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Soil nitrogen availability and nitrification in Mediterranean shrublands of varying fire history and successional stage

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Abstract. The short-term effect of a single fire, and the long-term effect of recent fire history and successional stage on total and mineral N concentration, net nitrogen mineralization, and nitrification were evaluated in soils from a steep semi-arid shrubland chronosequence in southeast Spain. A single fire significantly increased soil mineral N availability and net nitrification. Increasing fire frequency in the last few decades was associated with a sharp decrease in surface soil organic matter and total N concentrations and pools, and with changes in the long-term N dynamic patterns. The surface-soil extractable $NH_4^+:NO_3^-$ ratio increased throughout the chronosequence. All net mineralized N in laboratory incubations from all sites was converted to nitrate, suggesting that allelochemic inhibition of net nitrification is probably not important in this system. Net nitrification in samples during incubation increased through the sere. The maximum rate of net nitrification (k_{max}) increased through the first three stages of the sere. A linear relationship was found between total soil N and N mineralization, and both k_{max} and net nitrification for the first three stages of the sere, suggesting that total N and ammonification are likely to be the control mechanisms of nitrification within the sere. The oldest site exhibited the lowest specific k_{max} and the highest potential soil respiration rate suggesting that a lower N quality and increasing competition for ammonium might also limit nitrification at least in the long-unburned garrigue site.

Introduction

Fire intensity and burn frequency can strongly influence many ecosystem processes including the biogeochemical cycling of essential nutrients (Rundel 1983). It is thus important to distinguish between short-term changes occurring after a single fire event and the longer-term ecological effects resulting from different fire regimes (such as fire intensity, frequency and season of burn; Raison 1979).

Among the major nutrients, N is likely to be the most sensitive to fire disturbance (Christensen 1973). Significant increases in soil nitrate almost invariably have been found a few weeks after fire in mediterranean-type ecosystems such as the California chaparral (Rundel 1983), Israel pine forests (Kutiel & Naveh 1987) and the greek phrygana (Arianoutsou & Margaris 1982). Ammonium and readily decomposable organic N compounds added with ash deposition, along with post-fire temperature and moisture increases, may promote soil N mineralization and nitrification (Raison 1979). On the other hand, significant amounts of N can be volatilized (DeBell & Ralston 1970) or lost through erosion (DeBano 1976), especially in systems such as mediterranean shrublands where the largest fraction of the total N reserves accumulate in surface organic pools (Post et al. 1986). Thus, despite temporary post-fire enhanced mineral N availability, repeated burning in semi-arid steep areas may lead to a substantial depletion of the total N reserves of the ecosystem, causing marked changes in the long-term patterns of N cycling.

Considerable attention has been given in recent years to the study of changes in patterns of N cycling and nitrification in the face of disturbance and with successional development (e.g. Robertson & Vitousek 1981, Donaldson & Henderson 1990, Frazer et al. 1990), although few studies have specifically focused on fire disturbance. Most fire-related studies have considered only short-term changes, and few have dealt with longer-term effects of fire frequency on N availability, or interactions between repeated fires and interval successional processes (e.g. Covington & Sackett 1986). In this paper we analyze changes in soil N concentrations and net nitrification across a fire-related, semi-arid Mediterranean soil and vegetation chronosequence. Such a chronosequence provides the opportunity to assess variations in N cycling when either a single fire, or compounded post-fire successional age and fire frequency in the last decades are sources of variation.

Methods

Study sites

The study was conducted in the Hunting National Reserve of Sierra de Almijara, Malaga, Spain, in the upper Torrox river basin (36° 50′N, 3°57′E). The area, located in the boundary of the xeric southeastern of the Iberian peninsula, has a thermic semi-arid mediterranean climate (annual mean air temperature is 17–19 °C; annual precipitation is 350–600 mm, occurring mainly as torrential rain from November to March). The relief is characterized by discontinuous narrow ridges and deep V-shaped stream-cut valleys with numerous small steep sub-basins. Bedrocks consist of dolomitic kakiritized marble. Most soils are shallow sandy rendzinas with a very high carbonate content. Vegetation includes garrigue communities with *Juniperus oxycedrus, Rhamnus oleoides, Pistacia lentiscus* and *Quercus coccifera,* replaced by region-specific gorse-scrublands with *Cistus clusii, Ulex rivasgodayanus* and *Rosmarinus officinalis* as dominant species in frequently burned areas.

Four stands of at least 1 ha were choosen as sampling sites in adjacent, small, steep sub-basins, facing SE at between 300–400 m in elevation. They represented a chronosequence in which differences in time since the last burn and differences in fire frequency were compounded (Table 1). Since

Table 1. General characteristics of the recent fire history, and selected physical and chemical properties of surface soils at the four sampling units. Values of texture and cation exchange capacity listed above are means (±SD) of two subsite composites at each sampling unit, and of three subsites with three replicates per subsite for water holding capacity ($n = 3$). Values for the soil properties listed below are unweighted means (seasonal coefficient of variation, %) for the whole sequential sampling period (see text for further details). Within each column, different superscript letters indicate significantly different sites ($P < 0.05$, Tukey's HSD test following ANOVA).

Site	Years since last fire	No of fires since 1975	Vegetation cover (%)	Litter mass (g m^{-2})	Fraction >2 mm (volume %)	Bulk density (g cm^{-3})	Sand (%)	Clay (%)	C.E.C. (ME/100 g)	Base saturation (%)	Waterholding capacity (%)
Unit A	4	3	18	131	32	1.40	88.7 (3.0)	3.1 (0.7)	9.2 (1.5)	91	32.0 (2.1)
Unit B	9	2	55	252	19	1.21	59.0 (2.3)	8.8 (1.8)	31.2 (0.6)	100	34.9 (4.3)
Unit C	15	1	83	480	14	1.19	77.5 (4.2)	7.0 (0.5)	33.1 (3.0)	77	35.8 (2.0)
Unit D	32	0	94	985	7	1.04	74.8 (8.5)	6.6 (2.7)	32.7 (2.1)	78	37.9 (1.6)

Site	Soil moisture (%)	pH (1:1 soil: water)	Organic C (%)	Total N (mg/g)	Soil C/N	Litter C/N	NO_3-N (μg/g)	NH_4-N (μg/g)
Unit A	4.2^{a} (71.8)	8.1^{a} (0.9)	1.15^{a} (23.6)	0.80^{a} (35.0)	15.3^{a} (27.8)	71.1^{a} (21.4)	2.61^{a} (55.2)	3.34^{a} (17.6)
Unit B	10.0^{b} (55.2)	8.0^{a} (1.1)	4.21^{b} (13.5)	2.73^{b} (21.5)	15.9^{a} (13.5)	45.7^{b} (24.1)	3.37^{b} (76.4)	12.3^{b} (13.6)
Unit C	12.1^{c} (52.3)	8.0^{a} (0.7)	4.58^{c} (10.3)	3.50^{c} (15.3)	13.0^{b} (5.15)	35.0^{c} (16.4)	2.79^{ab} (69.9)	12.9^{bc} (10.7)
Unit D	15.3^{d} (54.2)	8.0^{a} (1.0)	5.31^{d} (8.31)	3.08^{bc} (8.14)	17.1^{c} (5.01)	34.1^{c} (11.3)	1.21^{c} (49.5)	14.1^{c} (15.8)

the chronosequence of stands resulted from fire caused by chance, no true replication of sites was possible.

