, com consequências no funcionamento de riachos de cabeceira tropicais. Para reduzir os efeitos negativos da agricultura intensiva e das mudanças climáticas, recomenda-se a implementação de florestas ripárias, com especial atenção às áreas de nascentes. Também se recomenda a implementação de boas práticas agrícolas na agricultura para garantir a sustentabilidade dos recursos hídricos tropicais.

Palavras-chave: Qualidade da água; Dinâmica de riachos; Florestas ripárias; Boas práticas agrícolas

ABSTRACT

Effects of intensive agriculture in the structure and functioning of tropical headwater streams

Tropical regions hold the planet's most biodiverse ecosystems. Despite its importance to biodiversity and conservation, anthropogenic activities are degrading these ecosystems, with unknown consequences for its functioning and structure. In between the several ecosystems through the tropics, freshwater ecosystems call attention, due to its small fraction of area comparing to terrestrial ecosystems, that represent an enormous importance for human surviving and developing. Headwater streams constitute the majority of water bodies in a catchment, and therefore, it is essential for the health of the entire freshwater ecosystems. Several headwater streams are inserted in agricultural lands, suffering from the pressures from agricultural intensification. Therefore, this thesis was developed aiming to understand what are the main pressures that tropical headwater streams has been suffering, worldwide and locally in the Corumbataí river basin. The first chapter will contribute to understanding what are the main issues that tropical streams have been experiencing, focusing on agricultural multiple stressors and climate change effects. The second chapter analyzes how the conversion of low-intensity pasturelands to high-intensity bioenergy crops changes the water quality parameters in streams located in the Corumbataí river basin and also examines how important are the riparian forests in the headwater zone to provide better water quality in bioenergy crops. The third chapter investigates how water quality and climatic characteristics affect benthic biofilm community dynamics in tropical headwater streams in the Corumbataí river basin. The results have shown the existence of several knowledge gaps about tropical streams, mainly related to the effects of climate change, multiple stressors and the lack of policies and mitigation strategies for climate change. In relation to water quality, we found that the conversion of low-intensity pastures to high-intensity bioenergy crops are degrading water quality. Riparian forests in the springhead zone have demonstrated to be essential in providing water quality in bioenergy crops, especially in the wet season. The benthic biofilm community seems to be controlled mainly by climate characteristics and not by nutrient availability as observed in temperate streams. Therefore, a climate change scenario, the benthic biofilm will be strongly affected, with consequences in the functioning of tropical headwater streams. To reduce the negative impacts of intensive agriculture and climate change, we recommend the implementation of riparian forests, with special attention to the springhead area and also the implementation of best agricultural practices in tropical agriculture to ensure the sustainability of tropical freshwater resources.

Keywords: Water quality; Stream functioning; Stream dynamics; Riparian forests; Best agricultural practices

1 INTRODUCTION

Nowadays, shifts in ecosystem dynamics due to human activities is a serious issue that environmental scientists have to deal. Croplands and pastures have become one of the largest biomes on Earth (FOLEY et al., 2005). Human activities have changed the planet, sufficiently to produce a stratigraphic signature in sediments and ice, distinguishing the human-dominated period, called “Anthropocene” (WATERS et al., 2016). These changes have caused extensive modifications of Earth’s ecosystems (FOLEY et al., 2005), and may have exceeded the biosphere capacity to regenerate (WACKERNAGEL et al., 2002).

Agriculture is one of the main causes of land-use changes on Earth. The land-use transition follows a timeline model proposed by Foley et al. (2005), from natural ecosystems to frontier clearing, then to subsistence agriculture and small-scale farms, and finally to intensive agriculture, urban areas and protected recreational lands (Figure 1.1). When the landscape reaches the intensive stage of land-use, several modifications on ecosystem dynamics are expected. For example, streamflow dynamics in agricultural areas was changed more than 100% in comparison to natural areas (DIAS et al., 2015). Changes in streamflow dynamics can alter nutrient dynamics and biological communities (POFF et al., 1997), with consequences for ecosystem functioning (CARDINALE et al., 2002).

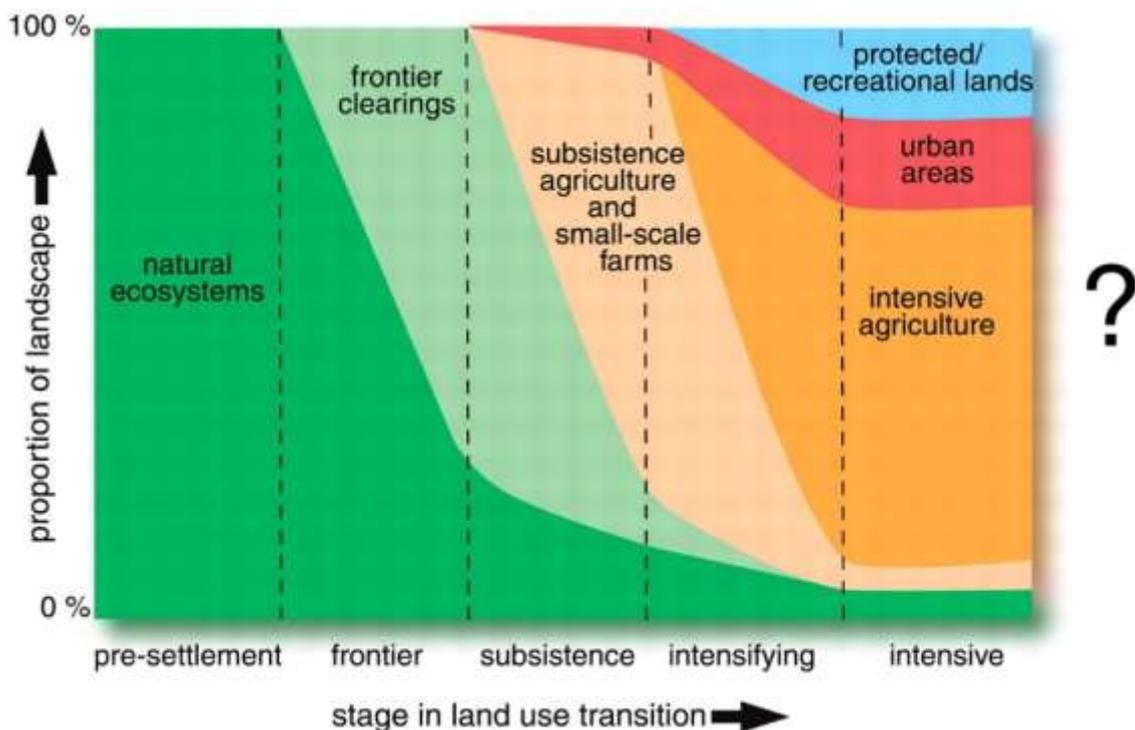


Figure 1.1 - Qualitative model of global land-use transition
Source: Foley et al. (2005)

Tropical areas present an intensive stage in land use, with extensive areas of agriculture and megacities (GUPTA, 2002; DEFRIES; ROSENZWEIG, 2010). These characteristics represent an important issue for biodiversity conservation, considering that the higher proportions of biodiversity occur in the tropics (GASTON, 2000). For example, more than 9,5% of known aquatic species are located in tropical freshwaters (DUDGEON, 2010). Also, tropical areas have greater energy inputs, faster rates of change and greater human-induced changes on hydrological cycle (WOHL et al., 2012).

Another topic that makes freshwater species extremely endangered in the tropics is the fast rates of deforestation. Deforestation changes the hydrological patterns in the catchments through the changes in the total water yield produced by precipitation, which increase with forest cutting due to the reduction of evapotranspiration and consequently, increase in the stream discharge (WOHL et al., 2012) (Figure 1.2). Changes in stream discharge modulate biological communities in streams (POFF et al., 1997; DODDS et al., 2015) with consequences for the entire aquatic ecosystem.

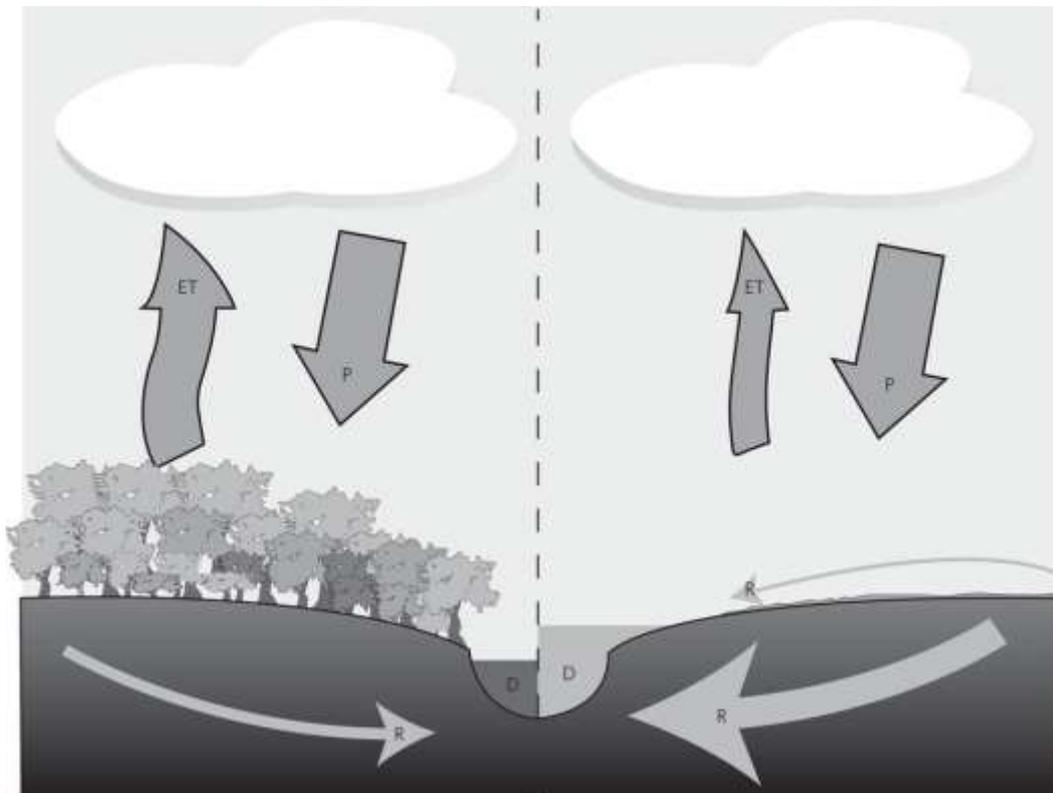


Figure 1.2 - Schematic view of the effects of deforestation on stream discharge: Evapotranspiration rates (ET) diminish with forest cutting; Total water yield (R) and discharge (D) increase with forest cutting
Source: Wohl et al. (2012)

Changes in runoff due to deforestation and agriculture have effects on water quality. Nutrient and sediments from agricultural lands are carried through runoff processes, polluting aquatic ecosystems and shifting biological communities, ecosystem functioning, and processes (DUDGEON et al., 2006; URIARTE et al., 2011). Especially in Brazil, we have an implication that draws attention for the stream water quality because of the modifications in the Forest Code - FC (Law N.: 12.651/2012), an important legal instrument to regulate land use and management on private properties. By the new FC, the length of riparian buffers to be recomposed (in permanent or intermittent streams with less than 10m wide) may be limited to 5m. The majority of first order streams have less than 10 m wide, and therefore, the majority of first order streams in Brazil are under pressures from riparian buffer reduction, which help in reducing the anthropogenic effects (URIARTE et al., 2011).

To detect the anthropogenic changes in aquatic ecosystems, the use of biological communities as bioindicators has been successfully applied in monitoring programs in temperate regions. The most common organisms that are used as bioindicators of anthropogenic activities in streams are the macroinvertebrate and benthic biofilm communities. In tropical streams, several studies have observed changes in macroinvertebrate communities due to changes in water quality. However, few studies have attempted to use the benthic biofilm communities to detect anthropogenic activities in tropical headwater streams.

The benthic biofilm is a community composed of algae, bacteria, fungi and protozoan, embedded in an exopolysaccharide matrix, living attached to an organic or inorganic substrate (SABATER et al., 2016) (Figure 1.3). Benthic biofilm responds for nutrient enrichment because is mainly composed of algae and bacteria and respond to sediment addition trough losses of colonization area due to sediment deposition. Therefore, we expected that this community could be a good indicator of agricultural activities in tropical headwater streams.

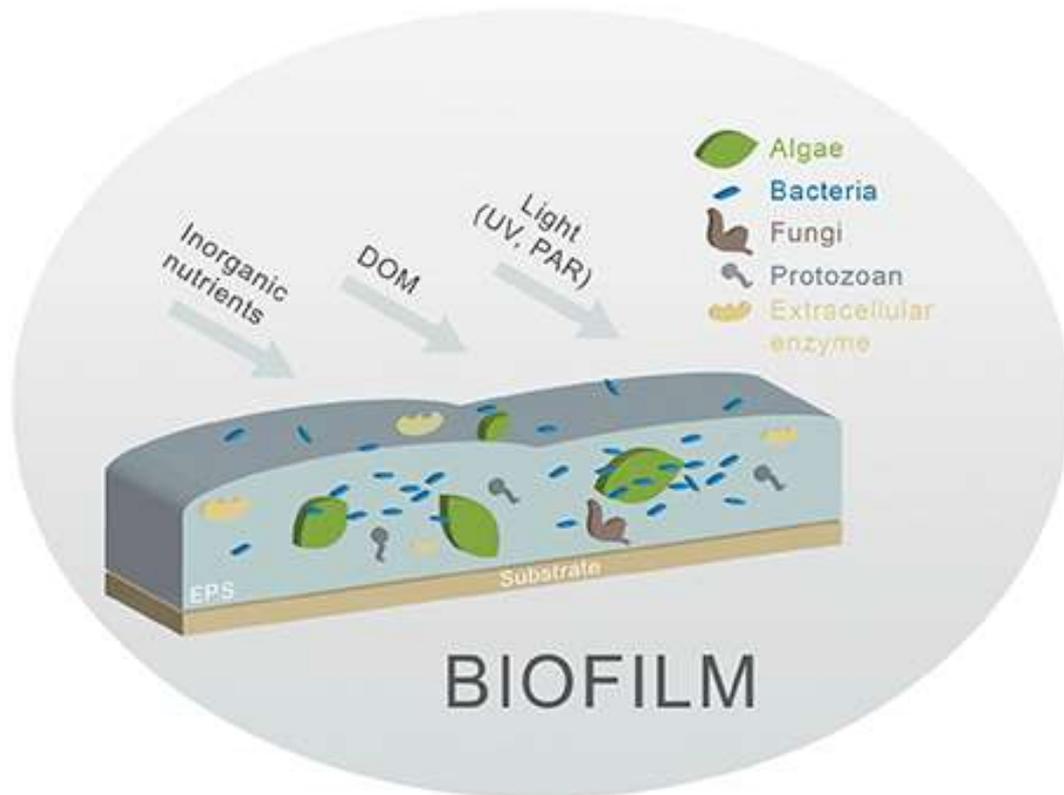
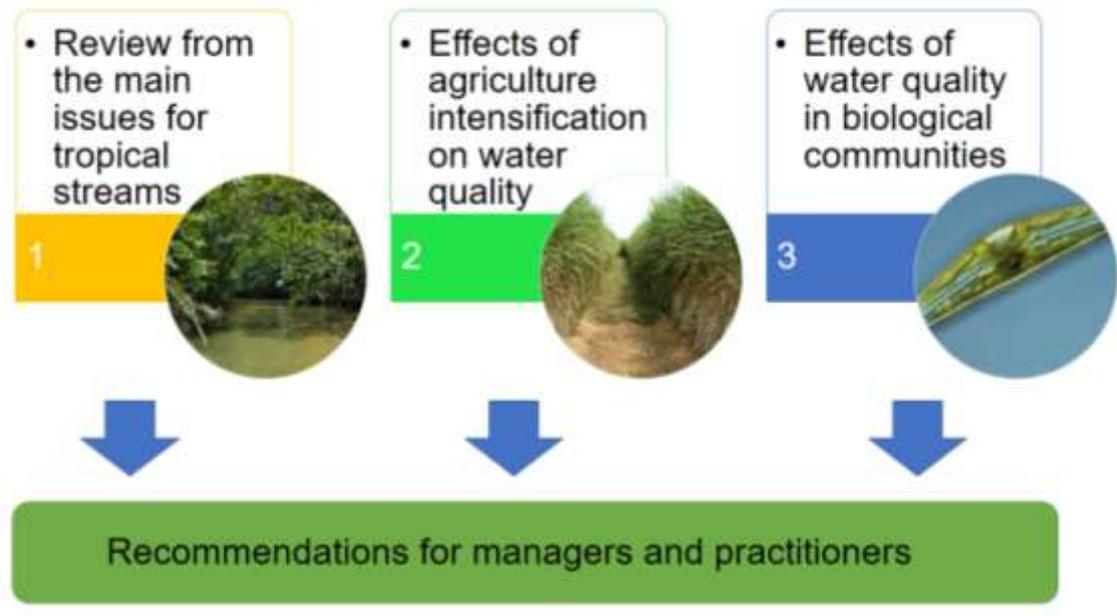


Figure 1.3 - Representation of the benthic biofilm and the main compositors of the community
 Source: Sabater et al. (2016). DOM – dissolved organic matter, UV – ultraviolet, PAR – photosynthetic active radiation, EPS - exopolysaccharide matrix.

Based on these assumptions, this thesis has been divided into three chapters, each one to address specific issues that streams located in the tropical zone has experienced (Figure 1.4):

- The first chapter will contribute to understanding what are the main issues that tropical streams have been experiencing, focusing on multiple stressors and climate change effects.
- The second chapter analyzes how the conversion of low-intensity pasturelands to high-intensity bioenergy crops changes the water quality parameters in streams located in the Corumbataí river basin (São Paulo, Brazil), and also analyzes how important are the riparian forests in the headwater zone to provide better water quality in bioenergy crops.
- The third chapter analyzes how water quality and climatic characteristics affect benthic diatom community dynamics in tropical headwater streams, which is the main instream autochthonous resource.

- Finally, we provide a broad conclusion and recommendations for managers and practitioners based on these three chapters.



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2 CLIMATE CHANGE AND MULTIPLE STRESSORS IN TROPICAL HEADWATER STREAMS

Abstract

Despite the importance of small tropical streams for maintaining freshwater biodiversity and providing essential ecosystem services to humans, relatively few studies have investigated multiple-stressor effects of climate and land-use change on these ecosystems, and how these effects may interact. To illustrate these knowledge gaps, we reviewed the current state of knowledge regarding the ecological impacts of climate change and catchment land use on small tropical streams. We consider effects of predicted changes in streamflow dynamics and water temperatures on water chemistry, habitat structure, aquatic biota and ecosystem processes. We highlight the pervasive individual effects of climate and land-use change on algal, macroinvertebrate and fish communities, and in-stream metabolism and decomposition processes. We also discuss expected responses of tropical streams in a multiple-stressor scenario, considering the predictions of higher temperatures and shifts in hydrological dynamics. Finally, we identify six key knowledge gaps in the ecology of low-order tropical streams and indicate future research directions that may improve catchment management in the tropics to help alleviate climate change effects and biodiversity losses.

