



## Influence of land-use on structural and functional macroinvertebrate composition communities associated on detritus in Subtropical Atlantic Forest streams

Influência do uso da terra sobre a composição estrutural e funcional da comunidade de macroinvertebrados associados a detritos em riachos subtropicais da Floresta Atlântica

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**Abstract: Aim:** Our aim in this study was to evaluate the effects of land use in drainage basins of the streams on the taxonomic and functional composition of aquatic invertebrate communities associated in leaf litter. **Methods:** We evaluated the colonisation of invertebrates in the incubated plant debris in streams with presence and absence of riparian vegetation and different land-uses in the drainage area. We used the litter bags approach. **Results:** The taxonomic and functional composition invertebrate associated with leaf litter ranged between streams. In addition, streams with presence of vegetation showed less variation taxonomic and functional composition communities. Still, the density of shredders invertebrates were lower in streams without vegetation. **Conclusions:** The riparian vegetation is an important environmental factor in the composition of invertebrates. However, the land-use throughout the drainage basin should be considered as relevant factor in structuring aquatic biota.

**Keywords:** invertebrates colonisation; functional feeding group; environmental quality; environmental integrity.

**Resumo: Objetivo:** O objetivo deste estudo foi avaliar os efeitos de usos da terra em bacias de drenagem de riachos sobre a composição taxonômica e funcional das comunidades de invertebrados aquáticos associados a detritos foliares. **Métodos:** Avaliamos a colonização de invertebrados em detritos de plantas incubadas em riachos com presença e ausência de vegetação ripária e diferentes usos da terra na área de drenagem. Utilizamos a abordagem de litter bags. **Resultados:** A composição taxonômica e funcional da fauna de invertebrados associados aos detritos variou entre os riachos. Além disso, os riachos com presença de vegetação mostraram comunidades com menor variação taxonômica e funcional. Ainda, a densidade de invertebrados fragmentadores foi menor nos riachos sem vegetação. **Conclusões:** A porcentagem de vegetação ripária é um fator ambiental importante na composição dos invertebrados. Porém, os usos da terra em toda a bacia de drenagem devem ser considerados como fatores relevantes na estruturação da biota aquática.

**Palavras-chave:** colonização de invertebrados; grupos tróficos funcionais; qualidade ambiental; integridade ambiental.



## 1. Introduction

Agricultural activities have increased the degradation of forests in recent decades (Carvalho et al., 2009; Diniz-Filho et al., 2009). When natural riparian vegetation is removed for agricultural uses, the water temperature, nutrient concentration and sediment input tend to increase in streams, causing negative effects to the ecological integrity of aquatic ecosystems (Allan, 2004; Blevins et al., 2013). However, it is unclear how the combined effect of different anthropogenic factors (e.g., riparian vegetation removal) can alter ecological processes and aquatic biota (Hagen et al., 2006; Arnaiz et al., 2011; Bonato et al., 2012; Chadwick et al., 2012).

Riparian zones are important for the maintenance and regulation of the aquatic environment (Naiman et al., 2005). The presence of riparian vegetation acts as a barrier to sediment input (Klapproth & Johnson, 2000), performing a hydrological role (e.g. superficial filtering effect and sub-surface water) and assisting in water quality maintenance. The removal of riparian vegetation in agricultural areas causes the loss of allochthonous material input, which is of key importance for maintaining the energy flow and balance of streams (Henry et al., 1994; Gonçalves Junior & Callisto, 2013).

Organic matter that is present in the water from the riparian zone has long been studied in a fragmented way as food webs and energy flow, and can reveal patterns and generate ecological benefits (Tank et al., 2010). These studies have demonstrated that changes in riparian conditions result in dynamic variations in the aquatic fauna (Angermeier & Karr, 1994; Gonçalves Junior et al., 2012), stream metabolic processes, and organic matter (Campbell et al., 1992; Webster et al., 1995; Sponseller & Benfield, 2001). In addition, removal of the vegetation has negative consequences on growth rates, abundance and the invertebrate community trophic structure (Chakona et al., 2009; Chadwick et al., 2012; Suga & Tanaka, 2013; Ferreira et al., 2015). In addition, changes in riparian vegetation affect the typical fragmentary distribution and decomposition of allochthonous organic matter (Sponseller & Benfield, 2001; Encalada et al., 2010).

