Invertebrate Biology 125(4): 306–313.

DOI: 10.1111/j.1744-7410.2006.00062.x

Community structure of land flatworms (Platyhelminthes, Terricola): comparisons between Araucaria and Atlantic forest in Southern Brazil

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Abstract. Land flatworms (Platyhelminthes: Terricola) are sensitive to environmental changes and might be used as biological indicators of the degree of disturbance of their habitats. Nevertheless, studies on the structure of land flatworm communities are rare. In the present study, we investigated the terrestrial flatworm communities in two types of ombrophilous forest, Atlantic forest and Araucaria forest, aiming to answer the following questions: (1) Is the community structure of the two types of ombrophilous forest different? (2) Are there differences, regarding community structure, among distinct fragments of Araucaria forest? (3) Are there indicators of edge effects in these communities? The study site, the National Park of Aparados da Serra, is mainly covered, at high altitudes, by fragments of Araucaria forest immersed in a matrix of open areas (grasslands) and, in low altitudes, by a continuous Atlantic forest. We conducted monthly surveys in the two types of forest with three replications per forest type over a period of 2 years. Results indicated that: (1) community structure is clearly distinct between the two types of forest, as well as among fragments of the Araucaria forest; (2) there are no apparent edge effects; (3) the diversity indices of the two types of ombrophilous forest are not significantly different (Atlantic forest—$H' = 2.87$, Araucaria forest—$H' = 2.55$; $p > 0.05$); and (4) there are significant differences in diversity indices between two fragments of Araucaria forest ($H' = 2.92$ and 2.47; $p < 0.001$). The following factors could affect community structure: type of vegetation, degree of human disturbance, and abiotic factors, such as pH and temperature.

Additional key words: diversity, planarians, ombrophilous forest

Land flatworms (Platyhelminthes: Terricola) are top predators within the soil ecosystem, preying on other invertebrates (Du Bois-Reymond Marcus 1951; Jones et al. 1995; Ogren 1995; Sluys 1999; Leal-Zanchet & Carbayo 2001; Carbayo & Leal-Zanchet 2003). These flatworms have restricted locomotion capacity over long distances; hence there are many endemic species (Sluys 1995). They are sensitive to the humidity and temperature of their environment (Froehlich 1955; Sluys 1998, 1999; Winsor et al. 1998). Boag et al. (1998a) considered soil pH in relation to the distribution of earthworm species in Scotland, and linked this to the possible spread of the vermivorous flatworm *Arthurdendyus triangulatus* (Dendy 1895). Owing to these biological characteristics and their habitat requirements, land planarians are possible indicator taxa in biodiversity and conservation studies (Sluys 1998). Carbayo et al. (2001, 2002), comparing the diversity of land planarians in habitats with different degrees of human disturbance, show that this diversity is inversely related to the degrees of habitat disturbance, and suggested that certain species are useful as biological indicators.

Although the highest species richness of land planarians is registered in areas that were originally covered by the southeastern Atlantic forest (Sluys 1998), which occurred in the Brazilian states of São Paulo, Rio de Janeiro, and Santa Catarina, the southern portions of this biome also harbor a large number of species (Leal-Zanchet & Carbayo 2000, 2001; Carbayo & Leal-Zanchet 2001, 2003; Carbayo et al. 2001, 2002; Froehlich & Leal-Zanchet 2003). For these communities, however, many questions on their ecology are still unanswered. At present, there is no published information on possible differences in community composition influenced by distinct forest

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types. Similarly, although their habitat requirements suggest that these animals are potentially susceptible to edge effects (alterations in microclimate characteristics associated with forest edges [Murcia 1995]) and also other effects related to habitat fragmentation, there is no published information focusing on such effects.

Habitat fragmentation has been severely affecting the Brazilian Atlantic forest, which originally ranged from the southern Brazilian state of Rio Grande do Sul to the state of Paraíba, in the north of Brazil (Câmara 1983). It has been largely destroyed, with only an estimated 5–7% of the original area still having any type of arboreal cover (Fonseca 1985; Tabarelli et al. 2005). The Brazilian Atlantic Forest shows a large number of strictly protected areas; however, there are many problems regarding their extent, distribution, and management (Tabarelli et al. 2005). The so-called political dominion of the Atlantic forest includes several phytophysiognomically distinct types of forest (Fundação SOS Mata Atlântica & INPE 1993). These forest types include the typical coastal Atlantic forest (dense ombrophilous forest) and the Araucaria (*Araucaria angustifolia* [Bert.] Kuntze Araucariaceae) forest (mixed ombrophilous forest).