Keywords: Tropical streams; Multiple stressors; Climate change; Agriculture

2.1 Introduction

More than 50% of inland freshwater habitats were lost during the twentieth century, and most of those that remain are degraded around the globe due to changes in land cover and land use, introduction of invasive species, hydrologic modification, overharvesting, pollution and climate change (HASSAN; SCHOLLES; ASH, 2005). Global climate change will affect not only multiple levels of biological organization but may also interact with other stressors to which freshwater ecosystems are also exposed (WOODWARD et al., 2010). Freshwater habitats hold a disproportionately high biodiversity relative to their area. For example, surface freshwater habitats represent only 0.01% of the world's water and 0.8% of Earth's surface, yet they contain about 9.5% of the animal species described on Earth (DUDGEON, 2010; DUDGEON et al., 2006).

Tropical streams represent one of the planet's most biodiverse freshwater ecosystems, but also one of the most threatened (BOULTON et al., 2008; DUDGEON et al., 2006; WANTZEN et al., 2006). These ecosystems located in biodiversity hotspots (MITTERMEIER et al., 1998; MYERS 1990) often differ from temperate streams, especially in terms of climatic conditions, geomorphology, landscape evolution and geological history

(WANTZEN et al., 2006). However, the main ecological processes in them are likely to be driven by the same variables that are important in temperate streams (BOULTON et al., 2008). Draining many different types of soils and vegetation, tropical streams have many specific characteristics that imply much is to be learned about their ecology (BOYERO et al., 2009). For example, tropical streams receive more solar radiation, have lower seasonal climatic variation and higher water temperatures than temperate streams, and are subjected to higher chemical weathering due to the year-round warm temperatures (LEWIS JR., 2008). Besides these natural factors, tropical streams are affected by multiple anthropogenic pressures, mostly due to changes in catchment/riparian land use but increasingly also by climate change (BOYERO et al., 2009; GUECKER; BOECHAT; GIANI, 2009). These anthropogenic pressures are more severe in tropical countries (SMITH et al., 2010), increasing the likelihood of ecosystem degradation. Due to all these factors acting simultaneously when affecting aquatic communities, it is likely that interactions of physical and chemical variables and biological communities in tropical streams will cause responses in a different range to that observed in temperate streams.

In recent decades, global climate change has become a “hot topic” in the biological sciences due to the numerous effects this change is expected to have on ecosystems (HELLER; ZAVALETA, 2009). In running waters, many of the predicted changes to ecosystems are linked to changes in the hydrological cycle and water temperature (DODDS et al., 2015). Evidence suggests that in the tropics these changes may be abrupt, with effects unprecedented in the last 5200 years (THOMPSON et al., 2006). Tropical areas will experience marked changes in temperature and alterations in the timing and amount of rainfall, and these are expected to occur earlier than in other parts of the globe (INTERGOVERNMENTAL PANEL ON CLIMATE CHANGE – IPCC, 2014; MORA et al., 2013; O’GORMAN, 2012). These changes, in turn, will increase the risk of droughts, floods and landslides and compounded stress on water resources (IPCC, 2014), with direct effects on the structure and functioning of tropical streams and rivers (DAVIES, 2010; JIMÉNEZ-CISNEROS et al., 2014). They also represent an important challenge for catchment management, especially because climate change effects on freshwater biota are likely to become evident earlier in the tropics than elsewhere, due to the relatively small natural climate variability in tropical regions that generates narrow climate bounds (MORA et al., 2013). A changing climate exceeding these bounds will likely be stressful for biological communities adapted to this narrow climate range (MORA et al., 2013), contributing to high extinction rates in the tropics (CEBALLOS et al., 2015). Considering the higher biodiversity

and faster human population growth in the tropics, tropical small streams may be disproportionately at risk compared to temperate streams in the face of ongoing global change, resulting in negative and interactive effects on ecosystem structure and function, ecosystem services, water quality, biodiversity and water availability.

Climate change effects on streams have been studied mainly in temperate systems to date. In these ecosystems, a number of potential effects on biogeochemical cycles and biological communities have been observed (BARON; SCHMIDT; HARTMAN, 2009; BUISSON et al., 2008; DURANCE; ORMEROD 2007), although potential interactions between key climate change drivers and other human-induced stressors such as those linked to agriculture remain largely unknown (PIGGOTT et al., 2015a, 2015b, 2015). By contrast, there has been little research into climate change effects on small tropical streams, and even less research on how predicted land-use changes will interact with climate change in these ecosystems. It is likely that the threats to the integrity of small tropical streams are different from temperate streams because tropical streams are subjected to a different range of temperatures and land use dynamics and are situated within a region that is home to almost half of the global human population (BOYERO et al., 2009; HARVEY et al., 2014).

Small headwater streams are fragile ecosystems highly susceptible to anthropogenic impacts, reflecting their direct connections to the adjacent landscape that influence the supply, transport and fate of water and solutes in a catchment (ALEXANDER et al., 2007). Modifications in these areas also expose downstream receiving water bodies to the cumulative effects of upstream activities (COVICH; CROWL; HEARTSILL-SCALLEY, 2006; LORION; KENNEDY 2009). In tropical streams, anthropogenic landscape modifications can result in short and unpredictable flood pulses, which often “reset” the physical and biotic environment (JUNK et al., 1989). Short and unpredictable flood pulses in tropical streams may occur more frequently under land-use and climate change, shifting the functional dynamics of the ecosystem. Despite increasing concern about how climate and land-use change will affect freshwater ecosystems globally, few studies have focused on small tropical streams, highlighting the need for climate change and multiple stressor research in these ecosystems (RAMIREZ et al., 2008).

In this paper, we reviewed global change-related studies in small streams (up to 3rd stream order) within the tropical zone (between 23 °N and 23° S). We focused on small streams because they are one of the most widespread freshwater ecosystems and generally represent the majority of water bodies in a catchment (BENDA et al., 2005). Some topics discussed hereafter remain largely unstudied in tropical systems. In these cases, we

extrapolate likely scenarios based on well-established general biological and physical principles or theories.

2.2 Flow dynamics

Flow regimes play a major role in determining the structure and functioning of running water ecosystems (BUNN; ARTHINGTON, 2002; POFF et al., 1997, 2010; RICHTER et al., 1996). The flow dynamics of a stream are controlled mainly by the distribution and amount of rainfall, the catchment relief, and land use characteristics in the catchment (DEFRIES; ESHLEMAN 2004; FOLEY et al., 2005; MACEDO et al., 2013; STANFIELD; JACKSON, 2011). In the tropics, the massive and ongoing conversion of forests to other land uses such as agriculture and urbanization are altering flow regimes (CARLSON et al., 2014; WU et al., 2007) and changing stream characteristics (Table 1). Given the forecast for increasing tropical precipitation extremes due to climate change (IPCC, 2014; O'GORMAN, 2012), further modifications of streamflow dynamics can be expected, with tropical small streams generally becoming flashier.

The tropical wet season, characterized in several regions by monsoonal rains, plays an important role in agriculture, hydroelectricity, industry and providing the basic needs for the human population (TURNER; ANNAMALAI, 2012). During this period, considerable changes occur in small stream ecosystem dynamics, starting with flow patterns that, in turn, affect nutrient and carbon concentrations, sediment inputs, channel structure and biological communities (Table 1). Thus, elevated stream flows limit benthic algal biomass accrual and affect benthic macroinvertebrate and fish communities directly via physical disturbance and indirectly via changes in resource availability, favoring species that are well-adapted to fast-flowing, frequently disturbed lotic environments (CARVALHO; TEJERINA-GARRO 2015; JUNK et al., 1989; NOLTE et al., 1997; PRINGLE; HAMAZAKI 1997; ROSSER; PEARSON 1995; TOWNSEND; DOUGLAS, 2014). Wet-season-induced changes occur in both natural and anthropogenic environments, but more strongly in the latter. Because of anthropogenic land use changes, lateral flow paths (surface and subsurface runoff) are more frequently active, eroding soil as well as nutrients and carbon previously stored in the soil and carrying them into streams (ALLAN, 2004; DUNNE, 1979; NEILL et al., 2006). Observational evidence suggests that land use changes associated with agricultural intensification can also reduce the monsoonal rainfall in some parts of the tropics (NIYOGI et al., 2010). Therefore, climate change effects during the wet season are likely to differ among

tropical small streams depending on land use intensity in the catchment and geographical location, because some regions may become drier whereas others may become wetter, with different consequences for the structure and dynamics of tropical small streams and shifts in aquatic biological communities.

During wet-season rainfall events (which are often intense in the tropics), high flows result in structural changes in the stream channel, its floodplain and along its banks, and surface runoff (i.e. overland flow). This results in increased inputs of allochthonous organic matter from the riparian zone (including large woody debris) and sediment from the adjacent catchment or due to landslides (Table 1). The addition of large woody debris increases the organic carbon concentration in the water and causes obstructions in the stream channel, which can create large pools favoring sediment retention (JOHNSON et al., 2006; WOHL; OGDEN, 2013). In a climate-change context, the magnitude of these processes will exceed their current natural variation due to the predicted changes in the global hydrological cycle and are likely to be more severe in modified landscapes. For example, in modified catchments, sediment export rates from terrestrial soils to streams are higher due to soil instability induced by vegetation removal, soil exposure and faster runoff (ALLAN, 1995; DUNNE, 1979). Changes in riparian vegetation are expected due to longer periods of exposure to flooding and other changes in streamflow dynamics (AUBLE; FRIEDMAN; SCOTT, 1994; GARSSSEN et al., 2015). Therefore, shifts in riparian vegetation composition and catchment sediment yield are expected in tropical small streams, depending on the land use and climate change intensity in the catchment, with consequences for biological community structure due to shifts in allochthonous organic matter inputs and streambed modifications.

Another flow-related consequence of climate change is that droughts are expected to become more severe, particularly in some tropical regions (HIRABAYASHI et al., 2008; IPCC, 2014; LI et al., 2009). Although recovery of stream ecosystems after droughts is often rapid, their impacts can be disproportionately severe once critical thresholds are exceeded (BOULTON, 2003). During the tropical dry season, streamflow dynamics are typically stable and, in the case of intermittent streams, flow often ceases completely. In natural environments, these changes in flow dynamics contribute to shaping biological communities (LAKE, 2000). When combined with changes in catchment land use such as agriculture and urbanization, the dry-season flow dynamics in tropical streams become more variable and pollutant concentrations in the water can be higher, which can then reduce or shift diversity of aquatic organisms (LONGO et al., 2010; NOLTE et al., 1997). Thus, because current climate

change forecasts for the tropics predict more frequent and severe droughts (IPCC, 2014), tropical stream ecosystem functioning may be compromised, especially in catchments strongly modified by human land use, because severe droughts followed by slow recolonization will alter biological communities and modify stream processes.

It is well established that some of the main problems related to flow regime modifications are due to human land-use activities and poor landscape management, such as dam construction and agricultural water abstraction, both in temperate regions (ALLAN, 2004; BUNN; ARTHINGTON, 2002; DUDGEON et al., 2006; POFF et al., 1997; POWER et al., 1996) and in the tropics (CHAVES et al., 2008; GERMER et al., 2009; WU et al., 2007). These problems are even more serious in the tropics where agricultural intensification is a key factor for human development, via food production and climate mitigation (DEFRIES; ROSENZWEIG, 2010). Evidence suggests that tropical forest cover is among the most important factors for the protection of streams, and existing research recognizes the critical role played by forests in maintaining soil permeability and thus producing more base flow in streams during the tropical dry season (BRUIJNZEEL, 2004; OGDEN et al., 2013). However, in many parts of the humid tropics, the areas covered by disturbed forests (e.g. logging, slash-and-burn agriculture, mining) have become larger than those covered by undisturbed forests, and this trend is ongoing (HANSEN et al., 2013; KIM et al., 2015; LAWRENCE; VANDECAR, 2015; WOHL et al., 2012). Because disturbed tropical forests are less efficient at reducing streamflow variability during high intensity rainfall events or droughts (FERRAZ et al., 2014), disturbed forests may not be able to mitigate climate change effects in agricultural landscapes. Therefore, well-designed, large-scale forest restoration programs are urgently needed throughout the tropics (LATAWIEC et al., 2015), in order to reinstate the original hydrological patterns and alleviate climate change effects in tropical small streams.

Table 2.1 - Stream responses to changes in rainfall and land use intensity in the tropical zone

Effects	Physicochemical responses	Biological responses	Location	Stream order	Reference
Increase in land use intensity	Streamflow increase	Not evaluated	Mexico	1st and 2nd	MUÑOZ-VILLERS; MCDONNELL, (2013)
			Africa	1st	RECHA et al., (2012)
			Brazil	1st	CHAVES et al., (2008); GERMER et al., (2009)
	Surface runoff	Not evaluated	Brazil	1st and 2nd	GUZHA et al., (2015); CHAVES et al., (2008); GERMER et al., (2009)
Rainfall increase	Wood and carbon export to streams	Not evaluated	Panama;	2nd	WOHL; OGDEN, (2013)
			Brazil	1st	JOHNSON et al., (2006)
	High flow	Reduction in epilithic biomass, high taxon richness due to rare taxa	Australia	1st to 5th	TOWNSEND; DOUGLAS, (2014)
	High flow	Steady algal biovolume in the presence of fishes, without fishes decreases in algal biovolume, richness and diversity in fish exclusion treatments	Costa Rica	3rd to 4th	PRINGLE; HAMAZAKI, (1997)
	High flow	Increased diversity and density of mayflies	Brazil	3rd to 4th	NOLTE et al., (1997)
	High flow	Reduced density and richness of macroinvertebrate communities	Australia	2nd and 3rd	ROSSER; PEARSON, (1995)
	High flow	Reduced abundance of macroinvertebrate communities in riffles	Costa Rica	4th	RAMIREZ; PRINGLE, (1998)
Rainfall decrease	Low flow, increase in flow variability and instability	Not evaluated	Hawaii	1st to 3rd	STRAUCH et al., (2015)
	Low flow	Reduced density and richness of macroinvertebrate communities, increase in the dominance of a mayfly genus	Brazil	3rd to 4th	NOLTE et al., (1997)
	Low flow, isolated pools	Increase in dominant species (<i>Atya lanipes</i> Hothuis, 1963), decrease in reproductive output, decrease in dominant shrimp (<i>Macrobrachium</i> spp. Bate, 1868)	Puerto Rico	1st and 2nd	Covich et al., (2006; 2003)

2.3 Water temperature

Rising air and water temperatures are the most commonly observed symptom of global climate change to date, with accelerating further increases forecast for the future (IPCC, 2014). Rising temperatures have been observed since the last century and are correlated with atmospheric carbon concentrations (IPCC, 2014). In streams, water temperature controls a multitude of processes, regulating the metabolic activity from individuals to ecosystems (BROWN et al., 2004). Tropical streams have lower seasonal climatic variation and higher water temperatures compared to temperate streams (MORA et al., 2013), are subjected to higher chemical weathering due to the year-round warm temperatures, receive more organic matter and have naturally higher biodiversity compared to temperate streams (BOULTON et al., 2008; DUDGEON et al., 2006; RAMIREZ et al., 2008). When combined, these characteristics suggest that tropical streams may be particularly vulnerable to climate-change-induced increases in temperatures. Despite this, the effects of rising water temperatures on tropical small streams remain poorly understood (Table 2). Due to the importance of water temperature on biological communities in tropical small streams, temperature-sensitive taxa may become excluded at elevated temperatures, causing reduced diversity.

Previous studies have documented the effects of water temperature on benthic macroinvertebrate communities in tropical small streams (JACOBSEN et al., 1997; SIQUEIRA et al., 2008; YULE et al., 2009). Benthic invertebrates play an important role in stream ecosystem functioning, for example for nutrient cycling, primary productivity, organic matter decomposition and translocation of materials (WALLACE; WEBSTER 1996). Tropical macroinvertebrate communities feature some special characteristics, including a rareness or absence of shredders - a functional group generally more abundant in temperate or cold environments where this guild plays an important role on organic matter decomposition (BOYERO et al., 2012). Few studies in tropical streams have investigated temperature effects on macroalgae, diatoms or fish (Table 2). These groups of organisms also play important roles in stream community structure and functioning, and changes in these communities have been shown to modify and destabilize aquatic food webs (COAT et al., 2009; MOTTA; UIEDA, 2005). Together, microbial communities and primary producers regulate stream metabolism rates, which are strongly temperature-dependent and may be compromised by climate change induced warming (ORTIZ-ZAYAS et al., 2005; REZENDE et al., 2014).

Existing research recognizes the critical role played by temperature on leaf litter decomposition in tropical small streams, an integral functional process for energy flux

through stream ecosystems (ABELHO; CRESSA; GRAÇA, 2005; ABELHO et al., 2010; BENSTEAD, 1996; MATHURIAU; CHAUVET, 2002). Recent evidence suggests that climate change effects on tropical streams will likely accelerate microbial litter decomposition via raising water temperatures while reducing detritivore-mediated decomposition via loss of detritivore species (BOYERO et al., 2011). Thus, although net decomposition rates may remain unchanged, carbon dioxide production by microbial activity is likely to increase, possibly further accelerating climate warming and having additional adverse ecological effects (BOYERO et al., 2011). Consequently, it is expected that carbon dioxide production in tropical streams will increase more than that in temperate streams as the Earth keeps warming, despite the higher rates of ecosystem respiration in tropical streams (ORTIZ-ZAYAS et al., 2005). The resulting increase in carbon dioxide production could be considerable, given that a recent study found that about 28 % of the carbon dioxide emission from stream and rivers in temperate regions was produced by aquatic metabolism (HOTCHKISS et al., 2015).

Table 2.2 - Ecological responses of stream communities to increasing water temperatures in the tropical zone

Response	Location	Stream order	Reference
Reduced abundance and richness of shredding aquatic insects	Malaysia	1st to 3rd	YULE et al., (2009)
Increase in aquatic insect orders and families	Ecuador	1st to 3rd	JACOBSEN et al., (1997)
Increase in the emergence of the aquatic insect Orthocladiinae (Chironomidae)	Brazil	1st	SIQUEIRA et al., (2008)
Shifts in macroalgae community	Mexico	3rd to 5th	BOJORGE-GARCÍA et al., (2010)
Increase in the respiration rates of stream whole metabolism	Puerto Rico	1st to 3rd	ORTIZ-ZAYAS et al., (2005)
Shifts in fish community structure	Brazil	3rd to 5th	ROLLA et al., (2009)
	Brazil	1st to 4th	PINTO et al., (2009)
Increase in leaf breakdown	Costa Rica	1st	BENSTEAD, (1996)
	Colombia	4th	MATHURIAU; CHAUVET, (2002)
	Brazil,	3rd	ABELHO et al., (2010)
	Brazil	1st to 4th	REZENDE et al., (2014)

2.4 Agriculture

Agriculture is one of the key human activities in the context of biodiversity loss and climate change in the tropics because tropical regions will need to increase their food production while, at the same time, mitigate climate change effects (DEFRIES; ROSENZWEIG, 2010). Although agriculture provides food, labor and improves the economy of tropical regions, it is one of the main causes of ecosystem degradation and biodiversity loss worldwide (TILMAN, 1999). When combined with climate change, the detrimental effects of agriculture in tropical regions may become even more severe, due to reduced freshwater availability and rising water demand (ELLIOTT et al., 2014; VÖRÖSMARTY et al., 2000), leading to increased water abstraction and eutrophication. Climate change is also predicted to have negative effects on agricultural development and production in the tropics, due to changes in temperature, precipitation and soil moisture (LAWRENCE; VANDECAR 2015). Therefore, tropical regions urgently need to improve their climate change mitigation/adaptation strategies to ensure the sustainability of agricultural systems.