However, in recent years, some studies have shown that land use in the drainage area of aquatic environments also generates significant effects on aquatic communities. Changes in land use at different landscape scales alter the structure and composition of aquatic communities (Sensolo et al.,

2012; Valle et al., 2013; De Toni et al., 2014). Although the attention of the scientific community to be directed to the riparian zone of the streams, the presence and amount of vegetation in the drainage areas of these environments is important for the maintenance of environmental integrity, as well as providing conditions for the establishment and colonization of benthic macroinvertebrates (Oliveira et al., 2014). Thus, in this study, we evaluated the modifications in the land-use of streams drainage areas and the relationship with the taxonomic and functional feeding composition of aquatic invertebrate communities associated with leaf detritus. We tested the hypotheses that drainage area with less percentage of vegetation and absence of the riparian vegetation influences the composition of invertebrate fauna in detritus, resulting in a low variability of the community.

## 2. Material and Methods

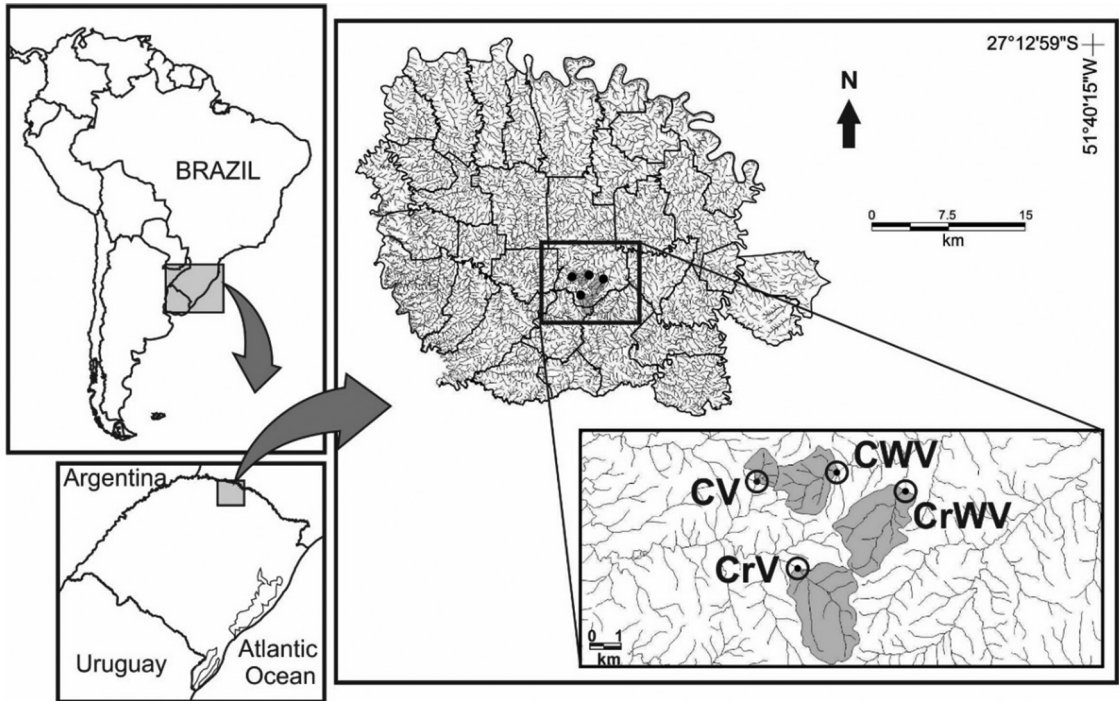
### 2.1. Study area

We conducted this study in four small streams (<3rd order) distributed in hydrographic sub-basin of Campo river (C) and hydrographic sub-basin of Cravo river (Cr) in southern Brazil (Figure 1). The climate is subtropical with a regular rainfall regime and well-defined annual seasons. The annual mean temperature is 17.6 °C and rainfall is well distributed throughout the year (range: 1750-2000 mm, annual mean: 1912 mm). The vegetation is characterized by a mixture of Subtropical Forest of the Alto Uruguay and Mixed Rain Forest (Oliveira-Filho et al., 2015). The predominant land use is agricultural practices (~77% of the area) and only 15% of the region area contains native vegetation (Decian et al., 2009).

