



The effects caused by the replacement of native riparian forest with *Eucalyptus sp.* on the benthic macro-invertebrate community

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Abstract

The replacement of native riparian forests with exotic plants, especially *Eucalyptus sp.* can cause changes in ecological processes of small orders rivers. The leaves of *Eucalyptus sp.* have lower nutrients concentration, higher hardness and secondary metabolites which inhibit the decomposing micro-organisms and benthic macro-invertebrate colonization, which are the connection between the dead organic matter and the rest of the food chain. Thus the aim of the study was to test the following hypothesis: leaf decomposition rate will be lower in the substrate composed of *Eucalyptus sp.* leaves, when compared to the substrate composed of native tree species leaves; the benthic macro-invertebrates diversity will be lower in the substrate composed of *Eucalyptus sp.* leaves than the one composed of native tree species leaves. Sixty litter bags were used in the experiment, 30 of them were filled with 5 g of *Eucalyptus sp.* leaves and the other 30 with a mixture of native trees leaves. From the results it was concluded that leaf decomposition rates were similar for both substrates, substrate composed of native trees leaves and substrate composed of *Eucalyptus sp.* leaves; the benthic macroinvertebrates diversity was lower in the substrate composed of *Eucalyptus sp.* leaves when compared to the substrate composed of native tree species leaves.

Key words: Litter decomposition, exotic plants, litter bag, degradative succession, diversity.

Introduction

In small second-order streams, where the amount of solar energy entering the water is a limiting factor, the main source of resources and energy that contributes to increased productivity is the organic matter from riparian vegetation^{1,2}. As a consequence, the allochthonous organic matter and its decomposition allow supporting important food chains³ as well as the cycling of nutrients and other chemical elements in such environments.

The decomposition rates of leaf litter vary considerably among the diverse vegetation species. This process is influenced by the physical and chemical characteristics of the litter and the environment^{4,5} as well as the micro and macro fauna of aquatic environments. Such relationship influences directly the breakdown of organic matter performed by fungi, bacteria and invertebrates (Gessner *et al.*⁶).

The aquatic ecosystems have suffered impacts due to the replacement of native riparian vegetation by exotic species (Richardson *et al.*⁷). Such changes affect the amount and quality of the leaf litters^{8,9}, causing alterations in the aquatic ecological processes, especially those related to leaf decomposition and nutrients cycling processes performed by the microbial communities and benthic detritivorous macro-invertebrates^{10,11}.

In Brazil in 2007, the total forest area planted with exotic species was 5,844,000 ha, and in 2011 it was 6,516,000 ha, and

the *Eucalyptus sp.* was the most common species. In 2007 the area planted with *Eucalyptus sp.* totaled 3,970,000 ha and in 2011 4,874,000 ha, an increase of 20% over the period¹².

Some authors have reported the negative impact of *Eucalyptus sp.* leaves on the decomposition micro-organisms; it reduces the colonization rate of filamentous fungi and yeasts^{13,14}. Sampaio *et al.*¹⁵ and Callisto *et al.*¹⁶ also demonstrated that for the benthic macro-invertebrates the consumption of *Eucalyptus sp.* leaves causes the breakdown of the trophic chain and reduction of the organisms diversity. In laboratory, the shredder *Tipula lateralis* Meig (Diptera: Tipulidae) rejected the eucalyptus leaves when mixed with leaves of other species, besides the fact that they do not grow on a diet based exclusively on *Eucalyptus sp.* leaves¹⁷.

Some of the problems associated with the decomposition and consumption of eucalyptus leaves by benthic macro-invertebrates involve physical characteristics such as hardness and the presence of cuticle^{18,19}, besides the chemical composition of the plant, which has low concentration of nitrogen and phosphorous²⁰, high contents of tannins and dense oil glands that may act as a toxic substance^{21,22} and cause a low decomposition rate.

Therefore, based on the premise that *Eucalyptus sp.*, when compared to other native species, has a greater amount of

chemical compounds that inhibit the biological activity, this study has the objective of testing the following hypotheses: the foliar decomposition rates are lower in substrates containing leaves of *Eucalyptus sp.* than in substrates comprised of leaves of native plant species; the diversity of benthic macro-invertebrates will be lower in the substrate containing leaves of *Eucalyptus sp.* than in the substrate with leaves of native plant species.

