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Heterogeneity of soil surface ammonium concentration and other characteristics, related to plant specific variability in a Mediterranean-type ecosystem

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Received 16 January 2007; received in revised form 26 November 2007; accepted 3 December 2007

In Mediterranean-type ecosystems ammonium is present in the soil throughout the year and its concentration is dependent on plant cover.

Abstract

Heterogeneity and dynamics of eight soil surface characteristics essential for plants—ammonium and nitrate concentrations, water content, temperature, pH, organic matter, nitrification and ammonification rates—were studied in a Mediterranean-type ecosystem on four occasions over a year. Soil properties varied seasonally and were influenced by plant species. Nitrate and ammonium were present in the soil at similar concentrations throughout the year. The positive correlation between them at the time of greatest plant development indicates that ammonium is a readily available nitrogen source in Mediterranean-type ecosystems. The results presented here suggest that plant cover significantly affects soil surface characteristics.

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Keywords: Soil nitrogen availability; Mediterranean; Nitrate; Ammonium; Plant functional types

1. Introduction

Heterogeneity in soil resource distribution arises from organic inputs and their subsequent microbial decomposition. These inputs vary as widely in their chemical and physical properties as the materials they are derived from (Hodge, 2003), while the spatial and temporal release of these materials is as complex (Tinker and Nye, 2000) as the inputs of organic and inorganic materials. Soil heterogeneity has been described at scales ranging from the landscape to the neighbourhoods of a few individual plants, and related to

community composition and ecosystem structure (McKane et al., 2002; Guo et al., 2004). Recently, attention has been directed to describing heterogeneity in the soil at the scale of individual plant neighbourhoods. Soil pH, organic matter content, and concentrations of various mineral elements have been shown to vary in some communities by an order of magnitude over 5 m or less (Jackson and Caldwell, 1993), and in a number of cases this variation has appeared to be associated with changes in plant species distribution (Montagini et al., 1986; Inouye et al., 1987; Tilman, 1989; Schlesinger et al., 1996).

The effects of individual plants on soil heterogeneity influence the functioning of several ecosystems (e.g. Hook et al., 1991; Schlesinger et al., 1996). Gallardo et al. (2005) pointed out that individual trees have an influence proportional to the

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projection of their crown area onto the soil surface. This heterogeneity is regularly invoked as being important for competitive interactions between plants (Chapin, 2000; McKane et al., 2002).

The most distinct features of Mediterranean-type ecosystems are their high biodiversity and the seasonality of air temperature and precipitation, characterised by a pronounced drought period in summer and a cool wet period in winter (Grove and Rackham, 2001). This climate has important implications for biological activity. The vegetation of the Mediterranean ecosystems may be grouped into two plant functional types (PFT): summer semi-deciduous and evergreen-sclerophylls. Each group has been characterised on the basis of its phenology, water relations and carbon exchange properties (Werner et al., 1999). Summer semi-deciduous species exhibit leaf dimorphism, shedding an important fraction of leaves and twigs in the summer (a stress avoidance strategy). Producing thicker leaves, they are better able to survive summer drought (stress tolerance strategies; Lösch et al., 1982; Correia and Catarino, 1994). Their development is very dependent on water availability in the upper soil layers. They are common in open and disturbed stands, but they are progressively eliminated under canopies of evergreen sclerophyllous species of the late successional stages (Correia and Catarino, 1994). Sclerophyll leaves are long-lived, consistent, hard and coriaceous (Mooney, 1983; Lo Gullo and Salleo, 1988). Sclerophyllous plants are able to develop deeper root systems (Werner et al., 1999).

The aim of this work was to determine whether plants with distinct metabolisms, chemical compositions and life spans influence soil surface characteristics in distinct ways. Eight soil surface characteristics essential for plant growth were assessed on four occasions over a year, to evaluate the relation between soil surface characteristics and plant species. Soil surface patterns were analysed considering sampling time and the dominant plant species, as well as the PFT (summer semi-deciduous or evergreen sclerophyllous) to which they belonged.

2. Materials and methods

2.1. Study site

Studies were carried out on a mixed sclerophyll scrub which had developed after fire 16 years previously, located in Serra da Arrábida, 50 km south of Lisbon (38°27'34" N, 9°0'20" W), at an altitude of 270 m. The soils of Serra da Arrábida have been classified as Calcic rhodo-chromic luvisols and calcareous chromic cambisols, according to the FAO system (Specht et al., 1988). The soils are skeletal, and true profiles cannot be discerned. The typical Mediterranean red soil "terra rossa" has accumulated in some places (Yaalon, 1997). Silt predominates in the soil (57%), while clay and sand contents are 28% and 15%, respectively (silt-sand-loam, Correia et al., 1992). The climate is sub-humid, warm variant according to Emberger's bi-climatic coefficient, with 650 mm average annual precipitation, and 16 °C mean annual temperature. The topsoil layer is very thin (maximum 15 cm), formed from calcareous bedrock (15.1% sand, 56.7% lime and 28.2% clay).

2.2. Field sampling

To study soil spatial variability at a medium scale, the approximately 50 × 50 m study area was divided into 575 cells of 2 × 2 m. Sampling took

place in November 2004, and February, April and July 2005. On each sampling occasion all 575 samples were cored within 10 days.

The study area was surveyed for shrub and tree species; herbaceous plants were not abundant and therefore not taken into consideration. The dominant plant species in each cell (the species occupying the most area) was identified and a central main plant stem selected for soil sampling. Whenever possible the identified species were grouped into two PFTs: summer semi-deciduous and evergreen-sclerophyllous (Correia and Catarino, 1994).

Each soil sample consisted of three cores, with 2 cm diameter and 15 cm length, taken within 1 cm distance of each other and 20 cm from the central main plant stem. A sub-sample for nitrate and ammonium analysis was immediately placed in 2 M KCl. Samples were stored at 4 °C for analysis. Soil water content was measured by time domain reflectometry (TDR) and temperature with a thermocouple. Both parameters were determined at 5 cm depth between 11:00 and 13:00 h.

Soil pH was determined in a water extract (1:10, w/v) with a selective electrode (micro pH 2001, Crison).

2.3. Laboratory analysis

Sieved (2 mm) soil samples were analysed for: organic matter, by loss of ignition; nitrate concentration, by electrophilic substitution of salicylate acid; and ammonium concentration, using the Berthelot reaction (Cruz and Martins-Loução, 2000). Organic matter concentrations were expressed as percentage of dry soil, nitrate and ammonium concentrations as µg N per gram of dry soil.

Nitrification potential was determined according to Kandeler (1996) on 10 g of fresh soil, amended with (NH₄)₂SO₄ and incubated at 60% of water holding capacity, at 25 °C for 2 weeks. Soil nitrate was extracted and determined at 0, 5, 10 and 15 days. Nitrification rates were obtained as the slope of the regression line fitting plots of NO₃⁻-N concentrations against time. In all cases a linear increase in NO₃⁻-N over time was observed. Results were expressed as µg NO₃⁻-N g⁻¹ day⁻¹.

Arginine ammonification (Alef and Kleiner, 1987) was used to estimate soil mineralisation. Soil samples (10 g dry weight) were mixed with 2.5 ml arginine solution (0.2%). Samples and controls with arginine solution substituted by distilled water were prepared in triplicate and incubated at 25 °C for 3 h. The reaction was stopped by freezing at -20 °C. After storage samples were brought to room temperature in the presence of 40 ml 2 M KCl, and shaken for 1 h, then centrifuged at 5000 × g for 10 min before determination of ammonium in the supernatant.

