

# Afforested sites in a temperate grassland region: influence on soil properties and methane uptake

M. E. Priano · V. S. Fusé · S. Mestelan · A. M. Berkovic · S. A. Guzmán ·  
R. Gratton · M. P. Juliarena 

Received: 30 December 2015 / Accepted: 10 July 2017  
© Springer Science+Business Media B.V. 2017

**Abstract** Methane (CH<sub>4</sub>) flux at the soil-atmosphere interface (SAI) results from the balance between CH<sub>4</sub> production (methanogenesis) and CH<sub>4</sub> consumption (methanotrophy). The latter predominates in well-aerated mineral soils; is affected by a combination of abiotic and biotic factors, especially soil diffusivity, which depends on soil properties, and methanotroph activity. This work reports results of CH<sub>4</sub> fluxes from afforested sites located in a temperate region of formerly native grassland in Buenos Aires Province (Argentina, Southern Hemisphere), taking a naturalized pasture as a reference. Methane concentration [CH<sub>4</sub>] and soil parameters along the soil profile were

also measured to understand intersite differences in CH<sub>4</sub> fluxes at the SAI, that could be related to vegetation cover and its influence on soil properties and therefore, on CH<sub>4</sub> soil diffusivity. At all sites soils were CH<sub>4</sub> sinks in the range of  $-3.55$  to  $-14.39$  ng CH<sub>4</sub> m<sup>-2</sup> s<sup>-1</sup>; the naturalized pasture presented the weakest one. Intersite differences in CH<sub>4</sub> fluxes may result from differences observed in [CH<sub>4</sub>] profiles and CH<sub>4</sub> diffusion coefficients. [CH<sub>4</sub>] variation could be explained mainly by differences in silt and clay content and bulk density that affect CH<sub>4</sub> soil diffusivity. These could be the result of afforestation that seems to improve the physical and biological soil attributes linked to CH<sub>4</sub> consumption as it meliorates its diffusivity.

---

M. E. Priano · V. S. Fusé · A. M. Berkovic ·  
M. P. Juliarena (✉)  
CIFICEN- Centro de investigaciones en Física e  
Ingeniería del Centro de la Provincia de Buenos Aires,  
Tandil, Argentina  
e-mail: pjuliarena@exa.unicen.edu.ar

S. Mestelan  
CRESCA - UNCPBA – Centro Regional de Estudios  
Sistémicos de Cadenas Agroalimentarias - Universidad  
Nacional del Centro de la Provincia de Buenos Aires,  
Buenos Aires, Argentina

S. A. Guzmán  
Universidad Nacional del Centro de la Provincia de  
Buenos Aires, Buenos Aires, Argentina

R. Gratton  
CONICET – Consejo Nacional de Investigaciones  
Científicas y Tecnológicas, Buenos Aires, Argentina

**Keywords** Methane concentration profile · Soils properties · Methane diffusion coefficient · Methane fluxes in soils

## Introduction

Both biotic and abiotic processes from natural and anthropogenic sources produce methane (CH<sub>4</sub>), a potent greenhouse gas whose current global mean concentration in the atmosphere is 1.8 ppmv (Curry 2007; Tate 2015). This concentration value has been increased since the start of the industrial period as a result of the growing imbalance between CH<sub>4</sub>

production and its consumption. Land conversion to agricultural use is a major contributor to this imbalance because it reduces the consumption of atmospheric  $\text{CH}_4$  by aerated soils (Hütsch 1998; Jensen and Olsen 1998; Ridgwell et al. 1999; Smith et al. 2000). The contribution of these soils to the global methane sink is small ( $\sim 30 \text{ Tg year}^{-1}$ ) compared to the photochemical  $\text{CH}_4$  oxidation that occurs in the atmosphere ( $450\text{--}510 \text{ Tg year}^{-1}$ ) but it is significant because it is the unique  $\text{CH}_4$  sink where anthropogenic activity (with the expansion of agriculture) has an influence (Boeckx et al. 1997; Dutaur and Verchot 2007; Tate 2015).

$\text{CH}_4$  flux at the soil-atmosphere interface (SAI) results from the balance between  $\text{CH}_4$  production (methanogenesis) and  $\text{CH}_4$  consumption (methanotrophy) where the latter predominates in well-aerated mineral soils (Smith et al. 2000, 2003). Between 30 and 50% of global soil  $\text{CH}_4$  consumption is apparently located in temperate latitudes. However, it has been poorly estimated because of its great spatial and temporal variability and the scarce measurements from the Southern Hemisphere (Dutaur and Verchot 2007; Price et al. 2003). The highest  $\text{CH}_4$  consumption was found in temperate and subtropical ecosystems, in soils under native forest (up to  $11 \text{ kg CH}_4 \text{ ha}^{-1} \text{ year}^{-1}$ ) followed by those under planted pine forest ( $4\text{--}6 \text{ kg CH}_4 \text{ ha}^{-1} \text{ year}^{-1}$ ) and undisturbed soils while the lowest rate was measured in most managed grassland and cropland soils ( $<1 \text{ kg CH}_4 \text{ ha}^{-1} \text{ year}^{-1}$ ) (Tate 2015).

Soil microbial processes in general and in particular  $\text{CH}_4$  oxidation (consumption) are affected by a combination of abiotic and biotic factors. Spatial and temporal variability in  $\text{CH}_4$  oxidation in aerated soils is caused by differences in soil diffusivity that depends on soil properties, methanotroph activity or some combination of the two (Von Fischer et al. 2009; Smith et al. 2003; Striegl 1993). The complex nature of the soil structure makes difficult to determine in situ soil diffusivity and, therefore, there is still uncertainty of its value in different and heterogeneous soils. Generally,  $\text{CH}_4$  oxidation capacity exceeds soil diffusivity; therefore soil physical characteristics are the limiting factor (Boeckx et al. 1997; Del Grosso et al. 2000; Hiltbrunner et al. 2012; Striegl 1993). Although  $\text{CH}_4$  oxidation rates vary markedly between terrestrial ecosystems, it is generally accepted that changes in soil moisture and texture mostly affect  $\text{CH}_4$

consumption due to their influence on the diffusion of atmospheric  $\text{CH}_4$  into the soil layer where methanotrophs are located (Boeckx et al. 1997; Dutaur and Verchot 2007; Prajapati and Jacinte 2014).

