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RESEARCH ARTICLE

Temporal variation in the Ephemeroptera, Plecoptera and Trichoptera community in response to environmental drivers in a subtropical stream

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Abstract: Aquatic insects are important elements in the ecological dynamics of lotic systems and their distribution in these environments can be driven by several environmental factors. Based in this assumption the aim of this study was to evaluate which environmental variables act as structural driver of the Ephemeroptera, Plecoptera and Trichoptera (EPT) community. We hypothesize that community structure will display strong temporal alterations due to characteristics in subtropical regions. We evaluated the effect of water temperature, conductivity, dissolved oxygen, depth, current velocity and rainfall on the distribution of EPT community. The insects were sampled using the litter bags incubation in a third-order subtropical stream located on Santa Catarina Island, Southern Brazil. The abundance of EPT was monitored for 24 months between January 2012 and December 2013. We identified 530 EPT specimens belonging to 11 families and 20 genera. The community showed a significant variation along the monitored time, mainly attributed to Trichoptera order. In addition to the results of this work the Ecnomidae family, *Austrotinodes* and *Neotrichia* genera represent new records for Santa Catarina State. Environmental variables varied significantly during the study time and depth and rainfall were the mainly environmental drivers acting under the community structure. We also observed a negatively correlation among rainfall and EPT community, especially to Trichoptera, which showed a strong temporal variation. In this study the EPT community displays a strong temporal variation along the monitored time, mainly attributed to rainfall events, characteristic of subtropical region. We also suggest that body adaptations also have an influence in the EPT

community structure, once under flood event conditions these adaptations may offer advantage or disadvantage to invertebrate establishment, as pointed in the Trichoptera order.

Key words: Environmental drivers; EPT community; subtropical stream; temporal distribution.

Introduction

The distribution and species composition of aquatic invertebrates in lotic environments are determined by a range of local factors, including current velocity, water temperature, dissolved oxygen (Bueno *et al.* 2003), rainfall (Bispo *et al.* 2004), conductivity (Scheibler & Debandi 2008), depth (Mollozzi *et al.* 2011) and the type of substrate (Buss *et al.* 2004). These variables show spatial and temporal variability and are therefore expected to drive structural changes in lotic invertebrate communities.

Generally, rainfall in lotic environments is considered to be the main factor that affects aquatic communities (Kim *et al.* 2013), since it interacts with local factors to affect lotic characteristics such as current velocity, depth, dissolved oxygen and type of substrate. In neotropical streams, these factors play an important role in the structure of aquatic communities and cause an increase or decrease in the abundance and diversity of the organisms that respond to changes in environmental characteristics. In tropical regions, the aquatic invertebrate community is generally more abundant during the dry season (Aburaya & Callil 2007), and declines during the rainy period (Righi-Cavallaro *et al.* 2010).

A clear distinction in the richness and diversity in the aquatic invertebrate community has also been found between dry and rainy periods. For example, Rios-Touma *et al.* (2011) observed a higher richness and diversity of the aquatic invertebrate community during the dry season in the Piburja stream (Ecuador), and Feeley *et al.* (2012) noted that high water levels due to increased rainfall induced significant losses in the richness and diversity of the invertebrate community. However, some studies have demonstrated that responses in invertebrate communities are probably not directly coupled to precipitation, but occur via related changes in stream characteristics, such as the carriage of sediments and organisms, due to an increase in current velocity and water flow (Oliveira *et al.* 1997).

Another environmental variable that can cause changes in aquatic invertebrate community is temperature; species composition and biological rates are temperature-dependent. Seasonal changes in water temperature can cause changes in the metabolic rate of organisms, as well affect their distribution along the course of a river and within geographic regions (Allan & Castillo 2007). The increased temperature during spring and summer is one of the main variables that influences the life cycle, egg incubation, fecundity, the dormancy of an organism (Sweeney & Vannote 1986), the foraging behavior and feeding strategies of aquatic organisms (Ward & Stanford 1982). Temperature also plays a major role in regulating seasonal changes in the growth rates of aquatic insects.