The main impacts of agriculture on stream ecosystems are related to increased inputs of nutrients, fine sediment and agrochemicals, changes in streamflow dynamics due to land use conversions, and water temperature changes caused by forest removal (LAWRENCE; VANDECAR, 2015; POFF et al., 1997; TILMAN, 1999). Tropical streams are likely to suffer increasingly from these impacts because agricultural intensification for food production is predicted to rise by at least 50% in the tropics by 2050 (DEFRIES; ROSENZWEIG, 2010; GODFRAY et al., 2010). Consequently, many tropical regions are subjected to high rates of deforestation, agricultural expansion and increasing erosion (RAMIREZ et al., 2008). When combined with climate change effects, the resulting challenges for tropical stream ecosystems are likely to worsen due to increasing surface runoff and erosion rates driven by rainfall increases (in quantity and/or intensity) (TUCKER; SLINGERLAND, 1997), and due to the interactive effects of rising water temperatures coupled with agricultural contaminants (see below).

Effects of agricultural practices on small tropical streams have been studied in various organisms. In fish communities, high functional redundancy (i.e., different species performing similar functions in the ecosystem) has been observed in agricultural landscapes and deforested streams, due to reduced habitat heterogeneity by grass dominance instead of forest in the riparian zone (CASATTI et al., 2015; TERESA et al., 2015). In macroinvertebrate communities, reduction of biodiversity and elimination of rare taxa was related to

deforestation for agricultural practices (BENSTEAD; DOUGLAS; PRINGLE, 2003; LORION; KENNEDY, 2009; SIQUEIRA et al., 2015). Key ecosystem processes in streams are also modified by agricultural practices, such as organic matter decomposition (SILVA-JUNIOR et al., 2014) and stream metabolism (GUECKER; BOECHAT; GIANI, 2009). Moreover, these changes in stream communities and ecosystem dynamics may be increased by climate change effects via non-additive interactions of nutrient and sediment addition, flow dynamics changes and temperature changes. Because it remains unknown how such interactions will affect tropical stream communities, this represents an important area for future research.

2.5 Climate change and multiple stressors

A stressor can be defined as a variable that, as a result of human activity, exceeds its range of normal variation and adversely affects individual taxa or community composition (TOWNSEND et al., 2008). Most present-day ecosystems, including those in running waters, are affected by multiple stressors acting simultaneously (CRAIN; KROEKER; HALPERN, 2008; NÖGES et al., 2015; ORMEROD et al., 2010; TOWNSEND et al., 2008; VINEBROOKE et al., 2004) or sequentially (CHRISTENSEN et al., 2006). Studies in freshwaters have shown that stressors interact frequently, resulting in complex, non-additive outcomes that cannot be predicted based on the effects of the individual stressors involved (FOLT et al., 1999). Synergistic interactions occur when the combined outcome of multiple stressors is larger than predicted based on the individual effects involved, and antagonistic interactions occur when the combined outcome is less than predicted (FOLT et al., 1999; PIGGOTT et al., 2015).

Studies examining effects of multiple stressors on running water ecosystems exist mainly from temperate regions. A meta-analysis considering 286 responses of freshwater ecosystems to paired stressors revealed more antagonistic responses than synergistic or additive ones, possibly due to the environmental variability of streams that foster the potential for acclimation and co-adaptation to multiple stressors (JACKSON et al., 2016). A streamside mesocosm experiment in New Zealand (PIGGOTT et al., 2015a, 2015b, 2015) manipulated two key agricultural stressors (nutrients and deposited fine sediment) and water temperature (to simulate climate warming up to 6 degrees Celsius above ambient temperatures), to determine the individual and combined effects of these stressors on periphyton and macroinvertebrate communities and organic matter decomposition. In this experiment,

stressors had pervasive individual effects but in combination produced many synergistic or antagonistic effects, both at the population and community level. For example, three invertebrate community-level metrics of stream health routinely used around the world all showed complex three-way interactions, with either a consistently stronger temperature response or a reversal of its direction when one or both agricultural stressors were also in operation (PIGGOTT et al., 2015b). Moreover, the negative effects of added sediment on invertebrate communities were often stronger at raised temperatures but less so when nutrients were added as well, suggesting that streams already impacted by high sediment loads may be further degraded under a warming climate, but the degree to which this will occur may also depend on in-stream nutrient conditions.

In another mesocosm study, one of the first manipulative experiments in a tropical stream (O'CALLAGHAN et al., 2015), increases in macroinvertebrate drift abundance were observed in response to elevated nutrient concentrations, and increases in macroinvertebrate drift abundance and taxon richness in response to elevated fine sediment levels.

As discussed above, small tropical streams are exposed to a number of stressors originating in agricultural activities (nutrient enrichment, sedimentation, deforestation, flow regime and water temperature changes due to deforestation), and these stressor impacts are likely to be modified by climate change effects. Consequently, complex interactions between multiple stressors and climate change are highly probable and studies in small tropical streams are urgently needed.

2.6 Conclusions and future research needs

We have identified six key areas urgently requiring future research efforts to advance our understanding and management of small tropical streams in the face of global change:

- (1) **More studies on temperature effects on tropical streams.** There is a lack of information about the influence of water temperature on tropical stream communities. Due to the ongoing effects of global climate change, we need to increase our knowledge on this topic in order to prevent biodiversity loss and maintain the functioning and ecosystem services provided by small tropical streams, such as provision of drinking water and nutrient recycling.

- (2) **Drought and flood effects on tropical streams.** More extreme droughts and floods are expected under a climate change scenario in tropical streams, and such extreme events are already happening in several parts of the tropics. These events can change the structure of streams and are stressful for aquatic communities, especially in systems where these events were less common in the past. Therefore, we encourage studies taking advantage of such events to help improve our understanding of shifts in aquatic communities.

- (3) **More studies on applied topics to help alleviate agricultural and climate change pressures.** Agriculture is a key human activity in the tropics, providing food, labor and development. Therefore, more research focusing on methods to alleviate agricultural impacts on tropical streams, combined with best landscape and catchment planning to avoid excessive surface runoff during high intensity storms and maximize the efficiency of fertilizers, should be conducted. Such research is required to maintain the sustainability of agriculture and water resources in the tropics in the face of climate change.

- (4) **Studies on multiple-stressor effects in tropical streams.** The role of multiple stressors in shaping tropical freshwater communities including streams is poorly understood. Therefore, more realistic and powerful experiments should be performed to understand the effects of the key agricultural stressors (e.g. nutrients, fine sediment, pesticides and light excess due to riparian deforestation), urbanization and climate change effects (e. g. water temperature, streamflow) on tropical stream communities.

- (5) **Climate change mitigation policies and strategies to ensure the sustainability of tropical freshwater resources.** There is scant information about how climate change will interact with agricultural stressors in tropical streams. Consequently, policies to manage tropical small streams and agriculture under climate change are absent or poorly informed. This is important due to potential unpredictable non-additive outcomes that are likely to affect freshwater biota and ecosystem services. Therefore, after a comprehensive study on multiple stressor effects in tropical streams, policies and strategies should be established to avoid damage to these ecosystems.

- (6) **Restoration of tropical streams.** Most existing studies of tropical stream restoration examined the implementation of riparian vegetation buffers, which is one of the most important components for the stream function and dynamics. However, implementations of other techniques to avoid habitat loss, invasive species, flow alterations, sedimentation and bank instability should also be investigated, especially due to the particular tropical soil characteristics and ongoing expansion of agricultural frontiers in the tropics.

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3 IMPACTS OF CONVERTING LOW INTENSITY PASTURELAND TO HIGH INTENSITY BIOENERGY CROPS ON THE WATER QUALITY OF TROPICAL HEADWATER STREAMS

Abstract

Sugarcane crops for biofuel production have transformed landscapes in São Paulo State due to the high demand for energy. The expansion of these crops, mostly occurring over low-intensity pasturelands helps to meet the growing demand for bioenergy production in the country without contributing to deforestation. However, it is expected that the high-intensity bioenergy crops will affect headwater streams, due to the close connections of these ecosystems to adjacent lands. Therefore, we have tested if the conversion of low-intensity pasturelands to high-intensity sugarcane crops are degrading water quality in tropical headwater streams. In addition, we tested if the conservation status of headwater's in bioenergy crops helps to alleviate the negative effects of bioenergy crops in the water quality. For this, field sampling was conducted in two sets of paired catchments, aiming to test the conversion effects and the headwater condition effects in the water quality parameters. We found that the conversion of low-intensity pasturelands to high-intensity sugarcane crops are elevating the concentrations of nitrate and suspended solids whereas reducing dissolved organic carbon and water temperature. The presence of forests in the headwater zone of sugarcane crops contributed to reducing the concentrations of nitrate, dissolved organic carbon and conductivity and increased dissolved oxygen concentrations. Most of the impacts on water quality in both treatments occurred mainly in the rainy season. Therefore, we conclude that the conversion of low-intensity pasturelands to high-intensity sugarcane crops are reducing the water quality in tropical headwater streams. The presence of well-preserved forests in the headwater area can help to prevent the water quality degradation, especially in the wet season. Best agricultural practices should be implemented to help alleviate the negative impacts of bioenergy crops in tropical headwater streams.

Keywords: Sugarcane; Tropical Streams; Water quality; Pasture; Season

3.1 Introduction

Streams around the world have been degraded due to several anthropogenic activities (DUDGEON et al., 2006; BOYERO et al., 2009; GUECKER; BOECHAT; GIANI, 2009), however, these pressures in tropical regions are likely to be more severe, due to agriculture intensification (DEFRIES; ROSENZWEIG, 2010). Expansion of crops for bioenergy is occurring at unprecedented rates in several regions of the world, especially in the tropics where bioenergy crops represent an important economic activity (LANGEVELD et al., 2014). For instance, Brazil is the second largest ethanol producer in the world with a sugarcane area of more than 10 million ha (LANGEVELD et al., 2014). These landscape conversions for bioenergy crops, such as sugarcane, are altering the structure and functioning of freshwater systems throughout the tropics (DIAZ-CHAVEZ et al., 2011) and as a consequence, altering

the structure of biological communities (SIQUEIRA et al., 2015) in one of the most biodiverse regions in the world (DUDGEON et al., 2006).

The expansion of sugarcane crops for biofuel production in Brazil has occurred mostly over pasturelands and annual crops, especially in the southeast region of the country, where approximately 1.5 million ha of pasturelands were converted to sugarcane fields between 2000 and 2009 (FILOSO et al., 2015). The conversion of degraded pasturelands to sugarcane crops in the region is defensible since it helps to meet the growing demand for bioenergy production in the country without contributing to deforestation (ALKIMIM et al., 2015). However, land intensification substantially increases the traffic of heavy machinery, soil displacement, and the use of fertilizer and pesticides (MARTINELLI; FILOSO, 2008). Such activities are known to have deleterious consequences for freshwater ecosystems (DIAZ-CHAVEZ et al., 2011; CIBIN et al., 2016), especially for headwater streams, which not only usually represent the majority of water bodies in a catchment (BENDA et al., 2005), but that are also vital to maintain the water quality and health of entire river ecosystems (ALEXANDER et al., 2007). Despite that, studies of the impacts of converting pastureland to sugarcane crops on headwater streams in Brazil are rare.

Riparian buffers and best agricultural practices are widely recommended to minimize impacts of intensive agriculture on streams (DOSSKEY et al., 2010; FERREIRA et al., 2012; FILOSO et al., 2015). In the humid tropics, such measures should be especially effective at protecting headwater streams during the rainy season, since high-intensity rainfall is associated with higher upland runoff, and transport of soil particles and pollutants to streams (WOHL et al., 2012; WOHL; OGDEN, 2013). However, the position and configuration of the riparian forest in the landscape play an important role in reducing the impacts of intensive agriculture on streams (DE SOUZA et al., 2013; FERNANDES et al., 2014; FERRAZ et al., 2014).

In this paper, we investigate how stream water quality changes with the conversion of pastureland into bioenergy crop in southern Brazil and evaluate the importance of landscape configuration to the effectiveness of riparian forests. We hypothesize that: (i) The conversion of pastureland to sugarcane reduces the input of organic material into streams but increases nutrient and sediment loads; (ii) in the rainy season, the impacts of pasture on the water quality of streams should be higher than that of sugarcane due to landscape runoff characteristics; (iii) the water quality of streams draining watersheds with sugarcane is higher when the top of the headwater is preserved than when it is not (iv) ecosystem function provided by preserved springheads in retaining sediment and nutrient loads to streams in

sugarcane crops are higher in wet season. In addition, we applied a water quality index to summarize the land-use effects on stream water quality and provide a tool to help guide landscape and water resources management. We expect that the conversion of pasture to sugarcane will result in a relatively low water quality index, but the use of conservation practices such as riparian buffers and forested headwater will increase the index values.

3.2 Methods

- Study area

Samples were collected in two sets of paired catchments located in the Corumbataí River basin, located in State of São Paulo, southeast Brazil (centered on 47°40'W, 22° 40' S) (Figure 3.1), a watershed of 1700 km² which has historically experienced intensive land use change (FERRAZ et al., 2014). Sugarcane crops cover approximately 32% of the arable area of the river basin and are the main economic activity in the region. Other land uses in the basin are pasture (~ 25%) and forestry (~ 20%). The regional climate is Cwa, according to Köppen classification, with dry winters and wet summers (CETRA; PETRERE, 2006). The average temperature in the winter is 17°C while in the summer average temperatures exceed 22°C (FERREIRA et al., 2012).

The impacts of land-use conversion and landscape structure on the water quality of small-order streams were tested using paired catchments. This method involves the comparison of water quality in two catchments adjacent to each other and with similar characteristics regarding topography, soils, area and climate, which allow climate differences to be accounted for in the analysis (BROWN et al., 2005).

The first paired catchments were used to evaluate the impacts of land use conversion from pasture to sugarcane on the water quality of streams. Both catchments drained first order streams and had similar topography, channel configuration and soil type (Ultisols). In 1995, the catchments were predominantly covered by pasture but, from 2008 on, most of the pasture in one of them was converted into sugarcane crops. The catchment predominantly covered by pasture, catchment “P”, is 1.35 km² and had approximately 68 % of its total area occupied by cattle pasture, 15% by sugarcane, and 7% by forest. The catchment predominantly covered by sugarcane “SC” has an area of 1.37 km², where 76% was occupied by sugarcane, 14% by cattle pasture, and 11% by forest.

The second paired catchments were used to test the influence of forested headwaters on the water quality of streams draining sugarcane crops. Again, the catchments drained first order streams with similar topography, drainage system and soil type (Latosols). In 1962, one of the catchments was mostly covered by pasture and the other by sugarcane. Since 1978, the land use in both catchments had been converted into sugarcane crops. However, while one catchment had an area of preserved forest at the top of the headwater (SCRS), the other (SR) did not. The catchment with the preserved headwater forest (SCRS) has a total area of 0.43 km², mostly covered by sugarcane (54%) followed by forests (40%) including upland forest, riparian buffer, and headwater forest). The catchment without the preserved headwater forest (SCR) has a total area of 0.47 km², with 45% of total area covered by sugarcane crops and 53% by forests. The top of the headwater is covered by an invasive grass species (*Brachiaria spp*).

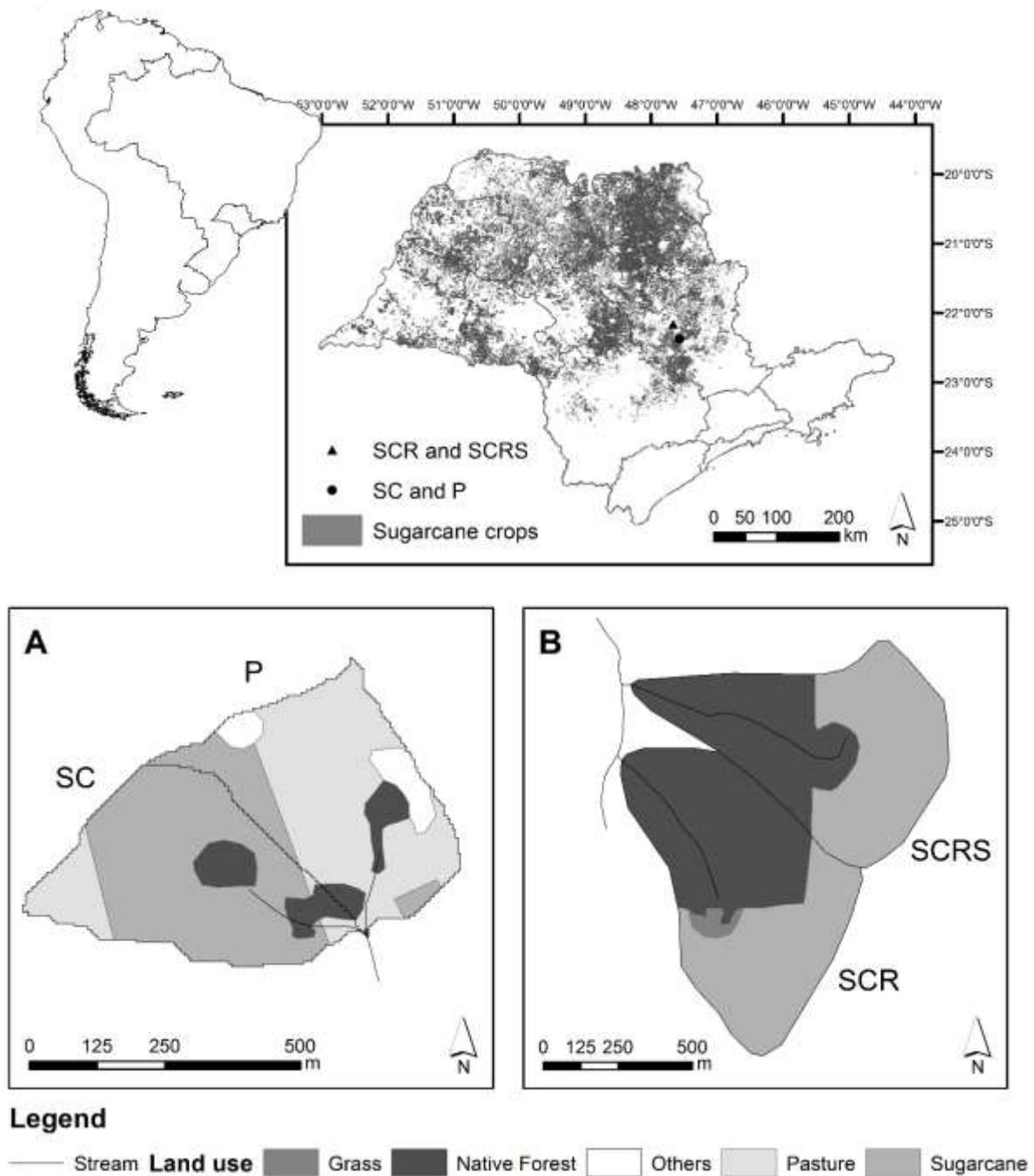


Figure 3.1 - Location of sampling sites Sugar Cane (SC), Pasture (P), SCR and SCRS in São Paulo state and the catchments land-use. (A) Conversion treatment, with pasture “P” and sugarcane “SC” land-uses. (B) Landscape configuration treatment, with preserved springhead streams “SCRS” and degraded springhead streams “SCR”. Figure adapted by author from Aguiar et al. (2011)

- Water chemistry

In each stream, water samples were collected twice a month during a period of one year, totalizing 96 samples ($n = 24 \times 4$). In the paired catchments used to investigate the

impacts of sugarcane conversion (“P” and “SC”), samples were collected between April 2011 and April 2012. Samples were collected from March 2013 to March 2014 in the catchments used to evaluate the influence of the headwater condition (catchments “SCR” and “SCRS”).

