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Indications of nitrogen-limited methane uptake in tropical forest soils

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Abstract

Tropical forest soils contribute 6.2 Tg yr^{-1} (28 %) to global methane (CH_4) uptake, which is large enough to alter CH_4 accumulation in the atmosphere if significant changes would occur to this sink. Elevated deposition of inorganic nitrogen (N) to temperate forest ecosystems has been shown to reduce CH_4 uptake in forest soils, but almost no information exists from tropical forest soils even though projections show that N deposition will increase substantially in tropical regions. Here we report the results from long-term, ecosystem-scale experiments in which we assessed the impact of chronic N addition on soil CH_4 fluxes from two old-growth forests in Panama: (1) a lowland, moist (2.7 m yr^{-1} rainfall) forest on clayey Cambisol and Nitisol soils with controls and N-addition plots for 9–12 yr, and (2) a montane, wet (5.5 m yr^{-1} rainfall) forest on a sandy loam Andosol soil with controls and N-addition plots for 1–4 yr. We measured soil CH_4 fluxes for 4 yr (2006–2009) in 4 replicate plots (40 m × 40 m each) per treatment using vented static chambers (4 chambers per plot). CH_4 fluxes from the lowland control plots and the montane control plots did not differ from their respective N-addition plots. In the lowland forest, chronic N addition did not lead to inhibition of CH_4 uptake; instead, a negative correlation of CH_4 fluxes with nitrate (NO_3^-) concentrations in the mineral soil suggests that increased NO_3^- levels in N-addition plots had stimulated CH_4 consumption and/or reduced CH_4 production. In the montane forest, chronic N addition also showed negative correlation of CH_4 fluxes with ammonium concentrations in the organic layer, which suggests that CH_4 consumption was N limited. We propose the following reasons why such N-stimulated CH_4 consumption did not lead to statistically significant CH_4 uptake: (1) for the lowland forest, this was caused by limitation of CH_4 diffusion from the atmosphere into the clayey soils, particularly during the wet season, as indicated by the strong positive correlations between CH_4 fluxes and water-filled pore space (WFPS); (2) for the montane forest, this was caused by the high WFPS in the mineral soil throughout the year, which may not only limit CH_4 diffusion from the atmosphere into the soil but also favour CH_4 production; and (3) both forest soils

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showed large spatial and temporal variations of CH_4 fluxes. We conclude that in these extremely different tropical forest ecosystems there were indications of N limitation on CH_4 uptake. Based on these findings, it is unlikely that elevated N deposition on tropical forests will lead to widespread inhibition of CH_4 uptake.

1 Introduction

Methane (CH_4) is an important atmospheric trace gas because it influences both the energy and the oxidant balance of the earth's atmosphere. Presently, the atmospheric concentration of CH_4 is about 1800 ppbv, which accounts for about 0.48 $W m^{-2}$ of the total anthropogenic radiative forcing (Denman et al., 2007). About 75 % of the global CH_4 source strength, which is about 600 $Tg yr^{-1}$, originates from biogenic sources wherein CH_4 is exclusively produced by methanogenic microorganisms (Conrad, 1989). Although CH_4 is especially produced in wetland soils, CH_4 production can also occur in upland soils during high rainfall or wet season, for example in anaerobic microsites inside soil aggregates (Keller and Reiners, 1994). In well-aerated soils, CH_4 is oxidized by methanotrophic microorganisms and CH_4 oxidation normally exceeds production, which results in a net CH_4 uptake. The largest biogenic sink of atmospheric CH_4 is through uptake by upland soils, which contributes about 5 % to the total removal of CH_4 from the atmosphere (Conrad, 2007).

Tropical ecosystems play an important role in the production and uptake of atmospheric CH_4 (Keller and Matson, 1994). In tropical forest areas, known wetland sources of CH_4 production do not suffice to explain the observed high CH_4 concentrations over neotropical forests (Frankenberg et al., 2008) and some "cryptic" wetlands may contribute significantly to the CH_4 production (Martinson et al., 2010). Most tropical forests grow on well-drained upland soils that are too dry to emit CH_4 but act instead as an important sink for atmospheric CH_4 (Kiese et al., 2003). In a recent review where measurements were stratified according to climatic zone, ecosystem and soil texture, the total global CH_4 uptake was estimated at 22.4 $Tg yr^{-1}$ of which 9.2 $Tg yr^{-1}$ (41 %)

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occurred in tropical ecosystems (Dutaur and Verchot, 2007). The contribution of tropical forest soils to global CH_4 uptake was estimated at 6.2 Tg yr^{-1} (28 %), which is large enough to alter the CH_4 accumulation in the atmosphere if significant changes would occur to this sink.