We selected two streams with riparian vegetation (V) in the adjacent area and two streams without riparian vegetation (WV) at drainage area. We used the riparian vegetation in the buffer zone as categorical variable. In addition, two streams (one with and other without riparian vegetation) are located in the same hydrographic sub-basin. In this form, we have four streams, two riparian vegetation situations, and two hydrographic sub-basins.

### 2.2. Field experiment

We used senescent leaves of *Campomanesia xanthocarpa* O. Berg (Myrtaceae). This species is native and occurs frequently in the riparian zone of streams in the forest formations in southern Brazil (Oliveira-Filho et al., 2006). We dried the leaves at environmental temperature (~25 °C/5 days).



**Figure 1.** Localization of the streams in the Campo river hydrographic sub-basin and Cravo river hydrographic sub-basin, Southern Brazil.

The senescent leaves of *C. xanthocarpa* contain 2% of nitrogen, 0.04% of phosphorous, 4.5% of tannin and have a C:N ratio = 23 (see details in Tonin et al., 2014).

We conducted the experiment in August 2010. We weighed  $2.5 \pm 0.1$  g leaves of *C. xanthocarpa* and added them to litter bags ( $15 \times 20$  cm, 10 mm mesh). We incubated the litter bags in low current areas in the streams, randomly, and after 7, 14 and 21 days of leaf immersion, we removed four litter bags from each stream. We stored the litter bags in a thermic cooler, and in the laboratory, washed the debris to remove the associated invertebrates. We used a total of 48 litter bags in this study.

### 2.3. Associated invertebrate fauna

In the laboratory, we gently washed the leaves in running water through a sieve (250  $\mu$ m mesh) for invertebrate retention. We dried the material remaining in an oven at 40 °C/72 h, and weighed it to determine the mass loss. We identified the benthic invertebrates associated with the detritus to the lowest possible taxonomic level according to Merritt & Cummins (1996), Fernandez & Domingues (2001) and Mugnai et al. (2010). The classification of functional feeding groups was performed following the recommendations

of Merritt & Cummins (1996), Fernandez & Domingues (2001), Kennet et al. (2005).

We excluded chironomid larvae from the analysis and the interpretation of results because they constitute a very abundant group in leaf litter decomposition (Ligeiro et al., 2010; Biasi et al., 2013). In addition, they perform an unclear ecological function in debris and have contained functional feeding groups (Sanseverino & Nessimian, 2008; Tonin et al., 2014).

### 2.4. Landscape and limnological variables

We analyzed the landscape using geoprocessing techniques. The base map was adapted to work at 1:35,000, with a spatial distribution of  $1 \times 1$  m, which were demarcated as catchment areas using the Triangle Irregular Network method. The classification of land use followed the method of rating Supervised Maximum Likelihood (maxlike). The ratings that fell within the area where the study was performed were: agriculture, exposed soil, pasture, and vegetation. We used MapInfo 8.5 and Idrisi 32 software.

We measured limnological variables in situ and collected water samples for laboratory analysis. The water variables measured in situ were: water temperature, dissolved oxygen, pH, conductivity

and turbidity and total dissolved solids, using a multiparameter Horiba® U-50PC analyzer. The variables measured *ex situ* were alkalinity via an acid-based titration, and total organic carbon and total nitrogen were measured using a TOC Analyzer Shimadzu®.

### 2.5. Data analysis

We evaluated the leaf mass loss during the study period as the decomposition rate ( $k$ ) by adjusting the values of the remaining dry weight by the negative exponential model  $W_t = W_0 \cdot e^{-kt}$ , where  $W_t$  is the weight at time  $t$  (in days),  $W_0$  is the initial weight and  $k$  is the decomposition coefficient (Webster & Benfield, 1986). We used an analysis of variance (two way ANOVA) to assess the differences between sample days during the study period, and the effect of vegetation on the decomposition rate. To explore the taxonomic data and functional similarity community composition, we used a non-Metric Multidimensional Scaling (NMDS) analysis, using the Bray-Curtis dissimilarity coefficient. The NMDS was performed with a biological matrix based on presence/absence data using the Jaccard index. We used a multivariate analysis of variance (PerMANOVA, 999 permutations) to assess taxonomic and functional similarity of community composition differences between incubation days and streams. We tested the environmental data and biological matrix (NMDS) relationship by multivariate non-parametric correlation (function 'envfit'). For all analyses, we used a significance level equal to  $p < 0.05$ . We used

the R software (R Development Core Team, 2013) with the 'vegan' package (Okansen et al., 2013).