Materials and Methods

Study conditions: The experiment was conducted at coordinates 25°9'10" S; 53°56'50" W, altitude 590 m, in western Paraná, from November to December 2007. The climate in the region, according to Köppen's classification, is mesothermal subtropical super humid, with average annual precipitation of 1800mm, hot summers, infrequent frosts, and a higher rainfall concentration during the summer months, but no definite dry season. Average temperature is 20°C and relative air humidity is 75% (Caviglione *et al.* ²³). The second-order stream has an average width of four meters and average depth of 0.4 meters. The riparian forest is preserved and consists of seasonal semi-deciduous vegetation having an average width of 25 meters on each side of the stream.

The experiment: Leaves of native plants were collected from the riparian vegetation along the stream, and leaves of *Eucalyptus sp.* were collected from a neighboring reforestation area. All indigenous plants were identified, and the collected leaves, together with the *Eucalyptus sp.* leaves were dried in forced-air ovens at a temperature of 60°C for 24 hours.

Sixty litter bags were made of high-density, 20 mm-polyethylene screen, sewn with nylon thread to form 250 × 200 mm bags. Later, five grams of dried leaves were introduced into each litter bag, resulting in 30 litter bags containing substrate of *Eucalyptus sp.* (SFE) and the other 30 containing substrate of native tree leaves (SFN). The native plants used in SFN are shown in Table 1.

Table 1. Native tree species used in the litter bag substrate of native tree leaves.

Family	Scientific name
Meliaceae	<i>Cedrella fissilis</i> Vellozo
Myrtaceae	<i>Campomanesia xanthocarpa</i> O. Berg
Tiliaceae	<i>Luehea divaricata</i> Martius & Zuccarini
Caesalpinaceae	<i>Bauhinia forficata</i> Benth
Bignoniaceae	<i>Tabebuia Alba</i> (Cham.) Sandwith
Mimosoideae	<i>Inga uruguensis</i> Hooker & Arnott
Aquifoliaceae	<i>Ilex paraguariensis</i> Saint Hilaire
Lauraceae	<i>Ocotea puberula</i> (Rich.) Nees
Apocynaceae	<i>Aspidosperma polyneuron</i> Muell. Arg.

The litter bags, sealed and numbered, were transported in plastic bags and incubated into the streambed using stones to fix them. The substrates were placed alternately along the longitudinal and transverse axis of the stream to keep environmental heterogeneity (Fig. 1).

Four litter bags of each type of substrate (SFE and SFN) were collected in intervals of 3, 7, 17, 24, 30, 37 and 46 days of incubation. The litter bags were removed from the stream using a 0.25 mm-mesh macro-invertebrates collector (D-frame net), and then were put into plastic bags and transported to the laboratory.

In the laboratory, the substrates were screened in a set of 1 mm, 0.50 mm and 0.25 mm screens to separate the leaves from the

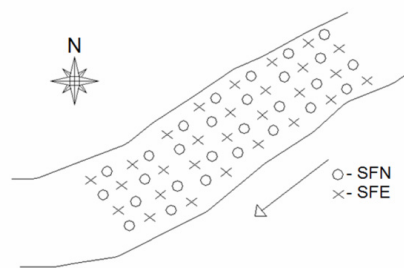


Figure 1. Arrangement of the leaf litter bags on the streambed. SFN - substrate comprised of native tree leaves; SFE - substrate comprised of *Eucalyptus sp.* leaves. The arrow indicates the stream flow.

macro-invertebrates. The individuals found were preserved in a 70% ethanol solution and then identified to family level, when possible, with a stereoscopic microscope ^{24,25}.

The leaves were washed, then dried in a forced-air oven at 60°C to reach constant weight, and weighed in a precision scale in order to determine loss of foliar mass.

Data analysis: The foliar decomposition coefficients (k) were determined by adjusting the weight loss data to the negative exponential model $W_t = W_0 e^{-kt}$, as described by Webster and Benfield ²⁶, where W_t is the remaining weight, W_0 is the initial weight, t is time (days), and k is the decomposition coefficients.