The increased use of nitrogen (N) fertilizers, fossil fuel, and cultivation of N-fixing crops have more than doubled the amount of "reactive" nitrogen (N_r) cycling worldwide (Vitousek et al., 1997). In the past decades, this has led to enhanced N_r input in forest ecosystems, especially in economically-developed regions of the temperate zone. Projections are that the input of N_r will increase substantially in tropical regions such as Southeast Asia and South and Central America due to increasing agricultural and industrial use of N (Galloway et al., 2008). A recent study suggested that elevated anthropogenic N_r deposition is probably already widespread in tropical forests (Hietz et al., 2011).

Elevated depositions of mineral N (ammonium (NH_4^+) and nitrate (NO_3^-)) and N fertilization to forest ecosystems have been shown to affect CH_4 fluxes from forest soils (Steudler et al., 1989; Brumme and Borken, 1999). Several mechanisms have been proposed to explain how mineral N affects CH_4 fluxes in upland soils. Most commonly, the inhibition of CH_4 oxidation in the soil by increased NH_4^+ levels is mentioned, not only in temperate soils (Steudler et al., 1989; Crill et al., 1994) but also in tropical soils (Veldkamp et al., 2001). The enzyme methane monooxygenase, which initiates the oxidation pathway of CH_4, is also able to oxidize NH_4^+. When NH_4^+ competes with CH_4 for reactive sites of methane monooxygenase, this may cause inhibition of CH_4 oxidation (Conrad, 1996). An osmotic effect may also contribute to the inhibition of CH_4 oxidation (Nesbit and Breitenbeck, 1992; Veldkamp et al., 2001). There is a discrepancy in published literature about the duration over which NH_4^+ can inhibit CH_4 oxidation. An inhibition effect of NH_4 for 13 yr has been reported (Mosier et al., 1996) whereas in another study inhibition lasted only about four weeks (Veldkamp et al., 2001). On the other hand, increased NO_3^- levels can inhibit CH_4 production because NO_3^- is preferred as an electron acceptor over bicarbonate (Conrad, 1989), and some intermediates if NO_3^-

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is denitrified (NO_2^-, NO, N_2O) can be toxic for methanogenic microorganisms (Conrad, 2007). While it was relatively early recognized that methanotrophic microorganisms also need a N source and thus could be N limited (Bender and Conrad, 1995), this was not explored in subsequent years. Only recently, Bodelier and Laanbroek (2004) showed through a literature review that many indications for N limitation of soil CH_4 consumption have been ignored in earlier studies. Apart from N limitation of growth and activity of CH_4-oxidizing bacteria, they also proposed that switching from fixation of molecular N to assimilation of mineral N can cause almost instantaneous changes in CH_4-oxidizing activity.

Presently, only one N-manipulation study is published about N effects on soil CH_4 fluxes from (sub)tropical forests and this was conducted in China (Zhang et al., 2008, 2011). In this study, N was applied monthly at rates ranging from 50 to 150 kg N ha^{-1} yr^{-1} and the studied forests were mature, disturbed and rehabilitated forests. In the mature forest, CH_4 uptake decreased with increasing N application rate whereas in the disturbed and rehabilitated forest no N-addition effect was observed. The authors concluded that the response of soil CH_4 uptake to N addition in tropical forests varied depending on the soil N status; the lack of effect from the disturbed and rehabilitated forest was explained by intense competition for N by the vegetation (Zhang et al., 2008).