One of the sub-basins (Unit D) had not been burned for at least 30 years when sampling began in 1988, and was selected as a reference long-unburned plot. At this site, vegetation consisted of a mature, dense (94% cover) garrigue with *Juniperus oxycedrus* and *Buxus balearicae*, some pine (*Pinus halepensis*), and small shrub species such as *Ulex rivasgodayanus* and *Rosmarinus officinalis*. Mean slope of the sampling site was 38° (±2°). The soils were slightly reddish, sandy clay loam to sandy loam, entic Haploxerolls (Carreira 1992).

The other three basins were affected by a forest fire that spread over 12,000 ha in the summer of 1975. One of them (unit C) had not reburned by our sampling date; vegetation consisted of a 13-yr-old mixed garrigue-gorse scrubland (83% shrub cover, 3064 g/m^2 of aboveground biomass, slope 41° ± 4°; Carreira 1992). Dominant species were *Genista spartioides, Rosmarinus officinalis, Ulex rivasgodayanus* and *Cistus clusii*, but *Juniperus oxycedrus* and *Buxus balearicae* were still important (ca. 30% of aboveground biomass). Soils were slightly shallower than in unit D and consisted of sandy loam entic haploxerolls.

The remaining two basins (Units A and B) were burned by chance in the summer of 1981. Unit B had not reburned by our sampling date. Vegetation was an 7-yr-old gorse-scrubland almost exclusively made up of *Rosmarinus officinalis, Ulex rivasgodayanus* and *Cistus clusii*. These three species accounted for more than 60% of the total aboveground biomass. Total shrub cover was 55%. Mean slope of the plot was 36°. Soil type, gravelly with sandy loam texture, varied from entic haploxeroll to typic xerorthent.

Unit A was affected by a third wild-fire in summer 1986. At the beginning of the sequential sampling period, vegetation was a 2-yr-old open regenerating gorse-scrubland made up of the same shruby species already cited but considerably enriched with very scattered herbaceous species colonizing the interespaces of bare soil (mean slope 38°, vegetation cover 18%). Soils, loamy sandy to sandy typic xerorthents, were highly eroded.

N-fixers such as *Ulex* and *Genista* accounted for 18.5, 35.8 and 32.8% of the aboveground biomass in units A, B and C, respectively (Carreira & Niell 1992). More information about the plant species composition of the sites and post-fire vegetation dynamic is given elsewhere (Carreira et al. 1991).

We hypothesized that these four stands represent a true chronosequence that combines the degradative effects of repeated fires and the aggradative effects of successional development. Several lines of evidence suggest that prior to 1975 all the four adjacent sub-basins had essentially the same type of vegetation and surface soil characteristics. Aerial photographs and reports from the Spanish Forest Service (ICONA) reveal that all the upper Torrox river basin was occupied by a uniform open *Pinus pinaster* forest with abundant understory vegetation. The area, a Hunting Reserve, was only minimally managed and grazing was excluded. In all the burned units a few scattered old individuals of *Pinus pinaster*, similar in size, still remain at present. In all cases, parent rock is a kakiritized dolomitic marble (Elorza et al. 1979). All sampling plots are within 1–3 km of each other and were located at similar elevation, aspect and slope within the small subbasins. Species composition was essentially the same in all units though with different proportions of abundance (Carreira et al. 1991). These factors, in addition to the consistent trends found along the chronosequence for such variables as plant tissue element concentration, phosphorus biogeochemistry, erosion and decomposition (Carreira 1992) give confidence that surface soil differences among sampling units are not merely due to preexisting variation but are due to their distinctive fire history and stand successional age.

In addition to this chronosequence, we sampled two recently burned sites. The first was a prescribed burn that was conducted in a sub-plot of unit B in the spring of 1988. More information about the prescribed fire, nutrient budgets and post-fire regeneration can be found in Carreira (1992) and Carreira et al. (1992). The second was a 3000 ha wild-fire that affected an area near the sampling units that was occupied by a gorse-scrubland vegetation similar to that found in units B and C.

Chronosequence soil sampling and analysis

To assess seasonal changes in surface soil N concentrations in relation to fire frequency, the four sampling units were sampled at 1–2 month intervals from October 1988 to November 1989. Five subsites were randomly located in each unit, and surface litter and soil samples (0–5 cm) were collected by excavating within a 0.16 m^2 frame. In each sampling unit, one maximum-minimum thermometer was placed beneath the canopy and another was buried between 0–5 cm into the soil. Monthly max/min temperatures were recorded. Rain gauges were also installed in each of the sampling units. Bulk density and gravel content were measured at the beginning of the sampling period by means of the sand funnel procedure (Blake and Hartge 1986).

At each sampling interval, soil samples were sieved to <2 mm and air-dried. A 50 g subsample of each soil sample was oven-dried (104 °C, 24h) to calculate moisture content. Total N was measured in ground samples using a 240-C Perkin-Elmer CNH analyzer. Organic carbon was analyzed by the Walkley-Black procedure (Jackson 1976). Mineral N was extracted from 10g samples by shaking and equilibrating over-night in 100 ml of IN KCl followed by centrifugation and Whatman GF/C paper filtration. Soil extracts were stored at -18 °C prior to analysis. Nitrite, nitrate and ammonium concentrations were determined colorimetrically with a Technicon Autoanalyzer II system (Technicon Instrument 1977, 1978).

