







Plant species invasion effects on litter dynamics in subtropical streams

Efeitos da invasão de espécies vegetais na dinâmica da serapilheira em riachos subtropicais

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Cite as: Fontana, L. E. et al. Plant species invasion effects on litter dynamics in subtropical streams. *Acta Limnologica Brasiliensia*, 2020, vol. 32, e302.

Abstract: Aim: We evaluated the effect of the presence of *Hovenia dulcis* Thunb. (Rhamnaceae) in riparian zones on the organic matter dynamics of small subtropical streams. **Methods:** We conducted this study in three subtropical Atlantic Forest streams with different densities of *H. dulcis* in riparian vegetation located in southern Brazil. In each stream, we quantified the input of allochthonous organic matter for one year using buckets (area: 0.04 m²/bucket) suspended about 1 m from the streambed in three different sections (15 buckets/stretch = 45 buckets/stream). Monthly, the plant material retained in the buckets was collected individually, dried (40±5 °C/72 h), identified (native litter together and *H. dulcis* litter alone) and weighed. **Results:** The largest input of native organic matter occurred during the winter months (~55 g.m⁻²), ranging from ~31 g.m⁻² (summer) to ~46 g.m⁻² (spring) over the year. The input of *H. dulcis* organic matter was concentrated in the autumn (~56 g.m⁻²) and summer (~28 g.m⁻²), being scarce in the other seasons (~3 g.m⁻² in the spring and winter). Only the contribution of native organic matter was associated with precipitation. Contrary to that observed with native vegetation (input of organic matter related with rainfall), *H. dulcis* input was related to the phenology of the species, which is deciduous, with leaf fall strongly marked, occurring especially during the autumn. **Conclusions:** When present at high densities (dominant), the presence of *H. dulcis* in riparian stream vegetation makes the supply of allochthonous plant resources scarce at some periods of the year, altering the energy availability in these ecosystems and, potentially, the functioning of subtropical streams.

Keywords: Atlantic Forest; biological invasion; exotic species; riparian zones.

Resumo: Objetivo: Avaliamos o efeito da presença de *Hovenia dulcis* Thunb. (Rhamnaceae) em zonas ripárias sobre a dinâmica da matéria orgânica de pequenos riachos subtropicais. **Métodos:** Realizamos este estudo em três riachos subtropicais de Mata Atlântica com diferentes densidades de *H. dulcis* na vegetação ripária localizados no sul do Brasil. Em cada riacho quantificamos o aporte de matéria orgânica alóctone durante um ano utilizando baldes (área: 0.04 m²/balde) suspensos a cerca de 1 m do leito dos riachos em três trechos distintos (15 baldes/trecho = 45 baldes/riacho). Mensalmente, o material vegetal retido nos baldes foi recolhido individualmente, seco (40±5 °C/72 h), identificado (detritos nativos conjuntamente e detritos de *H. dulcis* isoladamente) e pesado. **Resultados:** Em média,



o maior aporte de matéria orgânica nativa ocorreu durante os meses de inverno (-55 g.m^{-2}), tendo oscilado entre -31 g.m^{-2} (verão) e -46 g.m^{-2} (primavera) ao longo do ano. O aporte de matéria orgânica de *H. dulcis*, entretanto, foi concentrada nos meses de outono (-56 g.m^{-2}) e verão (-28 g.m^{-2}), sendo escassa nas outras estações do ano (-3 g.m^{-2} na primavera e no inverno). Apenas a contribuição da matéria orgânica nativa foi associada à precipitação mensal. A presença de *H. dulcis* alterou a dinâmica da matéria orgânica alóctone nos riachos. Ao contrário do observado com a vegetação nativa (aporte de matéria orgânica relacionada com a pluviosidade) o aporte de *H. dulcis* esteve relacionado com a fenologia da espécie, que é decídua e com queda foliar fortemente marcada, ocorrendo especialmente durante os meses de outono. **Conclusões:** Quando presente em elevadas densidades (dominante), a presença de *H. dulcis* na vegetação ripária de riachos torna a oferta de recursos vegetais alóctones escassa em alguns períodos do ano alterando a disponibilidade de energia nesses ecossistemas e, potencialmente, o funcionamento de riachos subtropicais.