Among aquatic insects, Ephemeroptera, Plecoptera and Trichoptera (commonly called EPT) consist of a diverse group of taxa that are sensitive to environmental changes, such as pollution and habitat fragmentation (Rosenberg & Resh 1993). Additionally, EPT taxa have a higher abundance and richness and play an important role in nutrient cycling and litter decomposition processes (Wiggins & Mackay 1978). The aim of this study was to evaluate which environmental variables act as structural drivers of the EPT community. Based in the assumption that the EPT community is sensitive to environmental changes, we hypothesize that taxonomic richness and abundance of community display strong temporal changes, due

to the four well-defined seasons, characteristics of subtropical regions, with a well-distributed rainfall throughout the year.

Material and methods

Study area

This study was conducted at the Parque Municipal da Lagoa do Peri (PMLP), a protected area on Florianópolis Island, Santa Catarina, Brazil (Fig. 1). The PMLP is located at the southern end of the island (27°44'S; 48°31'W), and consists of an area of 2.03 ha, which is covered by coastal vegetation in the coastal plain and primary forest area at higher altitudes next to the main headwaters of the watershed (Cecca 1997). The climate of the region is characterized as subtropical, with rainfall throughout the whole year; however, historical precipitation data since 1992 show higher rainfall in the warm months between December and March (Lisboa *et al.* 2015).

Samples of the EPT community were collected at Cachoeira Grande stream, a third-order stream according to the Strahler classification (1957), located in the western part of Lagoa do Peri and covered by a high cover Atlantic Forest with different stages of regeneration and succession. The stream has a maximum elevation of 280 m and drains a catchment of 1.66 km² (Dos Santos *et al.* 1989). The stream usually flow all-year-round, but can greatly increase in summer (December to March) due to a higher volume of rainfall.

Sampling design

Sampling of the EPT community was performed along a 100 m stretch of the Cachoeira Grande stream which was divided on five sampling sites, 20 m distant from each other. At each sampling site, litter bags were incubated in the stream, in triplicate, adding 15 litter bags *per* month, to colonization during 30 days to promote colonisation by aquatic invertebrates. This methodology was used to avoid the substrate drift.

The litter bags contained senescent leaves from riparian vegetation, collected along the Cachoeira Grande stream. The senescent leaves were obtained by eighteen leaf collectors placed along three strings, within 2 m of the stream course. The leaves retained in the collectors were incubated for 30 days in the stream, and the EPT community was sampled monthly between January 2012 and December 2013. The amount of material placed into litter bags in each month varied in terms of the relative species richness and total amount of litter (3.39 g ± 2.12 g).

In the laboratory, the EPT specimens were preserved in 70% alcohol and identified to the genus level under a stereomicroscope using the keys in Domínguez & Fernández (2009) and Pes *et al.* (2005).

Water parameters

The dissolved oxygen, electrical conductivity and water temperature were measured using a YSI 85 multi-parameter probe (YSI Incorporated, Yellow Spring, OH). Water depth was obtained using a measuring tape and the current velocity (m.s⁻¹) using the float method (Bain & Stevenson 1999). All abiotic variables were recorded monthly January 2012 through December 2013 in five sampling sites and grouped by season. Seasonal accumulated rainfall was obtained by summing daily precipitation data summed them up for three months. Daily rainfall was obtained from the Centro de Recursos Ambientais e de Hidrometeorologia of Santa Catarina, a rainfall station located approximately 15 km from the study site.

Data analysis

To improve the comparison and interpretation of our results, we grouped the data (water temperature, conductivity, depth, dissolved oxygen, current velocity, rainfall and EPT community) by season (Summer = January, February, March; Autumn = April, May, June; Winter = July, August, September; Spring = October, November and December). We used a one-way ANOVA to test the differences in the physical and chemical parameters of the water among the studied seasons. Rainfall data were used as monthly accumulated and grouped by season.