## 3. Results

### 3.1. Landscape and limnological variables

During the experimental period, the water in the Campo river hydrographic basin streams (CV and CWV) showed a temperature of  $15.3 \pm 0.9$  °C (mean $\pm$ SD), was well oxygenated ( $7.9 \pm 12.4$  mg/L), and slightly acidic (pH  $6.5 \pm 0.3$ ) (Table 1). Similarly, the streams in the Cravo river hydrographic basin (CrV and CrWV) had water with a temperature of  $17.1 \pm 0.3$  °C, which was well oxygenated ( $8.0 \pm 0.9$  mg/L) and slightly acidic (pH  $6.7 \pm 0.2$ ). The limnological variables of the four streams were similar, when the hydrographic basins and the presence of riparian vegetation were compared (Table 2).

In general, in the Campo river hydrographic basin, the main land uses in the drainage area were exposed soil (23.8%), pasture (30.5%), agriculture (36.9%), and vegetation (8.6%) (Table 1). The main land uses in the Cravo river hydrographic basin were exposed soil (30.2%), pasture (34%), agriculture (23.7%), and vegetation (12%) (Table 1). The land use varied between hydrographic basins and the presence of riparian vegetation (Table 2).

### 3.2. Leaf mass loss of *C. xanthocarpa* and associated invertebrate fauna

At the end of the study period, the higher mass loss of *C. xanthocarpa* was observed in riparian vegetation streams in the Cravo river hydrographic

**Table 1.** Limnological variables (mean  $\pm$  SD) and land uses (%) of the streams in the Campo river hydrographic sub-basin and Cravo river hydrographic sub-basin, Southern Brazil.

Variables	Streams			
	CV	CWV	CrV	CrWV
Geographic Coordinates	27°42'58"S 52°14'43"W	27°43'28"S 52°12'43"W	27°43'13"S 52°17'11"W	27°45'37"S 52°15'57"W
Water temperature (°C)	14.2 $\pm$ 0.5	16.1 $\pm$ 0.2	17.4 $\pm$ 0.6	18.1 $\pm$ 0.8
pH	6.4 $\pm$ 0.5	6.9 $\pm$ 0.3	6.4 $\pm$ 0.3	6.3 $\pm$ 0.5
Total dissolved solids (mg/L)	0.033 $\pm$ 0.001	0.048 $\pm$ 0.001	0.025 $\pm$ 0.001	0.040 $\pm$ 0.001
Dissolved Oxygen (mg/L)	7.9 $\pm$ 0.7	6.5 $\pm$ 1.3	6.5 $\pm$ 0.8	8.6 $\pm$ 0.3
Conductivity ( $\mu$ S/cm)	166 $\pm$ 20	74 $\pm$ 9	38 $\pm$ 8	198 $\pm$ 20
Alkalinity (mg/L)	1.8 $\pm$ 0.1	2.5 $\pm$ 0.1	1.1 $\pm$ 0.1	1.9 $\pm$ 0.1
Total organic carbon (mg/L)	16.7	17.1	4.2	25.6
Total nitrogen (mg/L)	1.71 $\pm$ 0.02	2.92 $\pm$ 0.05	1.57 $\pm$ 0.01	1.76 $\pm$ 0.01
Drainage area (ha)	668.3	1073.2	199.4	1284.8
Slope (%)	16.5	14.6	12.2	16.9
Agriculture (%)	32.8	28.6	38.3	29.3
Exposure soil (%)	10.4	20.9	35.7	31.6
Pasture (%)	42.5	39.0	5.3	24.4
Vegetation (%)	14.4	11.5	20.7	14.8

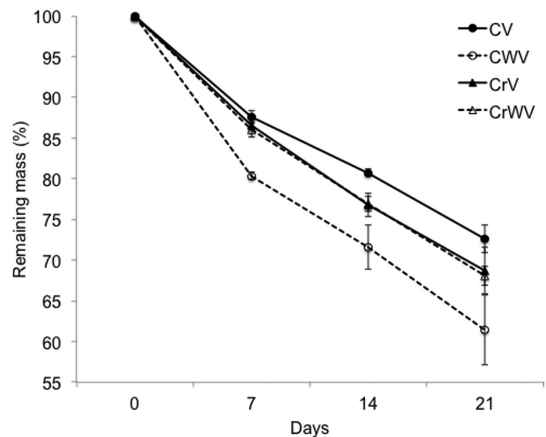
**Table 2.** PerMANOVA results of the environmental variables (limnological and land uses) and taxonomic and functional composition invertebrate fauna in the Campo river hydrographic basin and Cravo river hydrographic basin, Southern Brazil.