Water temperature, conductivity, and dissolved oxygen were measured on site with portable probes (YSI ProODO and Hanna HI 98360). Water samples were collected in the field using pre-washed 60 ml syringes coupled with plastic filter holders and 0.7 µm glass microfiber filter papers and transported to the laboratory in refrigerated containers to be processed and analyzed. Nitrate concentrations were determined using automatic continuous flow injection analysis (RUZICKA; HANSEN, 2000) by spectrophotometry after reduction with cadmium catalyst and reaction with sulfanilamide and N-naphthyl (GINE et al., 1980). Concentrations of dissolved organic carbon (DOC) and dissolved inorganic carbon (DIC) were analyzed in a Total Organic Carbon Analyzer (TOC 5000A, Shimadzu Scientific Instruments, Maryland, USA). Concentrations of total suspended solids were determined by the weight of residue retained in pre-dried 0.7 µm glass microfiber filter papers from unfiltered water samples (AMERICAN PUBLIC HEALTH ASSOCIATION - APHA, 2005).

- Integrated analysis of water quality

A water quality index was created to summarize the effects of land-use conversion and landscape configuration. For this, we calculated the quintiles of the concentrations of dissolved oxygen, conductivity, suspended solids and nitrate, which is known to have positive and negative effects on biological communities in each treatment (conversion and landscape configuration). Weights were applied on each quintile varying from 1 to 5, where the value 1 characterizes the higher values of conductivity, suspended solids and nitrate (above 80% of concentration values) and value 5 for the lowest concentrations (below 20% of concentration values). For dissolved oxygen, the weights were applied in the opposite way, assuming that higher values of dissolved oxygen represent better water quality. Therefore, the higher index values (value 5) represents the better water quality. After the application of the weights in each variable, a mean value of the weights was calculated for each sampling and summarized in box plot graphs, accounting the entire sampling period (one year). Mean values and coefficient of variation of the entire period for each catchment were calculated, assuming that the coefficient of variation represents the seasonal effects on the water quality index.

- Statistical analyses

To determine if water quality changes significantly with the conversion of pasture to sugarcane crops and with the presence or absence of preserved headwater, water quality parameters (water temperature, conductivity, dissolved oxygen, nitrate, dissolved carbon, inorganic dissolved carbon and suspended solids) were compared using Kruskal-Wallis test due to non-normal distributions of the data. Boxplots were used for graphical representation of the data variance.

To analyze seasonal influences on water quality parameters in the conversion and landscape configuration treatments, dry and wet periods were determined by the six driest months and the six wettest months through precipitation data obtained from CEAPLA – Centro de Análise e Planejamento Ambiental, São Paulo State University campus Rio Claro. The comparison between dry and wet season was made by Kruskal-Wallis test and box-plots for graphical representation of data variance.

3.3 Results

- Seasonal variation

We have classified dry and wet months separating the six driest months and the six wettest months through the year. The precipitation rates at SC and P catchments were characterized by wet summer and dry winter. In the catchments SCR and SCRS, the summer was mixed between dry and wet months, whereas the winter showed dry characteristics.

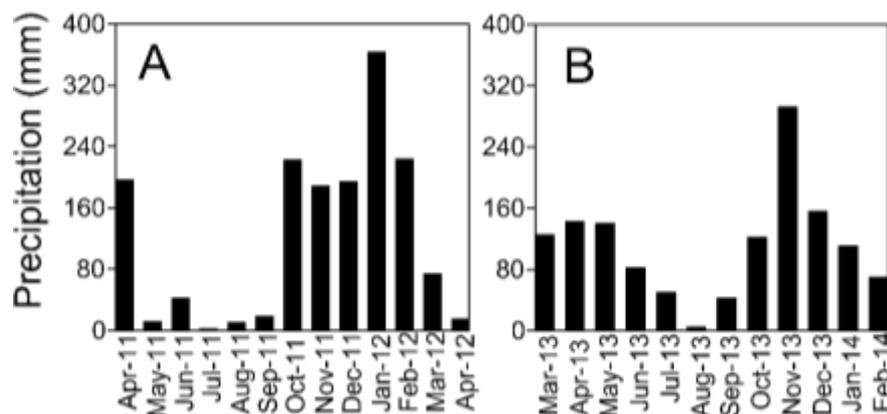


Figure 3.2 Total precipitation (mm) per month in the (A) SC and P catchments and (B) SCR and SCRS catchments during the sampling periods

- Changes in water quality due to conversion of pasture to sugarcane crops in tropical catchments

The conversion of pasture to sugarcane crops resulted in positive and negative effects on the water quality of the study streams (Figure 3.3). While water temperature, conductivity, and dissolved organic carbon concentrations decreased significantly with sugarcane crop conversion, concentrations of suspended solids and nitrate increased. Average nitrate concentrations were the most sensitive variable, increasing by a factor of four after sugarcane conversion (0.09 mg L^{-1} in the pasture, 0.44 mg L^{-1} in sugarcane). Suspended solid concentrations were also highly affected by land use conversion, with mean values about twice as high in sugarcane crops than in pasture (2.14 mg L^{-1} in the pasture, 6.04 mg L^{-1} in sugarcane).

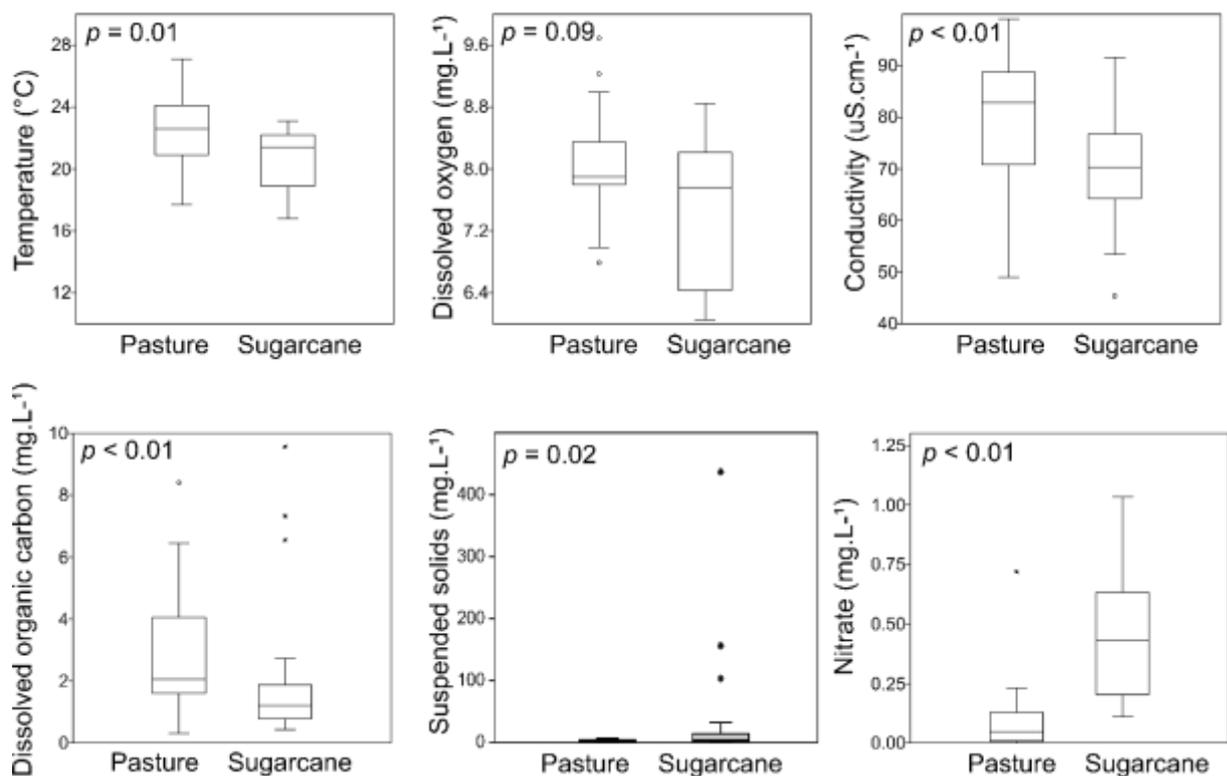


Figure 3.3 - Box plots and Kruskal-Wallis comparison of the conversion effects of pasture to sugarcane crops in tropical catchments

The physical and chemical characteristics of stream water also varied with seasonality (Table 3.1). Water temperature and dissolved organic carbon concentrations in sugarcane crops were significantly higher in the wet season than in the dry season. In the stream draining pastureland, water temperature and dissolved organic carbon were significantly higher in the

wet season. It is important to note that, despite the absence of statistical differences in nitrate concentrations in streams draining pasture during the wet and dry seasons, mean concentrations were far below those observed for sugarcane crops.

Table 3.1 - Mean values and standard errors (in brackets) of physical and chemical characteristics of water in dry and wet season, in sugarcane and pasture land uses

	Wet	Dry	Kruskal-Wallis
<i>Sugarcane</i>			
Temperature (°C)	21.76 (0.23)	18.88 (0.60)	$p < 0.01$
Dissolved oxygen (mg.L ⁻¹)	7.28 (0.26)	7.55 (0.35)	NS
Conductivity (µS.cm ⁻¹)	73.17 (2.78)	67.85 (2.85)	NS
Suspended solids (mg.L ⁻¹)	51.20 (36.03)	17.38 (12.71)	NS
Dissolved organic carbon (mg.L ⁻¹)	3.28 (0.82)	0.92 (0.09)	$p < 0.01$
Nitrate (mg.L ⁻¹)	0.38 (0.08)	0.50 (0.06)	NS
<i>Pasture</i>			
Temperature (°C)	23.60 (0.54)	21.32 (1.04)	$p = 0.05$
Dissolved oxygen (mg.L ⁻¹)	7.88 (0.12)	8.27 (0.35)	NS
Conductivity (µS.cm ⁻¹)	81.81 (2.98)	79.60 (3.85)	NS
Suspended solids (mg.L ⁻¹)	2.14 (0.58)	2.15 (0.69)	NS
Dissolved organic carbon (mg.L ⁻¹)	3.64 (0.44)	2.27 (0.60)	$p = 0.01$
Nitrate (mg.L ⁻¹)	0.04 (0.01)	0.14 (0.05)	$p < 0.05$

- Changes in stream water quality due to different landscape configurations

Water quality significantly differed in sugarcane catchments with different headwater forest condition (Figure 3.4). Dissolved oxygen concentrations were higher in the stream with headwater forest preservation than without it (headwater with invasive grass cover)) Mean conductivity values were about twice as low in preserved than in degraded headwaters, and average dissolved organic carbon and nitrate concentrations were about eight times lower.

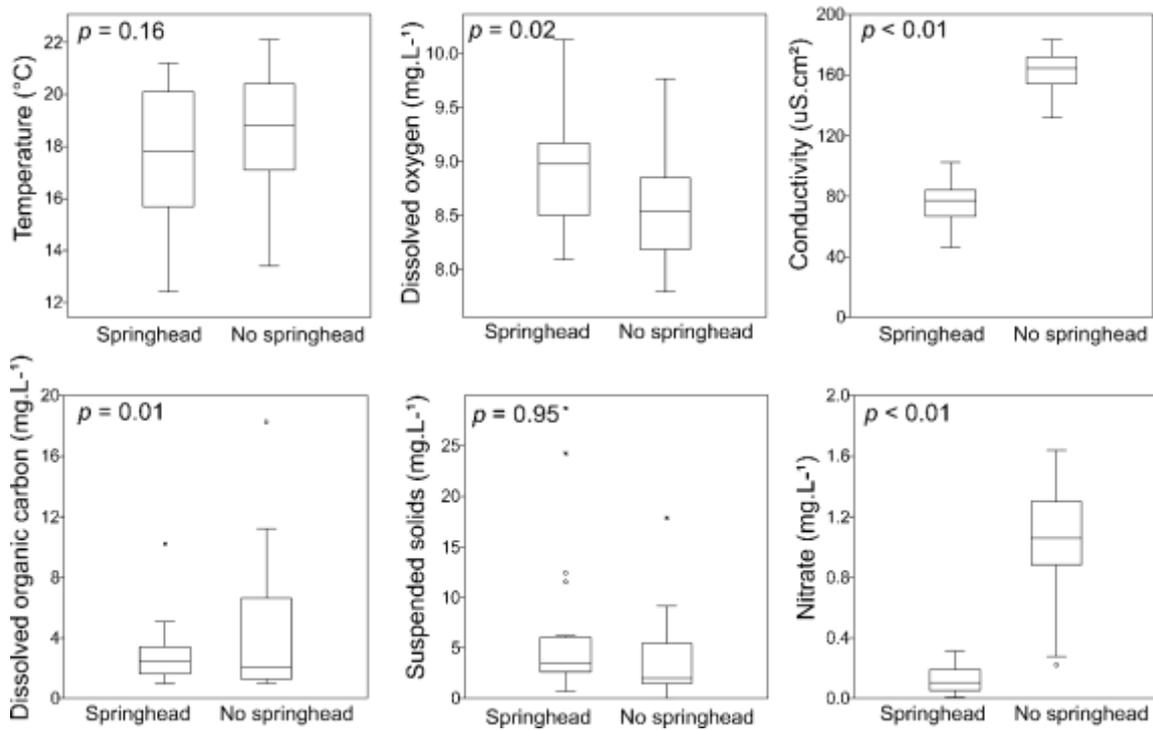


Figure 3.4 - Box plots and Kruskal-Wallis comparison of the landscape structure effects on tropical streams

Differences in the chemical characteristics of stream water between wet and dry season were not significant in the presence of preserved headwaters (Table 2.2). In the absence of preserved headwaters, concentrations of dissolved organic carbon were twice as high in the wet season than in the dry season while nitrate concentration was twice as low.

Table 3.2 - Mean values and standard errors (in brackets) of physical and chemical characteristics of water in dry and wet season, in streams with degraded and preserved springheads

	Wet	Dry	Kruskal-Wallis
<i>Preserved springhead</i>			
Temperature (°C)	18.63 (0.40)	17.16 (0.92)	NS
Dissolved oxygen (mg.L ⁻¹)	8.72 (0.09)	9.02 (0.19)	NS
Conductivity (µS.cm ⁻¹)	76.66 (4.49)	74.40 (4.06)	NS
Suspended solids (mg.L ⁻¹)	4.38 (0.83)	7.98 (2.92)	NS
Dissolved organic carbon (mg.L ⁻¹)	3.54 (0.69)	2.28 (0.36)	NS
Nitrate (mg.L ⁻¹)	0.13 (0.03)	0.12 (0.02)	NS
<i>Degraded springhead</i>			
Temperature (°C)	19.43 (0.33)	18.10 (0.80)	NS
Dissolved oxygen (mg.L ⁻¹)	8.39 (0.07)	8.68 (0.16)	NS
Conductivity (µS.cm ⁻¹)	165.13 (4.08)	160.87 (4.09)	NS
Suspended solids (mg.L ⁻¹)	3.22 (0.78)	4.23 (1.47)	NS
Dissolved organic carbon (mg.L ⁻¹)	5.61 (1.39)	2.83 (0.96)	$p < 0.05$
Nitrate (mg.L ⁻¹)	0.85 (0.10)	1.18 (0.09)	$p < 0.05$

- Integrated analysis of water quality in different conditions of land-use

The water quality index, used to summarize changes in the concentrations of dissolved oxygen, conductivity, suspended solids and nitrate through the year, show that the conversion of pasture to sugarcane crops decreased water quality by 11.7% and increased the coefficient of variation, interpreted here as a seasonal effect in 4.6%. In established sugarcane crops, catchments with riparian buffers and preserved headwaters had higher water quality index values and low coefficient of variation. The presence of preserved headwater and riparian buffers increased the water quality by 36.1% and reduced the variance by 26% (Figure 3.5).

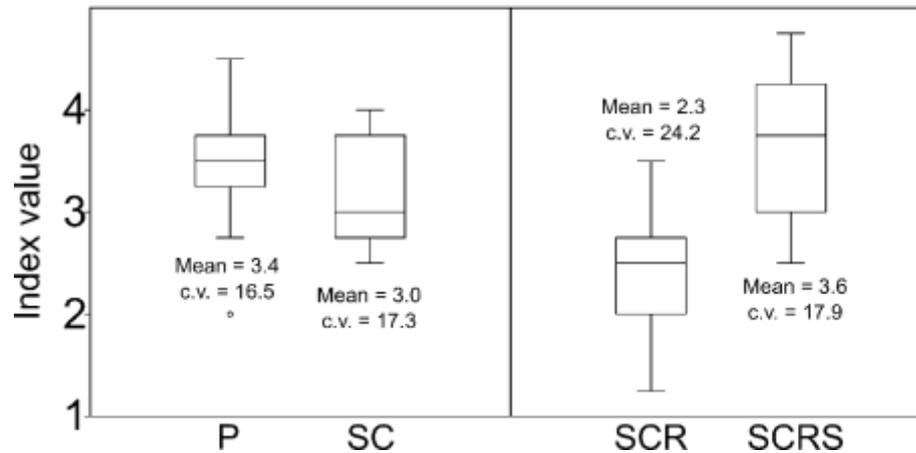


Figure 3.5 - Water quality index values in pasture (P), sugarcane (SC), sugarcane with riparian buffer (SCR) and sugarcane with riparian buffer and springhead (SCRs)

3.4 Discussion

- Conversion of pasture to sugarcane effects

Our initial hypothesis predicted that land-use conversion of pasture to sugarcane crops would reduce organic matter inputs but increase nutrient and sediment loads to streams. This prediction was based on the fact that transforming a landscape from pasture to sugarcane eliminates cattle waste and consequently, reduces organic loads to streams (SCHIESARI; CORRÊA, 2015). In addition, the intensive land management required for sugarcane production, including soil fertilization and the use of heavy machinery, increases sediment and nutrient loads to streams (MARTINELLI; FILOSO, 2008). Our results seem to support these predictions. Water temperature, conductivity, and dissolved organic carbon concentrations were lower in streams draining predominantly sugarcane crops than in streams draining pasture. However, variables that in elevated concentrations are known to have a negative impact on aquatic organisms, such as suspended solids and nitrate, were up to four times as high in sugarcane than in pasture catchments.