Soil sampling and analysis in recently burned sites

To assess short-term changes in surface soil N concentrations after a single fire, nine surface soil samples (0–5 cm) were randomly collected from within a 400 m^2 plot in the prescribed burn site before and immediately after the prescribed burning. Then, sampling was repeated every two weeks for the following three months and the nine samples were composited into three. Two months after the fire, soils in the unburned areas adjacent to the prescribed burning plot were also sampled and used as reference soils. Samples were measured for organic C, total N and extractable nitrate and ammonium as described above.

Soils from the wild-fire and its adjacent unburned areas were collected one week after the fire and were subject to laboratory incubations to measure N mineralization and nitrification rates.

Net Nitrogen Mineralization and Nitrification Estimates

In each of the four sampling units, as well as in the 3000 ha wild-fire and its adjacent unburned plots, three subplots were randomly located along a 100-m transect. Five soil samples (0–5 cm depth) were collected from within a 2-m diameter circle at each subplot in May (vegetation stands of units A, B, C and D were 4, 9, 15 and at least 32 years old, respectively, when this sampling took place; the fire in the wild-fire site had occurred one week before). The five samples were composited per subplot (n = 3), mixed in polyethylene bags and stored in an isothermic icebox for immediate transport to the laboratory. All samples were processed within 48-h after collection. Undried samples were sieved to <2 mm. Percent water was determined for each subsite composite by drying 50-g replicates for 48 h in an 80 °C oven. Water holding capacity (WHC) was estimated using a modification of the technique reported by Peters (1965). Initial concentrations of nitrite, nitrate and ammonium were analyzed in 1N KCl extracts of undried subsamples, and total N in air-dried ground subsamples, as described above.

To study the effect of moisture and temperature on net nitrification and N mineralization, 30-g subsamples were brought to approximately either 75 and 35% of WHC with distilled water, and were placed in polyethylene cups capped with a snap-on lid that had a 5-mm hole punched near its center. The cups were incubated for 1 month in the dark in a controlled-environment cabinet at 10, 20 and 30 °C. Water loss was monitored gravimetrically and original water content in all cups was restored twice a week. Net N mineralization and nitrification rates were estimated from the difference in mineral N and nitrate concentration of the soil before and after incubation.

To characterize the time-course of nitrate production, 50-g subsamples were incubated in the dark for up to 64 days (75% WHC, 20 °C). Approximately every 10 days, subsamples were removed from each cup and analyzed for mineral N. At the middle of the incubation period, the rate of CO_2 release was measured placing the samples in a small temperature and humidity controlled chamber connected to an ADC 225 InfraRed Gas Analyzer. Soil NO_3^--N concentrations were plotted against incubation time, and polynomial curves were fitted by least squares regression and characterized using the analysis proposed by Donaldson and Henderson (1990) on the data obtained within the time frame of the incubation experiment. The mathematical function of each soil sample incubation was used to calculate the following parameters describing the sample's potential to produce nitrate: The maximum rate of net nitrification (k_{max}) is defined as the maximum value of the first derivative of the fitted curves; and the delay period (t_o) as the x-intercept of the maximum rate line. When a negative value of the x-intercept was found, the delay period was computed as $t_o = 0$. Saturation time (T) is defined as the period comprised between the beginning of the incubation and the moment at which net nitrate production stops (when k = 0). The NO_3^--N concentration at time T (or the maximum value reached during the time frame of the experiment if nitrate production did not stop, in which case it would be a conservative estimate) was used as an index of the net nitrification potential (NP).

Statistical analysis

The significance of differences between means of soil properties for samples collected before and immediately after the prescribed burn was assessed using Student's t-tests ($n = 9$). To examine the effects of "site" and/or "sampling date", all data were analyzed using two-way or one-way ANOVA. When null hypothesis was rejected ($p < 0.05$), Tukey's HSD tests were used to asses for differences between means. Because the subplots in each site were considered replicates, our design is pseudo-replicated and results must be viewed with that in mind. The extent of seasonal variability in soil properties was evaluated as the coefficient of variation between means for the different sampling dates. Correlation between soil properties was calculated as Spearman product-moment coefficients of correlation. Error propagation was used to compute standard errors for soil and litter N and C pool size data that were calculated from the C and N data and separate measurements of bulk density and litter mass.

Results

Effects of recent fire frequency

General properties of soils of the chronosequence

Relevant physical and chemical properties of surface soils from the four chronosequence units revealed marked trends along the disturbance gradient (Table 1). The % by volume of the >2 mm fraction and the bulk density of the fine earth fraction increased with increasing fire frequency. Soil texture changed from sandy clay loam to loamy sandy, following observed differences in water holding capacity. In general, extreme values found in the most frequently burned site (Unit A) accounted for most of the significant variations in soil chemistry along the chronosequence. All soils were mildly alkaline (pH = 8.0–8.1). Cation exchange capacity increased and base saturation decreased through the sere (Table 1).

Changes in moisture and temperature

The older sites showed much stronger seasonal patterns in soil and litter moisture, and much less seasonal variation in soil temperature (Fig. 1), as can be expected from sites with nearly full canopy closure and higher organic matter content. Surface soil moisture varied significantly between sites and sampling dates ($p < 0.01$). Mean soil moisture increased from 4.2% in Unit A to 15.3% in Unit D (Table 1). There was a marked contrast between the wet (November to March) and the dry (May to September) seasons in all sampling units (Fig. 1). Similar patterns were found for litter moisture (annual means of 9.2, 9.8, 17.9 and 24.1% for units A to D respectively). The absolute difference between surface soil monthly max/min temperatures diminished through the sequence, with the lowest maximums and the highest minimums almost always found in the mature garrigue site (Unit D). Surface soils of Unit A and Unit B reached very high temperatures during the dry summer season (Fig. 1), presumably due to lower vegetation cover.