2.4. Statistical analyses

Soil properties were plotted and their summary statistics compared, for the different sample periods and for different plant species and related plant functional types. Soil property averages were compared by a Welch modified two-sided *t*-test.

Differences between samples in terms of surface soil properties were analysed through ordination, using CANOCO (Ter Braak and Smilauer, 1998). A principal component analysis (PCA), based on a correlation matrix to account for the different measuring units of the soil parameters, was computed for each sample occasion. Results are presented in biplots showing sample distribution in the ordination space (of the first two axes), as well as the relationship of the soil parameters to these axes and the samples. The relevance of the soil parameters in the determination of the PCA axes (their correlation to these axes) is shown graphically and by the respective axis scores. The samples in the PCA plots were marked with their respective dominant PFT or group. For ease of interpretation, a second plot was made presenting the position of the main resident species (centroid of the species' samples) in the same ordination space.

Statistical differences between sites occupied by different plant types were assessed through an analysis of similarities (ANOSIM) using Primer (Clarke and Green, 1988; Clarke, 1993). Analogously to the PCA, a (standardised normalised Euclidean distance) similarity matrix was used to cluster samples. Based on that matrix and on the three previously established groups (samples assigned to evergreen sclerophyllous, summer semi-deciduous or *Erica* sp./*Juniperus phoenicea* cover) similarities between groups are compared to similarities within groups. Distinctions between groups and statistical significance

of this distinction (global and pairwise differences between groups) are quantified.

Clustering in the distributions of individual plant species and plant functional types was assessed by Ripley's \hat{K} , comparing the observed point patterns of PFTs and individual species with the pattern of a homogeneous process with no spatial dependence. Clustering and basic statistical analyses were computed in R (R Development Core Team, 2005).

3. Results

No change in the dominant plant in each sampling cell was observed between the four sampling occasions. Most of the 13 identified plant species could be grouped into the two plant functional types: 5 summer semi-deciduous species dominated 205 cells, while 267 cells were dominated by 6 evergreen-sclerophyllous species (Table 1). *Erica* sp. and *Juniperus phoenicea* were considered separately.

All the analysed surface soil parameters (ammonium and nitrate concentrations, soil moisture, temperature, pH, organic matter, and nitrification and ammonification rates) varied spatially and temporally, which reflected in the variation per main plant species and per sample time (Fig. 1, Appendix A). Soil sample properties tended to display similar patterns of temporal and spatial variation according to the PFT which dominated their cell, thus a group of soil samples influenced by summer semi-deciduous, and another group, influenced by evergreen-sclerophyllous species and (generally also) *Erica* sp. and *Juniperus phoenicea* could be distinguished. Cell soil conditions were significantly different according to the dominant PFT (except for ammonium and nitrate concentrations in periods of low inorganic N availability and soil moisture). This distinction was especially evident when considering soil moisture, soil temperature, organic matter and nitrification. Ammonium and nitrate concentrations, as well as their relative proportion of availability are also distinct according to the PFT which dominated the cell, though to a lesser degree (Fig. 1). Samples associated with *Erica* sp. and *J. phoenicea* tended to be more similar to sclerophyllous-dominated cells (Table 2). Ammonium concentrations in the surface soils of all cells were higher in April and July than in November

and February. In April and July, evergreen-sclerophyllous-dominated cells presented significantly lower ammonium concentrations than cells dominated by summer semi-deciduous species. As with ammonium, nitrate concentrations in April and July were higher in summer semi-deciduous dominated cells. With the exception of the July sample, evergreen-sclerophyllous dominated cells had higher surface soil temperatures than those dominated by summer semi-deciduous species, especially in April. Irrespective of the pattern of variation, evergreen sclerophyll dominated cells had higher soil surface humidities than cells dominated by summer semi-deciduous species. Soil organic matter concentrations of summer semi-deciduous dominated cells were always lower than those of evergreen-sclerophyll dominated cells. In summer semi-deciduous dominated cells, the highest organic matter concentrations were found in July, while in cells dominated by evergreen-sclerophylls, the highest organic matter concentrations were observed in February. On all sampling occasions, the soil pH of summer-semi-deciduous dominated cells was lower than that of evergreen-sclerophyllous dominated cells. Pairwise comparison of consecutive time periods, for cells dominated by the same PFT, showed that changes in average values were significant for almost all soil properties and time steps (Table 2).

Principal component analyses revealed that the heterogeneities of the soil parameters varied seasonally, as did the relationships between them (Table 3, Fig. 2). The plots showing samples (marked according to the dominant plant species type) and soil properties (Fig. 2a,c,e,g) exhibit clustering of samples from cells dominated by the same PFT, a pattern which was most distinct in samples taken in April. The plots showing the central location of each plant species in relation to the PCA axes (Fig. 2b,d,f,h) highlight the clustering, revealing the proximity (similar soil conditions) of functionally related species.

Examining the two main clusters for each sampling occasion, we can see that, in comparison to samples from cells dominated by evergreen sclerophyllous species, samples from cells occupied by summer semi-deciduous species showed: lower soil temperatures and nitrate and higher nitrification rates in November; lower soil temperatures, organic matter contents and pH in February; lower soil moisture contents, soil temperatures and, to a lesser degree, pH in April; and higher soil temperatures and nitrate concentrations, and lower soil moisture contents in July. Just as soil parameters conditioned by PFT varied over time, the effect of the different soil conditions on the ordination (PCA scores) also varied over time (Table 3).

Analysis of similarity results (Table 4) suggests that ordination of the samples can be satisfactorily represented by the first two axes (all stress values are <0.2). The global distinction between PFT is clearly significant, with global R values varying between 0.3 and 0.5 (indicating distinct though overlapping groups). Pairwise differences between groups show that soil conditions related to the two PFT were always significantly different from each other. Cells dominated by *Erica* sp. and *J. phoenicea* were always significantly different from summer

Table 1
Identified dominant plant species, their frequency (occur.) assigned code and plant functional type (PFT) or group

Species	Code	Occur.	PFT/group
<i>Cistus albidus</i>	Ca	74	Summer semi-deciduous
<i>Cistus ladanifer</i>	Cl	2	Summer semi-deciduous
<i>Cistus monspeliensis</i>	Cm	89	Summer semi-deciduous
<i>Lavandula</i> sp.	La	2	Summer semi-deciduous
<i>Rosmarinus officinalis</i>	Ro	38	Summer semi-deciduous
<i>Cerantia siliqua</i>	Ce	11	Evergreen-sclerophyllous
<i>Myrtus communis</i>	Mu	17	Evergreen-sclerophyllous
<i>Olea europaea</i>	Ol	41	Evergreen-sclerophyllous
<i>Phillyrea angustifolia</i>	Ph	95	Evergreen-sclerophyllous
<i>Pistacea lentiscus</i>	Pi	57	Evergreen-sclerophyllous
<i>Rhamnus lycioides</i>	Rh	46	Evergreen-sclerophyllous
<i>Erica</i> sp.	Er	10	Erica/Juniperus
<i>Juniperus phoenicea</i>	Ju	93	Erica/Juniperus