Deposited sediments, which is related to suspended solids and elevated nutrients represent two important agricultural stressors for biological communities in stream ecosystems (PIGGOTT et al., 2015). Suspended fine sediments have been reported as a key stressor for aquatic organisms (MATTHAEI et al., 2006; PIGGOTT et al., 2012; ELBRECHT et al., 2016) while nitrate is not only one of the main causes of eutrophication (MARTINELLI; FILOSO, 2008), but in elevated concentrations can be also toxic for

invertebrates, fishes and amphibians (CAMARGO et al., 2005). In lentic ecosystems, the conversion of pastureland to sugarcane crops does not appear to have strong consequences to water quality when riparian forests are preserved (SCHIESARI; CORRÊA, 2015). However, in lotic ecosystems, our results suggest that the effects of the conversion are significant, possibly because of the direct connections between streams and their adjacent landscape that influence the supply, transport, and fate of water and solutes to streams (ALEXANDER et al., 2007). Therefore, the consequences of converting pasture to biofuel crops in tropical watersheds can be detrimental to aquatic organisms and ecosystem functioning of tropical headwater streams.

Our second hypothesis was that, during the rainy season, the impacts of pasture on the water quality of streams would be higher than the impacts of sugarcane due to more surface and subsurface runoff and lower water infiltration in pasture watersheds. This prediction was based on the fact that landscape physical characteristics such as water infiltration capacity and runoff coefficient vary among different land uses (ALLAN, 2004; ZIMMERMANN et al., 2006). Our results do not support this hypothesis since the same number of water quality variables differed significantly between the rainy and dry seasons in pasture and sugarcane catchments.

Differences in soil management between pasture and sugarcane crops are expected to influence runoff and infiltration. Usually, pastures show compacted soils due to cattle trampling and as a consequence, infiltration reduction occurs generating higher runoff rates, carrying organic loads and nutrients from pasture to streams (DUNNE et al., 2011). Elevated nitrate concentrations in streams draining pasturelands were observed in New Zealand, the United States and South Africa (QUINN et al., 1997; HAGGARD et al., 2003; ESTERHUIZEN et al., 2012). However, higher nitrate concentrations in sugarcane crops have been observed in a study comparing streams draining pasturelands and sugarcane crops and other land uses (MORI et al., 2015). In agreement with other studies, our result showed that mean nitrate concentration in sugarcane crops was higher than in pasture. This result is associated with soil fertilization in sugarcane crops, which is transported to aquatic ecosystem due to the high mobility of nitrogen (MARTINELLI; FILOSO, 2008), increasing nitrate concentrations. In Brazil, pastures usually have low management intensity, generally with low or without fertilization. This aspect is likely to contribute to low nitrate concentrations in the studied streams. Therefore, our results have shown that water quality in pastures and sugarcane crops were affected by seasonal changes.

- Headwater condition effects

Preserved headwaters have been associated with higher water quality in landscape management studies (ALEXANDER et al., 2007; VON FUMETTI et al., 2007; CANTONATI et al., 2015). Based on this, we expected that streams draining sugarcane crops but with preserved headwaters would have superior water quality in comparison to streams with degraded headwater. Our results supported our predictions, where the stream with preserved forest in the headwater had significantly higher values of dissolved oxygen and lower values of conductivity, dissolved organic carbon and nitrate concentrations. It is important to mention that both streams had more than 30 meters of riparian buffers and that both catchments had more than 35 percent of the total area covered by forest, hence, the only pronounced difference between the catchments regarding land cover was the headwater condition.

The importance of headwater conditions for the water quality and biodiversity of entire drainage networks are well established. Headwater streams receive much less attention than other aquatic ecosystems and are insufficiently covered by protective legislation (CANTONATI et al., 2012). Therefore, restoration programs should give more attention to headwater streams, as these environments can be key to maintain the water quality of stream ecosystems as agricultural land cover expand.

Nitrate and dissolved organic carbon concentrations were higher in the wet season in the stream with degraded headwater, supporting our fourth hypothesis that ecosystem function of the riparian forest is more evident during the wet season in the humid tropics. In the wet season, tropical storms can severely alter streamflow dynamics, causing impacts in biological communities and increasing nutrient concentration (RAMIREZ; PRINGLE, 1998; JOHNSON et al., 2006). During this period, riparian forests can play an important role by intercepting and retaining nutrient and particle transported from the upland areas (ZIEGLER et al., 2006; FERREIRA et al., 2012). However, our results show that if the headwater area is degraded, a riparian forest is not enough to guarantee good water quality in streamflow. This suggests that riparian buffers restoration and conservation should be accompanied by the restoration and conservation of headwaters to prevent stream channel sedimentation and other negative impacts to water quality.

- Management implications

We have known for quite a while that extensive pasture without conservation practices has a negative effect on aquatic ecosystems (NEILL et al., 2001; FOLEY et al., 2005; MATTHAEI et al., 2006), as confirmed by our water quality data. However, the conversion of pastureland to intensive agriculture for sugarcane production can reduce the water quality of streamflow even further, especially with regard to nutrient excess and erosion (FILOSO et al., 2015). Riparian forests can attenuate the negative effects of sugarcane cultivation (FILOSO et al., 2015). Nevertheless, while riparian buffers improve the overall ecological conditions of stream ecosystems (AARONS; GOURLEY, 2013; DE SOUZA et al., 2013; BURRELL et al., 2014), they are not able to mitigate all of the negative effects of intensive agriculture (FERREIRA et al., 2012), especially if the riparian buffer is not in a good structure (FERRAZ et al., 2014). In this study, we have also shown that riparian forests are less effective at preventing water quality degradation from the conversion of pastureland to sugarcane if the headwaters are degraded.

Despite the evidence that forest restoration can be the best option for improving water quality, recent studies have shown that this action can be very costly to farmers and managers, hindering the implementation at large scale (MANTYKA-PRINGLE et al., 2016). Aiming to reduce the implementation costs of forest restoration, governmental incentives and best agricultural practices can be helpful to improve water quality in agricultural lands. A recent review has shown that the improvements that still need to address in sugarcane crops in Brazil are soil erosion, prevention of soil degradation, water resources protection from pesticides and other toxic chemicals and the avoidance of sugarcane crops expansion in threatened biomes (FILOSO et al., 2015). These issues are likely to be addressed by riparian forest restoration, development of best agricultural practices and implementation of clear rules protecting threatened biomes within tropical zones.

3.5 Conclusion

Our results have shown that conversion of pasture to bioenergy crops has a negative impact on the water quality of tropical small-order streams. Such changes include the increase of nitrate concentrations, water temperature, and suspended solids. Well-preserved or restored headwater areas accompanied by robust riparian buffers can help prevent water quality

degradation, especially in the rainy season. Other best agricultural practices techniques should help as well.

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4 INVESTIGATING AGRICULTURAL AND DISTURBANCE INFLUENCES ON BENTHIC BIOFILM DYNAMICS IN TROPICAL HEADWATER STREAMS

Abstract

Agricultural intensification in the tropics has broadly changed the ecosystems. The main effects of agricultural activities are mainly linked with changes in nutrient and hydrological dynamics, which directly affects headwater streams. To detect these anthropogenic changes in streams, benthic biofilm community has been successfully used as a bioindicator of stream quality in temperate regions due to its rapid response to nutrient enrichment and flow dynamics. However, little is known about the responses of benthic biofilms in tropical streams, considering the intense rainfall patterns that frequently occurs during the tropical wet season. Therefore, we have investigated benthic biofilm community responses in relation to nutrient, flow and temperature dynamics in tropical streams. For this, field sampling was carried out, analyzing the benthic biofilm biomass, chlorophyll a content and functional structure of benthic diatoms in tropical headwater streams and investigated its relationship with water temperature, nitrate and flow dynamics. Our results demonstrated that benthic biofilm biomass is mainly controlled by flow dynamics. The functional structure of benthic diatoms is primarily controlled by water temperature and flow dynamics. Oppositely with studies from temperate streams, nutrient does not appear to have strong influences on benthic diatom community dynamics. These results show that in a climate change scenario, benthic biofilm will be strongly affected and therefore, shifts in the functioning of tropical streams are expected.

Keywords: Functional structure; Benthic diatoms; Benthic biofilm; Agriculture; Disturbance

4.1 Introduction

Tropical areas are constantly under anthropogenic pressures. Holding the remaining potentially cultivable areas for agriculture and increasing in urban areas and megacities, environmental problems in the tropics are continuously growing (GUPTA, 2002; SMITH et al., 2010). The accelerated development is degrading the freshwater habitats, which contain about 9.5% of the described animal species in one of the most biodiverse ecosystems on Earth (DUDGEON et al., 2006; WANTZEN et al., 2006; BOULTON et al., 2008). Tropical streams differ from temperate streams especially in terms of climatic conditions, geomorphology, landscape evolution and geological history (WANTZEN et al., 2006) therefore, it is likely that processes and functioning of tropical streams may differ from temperate streams due to differences in hydrological cycle and water temperature (DODDS et al., 2015). These characteristics make tropical streams an important subject of study, considering the lack of information about these ecosystems and the rapid development in tropical regions.

Agricultural land use and flow regimes are the main factors affecting resource supply, organic matter, energy cycling and physicochemical habitat characteristics in streams

(LANGE et al., 2015). The conversion of forests to agricultural landscapes shifts the surface runoff and stream discharge, increasing erosion, sediment loads and leaches nutrients and agricultural chemicals to streams (FOLEY et al., 2005). These problems are especially important in the tropics, where agricultural activities are a key factor for human development (DEFRIES; ROSENZWEIG, 2010) and are subjected to earlier climate change effects (MORA et al., 2013). Agricultural effects on tropical streams have been investigated and changes in fish (CASATTI et al., 2015; TERESA et al., 2015), macroinvertebrate (BENSTEAD; DOUGLAS; PRINGLE, 2003; LORION; KENNEDY, 2009; SIQUEIRA et al., 2015) and microbial (BOECHAT et al., 2011) communities have been observed. Consequences in stream processes also have been observed such as leaf litter decomposition (SILYA-JUNIOR et al., 2014) and stream metabolism (GUECKER; BOECHAT; GIANI, 2009).

To evaluate the anthropogenic effects on streams, benthic biofilm has been used in temperate systems in biomonitoring programs. In these systems, predictable changes can be observed following shifts in nutrient availability and flow dynamics (PASSY, 2007; BERTHON et al., 2011; LANGE et al., 2015), allowing the conception of a trait-based framework for stream algal communities to detect effects of land-use and climate change (LANGE et al., 2015). Indices and frameworks developed for temperate systems are often applied to tropical streams, due to lack of information on ecological preferences and tolerances of benthic diatoms (BERE, 2015), and consequences for applied issues are likely. For example, due to unpredictable and dynamic flow patterns induced by high-intensity rainfall and complex nutrient dynamics due to intense agricultural activities, different responses in benthic biofilm biomass and benthic algae community are expected in tropical streams.

An alternative to deal with the lack of information for benthic diatoms in tropical streams is the trait-based approach. This method is based on species attributes that govern its ability to deal with environmental problems and opportunities (VERBERK et al., 2013). Trait-based approaches can refine our mechanistic understanding of biological communities in a taxon-independent manner, allowing the comparison across ecosystems and transforming descriptive into predictive studies in community ecology (VERBERK et al., 2013; SCHMERA et al., 2015). For benthic diatoms, traits are classified according to the resistance to physical disturbance, nutrient enrichment, and life forms, in genus-level taxonomic resolution (PASSY, 2007; BERTHON et al., 2011; SCHNECK; MELO, 2012; LANGE et al., 2015). The use of coarse taxonomic resolution in genus-level has been applied in tropical

river floodplains, with similar responses to environmental variation as species-level (RIBAS; PADIAL, 2015). Therefore, we considered in this study that the same result found by Ribas and Padial (2015) could be applied to tropical streams.

In this paper, we investigate how benthic biofilm biomass and functional structure of benthic diatom change due to temperature, nutrient, flow disturbances and seasonal characteristics. We expect that benthic biofilm will be controlled mainly by physical characteristics and not by resource availability, due to the strong connection of tropical headwater streams with landscape and climate characteristics. Following these assumptions, we hypothesized that benthic biofilm biomass and algae biomass would be lower in wet season due to flow disturbances in tropical headwater streams. Oppositely, we expect that functional characteristics of benthic diatom community will be benefitted in the wet season, due to the structurally complex habitat provided by flow dynamics and higher temperatures and therefore, controlled mainly by physical characteristics and not by resource availability. We also provide first insights of the influence of landscape characteristics in benthic diatom traits in tropical headwater streams. These results would indicate if benthic diatom traits were a good approach for biomonitoring tropical headwater streams.

4.2 Methods

- Study area

Samples were collected in the Corumbataí river basin, located in São Paulo state, southeast Brazil (centered on 47°40'W, 22° 40' S), a region of 1700 km² which has historically experienced intensive land use change (FERRAZ et al., 2014). Sugarcane crops occupy approximately 32% of the cultivable area and is the main economic activity in the basin. Other main land uses are pasture (~ 25%) and forestry (~ 20%). The regional climate is classified as Cwa in Köppen classification, with dry winter and wet summer (CETRA; PETRERE, 2006). The average temperature in the winter is 17°C and exceeds 22°C in the summer (FERREIRA et al., 2012).

Three catchments draining first order streams with similar altitude, orientation and drainage were selected in the Corumbataí river basin, with area ranging from 0.43 km² to 1.40 km². The catchment “A” has 92.5% of sugarcane crops and 7.4% of forests. The catchment “B” has 76.7% of sugarcane crops and 23.3% of forest, and the catchment “C” has 54.3% of sugarcane crops and 45.7% of forest. Mean stream elevation ranged from 717 to 811 m a.s.l.

The canopy coverage in the middle of the streams was above 80% during the entire period of study.

- Field and laboratory methods

For stream water characterization at each field sampling, physical and chemical variables of water were measured. Streamflow (Q) was measured using the float method (EPA, 2014). Water temperature was recorded with an optical probe (ProODO, YSI, Ohio, USA). Spectrophotometry measured nitrate concentrations after reduction with cadmium catalyst and reaction with sulfanilamide and N-naphthyl (GINE et al., 1980). The wet and dry season were classified separating the six driest months and the six wettest months through streamflow values.

Benthic biofilm biomass, algae biomass, and benthic diatom samples were collected monthly from three random granite tiles in a set of 14 granite tiles disposed along the streambed for one year (March 2013 to February 2014). Granite tiles surface (264.5 cm²) were scrubbed with a toothbrush with jets of ultrapure water and fixed with 4% formalin. Benthic biofilm biomass was measured through ash-free dry weight and algae biomass was measured through chlorophyll-a by methods described in APHA (2005). Benthic diatom community was assessed using Hydrogen Peroxide and Potassium dichromate (HAUER; LAMBERTI, 2007). This method was necessary due to the high amount of exopolysaccharide matrix and organic matter in the samples. The identification was performed at 1000x magnification using permanent slides prepared with Naphrax®. The counts were performed at 400x magnification in an inverted microscope following transects along the chamber. At least 300 units were counted in each sample or until the stabilization of collector's curve. Counts were converted to the density of cells per surface area of rock (number of cells cm²) using the methods described in Hauer and Lamberti (2007). For taxonomic analysis, samples were identified to genus or species level according to the specific bibliography for tropical and temperate regions.

Benthic diatom functional traits were classified according to the resistance to physical disturbance and nutrient enrichment (low-profile, high-profile and motile) described in Passy (2007) and life forms (mobile, colonial, tube-forming, stalked and pioneer) described in Berthon et al. (2011). After the assignment of functional traits, we calculated the following functional indices: (i) Functional richness - represents the amount of functional space filled by the benthic diatom community (VILLEGER et al., 2008). (ii) Functional evenness – measures

the regularity of spacing between benthic diatom species in the trait space and the distribution evenness of benthic diatom abundance (VILLEGER et al., 2008). (iii) Functional divergence – relates how benthic diatoms abundances are distributing within the volume of functional trait space occupied by species (VILLEGER et al., 2008) and (iv) Functional dispersion – average distance of individual species to the centroid of all species in the benthic diatom community trait space, accounting the relative abundances (LALIBERTE; LEGENDRE, 2010). All the functional indices were calculated in the software FDiversity (CASANOVES et al., 2011).

- Statistical analysis

Seasonal effects on physical and chemical characteristics of water were analyzed by Kruskal-Wallis test, comparing dry and wet season. To analyze the controlling factors of benthic biofilm biomass and benthic diatom functional traits, each response variable was submitted to a linear regression, using water temperature and log transformed nitrate as covariates in the regression, with some cases of outlier's elimination for best model fit. To analyze flow effects on biomass and functional indices, streamflow values was divided into two categories: high flow (flow above $0.02 \text{ m}^3 \cdot \text{s}^{-1}$) and low flow (flow below $0.02 \text{ m}^3 \cdot \text{s}^{-1}$), and then Kruskal-Wallis test was applied. To provide the insights of the influence of landscapes characteristics on benthic diatom functional traits, we calculated the functional indices values for each stream and discussed based on the landscape characteristics (forest and sugarcane crops proportions in each catchment). Statistical analyses were performed in R (R Core Team, 2013), Fdiversity (CASANOVES et al., 2011) and PAST (HAMMER et al., 2001) software.

Results

- Physicochemical characteristics of stream water between dry and wet seasons

Small differences were observed in the analyzed physical and chemical characteristics of stream water between dry and wet season (Table 4.1). Only streamflow patterns showed significant differences between seasons, with mean values three times higher in the wet season. Temperature and nitrate did not differ between seasons, however, showed higher mean values on wet season.

Table 4.1 - Mean, standard deviation (in brackets) and Kruskal-Wallis comparisons of physicochemical variables measured at each hydrological period

	Dry season	Wet season	Kruskal-Wallis
Temperature (°C)	18.1 (1.97)	19.8 (1.97)	NS
Streamflow (m ³ .s ⁻¹)	0.01 (0.00)	0.03 (0.02)	$p < 0.01$
Nitrate (mg.L ⁻¹)	0.67 (0.23)	0.80 (0.37)	NS

- Controlling factors of benthic biofilm and benthic algae biomass in tropical headwater streams

Benthic biofilm biomass and benthic algae biomass were influenced mainly by flow dynamics in tropical headwater streams (Figure 4.1). Higher values of ash-free dry weight and chlorophyll-*a* were found in dry season, characterized by low values of streamflow. Algae and benthic biomass did not show a linear relationship with water temperature and nitrate.

