

Soil-atmosphere greenhouse gases (CO₂, CH₄ and N₂O) exchange in evergreen oak woodland in southern Portugal

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ABSTRACT

A 10–20% decrease in annual precipitation is predicted in the Mediterranean basin, and in particular to the Iberian Peninsula, with foreseen effects on the exchange of soil-atmosphere greenhouse gases (GHGs; CO₂, CH₄, and N₂O). To simulate this scenario, we setup an experimental design in the particularly dry period of 2008–2009 using rainfall exclusion and irrigation, to obtain plots receiving 110% (538 mm), 100% (493 mm) and 74% (365 mm) of the natural precipitation. Soil CO₂ fluxes showed a strong increase from summer to autumn as a consequence of increasing soil heterotrophic respiration that resulted from rewetting. Fluxes of N₂O were negligible. According to our data, soil was a permanent CH₄ sink independent of the soil water content (in the range between 6–26% WFPS – water-filled pore space) and of soil temperature (in the range of 7–28°C), supporting the concept that seasonally dry ecosystems (Mediterranean) may represent a significant sink of atmospheric CH₄. The study provides evidence that the 26% decrease or 10% increase in the ambient rainfall from annual precipitation of ca 500 mm did not significantly affect soil functionality and had a limited impact on soil-atmosphere net GHGs exchange in evergreen oak woodlands in southern Portugal.

Keywords: climate change; drought; Mediterranean; precipitation

Climate change, caused by the increasing atmospheric concentration of greenhouse gases (GHGs), is modifying the hydrological cycle, increasing the frequency of extreme drought and of flood events in many places of the globe (IPCC-WGI 2007). In the Mediterranean basin, present spring precipitation is significantly lower than in the 1970s and inter-annual variability of winter precipitation and the frequency of drought is significantly higher (Xoplaki et al. 2004, Do Ó and Roxo 2008). According to Giannakopoulos et al. (2005), a 10–20% drop in precipitation seems to be the dominant feature of the predicted precipitation regime for the Iberian Peninsula.

The expected changes in precipitation (pattern and amount) in the Mediterranean region will

likely affect soil respiration (R_s), carbon cycling and net ecosystem carbon exchange (NEE) in these terrestrial ecosystems (Inglisma et al. 2009, Jongen et al. 2011). However, the impact of a reduction in precipitation on soil-atmosphere exchange of GHGs, such as carbon dioxide (CO₂), methane (CH₄) and nitrous oxide (N₂O) is not well known. In the case of Mediterranean oak woodlands, which covers a wide area in the Iberian Peninsula, studies of intra- and inter-annual carbon balance and variation in CO₂ fluxes (Pereira et al. 2007) have evidenced that seasonal drought and drying-rewetting cycles are a major feature in determining soil CO₂ exchange. However, the potential effect of reduction in precipitation on soil-atmosphere net CH₄ and N₂O exchange is less straightforward.

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The magnitude of GHGs net fluxes are known to vary considerably with vegetation, microbial community, soil organic matter, ammonia (NH_4^+) and nitrate (NO_3^-) concentrations, soil conditions, including wetness and soil temperature and are, therefore, controlled or significantly influenced by climate conditions (Mariko et al. 2007). In the soils, where gas diffusion represents the main controlling factor of CH_4 oxidation, soil water content will be of critical importance for determining the potential of the ecosystem as a CH_4 sink (Striegl et al. 1992). This is especially true for Mediterranean ecosystems with warm and dry summers and precipitations in winter, which might create favorable conditions for CH_4 oxidation throughout the year (Castaldi et al. 2007). Although soils are usually considered as net source of atmospheric N_2O , they can also act as a sink, at least temporarily (Slemr and Seiler 1991). Rosenkranz et al. (2006) found weak and significantly negative N_2O fluxes in Mediterranean forest soils and linked fluxes to the very low N availability.

In most publications, temperature is considered the main limiting factor of microbiological activity (Skiba et al. 1998, Cruz et al. 2008). However, in Mediterranean ecosystems the combined effects of soil water content (drought), low and poor organic matter and low soil P, C, and N contents play a key influence on microbial activity and, as a consequence, in greenhouse gas fluxes. The aim of the present work was to understand the dynamics of soil-atmosphere net GHGs fluxes in evergreen oak woodlands of southern Portugal as well as to evaluate the effect of the predicted decrease in rainfall in the Iberian Peninsula. The quantification of soil-atmosphere GHGs exchange in Mediterranean region are poorly reported while observational data of non- CO_2 fluxes as far as we know, has not yet been reported for Portugal.