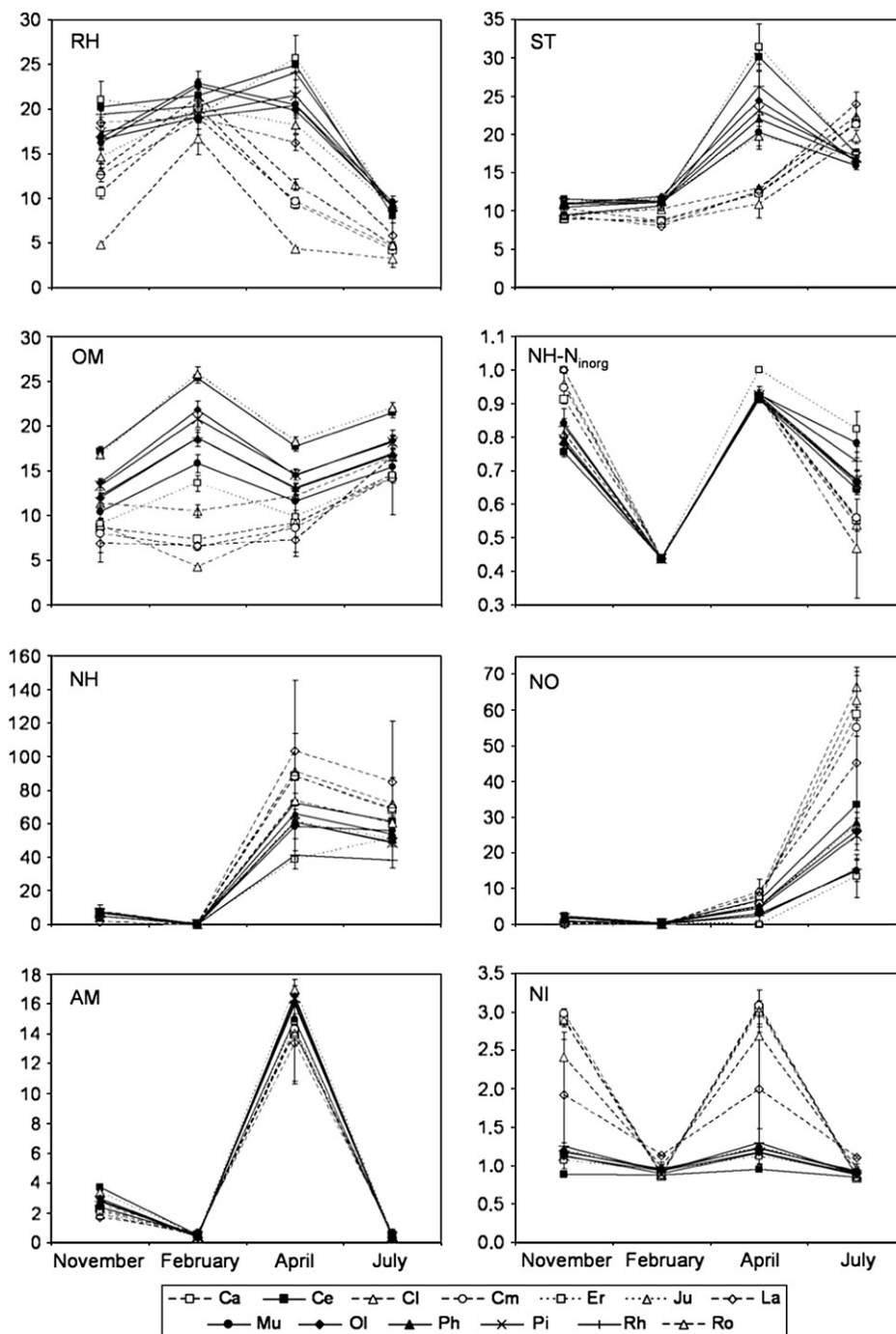


Fig. 1. Temporal variation of average (\pm SE) surface soil properties for cells dominated by the 13 observed plant species; evergreen-sclerophyllous with continuous line and filled symbol; semi-deciduous with dashed line and open symbol; others with dotted line and grey-filled symbol. RH, soil moisture (%); ST, soil temperature ($^{\circ}$ C); OM, organic matter (% soil DW); $\text{NH-N}_{\text{inorg}}$, proportion of ammonium in inorganic N; NH, ammonium ($\mu\text{g N g}^{-1}$ soil DW); NO, nitrate ($\mu\text{g N g}^{-1}$ soil DW); AM, ammonification ($\mu\text{g NH}_4 \text{ g}^{-1} \text{ day}^{-1}$); NI, nitrification ($\mu\text{g NO}_3 \text{ g}^{-1} \text{ day}^{-1}$); species codes according to Table 1.

semi-deciduous dominated cells, especially in April, with $R > 0.75$ indicating large differences and no overlap between groups. Soil conditions of cells dominated by *Erica* sp. and *J. phoenicea* were quite similar to those of cells dominated by sclerophylls, since R values of less than 0.25 indicate very poor separation between groups. They were not significantly different from cells dominated by evergreen sclerophylls

in February and July, yet significantly different in November and April.

The spatial distributions of *Rosmarinus officinalis*, *Juniperus phoenicea*, *Olea europaea*, *Pistacea lentiscus* and *Rhamnus lycioides* approximated complete randomness (according to Ripley's \bar{K} , data not shown). Slight clustering was observed for *Cistus albidus*, *Myrtus communis* and *Phillyrea*

Table 2

Comparison of mean values and their SE (in parentheses) of the soil properties measured, in time and per PFT