A Principal Coordinate Analyses (PCoA), using the Bray-Curtis index for similarity between samples was performed to visualize the scatter of the EPT community based on their seasonality distribution. The relationship between environmental variables and EPT community structure was explored by the BIO-ENV routine (method: BEST), tested with Monte Carlo significance levels (Clarke & Gorley 2006). All statistical analyses and graphs were performed and created in the R-Program version 3.2.2 and PRIMER6 & PERMANOVA+.

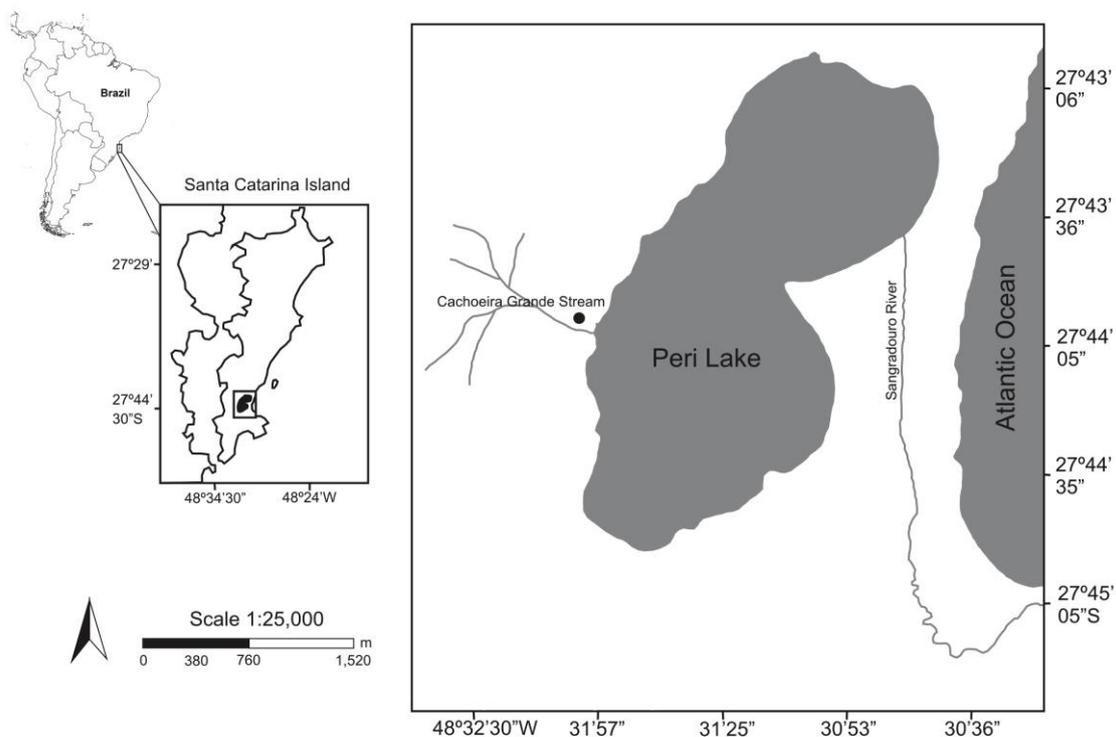


Figure 1. Geographic location of Cachoeira Grande stream, in the Parque Municipal da Lagoa do Peri, Florianópolis, Santa Catarina, Brazil. Modified from Lemes-Silva *et al.* (2016).

Results

Abiotic variables

The water temperature ($F_{7,76} = 23.0$; $p = 0.0001$), conductivity ($F_{7,76} = 5.206$; $p = 0.0001$), dissolved oxygen ($F_{7,76} = 5.182$; $p = 0.0001$), depth ($F_{7,76} = 14.53$; $p = 0.0001$), current velocity ($F_{7,76} = 8.323$; $p = 0.0001$) and rainfall ($F_{7,76} = 49.57$; $p = 0.0001$) varied

significantly among the seasons (Fig. 2). Water temperature was highest in the Summer 2012 and lowest in the Winter 2013. Conductivity showed the highest values during the Autumn 2012. Dissolved oxygen was highest in the Autumn 2013 and lowest in Summer 2013. Depth was highest in the Summer 2013 and lowest in Spring 2012. Current velocity and rainfall showed the highest values in the Spring 2012 and Summer 2013, respectively.