Palavras-chave: Mata Atlântica; invasão biológica; espécies exóticas; zonas ripárias.

1. Introduction

In small and shaded streams, the contribution of allochthonous organic matter is fundamental for the maintenance of aquatic communities because the primary productivity in these environments is limited by the shading generated by riparian vegetation (Graça, 2001; Neres-Lima et al., 2017). Within streams, the allochthonous organic matter is decomposed in a combination of chemical, physical and biological processes that result in its transformation and incorporation into the aquatic food chain (Webster & Benfield, 1986; Marks, 2019). Therefore, the conservation of riparian vegetation is essential for the maintenance of fundamental ecosystem processes (e.g. nutrient cycling) and aquatic communities (Graça, 1993; Abelho, 2001; Graça et al., 2015).

Allochthonous organic matter originated by riparian vegetation is composed of leaves ($\sim 60\%$ to 80% of the total), branches ($\sim 30\%$), and reproductive material (i.e. flowers and fruits) (Gonçalves Júnior & Callisto, 2013; Rezende et al., 2017). The contribution of allochthonous organic matter to streams occurs in different ways (Gonçalves Júnior et al., 2006) and is influenced by the structure and composition of the riparian vegetation (França et al., 2009; Rezende et al., 2017). Many plant species contribute to the entry of organic matter into tropical (França et al., 2009; Gonçalves Júnior et al., 2014; Rezende et al., 2017; Jijón & Molinero, 2019) and subtropical streams (Lisboa et al., 2015). Due to the specific phenology of tree species, the contribution of organic matter occurs in different periods, representing a constant energy input throughout the year in aquatic ecosystems (Lisboa et al., 2015; Rezende et al., 2017). Additionally, rainfall is considered the main climatic factor affecting the dynamics and input of organic matter on Brazilian streams (Lisboa et al., 2015; Rezende et al., 2016; Tonin et al., 2017).

The effect of exotic species presence on the ecosystems functioning is, currently, one of the main interest topics among ecologists (Sutherland et al., 2013). Many exotic species can occur associated with different aquatic ecosystems (Strayer, 2010). Specifically, the impact of exotic species on the functioning of streams, if existent, is still unclear and variable. In this context, the effect of the exotic species presence in the riparian vegetation of streams over, for example, ecosystem processes (e.g. decomposition of organic matter) and aquatic communities (e.g. aquatic invertebrates) are variable and can be positive, neutral or negative (Castro-Díez & Alonso, 2017). However, in the Iberian Peninsula, studies have shown that replacing native riparian vegetation (i.e. deciduous forest) with *Eucalyptus* monocultures alters the dynamics of organic matter in streams (Abelho & Graça, 1996; Pozo et al., 1997; Graça et al., 2002; Molinero & Pozo, 2004).

The exotic and invasive species *Hovenia dulcis* Thunb. (Rhamnaceae) has a high invasive capacity in native forest remnants (Meyer et al., 2012; Dechoum et al., 2015). Part of the successful establishment of *H. dulcis* is due to the allelopathic potential of its pseudo-fruits and leaves on seeds germination and seedling growth (Wandscheer et al., 2011) of native species (Ribeiro et al., 2019) and seeds dispersion by zoochory (pseudo-fruits are widely consumed, especially by mammals and birds) (Rocha et al., 2008; Hendges et al., 2012; Lima et al., 2015). The invasion of native forest remnants by *H. dulcis*, including protected areas and riparian zones in Brazil, has been reported in several studies (Zenni & Ziller, 2011; König et al., 2014; Dechoum et al., 2015; Lazzarin et al., 2015; Padilha et al., 2015). However, knowledge about the effect of *H. dulcis* on aquatic ecosystems is still scarce.