The Araucaria forest occurs at higher altitudes between latitudes 24° and 30°S, primarily at altitudes 500–1400 m in southern Brazil, and in isolated islands between 18° and 24°S at elevations 1400–1800 m in southeastern Brazil (Hueck 1953; Rambo 1994). The typical coastal Atlantic forest occurs in southern Brazil as a 100–200-km-wide belt in the coastal lowlands, near the border between the states of Santa Catarina and Rio Grande do Sul, and on the slopes of the "Serra Campos Gerais" mountains at elevations up to 1000 m (Hueck 1972; Behling et al. 2004). Transition between both forest types in southern Brazil may occur gradually or abruptly, depending on the slope of the ecotonous areas.

The Araucaria forest has been strongly affected mainly by logging and agricultural land-use practices (Auler et al. 2002; Behling et al. 2004). The process of fragmentation of this forest may negatively influence the populations of land planarians, being responsible for decreases in diversity as well as possibly causing local extinction of species.

In this study we compared communities of the terrestrial flatworms in two types of ombrophilous forests, namely the Atlantic forest and the Araucaria forest. We focused on the following questions: (1) Is the community structure of the two types of ombrophilous forest different? (2) Are there differences in community structure among fragments of the Araucaria forest? (3) Are there indicators of edge effects in these communities?

Methods

The National Park of Aparados da Serra (NPAS) (Fig. 1) is located in the northeast of the state of Rio Grande do Sul, southern Brazil (29°05–29°15S to 50°00–50°15W). Together with the neighboring National Park of Serra Geral it constitutes one of the most important protected areas of Rio Grande do Sul, covering an area of 27,000 ha. The study was conducted in the two main forest types of the park: (1) mixed ombrophilous forest (MO) with araucaria pine, *Araucaria angustifolia* (Bert.) Kuntze, called Araucaria forest, occurring in fragments surrounded by dry and wet grassland fields, at an altitude of ~900 m, and (2) continuous Atlantic forest (AF) located at ~40 m above sea level. The climate of the region is warm temperate (subtropical) and humid without marked dry periods (Nimer 1989), with an annual rainfall varying between 1500 mm and 2250 mm year^{-1} (IBAMA, www.ibama.gov.br).

Samplings were conducted during the day by an experienced collector (I.A.F.) between March 2000 and March 2002. We sampled three fragments of Araucaria forest, presenting sizes of 24.9 ha (MO1), 3.6 ha (MO2), and 1.6 ha (MO3), respectively, and three randomly selected areas in the continuous Atlantic forest. For each area we randomly selected two areas to establish 49-m^2 plots (7 × 7 m). One of the plots was placed on the forest edge and the other in the interior of the area (50 m away from the forest edge). The position of both plots was altered monthly (also by random selection). The edge was estimated as an extension of 50 m from the ecotone between forest and grassland in the direction of the forest. We searched for flatworms inside the plots by direct sampling in soil litter, under and inside fallen logs and branches, and under rocks. After inspection, the branches, logs, and rocks were returned to their original position so as to avoid alteration of the soil fauna (Ball & Reynoldson 1981; Winsor 1997). Each plot was sampled once a month, thus totalling 48 samplings. For identification purposes, we used techniques described by Leal-Zanchet & Carbayo (2001).

During sampling we registered the temperature (to the nearest 0.1°C), using an analog thermometer on the soil. Soil pH was measured by calculating the mean of two samples composed of ten subsamples of superficial soil randomly collected in the studied areas. These samples were collected in the winter and summer of 2001, and analyzed at the Geochemistry