Soil property	Time	Sclerophyllous (S)	Semi-deciduous (D)	<i>Erica/J. phoenicea</i> (E)	S–D	S–E	D–E
Soil moisture (%)	Nov	17.36 (0.40) ^a	12.00 (0.43) ^a	15.29 (0.77) ^a	***	*	***
	Feb	20.26 (0.31) ^b	19.77 (0.32) ^b	20.03 (0.41) ^b	n.s.	n.s.	n.s.
	Apr	21.38 (0.50) ^b	9.92 (0.30) ^c	18.97 (0.95) ^b	***	*	***
	Jul	9.06 (0.15) ^c	4.51 (0.11) ^d	8.99 (0.23) ^c	***	n.s.	***
Soil temperature (°C)	Nov	10.66 (0.06) ^a	9.14 (0.06) ^a	11.01 (0.10) ^a	***	**	***
	Feb	11.25 (0.09) ^b	8.97 (0.11) ^a	11.38 (0.14) ^b	***	n.s.	***
	Apr	23.55 (0.66) ^c	12.39 (0.14) ^b	20.90 (1.21) ^c	***	n.s.	***
	Jul	16.69 (0.12) ^d	21.51 (0.21) ^c	16.91 (0.20) ^d	***	n.s.	***
Organic matter (% soil DW)	Nov	12.70 (0.23) ^a	8.81 (0.19) ^a	15.99 (0.49) ^a	***	***	***
	Feb	19.71 (0.38) ^b	7.48 (0.24) ^b	24.67 (0.80) ^b	***	***	***
	Apr	13.69 (0.26) ^c	9.43 (0.22) ^c	17.42 (0.52) ^c	***	***	***
	Jul	17.47 (0.26) ^d	14.68 (0.21) ^d	21.35 (0.52) ^d	***	***	***
Nitrate ($\mu\text{g N g}^{-1}$ soil DW)	Nov	1.63 (0.05) ^a	0.43 (0.04) ^a	1.57 (0.08) ^a	***	n.s.	***
	Feb	0.26 (0.03) ^b	0.33 (0.04) ^a	0.24 (0.06) ^b	n.s.	n.s.	n.s.
	Apr	4.52 (0.13) ^c	7.77 (0.16) ^b	4.33 (0.22) ^c	***	n.s.	***
	Jul	24.50 (1.12) ^d	58.49 (1.97) ^c	26.25 (2.00) ^d	***	n.s.	***
Ammonium ($\mu\text{g N g}^{-1}$ soil DW)	Nov	6.76 (0.16) ^a	5.59 (0.59) ^a	6.65 (0.21) ^a	n.s.	n.s.	n.s.
	Feb	0.20 (0.03) ^b	0.26 (0.03) ^b	0.18 (0.05) ^b	n.s.	n.s.	n.s.
	Apr	59.75 (1.46) ^c	88.65 (1.80) ^c	58.94 (1.84) ^c	***	n.s.	***
	Jul	49.76 (1.11) ^d	69.25 (1.47) ^d	50.90 (1.68) ^d	***	n.s.	***
pH	Nov	7.26 (0.03) ^a	6.82 (0.04) ^a	7.48 (0.09) ^a	***	*	***
	Feb	6.64 (0.04) ^b	6.18 (0.05) ^b	6.88 (0.10) ^b	***	*	***
	Apr	7.93 (0.05) ^c	6.50 (0.03) ^c	8.22 (0.11) ^c	***	*	***
	Jul	6.96 (0.10) ^d	5.60 (0.08) ^d	6.91 (0.14) ^d	***	n.s.	***
Ammonification ($\mu\text{g NH}_4 \text{ g}^{-1} \text{ day}^{-1}$)	Nov	2.84 (0.05) ^a	2.23 (0.04) ^a	3.25 (0.09) ^a	***	***	***
	Feb	0.43 (0.01) ^b	0.54 (0.01) ^b	0.46 (0.01) ^b	***	n	***
	Apr	16.13 (0.13) ^c	14.14 (0.13) ^c	16.78 (0.23) ^c	***	*	***
	Jul	0.39 (0.01) ^d	0.50 (0.01) ^d	0.42 (0.01) ^d	***	*	***
Nitrification ($\mu\text{g NH}_4 \text{ g}^{-1} \text{ day}^{-1}$)	Nov	1.16 (0.01) ^a	2.92 (0.04) ^a	1.20 (0.03) ^a	***	n.s.	***
	Feb	0.93 (0.01) ^b	0.89 (0.01) ^b	0.92 (0.02) ^b	**	n.s.	n.s.
	Apr	1.21 (0.01) ^c	3.05 (0.04) ^c	1.24 (0.03) ^c	***	n.s.	***
	Jul	0.91 (0.01) ^d	0.86 (0.01) ^d	0.90 (0.02) ^d	**	n.s.	n.s.

Temporal variability: mean values followed by different letters within a column of one soil property are significantly different ($p < 0.05$); PFT variability: S–D comparison between sclerophyll and semi-deciduous dominated sites, S–E between semi-deciduous and *Erica/J. phoenicea* dominated sites, D–E between sclerophyllous and *Erica/J. phoenicea* dominated sites. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$; n.s., not significant: $p \geq 0.05$.

angustifolia. The most clustered species were *Ceratonia siliqua*, *Erica* sp. and *Cistus monspeliensis* (data not shown). Clustering at PFT level was not evident (data not shown). *Cistus ladanifer* and *Lavandula* sp. were too rare in our study for this analysis.

4. Discussion

Irrespective of the sampling time, and considering all the soil parameters assessed, it was possible to group the soil samples according to the dominant plant species in the cells from which they were obtained: summer semi-deciduous species or evergreen sclerophyllous species, *Erica* sp. and *Juniperus phoenicea*. The plants found are representative of the most abundant and characteristic machia species (Thompson, 2005). Plant distribution across the sampling grid did not

change during the study (approximately 9 months), mainly because only shrub and tree species were considered.

Multivariate analyses showed that the surface soil characteristics displayed spatial heterogeneity related to the dominant resident plant species and their plant functional types. This relation was most evident for the spring sampling in April (Fig. 2, Table 4). Given the distinct strategies displayed by these PFT (Correia; 1998, Peñuelas and Fillela, 2003), it was to be expected that the full potential of their specific traits would become evident under differing conditions, namely different seasons. The summer semi-deciduous plants' strategy favours their growth whenever their root system can access water. This situation occurs in all seasons except summer; however, the highest growth rates are achieved during spring (Thompson, 2005). In contrast, the advantages of the evergreen sclerophylls' strategy become evident during the

Table 3

Proportion of variance (prop. var.) explained and soil-property scores for the first and second principal component axes (PC) of the PCA presented in Fig. 2

	November		February ^a		April		July	
	1st PC	2nd PC	1st PC	2nd PC	1st PC	2nd PC	1st PC	2nd PC
Prop. var.	0.448	0.144	0.259	0.160	0.474	0.156	0.406	0.175
Scores								
RH	0.522	1.704	0.508	−1.219	1.052	0.557	−0.969	1.127
ST	1.124	0.725	1.534	−0.526	0.955	0.676	0.972	−1.014
OM	1.190	−1.165	1.480	0.916	0.779	−1.304	−0.317	1.600
NO	1.118	0.238			−1.040	−1.127	1.029	−0.679
NH	0.827	0.415	−0.319	1.164	−0.934	−1.110	1.336	0.833
PH	0.896	−0.151	1.132	1.305	1.114	−1.075	−1.231	−0.673
AM	0.998	−1.580	−0.824	1.113	0.809	−1.366	1.282	1.102
NI	−1.148	−0.683	0.369	−0.316	−1.236	0.098	−0.124	0.567

Results are given for each of the four sampling occasions; soil variable codes are according to Fig. 1.

^a For February the soil property NO was omitted from the analysis because it was very correlated with NH.

Mediterranean summer. Thus, it was expected that the clearer separation/distinction between the studied soil samples influenced by plant species of the two PFT would occur during these two seasons: spring, the maximum growing season highlighting the summer semi-deciduous species; and summer, the survival season, highlighting the evergreen sclerophyllous species' influence. Interestingly, the data allowed the separation of the cells dominated by summer semi-deciduous species from all others, but did not convincingly distinguish between the cells dominated by evergreen sclerophylls and those dominated by other species such as *Erica* sp. and *Juniperus phoenicea* (Tables 2 and 4).

The factors determining the distinction between the two groups were not the same on all sampling occasions (Fig. 2, Table 3). In April, when conditions are generally favourable to biological activity, the sites with higher soil temperatures had lower nitrate and ammonium concentrations, possibly due to immobilisation (Ryel et al., 1996). In July, when weather conditions in Mediterranean ecosystems are less favourable, the combination of lower soil temperatures and higher water availability in the soils occupied by the evergreen sclerophylls (Table 2, Figs. 1 and 2) emphasise the differences between the characteristics of the soils occupied by the two PFT.

Throughout the year, soil surface properties may differ according to plant cover, suggesting that plant canopies and root systems influence surface soil properties (Gallardo et al., 2002; Castells and Peñuelas, 2003). Rutigliano et al. (2004) found that plant cover type was the key factor influencing the soil surface properties in Mediterranean-type ecosystems. Canopy shape and leaf morphology may affect interception of light and water (Lavelle, 1997; Hedlund et al., 2003), perhaps in turn influencing soil temperature and water content (Cruz et al., 2003), and be related with the range of soil temperatures observed over time (Fig. 1) and space in a particular area. In general, soils developing under trees are richer in organic matter and water availability (Joffre and Ramball, 1993). Leachates from

the canopy and leaf chemical composition are also known to interfere with soil biological activity (Castells and Peñuelas, 2003). The relevance of soil heterogeneity, promoted by plant cover, for ecosystem functioning may be greater when resources are very limited by biological activity (Thompson, 2005), as in Mediterranean-type ecosystems.