The static chambers technique is the most widely used method to measure  $\text{CH}_4$  flux in situ at the SAI. It is simple, low cost and allows repeated measurements on a study site with minimal disturbance and relatively little labor. Moreover, the  $\text{CH}_4$  concentration profile can be obtained with a simple probe inserted into the soil and this information can be used both to supplement  $\text{CH}_4$  flux measurements and to identify the soil depth where  $\text{CH}_4$  oxidation mostly occurs or where the diffusion process is predominant (Hou et al. 2012; Kusa et al. 2008). How these two processes co-participate in regulating  $\text{CH}_4$  flux at the SAI remains unclear—little is known about either the vertical heterogeneity of soil diffusivity and  $\text{CH}_4$  oxidation (Von Fischer et al. 2009) and about how these two parameters could explain intersite differences in  $\text{CH}_4$  fluxes.

In this work we present results of  $\text{CH}_4$  fluxes from afforested sites located in a temperate region of formerly native grassland in Buenos Aires Province (Argentina, Southern Hemisphere) taking a naturalized pasture as a reference. The aim of this work was to understand intersite differences in  $\text{CH}_4$  fluxes that could be related to vegetation cover and its influence on soil properties and therefore, on  $\text{CH}_4$  soil diffusivity.

## Materials and methods

### Sites and soil description

The experimental sites were located in the piedmont of Tandil Sierras ( $50\text{--}250 \text{ MAMSL}$ ), near Tandil city ( $37^\circ 19' 00'' \text{S } 59^\circ 08' 00'' \text{W}$ ), where annual mean air temperature is  $13.7^\circ \text{C}$  and average annual precipitation is  $889 \text{ mm}$ . Soils are developed in a Quaternary loess mantle composed by pebbles, eolianites, lacustrine clays and alluvial deposits with a variable thickness covering a Pre-Cambrian granitic basement (Chaparro et al. 2003). Table 1 presents a brief site description along with its associated soils. A more detailed description can be found in Priano et al. (2014).

At each experimental site, one pit was hand excavated and described according to the NRCS soil

**Table 1** Sites and description

Site ID	Landforms	Parent material	Vegetation	Soil classification
Pasture field (PF)	Gently undulating slopes (<2%)	Loessic sediments	Pastureland with enclosure; herbage naturally growing*	Typic Argiudoll
Eucalyptus grove (EG)	Upland; shoulder of a loessic crest. Anthropogenic disturbance nearby (crest dissection for a road). Predominant slope >3%	Loessic sediments	<i>Eucalyptus viminalis</i>	Typic Argiudoll
Afforested pine grove (PG)	Upland slope (>10%)	A loessic mantle mixed with colluvial debris from a Precambrian granite outcrop (Tandilia System or Tandilia sierras)	<i>Pinus radiata</i>	Typic Argiudoll
Mixed deciduous grove (DG)	Upland slope contiguous to a floodplain. Anthropogenic disturbance nearby; slope >2%	Loessic mantle reworked by water	Grassland + trees ( <i>Celtis sp.</i> , <i>Eucalyptus camaldulensis</i> , <i>Eucalyptus viminalis</i> , <i>Laurus nobilis</i> , <i>Quercus robur</i> and <i>Cedrus sp.</i> )	Petrocalcic Argiudoll

PF pasture field, EG eucalyptus grove, PG afforested pine grove, DG mixed deciduous grove

\* Prior to the study, the site was used for cattle raising purposes and included pasture sown with NP-fertilization

description manual (Schoeneberger et al. 2002). Undisturbed soil samples were taken to estimate porosity using the cylinder method (SSLS 2004). Disturbed soil samples were taken for soil physico-chemical characterization. Soil characterization at LASFA (Laboratorio de Análisis de Suelos de la Facultad de Agronomía–UNCPBA) facilities included organic C determination (OC, wet oxidation), total N (micro Kjeldahl with steam distillation), pH (potentiometric method, 1:2.5 soil:water), salinity (electrical conductivity in 1:2.5 soil:water extracts, EC), and cation exchange capacity (CEC) extracted in 1 N NH<sub>4</sub>Ac, pH 7 or 8.2 (the latter for samples with reaction to HCl 10% v/v, indicating the presence of carbonates). The CEC involved the displacement of ammonium by 1 N NaAc and quantification of ammonium by steam distillation. Mechanical analysis (texture) was performed using the hydrometer method after labile OC oxidation with H<sub>2</sub>O<sub>2</sub> and carbonates dissolution with HCl and removal of cations by washing. All soil routines were performed according to SSLS (2004) protocols, with the exception of OC, that followed a modified Walkley and Black procedure according to SAMLA (2004). After soil morphological and physicochemical characterization, and considering climate information, soils were classified according to the Soil Taxonomy (SSS 2014).

#### Methane flux and methane soil air concentration

At each site, CH<sub>4</sub> fluxes from the SAI were measured by the static chamber technique described in Priano et al. (2014). CH<sub>4</sub> fluxes in each static chamber were determined by the linear regression of methane concentration versus time, always with R<sup>2</sup> > 0.94.

Simultaneously, sub-surface CH<sub>4</sub> concentration [CH<sub>4</sub>] was measured to obtain the [CH<sub>4</sub>] profile of the soil atmosphere and to determine gas diffusion coefficients for the first layer (10 cm) at each site (A horizons) by Fick's Law using a stainless steel probe (6 mm outer diameter and 4 mm inner diameter) of 550 mm length and with four lateral small holes (<5 mm) in its lower end. The probe was randomly placed three times at each site to obtain three [CH<sub>4</sub>] profiles after driving the probe into the soil and taking soil air samples every 5 cm until reaching a depth of 50 cm. This technique allowed us to obtain a gas sample at a selected depth with the least disturbance to the soil gas composition and the soil environment. Gas samples were drawn using 20-mL syringes with the three stopcocks that were flushed with approximately 10 mL of gas at the desired sampling depth and then a 10-mL gas sample was withdrawn.

Both measurements were carried out during June–July 2013 because it was the time when we recorded

the highest CH<sub>4</sub> consumption for these sites (Priano 2014).

All gas samples were analyzed immediately with a gas chromatograph (GC, Agilent 7890A) equipped with a FID and 1.8 m Poropak Q (80/100 mesh) column. The oven, injector and detector temperatures were 50, 250 and 250 °C respectively. The flow rate of the carrier gas (He) was 30 mL min<sup>-1</sup>. Flame gases (H<sub>2</sub> and O<sub>2</sub>) were set at 30 and 400 mL min<sup>-1</sup>.