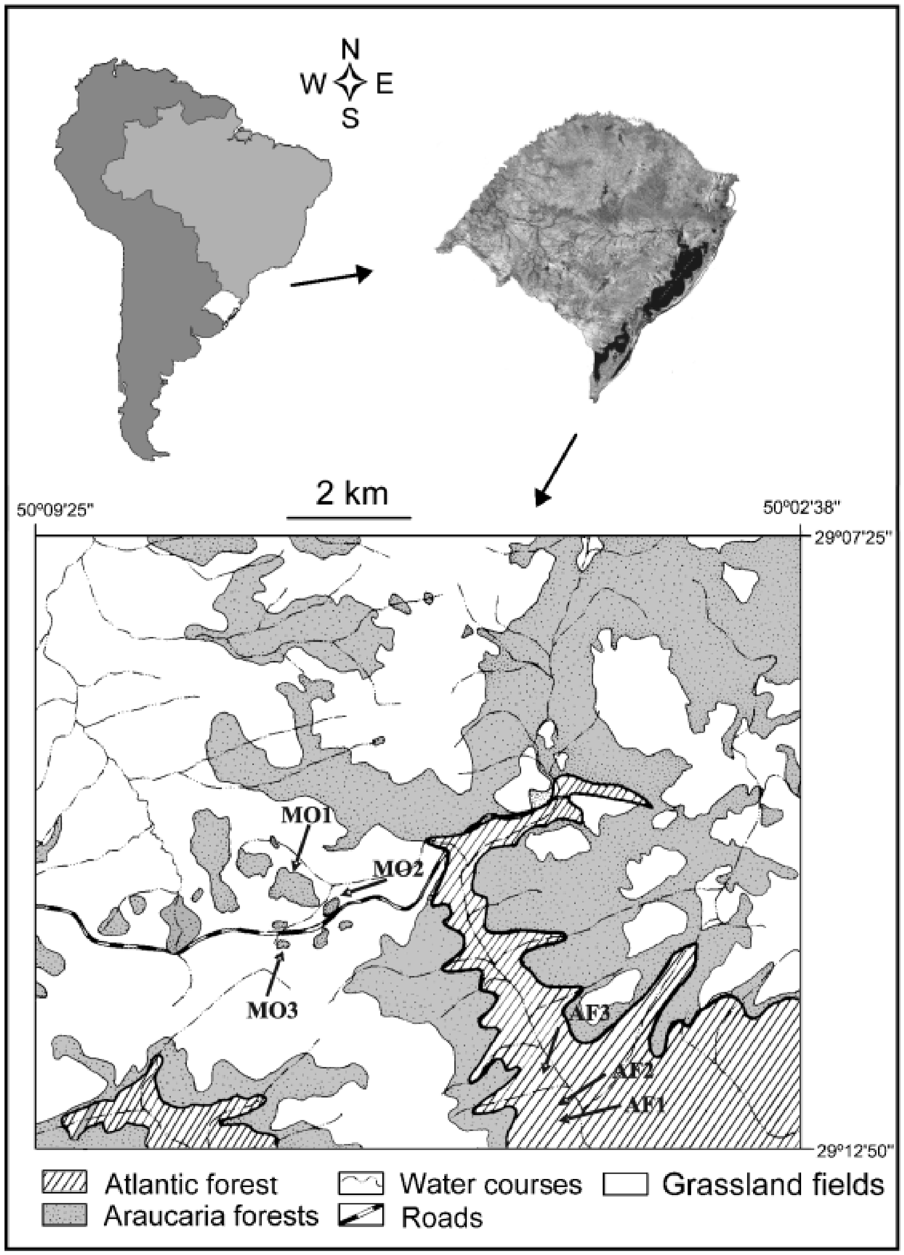

Fig. 1. Location of the study area (National Park of Aparados da Serra) in southern Brazil. MO: Araucaria forest; AF: Atlantic forest; numbers indicate the studied MO fragments and areas inside the continuous AF.

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We estimated the available food supply for the land flatworms in the Araucaria forest fragments by using 300-mL plastic cups filled with water and 4% formaldehyde (8:1) as pitfalls. These pitfalls were buried in the soil with their opening placed flush to the ground and were left for 72 h per month. We placed 48 pitfalls on each fragment of the Araucaria forest between February 2001 and March 2002. The collected invertebrates were identified at the level of order and divided into size classes, followed by measurement of their biomass. We considered as an index of the available monthly biomass of invertebrates the overall biomass at each fragment per month divided by the number of cups placed at the fragment at that month.