Nitrogen constraints on biological activity may be imposed by concentration and/or form. Both act through many interdependent factors: soil pH and temperature, water content, organic matter, biological activities, etc. If plant cover influences some of these properties, then they may consequently influence inorganic nitrogen availability. Since most soil properties are interdependent, but do not vary in a synchronised way, nitrate and ammonium pools in soils are very labile, heterogeneous (Table 2, Fig. 1) and unpredictable (Fitter and Hay, 2002).

The patterns of ammonium and nitrate temporal and spatial distribution are far from universal, but there is evidence for the presence of ammonium (Fig. 1) in Mediterranean-type ecosystems in concentrations comparable to (or higher than) those of nitrate (Castaldi and Aragosa, 2002; Castells and Peñuelas, 2003; Gallardo et al., 2005). Differences in nitrate and ammonium's spatial and temporal distributions may have important consequences for individual performance, since plants and soil biota differ in their preference for nitrate or ammonium as nitrogen source (Min et al., 2000; Gallardo et al., 2005; Cruz et al., 2006). Apart from biological preferences, the assimilation of each form has implications for other surface soil properties (Marschner, 1995). Therefore, for the same soil type, rhizosphere pH may differ greatly between plant species. Differences in pH of soils occupied by plants belonging to distinct species may reflect distinct nutrient acquisition strategies related to the fine scale pattern of nutrient distribution in the soil (Farley and Fitter, 1999).

Nitrate and ammonium are the nitrogen sources readily available to plants, but usually they do not represent more than 10% of the total nitrogen in the soil. Most is bound in organic matter, so the distribution of total nitrogen follows that of organic matter. Organic matter concentration in the soil was very heterogeneous, with high values associated with soils dominated by sclerophyllous plants (Table 2, Figs. 1 and 2). This may reflect the relative ability of each group to retain litter produced by the plants (Thompson, 2005). Since both groups present high contents of phenolic compounds (Castells and Peñuelas, 2003), this cannot be responsible for differences in decomposition rates of shed leaves.

Ammonification and nitrification are the main biological processes contributing to the production of inorganic nitrogen in natural and semi-natural systems. However, organic matter, nitrate and ammonium present distinct temporal (Fig. 1) and spatial (Cruz et al., 2003) variation patterns, suggesting that distributions are determined by distinct processes and therefore by different regulating factors. Nitrification rates observed under evergreen-sclerophyllous canopies were constant over time, which may be due to the presence of allelopathic compounds leached from plants, and to the quality of the sclerophyllous leaves (Gallardo and Menino, 1992). Nitrification

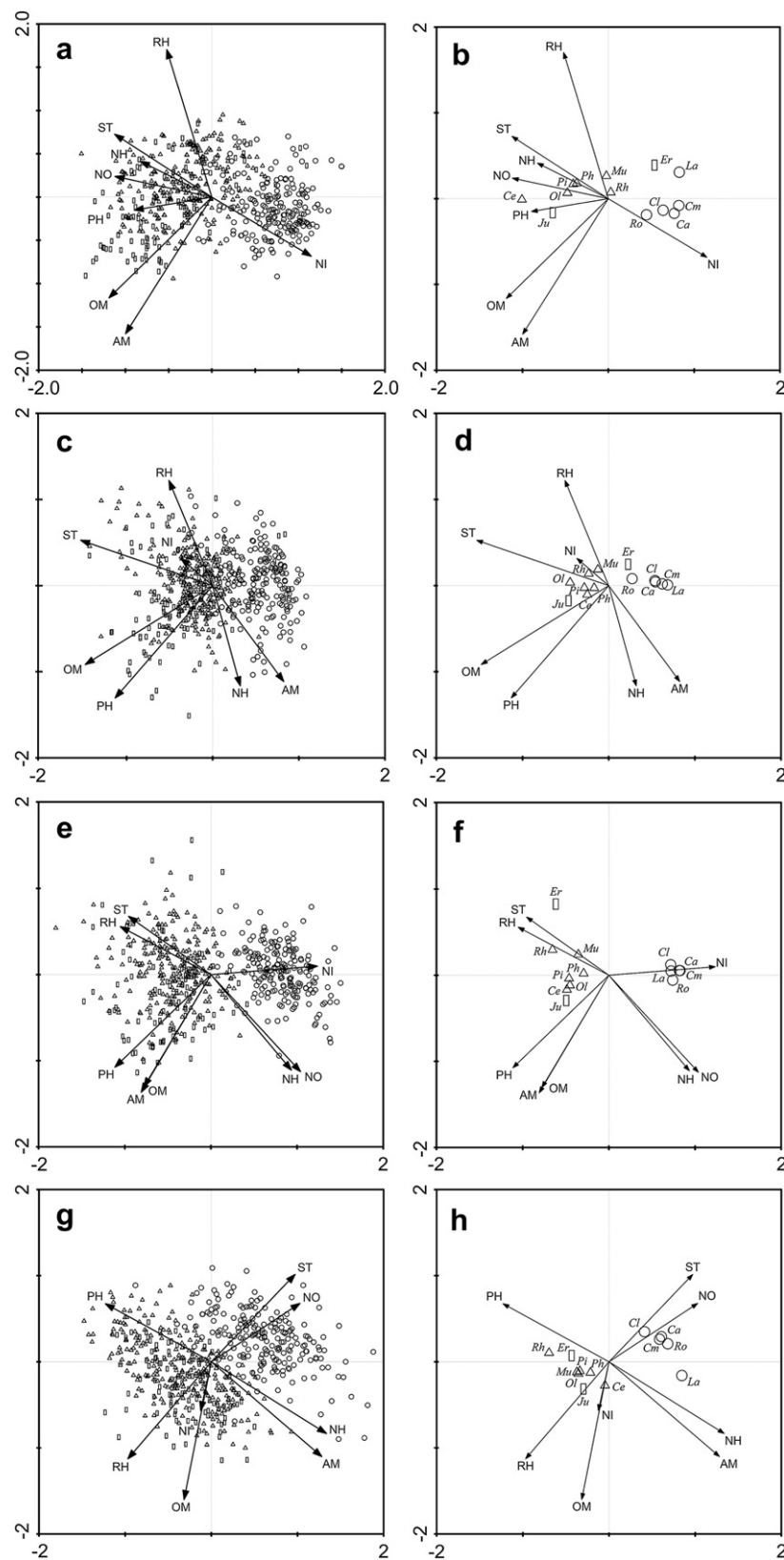


Fig. 2. (a) First two axes of the PCA ordination of the November samples according to their soil surface properties; vectors of soil properties are projected showing their correlation (vector direction) and effect on the axes (vector length). Samples are identified by the PFT of the dominant species: circle, summer semi-deciduous; triangle, evergreen sclerophyllous; box, *Erica* sp. and *Juniperus phoenicea*. (b) The same PCA, placing the dominant species in the ordination space; species codes according to Table 1, soil property codes according to Fig. 1. (c, d) *Idem* for the February samples; (e, f) *Idem* for the April samples; (g, h) *Idem* for the July samples.