A Bray-Curtis similarity analysis with the substrate type and incubation time, the number of taxa rate, number of individuals, dominance, diversity and Simpson's dominance, Shannon-Wiener's diversity, Margalef's diversity, equitability indices and Berger-Parkers dominance were calculated according to the procedures implemented in the PAST 2.15 (Hammer *et al.* ²⁷) software.

Results and Discussion

The k values at the end of the experiment were nearly 0.1015 d⁻¹ for SFE and 0.1061 d⁻¹ for SFN.

The results found for SFE are approximately 8 to 12 times higher than those found by Galizzi and Marcheses ²⁸ and Sampaio *et al.* ²⁹, respectively.

The results obtained for SFN are difficult to compare with other studies due to the following situations: the breakdown of the mixed litter can be altered by additive effects resulting from the species interaction or determined by some dominant species in the process ^{30,31}. Decomposition of the species used in the SFN has not been sufficiently studied. Only two genera have previous studies. Ardón *et al.* ³² studied the decomposition of the species *Luehea seemanii* Triana and Planch, and found 0.033 d⁻¹, three times lower than SFN. Moretti *et al.* ³³ studied the genus *Ocotea sp.* and found 0.009 d⁻¹, 12 times lower than SFN.

The foliar mass loss at the end of the experiment was higher in SFN when compared to SFE, though it was only 4% higher. However, comparison between the weight losses of the two substrates during collection shows interesting results (Table 2 and Fig. 2).

In the first sample collection, during the period of higher precipitation (96 mm), three days of incubation, the weight loss for SFE was of 17% and for SFN it was of 29.3%. At the sixth collection, the second period of higher precipitation (50 mm), with 37 days of incubation, the weight loss for SFE was of 11%, and for SFN of 18%. These differences in mass loss between the

Table 2. Weight loss of the substrate containing leaves of *Eucalyptus sp.* and the substrate containing leaves of native trees.

T	P	PME	PMN	PMAE	PMAN
3	96	17.00	29.30	17.00	29.30
7	70	18.05	17.70	35.05	47.00
17	16	7.75	7.00	42.80	54.00
24	15	2.80	3.00	45.60	57.00
30	8	9.65	8.00	55.25	65.00
37	50	11.00	18.00	66.25	83.00
46	43	15.70	3.00	81.95	86.00

T: Time (days); P: Precipitation in sampling intervals (mm); PME: Percentage of mass loss in the substrate of *Eucalyptus sp.* leaves at each interval; PMN: Percentage of mass loss of the substrate of native tree leaves at each interval; PMAE: percentage of accumulated mass loss in the substrate of *Eucalyptus sp.*; PMAN: percentage of accumulated mass loss of the substrate of native tree leaves.

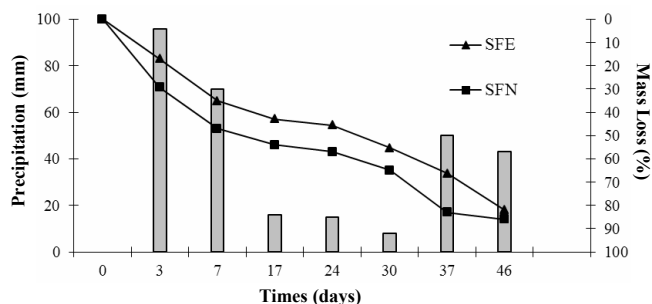


Figure 2. Precipitation and foliar weight loss of the substrate composed of leaves of *Eucalyptus sp.* (SFE) and of the substrate composed of indigenous plant leaves (SFN) during the sampling period.

two substrates were due to the fact that the *Eucalyptus sp.* leaves have thick cuticles, which diminish the mass loss resulting from abrasion with water^{18,19}. However, at the seventh sample collection, after 46 days of incubation, despite the occurrence of a rainfall of 43 mm, the weight loss in SFE corresponded to 15.7%, and in SFN to 3%. Therefore, it is clear that the cuticle effect was inexistent at the 46th day of incubation.

Since higher rainfall levels cause more mass loss, one can conclude that leaching and fragmentation caused by the stream flow speed and turbulence accelerate leaf weight loss⁴. This is evidenced by Pearson's correlation between rainfall and the percentage of weight loss in each sampling period. Correlations were positive for SFE (0.85) and for SFN (0.89). Paul and Meyer³⁴ noticed that the decomposition rate of leaves of *Rhododendron maximum* L. increased four times during the rainy season.