Changes in organic C and total N

Organic C and N concentrations of the soils varied both between sites and sampling dates ($p < 0.01$, except $p < 0.05$ for the effect of date on total N). Mean C concentrations decreased significantly with increasing degree of fire disturbance, from 5.3% in Unit D to 1.15% in Unit A (Table 1). Total N showed a similar trend to that of organic C ($r = 0.941$). Very low values were found at the most eroded, frequently burned site, in contrast with those of older stands. Since both variables increased at similar rates through the chronosequence, the C/N ratio changed very little except for Unit D where a total N concentration slightly lower than that in Unit C accounted for the

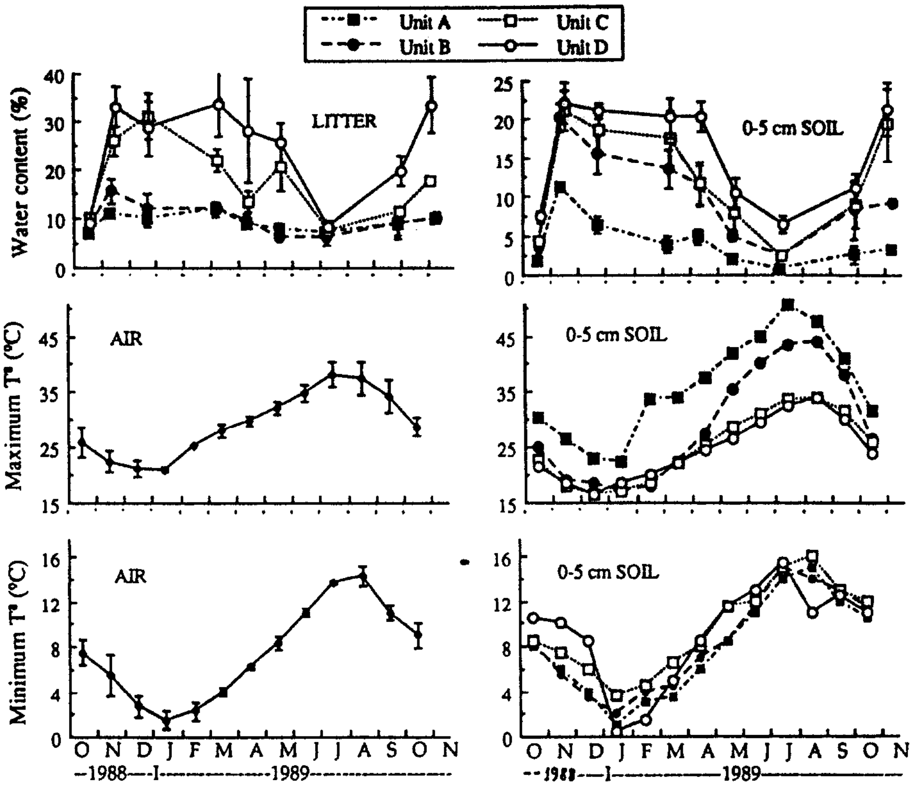

Fig. 1. Seasonal changes in percent moisture of litter and 0–5 cm soil samples, and monthly maximum and minimum temperatures under the canopy and at 2.5 cm depth into the soils, in the four chronosequence units. Moisture values are the mean ± standard deviation of five subsites per site.

significantly higher C/N found in that site. The extent of seasonal variation decreased along the sere for both organic C and total N (Table 1). Except for Unit A, values of total N in soils were slightly lower in autumn (November) and spring (April) compared to mid-winter and summer ($P < 0.05$). Soil C/N showed very little seasonal variation in Unit B, C and D, in contrast to the very sharp and irregular pattern of variation found in Unit A (Fig. 2). Litter C/N significantly decreased through the sequence (Table 1).

Pools of organic C and total N in the 0–5 cm soil and litter were calculated from actual C and N data, and values of % volume of the fine earth fraction, bulk density and litter mass presented in Table 1. Trends similar to those already reported for concentrations held through the chronosequence. Litter N pools were 12.4 g m^{-2} in unit D, 5.9 g m^{-2} in unit C, 2.5 g m^{-2} in unit B and 0.9 g m^{-2} in unit A. When expressed as % of pool size in the long-

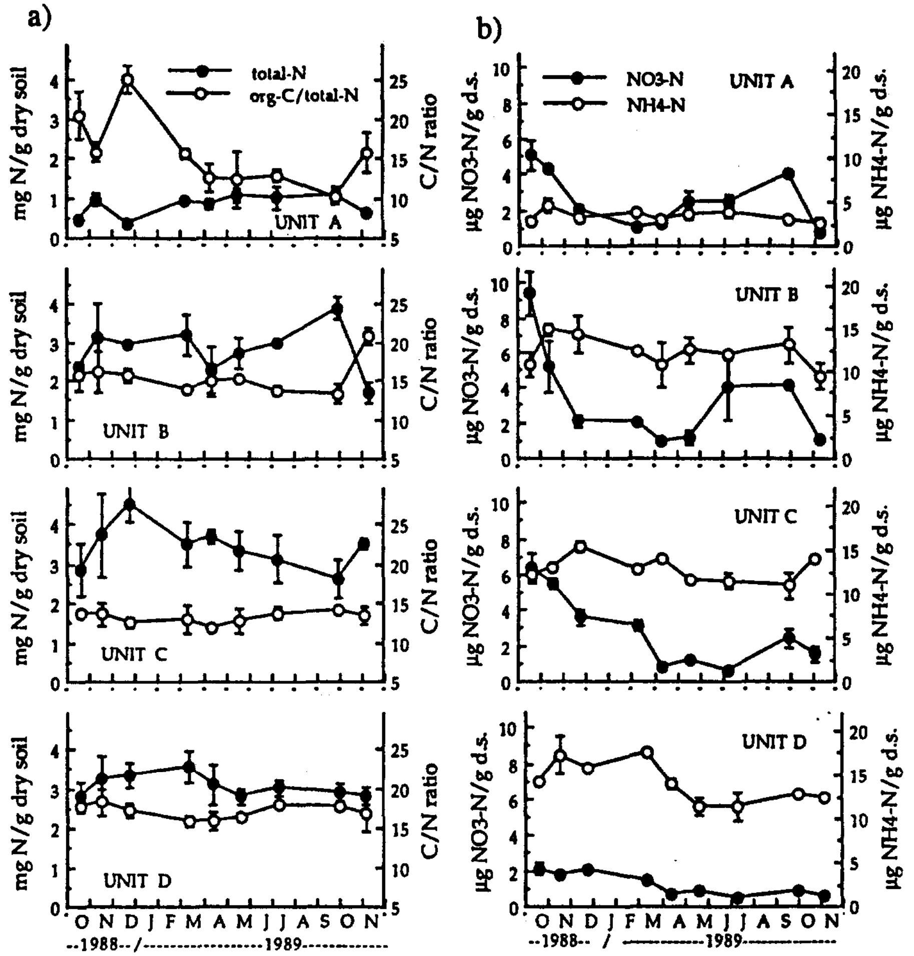

Fig. 2. Seasonal changes in (a) the C/N ratio and total N concentration, and (b) extractable NO_3^- and NH_4^+ concentrations of 0–5 cm surface soils in the four chronosequence units. Values are the mean ± standard deviation of 5 to 2 subsite composites per site.

unburned site, litter N pools exponentially decreased with increasing annual fire probability (Fig. 3). Surface soil organic C pools were 2.6 kg m^{-2} in unit D, 2.3 kg m^{-2} in unit C, 2.1 kg m^{-2} in unit B and 0.6 kg m^{-2} in unit A. Surface soil N pools were 38.5 g m^{-2} in unit A, 134.9 g m^{-2} in unit B, 179.1 g m^{-2} in unit C and 150.1 g m^{-2} in unit D. A low to medium fire frequency has little effect in soil organic C pools, and may even promote soil N pools, compared to pool sizes in the long-unburned site (Fig. 3). However, soil C and N pools are clearly depleted in the most frequently burned site (unit A).