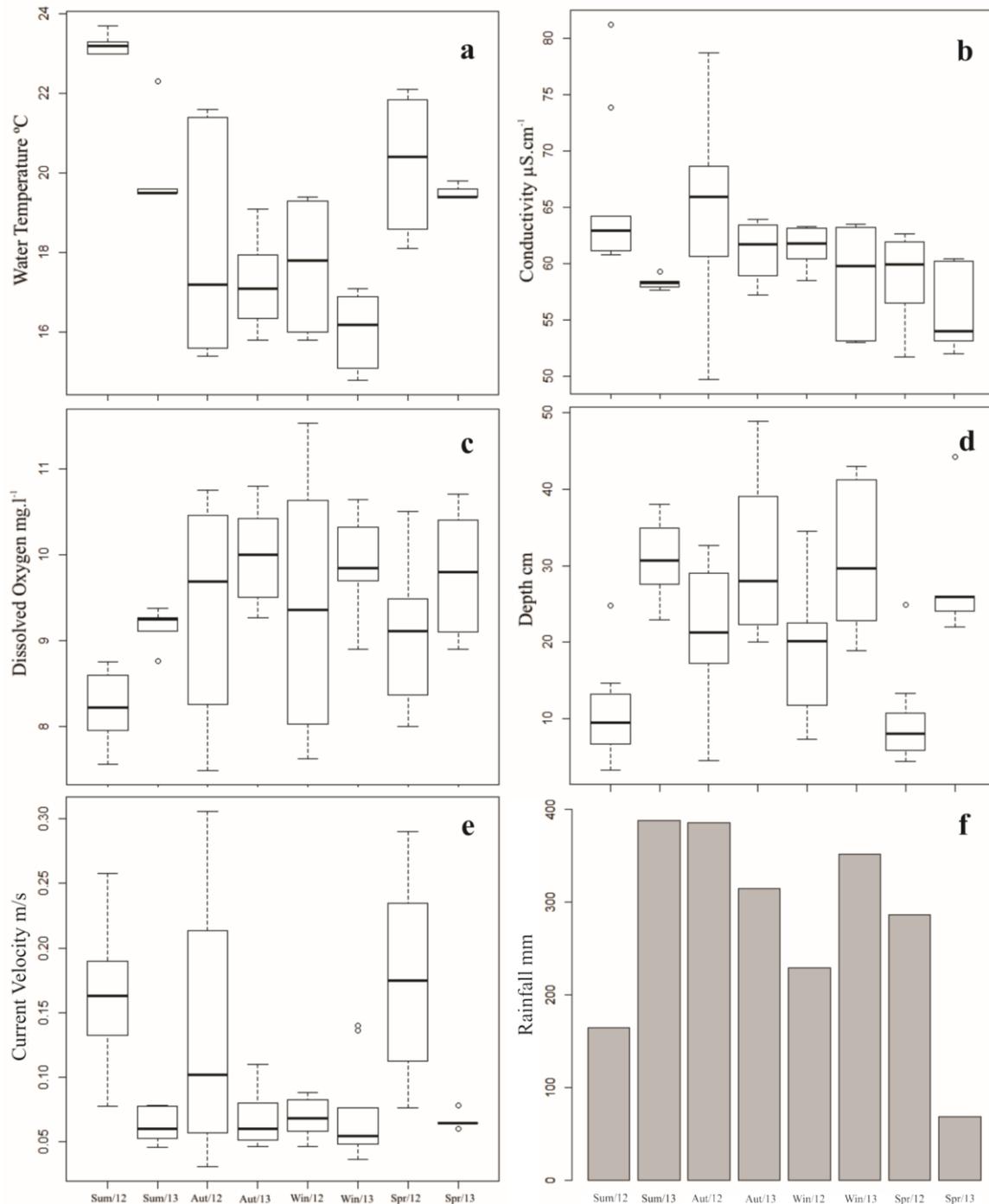


Figure 2. Seasonal comparison of (a) water temperature; (b) conductivity; (c) dissolved oxygen; (d) depth; (e) current velocity (boxes represent the upper quartile, median and lower quartile, and whiskers represent the 5th and 95th percentiles, central squares represent the median) and (f) seasonal accumulated rainfall. Sum = Summer (January to March); Aut = Autumn (April to June); Win = Winter (July to September); Spr = Spring (October to December).

EPT community

We identified a total of 530 EPT individuals, distributed among 11 families and 20 genera (Table 1). Trichoptera were most abundant (85.1% of collected individuals) and showed the greatest richness of genera, with eight families and 13 genera, of which *Macronema*, *Triplectides*, *Phylloicus* and *Polycentropus* were the more representative genera. In addition, the Ecnomidae family, and the genera *Austrotinodes* and *Neotrichia* represent new records for the Santa Catarina State. Immature individuals of Ephemeroptera represented 14.5% of the total, distributed among two families and five genera, of which *Farrodes* and *Miroculis* were the most abundant. The Plecoptera was lowly abundant (0.4% of collected individuals) and was represented by only two genera: *Gripopteryx* and *Paragripopteryx*.

The genera *Macronema* and *Triplectides* were dominant in all seasons; whereas *Gripopteryx*, *Paragripopteryx*, *Austrotinodes*, *Smicridea*, *Neotrichia* and *Nectopsyche* were only recorded in one season throughout the study period. The highest abundance and richness of the EPT community was recorded during spring and the lowest during summer (Table 1).