Table 3 shows that 1622 specimens of benthic macro-invertebrates were found, representing 30 taxa. Out of this amount, 542 (33.4%) are representative of the family Chironomidae, 446 (27.5%) of the family Elmidae and 223 (13%) of the class Bivalvia. The number of taxa found is similar to that found by Moretti *et al.*³⁵, but the number of individuals was twice as high. Gonçalves Jr. *et al.*³⁶ also reported high densities of the Chironomidae and Elmidae. It is worth noting that the family Chironomidae is often numerically dominant in freshwater environments in different climatic regions³⁷.

Specimens of the families Corydalidae, Gripopterygidae, Libellulidae, Psephenidae and Tipulidae were found only in SFN. In laboratory experiments, Canhoto and Graça^{17,18} noticed that specimens of the family Tipulidae did not colonize SFE. However, individuals of the families Leptophlebiidae, Hydrophilidae, Notonectida and Tabanidae were found only in SFE. Representatives of the families Hydrophilidae, Notonectidae, and Tabanidae are

Table 3. Taxons and number of individuals found in the substrate comprised of leaves of *Eucalyptus sp.* and in the substrate containing leaves of native trees.

Class	Order	Family	N	SFN	SFE
Insecta	Ephemeroptera	Beatidae	41	17	24
		Caenidae	46	20	26
		Leptophlebiidae	1	0	1
		Hydropsychidae	19	16	3
		Leptoceridae	18	12	6
		Limnephilidae	34	23	11
	Trichoptera	Philopotamidae	49	38	11
		Polycentropodidae	3	1	2
		Calopterygidae	11	6	5
	Odonata	Coenagrionidae	4	2	2
		Libellulidae	1	1	0
	Plecoptera	Gripopterygidae	1	1	0
		Perlidae	6	5	1
	Coleoptera	Elmidae	446	241	205
		Gyrinidae	8	2	6
		Hydrophilidae	4	0	4
		Psephenidae	1	1	0
		Ceratopogonidae	42	13	29
		Chironomidae	542	213	329
	Diptera	Empilidae	3	2	1
		Simuliidae	12	9	3
		Tabanidae	2	0	2
		Tipulidae	3	3	0
Megaloptera	Corydalidae	1	1	0	
Hemiptera	Notonectidae	1	0	1	
Crustacea	Decapoda	-	62	31	31
Bivalvia	-	-	233	119	114
Gastropoda	-	-	10	3	7
Oligochaeta	-	-	8	2	6
Hirudinea	-	-	8	7	1

N: Total number of individuals found; SFN: Number of individuals found in the substrate composed of leaves of native trees; SFE: Numbers of individuals found in the substrate composed of *Eucalyptus sp.* leaves.

predators, and those of the family Leptophlebiidae are Collector / Gatherers (Poff *et al.*³⁸), so neither the SFE substrate nor the SFN have any direct effect upon them. Moreover, these families were found in low densities, what may indicate that they were captured because of the drift, suspended on the water column, and not necessarily colonizing the substrate³⁹.

The community descriptive indices (Table 4) show that the benthic macro-invertebrates succeeded in colonizing SFN after the first collection at the third day of incubation until the fifth collection in 30 days of incubation. In the following samples, the indices show that in SFE the community was more balanced. This is because the slow leaf loss in SFE provided a larger area and a more resistant shelter for the benthic macro-invertebrates that take nutrients from the water column⁴⁰; at the 37th day the leaf weight in SFE was nearly 23% higher than in SFN. SFN had a larger number of taxa per weigh unit from the first to the sixth sample collection, and only in the last collection the number of taxa per unit was higher in SFE (Table 4).

The community reached its climax between the third collection after 17 days of incubation and the fifth collection after 30 days of incubation; in the 4th collection after 24 days incubation, it was found the greatest number of taxa in both substrates (Table 4). These results can be explained by the theory of degradative ecological succession of Begon *et al.*⁴¹. This theory sustains that the substrates colonization begins with some generalist species, and then specialized species begin to colonize until the climax. After the climax, the community diversity begins to decline, because the substrate reaches an advanced stage of decay and can no longer feed the community.