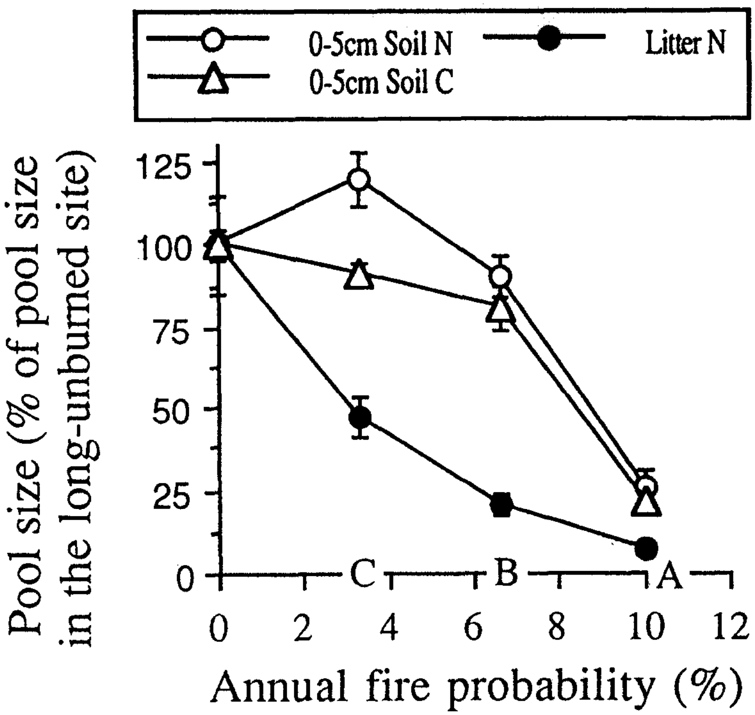

Fig. 3. Pool sizes of litter N and 0–5 cm soil C and N as a function of site annual fire probability (over the last 30 years), expressed as percent change from the long-unburned site (unit D).

Changes in mineral N concentrations

NO_3^- and NH_4^+ concentrations in the top 0–5 cm of soil varied significantly with site ($P < 0.01$), sampling date ($P < 0.01$) and the site*date interaction ($P < 0.01$ for NO_3^-, $P < 0.05$ for NH_4^+). Mean NH_4^+ concentrations increased from 3.3 μg NH_4^+-N g^{-1} in the regenerating open gorse scrubland to 14.1 μg NH_4^+-N g^{-1} in the oldest garrigue stage (Table 1). Seasonal variation in NH_4^+ was relatively muted, especially in comparison with NO_3^-, and resembled that of total N and organic C in all sampling units ($r = 0.934$ and $r = 0.973$, respectively; Fig. 2). Seasonally-averaged NO_3^- concentrations slightly increased from Units A to B, and then decreased through the last three stages of the chronosequence (Table 1). A regular pattern of seasonal variation was observed for NO_3^- in all sampling units (Fig. 2) that was the opposite to that of soil moisture (Fig. 1). Maximum NO_3^- concentrations were found in October, and values progressively decreased during the wet season until minimums were reached in early to late spring. NO_3^- accumulated again through the dry season (except in unit D). Though that pattern was qualitatively similar between units, quantitative differences appeared among them, with higher maximum NO_3^- concentrations in units A and B. Minimum NO_3^- concentrations appeared earlier in these units (December-March) than in units C and D (April-June), and accumulation of NO_3^- during the dry season also began sooner in units A and B. NO_3^- concentrations were considerably lower in October/November 1989 than in the same period of 1988 (Fig. 2), perhaps due to heavy rains that affected the region in the autumn of 1989 (more than 200 mm of rain in a few days). In contrast, the preceding hydrologic year experienced a relative drought (Fig. 1).

Net nitrification

Patterns of NO_3^- accumulation during incubations differed among sites in terms of the shape of the curves and net amounts of NO_3^- produced (Fig. 4). Net nitrification potential increased along the fire disturbance sequence, ranging from 20.8 in unit A to 47.4 $\mu g\ NO_3^-\text{-N}\ g^{-1}$ in unit D (Table 2). Net NO_3^- production stopped within the time frame of the incubation experiment in units A and B (at 49 and 59 days, respectively) but not in units C and D (> 64 days). The k_{max} significantly increased from unit A to unit C, but the minimum value was found in unit D (0.67 $\mu g\ NO_3^-\text{-N}\ g^{-1}\ d^{-1}$). The best fit for the time course of NO_3^- accumulation in unit D was linear, in contrast with third-order saturation-shaped curves for the three other sites (Fig. 4).

The *specific rate of nitrification* is defined as the amount of nitrified N per unit of total N and may be considered as a relative index of N quality (Power 1980). When total N was plotted against k_{max}, soils from all sites except the most recently burned site fell close to the same isoline of specific nitrification, although samples from unit D clustered slightly to the left of that isoline (Fig. 5). Recently burned soils from the wild-fire site were clearly segregated from the other sites and clustered in an area of high specific nitrification rate, perhaps because of improved substrate quality and conditions for N mineralization as a consequence of the fire. The lowest mean maximum specific nitrification rate was found in unit D (0.24 $mg\ NO_3^-\text{-N}\ g^{-1}\ N\ d^{-1}$, a value significantly lower than that found for unit A and near the 0.05 significance level compared to unit C; Table 2).

Changes in NH_4^+ concentrations in incubated samples were similar for all sampling units, with maximum values ten days after the beginning of the incubation, followed by the return to or decrease below the initial levels by the end of the incubation period (Fig. 4). Nevertheless, quantitative differences were consistent through the sere, with ammonium concentrations at any one incubation time being higher in soils from sites of successively older vegetation.