MATERIAL AND METHODS

Site description, climate and vegetation. The experiment was part of the EU project NitroEurope

IP and was located at Mitra Campus, 12 km southwest of Évora in southern Portugal ($38^\circ 31' 40''\text{N}$, and $8^\circ 00' 01''\text{W}$). The region has a typical Mediterranean climate, with hot and dry summers and moderately cold winters. Long-term (1961–1990) average meteorological data for this area show that more than 80% of annual precipitation (ca 669 mm) occurs from October to April and a mean annual temperature of 15.5°C (Pereira et al. 2007). The work was developed in a particularly dry and cool year, (2008–2009), where annual precipitation was 74% of the long-term average with a mean annual temperature of 13.4°C (Table 1). The study site (0.264 ha) was covered with *Quercus suber* L. trees. From the surface to 1 m depth, the soil is constituted by 88.9% of sand, 4.9% of silt, and 6.3% of clay, with a low water retention capacity of 5% (v/v) (Kurz-Besson et al. 2006). The soil C/N ratio is 14.3 ± 1.8 .

Treatments. An experiment was previously designed as described in (Grant et al. 2010) according to prediction of 20% reduction in precipitations (Miranda et al. 2006) and established three treatments by means of rainfall capture and irrigation: control (C), rain exclusion (RE) and water addition (WA). Each treatment was replicated randomly within each of three blocks, with two chambers per treatment per block. Control with full natural rainfall received 493 mm of precipitation during the experimental period April 2008–April 2009. In the RE treatment water was prevented from reaching the soil by 26% less than the control by using below-canopy plastic covers (covering 26% of the soil surface). During the experimental period (2008–2009), RE treatment plots received 365 mm of natural precipitation. Additional water was supplied to the WA treatment plots via irrigation pipes with drip emitters. Irrigation was applied randomly once per month in days without precipitation to make irrigation more effective due to low soil water holding capacity. The WA treatment plots received 10% more than the control, i.e. 538 mm of precipitation.

Soil CH_4 , N_2O and CO_2 measurement, soil temperature and soil moisture. Fluxes of CH_4 and

Table 1. Long-term average meteorological data for (1961–1990 and 2000–2009) and for study period (April 2008–April 2009) of precipitation and air temperature study site. Data from Centre of Ecophysics of Évora

	1961–1990	2000–2009	Study period (April 2008–April 2009)		
			control	rain exclusion	water addition
Precipitation (mm)	669	561	493	365	538
Air temperature ($^\circ\text{C}$)	15.5	15.4		13.4	

N₂O were measured with 18 static chambers (six chambers per treatment) using the technique described by Rosenkranz et al. (2006) every 4 weeks during one year. Gas samples were analysed by gas chromatograph (Fison series 800, Ontario, Canada) according to (Castaldi et al. 2007). Soil CO₂ effluxes were measured with a PP-Systems EGM-3 closed chamber, dynamic soil system (PP-System, Amesbury, USA). Soil temperature was measured near the collars with a handheld digital thermometer. Water filled pore space (WFPS) was determined according to Linn and Doran (1984).

Soil analyses. To determine soil mineral nitrogen concentration, four samples from the area around the static chambers were collected on the same days of gas sampling. NO₄⁺ and NH₃⁻ concentrations were analysed as described in Cruz and Martins-Loução (2000).

Statistical analysis. Data were subjected to one-way ANOVA using a randomized block design, to test for the effects of water treatments. All variables were tested for normality and homogeneity of variances. Differences were considered statistically significant at $P < 0.05$. Data analysis was carried out using SAS 9.1 (SAS Institute, Cary, USA).