CH<sub>4</sub> diffusion coefficient in soil (D<sub>s</sub>) was obtained for the first 10 cm soil layer using Fick's Law:

$$F_{\text{CH}_4} = -D_s \frac{\Delta[\text{CH}_4]}{\Delta Z}, \quad (1)$$

where F<sub>CH<sub>4</sub></sub> is the CH<sub>4</sub> mass flux (ng CH<sub>4</sub> m<sup>-2</sup> s<sup>-1</sup>), D<sub>s</sub> is the soil CH<sub>4</sub> diffusivity coefficient (cm<sup>3</sup> soil air cm<sup>-1</sup> soil s<sup>-1</sup>) and Δ[CH<sub>4</sub>]/ΔZ the concentration gradient (ppm CH<sub>4</sub> soil air cm<sup>-1</sup> soil depth). The diffusivity coefficient D<sub>s</sub> was obtained by

$$D_s = D_a * a \varphi_g^b, \quad (2)$$

according to Price et al. (2003), where D<sub>a</sub> is the diffusion coefficient for CH<sub>4</sub> in air (cm<sup>2</sup> s<sup>-1</sup>), φ<sub>g</sub> the air filled porosity (m<sup>3</sup> air m<sup>-3</sup> soil) and *a* and *b* are the dimensionless coefficients that account for the tortuosity and size distribution of the soil pore respectively and, whose empirical values are *a* = 0.9 and *b* = 2.3–, the most commonly values used in soil CH<sub>4</sub> oxidation studies. CH<sub>4</sub> diffusion coefficient in air (D<sub>a</sub>) is 0.195 cm<sup>2</sup> s<sup>-1</sup> at standard temperature and pressure conditions (STP) (Ridgwell et al. 1999) and the term φ<sub>g</sub> is

$$\varphi_g = \varepsilon(1 - WFPS), \quad (3)$$

where ε is the porosity (m<sup>3</sup> m<sup>-3</sup>) and WFPS the water filled pore space (m<sup>3</sup> m<sup>-3</sup>). D<sub>a</sub> depends on temperature and pressure; therefore corrections for temperature were made according to Price et al. (2003):

$$D_{a(T)} = D_{a273K} * \left[ \frac{T(K)}{273K} \right]^2, \quad (4)$$

Pressure corrections were not taken into account here because sampling was performed in the same area (Tandil city) (Price et al. 2003).

#### Environmental parameters

Soil temperature at a 5 cm depth, air temperature inside the chamber and air ambient temperature were

logged with a temperature data logger ibutton DS1921G in each case during CH<sub>4</sub> flux measurements. In addition, soil water content (w%, n = 5) was measured gravimetrically for the first soil layer from each site with a small stainless steel cylinder (6 cm in height, 4 cm in internal diameter), which afterwards allowed us to calculate bulk density (ρ<sub>a</sub>), water filled pore space (WFPS) and porosity (ε) through:

$$\rho_a = \frac{\text{drymass}}{\text{Totalvolume}}; WFPS = \frac{w\% * \rho_a}{\rho_w * \varepsilon}; \varepsilon = 1 - \frac{\rho_a}{\rho_r}, \quad (5)$$

where ρ<sub>w</sub> is the water density (1 g cm<sup>-3</sup>) and ρ<sub>r</sub> is the particle density (2.65 g cm<sup>-3</sup>). Then, φ<sub>g</sub> was calculated according to Eq. 3.

#### Statistical analysis

ANOVA test was used to establish if there were any statistical differences between sites regarding soil parameters within the first layer, CH<sub>4</sub> fluxes and CH<sub>4</sub> diffusion coefficients in soil. Levene test was used to determine homogeneity of variance that allows defining which post hoc test can be used (LSD Fisher or Games-Howell). Linear correlations (Pearson) were used to determine the significance of the relationship between methane concentration in soil profile and soil parameters and subsequently, simple and multiple regression analysis were carried out.

## Results

### Soil parameters at the studied sites

Soil morphology of the four studied sites (Table 2) was the expected for soils in the area, developed on loess primarily under grassland vegetation (INTA-CIRN 1989). These Mollisols had surface horizons rich in OC and Bt horizons with clay accumulation due to illuviation (Pazos 1984). The soil profiles were free of carbonates with the exception of the BCK horizon of the DG site that accumulated inorganic C as concretions and was underlain by a petrocalcic horizon, whose cementation was due to carbonates that accumulated as result of a past semiarid climate (Pazos and Mestelán 2002). Trees at the EG, PG and DG were the result of afforestation and the gross biomass

**Table 2** Soil parameters for the different sites

Site	Horizons	Depth (cm)	PH (1:2.5, water)	EC ( $\mu\text{S cm}^{-1}$ )	OC ( $\text{g kg}^{-1}$ )	Total N ( $\text{g kg}^{-1}$ )	C:N ratio	CEC ( $\text{cmoc kg}^{-1}$ )	Clay %	Silt %	Sand %	$\rho_a$ ( $\text{g cm}^{-3}$ )	$\varepsilon$ %
PF	Ap1	0–7	5.8	n.d	33.0	3.42	9.6	22.3	26.09	37.04	36.88	1.18	55.47
	Ap2	7–20	6.1	n.d	23.2	2.10	11.0	19.8	25.83	37.78	36.39	n.d	n.d
	AB	20–35	6.3	n.d	12.1	1.20	10.1	18.2	27.14	35.44	37.42	n.d	n.d
	Bt1	35–57	6.4	n.d	8.4	0.64	13.1	25.6	45.22	25.72	29.06	n.d	n.d
	Bt2	57–70	6.5	n.d	4.6	0.45	10.2	23.1	39.59	25.28	35.14	n.d	n.d
	BC	70–78	6.7	n.d	3.0	0.30	10.0	17.5	19.72	36.48	43.80	n.d	n.d
	C	78+	6.8	n.d	2.0	0.15	13.3	15	7.80	39.13	53.07	n.d	n.d
EG	A	0–17	5.06	74.7	67.8	4.27	15.9	21.9	25.86	29.71	44.43	0.84	68.14
	AB	17–30	5.46	67.4	15.3	1.19	12.9	20.2	31.87	40.94	27.19	1.17	55.88
	Bt1	30–47	6.35	76.5	12.4	0.79	15.7	25.2	32.25	39.76	27.99	1.07	59.71
	Bt2	47–64	6.76	158.4	7.1	0.28	25.4	21.9	42.91	30.56	26.54	1.16	56.36
PG	Oa	5–0	4.18	300	205.9	7.47	27.6	65.3	n.d	n.d	n.d	0.65	75.42
	A1	0–9	5.55	66.8	31.4	1.8	17.4	20.2	25.74	38.66	35.6	0.94	64.5
	A2	9–16.5	5.83	42.2	15.3	1.43	10.7	16.8	26.28	37.64	36.08	1.23	53.64
	AB	16.5–32	5.84	49.1	14.9	0.94	15.9	35.3	39.18	28.15	32.66	1.14	56.98
	Bt1	32–44	6.2	26.4	7.4	0.28	26.4	32.9	48.50	25.73	25.77	1.17	55.96
	Bt2	44–63	6.32	37.3	4.0	0.19	21.1	26	42.53	26.68	30.79	1.12	57.79
	C	63–74+	6.45	60	3.7	0.16	23.1	20.2	32.58	26.88	40.53	1.11	58.26
DG	A1	0–10	5.15	87	44.2	3.18	13.9	31.9	19.60	24.85	55.54	0.97	63.21
	A2	10–20	5.12	60	27.7	1.93	14.4	30.6	15.80	29.94	54.25	1.14	56.96
	Bt1	20–28	6.05	95	24.6	1.1	22.4	29.6	48.00	22.41	29.60	1.03	60.97
	Bt2	28–38/40	6.82	219	6.6	0.72	9.2	27.5	58.05	17.02	24.94	1.09	58.92
	BCK	38/40–55	7.44	226	3.7	0.32	11.6	24.6	22.35	8.04	49.39	1.09	58.82
	2Ckm	55+	n.d	n.d	n.d	n.d	n.d	n.d	n.d	n.d	n.d	n.d	n.d