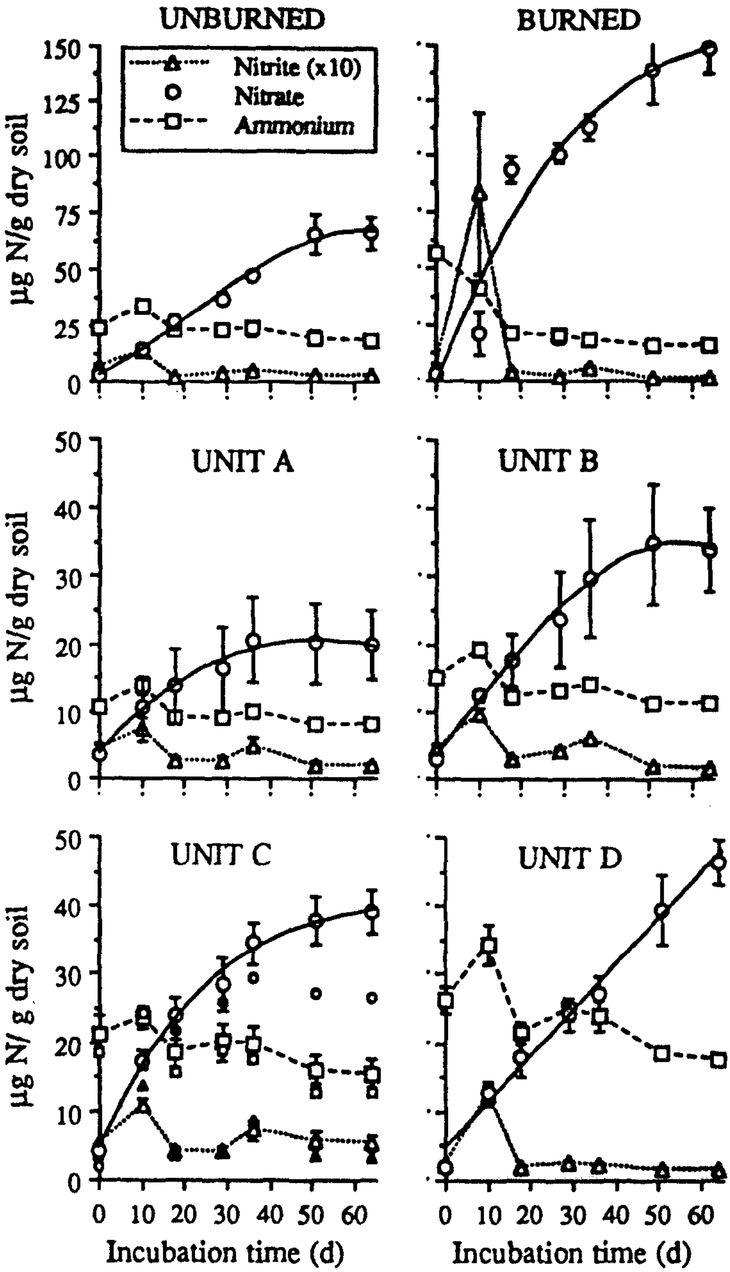

Fig. 4. NO_3^- accumulation fitted curves and changes in nitrite and ammonium concentrations during aerobic incubation (20 °C, 75% water holding capacity) of 0–5 cm soil samples from the four chronosequence units and a recently burned (wild-fire site) and its adjacent unburned gorse scrubland. Values are the mean ± standard deviation of three subsite composites per site. Scale for nitrite is multiplied by 10. Small symbols in the figure corresponding to unit C are values for 5–15 cm soils.

Table 2. Maximum net nitrification rate (k_{max}, μg NO_3-N g^{-1} dry soil d^{-1}), specific maximum nitrification rate (k'_{max}, mg NO_3-N g^{-1} total N d^{-1}), saturation time (T, days of incubation when k = 0), net nitrification potential (NP, mg NO_3-N g^{-1} dry soil at time T), specific nitrification potential (NP′, mg NO_3-N g^{-1} total N at time T) and CO_2 release rate (CO_2^, μMol CO_2 g^{-1} dry soil d^{-1}) in incubated soils from the four chronosquence units and a recently burned (wild-fire site) and its adjacent unburned gorse scrublands. Within columns, means followed by the same letter are not significantly different at $P < 0.05$ (Tukey's HSD test following ANOVA).

Site	k_{max}	k'_{max}	T	NP	NP′	CO_2^
Unit A	0.72[a]	0.73[a]	49 ± 6	20.8[a]	21.2[a]	1.05[a]
Unit B	0.89[ab]	0.54[ab]	59 ± 7	36.5[ab]	18.8[a]	1.59[ab]
Unit C	1.42[b]	0.63[ab]	>64	39.4[ab]§	17.3[a]§	1.76[abc]
Unit D	0.67[a]	0.24[b]	>64	47.4[b]§	17.3[a]§	3.01[c]
Unburned	1.51[b]	0.87[a]	60 ± 2	66.8[b]	38.3[b]	2.91[bc]
Burned	5.08[c]	1.51[c]	>64	148.0[c]§	43.6[b]§	2.45[abc]

§ These values have to be consider as conservative estimates since no saturation of the net nitrate production was achieved within the time frame of the incubation experiment.

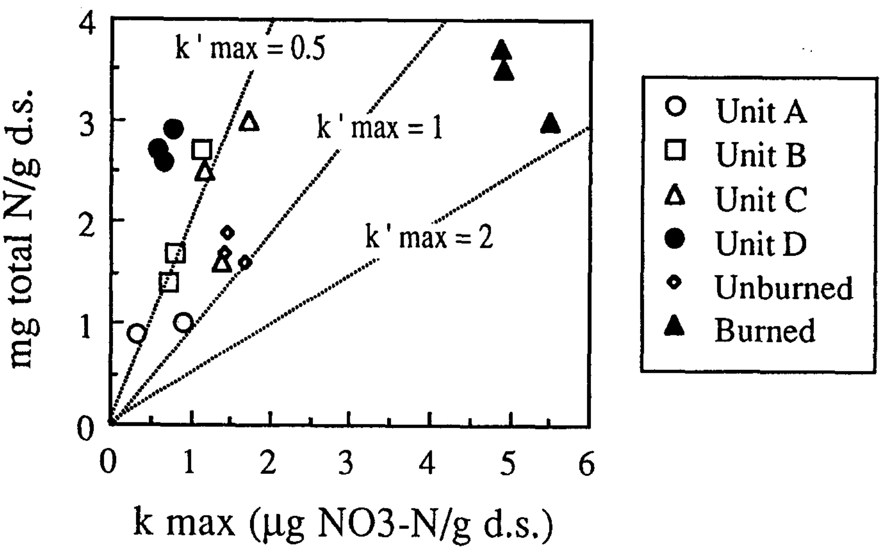

Fig. 5. Relationships between net maximum nitrification rate (k_{max}) and total N concentration of 0–5 cm soil samples from the four chronosequence units and a recently burned (wild-fire site) and its adjacent unburned gorse scrublands, with indication of isolines for specific net nitrification rates of 0.5, 1 and 2 mg NO_3^--N produced per g of total nitrogen per day.