Table 1. Taxonomic composition of Ephemeroptera, Plecoptera and Trichoptera (EPT) collected during January 2012 and December 2013 in Cachoeira Grande stream, Florianópolis, Santa Catarina, Brazil. Sum = Summer (January to March); Aut = Autumn (April to June); Win = Winter (July to September); Spr = Spring (October to December).

Taxa	2012				2013			
	Sum	Fall	Win	Spr	Sum	Fall	Win	Spr
Ephemeroptera								
Baetidae								
<i>Americabaetis</i> Kluge, 1992	1	-	-	4	-	1	-	7
<i>Zelusia</i> Lugo-Ortiz & McCafferty, 1998	-	-	-	4	-	-	-	1
Leptophlebiidae								
<i>Farrodes</i> Peters, 1971	-	1	1	8	6	3	5	5
<i>Massartela</i> Lestage, 1930	-	-	-	2	-	-	-	1
<i>Miroculis</i> Edmunds, 1963	-	4	4	9	1	-	5	4
Plecoptera								
Gripopterygidae								
<i>Gripopteryx</i> Pictet, 1841	-	-	-	-	-	-	-	1
<i>Paragripopteryx</i> Enderlein, 1909	-	-	1	-	-	-	-	-
Trichoptera								
Calamoceratidae								
<i>Phylloicus</i> Müller, 1880	2	2	3	7	1	-	8	19
Ecnomidae								
<i>Austrotinodes</i> Schmid, 1955	-	-	2	-	-	-	-	-
Hydrobiosidae								
<i>Atopsyche</i> Banks, 1905	-	-	-	2	0	1	-	-
Hydropsychidae								
<i>Macronema</i> Pictet, 1836	3	7	8	69	20	26	13	70
<i>Smicridea</i> McLachlan, 1871	-	-	-	-	1	-	-	-
Hydroptilidae								
<i>Metrichia</i> Ross, 1938	-	3	-	1	-	1	-	1
<i>Neotrichia</i> Morton, 1905	-	-	-	-	-	-	-	1
Leptoceridae								
<i>Nectopsyche</i> Müller, 1879	-	-	-	-	-	-	-	1
<i>Oecetis</i> McLachlan, 1877	-	-	-	-	-	1	1	-
<i>Triplectides</i> Kolenati, 1859	7	31	27	23	4	7	4	18
Philopotamidae								
<i>Chimarra</i> Stephens, 1829	-	-	-	2	-	8	-	-
Polycentropodidae								
<i>Cernotina</i> Ross, 1938	-	1	2	1	-	-	1	5
<i>Polycentropus</i> Curtis, 1835	1	1	4	5	-	-	2	23
Abundance	14	50	52	137	33	48	39	157
Richness	5	8	9	13	6	8	8	14