PF pasture field, EG eucalyptus grove, PG afforested pine grove, DG mixed deciduous grove, EC electrical conductivity, OC organic content, Total N total nitrogen content, C:N ratio proportion of carbon to nitrogen, CEC cation exchange capacity,  $\rho_a$  bulk density and  $\varepsilon$  porosity, n.d not determined

productivity just allowed the formation of O horizons at the PG site, probably due to a more acidic and recalcitrant organic residues. In fact, the lowest pH value was found for the Oa horizon of the PG site. All the afforested sites had increased OC content in the 0–30 cm layer respective to the PF site (Table 2), having the vegetation a relevant imprint also on the OC concentration profile, which might be related to the migration of C soluble forms and deep root C allocation. An increased N concentration could be seen on the afforested surface horizons by biomass extraction and residues return, particularly at PG and EG sites, compared to PF site. However, the rate of soil OC built up was higher than the soil total N rate, yielding higher C: N ratios than those for PF site,

settled around the 10–12 classical ratio for Mollisols. Soil reaction was either influenced by soil vegetation as it is shown in surface horizons of the afforested sites, or could be influenced by ammonium based fertilizers for PF site. However, most of the subsurface horizons exhibit pH values related to adequate bases provision and in the BCK horizon of the DG site, carbonate activity could be ascertained from the soil reaction. Cation exchange capacity (CEC) was high through all the four soil profiles and was either related to the OC concentration or clay content. Argillic horizons (Bts), with evidence of clay illuviation (argillans) and peaks in clay content, were found at the four pits. Despite the vegetation type, elluviated (E) horizons were not found, and there were not

significant sand to clay ratio shifts suggesting its incipient formation with exception of the A2 to Bt1 horizons of the DG site. Bulk density correlated negatively to OC concentration and defined total porosity that was always high.

### Methane fluxes

All sites presented soils that resulted CH<sub>4</sub> sinks as it is indicated by their negative flux values (Table 3) which were in the range of  $-3.55$  to  $-14.39$  ng CH<sub>4</sub> m<sup>-2</sup> s<sup>-1</sup>. Lower flux values (absolute values) were found at PF and EG sites and the highest were found at DG site with statistically significant differences between them (ANOVA Test, post hoc Games Howell,  $p < 0.0001$ ). These results are similar to those reported previously for the same sites (Priano et al. 2014) and consistent with the literature (Dutaur and Verchot 2007; Hiltbrunner et al. 2012; Smith et al. 2000).

### Methane concentration profiles

Mean [CH<sub>4</sub>] profiles are shown in Fig. 1. In each case, the obtained profile (mean CH<sub>4</sub> concentration) is compared to three theoretical profiles that could exist if there were only diffusive processes that explain [CH<sub>4</sub>] at a particular depth. In this simulation, three CH<sub>4</sub> diffusion coefficients in soils ( $D_s$ ) were employed, that are related to its value in air at the STP ( $D_a$ ). To obtain these three theoretical profiles, Fick's Law (Eq. 1) and the mean CH<sub>4</sub> flux value for each site (Table 3) were used.

Each case shows a decrease in [CH<sub>4</sub>] reaching values between 0.2 and 1.4 ppm at a depth of 40 cm. Within the first layer (10 cm-deep), which is involved in CH<sub>4</sub> fluxes from the SAI, both PG and DG sites

showed a slope according to the theoretical profile  $D_s = 0.1 D_a$  with values close to  $0.02$  cm<sup>2</sup> s<sup>-1</sup> (lineal gradient,  $R^2 = 0.8$ ); this indicate that Fick's Law could be a good approximation to estimate CH<sub>4</sub> fluxes in these cases.

On the contrary, PF site presented a slope far away from the theoretical profile previously mentioned ( $D_s = 0.1 D_a$ ) that could suggest either the presence of CH<sub>4</sub> oxidation or diffusion processes that may prevail in this layer as a result of a  $D_s$  value much smaller than  $0.02$  cm<sup>2</sup> s<sup>-1</sup>. In this site, deeper layers show an important change in slope with an increase in CH<sub>4</sub> concentration reaching values close to atmospheric ones, which could indicate the presence of methanogenesis.

EG mean profile presented a decrease in [CH<sub>4</sub>] within a first thin layer of 5 cm and then at a depth of 25 cm. The slope in the first case (5 cm) is similar to that of a theoretical profile if  $D_s$  takes values of  $0.059$  cm<sup>2</sup> s<sup>-1</sup> ( $D_s = 0.3 D_a$ ).

### Soil water content and related parameters

There were statistically significant differences between sites in  $w$  %,  $\rho_a$ , WFPS and  $\varphi_g$  from the first 10 cm of the soil layer (Table 4); PF site presented the lowest value of  $\varphi_g$  and the highest  $\rho_a$ .