Table 4. Descriptive indices of the community in the substrate composed of *Eucalyptus sp.* leaves and in substrate composed of native tree leaves during the experiment period of time.

	S	N	D	D _s	H'	A	J	D	S/M
<i>Eucalyptus spp</i> 3 days	8	30	0.331	0.669	1.446	2.058	0.696	0.500	1.93
Native 3 days	11	35	0.159	0.841	2.064	2.813	0.861	0.257	3.11
<i>Eucalyptus spp</i> 7 days	9	28	0.214	0.786	1.846	2.401	0.840	0.393	2.77
Native 7 days	11	50	0.186	0.814	1.916	2.556	0.799	0.280	4.15
<i>Eucalyptus spp</i> 17 days	12	147	0.346	0.654	1.525	2.204	0.614	0.551	4.20
Native 17 days	17	249	0.290	0.710	1.737	2.900	0.613	0.498	7.39
<i>Eucalyptus spp</i> 24 days	17	190	0.277	0.723	1.764	3.049	0.623	0.458	6.25
Native 24 days	21	158	0.214	0.786	2.041	3.951	0.671	0.329	9.77
<i>Eucalyptus spp</i> 30 days	15	234	0.345	0.655	1.437	2.566	0.531	0.457	6.70
Native 30 days	18	247	0.283	0.717	1.840	3.086	0.637	0.494	10.29
<i>Eucalyptus spp</i> 37 days	9	50	0.203	0.797	1.782	2.045	0.811	0.300	5.33
Native 37 days	6	35	0.295	0.705	1.401	1.406	0.782	0.429	7.06
<i>Eucalyptus spp</i> 46 days	10	47	0.273	0.727	1.703	2.338	0.739	0.468	11.08
Native 46 days	6	115	0.387	0.614	1.144	1.054	0.639	0.530	8.57
Total <i>Eucalyptus spp</i>	25	831	0.242	0.759	1.878	3.570	0.583	0.396	-
Total native	26	789	0.196	0.804	2.071	3.748	0.636	0.305	-

S: Number of taxons; N: Number of individuals; D: Dominance; D_s: Simpson's diversity and dominance; H': Shanon-Wiener's diversity; α: Margalef's diversity; J: Equitability; d: Berger-Parker's dominance; S/M: number of taxa per gram of substrate.

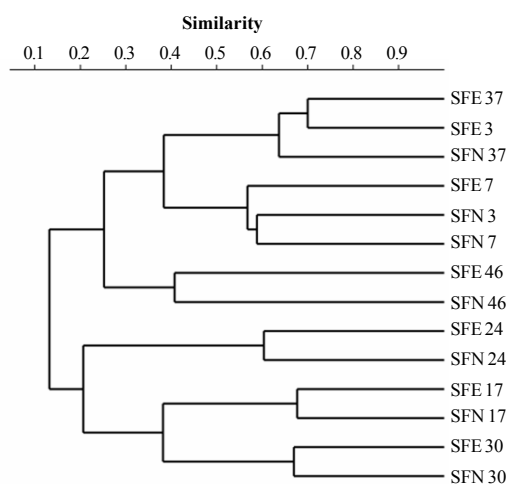


Figure 3. Bray-Curtis similarity analysis between the type of substrate and incubation time. SFN: substrate composed of native tree leaves; SFE: substrate composed of *Eucalyptus sp.* leaves. The numbers following the abbreviations indicate the incubation time (days).

Bray-Curtis similarity analysis (Fig. 3) grouped the collection that presented the greatest richness and diversity. It shows that in the third collection, after 17 days of incubation, the difference between SFN and SFE was of 31%. At the 4th collection, after 24 days of incubation, the difference was of 40%, and in the fifth collection, 30 days incubation, difference was of 32%. This difference may be attributed to the kind of substrate. SFE has low concentration of nitrogen and phosphorous, high levels of tannins and dense oil glands that can act as a benthic macro-invertebrates repellent, thus decreasing the community diversity¹⁸⁻²².

Conclusion

The rate of foliar decomposition was the same for both the substrate composed of *Eucalyptus sp.* leaves and the substrate composed of indigenous tree leaves.

Diversity of benthic macro-invertebrates was lower in the substrate composed of leaves of *Eucalyptus sp.*, when compared to the substrate containing leaves of native species.

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