RESULTS

Seasonal changes and soil properties. Soil temperature, air temperature and precipitation (plus water addition from irrigation) from April 2008 to April 2009 are shown on Figure 1a. Soil temperature varied between 28°C in summer (July) and 7°C in winter (January), closely following air temperature. The differences between treatments were not statistically significant. WFPS ranged between 6% till 26% during the experimental period at 20 cm depth soil (Figure 1b). The WA treatment plots were significantly wetter on average by 1.3-fold than the RE treatment plots throughout the entire year, except in October. However, when compared to controls WA treatments were wetter only in June, July, and August. In June soil moisture in all treatments dropped noticeably as compared to previous months. WFPS was negatively correlated with soil temperature during the whole period of observation ($r = -0.72$, $P < 0.001$). No significant differences in soil pH among treatments were observed except in December, when soil pH in WA plot was slightly but significantly higher, as compared with control and RE plots.

Net soil CO₂ fluxes. Net soil CO₂ flux varied significantly along the year (Figure 1c). The values

ranged from 20 mg CO₂/m²/h to 240 mg CO₂/m²/h with the lowest values occurring in the dry summer period (July, August, and September) when soil water content exhibited the lowest values in the experimental period (Figure 1b). Soil rewetting stimulated ecosystem respiration, i.e. after the dry summer period, rainfall in September led to a significant increase in carbon dioxide fluxes in all treatments on average by 2.6-fold, maintaining the same magnitude during the following autumn-winter months (Figure 1c). In the wet period (October 2008–April 2009), carbon dioxide fluxes ranged from 70 mg CO₂/m²/h to 130 mg CO₂/m²/h, when WFPS ranged between 11% to 26% (Figure 1b). Rainfall events were accompanied by substantial drop in soil temperature till March (Figure 1a). There was a negative correlation between net soil CO₂ fluxes and soil temperature during summer ($r = -21$, $P < 0.01$), and positive correlation between CO₂ fluxes and soil temperature in wet months ($r = 0.54$, $P < 0.01$). However, our results did not show sustained statistically significant differences in soil CO₂ fluxes between the different irrigation regimes.

Net soil CH₄ fluxes. Our results showed that soil was a permanent sink for CH₄ independently of WFPS (Figures 1c, d). Methane consumption tends to have higher values in all treatments during summer (in June, July, and August) than those during other months of the study period. Methane consumption was significantly affected by treatments in September and October: CH₄ fluxes in the water addition treatment plots were significantly higher than those in control plots. Overall, comparing all samples, there was a large variability in methane uptake after the rains, especially in April 2008 and January and February 2009. So, CH₄ fluxes varied between -104.17 μmol CH₄-C/m²/h to 54.93 μmol CH₄-C/m²/h, between -8.37 μmol CH₄-C/m²/h to -70.30 μmol CH₄-C/m²/h, and between -90.49 μmol CH₄-C/m²/h to 76.77 μmol CH₄-C/m²/h in April 2008, January 2009 and February 2009, respectively. There was a negative correlation between methane consumption and soil temperature in summer ($r = -0.32$, $P < 0.001$), and CH₄ fluxes positively correlate with soil CO₂ fluxes ($r = 0.26$, $P < 0.001$).

Net soil N₂O fluxes. N₂O fluxes were very low and both net emission and net uptake of N₂O were found to occur with fluxes varying between -7.38 mg N₂/m²/h and 47.28 mg N₂/m²/h (Figure 2a). No significant differences between treatments were observed in any occasion. In wet months nitrous oxide fluxes were positively correlated with soil temperature.