In this study, we sought to understand how the presence of *H. dulcis* in riparian zones affects the dynamics of organic matter in small order streams (i.e. streams in which allochthonous organic matter input is the main energy source). For this, we quantified the annual contribution of organic matter from native species and *H. dulcis* in subtropical streams. Our hypothesis is that the input of organic matter by native species is constant throughout the year and associated with periods of high rainfall while the input of organic matter by *H. dulcis* is concentrated at the period of the year associated with the deciduous foliar habit of the species.

2. Material and Methods

2.1. Study area

We performed this study in a forest remnant located in southern Brazil within a protected area (Parque Natural Municipal Mata do Rio Uruguai Teixeira Soares; 27°30'02"S and 51°57'11"W). This area is inserted in the Atlantic Forest biome, comprising a transition zone between Semideciduous Seasonal Forest and Atlantic Forest with Araucaria (Oliveira-Filho et al., 2015). The vegetation is characterized by an elevated diversity of tree species,

specially of the families Lauraceae (e.g. *Nectandra megapotamica* (Spreng.) Mez, *Ocotea puberula* (Rich.) Nees, *Ocotea odorifera* (Vell.) Rohwer), Fabaceae (e.g. *Parapiptadenia rigida* (Benth.) Brenan, *Dalbergia frutescens* (Vell.) Britton, *Inga marginata* Willd. and *Apuleia leiocarpa* (Vogel) J.F. Macbr.), Euphorbiaceae (e.g. *Gymnanthes concolor* (Spreng.) Müll. Arg., *Sebastiania commersoniana* (Baill.) L.B. Sm. & Downs and *Sebastiania brasiliensis* Spreng.), Sapindaceae (e.g. *Allophylus edulis* (A.St.-Hil, Cambess. & A. Juss.) Radlk, *Allophylus puberulus* (Cambess.) Radlk. and *Cupania vernalis* Cambess.) and Meliaceae (e.g. *Cedrela fissilis* Vell., *Trichilia elegans* A. Juss. and *Cabralea canjerana* (Vell.) Mart.) (Socioambiental Consultores Associados, 2012). Additionally, *H. dulcis* Thunb. (Rhamnaceae) is commonly found within this protected area (Socioambiental Consultores Associados, 2012). The region is part of the Serra Geral geological formation and the climate is classified as humid subtropical (Köppen classification), with an annual mean temperature of 18 °C and mean precipitation of 1900 mm (Alvares et al., 2013).

We selected three $\leq 2a$ order streams: PM (27°29'42" S and 51°56'9" W), SC (27°29'50" S and 51°56'8" W) and BO (27°30'5" S and 51°56'55" W) (approximate coordinates) (Figure 1). The selected