## Discussion

### Soil parameters and soil methane concentration profiles

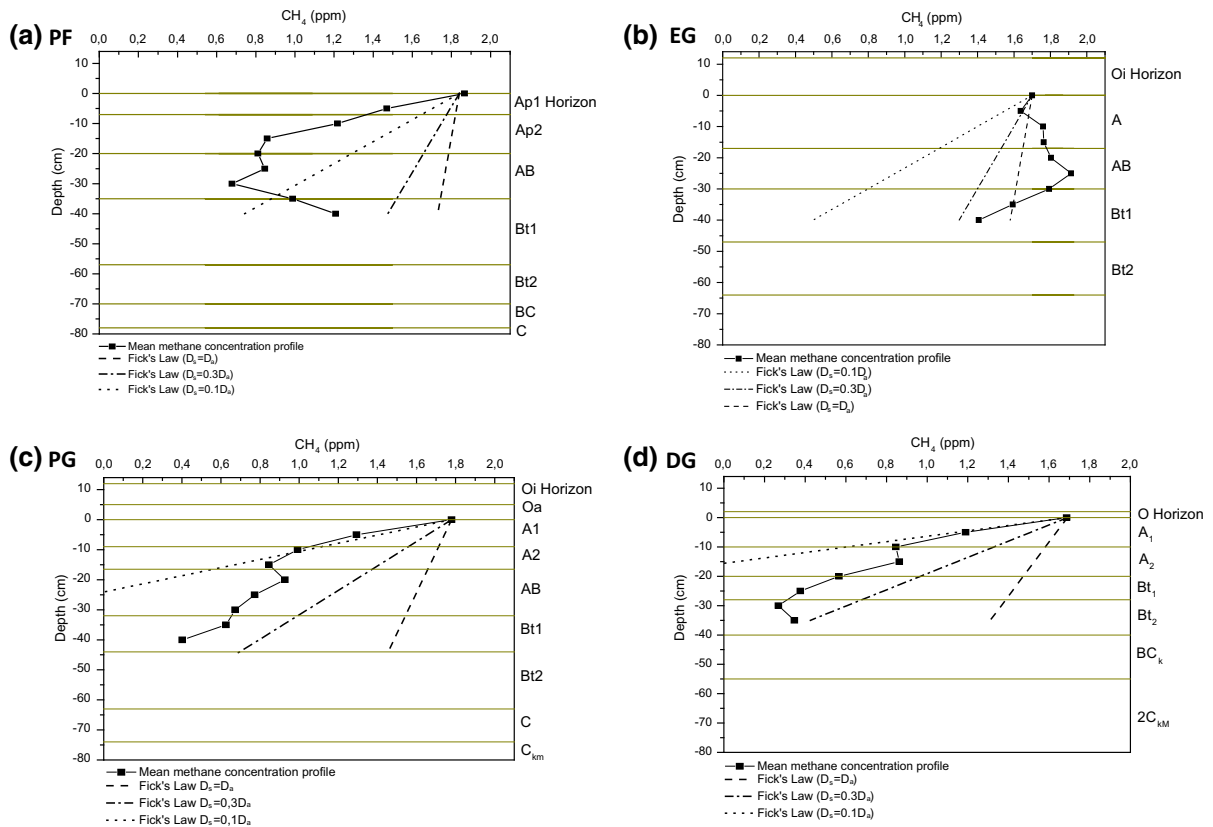
[CH<sub>4</sub>] showed a significant and positive lineal correlation with the proportion of silt in soils ( $R = 0.59$ ,

**Table 3** Mean CH<sub>4</sub> fluxes in ngCH<sub>4</sub> m<sup>2</sup> s<sup>-1</sup> for each site

Site	Mean methane flux (ngCH <sub>4</sub> m <sup>-2</sup> s <sup>-1</sup> )	CV	Min	Max	N
PF	$-3.55 \pm 1.10^{(a)}$	31.11	-4.94	-1.84	7
EG	$-3.99 \pm 2.29^{(a)}$	57.28	-7.29	-2.22	4
PG	$-9.81 \pm 2.42^{(b)}$	24.66	-11.92	-5.85	5
DG	$-14.39 \pm 4.79^{(c)}$	33.29	-20.09	-9.00	4

Different letters a, b and c indicate statistically significant intersite differences in CH<sub>4</sub> fluxes (ANOVA test Games Howell,  $p < 0.0001$ )

PF pasture field, EG eucalyptus grove, PG afforested pine grove, DG mixed deciduous grove, CV coefficient of variation, Min minimum value, Max maximum value, N numbers of measurements



**Fig. 1** Mean CH<sub>4</sub> concentration profiles measured during June–July 2013 for: **a** PF Pasture field; **b** EG Eucalyptus grove; **c** PG Afforested pine grove and **d** DG Mixed deciduous grove (solid lines), with its respective horizons. Lines represent three different theoretical profiles (Fick’s Law) for the three different

CH<sub>4</sub> diffusion coefficient values in soil ( $D_s$ ) compared with the value in air at the STP ( $D_a$ ): Fick’s Law with  $D_s = D_a$  (dashed lines), Fick’s Law with  $D_s = 0.3 D_a$  (Dash-dot lines) and Fick’s Law with  $D_s = 0.1 D_a$  (dotted lines)

**Table 4** Mean values of soil water content (w%), at the sampling dates, bulk density ( $\rho_a$ ), water filled pore space (WFPS) and air filled porosity ( $\phi_g$ )

Site	w%	$\rho_a$ (g cm <sup>-3</sup> )	WFPS	$\phi_g$
PF	28.25 <sup>(ab)</sup>	1.18 <sup>(a)</sup>	59.96 <sup>(a)</sup>	0.23 <sup>(a)</sup>
EG	32.93 <sup>(b)</sup>	0.94 <sup>(c)</sup>	28.99 <sup>(c)</sup>	0.53 <sup>(c)</sup>
PG	24.04 <sup>(a)</sup>	0.91 <sup>(b)</sup>	34.43 <sup>(b)</sup>	0.43 <sup>(bc)</sup>
DG	30.79 <sup>(ab)</sup>	0.96 <sup>(abc)</sup>	45.58 <sup>(ab)</sup>	0.35 <sup>(b)</sup>

PF pasture field, EG eucalyptus grove, PG afforested pine grove, DG mixed deciduous grove

Letters a b and c indicate statistically significant intersite differences (ANOVA test  $p < 0.03$  for w %,  $p < 0.001$  for the rest of the parameters)