Table 4
Analyses of similarity (ANOSIM) results for the comparison between samples with dominated species identified as summer semi-deciduous (D), evergreen sclerophyllous (S), or *Erica sp./I. phoenicea* (E), for each sampling occasion; stress, *R* values and significance levels are given for each comparison

Time	Stress	Comparison	<i>R</i>	Sig. level
November	0.18	Global	0.366	0.0001
		S–D	0.466	0.0001
		S–E	0.104	0.0001
		D–E	0.456	0.0001
February	0.15	Global	0.357	0.0001
		S–D	0.473	0.0001
		S–E	0.039	0.066 n.s.
		D–E	0.456	0.0001
April	0.13	Global	0.492	0.0001
		S–D	0.583	0.0001
		S–E	0.057	0.021
		D–E	0.775	0.0001
July	0.11	Global	0.285	0.0001
		S–D	0.341	0.0001
		S–E	0.010	0.302 n.s.
		D–E	0.528	0.0001

rates under summer semi-deciduous species changed over time. This may have been due to combined effects of some other surface soil characteristics. Note that although low nitrification rates were observed in July, nitrate content in the soil solution was the highest at that point (Fig. 1). This accumulation of nitrate in the soil solution may result from a severe inhibition of denitrification, due to low soil water content and high soil temperature (Fig. 1, Carreira et al., 1994).

The distinct effects of each soil parameter on soil heterogeneity over the year (Fig. 2, Table 3) suggest asynchrony between the specific variations of the factors having the largest influence on soil surface properties. The distinct correlations between factors according to the time of the year reflect an intrinsic degree of dependence of the variables, particularly evident under certain limiting conditions, and their influence on nutrient pool size and biological activities (Stark and Schmel, 2001).

Together the data clearly show that plant species did affect surface soil properties in a concerted form, indicating that the current trait-based functional classifications might be useful for understanding plant and ecosystem functioning. This distinction based on the properties of soil under the influence of plants belonging to different functional types is of great importance because research data on functional traits that surpass morphological or taxonomical descriptions are rare, though necessary to predict changes in ecosystem function.

Acknowledgements

This study was supported by the Fundação para a Ciência e Tecnologia FEDER through the project POCI/BIA-BDE/59183/2004.

Appendix

Soil property means (standard errors) per dominant plant species and sampling occasion; species coded according to Table 1

Sp.	Soil moisture				Soil temperature				Organic matter				Ammonium			
	Nov	Feb	Apr	Jul	Nov	Feb	Apr	Jul	Nov	Feb	Apr	Jul	Nov	Feb	Apr	Jul
Ca	10.67 (0.67)	20.07 (0.53)	9.33 (0.49)	4.23 (0.14)	8.95 (0.09)	8.76 (0.17)	12.31 (0.26)	21.40 (0.34)	8.58 (0.26)	7.29 (0.39)	9.12 (0.28)	14.28 (0.37)	7.00 (1.59)	0.28 (0.06)	88.13 (3.07)	68.10 (2.54)
Cl	4.81 (0.00)	16.74 (0.35)	4.33 (0.21)	3.21 (0.92)	9.15 (0.65)	8.44 (0.43)	10.83 (1.71)	19.66 (0.89)	8.86 (3.06)	4.26 (0.05)	9.04 (3.12)	14.53 (4.42)	7.80 (3.90)	0.00 (0.00)	73.64 (40.30)	60.45 (26.91)
Cm	12.61 (0.73)	18.94 (0.46)	9.69 (0.44)	4.62 (0.15)	10.27 (1.12)	8.66 (0.16)	12.22 (0.18)	21.24 (0.34)	7.95 (0.27)	6.44 (0.26)	8.60 (0.35)	14.18 (0.27)	4.78 (0.27)	0.30 (0.06)	87.99 (2.39)	68.89 (2.03)
La	18.45 (0.00)	18.96 (4.05)	16.23 (0.86)	5.82 (1.47)	9.39 (0.39)	7.97 (0.08)	12.62 (0.61)	23.91 (1.59)	6.86 (2.04)	6.60 (0.13)	7.24 (1.84)	16.60 (2.96)	1.56 (0.00)	0.00 (0.00)	103.17 (41.94)	85.02 (35.88)
Ro	13.17 (0.80)	21.35 (0.84)	11.55 (0.63)	4.78 (0.36)	9.42 (0.17)	10.20 (0.29)	13.03 (0.34)	22.31 (0.47)	11.30 (0.44)	10.47 (0.70)	12.09 (0.48)	16.53 (0.49)	4.82 (0.41)	0.12 (0.05)	91.20 (4.71)	71.99 (3.57)
Ce	20.28 (0.46)	21.51 (1.31)	24.96 (0.74)	8.04 (0.72)	11.47 (0.03)	11.29 (0.21)	30.08 (0.91)	17.58 (0.54)	17.19 (0.50)	25.33 (0.53)	17.69 (0.53)	21.51 (0.53)	7.73 (0.71)	0.28 (0.19)	72.19 (6.01)	61.12 (3.36)
Mu	16.82 (1.11)	22.84 (1.40)	20.54 (1.33)	9.54 (0.75)	10.47 (0.21)	11.17 (0.50)	20.24 (2.22)	15.90 (0.52)	10.37 (0.74)	15.81 (0.99)	11.60 (1.01)	15.42 (1.01)	6.65 (0.77)	0.18 (0.10)	58.25 (7.18)	55.75 (6.18)
Ol	16.26 (0.72)	22.59 (0.78)	19.88 (0.90)	9.16 (0.34)	10.92 (0.11)	11.86 (0.25)	24.40 (1.13)	16.46 (0.31)	13.60 (0.51)	21.84 (0.92)	14.49 (0.55)	18.31 (0.55)	6.77 (0.38)	0.22 (0.06)	61.00 (3.64)	49.25 (2.54)
Ph	16.59 (0.71)	19.04 (0.50)	20.39 (0.88)	9.00 (0.26)	10.86 (0.06)	11.19 (0.11)	21.99 (1.17)	16.78 (0.21)	12.20 (0.34)	18.69 (0.59)	13.10 (0.40)	16.91 (0.40)	7.48 (0.26)	0.24 (0.05)	66.10 (2.30)	53.82 (1.71)
Pi	17.40 (0.71)	19.55 (0.63)	21.51 (0.90)	9.22 (0.34)	11.04 (0.10)	11.35 (0.20)	23.09 (1.36)	17.01 (0.23)	13.34 (0.56)	20.81 (0.93)	14.53 (0.63)	18.19 (0.63)	6.88 (0.30)	0.18 (0.05)	60.87 (2.66)	48.58 (2.18)
Rh	19.41 (1.30)	20.31 (0.77)	24.04 (1.63)	8.94 (0.34)	9.45 (0.16)	10.73 (0.19)	26.22 (1.99)	16.37 (0.31)	11.94 (0.58)	18.68 (0.95)	13.00 (0.66)	16.80 (0.66)	4.92 (0.34)	0.12 (0.04)	41.34 (3.15)	38.37 (2.58)
Er	21.09 (2.04)	19.27 (1.50)	25.73 (2.49)	8.76 (0.52)	9.29 (0.23)	10.37 (0.36)	31.41 (3.07)	17.27 (0.52)	9.07 (0.62)	13.59 (0.91)	9.88 (0.68)	14.52 (0.87)	4.52 (0.62)	0.28 (0.20)	38.79 (4.89)	51.64 (8.09)
Ju	14.67 (0.79)	20.11 (0.43)	18.24 (0.99)	9.01 (0.25)	11.20 (0.08)	11.49 (0.15)	19.76 (1.25)	16.87 (0.22)	16.74 (0.48)	25.86 (0.79)	18.23 (0.51)	22.08 (0.51)	6.88 (0.21)	0.17 (0.05)	61.11 (1.84)	50.82 (1.67)
Sp.	Nitrate				pH				Ammonification				Nitrification			
	Nov	Feb	Apr	Jul	Nov	Feb	Apr	Jul	Nov	Feb	Apr	Jul	Nov	Feb	Apr	Jul
Ca	0.42 (0.06)	0.36 (0.08)	7.70 (0.27)	58.75 (3.65)	6.80 (0.07)	6.11 (0.12)	6.46 (0.05)	5.64 (0.14)	2.25 (0.07)	0.53 (0.02)	13.99 (0.24)	0.50 (0.02)	2.90 (0.06)	0.89 (0.02)	3.05 (0.06)	0.87 (0.02)
Cl	0.55 (0.55)	0.00 (0.00)	6.45 (3.53)	62.75 (8.05)	6.75 (0.35)	6.52 (0.67)	6.56 (0.53)	6.74 (2.11)	1.88 (0.32)	0.48 (0.14)	14.17 (3.50)	0.48 (0.14)	2.41 (0.23)	0.98 (0.03)	2.69 (0.05)	0.96 (0.03)
Cm	0.22 (0.04)	0.39 (0.07)	7.70 (0.21)	55.06 (2.27)	6.78 (0.06)	6.09 (0.08)	6.51 (0.05)	5.50 (0.12)	2.11 (0.07)	0.54 (0.01)	14.33 (0.20)	0.49 (0.01)	2.98 (0.06)	0.88 (0.02)	3.08 (0.06)	0.85 (0.02)