The PCoA ordination showed that EPT community among seasons revealed a strong compositional difference (Fig. 3). The ANOVA test also showed the EPT abundance varied significantly between seasons ($F_{7,15} = 3.311$; $p = 0.024$). The differences were mostly attributed to the changes in the order Trichoptera, which dominated in all seasons and varied significantly over time, with the highest abundance mainly in spring and a lower abundance in the summer.

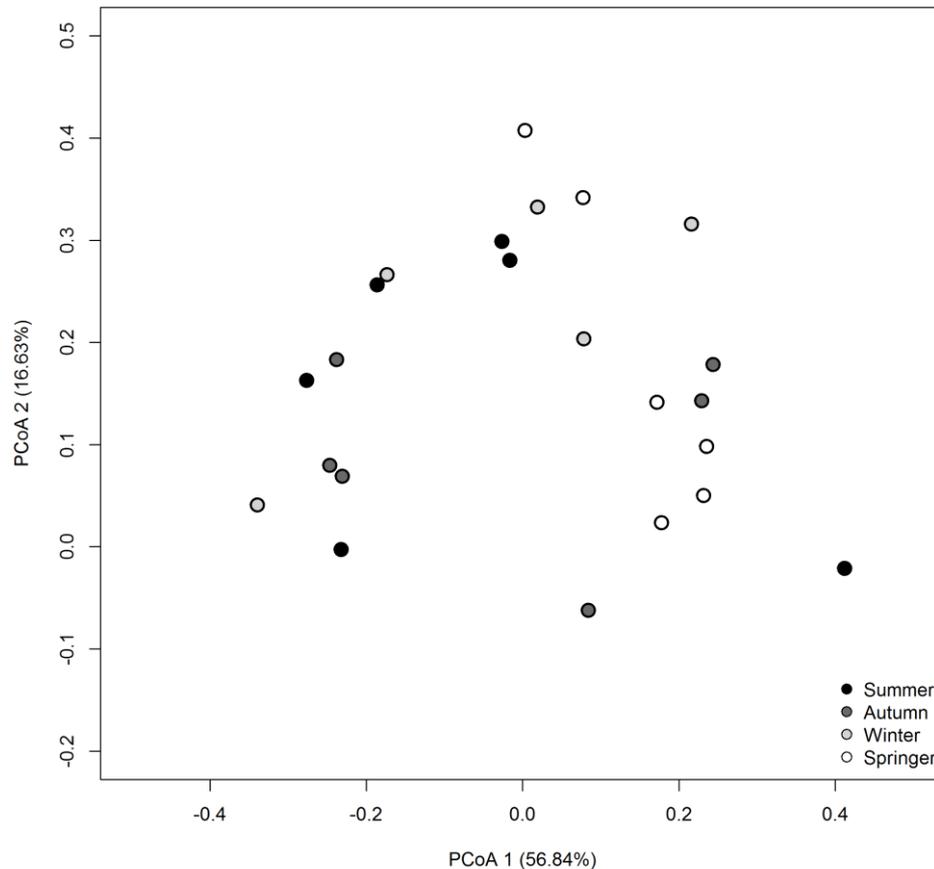


Figure 3. Scatter of Ephemeroptera, Plecoptera and Trichoptera (EPT) community across seasons sampled in the Cachoeira Grande stream between January 2012 and December 2013.