We calculated the Shannon–Wiener diversity index (H'; see Krebs 1989) and tested for significant differences between diversity indices of pairs of habitats (modified t-test for the Shannon–Wiener diversity index; see Zar 1999). To evaluate community grouping in the different areas, we used cluster analysis and correspondence analysis (CA) using PC-ORD software (MjM Software Design, Gleneden Beuch, OR, USA) (McCune & Mefford 1999) and Systat 11.0 for Windows software (Systat Software Inc., Richmond, OR, USA).

Results

We found 712 individuals, 686 of which could be identified, belonging to 19 species and five genera (Table 1). Habitat selection was noted for certain species. *Geoplana ladislavii* von Graff 1899, *Notogynaphallia graffi* Leal-Zanchet & Froehlich 2002, *Geoplana* sp. 6, and *Pasipha* sp. 1 were restricted to the Atlantic forest (AF). Ten other species were restricted to the Araucaria forest (MO) (Table 1). Five species, i.e., *Cephaloflexa bergi* (von Graff 1899), Geoplanidae 2, Geoplanidae 3, *Geoplana* sp. 3, and *Geoplana* sp. 5, were restricted to one fragment of MO, *Geoplana* sp. 3 being the most abundant in fragment MO2 (Table 1).

The overall richness of MO (15 species) was greater than that of AF (nine species) (Table 1). Shannon–Wiener diversity was greater in MO ($H' = 2.87$) than in AF ($H' = 2.55$), but there was no significant difference ($p > 0.05$). However, we detected significant differences between fragment MO1 and fragment MO3 (Table 1; $p < 0.001$). The similarity between MO and AF was 45.2%.

Cluster analysis demonstrated that the land flatworm communities of each fragment of MO are distinct (Fig. 2), but there is high similarity between species composition on the edge and in the interior of each fragment. In AF, community composition was similar among the studied areas, without a clear distinction among edge and interior areas.

CA ordination analysis showed a clear differentiation between communities of the two types of forest (Fig. 3). *Notogynaphallia graffi, Pasipha* sp. 1, *G. ladislavii,* and *Geoplana* sp. 6 are the most important species for AF, and *G. franciscana* Leal-Zanchet & Carbayo 2001, Notogynaphallia sp. 1, *Geoplana* sp. 1, and Geoplanidae 4 are the most important ones for MO. Specimens of *C. bergi, Geoplana* sp. 4, and Geoplanidae 2 do not affect the community structure of the two types of forest.

During the sampling period, the mean temperatures on the soil were 18°C (minimum of -1°C and maximum of 26°C) in the MO areas and 22°C (minimum of 8°C and maximum of 38°C) in the AF areas. Soil pH was 4.0 for the MO areas and 6.8 for the AF areas.

Table 1. Abundance, richness, and diversity (Shannon–Wiener, H') of land planarian species in the two types of forest, Araucaria forest (MO) and Atlantic forest (AF), in the National Park of Aparados da Serra, Brazil. MO and AF numbers indicate the studied fragments and areas.

		Areas					
Species	Abbreviations	MO1	MO2	MO3	AF1	AF2	AF3
Choeradoplana iheringi	Chi	39	61	53	5	14	12
Cephaloflexa bergi	Ceb	2	0	0	0	0	0
Geoplana franciscana	Gef	16	16	105	0	0	0
G. josefi	Gej	41	5	14	5	7	5
G. ladislavii	Gel	0	0	0	7	10	2
Notogynaphallia graffi	Gem	0	0	0	6	2	2
Geoplana sp. 1	Ge1	0	4	2	0	0	0
Geoplana sp. 2	Ge2	15	2	12	11	20	14
Geoplana sp. 3	Ge3	0	9	0	0	0	2
Geoplana sp. 4	Ge4	18	18	39	4	2	0
Geoplana sp. 5	Ge5	4	0	0	0	0	0
Geoplana sp. 6	Ge6	0	0	0	2	0	0
Geoplanidae 1	Gp1	2	0	2	0	0	0
Geoplanidae 2	Gp2	2	0	0	0	0	0
Geoplanidae 3	Gp3	2	0	0	0	0	0
Geoplanidae 4	Gp4	0	2	2	0	0	0
Notogynaphallia sp. 1	N1	4	11	15	0	0	0
Pasipha sp. 1	Pa1	0	0	0	0	2	0
Pasipha sp. 2	Pa2	21	0	14	0	0	0
Total		166	128	258	40	57	37
Richness		12	9	10	7	7	6
Diversity (H')		2.92	2.39	2.47	2.66	2.35	2.13