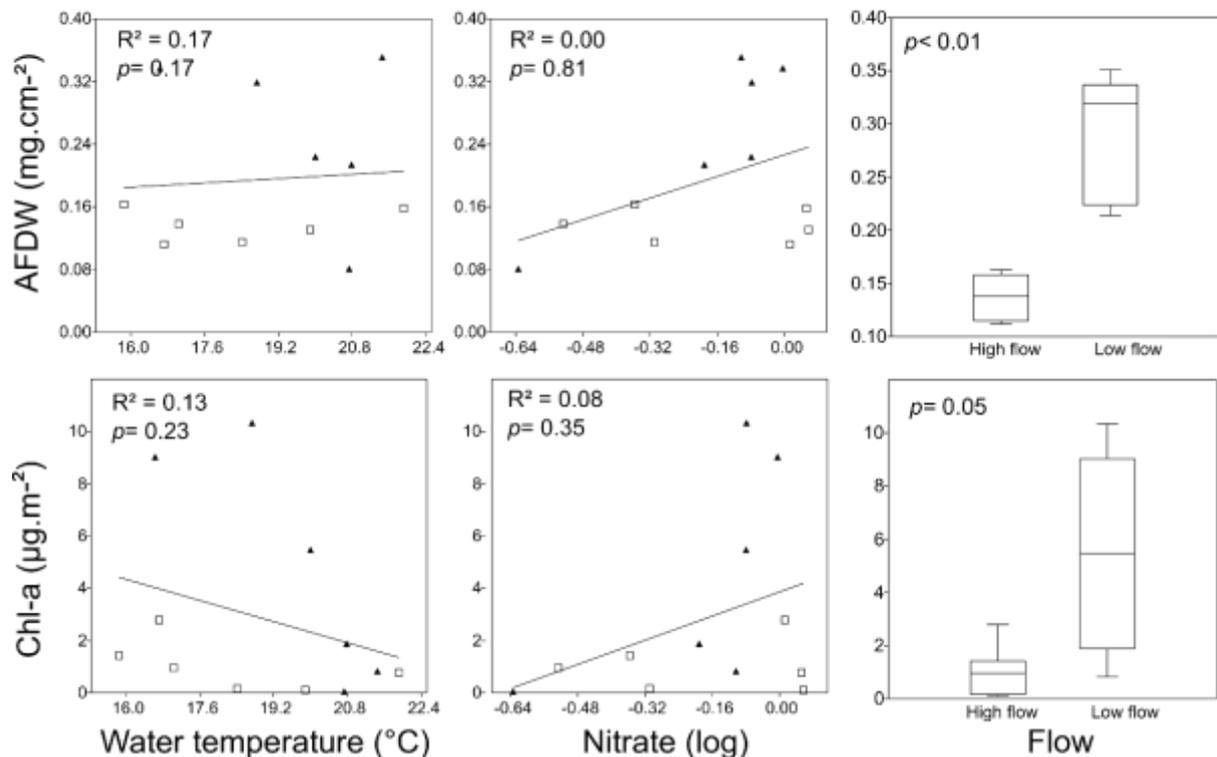


Figure 4.1 - Linear regressions of biofilm and algae biomass with water temperature and log transformed nitrate concentrations (Log NO₃) and boxplots of biofilm and algae biomass on low (> 0.02 m³.s⁻¹) and high (< 0.02 m³.s⁻¹) streamflow. Triangle symbols are samples from dry season; square symbols are samples from wet season

- Controlling factors of benthic diatom functional diversity in tropical headwater streams

Physical characteristics of stream water were the main controlling factor of benthic diatom functional indices. High water temperature promoted higher values of functional richness and functional dispersion. Functional richness was also influenced by flow, with higher values in high flow periods. Functional evenness and functional diversity were not affected by physical and chemical characteristics of water in the studied streams (Figure 4.2).

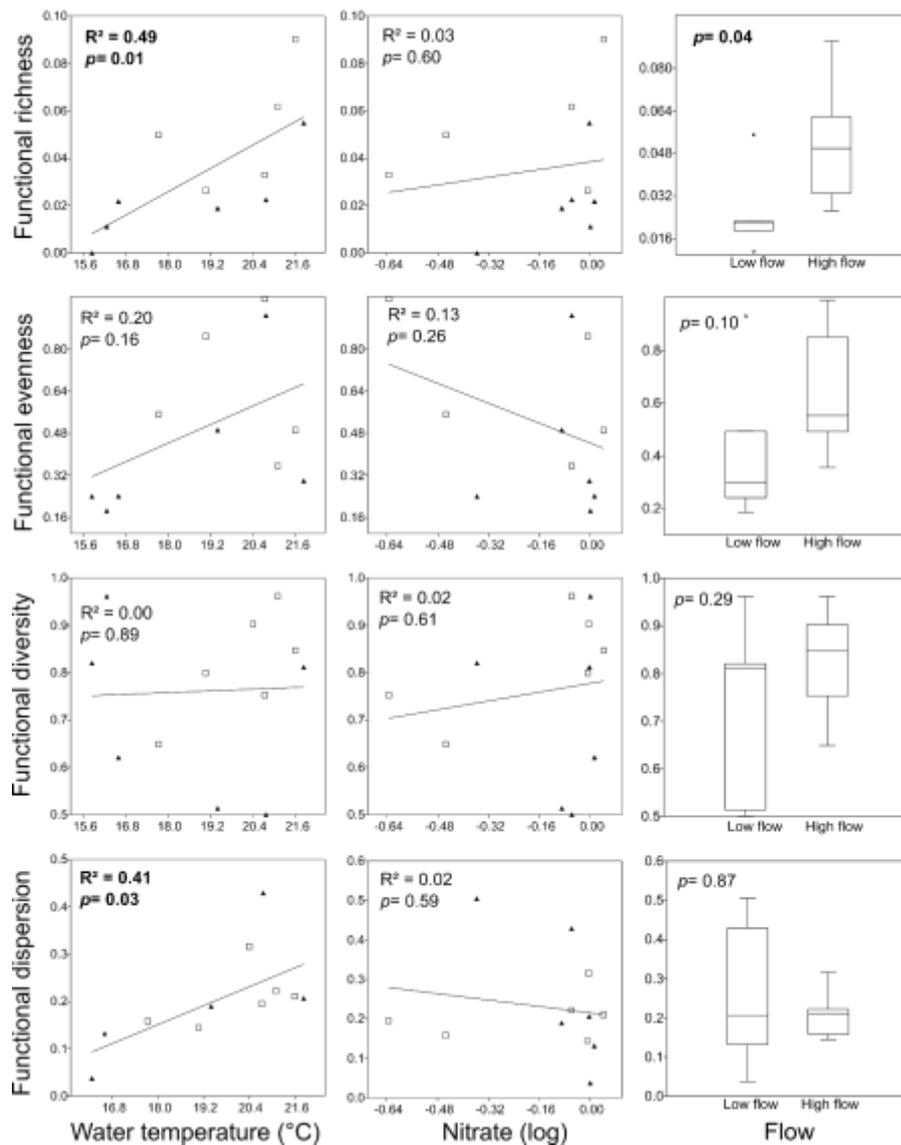


Figure 4.2 - Linear regressions of functional diversity indices with water temperature and log transformed nitrate concentrations (Log NO_3^-) and boxplots of functional diversity indices on low ($> 0.02 \text{ m}^3 \cdot \text{s}^{-1}$) and high ($< 0.02 \text{ m}^3 \cdot \text{s}^{-1}$) streamflow. FRic = functional richness, FEve = Functional evenness, FDiv = Functional diversity and FDis = Functional dispersion. Triangle symbols are samples from dry season; square symbols are samples from wet season

- Influences of landscape characteristics in benthic diatom functional characteristics in tropical headwater streams

Landscape characteristics possibly influenced functional characteristics of benthic diatoms. Benthic diatom functional traits showed higher values of functional diversity and functional dispersion in the stream with low coverage of riparian vegetation and higher percentages of agricultural coverage. The stream with the highest coverage of forest in the catchment showed the lowest values of functional richness, evenness, and dispersion (Table 4.2).

Table 4.2 - Values of functional richness (FRic), functional evenness (FEve), functional diversity (FDiv) and functional dispersion (FDis) in the sampled streams in the catchments A, B and C

Catchment	Agriculture cover	Forest cover	FRic	FEve	FDiv	FDis
A	92.5%	7.4%	9.2E-09	0.495	0.985	0.167
B	76.7%	23.3%	5.5E-10	0.507	0.857	0.119
C	54.3%	45.7%	4.5E-10	0.322	0.959	0.041

4.3 Discussion

Benthic biofilm and algae biomass was mainly influenced by flow dynamics, which is connected with seasonal patterns in tropical headwater streams. This result is attributed to the strong hydrological connection between landscape characteristics in headwater streams (ALEXANDER et al., 2007; FREEMAN et al., 2007), causing natural or anthropogenic shifts in streamflow (POFF et al., 1997; LYTLE; POFF, 2004). In the tropics, the hydrological processes have greater energy inputs, faster rates of change and faster rates of human-induced changes (WOHL et al., 2012), and therefore, represent the main factor for the development of benthic biofilm and algae biomass in tropical headwater streams. Benthic biofilm biomass is composed of diverse organisms, including algae, bacteria, and fungi, therefore, is an important autochthonous resource for in-stream consumers (SABATER et al., 2016). Previous studies in tropical and subtropical streams have demonstrated the effects of flow on benthic biofilm biomass (KOHLENER et al., 2012; SCHNECK; MELO, 2012), supporting our predictions. Streamflow is one of the most affected characteristic under a land-use and climate change scenario (ALLAN, 2004; BRUIJNZEEL, 2004), representing a concern for tropical regions, where agricultural development is an important social and economic activity

(DEFRIES; ROSENZWEIG, 2010) and is affected due to climate change (O'GORMAN, 2012).

Benthic diatom functional characteristics were connected with seasonal/climate characteristics in tropical headwater streams. Analyzing the controlling factors of benthic diatom functional diversity, we found that functional richness and functional dispersion showed a positive relationship with water temperature, which is known to have exponential effects in nearly all rates of biological activity (BROWN et al., 2004) and has been observed in several organisms, including diatom distribution (ALLEN et al., 2002; PAJUNEN et al., 2015). Streamflow also demonstrated effects on functional richness, with higher values of functional indices on high flow periods. In this period, instable flow washes the accumulated sediment on the streambed, exposing rocks and other types of substrata, creating a high degree of niche differentiation and therefore, low competition (MASON et al., 2005; CIBILS et al., 2015), favoring the establishment of species with different functions. The same result has been observed in subtropical streams (SCHNECK; MELO, 2012; CIBILS et al., 2015), reinforcing our expectations that benthic diatom traits are mainly controlled by climatic characteristics in the tropical headwater streams. One of the issues that emerges from these findings is the observed relationship between converted lands and higher water temperatures and higher streamflow in different parts of the globe (BROWN et al., 2005; BROADMEADOW et al., 2011; SIMMONS et al., 2015), which demonstrated effects on functional dynamics of benthic diatom traits. It can thus be suggested that deforestation is changing the community dynamics of stream benthic diatom, with unknown consequences for tropical stream functioning.

The effects of deforestation on benthic diatom community dynamics can be inferred by the functional diversity values per site, where the stream with less forest coverage and higher agriculture proportions in the catchment showed the highest values of functional diversity and functional dispersion. Streams located in non-forest catchments displays instable flow and high nutrient inputs (WU et al., 2007; CARLSON et al., 2014), turning these ecosystems heterogeneous and as a consequence, opening space for the development of species with different niche occupying and exploration (SIMPSON, 1949). Oppositely, forested streams exhibit a higher number of environmental filters for stream algae development, e. g. light deprivation due to canopy cover and low nutrients due to riparian forest buffers, which are the main resources for algal development (STEVENSON et al., 1996). These results are in accord with recent studies that does not support the argument that land use always leads to loss of species and functional traits (MAYFIELD et al., 2010).

Studies on phytoplankton dynamics in shallow lakes also demonstrated higher functional properties on transient non-stable environmental state (WEITHOFF et al., 2001). Therefore, the idea that high functional diversity maintains stability in ecosystem functioning are dependent on ecosystem disturbance history, species pool and environmental filter that are different between communities (MAYFIELD et al., 2010; BISWAS; MALLIK, 2011), and therefore should be interpreted with caution on conservation biology and biomonitoring programs of tropical headwater streams.

4.4 Conclusions

We demonstrated here that disturbances play an important role in benthic diatom community in tropical headwater streams, and these disturbances are closely linked with climate characteristics, such as temperature and streamflow. These results are in agreement with the Stream Biome Gradient Concept, which hypothesizes that stream biological features changes predictably along climate characteristics (DODDS et al., 2015). Interestingly and supporting our hypothesis, nitrate concentrations did not demonstrate effects on diatom functional diversity, as commonly observed in temperate streams (BIGGS; CLOSE, 1989; PASSY, 2007; LANGE et al., 2015). Because of highly dynamic environmental variability in headwater streams, linked with the high-intensity storms that are commonly observed in tropical regions, climate characteristics modulate benthic diatom community in tropical headwater streams. In a climate change scenario, with the forecast of high intensity rainfall, prolonged drought and higher temperatures in the tropics (IPCC, 2014), shifts in benthic diatom communities are expected, with unknown consequences for the entire stream ecosystem, once diatom community plays a key role in in-stream metabolism, primary production, and resource availability for aquatic organisms.

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5 SYNTHESIS AND RECOMMENDATIONS

Tropical streams are facing multiple anthropogenic pressures that are shifting water quality and biological communities. These changes, linked with climate change effects can result in deleterious effects for stream biological communities in one of the most biodiverse ecosystems on Earth, causing biodiversity loss. In addition, these negative impacts will also affect the agriculture development, which is one of the most important activities in the tropics, which generates labor and creates opportunities for human development.

Our results have shown that we have important challenges to be addressed that are essential to help alleviate the anthropogenic pressures in tropical headwater streams. For example, little is known about the climate change effects in tropical streams. Few studies have analyzed the effects of elevated temperature, floods and droughts in the structure and functioning in these ecosystems, and fewer studies have analyzed the interactions between these effects in a multiple stressor scenarios. Another knowledge gap is that fewer studies have analyzed the possible mitigation policies and strategies to ensure the sustainability of tropical freshwater resources facing land-use and climate change.

In relation to water quality, we found that the conversion of low-intensity pastures to high-intensity bioenergy crops are reducing the water quality in tropical headwater streams. The main issue from this conversion is the high nitrate inputs in the streams, which is one of the main causes of eutrophication, that contributes to biodiversity loss. However, for benthic biofilm communities, it seems that the climate effects are more important in regulating the community dynamics. Due to the high-intensity rainfall patterns in the tropics and the close connections of headwater streams to adjacent lands, benthic biofilms are washed during the wet seasons and as a consequence, do not respond according to nitrate concentrations, as observed in temperate streams. Therefore, it is expected that benthic biofilm communities will be strongly affected by climate change.

Based on these conclusions, we recommend to managers and practitioners to focus on riparian forest restoration programs to avoid the entering of sediments and nutrients to streams, which will reduce the water quantity through sedimentation of small water bodies and will reduce water quality through eutrophication. Riparian forests also can help to alleviate the climate change effects through reductions in water temperature. It is crucial to establish forests in the springhead zone, otherwise, sediments and nutrients will continue to enter in streams. Although riparian forests be one of the best options to protect headwater streams, these forests are not able to protect these ecosystems entirely. Therefore, it is indispensable

that best agricultural practices are applied (i.e. reduction of roads inside the fields, reduction of vinasse application, controlled use of fertilizers and pesticides) to avoid the negative impacts on the agriculture, especially during the wet season.

Another important recommendation based on the results of this study is to not reduce the length of riparian buffers. As we have shown and repeat here, the riparian vegetation has an important ecosystem function in retaining the sediment and nutrient loads to streams. Reducing the riparian vegetation buffers will increase the nutrient exports to streams, increase the stream sedimentation and therefore, reduce water quality and availability for the agricultural activities and also for the population.