Here we report the impact of chronic N additions on soil CH_4 fluxes from two species-rich, old-growth forests in Panama: a lowland, moist forest on clayey Cambisol and Nitisol soils, and a montane, wet forest on a sandy loam Andosol soil covered with an organic layer. We hypothesized that: (1) in the lowland forest, with large soil N-cycling rates (Corre et al., 2010) and tree stem diameter growth ($\geq$ 10 cm diameter trees) and fine litterfall that were not N limited (Wright et al., 2011), long-term N addition will inhibit CH_4 uptake; (2) in the montane forest, with smaller soil N-cycling rates (Corre et al., 2010) and tree stem diameter growth (10–50 cm diameter trees) and fine litterfall that were N limited (Adamek et al., 2009), long-term N addition will stimulate CH_4 uptake. We tested these hypotheses by comparing soil CH_4 fluxes over a period of four years

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(2006–2009) in the lowland forest between control and N-addition plots during 9–12 yr of N additions and in the montane forest between control and N-addition plots during 1–4 yr of N additions. Our objectives were to (1) assess changes in soil CH_4 fluxes as a result of long-term N addition, and (2) relate these changes to soil-extractable NO_3^-, NH_4^+ and soil water-filled pore space (WFPS), which are factors that potentially control soil CH_4 fluxes. This is the first study to report how CH_4 fluxes change under chronic N addition to diverse, old-growth neotropical forests.

2 Materials and methods

2.1 Approach

We applied N fertilizer to create N-enriched condition, which ultimately will be the result of increased atmospheric N deposition. N deposition normally enters the ecosystem at the canopy level at low N concentrations with frequent occurrence in a year whereas we applied N fertilizer to the soil at high N concentration in four doses per year (see below). One of the artefacts of N fertilization is the occurrence of pronounced peaks of soil mineral N concentrations, which can affect short-term CH_4 fluxes within the first weeks following N application (Veldkamp et al., 2001). We therefore did a separate statistical analysis for CH_4 fluxes that include all measurements conducted from 1 day to 3 months after a N application and for CH_4 fluxes that were measured $\geq$ 6 weeks after the last N application (hereafter referred as long-term CH_4 fluxes). The long-term CH_4 fluxes are unlikely to be affected by the artificially high mineral N concentrations directly following N application. Furthermore, the type of N fertilizer (in our case urea) will be less important for the long-term CH_4 fluxes because within six weeks following urea application in our study sites urea-N was hydrolyzed and processed in the internal soil N cycle (Koehler et al., 2009).

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2.2 Site description and experimental design

The lowland forest (25–61 m elevation) consists of an old-growth (> 200 yr), semi-deciduous forest and is located on the Gigante Peninsula (9° 06′ N, 79° 50′ W) which is part of the Barro Colorado Nature Monument, Republic of Panama. On the nearby Barro Colorado Island, annual rainfall averages 2715 ± 139 mm (1999–2010) with a dry season from January to April. Ambient N deposition from rainfall was 9 kg N ha^{-1} yr^{-1}, measured bi-weekly in 2006–2007 at the shore of Gigante Peninsula near the study site (Corre et al., 2010). The mean annual air temperature is 27.2 ± 0.1 °C. Stem diameter growth of trees with ≥ 10 cm diameter at breast height (dbh), fine litter production, and fine-root biomass within 0–10 cm depth were not affected by 11 yr of N addition (Wright et al., 2011). The soils are Endogleyic Cambisol in the lower parts of the landscape and Acric Nitisol in the upper parts of the landscape, both with heavy clay texture; after 8 yr of N addition significant decreases in soil pH (control = 5.1 ± 0.1, N addition = 4.8 ± 0.1) and base saturation (control = 67 ± 8 %, N addition = 41 ± 7 %) and increase in exchangeable aluminium (Al) (control = 213 ± 39 g Al m^{-2}, 8 yr N addition = 297 ± 44 g Al m^{-2}) were observed in the 0–50 cm depth mineral soil.

The montane forest (1200–1300 m elevation) consists of an old-growth lower montane forest and is located in the Fortuna Forest Reserve in the Cordillera Central (8° 45′ N, 82° 15′ W), Chiriquí province, Republic of Panama. Mean annual rainfall is 5461 ± 250 mm (1997–2010) with no dry season. Ambient N deposition from rainfall was 5 kg N ha^{-1} yr^{-1}, measured biweekly in 2006–2007 at a forest clearing near the study site (Corre et al., 2010). The annual mean air temperature is 20.3 ± 0.2 °C. Stem diameter growth of trees with 10–50 cm dbh and fine litter production increased during the first 2 yr of N addition compared with the control plots (Adamek et al., 2009) whereas fine-root biomass and production (from organic layer down to 20 cm depth of the mineral soil) were not affected by N addition (Adamek et al., 2011). The soil is Aluandic Andosol with sandy loam texture and has an organic layer thickness of 10 ± 1 cm; after 3 yr of N addition no significant changes in pH (control = 4.7 ± 0.1,

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N addition = 4.6 ± 0.2), base saturation (control = 8 ± 3 %, N addition = 11 ± 4 %), and exchangeable Al (control = 252 ± 16 g Al m^{-2}, N addition = 280 ± 24 g Al m^{-2}) were observed in the 0–50 cm depth mineral soil.