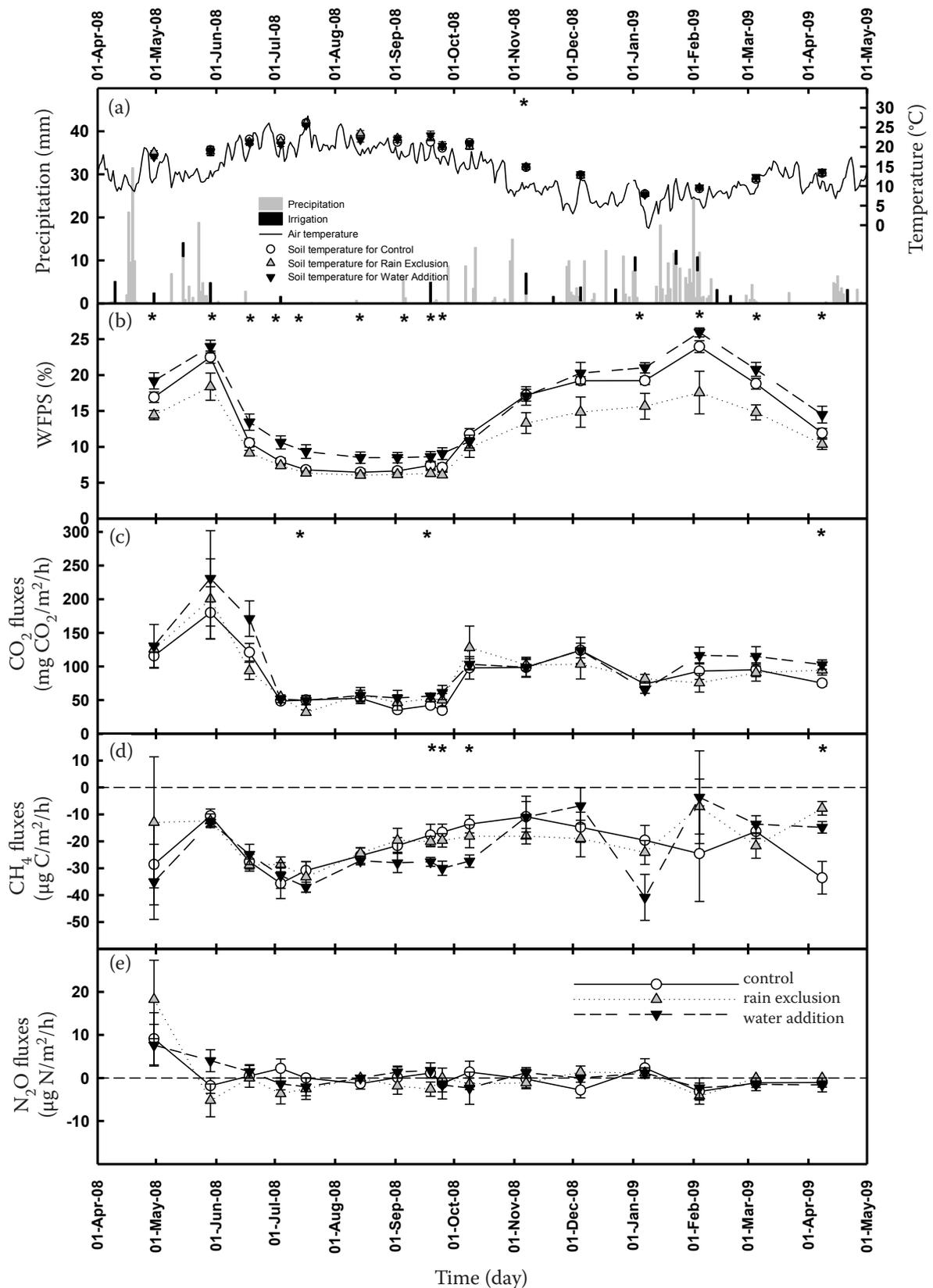


Figure 1. (a) Precipitation (mm), irrigation applied to the water addition (WA) treatment (mm), air temperature ($^{\circ}\text{C}$) and soil temperature recorded in the upper 5 cm of soil; (b) water field pore space (%); (c) soil CO_2 fluxes; (d) soil CH_4 fluxes, and (e) soil N_2O fluxes measured in the control (C), rain exclusion (RE) and water addition (WA) treatments at the study site from April 2008 to April 2009. Values are means \pm SE ($n = 6$). A significant effect of rain exclusion (RE) and water addition (WA) treatments ($P < 0.05$) is indicated by stars directly above the data points

Soil $\text{NH}_4^+\text{-N}$. Soil $\text{NH}_4^+\text{-N}$ content was low all over the year ranging from 0 to 13.53 mg $\text{NH}_4^+\text{-N}$ /kg dry soil (Figure 2a). Monthly measurements demonstrated that $\text{NH}_4^+\text{-N}$ level in control and RE treatment plots varied between 0.94 mg $\text{NH}_4^+\text{-N}$ /kg dry soil to 13.53 mg $\text{NH}_4^+\text{-N}$ /kg dry soil, while WA plots maintained a relatively constant level of $\text{NH}_4^+\text{-N}$. The most significant difference among treatments was observed in September when NH_4^+ content in both control and RE plots attained their maximum and were 3-fold higher, as compared with WA plots. Although smaller, additionally significant differences ($P < 0.05$) also occurred in July and January.