	df	SS	MS	F-value	p-value
<i>Limnological variables</i>					
Sub-basins	1	0.069	0.069	1.289	0.255
Presence of Vegetation	1	0.050	0.050	0.941	0.374
Residuals	9	0.485	0.053		
<i>Land use variables</i>					
Sub-basins	1	0.036	0.036	9.316	0.018
Presence of Vegetation	1	0.204	0.204	52.596	0.001
Residuals	9	0.034	0.004		
<i>Invertebrates taxonomic composition</i>					
Sub-basins	1	0.317	0.317	6.621	0.001
Presence of Vegetation	1	0.864	0.864	18.004	0.001
Time	1	0.041	0.041	0.868	0.468
Residuals	44	2.112	0.048		
<i>Invertebrates functional feeding group composition</i>					
Sub-basins	1	0.216	0.216	2.282	0.069
Presence of Vegetation	1	1.713	1.713	18.069	0.001
Time	1	0.044	0.044	0.468	0.729
Residuals	44	4.171	0.948		

basin (Figure 2). The percentage residual leaf weight in the Campo river was lower in the stream with riparian vegetation (80.4% of the remaining weight;  $k=-0.0148$  g/day). In contrast, leaves in the stream without riparian vegetation showed the lowest residual weight (71.1%;  $k=-0.0229$  g/day; Figure 2). Leaves from both streams of the Cravo river hydrographic basin showed a similar residual weight (77.4%,  $k=-0.0179$  g/day; 77%,  $k=-0.0181$  g/day; Figure 2).

During the experimental period, 2674 invertebrates were collected (Table 3). Of the total, 363 individuals were collected in the stream of the Campo river with riparian vegetation (density = 16 ind/g DW), and 713 individuals in the stream without vegetation (30 ind/g DW). In the Cravo river basin, 315 individuals were collected in the stream with riparian vegetation (9 ind/g DW) and 1,195 individuals in the stream without vegetation (62 ind/g DW). The most frequent functional feeding group (FFG) was scrapers (62%), followed by filterers (18.8%), shredders (15%), collectors (4.1%), and predators (0.1%). Moreover, there were a higher percentage of shredders in the two riparian vegetation streams.

The NMDS ordination showed segregation in taxonomic and functional feeding-group composition among streams with and without vegetation (Figure 3). The invertebrate taxonomic composition was different in the hydrographic basins and in the presence of riparian vegetation