The N-addition experiment in the lowland forest was part of an on-going nutrient manipulation study established in 1998 (Wright et al., 2011) whereas the N-manipulation experiment in the montane forest started in 2006 (Corre et al., 2010). At both sites, four replicates of N-addition plots and four controls were established. The size of the plots was 40 m × 40 m, separated by at least 40 m of buffer zone where no manipulation was done. The N-addition plots received 125 kg urea – N ha^{-1} yr^{-1} split in four applications (i.e. during the rainy season (May–December) for the lowland forest, and every quarter of the year for the montane forest). Measurements were conducted in the central 20 × 20 m area of the plot to prevent possible edge effects (e.g. roots from trees outside the plots growing into the N-fertilized plots).

2.3 CH_4 flux measurements

Soil CH_4 fluxes were measured using vented static chambers. Four permanent chamber bases (area 0.04 m^2, height 0.25 m, total volume with cover 11 L) were installed on each plot in a stratified random design along two perpendicular 20 m long transects that crossed in the plot's center. Four gas samples (100 mL each) were removed at 2, 12, 22 and 32 min after chamber closure and stored in pre-evacuated glass containers with a teflon-coated stopcock. Gas samples were analyzed in the field station in Panama using a gas chromatograph (Shimadzu GC-14B, Columbia, MD, USA) equipped with a flame ionization detector and an autosampler (Loftfield et al., 1997). CH_4 concentrations were determined by comparison of integrated peak areas of samples with those of three to four standard gases (depending on concentrations: 250, 1499, 1996, 9900 and 20010 ppb CH_4; Deuste Steininger GmbH, Mühlhausen, Germany). Gas fluxes were calculated from the concentration change in the chamber versus time and were adjusted for air temperature and atmospheric pressure measured at the time of sampling. To account for the decreasing diffusion gradient over time caused by the chamber

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feedback, we fitted both a linear and a quadratic regression model if CH_4 concentrations increased or decreased asymptotically (Wagner et al., 1997). We chose the statistically more adequate model based on the Akaike Information Criterion. The quadratic model was used in 14 % of the flux calculations in the montane forest and in 20 % of the gas flux calculations in the lowland forest. If CH_4 concentrations leveled out over time and the quadratic model was statistically inferior, we excluded the last data point and calculated the flux based on a linear model. These data screening and calculation procedures ensure that we minimized underestimations which may occur if a linear model was uncritically applied to static chamber flux data (Livingston et al., 2006). Positive fluxes indicate CH_4 emission from the soil; negative fluxes indicate CH_4 uptake by the soil. Zero fluxes were included. The annual CH_4 fluxes were approximated by applying the trapezoid rule on time intervals between measured flux rates, assuming constant flux rates per day.

2.4 Soil mineral N and moisture

From earlier experience in tropical forests, we learned that short storage of disturbed soil samples can considerably alter mineral N concentrations (Arnold et al., 2008). We therefore conducted mineral N extractions in the field. Parallel to gas sampling, four samples of mineral soil (0–0.05 m depth) were collected within the central 10 m × 10 m of each plot. For the montane site, we sampled the organic layer and 0–5 cm depth mineral soil separately. While in the field, samples were pooled for each plot, leaves and roots were manually removed, and a subsample (50–60 g fresh weight) was added to a prepared extraction bottle containing 150 mL of 0.5 mol L^{-1} K_2SO_4. Shaking (1 h) and filtering continued upon arrival in the field station which was at most 6 h from field extraction. Soil extracts were frozen and kept that way during air transport to the University of Goettingen (Germany), where NH_4^+ and NO_3^- contents were analyzed using continuous flow injection colorimetry (Cenco/Skalar Instruments, Breda, Netherlands). NH_4^+ was determined using the Berthelot reaction method (Skalar Method 155–000) and NO_3^- was measured using the copper-cadmium reduction method (NH_4Cl buffer

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but without ethylenediamine tetraacetic acid; Skalar Method 461–000). The rest of the field-moist sample was stored in plastic bags for gravimetric moisture determination, conducted in the field station on the same sampling day. We dried 40–100 g of fresh-weight soil for 24 h at 105 °C. We expressed moisture content as water-filled pore space (WFPS) using measured bulk density and particle densities of 2.65 g cm^{-3} for mineral soil (Linn and Doran, 1984) and 1.4 g, cm^{-3} for organic layer (Breuer et al., 2002).