CO_2 release rates from soil samples incubated for 1 month increased significantly through the age sequence ($p = 0.003$, Table 2). However, no differences appeared between the recently burned and unburned samples.

Effects of temperature and moisture on net nitrification and N mineralization
Means (± SE) of net nitrification and N mineralization rates for each combination of temperature and moisture are shown in Table 3. After 30 days, mineralized N was always completely nitrified, so that net N mineralization was similar or even slightly lower than nitrification. Although differences were rarely significant, nitrification and N mineralization rates across the chronosequence followed a general pattern of low rates in unit A, maximum values in unit C (or unit B if incubation temperature was 30 °C), and intermediate values in unit D. Significant differences were found between sites when incubation was carried out at 75% but not at 35% of WHC (Table 3). Both temperature and moisture significantly affected nitrification ($P < 0.001$ and $P = 0.028$ respectively, ANOVA main effects), although the influence of temperature was more important (overall means of 0.31, 0.62 and 1.04 μg NO_3^--N g^{-1} d^{-1} for 10°, 20° and 30 °C respectively; versus 0.60 and 0.71 μg NO_3^--N g^{-1} d^{-1} for 35 and 75% of WHC).

Effects of a single fire

Soil N concentrations (prescribed fire)
No significant changes were found for organic C and total N concentrations, or for the soil C/N ratio, in the top 0–5 cm of the soil immediately after the prescribed fire in Unit B ($p > 0.1$ for all three). Organic C and soil C/N did not show significant changes with time during the first 3 months following the fire ($p = 0.59$ and $p = 0.09$, respectively). No significant differences from preburn nitrate concentrations appeared until after the first rain that occurred 24 days after the fire, when nitrate increased significantly (Fig. 6). Two months after the fire, nitrate concentration was two-fold higher in the burned plot than in the adjacent unburned plot. Ammonium concentrations did not change significantly. The NH_4^+:NO_3^- ratio asymptotically decreased from 5 to less than 1 during that period (Fig. 6).

Net nitrification and net N mineralization (wild-fire site)
Net nitrification potential was considerably higher in the recent wild-fire site than in the control unburned site (Table 2). Nitrate accumulation in the unburned samples showed a very small delay period (t_o = 3 days). Once the maximum nitrification rate was reached, NO_3^- concentrations increased linearly for approximately 50 days after which net nitrate production stopped

Table 3. Net nitrate and total mineral N (ammonium plus nitrate) production (μg N per g dry soil per one month incubation period) in soils of the four chronosequence units incubated under different temperature and moisture conditions. Each value is the mean (±SE) of three subsites with one composite sample per subsite. Within each individual column, means with different superscript letters are significantly differents at the 0.05 level (Tukey's HSD test following ANOVA).

	35% water holding capacity			75% water holding capacity		
	10 °C	20 °C	30 °C	10 °C	20 °C	30 °C
	NET NITRATE PRODUCTION (μg NO_3-N g^{-1} $month^{-1}$)					
UNIT A	11.4^a (0.8)	19.5^a (0.6)	29.8^a (0.5)	1.9^a (2.8)	17.1^a (6.6)	15.0 ^{a} (4.6)
UNIT B	10.4^a (1.3)	19.8^a (1.9)	30.0^a (8.3)	12.3^b (1.2)	27.1^a (8.9)	65.8 ^{b} (2.0)
UNIT C	15.0^a (2.5)	21.7^a (2.8)	38.0^a (8.5)	15.1^b (1.8)	30.4^a (2.4)	47.8^{bc} (8.5)
UNIT D	10.8^a (1.3)	18.1^a (1.2)	35.4^a (1.8)	12.6^b (1.7)	25.3^a (2.3)	44.6 ^{c} (6.3)
	NET MINERAL-N PRODUCTION (μg mineral N g^{-1} $month^{-1}$)					
UNIT A	11.0^a (1.1)	18.1^a (1.0)	28.8^a (0.4)	2.1^a (2.3)	16.3^a (6.7)	15.1 ^{a} (3.7)
UNIT B	10.1^a (1.9)	17.6^a (1.9)	28.0^a (8.1)	12.1^b (1.2)	26.3^a (8.0)	64.4 ^{b} (2.0)
UNIT C	14.6^a (1.5)	19.4^a (1.9)	34.8^a (7.3)	14.1^b (0.4)	29.1^a (2.0)	46.1 ^{c} (6.8)
UNIT D	13.4^a (1.8)	16.7^a (2.5)	31.4^a (0.8)	15.7^b (0.9)	23.1^a (2.4)	43.1 ^{c} (6.4)

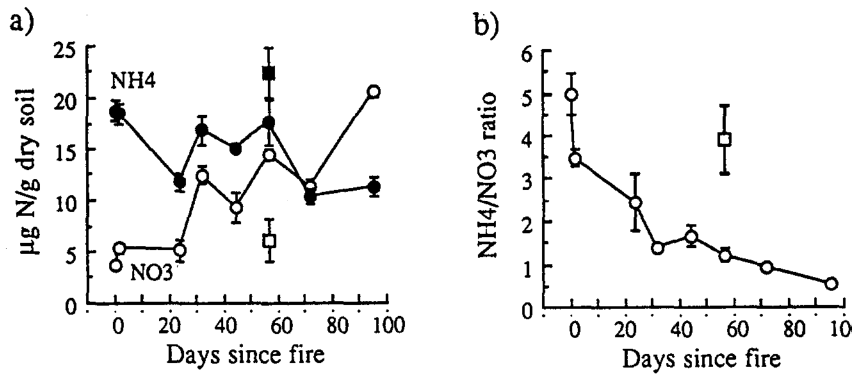

Fig. 6. Short-term changes in (a) extractable NO_3^- (open dots) and NH_4^+ (closed dots) concentrations, and (b) the NH_4^+:NO_3^- ratio of 0–5 cm surface soils following prescribed burning of .a gorse-scrubland stand in Unit B. Squares represent values found in the adjacent unburned plot two months after the fire. Values are the mean ± standard deviation of nine subsites for samples collected before and immediately after the fire, and of three subsite composites for the rest of the sampling dates.