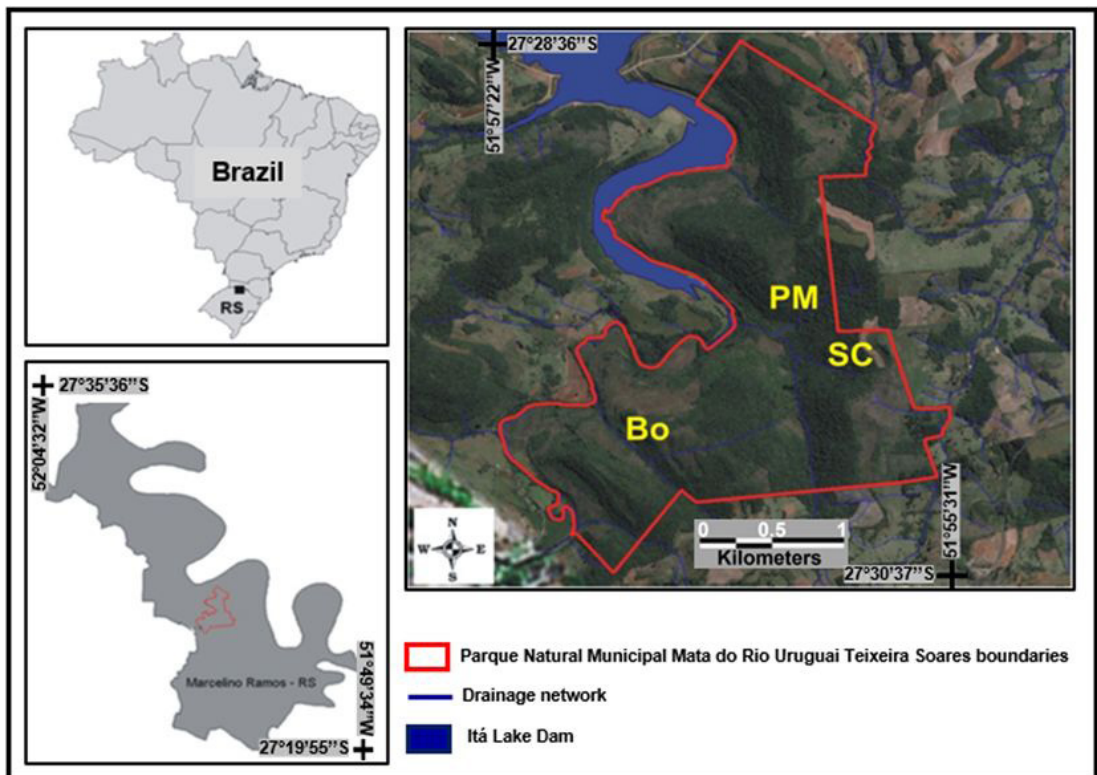


Figure 1. Geographic location of the three subtropical streams (PM, BO and SC) studied in southern Brazil. Prepared in collaboration with the Laboratório de Geoprocessamento e Planejamento Ambiental (URI Erechim).

streams are morphologically similar with widths ranging between 0.5 and 2.0 m and depths ≤ 0.5 m. Each stream has, at least, 50 m of riparian vegetation on each side bank. The riparian vegetation of each stream presents distinct densities of *H. dulcis*. We measured *H. dulcis* density in the riparian vegetation of each stream by counting the individuals with circumference at chest height ≥ 0.3 m from a two 10 x 50 m plots (one in each stream margin). The BO stream presented the high density of *H. dulcis* on riparian vegetation (360 ind.ha⁻¹) being followed by SC stream (230 ind.ha⁻¹), while PM stream presented the lower *H. dulcis* density (10 ind.ha⁻¹). The rainfall monthly data were obtained on a meteorological station located 8 Km of the studied area.

2.2. Allochthonous organic matter sampling

In each stream, we quantified the contribution of total native and exotic allochthonous organic matter over one year (monthly, from June 2016 to May 2017), using methods adapted from Gonçalves Júnior & Callisto (2013). We collected organic matter with buckets with perforated bottom for rainwater drainage (area 0.04 m² per bucket) suspended about 1 m above the streambed in three sections located 15-20 m apart from each other. On each section we suspended 15 buckets, totaling 45 buckets per stream. Every month, we collected all the plant organic matter from each bucket individually, conditioned it on plastic bags and transported all samples to the laboratory for drying (40 \pm 5 °C/72h), identification and weighing. From the collected plant material, we considered a pool containing all the fractions of organic matter (i.e. leaves, branches, fruits, flowers, and seeds). We considered the pool of plant fractions as the total content of allochthonous organic matter since the main idea this study was to evaluate the total contribution of allochthonous organic matter to streams and not the individual contribution of distinct fractions. We identified the organic matter of *H. dulcis* and weighed it separately, while organic matter from native species was counted all together.