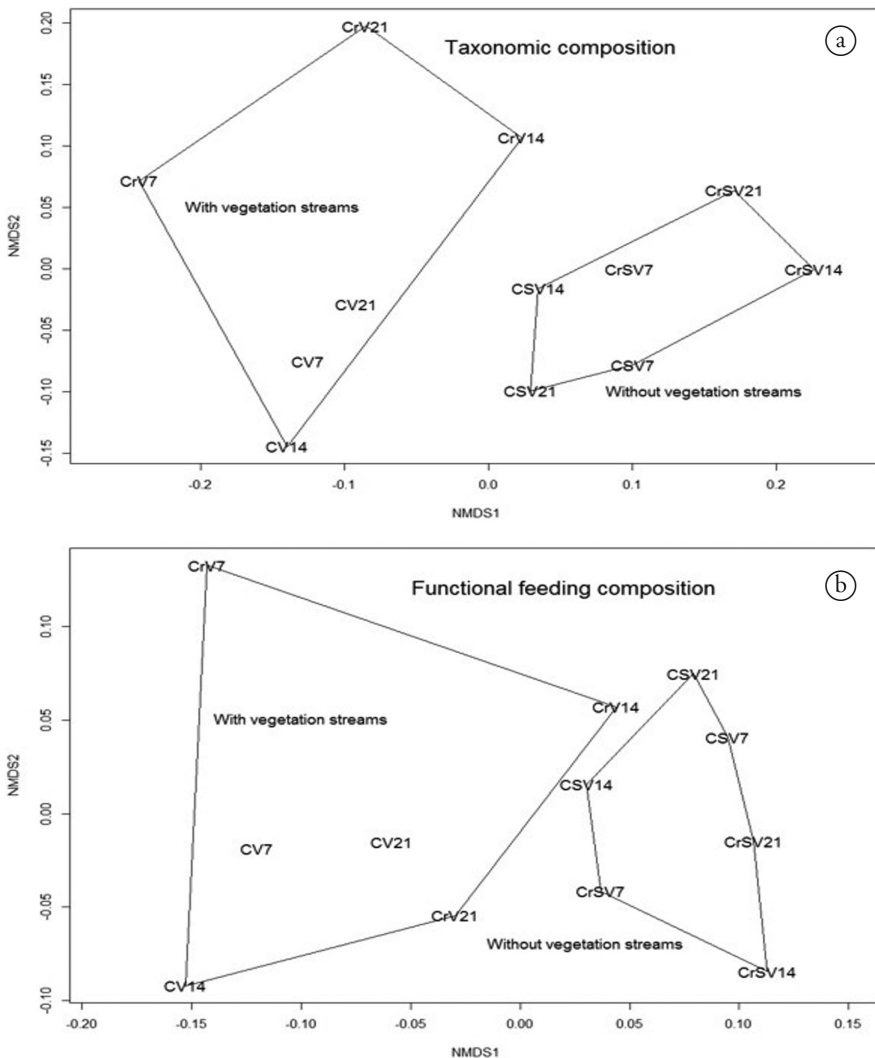
**Figure 2.** Remaining mass ( $\pm$  SE) of *Campomanesia xanthocarpa* leaf litter in the streams in the Campo river hydrographic basin and Cravo river hydrographic basin, Southern Brazil.

(Table 2). Moreover, the functional feeding-group composition differed only in the presence of riparian vegetation (Table 2). The sampling unit ordination of taxonomic composition was correlated with vegetation ( $r=0.94$ ,  $p=0.001$ ) and anthropogenic uses ( $r=-0.94$ ,  $p=0.001$ ), water temperature ( $r=0.88$ ,  $p=0.007$ ), and total organic carbon ( $r=-0.72$ ,  $p=0.002$ ). On the other hand, FFG composition only correlated with electrical conductivity ( $r=-0.99$ ,  $p=0.05$ ).

**Table 3.** Invertebrates abundance and functional feeding group (FFG) associated in *Campomanesia xanthocarpa* in the Campo river hydrographic basin and Cravo river hydrographic basin, Southern Brazil.

Taxa	FFG	Campo river		Cravo river	
		V	WV	V	WV
Simuliidae	Filterer	15	156	129	186
Caenidae	Scraper	0	0	0	7
Baetidae	Scraper	130	327	8	94
Leptohiphidae	Scraper	5	0	0	0
Leptophlebiidae	Scraper	2	4	0	37
Gripopterygidae	Collector/Shredder	166	13	69	119
Hydropsychidae	Scraper	37	204	101	641
Hydroptilidae	Collector/Scraper	3	4	3	96
Glossosomatidae	Scraper	0	0	0	1
Odontoceridae	Scraper	0	0	0	1
Elmidae	Collector/Shredder	3	3	2	12
Psephenidae	Scraper	0	1	0	0
Calopterygidae	Predator	0	1	1	1
Hydrobiidae	Scraper	2	0	1	0
Ancilidae	Scraper	0	0	1	0

V: with vegetation; WV: without vegetation.



**Figure 3.** NMDS ordination of (a) taxonomic and (b) functional invertebrate fauna composition associated in *Campomanesia xanthocarpa* leaf litter in the streams in the Campo river hydrographic sub-basin and Cravo river hydrographic sub-basin, Southern Brazil.

#### 4. Discussion

The limnological variables were similar among streams, although the land use varied between the streams in the two hydrographic basins. As expected, the riparian vegetation of the streams was the determining factor for this difference. Land uses affect the river basin characteristics, and alter the hydrological characteristics, substrate availability, water quality, and aquatic biota (Smith & Lamp, 2008). Changes in landuse (e.g., agricultural and pasture) have dramatically impacted aquatic ecosystems and are the major cause of riparian zone deforestation (Harding et al., 2006; Sensolo et al., 2012), with the low slope in both hydrographic basins favours agricultural landuse.