APPENDIX

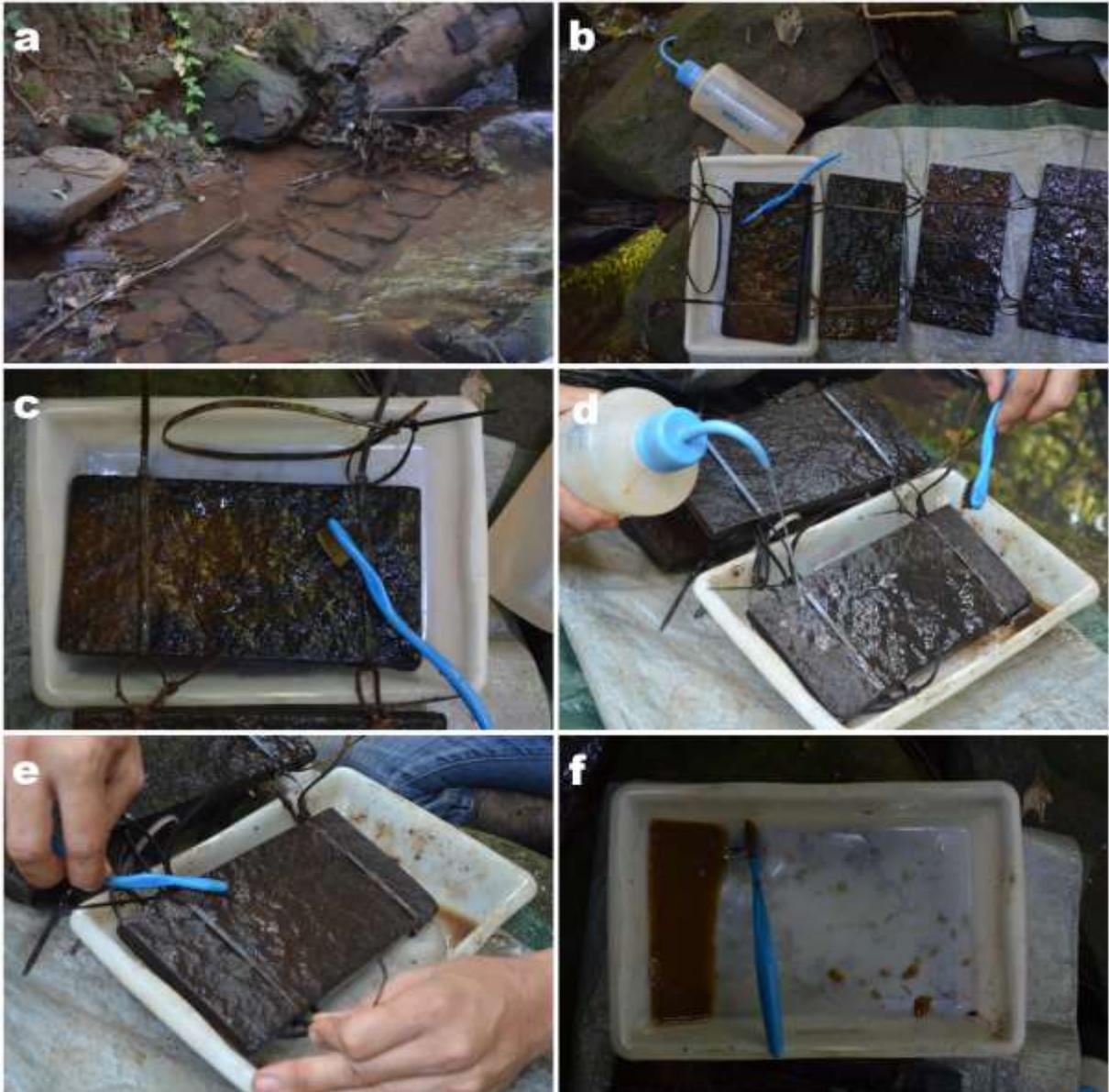
Appendix A

Pictures of the sampling stations. (a and b)- sampling station A. (c and d) – sampling station B. (e and f) – sampling station C in the chapter 3 or SCRS in the chapter 2. (g and h) – sampling station C2 or SCR in the chapter 2



Appendix B

Methodology used to collect stream biofilm community in the sampled streams. (a) tiles along streambed; (b) tiles after colonization period (4 weeks); (c) removing the biofilm from the tiles; (d) washing the biofilm from the tiles with ultrapure water; (e) re-washing the biofilm from the tiles with ultrapure water and (f) removed biofilm for laboratory analysis



Appendix C

Identified benthic diatoms, used to extract the traits in the chapter 4

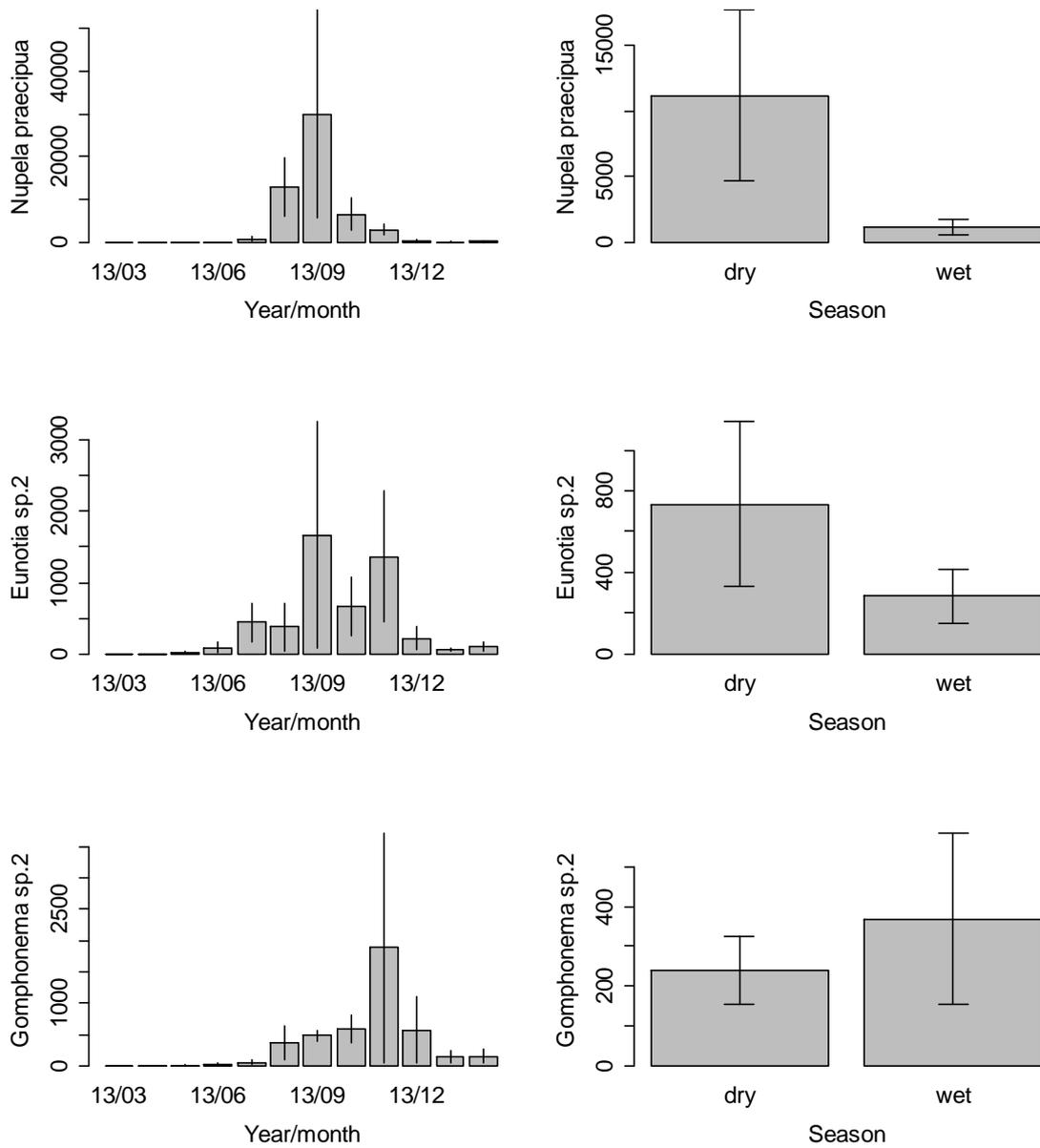
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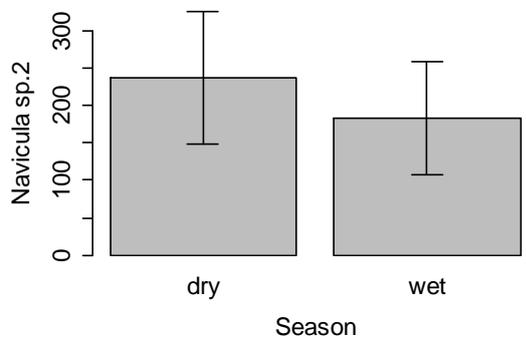
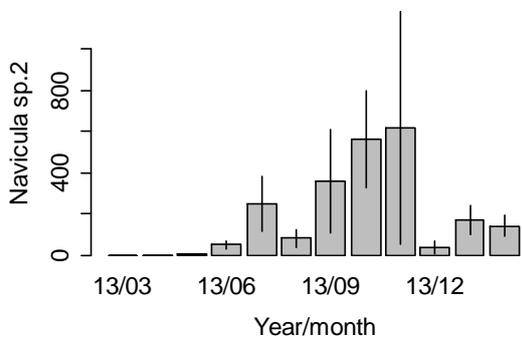
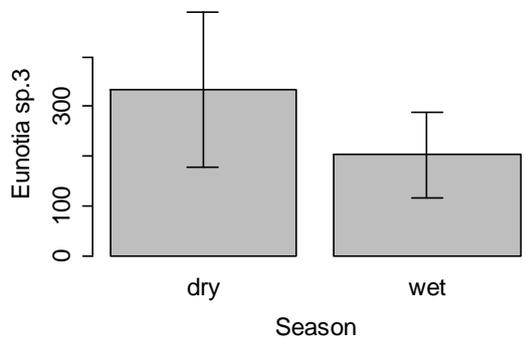
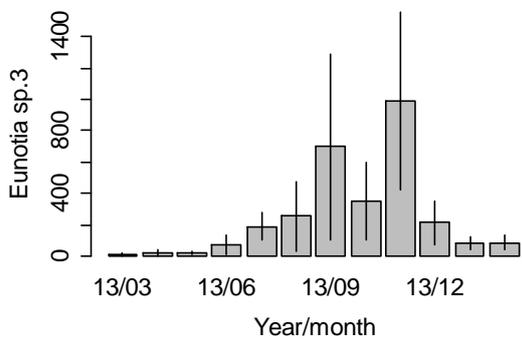
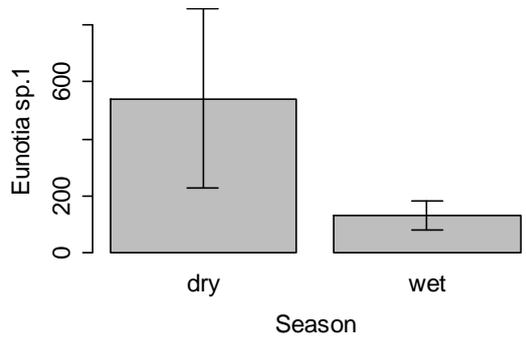
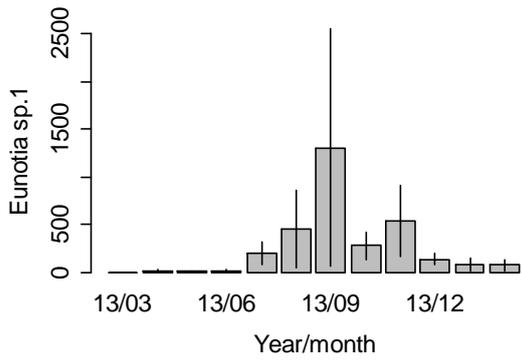
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<i>Aulacoseira</i> sp.	Thwaites, 1848
<i>Cocconeis</i> sp.	Ehrenberg, 1836
<i>Diploneis</i> sp.1	Ehrenberg ex Cleve, 1894
<i>Diploneis</i> sp.2	Ehrenberg ex Cleve, 1894
<i>Encyonema</i> sp.	Kützing, 1834
<i>Eunotia bilunaris</i>	(Ehrenberg) Schaarschmidt, 1880
<i>Eunotia bilunaris</i>	(Ehrenberg) Schaarschmidt, 1880
<i>Eunotia bilunaris</i>	(Ehrenberg) Schaarschmidt, 1880
<i>Eunotia</i> sp.	Ehrenberg, 1837
<i>Eunotia</i> sp.1	Ehrenberg, 1837
<i>Eunotia</i> sp.2	Ehrenberg, 1837
<i>Eunotia</i> sp.3	Ehrenberg, 1837
<i>Eunotia</i> sp.4	Ehrenberg, 1837
<i>Eunotia</i> sp.5	Ehrenberg, 1837
<i>Eunotia</i> sp.6	Ehrenberg, 1837
<i>Eunotia</i> sp.7	Ehrenberg, 1837
<i>Fragilaria</i> sp.1	Lyngbye, 1819
<i>Fragilaria</i> sp.2	Lyngbye, 1819
<i>Fragilaria</i> sp.3	Lyngbye, 1819
<i>Fragilaria</i> sp.4	Lyngbye, 1819
<i>Frustulia rhomboides</i> var. <i>saxonica</i>	(Rabenhorst) De Toni, 1891
<i>Frustulia</i> sp.	Rabenhorst, 1853
<i>Gomphonema parvulum</i>	(Kützing) Kützing 1849
<i>Gomphonema parvulum</i> var. <i>lagenula</i>	(Kützing) Otto Müller 1905
<i>Gomphonema</i> sp.1	Ehrenberg, 1832
<i>Gomphonema</i> sp.2	Ehrenberg, 1832
<i>Navicula cryptocephala</i>	Kützing, 1844
<i>Navicula cryptotenella</i>	Lange-Bertalot, 1985
<i>Navicula</i> sp. 3	Bory de Saint-Vincent, 1822

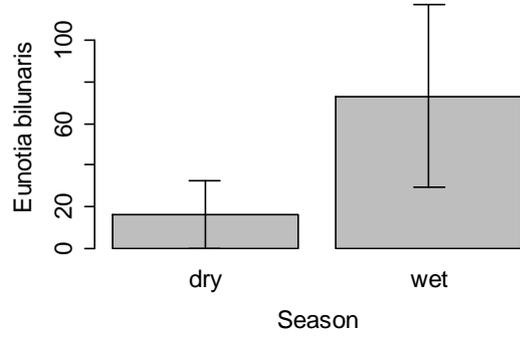
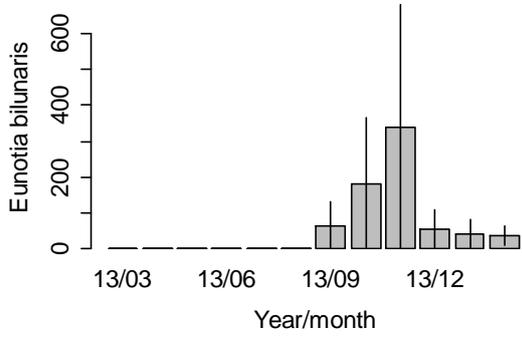
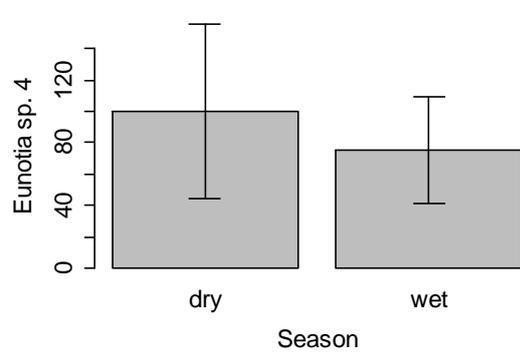
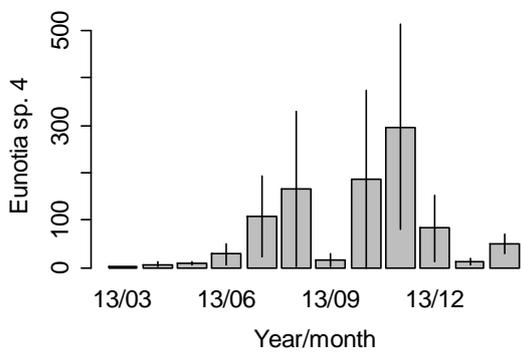
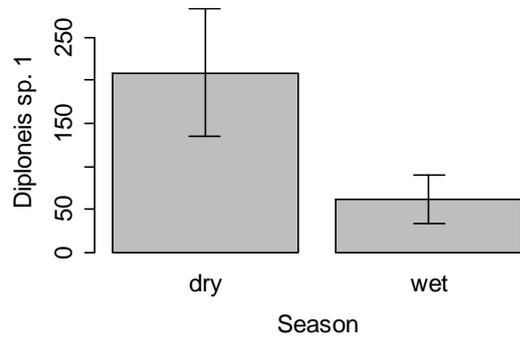
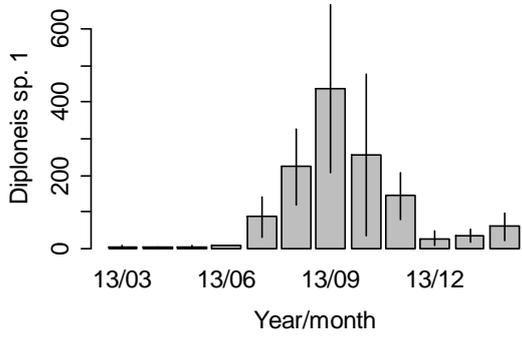
Genus	Authority
<i>Navicula</i> sp. 4	Bory de Saint-Vincent, 1822
<i>Navicula</i> sp. 5	Bory de Saint-Vincent, 1822
<i>Navicula</i> sp. 6	Bory de Saint-Vincent, 1822
<i>Navicula</i> sp.1	Bory de Saint-Vincent, 1822
<i>Navicula</i> sp.2	Bory de Saint-Vincent, 1822
<i>Neidium subdubium</i>	D.Metzeltin & K.Krammer
<i>Nitzschia</i> cf. <i>parvula</i>	Lewis, 1862
<i>Nitzschia clausii</i>	Hantzsch, 1860
<i>Nitzschia</i> sp.1	Hassall, 1845
<i>Nitzschia</i> sp.2	Hassall, 1845 Hassall, 1845
<i>Nitzschia</i> sp.3	
<i>Nitzschia</i> sp.4	Hassall, 1845
<i>Nitzschia</i> sp.5	Hassall, 1845
<i>Nitzschia</i> sp.6	Hassall, 1845
<i>Nitzschia</i> sp.7	Hassall, 1845
<i>Nitzschia</i> sp.8	Hassall, 1845
<i>Nupela praecipua</i>	(E.Reichardt) E.Reichardt, 1988
<i>Pinnularia</i> sp.1	Ehrenberg, 1843
<i>Pinnularia</i> sp.2	Ehrenberg, 1843
<i>Pinnularia</i> sp.3	Ehrenberg, 1843
<i>Pinnularia</i> sp.4	Ehrenberg, 1843
<i>Pinnularia</i> sp.5	Ehrenberg, 1843
<i>Pinnularia</i> sp.6	Ehrenberg, 1843
<i>Rhopalodia operculata</i>	(C.Agardh) Håkanasson, 1979
<i>Staurosira</i> sp.	Ehrenberg, 1843

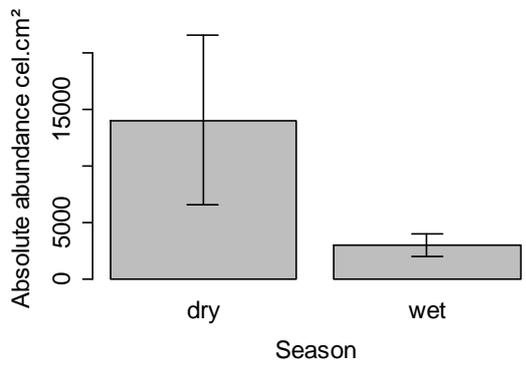
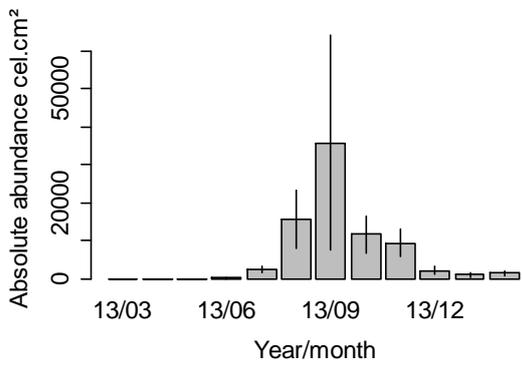
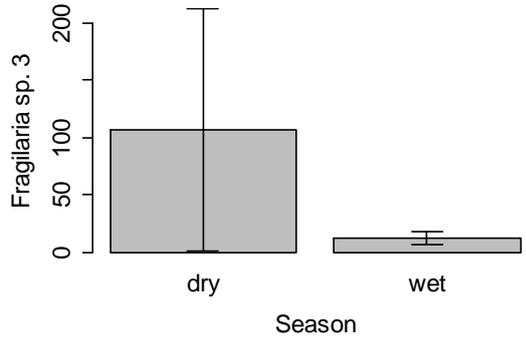
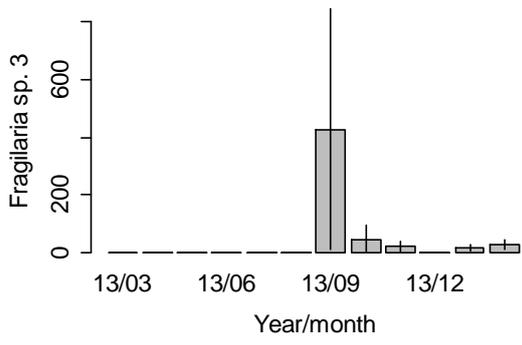
Appendix D