BIO-ENV analyses indicated that depth and rainfall were the main drivers of the EPT community ($r = 0.307$). We also observed a negative relationship between EPT abundance and the amount of rainfall, suggesting a lower EPT abundance during periods of heavy rains, and the community was more abundant in months with lower rainfall values (Fig. 4).

Discussion

Occurrence of EPT in relation to abiotic characteristics

Studies in tropical and subtropical streams have shown that temporal changes in temperature and rainfall regimes affect the distribution of aquatic communities in streams (Bispo & Oliveira 2007, Righi-Cavallaro *et al.* 2010). In this study, the EPT community assemblages showed different temporal patterns in the prevalence of some groups of

organisms (Trichoptera) to the detriment of others (Ephemeroptera and Plecoptera). The results corroborate our first hypothesis that rainfall strongly influences the EPT community, which displays a lower abundance in rainy months. However, the influence of rainfall indirectly characterizes the river environment via its influence on several of the important physical and chemical conditions, because water flow that carries the insects downstream increases the oxygenation level and contributes to the input of sediments and organic matter. The highest current velocity was not necessarily observed in the seasons with the highest rainfall, probably because the current velocity did not reflect rainfall data on the sampled day, but in a general way reflecting in EPT community.

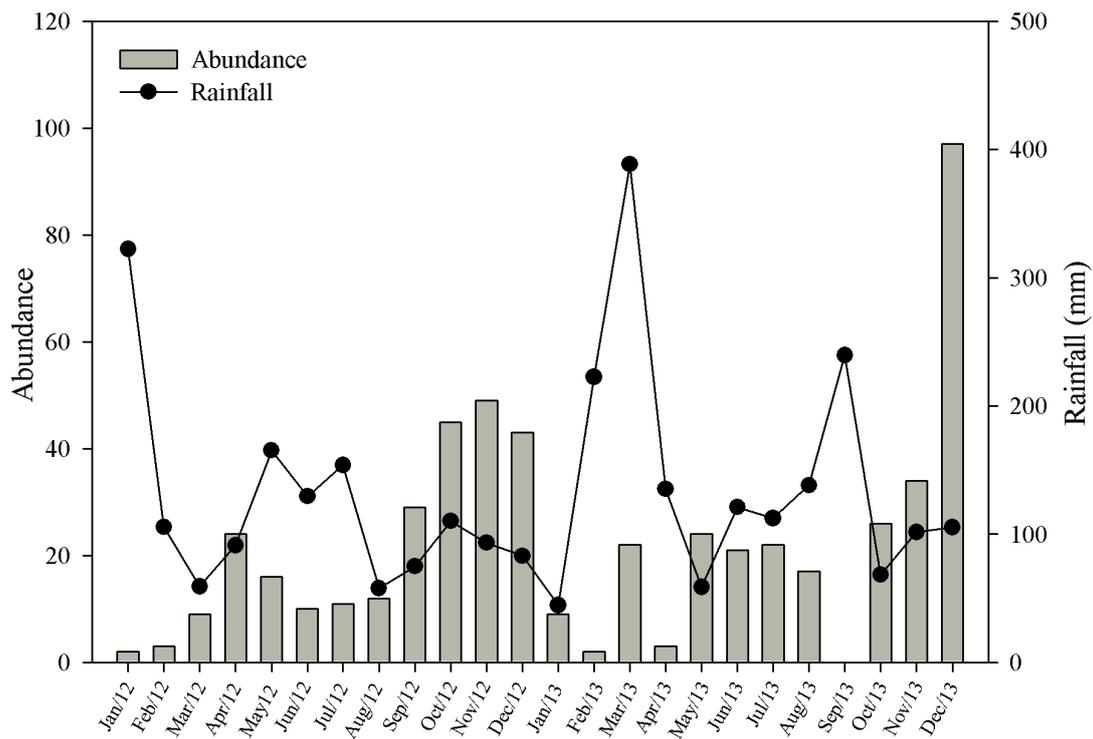


Figure 4. Abundance of the Ephemeroptera, Plecoptera and Trichoptera (EPT) community sampled in the Cachoeira Grande stream between January 2012 and December 2013 and its relationship with rainfall.