The leaves of *C. xanthocarpa* used in the litter bags experiments have a low nutritional quality (Tonin et al., 2014) and are essential to understand the effect of the riparian vegetation presence on the taxonomic and functional composition of the invertebrate community. The input of allochthonous organic matter into the stream increases the environmental heterogeneity, and provides a refuge and food source for aquatic biota (Biasi et al., 2013; König et al., 2014). However, in this study, we believe that the litter bags acted specifically as a refuge, because of the lower density of shredders than in the streams with vegetation. The removal of the riparian vegetation caused a reduction in the density of shredders in this study. Shredders are clearly very important in the transformation process of organic matter in forested and deforested streams (Masese et al., 2014).

The taxonomic composition of the invertebrate communities was more sensitive to variation in the streams. The change in land use (e.g., the removal of vegetation and increase in agricultural area) causes variations in community composition by excluding some less sensitive taxa (Sensolo et al., 2012). The water temperature and total organic carbon are influenced by agricultural activity. The replacement of vegetation by agriculture decreases the canopy and favours light penetration and an increase in water temperature. On the other hand, the higher sediment input contributes to an increase in the nutrients and organic matter concentration in streams (Nava et al., 2015), which might affect the availability of food (e.g., biofilm). Thus, the main carbon source for organisms becomes autochthonous instead allochthonous. This change in food for the benthic community decreases the shredder density and increases the density of scrapers.

Biological factors are important during the decomposition process and relate to colonisation by fungi, bacteria and invertebrates (Abelho, 2001). Microorganisms are responsible for conditioning the detritus, and make organic matter more palatable to invertebrates due to the degradation of refractory compounds (Bärlocher, 1992; Graça & Cressa, 2010). In this study, the experimental period might not have been favourable for colonisation by microorganisms such as shredders. Some studies have reported the importance of incubation time on the fragmentation of debris (Ligeiro et al., 2010; Biasi et al., 2013), which is essential for microbial conditioning, to accelerate the decomposition process (Encalada et al., 2010). In this study, these arguments are relevant, since the higher abundance of shredders is not reflected in the mass loss of detritus. Therefore, this mass loss might have been caused by the action of the stream, but the debris also constitutes a habitat that is extremely favorable to invertebrates. Furthermore, the additive effects of nutrients and/or shredder organisms on litter processing rates can be confounded by others factors (Huryn et al., 2002).

The remaining percentage of detritus mass in the streams did not affect the presence of riparian vegetation. In the Campo river hydrographic basin, leaves in the two streams had similar loss in mass, whereas in the Cravo river hydrographic basins, leaves in the stream without vegetation showed a lower residual mass. Among the many factors that might contribute to loss in mass of debris, water physico-chemical characteristics and leaf chemicals are the factors that affect the leaf leaching process. However, although the same plant species was used and the streams were limnologically similar, the most plausible explanation for the mass loss of debris is the flow action of the streams; water flow increases the mass loss, due to the removal of soluble compounds by leaching. In addition, flow stress the fibers that comprise the leaf structure and increases the leaching of compounds (Fonseca et al., 2013). The decomposition rates observed in this study can be considered to be rapid ( $k > -0.0173$ ), except for those of incubated debris in the stream without vegetation in the Campo river hydrographic basin, whose coefficient of decomposition is considered to be intermediate ( $-0.0041 > k > -0.0173$ ) according to the classification proposed by Gonçalves et al. (2013).

The presence of riparian vegetation was essential to modify the taxonomic and functional invertebrate composition. In this study, the density of shredders

was lower in the streams without vegetation. This is important when considering the drainage areas of small streams in areas with intensive agricultural practice. In small streams, the main source of energy is allochthonous material (Gonçalves Junior & Callisto, 2013). In addition, shredders are responsible for transforming coarse particulate organic matter to fine particulate organic matter (Vannote et al., 1980). Thus, nutrient cycling, especially that of carbon, is complex in streams that are under the influence of agriculture. Finally, this study showed that the riparian vegetation has an importance in the structure of invertebrate communities, but the land uses in drainage basin streams should be considered. The intensive occupation of the small streams drainage basin can cause serious changes on aquatic biota. A fragile riparian zone not support a high anthropogenic pressure from adjacent areas. Our study contributes to information for managers. The managers should to observe, not only the riparian zone of lotic environments conditions, but all land uses of the watershed.

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