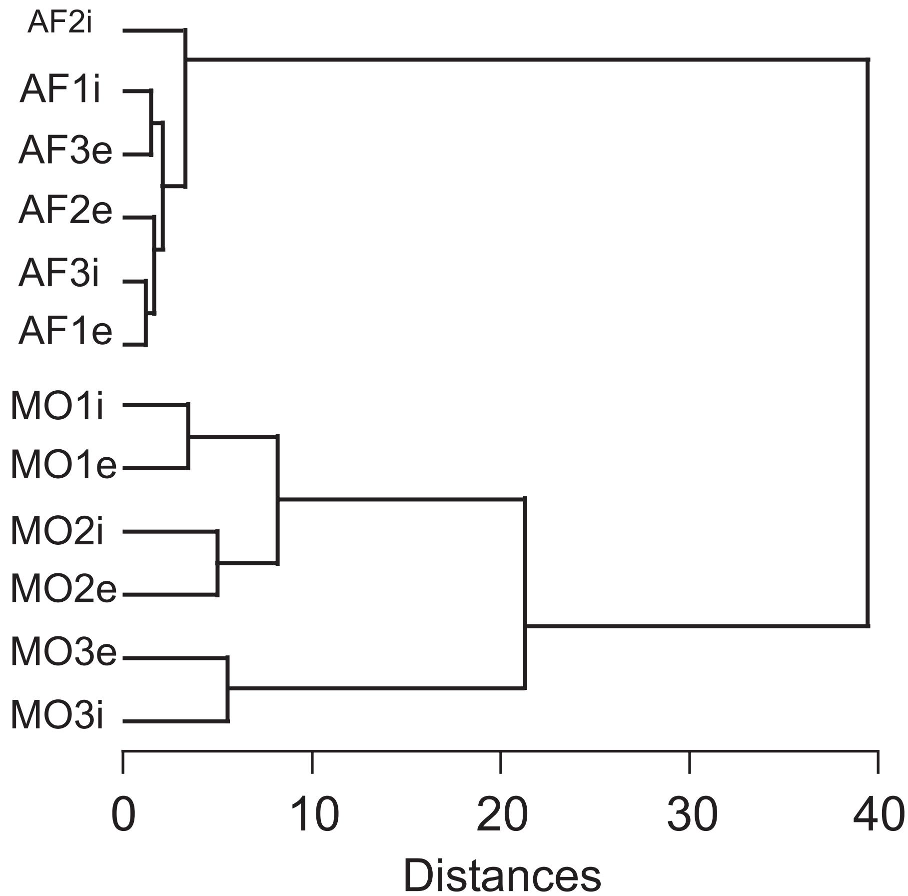

Fig. 2. Results of cluster analysis of edge (e) and interior (i) plots in the two types of forest, Araucaria forest (MO) and Atlantic forest (AF), in the National Park of Aparados da Serra. Numbers indicate the studied MO fragments and areas inside a continuous AF.

Discussion

Several authors (Springett 1976; Alford et al. 1998; Boag et al. 1998a,b; Sluys 1998; Winsor 1998) indicated that pH, depth, temperature, texture and humidity of the soil, presence of refuge in the soil, and prey abundance potentially influence the occurrence and distribution of land flatworms. In the present study, CA showed that communities of the Araucaria forest and Atlantic forest of the NPAS are different, which might be the result of the diverse thermal amplitudes and differences in the pH of the soil registered for the two types of habitats. The distinct separation between the two communities of land flatworms in these two types of habitats was mainly caused by two groups of species, each characteristic of each type of habitat, indicating a dominant fauna for each type of forest.

