



Habitat structure, soil properties, and food availability do not predict terrestrial flatworms occurrence in Araucaria Forest sites

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ABSTRACT

Flatworm community structure responds strongly to large-scale habitat alteration, with low diversity in disturbed environments. However, the ecological factors influencing small-scale distribution of flatworms are poorly understood. The aim of this study is to test if the occurrence of land planarians is influenced by microhabitat traits. The study was developed in two Araucaria Forest sites located in the São Francisco de Paula National Forest, southern Brazil. In each site, a 0.5 ha grid subdivided in 10 m² plots, was established. In 100 randomly selected plots, the occurrence and abundance of each land planarian species were recorded. For each plot, we also recorded information on six habitat structural traits, 21 soils properties, and 15 prey availability parameters. Sixty-six specimens were recorded, belonging to 21 species of land planarians. Although the two areas differed in relation to 17 ecological factors analyzed, mean flatworm density did not significantly differ between the two studied areas. The pattern of their abundance followed a Poisson distribution, suggesting that flatworms were randomly distributed throughout the study area. Simple logistic regressions and a multiple logistic regression, controlled for spatial autocorrelation, showed no evidence that the occurrence of terrestrial flatworms is controlled by habitat structure, prey availability or soil properties. In addition, a Principal Component Analysis, controlled for spatial autocorrelation, generated three main axes that represented 40.7% of the variation of the 42 ecological traits. This analysis showed very similar distribution of plots with and without flatworms along these three axes. The results suggest that land flatworms exhibit no pronounced microhabitat preferences inside undisturbed Araucaria Forest sites.

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Introduction

Organisms are not evenly distributed across their potential range, as species prefer certain habitats. This habitat selection is of critical importance to many species, being based to a large extent on food availability, and the presence of predators and competitors, but also on the abiotic conditions and physical structure of the habitat (Orians and Wittenberger 1991; Ward and Lubin 1993; Stilling 1999).

For soil invertebrates, in particular, several environmental factors can determine microhabitat selection. Litter-dwelling invertebrate communities, for example, are influenced by the presence of logs (Evans et al. 2003). The richness and diversity of lapidicolous communities were significantly correlated to patch area, terrain slope, and the amount of organic matter and water in the substrate (Ferreira and Silva 2001). In another investigation, spiders, mites, springtails and cockroaches belonging to

rock-dwelling fauna on sandstone outcrops were linked with high percentages of rock substrate, whereas ant communities were associated with rocks that showed a deep soil substrate, those partly embedded into the soil, and those with high canopy cover (Goldsbrough et al. 2003). Also, biotic factors, such as prey availability and interspecific competition, are certainly important for microhabitat selection (Ward and Lubin 1993; Churchfield and Rychlik 2006; Birkhofer et al. 2010).

Terrestrial flatworms can be commonly found in the leaf litter and under stones, fallen logs, and branches. Some studies have suggested that flatworms remain hidden during the day in humid refuges because they cannot adapt to dry conditions (Kawaguti 1932; Froehlich 1955; Winsor et al. 1998). Furthermore, they are also sensitive to high humidity, leading to avoidance of more humid areas (Froehlich 1955; Winsor et al. 1998; Sluys 1998, 1999). Terrestrial flatworms are carnivorous, preying upon slugs, snails, earthworms, nemertean, land isopods, insect larvae, termites, springtails, mites, and other terrestrial flatworms (Froehlich 1955; Ogren 1995; Prasniski and Leal-Zanchet 2009). Therefore, flatworm abundance could also be expected to follow the abundance of their preferable prey.

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Studies on the influence of ecological factors on the occurrence or abundance of terrestrial flatworms are rare. They are mainly related to the occurrence in Europe of two exotic species, the “New Zealand” (*Arthurdendyus triangulatus*) and the “Australia” (*Australoplana sanguinea alba*) flatworms which are earthworm predators (Boag et al. 1998a; Christensen and Mather 1998; Jones and Cumming 1998; Mather and Christensen 1998). Boag et al. (1998b) used Geographic Information System techniques, soil pH, and land use information to predict where *A. triangulatus* could have the potential to cause a detrimental effect on agriculture. They showed that low annual rainfall and acidic soils probably restricts earthworm occurrence, and hence the occurrence of *A. triangulatus*. Jones et al. (1998) found a positive relationship between the abundance of *A. sanguinea alba* and monthly rainfall in the previous three months. In a study looking at the influence of air and soil humidity as well as soil organic matter contents on a flatworm community in areas originally covered by deciduous forest, the abundance of terrestrial flatworms was negatively correlated to soil organic matter content (Baptista and Leal-Zanchet 2010). Thus, the factors which influence the occurrence of flatworms in their natural environment remain poorly understood.