(continued on next page)

Appendix (continued)

Sp.	Soil moisture			Soil temperature			Organic matter			Ammonium		
	Nov	Feb	Apr	Jul	Nov	Feb	Apr	Jul	Nov	Feb	Apr	Jul
La	0.00 (0.00)	0.00 (0.00)	9.04 (3.68)	45.22 (24.44)	6.72 (0.42)	6.15 (0.39)	6.60 (0.50)	5.83 (0.85)	1.71 (0.02)	0.66 (0.22)	13.40 (2.58)	0.66 (0.22)
Ro	0.96 (0.09)	0.15 (0.06)	8.06 (0.39)	66.48 (5.61)	6.97 (0.07)	6.33 (0.11)	6.56 (0.06)	5.66 (0.21)	2.56 (0.11)	0.57 (0.02)	14.04 (0.26)	0.51 (0.02)
Ce	2.42 (0.11)	0.36 (0.25)	6.74 (0.34)	33.47 (2.11)	7.48 (0.07)	6.39 (0.15)	7.82 (0.13)	6.19 (0.23)	3.72 (0.20)	0.53 (0.02)	15.98 (0.61)	0.43 (0.02)
Mu	1.02 (0.19)	0.23 (0.12)	3.01 (0.53)	15.04 (2.95)	7.05 (0.08)	6.46 (0.13)	7.94 (0.20)	7.06 (0.50)	2.39 (0.14)	0.46 (0.04)	14.97 (0.46)	0.40 (0.04)
Oi	1.88 (0.13)	0.29 (0.08)	5.20 (0.38)	26.28 (1.87)	7.46 (0.09)	6.87 (0.14)	8.29 (0.12)	6.92 (0.22)	2.99 (0.14)	0.43 (0.02)	16.55 (0.36)	0.39 (0.02)
Ph	1.89 (0.06)	0.30 (0.07)	5.22 (0.18)	28.62 (1.98)	7.15 (0.03)	6.58 (0.06)	7.69 (0.07)	6.48 (0.16)	2.82 (0.09)	0.46 (0.01)	16.01 (0.21)	0.41 (0.01)
Pi	1.68 (0.07)	0.23 (0.06)	4.63 (0.21)	24.83 (2.37)	7.31 (0.09)	6.70 (0.09)	8.03 (0.12)	6.81 (0.19)	2.77 (0.13)	0.43 (0.02)	16.38 (0.32)	0.39 (0.01)
Rh	0.85 (0.10)	0.15 (0.05)	2.37 (0.26)	15.34 (3.08)	7.28 (0.07)	6.63 (0.09)	8.01 (0.09)	8.32 (0.24)	2.80 (0.10)	0.36 (0.02)	16.17 (0.30)	0.32 (0.02)
Er	0.00 (0.00)	0.35 (0.25)	0.00 (0.00)	13.48 (6.00)	6.17 (0.44)	5.65 (0.45)	6.84 (0.49)	8.42 (0.38)	2.03 (0.24)	0.43 (0.05)	14.83 (1.00)	0.42 (0.05)
Ju	1.74 (0.07)	0.22 (0.06)	4.80 (0.19)	27.63 (2.08)	7.62 (0.08)	7.01 (0.09)	8.37 (0.10)	6.74 (0.14)	3.38 (0.09)	0.46 (0.01)	17.00 (0.23)	0.42 (0.01)

This paper results from the ESF-FWF Conference on Reduced Nitrogen in Ecology and the Environment, organized in the Universitätszentrum Obergurgl, Austria on October 14–18 2006 (<http://www.esf.org/conferences/lc06203>). This conference was organised by the European Science Foundation (ESF) in partnership with the Fonds zur Förderung der wissenschaftlichen Forschung in Österreich (FWF) and the Leopold-Franzens-Universität Innsbruck (LFUI). We also acknowledge the support of the COST Action 729, and the ESF program Nitrogen in Europe (NinE).

References

- Alef, K., Kleiner, D., 1987. Applicability of arginine ammonification as indicator of microbial activity in different soils. *Biology and Fertility of Soils* 5, 148–151.
- Carreira, J.A., Niell, F.X., Lajtha, K., 1994. Soil nitrogen availability and nitrification in Mediterranean shrublands of varying fire history and successional stage. *Biogeochemistry* 26, 189–209.
- Castaldi, S., Aragosa, D., 2002. Factors affecting nitrification and denitrification variability in a natural and fire-disturbed Mediterranean Shrubland. *Biology and Fertilization of Soils* 36, 418–425.
- Castells, E., Peñuelas, J., 2003. Is there a feedback between N availability in siliceous and calcareous soils and *Cistus albidus* leaf chemical composition? *Oecologia* 136, 183–192.
- Chapin, F.S., 2000. Driving forces for changes of biodiversity. *Nature* 405, 234–242.
- Clarke, K.R., 1993. Nonparametric multivariate analyses of changes in community structure. *Australian Journal of Ecology* 18, 117–143.
- Clarke, K.R., Green, R.H., 1988. Statistical design and analysis for a biological effects study. *Marine Ecology Programme Series* 46, 213–226.
- Correia, O., Catarino, F.C., 1994. Seasonal changes in soil-to-leaf resistance in *Cistus* sp. *Pistacia lentiscus*. *Acta Oecologica* 15, 289–300.
- Correia, O., Martins, A.C., Catarino, F., 1992. Comparative phenology and seasonal nitrogen variation in Mediterranean species of Portugal. *Ecologia Mediterranea* 18, 7–18.
- Cruz, C., Martins-Loução, M.A., 2000. Nitrogen in a sustainable environment: a matter of integration. In: Martins-Loução, M.A., Lips, S.H. (Eds.), *Nitrogen in a Sustainable Ecosystem*. Backhuys, The Netherlands, pp. 415–419.
- Cruz, C., Dias, T., Matos, S., Tavares, A., Neto, D., Martins-Loução, M.A., 2003. Nitrogen availability and plant cover: the importance of nitrogen pools. In: Tiezzi, E., Brebbia, C.A., Usó, J.L. (Eds.), *Ecosystems and Sustainable Development IV*. Witpress, Southampton, pp. 123–135.
- Cruz, C., Bio, A.M.F., Dominguez-Valdivia, M.D., Aparicio-Tejo, P.M., Lamsfus, C., Martins-Loução, M.A., 2006. How does glutamine synthetase activity determine plant tolerance to ammonium? *Planta* 223, 1068–1080.
- Farley, R.A., Fitter, A.H., 1999. Temporal and spatial variation in soil resources in a deciduous woodland. *Journal of Ecology* 87, 688–696.
- Fitter, A.H., Hay, R.K.M., 2002. *Environmental Physiology of Plants*. Academic Press, London.
- Gallardo, A., Menino, J., 1992. Nitrogen immobilization in leaf litter at two mediterranean ecosystems of SW Spain. *Biogeochemistry* 15, 213–228.
- Gallardo, A., Paramá, R., Covel, F., 2005. Soil ammonium vs. nitrate spatial pattern in six plant communities: simulated effect on plant populations. *Plant and Soil* 277, 201–219.
- Grove, A.T., Rackham, O., 2001. *The Nature of Mediterranean Europe*. Yale University Press, London.
- Guo, D., Mou, Pu, Jones, R.H., Mitchell, R.J., 2004. Spatio-temporal patterns of soil available nutrients following experimental disturbance in a pine forest. *Oecologia* 138, 613–621.
- Hedlund, H., Santa-Regina, I., Van Der Putten, W.H., Leps, J., Dias, T., Korthals, G.W., Lavorel, S., Brown, V.K., Gormsen, D., Mortimer, S.R., Rodriguez-Barrueco, C., Roy, Y.J., Smilauer, P., Smilauerova, M., Van