Soil $\text{NO}_3^-\text{-N}$. The differences between treatments in $\text{NO}_3^-\text{-N}$ were more marked in April 2008, when its content in both RE and WA treatment plots was 3.8-fold higher than in control plots (Figure 2b). In July, September, October, and January, the differences between treatments were lower than in April. We observed a seasonal trend of soil $\text{NO}_3^-\text{-N}$ content in all treatments in winter months: the lowest values of $\text{NO}_3^-\text{-N}$ were found in December, January and February, and ranged between 0.98 mg $\text{NO}_3^-\text{-N}$ /kg dry soil and 4.48 mg $\text{NO}_3^-\text{-N}$ /kg dry soil, whereas in summer months it was between 4.87 mg $\text{NO}_3^-\text{-N}$ /kg dry soil and 15.09 mg $\text{NO}_3^-\text{-N}$ /kg dry soil.

DISCUSSION

The results presented show that reducing 26% of rainfall reaching the soil or adding 10% water affected WFPS throughout the experimental period (Figure 1b). Our data are in good agreement with those of Inglima et al. (2009), indicating that net carbon dioxide fluxes were water-limited since higher net CO_2 fluxes were observed in April 2008 in comparison to April, 2009 (April 2008 was wetter than April 2009). During the hot and dry period (June–September), low soil moisture (6%) suppressed R_s in all treatments confirming the evidence that soil temperature is another important variable controlling CO_2 emission rates in Mediterranean forest ecosystems (Castaldi et al. 2007). Furthermore, the summer decrease of R_s may be due to a combined effect of low soil water content, SWC (quantity of water in soil), low mineralization and low relative amount of easily degradable soil organic matter, i.e. low C/N ratio (in our study C/N = 14.37).

The first rain events in September led to increases in net CO_2 fluxes. These data are in agreement with previously observed strong increases in ecosystem respiration by autumn rains in the Mediterranean basin (Jarvis et al. 2007), where soil CO_2 fluxes