$p < 0.01$ ), Table 2; Fig. 1, but a negative one with the clay content ( $R = -0.48$ ,  $p < 0.01$ ) and  $\rho_a$  ( $R = -0.40$ ,  $p < 0.01$ ). These negative relationships

with [CH<sub>4</sub>] could be supported by the effects that clay has on the pore size distribution, leading to a dominant microporosity (Hillel 2006). Additionally, it was yet established a negative relationship between clays and methanotrophic activity (Bender and Conrad 1995); although the maximum clay content of the studied soils occurred at a depth (Bt horizons) where restriction to oxygen diffusion probably will create an environment not prone to methanotrophy, as the change in the slope of [CH<sub>4</sub>] indicates at PF and DG sites. On the other hand, the increase on  $\rho_a$  implies a decrease in  $\epsilon$ , with probable implications on CH<sub>4</sub> diffusivity. These two parameters, clay content and  $\rho_a$ , determine  $D_s$  value according to Prajapati and Jacinthe (2014), affecting [CH<sub>4</sub>] gradient and, in consequence, CH<sub>4</sub> oxidation. PF site presented the highest values for both parameters in the first layer and the lowest CH<sub>4</sub> oxidation (Table 3).

At the same time,  $[\text{CH}_4]$  correlated negatively with soil pH values ( $R = -0.52$ ,  $p < 0.01$ ) suggesting a restriction to methanotrophic activity in the more acid O–A horizons developed at the afforested sites, as it was observed by Smith et al. (2000). Moreover,  $\varepsilon$  and  $w\%$  showed significant and positive lineal correlation with  $[\text{CH}_4]$  ( $R = 0.40$ ,  $p < 0.01$  and  $R = 0.32$ ,  $p < 0.01$  respectively). Finally, a significant and positive lineal correlation between  $[\text{CH}_4]$  and total nitrogen content was found ( $R = 0.52$ ,  $p < 0.01$ ) for the afforested sites. High values of  $[\text{CH}_4]$  indicate low  $\text{CH}_4$  oxidation which is related with high amount of total nitrogen content. This is in agreement with findings of Hütsch (1998).

A multiple regression with the most significant physical parameters (silt and clay content,  $\rho_a$  and  $\varepsilon$ ) showed that these parameters explain 68% of the  $[\text{CH}_4]$  variation in the soil profile ( $R^2 = 0.68$ ,  $p < 0.01$ ). The adjustment improved when  $\varepsilon$  was excluded from the analysis as it was well correlated with  $\rho_a$  (they are covariants) ( $R^2 = 0.69$ ,  $p < 0.01$ ). These results indicate that, at least for the first layer,  $[\text{CH}_4]$  profiles could be explained mainly by differences in physical parameters among soil layers as a result of the diffusion of atmospheric  $\text{CH}_4$  into the soil. Subsequently it should be evaluated if there are differences in methanotrophic population between sites, as a result of afforestation.

A mean  $[\text{CH}_4]$  statistical analysis between sites showed that DG and PG sites presented the lowest  $[\text{CH}_4]$  values along the soil profile (0.61–0.88 ppm), as it is shown in Fig. 1. These results agree with the  $\text{CH}_4$  fluxes values from the SAI (Table 3).

Methane diffusion coefficient in soils ( $D_s$ ) and estimated methane flux (Fick's Law)

According to Eq. 2, 3 and 4  $\text{CH}_4$  coefficient values ( $D_s$ ) were obtained for the first layer of each site

(Table 5) which then allowed us to estimate  $\text{CH}_4$  flux (Fick's law, Eq. 1) base on  $[\text{CH}_4]$  profiles (Fig. 1), assuming no oxidation or emission processes of  $\text{CH}_4$  at the analyzed depth.

Coinciding with what was previously observed (Fig. 1), the lowest  $D_s$  values were for the reference site, PF site ( $\ll 0.01 \text{ cm}^2 \text{ s}^{-1}$ ), lower than 10% of  $D_a$  value but similar to those reported in other works (Prajapati and Jacinthe 2014). EG site showed the highest  $D_s$  values close to  $0.05 \text{ cm}^2 \text{ s}^{-1}$  which could explain the small variation in  $[\text{CH}_4]$  along the profile, and PG and DG presented intermediate values. As other works report (Kusa et al. 2008; Prajapati and Jacinthe 2014), a marked variation occurred in each case (CV values). However, intersite differences in  $D_s$  values are statistically significant ( $p < 0.001$ ) as letters a, b and c show (Table 5) and they could be explained by differences in  $w\%$  and its related parameters (Table 4) that were used to calculate the  $D_s$  values.

Furthermore, a new methane flux was estimated for the first soil layer in each site according to Fick's Law employing mean  $D_s$  values (Table 5) and  $[\text{CH}_4]$  measured in each case (Fig. 1). Results are also shown in Table 5; letters a, b and c indicate the statistically significant differences between sites (ANOVA test,  $p < 0.001$ ). According to this, the estimated  $\text{CH}_4$  flux for PF site was similar to that registered by the static chamber technique (Table 3). This result could indicate that, in this case, diffusive processes prevail in the first layer and most of the  $\text{CH}_4$  oxidation could occur in deeper layers as Fig. 1 shows. Mean  $\text{CH}_4$  flux estimated for EG site was 26% of that measured using the static chamber technique (Table 3) and showed great variation. In PG site, mean  $\text{CH}_4$  flux estimated was 82% higher than that on Table 3. As mean  $D_s$  value was estimated to be close to 15% of  $D_a$

**Table 5** Mean  $\text{CH}_4$  diffusion coefficient values ( $D_s$ ,  $\text{cm}^2 \text{ s}^{-1}$ ) and estimated  $\text{CH}_4$  flux ( $\text{ngCH}_4 \text{ m}^{-2} \text{ s}^{-1}$ ) for each site

Site	$D_s$ Mean values ( $\text{cm}^2 \text{ s}^{-1}$ )	CV %	N	Estimated $\text{CH}_4$ flux ( $\text{ngCH}_4 \text{ m}^{-2} \text{ s}^{-1}$ )	CV %	N
PF	$0.0067 \pm 0.0036^{(a)}$	53.70	6	$-3.32 \pm 2.13^{(a)}$	64.05	6
EG	$0.0451 \pm 0.0096^{(c)}$	21.38	5	$-1.03 \pm 8.64^{(ab)}$	835.4	6
PG	$0.0294 \pm 0.0147^{(b)}$	50.03	5	$-17.90 \pm 5.67^{(c)}$	31.68	6
DG	$0.0179 \pm 0.0105^{(b)}$	58.32	5	$-10.61 \pm 2.21^{(b)}$	20.81	8