- Dijk, C., 2003. Plant species diversity, plant biomass and responses of the soil community on abandoned land across Europe: idiosyncrasy or above-belowground time lags. *Oikos* 103, 45–58.
- Hodge, A., 2003. N capture by *Plantago lanceolata* and *Brassica napus* from organic material: the influence of spatial dispersion, plant competition and an arbuscular mycorrhizal fungus. *Journal of Experimental Botany* 54, 2331–2342.
- Hook, P.B., Burke, I.C., Lauenroth, W.K., 1991. Heterogeneity of soil and plant N and C associated with individual plants and openings in North American shortgrass steppe. *Plant and Soil* 138, 247–256.
- Inouye, R.S., Huntly, N.J., Tilman, D., Tester, J.R., 1987. Pocket gophers (*Geomys bursarius*), vegetation, and soil nitrogen along a successional sere in east central Minnesota. *Oecologia* 72, 178–184.
- Jackson, R.B., Caldwell, M.M., 1993. The scale of nutrient heterogeneity around individual plants and its quantification with Geostatistics. *Ecology* 74, 612–614.
- Joffre, R., Ramball, S., 1993. How tree cover influences the water balance of Mediterranean Rangelands. *Ecology* 74, 570–582.
- Kandeler, E., 1996. Nitrification during long term incubation. In: Schinner, F., Kandeler, E., Ohlinger, R., Margesin, R. (Eds.), *Methods in Soil Biology*. Springer, Berlin, pp. 149–151.
- Lavelle, P., 1997. Faunal activities and soil processes: adaptive strategies that determine ecosystem function. *Advances in Ecological Research* 27, 93–132.
- Lo Gullo, M.A., Salleo, S., 1988. Different strategies of drought resistance in three mediterranean sclerophyllous growing in the same environmental conditions. *New Phytologist* 108, 267–276.
- Löscher, R., Tenhunen, J.D., Pereira, J.S., Lange, O.L., 1982. Diurnal courses of stomatal resistance and transpiration of wild and cultivated Mediterranean perennials at the end of the summer dry season in Portugal. *Flora* 172, 138–160.
- Marschner, H., 1995. *Mineral Nutrition of Higher Plants*. Academic Press, London.
- McKane, R.B., Johnson, L.C., Shaver, G.R., Nadelhoffer, K.J., Rastetter, E.B., 2002. Resource based niches provide a basis for plant species diversity and dominance in arctic tundra. *Nature* 415, 68–70.
- Min, X., Siddiqi, M.Y., Guy, R.D., Glass, A.D.M., Kronzucker, H.J., 2000. A comparative kinetic analysis of nitrate and ammonium influx in two early-successional tree species of temperate and boreal forest ecosystems. *Plant Cell and Environment* 23, 321–328.
- Montagini, F., Haines, B.L., Boring, L.R., Swank, W., 1986. Nitrification potentials in early successional black locust and in mixed hardwood forest stands in the southern Appalachians, USA. *Biogeochemistry* 2, 197–210.
- Mooney, H.A., 1983. Carbon gaining capacity and allocation patterns of Mediterranean-climate plants. In: Kruger, F.J., Mitchell, D.T., Jarvis, J.U.M. (Eds.), *Mediterranean-Type Ecosystems. The Role of Nutrients*. Springer, Berlin, pp. 103–119.
- Peñuelas, J., Fillela, I., 2003. Deuterium labelling of roots provides evidence of deep water access and hydraulic lift by *Pinus nigra* in a Mediterranean forest of NE Spain. *Environmental and Experimental Botany* 49, 201–208.
- R Development Core Team, 2005. R: A language and environment for statistical computing. R Foundation for Statistical Computing. Vienna, Austria, URL: <http://www.R-project.org>.
- Rutigliano, F.A., Ascoli, R.D., de Santo, A.V., 2004. Soil microbial metabolism and nutrient status in a mediterranean area as affected by plant cover. *Soil Biology and Biochemistry* 36, 1719–1729.
- Ryel, R.J., Caldwell, M.M., Manwaring, J.H., 1996. Temporal dynamics of soil spatial heterogeneity in sagebrush-wheatgrass steppe during a growing season. *Plant and Soil* 184, 299–309.
- Stark, J., Schimel, J., 2001. Errors in overestimating of gross N transformation rates in grassland soils. *Soil Biology and Biochemistry* 33, 1433–1435.
- Schlesinger, W.H., Raikes, J.A., Hartley, A.E., Cross, A.F., 1996. On the spatial pattern of soil nutrients in desert ecosystems. *Ecology* 77, 364–374.
- Specht, R.L., Rundel, P.W., Westman, W.E., Catting, P.C., Majer, J.D., Greenslade, P., 1988. *Mediterranean Type Ecosystems. A Data Source Book*. Kluwer Academic Publishers, Dordrecht.
- Ter Braak, C.J.F., Smilauer, P., 1998. *CANOCO Reference Manual and User's Guide to Canoco: Software for Canonical Community Ordination (version 4)*. Centre for Wageningen, Wageningen.
- Thompson, J.D., 2005. *Plant Evolution in the Mediterranean*. Oxford University Press, New York.
- Tilman, D., 1989. Competition, nutrient reduction and the competitive neighbourhood of a bunchgrass. *Functional Ecology* 3, 215–219.
- Tinker, P.B., Nye, P.H., 2000. *Solute Movement in the Rhizosphere*. University Press, Oxford.
- Werner, C., Correia, C., Beyschlag, W., 1999. Two different strategies of Mediterranean macchia plants to avoid photoinhibition damage by excessive radiation levels during summer drought. *Acta Oecologica* 20, 15–23.
- Yaalon, D.H., 1997. Soils in the Mediterranean region: what makes them different? *Catena* 28, 157–169.