



Herbaceous layer development during spring does not deplete soil nitrogen in the Portuguese montado

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ABSTRACT

Nitrogen (N) content in the soil and in the herbaceous biomass were monitored during spring of 2004–2006 to determine how the herbaceous layer development influences soil N availability in the montado ecosystem of southern Portugal. Highest ($246.6 \pm 52.7 \text{ g m}^{-2}$) and lowest ($123.2 \pm 89.5 \text{ g m}^{-2}$) peak biomass occurred in 2006 and 2005 respectively. Total soil N within the top 20 cm soil profile ranged between $0.2 \pm 0.1\%$ in February and $0.41 \pm 0.2\%$ in May, while available soil N was lowest ($5 \pm 2 \mu\text{g g}^{-1}$ soil) in February but increased three-to-five fold in March and was $>17.5 \mu\text{g g}^{-1}$ soil at senescence in May. Significant ($p < 0.001$) increase in total N in the aboveground pool occurred between February and May. There was however, no decay in soil N content. Instead, the herbaceous vegetation enhanced soil N input and N retention in the ecosystem. Most of the herbaceous plants were annuals with large reserves of organic N at senescence, which returned to the soil as detritus. The herbaceous vegetation is a critical component of the montado that contributes to N recharge and cycling within the ecosystem.

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1. Introduction

Montado, a two-layered vegetation structure covers approximately 2–2.5 million ha in the Iberian Peninsula and is dominated by a continuous layer of herbaceous biomass and widely distributed trees (Joffre and Lacaze, 1993; Joffre et al., 1988, 1999). The tree-component is comprised of the evergreen oak species, mainly *Quercus suber* and *Quercus rotundifolia* (Bugalho et al., 2009). The herbaceous layer has a rich biological diversity and is considered as habitats to be preserved (Moreno et al., 2005a). Due to its extent, the herbaceous layer provides a larger photosynthetic surface compared to the trees and could play a significant role in carbon fixation and ecosystem productivity (Pereira et al., 2007; Aires et al., 2008; Li et al., 2008) as well as influence soil nutrient and water balance (Joffre, 1990; Moreno et al., 2007). Due to the semi-arid nature of the montado, the life cycle of the herbaceous plant

community is confined to spring when soil moisture and temperatures favor their growth and development (Otieno et al., 2006; Pereira et al., 2007).

Most studies in the Mediterranean tree-grass mosaic focus on the effects of trees on the herbaceous layer production (Joffre and Rambal, 1988; Joffre, 1990; Jackson et al., 1990; Gallardo, 2003; Moreno et al., 2005a; Moreno, 2008). Limited information however, exists on how the understory vegetation influences soil nutrient availability and hence the overall ecosystem production. The nature of the herbaceous layer development suggests that it may play a role in the availability and distribution of soil resources, with significant implications on ecosystem functioning (Whitehead, 1995) since the soil nutrient and water store is limited (Aires et al., 2008). Studies have shown positive effects of understory clearance on the physiological status (Infante et al., 1999; Moreno and Cubera, 2008) and productivity (Diaz et al., 1997) of the remaining trees. The improved physiological status of the trees could be due to an increase in the available soil volume, and thus water and nutrients for the individual trees (Moreno et al., 2005b). Woodmansee and Duncan (1980), Joffre (1990) and Gallardo et al. (2000) have shown a rapid increase in

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nitrogen (N) in the herbaceous layer biomass between February and April in the Mediterranean regions, while soil extractable nitrogen was at its lowest. This however, is also the period when maximum growth of trees occurs (Otieno et al., 2006). The proliferation of the herbaceous layer in spring results in the accumulation of nitrogen in the aboveground biomass and may lead to a significant reduction in soil N and its limitation. Although extensive rooting system of trees may allow them to operate outside the boundaries of the herbaceous layer and they could access nutrient resources located in the deep soil layers (Moreno et al., 2007), the strategy is only relevant when the system allows deep soil layers to be systematically recharged.

Mediterranean ecosystems are water controlled and microbial biomass and N mineralization are regulated by moisture dynamics (Joffre, 1990; Gallardo et al., 2000). In the Mediterranean region of Spain, which resembles the Portuguese montado, major organic matter input into the soil occurs during spring and summer while potential maximum nitrification is reached in spring, with a significant increase in soil extractable nitrogen between February and June (Gallardo et al., 2000). Low mineralization and soil N input in winter is due to low temperatures and anaerobic conditions (Whitehead, 1995), while nitrification is inhibited by dry soil conditions, restricting the amount of available N (Garwood and Tyson, 1973). Thus nitrogen input in spring should be adequate to support biomass development of the herbaceous layer without depleting the soil N store during spring and early summer, since the period also coincides with active mineralization and soil N input. In this study, we examined the dynamics of nitrogen in relation to biomass development of the herbaceous layer community during the active growing period in spring. We attempt to answer the following questions: 1. How does N distribution in the soil and plant pools relate to biomass development of the herbaceous layer plant community? 2. Is there a possible limitation of soil N created by the rapid growth of the herbaceous layer community in spring?