PF pasture field, EG eucalyptus grove, PG afforested pine grove, DG mixed deciduous grove, CV coefficient of variation, N numbers of measurements; and letters a, b and c indicate statistically significant intersite differences (ANOVA Test, Games Howell  $p < 0.001$ )



(Table 5), the  $[\text{CH}_4]$  gradient in the first 10 cm layer shown in Fig. 1 moved away from which should be obtained if only diffusion occurs which a  $D_s$  value between  $0.1 D_a$  and  $0.3 D_a$ . Therefore, the gradient difference observed could be due to  $\text{CH}_4$  oxidation in this layer ( $A_1$  horizon) and Fick's Law would not be a good approximation for this site as it was mentioned in 3.3, at least for this layer. In contrast, in DG site, the mean  $\text{CH}_4$  flux estimated was 26% lower than that detected by the static chamber (Table 3) but the  $[\text{CH}_4]$  gradient for the first layer (Fig. 1) was similar to the theoretical one for a  $D_s$  value close to 10% of  $D_a$  (and similar to the estimated  $D_s$  value, Table 5), which would suggest predominance of diffusive processes. Similar results were found in Hütsch (1998) where a strong  $\text{CH}_4$  oxidation was reported at a subsurface horizon in forest soils.

Therefore, differences in  $\text{CH}_4$  fluxes at the SAI from PF and DG sites could be explained mainly by differences in soil physical parameters ( $\rho_a$ ,  $\phi_g$ , Table 4) that regulate  $\text{CH}_4$  diffusion into the soil and determine  $\text{CH}_4$  diffusion coefficient for which, PF presented the lowest value. Also, methanogenesis in deeper layers in PF site (mean  $[\text{CH}_4] = 1.2$  ppm) could account for intersite differences. For this reason, Fick's Law could be a good approximation to estimate  $\text{CH}_4$  fluxes for these sites with a certain  $\text{CH}_4$  diffusion coefficient.

In contrast, in PG site diffusion likely prevails in horizons  $O_1$  and  $O_2$  and Fick's law could be applicable to estimate  $\text{CH}_4$  fluxes considering solely the  $[\text{CH}_4]$  gradient in horizons  $O_1$  and  $O_2$ , which was not measure in this work.

Finally, EG site presented a  $[\text{CH}_4]$  profile that agrees with its estimated  $\text{CH}_4$  diffusion coefficient, which was the highest of all ( $\sim 0.05 \text{ cm}^2 \text{ s}^{-1}$ ).  $\text{CH}_4$  flux at the SAI was low in this case and the reason could be methanotrophic activity present in deeper layers as it is shown in the  $[\text{CH}_4]$  profile and differences in its bacterial community. For this reason, Fick's law would not be appropriate for estimating  $\text{CH}_4$  flux.

## Conclusions

Statistically significant differences between sites were found regarding  $\text{CH}_4$  fluxes at the SAI that may result from differences observed in  $[\text{CH}_4]$  profiles and  $D_s$ ,

The strongest  $\text{CH}_4$  sinks were found in DG and PG sites which also showed the lowest  $[\text{CH}_4]$  values. Such values could be partly explained by differences in silt and clay content and  $\rho_a$  along the soil profile, but they are also the result of methanotroph activity which was not measured in this work. Afforestation can improve the physical and biological soil attributes linked to  $\text{CH}_4$  consumption as it meliorates its diffusivity.

Besides,  $\text{CH}_4$  fluxes could be estimated by Fick's Law, for some cases, using a particular  $D_s$  and a  $[\text{CH}_4]$  profile; taking into account that  $\text{CH}_4$  consumption seems to be present in deeper layers and it is limited by physical factors.

However, more measurements must be performed to determine the spatial and temporal variability of the parameters that regulate  $\text{CH}_4$  fluxes in these sites and studies of the potential  $\text{CH}_4$  oxidation must be carried out to confirm and understand the differences in  $\text{CH}_4$  fluxes between these sites.

**Acknowledgements** Financial support was provided by PICT 2010-1010 and PICT 2015-2540 of the National Agency for Scientific and Technological Research (ANPCyT) of the Ministry of Science, Technology and Innovation (MINCYT), Argentina.

## References

- Bender M, Conrad R (1995) Effect of  $\text{CH}_4$  concentrations and soil conditions on the induction of  $\text{CH}_4$  oxidation activity. *Soil Biol Biochem* 27(12):1517–1527. doi:[10.1016/0038-0717\(95\)00104-M](https://doi.org/10.1016/0038-0717(95)00104-M)
- Boeckx P, Van Cleemput O, Villaralvo I (1997) Methane oxidation in soils with different textures and land use. *Nutr Cycl Agroecosyst* 49:91–95. doi:[10.1023/A:1009706324386](https://doi.org/10.1023/A:1009706324386)
- Chaparro MAE, Bidegain JC, Sinito AM, Gogorza CSG, Jurado S (2003) Preliminary results of magnetic measurements on stream-sediments from Buenos Aires Province, Argentina. *Stud Geophys Geod* 47:121–145. doi:[10.1023/1022255706899](https://doi.org/10.1023/1022255706899)
- Curry CL (2007) Modeling the soil consumption at atmospheric methane at the global scale. *Glob Biogeochem Cycles* 21:1–15. doi:[10.1029/2006GB002818](https://doi.org/10.1029/2006GB002818)
- Del Grosso SJ, Parton WJ, Mosier AR, Ojima DS, Potter CS, Borken W, Brumme R, Butterbach-Bahl K, Crill P M, Dobbie K, Smith KA (2000) General  $\text{CH}_4$  oxidation model and comparisons of  $\text{CH}_4$  Oxidation in natural and managed systems. *Glob Biogeochem Cycles* 14:999–1019. doi:[10.1029/1999GB001226](https://doi.org/10.1029/1999GB001226)
- Dutaur L, Verchot LV (2007) A global inventory of the soil  $\text{CH}_4$  sink. *Glob Biogeochem Cycles* 21:1–9. doi:[10.1029/2006GB002734](https://doi.org/10.1029/2006GB002734)