2.3. Data analysis

We compared the total annual input from native species and *H. dulcis* organic matter of the streams studied together using a paired t-test. Similarly, we compared the input from native species and *H. dulcis* organic matter of each stream individually using paired t-tests. In addition, we analyzed the monthly variation in the input of organic matter

from native species and *H. dulcis* using an Analysis of Variance (two-way ANOVA), considering the streams (3 levels) and sampling months (12 levels) as factors. For the t-tests and ANOVA analyses, we tested the normality of the organic matter input data using a Shapiro-Wilk test. As the data were not in accordance with the normal distribution, we transformed them into $\ln(x + 1)$ in order to reduce variability and to better correspond to the assumptions for performing parametric tests. Finally, we evaluated the association between monthly precipitation and inputs of organic matter from native species and *H. dulcis* using Pearson's correlation. Statistical analyses were performed using the statistical software R (R Core Team, 2019), using the "stats" package.

3. Results

The total annual (sum of streams) input of native organic matter and *Hovenia dulcis*, regardless of the three streams studied, was similar (-1629 g.m⁻² and -873 g.m⁻², respectively; $t = 1.0$, $df = 2$, $p = 0.40$). However, the total input of native and *H. dulcis* organic matter varied between streams (Figure 2). In the PM stream, the total input of native organic matter was greater than that of *H. dulcis* (663.4 g.m⁻² and 14.3 g.m⁻², respectively; $t = -29.8$, $df = 35$, $p < 0.001$). Similarly, in the BO stream, the total native input was higher than that of *H. dulcis* (616.9 g.m⁻² and 327.0 g.m⁻², respectively; $t = -4.7$, $df = 35$, $p < 0.001$). Finally, in the SC stream, the input of *H. dulcis* was greater than the input of native organic matter (531.5 g.m⁻² and 348.8 g.m⁻², respectively; $t = -2.3$, $df = 35$, $p = 0.025$).

The input of native organic matter varied among streams over months (Table 1). On mean, the highest organic matter input of native species occurred in the winter months (mean \pm standard error; 55.5 ± 0.9 g.m⁻²; Figure 2A-2D; Table 2), while spring (46.2 ± 6.5 g.m⁻²), summer (31.2 ± 5.3 g.m⁻²) and autumn (44.3 ± 10.7 g.m⁻²) months presented organic matter input slightly lower (Figure 2A-2D; Table 2). Additionally, PM (55.3 ± 4.4 g.m⁻²; Figure 2A) and BO streams (48.3 ± 6.2 g.m⁻²; Figure 2C) presented the greatest input of native species organic matter. Similarly, *H. dulcis* organic matter input also differed between streams over the months (Table 1). On mean, the highest *H. dulcis* organic matter input occurred in the autumn (56.5 ± 27.0 g.m⁻²) and summer (28.1 ± 17.8 g.m⁻²) months (Figure 2A-2D; Table 2). However, on winter (2.0 ± 0.7 g.m⁻²) and spring (2.0 ± 0.8 g.m⁻²) months the *H. dulcis*