Regarding habitat disturbance, the studied areas showed different kinds of environmental alterations, which might affect terricolan diversity. In the area of the Atlantic forest, the highest degree of habitat disturbance occurred in AF1 rather than in the areas AF2 and AF3, the latter two presenting more new trees and a higher density of bush plants (I.A. Fick, unpubl. data). However, in the three studied AF areas, the exotic rodent *Rattus rattus* (E.M. Vieira, unpubl. data) occurs, being an indicator of human disturbance. In the fragments of Araucaria forest, the intermittent presence of cattle occurs, thus hindering plant regeneration and increasing soil compaction, as well as the incidence of solar light. Soil compaction probably hinders vertical migrations (Carbayo et al. 2002), which is the usual behavior of land flatworms (Boag et al. 1998a; Jones et al. 1998). The occurrence of domestic pigs was also observed in the studied site, but with less frequency (three times during the sampling period). Winsor (1998) commented that wood destruction caused by wild pigs might be a negative long-term factor for the conservation of land planarian fauna. Comparing four types of habitats with different degrees of disturbance, Carbayo et al. (2002) showed that terrestrial flatworm diversity is inversely related to the degree of habitat disturbance (selective logging and reforestation with exotic pines, in this case).

Comparing terricolan richness and diversity of the Araucaria forest in the present study (NPAS) with

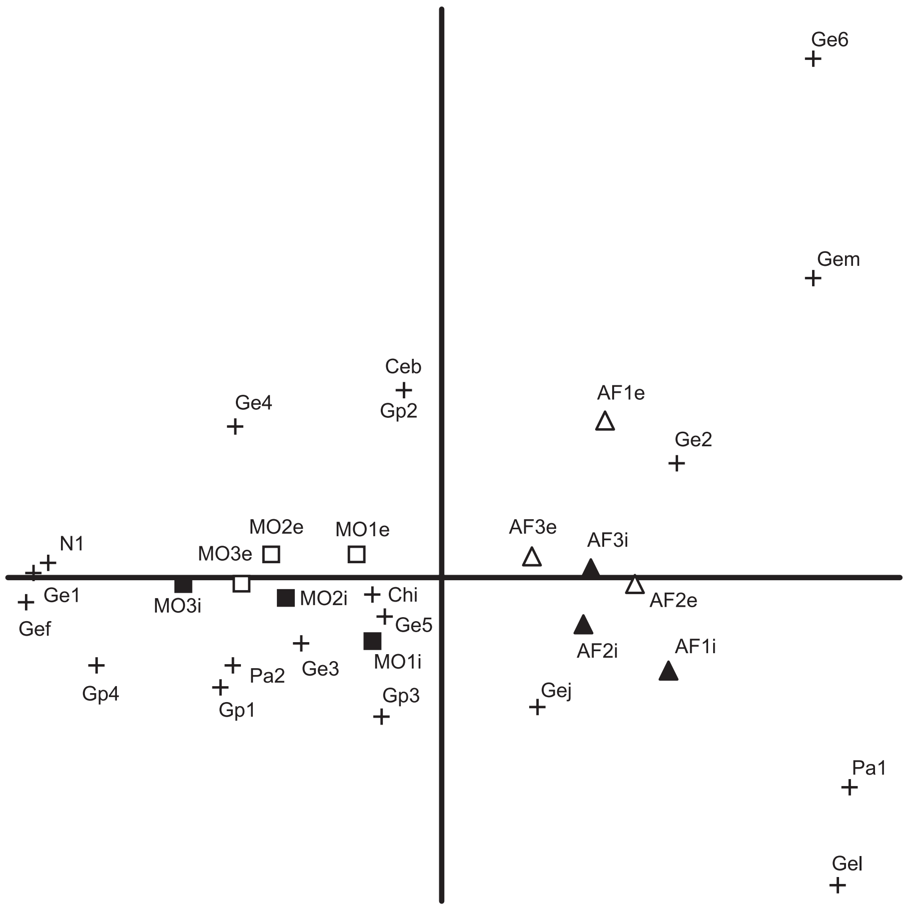

Fig. 3. Results of correspondence analysis ordination of the terrestrial flatworm communities of the National Park of Aparados da Serra. The triangles and quadrates indicate, respectively, the areas of Atlantic forest and the fragments of Araucaria forest. The filled and empty figures indicate, respectively, the interior (i) and edge (e) of the fragments or areas. The crosses indicate the species; abbreviations as in Table 1.

those of the same type of habitat in the National Forest of São Francisco de Paula (NF-SFP), studied by Carbayo et al. (2002), both studies with a similar sampling effort, we found higher richness and diversity for NF-SFP, showing 22 species registered for this type of habitat (with and without selective araucaria pine logging), with a diversity index of $H' = 3.272$ for an area without araucaria pine logging and $H' = 3.467$ for an area with selective araucaria pine logging (Carbayo et al. 2002). The areas of Araucaria forest of NPAS and NF-SFP are different with respect to both structure and habitat disturbance. NF-SFP shows an almost continuous area of Araucaria forest presenting a mosaic distribution, with plantations of native and exotic plants (especially araucaria pine and *Pinus* LINNAEUS), the main habitat disturbances being selective araucaria logging as well as the planting of exotic plants.