Species abundance per month and per season (in order, from the most abundant to the less abundant until 1% of total abundance)

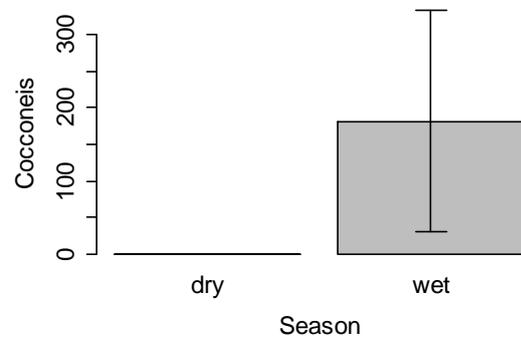
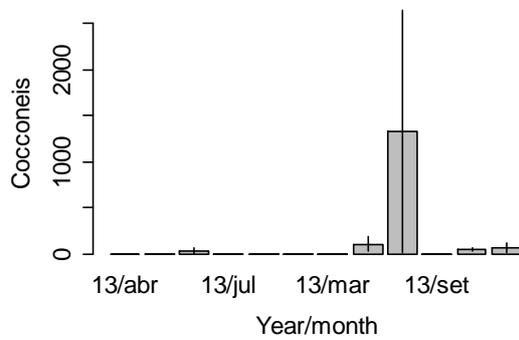
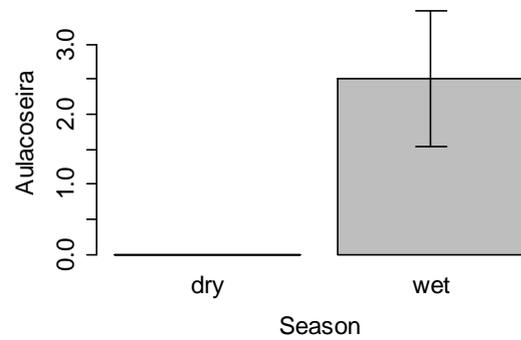
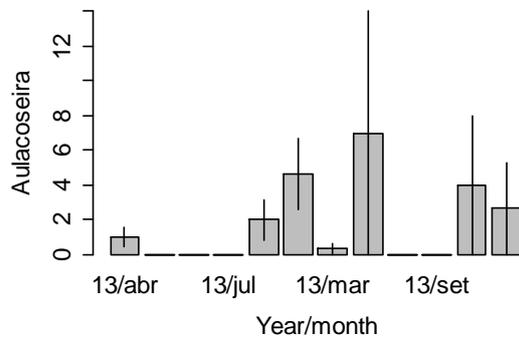
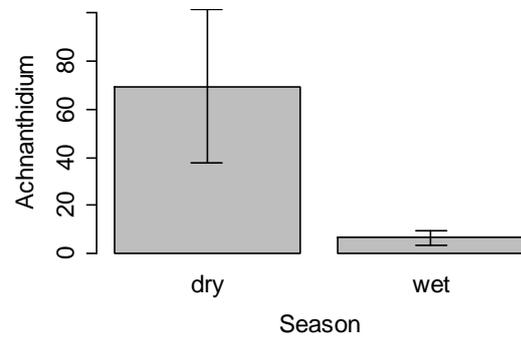
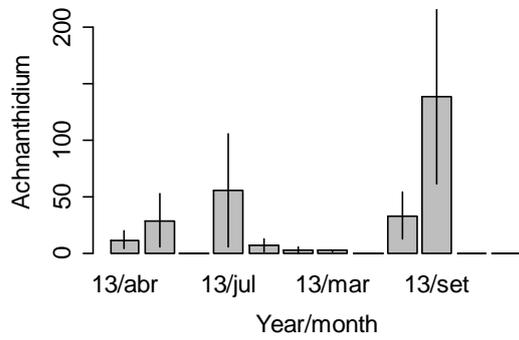


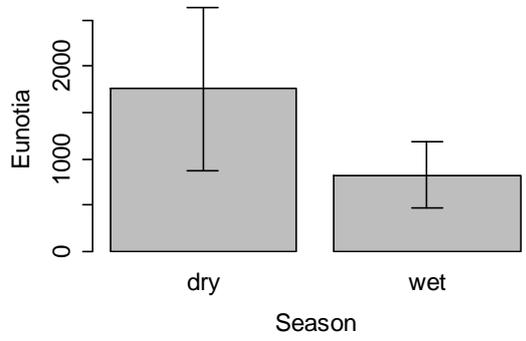
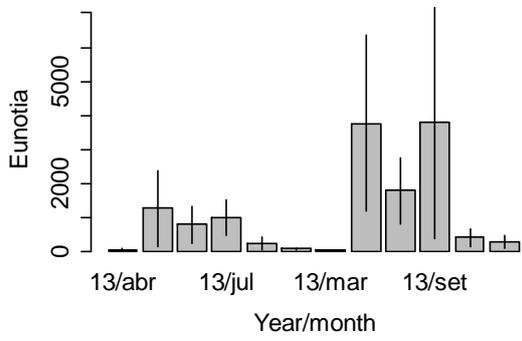
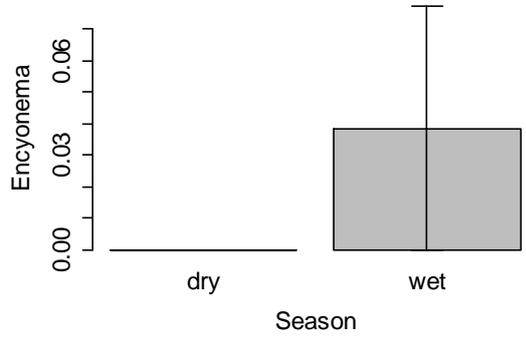
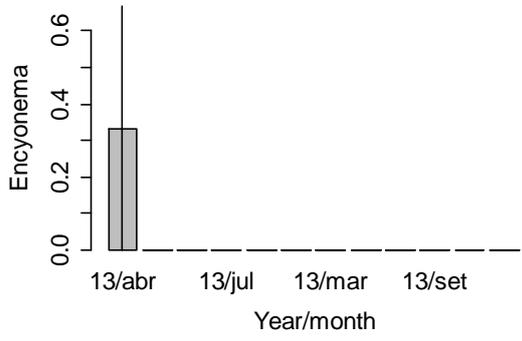
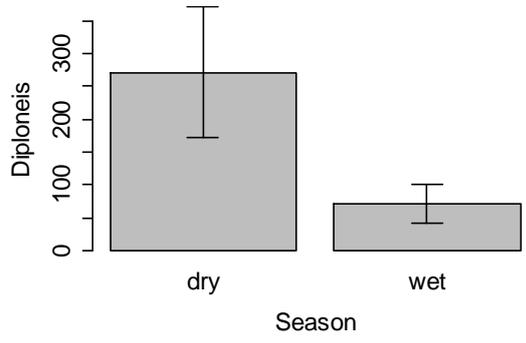
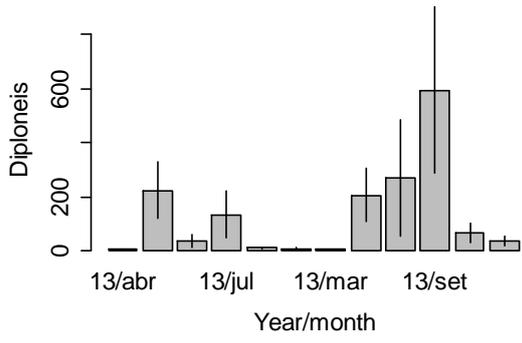


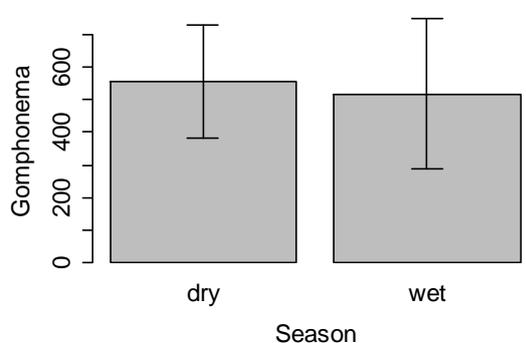
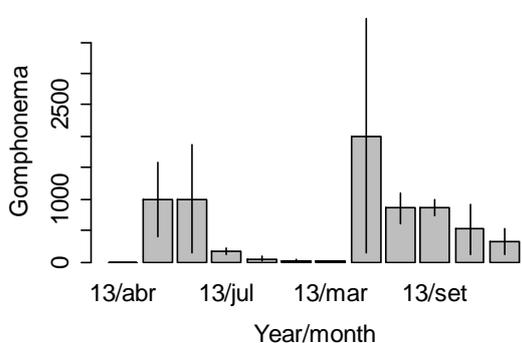
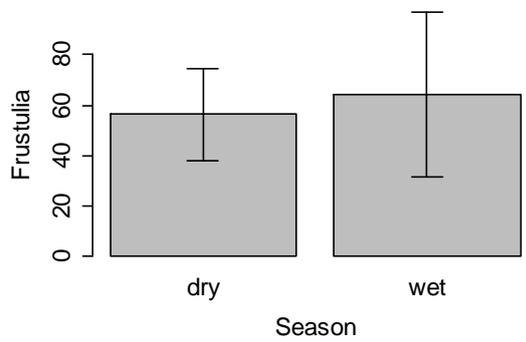
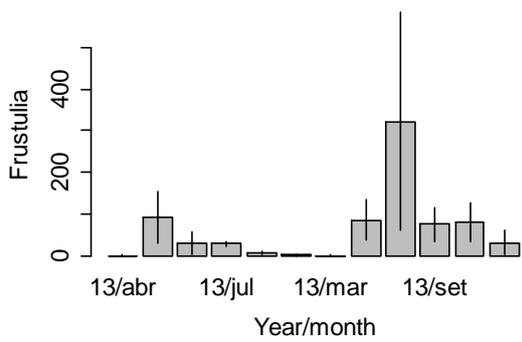
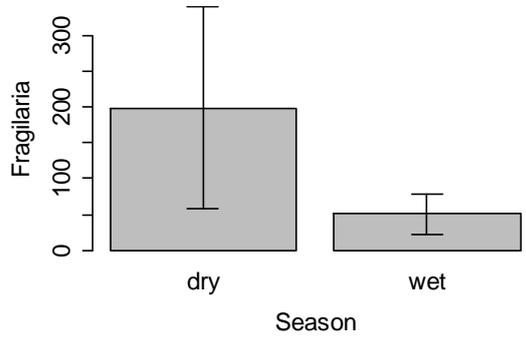
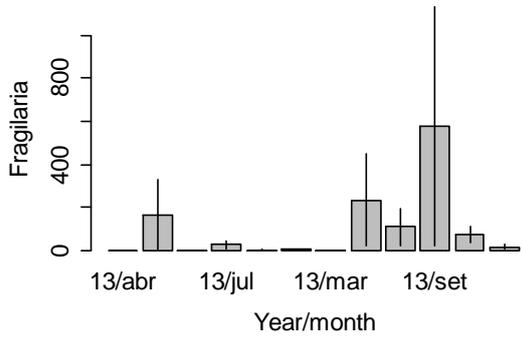


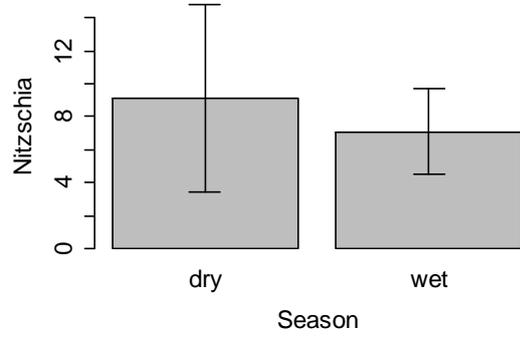
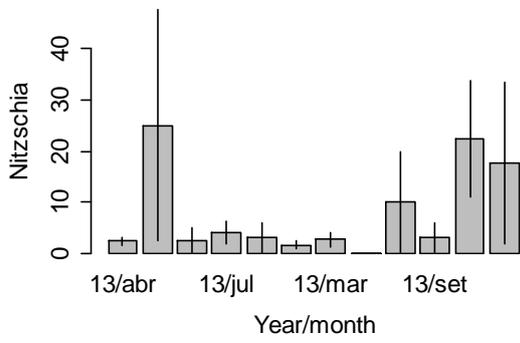
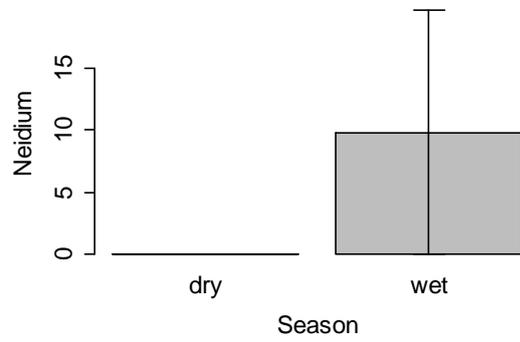
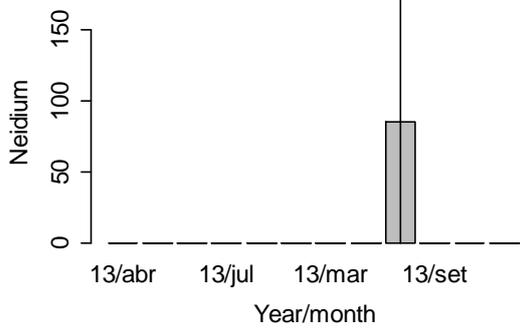
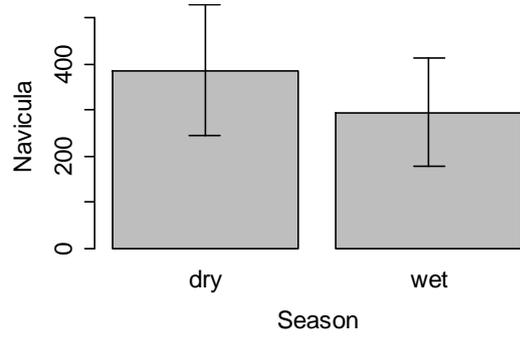
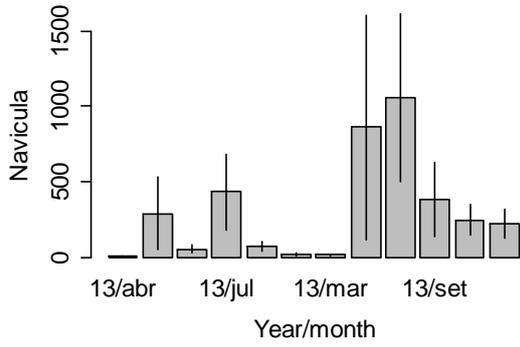


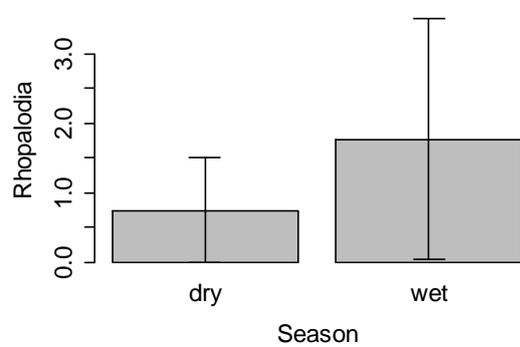
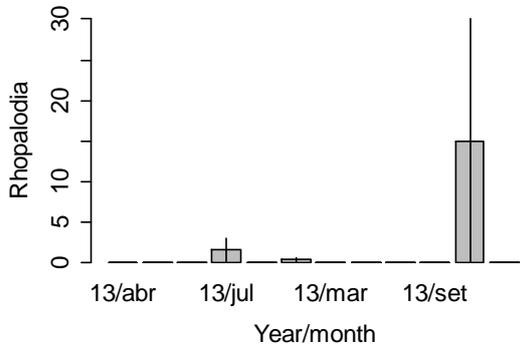
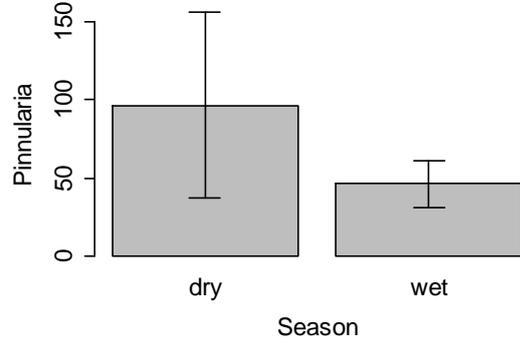
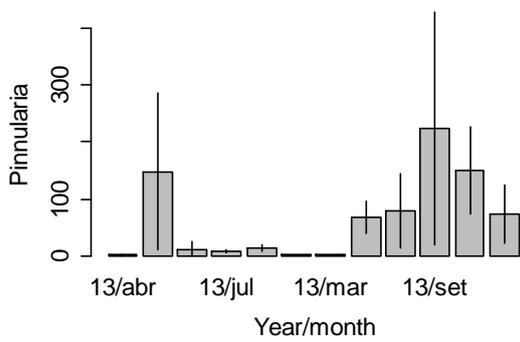
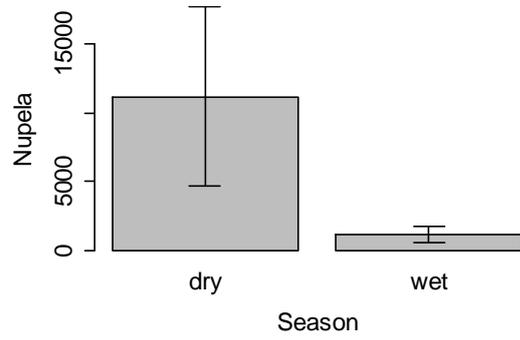
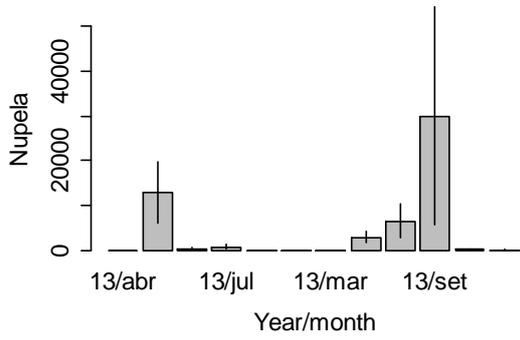
Genus absolute abundance (cel.cm²) per month and per season (all identified genus)











Diatom traits per month and per season – relative abundances. Length in μm