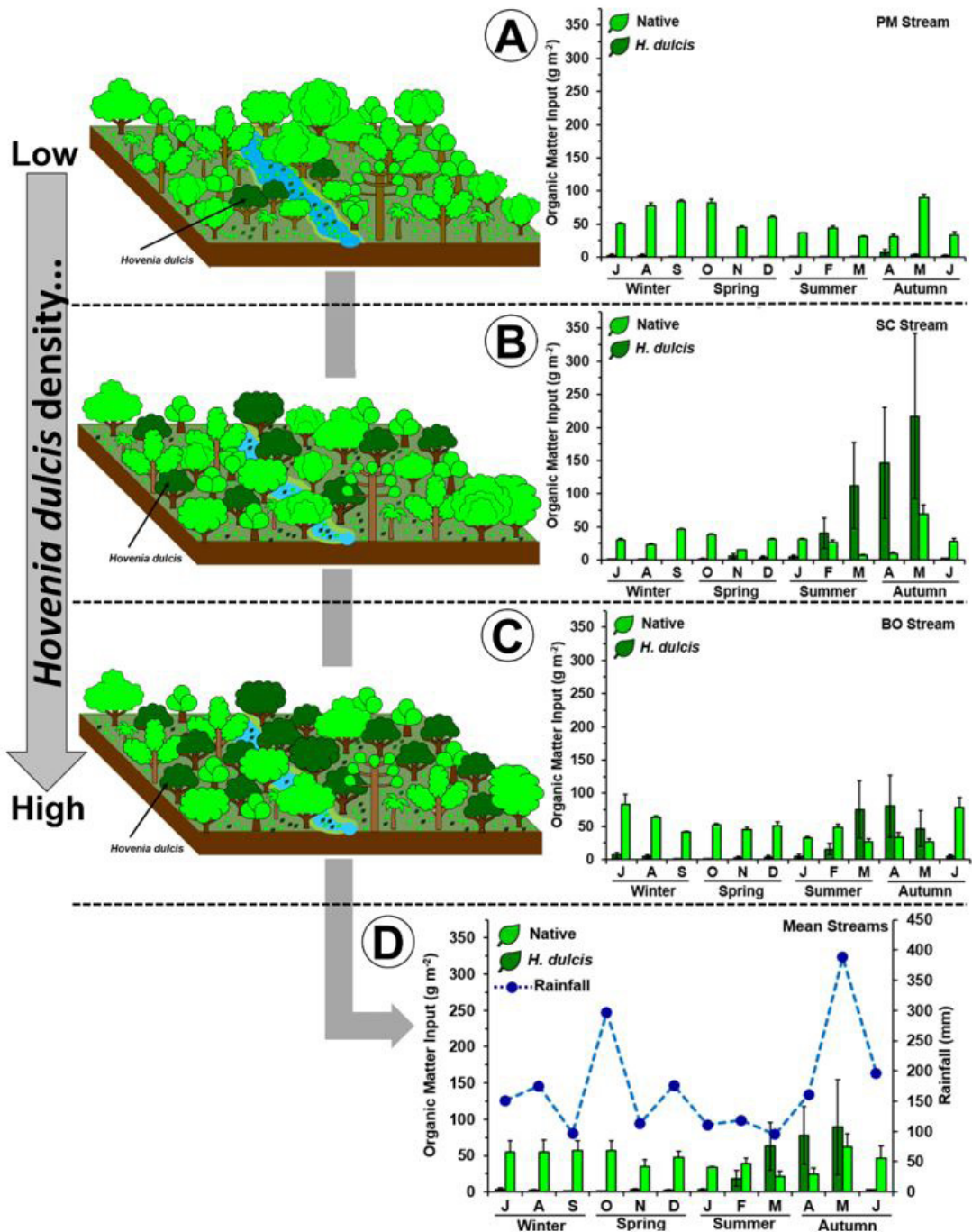


Figure 2. Monthly organic matter input (mean \pm standard error) of native species and *H. dulcis* in three subtropical streams with different *H. dulcis* densities on riparian vegetation: PM stream (A), SC stream (B) and BO stream (C). Also shown is the mean (\pm standard error) organic matter input by the three streams and the monthly rainfall observed during the study period (D). (Initial "J": July 2016).

organic matter input presented a strongly decrease (Figure 2A-2D; Table 2).

The input of organic matter of native species was positively related to rainfall ($r = 0.60$; $df = 10$; $p = 0.04$). On the other hand, we did not observe significant association between rainfall and *H. dulcis*

organic matter input ($r = 0.13$; $df = 10$; $p = 0.68$) (Figure 2D).

4. Discussion

The annual similar input of organic matter of native species and of *Hovenia dulcis* in the study area

Table 1. Result of Analysis of Variance (two-way ANOVA) on organic matter input of native species and *H. dulcis* among streams and months. Degrees of freedom (df), sum of squares (SS), mean of squares (MS), test value (F) and test significance (p) are described.

	df	SS	MS	F	p
Native input					
Stream	2	12.8	6.4	13.9	<0.001*
Month	11	19.9	1.8	3.4	<0.001*
Stream:Month	22	11.5	0.5	1.1	0.808
Residuals	72	33.1	0.5		
<i>Hovenia dulcis</i> input					
Stream	2	50.7	25.4	63.4	<0.001*
Month	11	127.7	11.6	29.0	<0.001*
Stream:Month	22	61.3	2.8	6.9	<0.001*
Residuals	72	28.8	0.4		

(*) Significant values for $p < 0.05$.