Rainfall events can promote an increase in the amount of allochthonous organic matter from terrestrial sources (Lisboa *et al.* 2015) and change the electric conductivity of the environment. Changes in depth also are expected, due to the drift and deposition of substrates, and consequently change the composition and habitat diversity available to invertebrates (Buss *et al.* 2004). The substrate becomes more uniform, which can change the dissolved oxygen concentration and temperature (Souza *et al.* 2014), because an increase in rainfall is expected to result in an increase in the current velocity.

Although our results do not point to the water temperature as a variable responsible for the EPT community structure, the same variable seems to have influence on the macroinvertebrate community structure of Brazilian subtropical region (Beghelli *et al.* 2014). The increase in temperature during warm months combined with a higher light intensity,

favours primary production, including the periphyton growth (Von Schiller *et al.* 2007), which is one of the main food resources in low order streams (Vannote *et al.* 1980). Then, in summer the habitat can support more abundant communities. A higher EPT abundance was expected in summer, but it is supposed that the effects of high rainfall exceeded the influence of temperature on the community and therefore, temperature, in our case, is probably less important than rainfall in determining the distribution patterns of the EPT community.

Variation of EPT community

The low EPT abundance during periods of high rainfall was probably caused by an increase in water flow and consequently caused individuals drift. Bispo *et al.* (2001, 2004, 2006) observed a low abundance of the EPT community in the rainy season in a tropical climate and a greater number of individuals in drier months. In an equatorial climate, Suhaila *et al.* (2014) also observed a reduction in the abundance of macroinvertebrates that was correlated with an increase in the amount of rainfall.

The observed difference in the structure of the EPT community might only relate to the Trichoptera. They are abundant in lotic streams and are sensitive to environmental changes (Ross 1967). According to the same author, the Trichoptera community in small streams is affected by the distribution of local rainfall and periodicity of leaf-fall, which causes the ecological conditions for this group to change. Our results corroborate this finding, since a lower abundance and richness was observed during the rainy season (summer) and a higher abundance and richness was present during the dry season (spring).

The availability of food is an obvious factor controlling the occurrence and abundance of species. The higher abundance of the genera *Macronema*, *Phylloicus* and *Triplectides* might be related to the organic substrate deposited in areas with lower water velocity, which is more attractive to these organisms, since they are collectors and shredders, respectively (Oliveira & Bispo 2001).

The greater effect of precipitation on the community of Trichoptera abundance than on Ephemeroptera and Plecoptera could be associated with their smaller escape capacity associated with the building of a shelter which probably limits their movement during flood periods. For example, *Macronema* constructs a type of sand funnel shelter attached to the substrate, *Phylloicus* uses leaves in the construction of shelters (Prather 2003) and individuals of *Triplectides* use small pieces of wood (Crisci-Bispo *et al.* 2004). The other trichopteran genera also builds portable or fixed shelters attached to a substrate and the free-living individuals anchor themselves clinging from a silk thread and as consequence some of them can be extremely sluggish in their movements (Ross 1967). Consequently, they can be easily drifted with higher flows than Ephemeroptera and Plecoptera.

Feeley *et al.* (2012) studied the effects of catastrophic storm events on the benthic macroinvertebrate community found no significant changes in the richness of the Ephemeroptera and Plecoptera communities following the storm events, but did observe a severe loss in the richness and diversity of the Trichoptera community after the disturbance, which might be attributable to the larval characteristics of Trichoptera.

In general, we observed that EPT community were driven mainly rainfall influence, factor that can change the environmental hydrological conditions establishing disturbed conditions corroborating with the hypothesis that community structure will respond temporally to subtropical region characteristics. Besides, knowledge about morphology and body adaptations in invertebrates' communities also need be considered to understand changes in the communities' structure and ecological assessment.

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