(Fig. 4). The k_{max} in the burned soils was more than 3 times higher than in unburned samples, and net nitrate production did not stop within the time frame of the incubation experiment. As nitrate levels increased in the burned samples, ammonium concentrations progressively decreased throughout the incubation period up to about a 25% respect to the initial concentrations (Fig. 4). A high nitrite accumulation in samples from the burned site was found early during incubations.

Discussion

Ash can be rich in NH_4^+ and authors have reported substantial increases in surface soil NH_4^+ contents immediately following fire (DeBano et al. 1979, Kovacic et al. 1986, Rapp 1990, Klopatek et al. 1990). However, we found no immediate changes in soil NH_4^+ after the prescribed burn, agreeing with other studies of medium to low intensity burns (e.g. Herman & Rundel 1989). Similarly, we found no immediate changes in soil NO_3^- after the fire (Fig. 5). This is a common result in Mediterranean-type shrublands with medium to low organic matter levels in the upper soil horizon such as the California chaparral (Christensen 1973, Dunn et al. 1979). However, soil NO_3^- almost invariably increases a few weeks after fire (Christensen 1973, Kutiel & Naveh 1987, Rapp 1990) which is probably related to enhanced microbial mineralization and nitrification (Arianoutsou & Margaris 1982, Herman & Rundel 1989). In our study, net nitrification clearly increased in burned vs. unburned samples collected following the wild-fire in a gorse-scrubland (Fig. 4).

Although fire almost always brings about a temporary enhancement in mineral N availability, total N losses, including hydrological exports of N during post-fire regeneration, can be greatly elevated, especially in nutrient-poor steep semiarid ecosystems (Boerner 1982). In such systems, repeated fires separated by short intervals of time may lead to a substantial depletion of total N reserves if not rapidly replaced by N_2 fixation (Rundel 1983). The results from our study suggest that chronic fire history affects current surface soil C and N levels in the most frequently burned sites so that the younger, early successional stands are most nutrient poor (Table 1, Figs. 2 and 3). Cumulative erosion losses, and net mineralization losses in the absence of adequate replacement by litter production in successively young stands, can explain the very low levels of total N and organic C in Unit A. Cumulative erosion fluxes increased exponentially with increasing disturbance intensity, being almost three orders of magnitude higher in Unit A than in Unit D for the period between winter 1989 and spring 1990 (Carreira 1992).

Soil mineral N concentrations also showed marked changes, with a sharp rise in the NH_4^+:NO_3^- ratio along the sequence. Such a pattern has been observed in many other studies of post-disturbance succession (Robertson & Vitousek 1981). While such a pattern has been described as the result of progressive inhibition of nitrification in older sites (Rice & Pancholy 1972), our incubations showed 100% nitrification over a 30 day incubation, with no lags (Tables 2 and 3). Field concentrations of ions may simply reflect differences between plant uptake and loss. The fact that cation exchange capacity was not saturated and increased in older stands (Table 1) could contribute to the elevation in the NH_4^+:NO_3^- ratio by allowing the buildup of a larger pool of exchangeable NH_4^+ in these less eroded soils.

Soil nitrate and moisture showed qualitatively similar seasonal trends in all sampling units (Figs. 1 and 2), indicating that they are highly influenced by the year-round fluctuations typical of mediterranean-type climates. The results from the temperature/moisture experiment (Table 3) suggest that substrate quality is less important than microenvironmental conditions in controlling nitrification rates in these soils. However, some quantitative differences in soil nitrate did appear between sites that suggest differences in patterns of N cycling throughout the sere. For instance, the degree of seasonal variation in soil NO_3^- levels decreased in less disturbed sites (Fig. 2). Another shift through the sequence is the progressive delay in the appearance of, and a decrease in the maximum amount of summer nitrate accumulation. We suggest that the lower seasonality and lower summer nitrate accumulation in less disturbed sites is mainly due to increased biological sequestering (plant uptake and immobilization) of the mineral N produced rather than due to decreased mineralization. Net N mineralization and soil respiration rates were the highest in soils from the least disturbed sites (Table 2). Similarly, summer soil moisture conditions improved through the sequence (Fig. 1), thus presumably allowing plant uptake in older stands to continue further into the dry season.

Robertson & Vitousek (1981) suggested that N availability and mineralization are the most likely factors to limit nitrification within any given sere. High nitrification rates following disturbance and a later decrease as succession progresses is a common pattern observed after clear-cutting in temperate (Likens et al. 1970, Matson & Vitousek 1981) and Mediterranean-type forests (Frazer et al. 1990), after shrubland fires (Arianoutsou & Margaris 1982) or after abandonment of old-fields (Haines 1977). Mineral N availability is generally higher in early successional stages, due to accelerated N mineralization resulting from elevated temperature and moisture, and to low plant uptake, after fire and in disturbed forests (Dunn et al. 1979, Vitousek & Melillo 1979), and to N fertilization in old-fields. As a consequence, a pattern of decreasing nitrification with succession is common. In this study we similarly observed high nitrification rates after a single shrubland fire (Fig. 6, Table 2).

In contrast to the studies discussed above, however, nitrification potentials in our chronosequence generally increased from more recently disturbed to older sites (i.e. units A to D, Fig. 4, Table 2). We suggest that this discrepancy is in part due to the fact that our sites represent a chronosequence of recovery following *different* intensities of disturbance in contrast to a single similar disturbance. A higher fire frequency and intensive erosion (Carreira 1992) in units A and B left them with depleted soil N and litter pools (Table 1, Fig. 3), apparently setting ecosystem nitrogen cycling back, and thus limiting N mineralization and nitrification at these younger successional sites. As the total N pools in unit B and C increased, nitrification potentials similarly increased.

Conclusion

Current fire disturbance regimes in steep, semi-arid, dolomitophile shrublands on sandy soils of Southern Spain appear to set ecosystem N cycling back. Despite a temporary post-fire enhancement of mineral N availability, high fire frequency leads to a substantial depletion of soil total N pools, apparently because losses due to erosion and possibly other solution or gas pathways are not replaced fast enough by N_2 fixation even though leguminous N-fixing species are abundant in regenerating stands. Low total N and NH_4^+ concentrations, and low soil moisture, in highly eroded soils limit N mineralization and nitrification rates and, thus, mineral N availability to growing plants. These processes may be playing a role in desertification processes that are under way in the region.

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