In Southern Brazil, it has been demonstrated that the abundance, richness, and composition of terrestrial flatworms respond dramatically to major habitat shifts (Carbayo et al. 2002; Leal-Zanchet et al. 2006; Fonseca et al. 2009; Leal-Zanchet et al. 2011). The present study aims to test if terrestrial flatworms exhibit microhabitat preferences within an undisturbed native forest patch, particularly in relation to habitat structure, soil properties, and prey availability.

Materials and methods

Study area

This study was performed in the São Francisco de Paula National Forest (29°23′–29°27′S, 50°23′–50°25′W) which is located on the eastern border of the Araucaria Plateau, in Rio Grande do Sul, Brazil. This National Forest encompasses 1606.70 ha, with altitudes ranging from 600 to 950 m a.s.l. The landscape was originally dominated by Araucaria Forest and native grasslands (Pillar et al. 2009). Nowadays, remnants of this forest type form a heterogeneous mosaic landscape mixed with old-tree monocultures of *Araucaria angustifolia*, *Pinus*, and *Eucalyptus* (Fonseca et al. 2009).

The regional climate is subtropical and humid, without an extremely dry period and an annual rainfall of around 1750–2500 mm year⁻¹. The mean annual temperature is about 16 °C (Backes 1999).

Sampling design

Terrestrial flatworm surveys were carried out in two roughly adjacent Araucaria Forest sites located on the opposite sides of a forest stream (eastern and western sides, respectively). In each area, a half hectare grid, subdivided in 10 m × 10 m plots, was established. In total, 100 plots were randomly selected for flatworm sampling; 56 and 44 plots being located in the eastern and western sides, respectively.

The occurrence and abundance of terrestrial flatworms were surveyed once in each plot in July 2007. Each plot was subdivided in four subplots (2.5 m × 2.5 m), each one being sampled for 10 min. Furthermore, for each plot, we recorded information on 42 ecological factors, as follows:

Shrub density and depth of leaf litter

Shrub density and leaf litter depth were measured at 22 points (spaced 50 cm from each other) located in a plot-centered cross design.

Refuges availability

In each plot we counted the number of stones with a width of at least 10 cm. Also, we counted and measured the length and width of all tree logs and branches with diameter greater than 2 cm.

Prey availability

The availability of potential prey items was estimated by one pitfall trap in the middle of each plot. The pitfall (6.5 cm × 7 cm), containing 200 ml of an aqueous solution of ethyl alcohol, remained open during seven consecutive days. Animals captured by the traps were identified to the order or class level. Also, they were classified into the following size class categories: 1 (0–2 mm), 2 (2.1–4 mm), 3 (4.1–8 mm), and 4 (more than 8 mm).

Canopy openness

The image of the canopy of each plot was recorded by a hemispheric lens digital camera, located in the center of the plot and 1 m above ground level. Canopy openness (%) was estimated by the software Gap Light Analyzer 2.0 (Frazer et al. 1999).

Physical and chemical soil variables

Standardized soil samples (20 cm deep and 10 cm wide), extracted with a soil borer, were collected in the center and on the four corners of each plot. For each plot, the samples were analyzed in the laboratory for texture, clay composition, pH, moisture, organic matter, phosphate, potassium, calcium, magnesium, and aluminum contents. Texture and clay composition were determined by the densimetry method; pH was established in a soil:water ratio of 1:1. Soil moisture was determined by finding the difference between the soil before and after oven drying. Organic matter was extracted using the wet digestion method. P and K were determined by the Mehlich-1 solution. Exchangeable Ca, Mg, and Al were extracted with 1 mol L⁻¹ KCl.

Data analyses

Spatial heterogeneity of the environmental variables was characterized by the coefficient of variation (CV = SD/mean). Differences between sites for the 42 ecological variables were analyzed by *t*-tests after the data were log-transformed (Kolmogorov–Smirnov, *P* > 0.05), except pH, leaf litter depth, calcium content, and base saturation index. Differences in mean flatworm density between the two sites were evaluated by the non-parametric Mann–Whitney U Test Statistic. The spatial distribution pattern of the flatworms was contrasted with the expected values based on a Poisson distribution with a chi-square goodness of fit test. Spatial autocorrelation was tested by Moran's I, using 10 distance classes containing equal number of pairs (analyses using from 8 to 12 classes showed the same results). The significance of Moran's I for each distance class was tested by 999 permutations, after correcting the α -value by Bonferroni. Simple logistic regressions, corrected for spatial autocorrelation, were used to test the influence of each one of the 42 environmental variables on the occurrence of the terrestrial flatworms. The correction for spatial auto-correlation is made by including in the model an auto-covariate which obtained an autoregressive model approach (Dormann 2007). For the analyses, tolerance was fixed as 0.75 and for each run 100 iterations were performed (Rangel et al. 2010). Afterwards, the 42 variables were grouped in three variable types: (1) habitat structure (Hab) – number of stones, number and size of fallen tree logs and branches, shrub density, litter height, canopy openness, (2) soil property (Soil)

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