Some species, considered to be generalist concerning habitat use and tolerance, such as *G. ladislavii* (Carbayo et al. 2002) and *N. graffi,* were only registered in the AF areas of NPAS and not in the MO fragments. Both species are widely distributed in the northeast region of Rio Grande do Sul (A.M. Leal-Zanchet, unpubl. data), *G. ladislavii* also having been registered more northerly in the state of Santa Catarina (Froehlich 1959). Both species were registered for areas of Araucaria forest and plantations of araucaria in NF-SFP, members of *G. ladislavii* being the most abundant at that site (Carbayo et al. 2002). The absence of these species in the MO fragments of NPAS might be explained by the fact that this area has been burned during the last 50 years (A. Backes, pers. comm.), thus causing local extinction of these species, as they are unable to recolonize the fragments of Araucaria forest. Furthermore, the surrounding grassland areas of the NPAS are burned annually. According to Springett (1976), frequent rotational fuel reduction burning leads to simplification of the flatworm fauna.

Some species, such as *Choeradoplana iheringi* VON GRAFF 1899 and *G. franciscana,* occurred in great abundance in the three fragments of Araucaria forest, these being more generalist species, whereas others, such as *C. bergi, Geoplana* spp. 3 and 5, and Geoplanidae 2 and 3, being more specific, were only registered for one fragment. *Choeradoplana iheringi,* indicated by Carbayo et al. (2002) as a biological indicator of more disturbed habitats, was the most abundant species in NPAS, being registered for the two types of forest that we studied. According to Carbayo et al. (2002), *G. franciscana,* registered in NF-SFP for areas of Araucaria forest and araucaria plantations, prefers well-preserved habitats. Members of this species were the second most abundant of species in NPAS, occurring in the three fragments of Araucaria forest. Such a pattern could indicate the strong association of the species with araucaria formation. Besides the lower richness of NPAS, when compared with that of NF-SFP, there was the dominance of two species, *C. iheringi* and *G. franciscana,* individuals of which were among the four most abundant species registered for the NF-SFP.

Cluster analysis demonstrated that there was apparently no edge effect in communities of the studied site, as communities from the interior of the areas are more similar to those of the edge than to those from the interior of others in each kind of habitat. Considering that the forest edge receives a high incidence of solar light, due to the ecological requirements of land planarians, we expect that there would be differences between communities of the edge and the interior of the areas. However, the disturbance level of the studied area might extend some edge characteristics of the microclimate, such as temperature and humidity, to the interior of the forest. MO areas especially present relatively few seedlings and a minimally developed understory (I.A. Fick, unpubl. data), thus increasing the incidence of solar light when compared with other areas of Araucaria forest without the occurrence of cattle. For the Atlantic forest areas, we also did not detect apparent edge effects. This might be a real pattern, indicating that land flatworms are not affected by forest edges, or else that in order to sample the real "interior" communities it will be necessary to collect planarians at >50 m from the forest edge. Alternatively, this pattern could be influenced by the successional stage of the forest, which was not a primary forest.

The clear separation of the communities of the two kinds of forest in the NPAS, and the preference of some species for only one fragment of Araucaria forest, suggest the potential usefulness of such species indicators for vegetal formations and also in conservation studies. This preference also indicates the importance of the preservation of this protected area, where the elimination of a fragment of Araucaria forest could mean local or regional extinction of a species.

Acknowledgments. We acknowledge the Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) and the Fundação de Amparo à Pesquisa do Rio Grande do Sul (FAPERGS) for providing fellowships and grants that supported this study and the Instituto Nacional do Meio Ambiente e Recursos Renováveis (IBAMA) for supporting field activities. Dr. Fernando Carbayo Baz provided suggestions during field activities. Fabiano Gil is acknowledged for his help with the preparation of the final version of the figures.

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