Table 2. Results of Tukey test carried *a posteriori* of the Analysis of Variance (two way ANOVA) to verify the difference of native and *H. dulcis* organic matter input between the months.

Native	Jan	Feb	Mar	Apr	May	June	July	Aug	Sep	Oct	Nov	Dec
Jan	-											
Feb	1.000	-										
Mar	0.315	0.239	-									
Apr	0.240	0.177	1.000	-								
May	0.999	0.999	0.077	0.053	-							
June	1.000	1.000	0.310	0.236	0.999	-						
July	0.999	0.999	0.048*	0.032*	1.000	0.999	-					
Aug	0.992	0.997	0.020*	0.013*	0.999	0.993	1.000	-				
Sep	0.916	0.956	0.005*	0.003*	0.998	0.919	0.999	0.999	-			
Oct	0.941	0.971	0.006*	0.004*	0.999	0.944	0.999	0.999	1.000	-		
Nov	1.000	0.999	0.485	0.391	0.998	1.000	0.994	0.960	0.794	0.839	-	
Dec	0.999	0.999	0.046*	0.031*	1.000	0.999	1.000	1.000	0.999	0.999	0.993	-
<i>H. dulcis</i>												
	Jan	Feb	Mar	Apr	May	June	July	Aug	Sep	Oct	Nov	Dec
Jan	-											
Feb	0.031*	-										
Mar	<0.001*	0.258	-									
Apr	<0.001*	0.001*	0.820	-								
May	<0.001*	0.015*	0.993	0.999	-							
June	1.000	0.023*	<0.001*	<0.001*	<0.001*	-						
July	1.000	0.054	<0.001*	<0.001*	<0.001*	1.000	-					
Aug	0.999	0.003*	<0.001*	<0.001*	<0.001*	0.999	0.998	-				
Sep	0.131	<0.001*	<0.001*	<0.001*	<0.001*	0.162	0.080	0.493	-			
Oct	0.277	<0.001*	<0.001*	<0.001*	<0.001*	0.328	0.185	0.734	0.999	-		
Nov	0.999	0.005*	<0.001*	<0.001*	<0.001*	0.999	0.999	1.000	0.407	0.649	-	
Dec	0.999	0.008*	<0.001*	<0.001*	<0.001*	0.999	0.999	1.000	0.312	0.549	1.000	-

(*) Significant values for $p < 0.05$.

reveals the effect of the exotic species on organic matter dynamic in streams. The presence of *H. dulcis* in the riparian zone of streams has altered the allochthonous organic matter dynamic to streams, which may cause changes in these ecosystems functioning. These arguments are corroborated by the comparative results between the organic matter input of native species and *H. dulcis* in each stream individually. In SC and BO streams, *H. dulcis*

density in the riparian zone was greater than in PM stream. Thus, the high abundance of *H. dulcis* greatly influenced the organic matter input in the streams, especially in SC stream, where *H. dulcis* input was significantly greater than native input. These results are ecologically relevant because, although higher when compared with native organic matter input in the SC stream, the *H. dulcis* organic matter input it was available in large quantities

in only 3 or 4 months of the year. In this sense, a riparian zone with predominance of *H. dulcis* may generate an excessive organic matter input in a short period of the year, depleting the aquatic ecosystem from an allochthonous energy source for long periods.