2. Materials and methods

2.1. Study site and plant species

The experimental site was located in the center of the Alentejo Province, southern Portugal, at Herdade da Mitra (38°32'N, 88°00'W, 243 m a.s.l.) near the Mitra campus of the University of Evora. Measurements were carried out during the spring periods of 2004, 2005 and 2006. The climate in Mitra is Mediterranean, with hot and dry summers. Annual precipitation ranges between 350 and 650 mm, occurring between October and April. Between 2004 and 2006 rainfall patterns typical of this Mediterranean region of Portugal (Corte-Real et al., 1998) were experienced, but with significant inter-annual variations. Compared to 2004 and 2006, the hydrological year 2005 was drier, with precipitation amounts below the long-term mean. The hydrological year 2006 was the wettest with rainfall record between October and April amounting to 561 mm (2005 = 314 mm and 2004 = 481 mm). The soils were shallow (ca. 30 cm deep) sandy Cambisols (FAO, 1988) overlying a fractured gneiss rock, with low water retention capacity. Soil characteristics of the top 20 cm soil profile are shown in Table 1.

The vegetation is typical for the montado and the dominant tree species are *Q. suber* and *Q. rotundifolia* at a density of 40–50 trees ha⁻¹, comprising an overstory with ca. 35% of tree canopy

cover. The understory comprises a rich diversity of plant species that can be grouped into 3 functional types of forbs, grasses and legumes. Approximately 70% of the herbaceous community consisted of annuals, while 30% were perennials (Supplementary material).

2.2. Measurements

2.2.1. Above- and belowground biomass sampling

Monthly biomass determinations were conducted during spring of the three years. Active growth of the herbaceous layer was confined to the spring period between March and May, due to low temperatures between November and February and drought that occurs from June to September (Otieno et al., 2006; Pereira et al., 2007). The choice of sampling period was, therefore, based on the active vegetative period (spring) of the herbaceous layer. During January 2004, an area measuring 100 m by 100 m was demarcated for our studies. The study area was relatively uniform, with an even topography. The demarcated area was representative of the Mitra region. Grazing by domestic animals was excluded between February and May during the period when we conducted sampling, otherwise the fields were grazed during the rest of the year. This was necessary in order to allow for the quantification of the total N shifted to the aboveground biomass pool during plant growth.

During each monthly sampling, 15 sampling plots (quadrats) measuring 25 cm by 25 cm were randomly established across the study site for aboveground biomass harvesting. Each plot was established at least 10 m from the nearest tree to avoid the influence of trees and their roots on nutrient and biomass. From each plot, aboveground biomass was cut to the ground level and the harvested biomass separated into forbs, grass and legumes before being oven-dried at 80 °C for 48 h and weighed to determine aboveground shoot biomass. The harvested plots were clearly marked and numbered to avoid repeated harvesting in the subsequent months. New plots were established on fresh, undisturbed locations during subsequent months. After cutting the aboveground biomass, soil cores were obtained from the center of each of the plots using an 8 cm-diameter core sampler. The soil cores were separated into layers of 0–3, 3–7 and 7–12 cm depths. Roots were separated from the soil by washing the soil samples under running tap water in a 0.5 mm sieve. The roots were oven-dried at 80 °C for 48 h and weighed to determine root biomass.

2.2.2. Soil and plant nitrogen (N) determination

On the same day as biomass sampling, a second soil core was obtained from the same location, next to the root sampling point, down to 20 cm soil depth for soil N analysis using an 8 cm-diameter core sampler. A set of 15 soil samples was obtained from the study site at any monthly sampling period. The fresh soil samples from the respective plots were divided into layers of 0–3, 3–7, 7–12 and 12–20 cm depths. Samples from each layer were then divided into two equal parts. One set of samples was used for the determination of soil nitrogen. The second set, from the 4 respective layers, was used for the determination of soil pH. Soil pH was determined by filling 20 g of fresh ground and homogenized soil into plastic bottles with 50 ml of distilled water and shaking for 1 h. A calibrated pH-meter was used for pH determination.

Available soil nitrogen was extracted by shaking 100 g of fresh, homogenized soil with 250 ml of 1 M KCl. The extracted solution

Table 1
Physical and chemical soil characteristics of the study site in Mitra.

Depth (cm)	Bulk density (g cm ⁻³)	Texture			pH	%C	%N	C:N
		%Sand	%Silt	%Clay				
0–20 cm	1.54 ± 0.3	70	21	9	6.4 ± 0.5	1.4 ± 0.9	0.1 ± 0.1	11.5 ± 2

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