- Hillel D (2006) Environmental soil physics. fundamentals, applications, and environmental considerations. Academic Press, New York
- Hiltbrunner D, Zimmermann S, Karbin S, Hagedorn F, Niklaus PA (2012) Increasing soil methane sink along a 120-year afforestation chronosequence is driven by soil moisture. *Glob Change Biol* 18:3664–3671. doi:[10.1111/j.1365-2486.2012.02798.x](https://doi.org/10.1111/j.1365-2486.2012.02798.x)
- Hou L-Y, Wang Z-P, Wang J-M, Wang B, Zhou S-B, Li L-H (2012) Growing season in situ uptake of atmospheric methane by desert soils in a semiarid region of northern China. *Geoderma* 189–190:415–422. doi:[10.1016/j.geoderma.2012.05.012](https://doi.org/10.1016/j.geoderma.2012.05.012)
- Hütsch BW (1998) Tillage and land use effects on methane oxidation rates and their vertical profiles in soil. *J Biol Fert Soils* 27:284–292. doi:[10.1007/s003740050435](https://doi.org/10.1007/s003740050435)
- INTA-CIRN (1989) Cartas de Suelos de la República Argentina, Instituto de Suelos. INTA, Buenos Aires
- Jensen S, Olsen RA (1998) Atmospheric methane consumption in adjacent arable and forest soil systems. *Soil Biol Biochem* 30:1187–1193. doi:[10.1016/S0038-0717\(97\)00149-1](https://doi.org/10.1016/S0038-0717(97)00149-1)
- Kusa K, Sawamoto T, Hu R, Hatano R (2008) Comparison of the closed-chamber and gas concentration gradient methods for measurement of CO<sub>2</sub> and N<sub>2</sub>O fluxes in two upland field soils. *J Soil Sci Plant Nutr* 54:777–785. doi:[10.1111/j.1747-0765.2008.00292.x](https://doi.org/10.1111/j.1747-0765.2008.00292.x)
- Pazos MS (1984) Relación arcilla iluvial/arcilla total en moli-soles del sudeste de la Provincia de Buenos Aires. *Ciencia del Suelo* 2:132–136
- Pazos MS, Mestelán S (2002) Variability and soil classification of Udolls with tosa (caliche). *Soil Sci Soc Am J* 66:1256–1264. doi:[10.2136/sssaj2002.1256](https://doi.org/10.2136/sssaj2002.1256)
- Prajapati P, Jacinthe PA (2014) Methane oxidation kinetics and diffusivity in soils under conventional tillage and long-term no-till. *Geoderma* 230–231:161–170. doi:[10.1016/j.geoderma.2014.04.013](https://doi.org/10.1016/j.geoderma.2014.04.013)
- Priano ME (2014) Gases de efecto invernadero: Mediciones de flujo en la interfaz suelo-atmósfera y sus concentraciones en el aire del suelo. Ph.D Thesis, Universidad Nacional del centro de la Provincia de Buenos Aires, Tandil, Buenos Aires, Argentina
- Priano ME, Fusé VS, Gere JI, Berkovic AM, Williams KE, Guzmán SA, Grattón R, Juliarena MP (2014) Tree plantations on a grassland region: effects on methane uptake by soils. *Agrofor Syst* 88:187–191. doi:[10.1007/s10457-013-9661-6](https://doi.org/10.1007/s10457-013-9661-6)
- Price SJ, Sherlock RR, Kelliher FM, Mc Seveny TM, Tate KR, Condron LM (2003) Pristine New Zealand forest soil is a strong methane sink. *Glob Change Biol* 10:16–26. doi:[10.1046/j.1529-8817.2003.00710x](https://doi.org/10.1046/j.1529-8817.2003.00710x)
- Ridgwell AJ, Marshall SJ, Gregson K (1999) Consumption of atmospheric methane by soils: a process-based model. *Glob Biogeochem Cycles* 13:59–70. doi:[10.1029/1998GB900004](https://doi.org/10.1029/1998GB900004)
- Schoeneberger PJ, Wysocki DA, Benham EC, Broderon WD (editors) (2002) Field book for describing and sampling soils, version 2.0. Natural Resources Conservation Service, National Soil Survey Center, Lincoln, NE
- Sistema de Apoyo Metodológico a los Laboratorios de Análisis de Suelos, Agua, Vegetales y Enmiendas Orgánicas (SAMLA) (2004) Guidelines for soil testing analysis. 1st Ed. SAGPyA (Secretary of Agriculture and Fisheries). CD Rom. ISBN 987-918440-8
- Smith KA, Dobbie KE, Ball BC, Bakken LR, Sitaula BK (2000) Oxidation of atmospheric methane in Northern European soils, comparison with other ecosystems, and uncertainties in the global terrestrial sink. *Glob Change Biol* 6:791–803. doi:[10.1046/j.1365-2486.2000.00356.x](https://doi.org/10.1046/j.1365-2486.2000.00356.x)
- Smith KA, Ball T, Conen F, Dobbie KE, Massheder J, Rey A (2003) Exchange of greenhouse gases between soil and atmosphere: interactions of soil physical factors and biological processes. *Eur J Soil Sci* 54:779–791. doi:[10.1046/j.1365-2389.2003.00567.x](https://doi.org/10.1046/j.1365-2389.2003.00567.x)
- Soil Survey Laboratory Staff (SSLS) (2004) Soil Survey Laboratory Methods Manual. Soil Survey Investigations Report No. 42. Version 2.5. USDA-NRCS, Washington, DC
- Soil Survey Staff (SSS) (2014) Keys to Soil Taxonomy. 12th Ed. USDA (United States Department of Agriculture)-NRCS (National Resources Conservation Services), Washington, DC
- Striegel RG, Proceedings of the NATO Advanced Research Workshop (1993) Diffusional limits to the consumption of atmospheric methane by soils. *Chemosphere* 26:715–720, ISSN 0045-6535. doi:[10.1016/0045-6535\(93\)90455-E](https://doi.org/10.1016/0045-6535(93)90455-E)
- Tate KR (2015) Soil methane oxidation and land-use change – from process to mitigation. *Soil Biol Biochem* 80:260–272. doi:[10.1016/j.soilbio.2014.10.010](https://doi.org/10.1016/j.soilbio.2014.10.010)
- Von Fischer JC, Butters G, Duchateau PC, Thelwell RJ, Siller R (2009) In situ measures of methanotroph activity in upland soils: a reaction-diffusion model and field observation of water stress. *J Geophys Res Biogeosci* 114:1–12. doi:[10.1029/2008JG000731](https://doi.org/10.1029/2008JG000731)