The contribution of native organic matter was, on mean, slightly higher in the winter months and, then, in the spring and autumn months and was related to the rainfall. During the study period the rainfall was well distributed throughout the year, with peaks (>200 mm.months⁻¹) in the October and May which may have contributed to the increase in the native organic matter input in the spring and autumn. In addition to rainfall, the pattern of native organic matter dynamics observed may also be related to the riparian vegetation composition, which, in the study region, is characterized by the occurrence of perennial, semi-deciduous and deciduous species (Loregian et al., 2012; Mélo et al., 2013). Thus, the native organic matter input was possibly related to the leaf habit and to a greater leaf fall of deciduous and semi-deciduous species (especially in the autumn / winter transition) or the replacement of senescent leaves for the appearance of new leaves in the growing season (especially in the winter / spring transition) (Athayde et al., 2009; Turchetto & Fortes, 2014; Dick et al., 2015; Capellesso et al., 2016).

The contribution of organic matter of native species observed in this study was similar to the vertical contribution observed in another subtropical Atlantic Forest stream (Lisboa et al., 2015) and in a tropical stream of Cerrado (Leite et al., 2016), but was lower than that observed in tropical Atlantic Forest stream (Gonçalves Júnior et al., 2014). Although studies with this approach are relatively scarce in subtropical climates, our results highlight the influence of rainfall and temperature decrease on the contribution of terrestrial organic matter to subtropical streams.

The highest contribution of *H. dulcis* organic matter occurred during autumn and late summer. This result was clearly related to the deciduous foliar habit of this species, which has the highest leaf fall in autumn (Carvalho, 1994; Schumacher et al., 2008). Additionally, the high organic matter input of *H. dulcis* observed at the end of the summer (i.e. March) is related to the early occurrence of days with low temperatures like the days of autumn, anticipating the fall of the leaves. In Brazil, the literature indicates that the leaf fall period of *H. dulcis* is concentrated in the months of April-

May to August (i.e. autumn and winter) (Carvalho, 1994), but studies evaluating the production of *H. dulcis* litterfall are very scarce. The results of this study indicated that during the autumn, the contribution of *H. dulcis* was 27% higher than the contribution of native species. Unlike what is sometimes observed with *Eucalyptus* (low organic matter input when compared to the native deciduous organic matter input) (Graça et al., 2002; Molinero & Pozo, 2004), *H. dulcis* effects at the organic matter dynamic are mainly related to the greater organic matter input during a restricted period of the year.

The occurrence of increased organic matter input (e.g. native or exotic) only in a short period of the year can affect the functioning of these ecosystems. In a broader context, *H. dulcis* has a high invasive capacity, especially in impacted forests remnants with high canopy openings, less species diversity and simplified communities structure (Dechoum et al., 2015; Lazzarin et al., 2015). Thus, in riparian zones with greater invasiveness of *H. dulcis*, especially where this species becomes dominant, *H. dulcis* presence may affect the organic matter dynamic of small streams because of its short period of abundant foliar fall. On the other hand, in streams that have native riparian vegetation, we observed a substantial presence of native organic matter during the whole study period, despite the greater contribution during winter and spring, which guarantees an “always present” energy supply in these streams (Lisboa et al., 2015).

Our results demonstrate that the invasion of *H. dulcis* in riparian zones may affect the dynamics of organic matter in streams, especially where this species is dominant on riparian vegetation. While the contribution of native species organic matter was relatively constant throughout the year, the contribution of *H. dulcis* occurred predominantly in a short period of the year (especially in autumn). This timely supply of organic matter makes the availability of allochthonous plant resources scarce for most of the year, compromising the maintenance of key ecosystem processes (e.g. nutrient cycling) and aquatic communities in these streams.

Acknowledgements

The authors are grateful to the Chico Mendes Institute for Biodiversity Conservation for the issuance of a Collection License (ICMBio; Collection License No. 53164-1), to the Parque Natural Municipal Mata do Rio Uruguai Teixeira Soares for the assistance during the field activities and

the ENGIE Tractebel Energia for the provision of rainfall data. LEF is grateful for the scholarship grant (CAPES-PROSUP) linked to the Graduate Program in Ecology of the URI Erechim. LUH receives financial support from CNPq (nº 421632/2016-0) and CNPq grants (nº 305203/2017-7).

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Received: 29 September 2019

Accepted: 17 August 2020

Associate Editors: Fabiana Schneck.