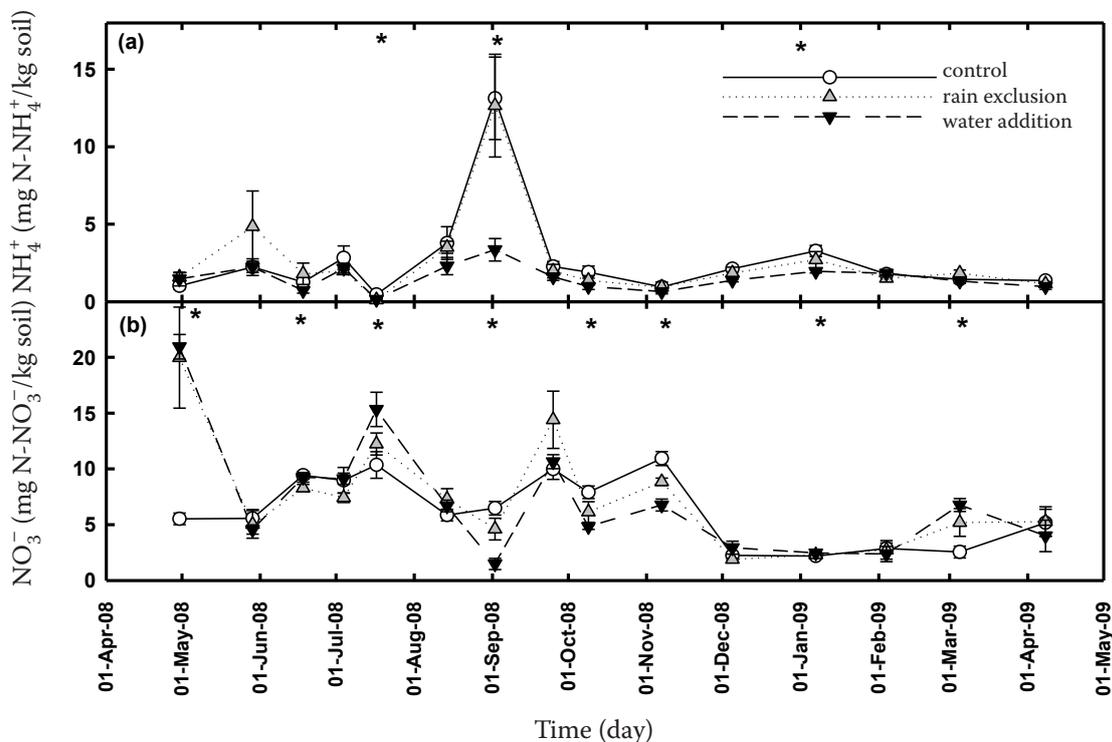


Figure 2. (a) Soil $\text{NH}_4^+\text{-N}$ (mg/kg N DW) and (b) $\text{NO}_3^-\text{-N}$ (mg/kg N DW) contents measured during the study period in the control, rain exclusion and water addition treatments on the same days of gases sampling. Values are means \pm SE ($n = 6$). A significant effect of rain exclusion (RE) and water addition treatments (WA) ($P < 0.05$) is indicated by stars directly above the data points

after the first autumn precipitations are, probably due to easily degradable substrates, accumulated in dry soils over the summer and the stimulation of microbial activity. New water potential equilibrium is established and the microbial cells begin to re-absorb and release metabolites (Fierer and Schimel 2002), generating net CO₂ fluxes. From October, 2008 till April, 2009, soil CO₂ flux was constant independently of the WFPS (Figures 1b, c). This could be explained by the fact that in woodlands, microbial activity appears not to be moisture-limited before wetting, showing a mild response to the additional water-input to the soil (Inglisma et al. 2009). According to our data, the soil in evergreen oak woodland presented a permanent CH₄ sink independent of WFPS and soil temperature, and supported the concept that seasonally dry ecosystems (Mediterranean) are a significant sink of atmospheric CH₄ (Rosenkranz et al. 2006, Castaldi et al. 2007). It seems that regardless of the treatment, the soil maintained a fairly consistent potential for methane oxidation over the course of the years 2008–2009. The methanotrophic community was apparently able to remain viable during unfavorable winter conditions of low soil temperature (7°C), and high WFPS (more than 25%), and low rates of gas diffusion and CH₄ oxidation. During wet months (May, 2008 and November, 2008–April, 2009) the rate of methane oxidation had a large variability in all treatments which could be explained by the variability in oxygen content of the wet soil (Rower et al. 2010). However, CH₄ consumption tends to have higher values and lower variability in the dry summer, as a result of increased gas diffusivity under low WFPS (less than 10%) and high temperature. In the absence of rainfall, the rate of methane oxidation increased, probably because of the increased oxygen content of the soil and reduced water filling pore space, therefore allowing more atmospheric CH₄ to easily diffuse along the soil profile and reach methane oxidizing microorganisms (Otter and Scholes 2000).

Negative correlation between CH₄ consumption and soil temperature in dry months might be explained by the fact that temperature can influence CH₄ fluxes in combination with soil water content (Borken et al. 2000), with microbiological activity being limited by water deficit. Therefore the response to temperature did not follow an Arrhenius-type relation with rising temperature in dry period. A positive correlation between soil CH₄ and CO₂ in our study may be explained by the close links between C and N cycles involving microorganisms that contribute directly to CO₂, N₂O and CH₄ fluxes (Osler and Sommerkorn 2007).

Our data show that the mineral N content of soil, is compatible with its use for growth by methanotrophic bacteria, whereas high rates of nitrification can produce toxic effects on CH₄-consuming bacteria through production of NO₂ and NH₂OH (Bowden et al. 2000). For example, soil nitrate concentrations around 40–50 ppm were shown to inhibit methane oxidation by soil microorganisms in 10–20% (Cruz et al. 2008). Since the nitrate concentrations determined in the oak woodland soils in our study were 2 to 3 times lower, we may conclude that there would be little or no inhibitory effect on soil methane oxidation by methanotrophic bacteria. Low soil inorganic nitrogen content was presumably the main factor determining that our studied soils are a methane sink. We have observed very low net N₂O exchange between soil and atmosphere and this consisted of the low mineral N available in our soil. Indeed, many authors reported links between net negative N₂O fluxes and very low concentrations of inorganic N, low rates of denitrification and low soil C/N ratio (Ryden 1981, Skiba et al. 1998). During the experimental period of 2008–2009 the conditions for net N₂O uptake in the evergreen oak woodland we studied were in the range of values described by Ryden (1981).

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