2.5 Statistical analyses

For CH_4 fluxes, statistical analysis was conducted on the plot means (average of 4 chambers) of each sampling day. Linear mixed effects models were used to test for the fixed effects of site (lowland vs. montane control plots) or treatment (control vs. N addition for each site) on the repeated measurements of soil CH_4 fluxes and soil factors (WFPS, soil temperature, NH_4^+ and NO_3^- concentrations). The spatial replication and time (sampling days) were included as random effect. A function which allows different variances of the response variable per level of the fixed effect and/or a first-order temporal autoregressive process was included if this improved the relative goodness of the model fit based on likelihood ratio tests. The significance of the fixed effect was evaluated using analysis of variance (Crawley, 2009). If residual plots revealed non-normal distribution or non-homogenous variance, square-root or logarithmic transformation was used for right-skewed data and quadratic transformation for left-skewed data, and the analysis was repeated. Effects were considered significant if P value ≤ 0.05. Pearson correlation tests were conducted on treatment means (average of 4 plots) of each sampling day to investigate the linear influences of WFPS, soil temperature, NH_4^+ and NO_3^- concentrations on soil CH_4 fluxes. A few CH_4 fluxes from the N-addition plots of the montane forest were exceptionally high (21 out of 196 plot means with emissions > 60 µg CH_4–C m^{-2} h^{-1}), and correlation analyses were conducted both including (using logarithmic transformation) and excluding these high emissions. We also used Pearson correlation to test the influences of annual rainfall, soil clay and sand contents, organic layer thickness, and annual N deposition on annual soil CH_4-C

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fluxes of tropical forests published so far. Mean values in the text are given with ±1 standard error. Analyses were conducted using R 2.15.2 (R Development Core Team, 2011).

3 Results

3.1 Soil water content, temperature and mineral N

In the lowland forest, the pronounced dry season from January to April caused a strong seasonality in WFPS, which ranged from approximately 55–70 % during rainy season to 35–45 % during dry season (Fig. 1a). Mean annual soil temperature was 25.5 °C and the seasonal variation was 2.5 °C (Fig. 1c). Neither WFPS nor soil temperature differed between the control and N-addition plots. In the montane forest, where dry season is absent, high WFPS in the mineral soil (70–80 %) occurred throughout the year. The organic layer with its low bulk density had a much lower WFPS (20–35 %; Fig. 1b). Mean annual soil temperature was 18.1 °C and the seasonal variation was 3.8 °C (Fig. 1d). Also, WFPS and soil temperature were similar between the control and N-addition plots.

In the lowland forest, NH_4^+ concentrations did not differ between the control and N-addition plots (Fig. 2a) but NO_3^- concentrations increased with N addition ($P < 0.01$) (Fig. 2b). In the montane forest, mineral N was dominated by NH_4^+ in both organic layer and mineral soil. N addition increased NH_4^+ concentrations in the mineral soil ($P < 0.01$) but did not show an effect on NH_4^+ concentrations in the organic layer (Fig. 2c, e). NO_3^- concentrations increased in both mineral soil ($P = 0.01$) and organic layer ($P = 0.03$) with very large increases in the fourth year of N addition (Fig. 2d, f).

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3.2 CH_4 fluxes from control forest soils

CH_4 fluxes from the lowland forest control plots ($-21.47 \pm 1.57\,\mu g\,CH_4{-}C\,m^{-2}\,h^{-1}$) did not differ from the fluxes of the montane forest control plots ($-3.99 \pm 3.40\,\mu g\,CH_4{-}C\,m^{-2}\,h^{-1}$; Fig. 3, Table 1). This seemingly larger CH_4 uptake rates in this moist ($2.7\,m\,yr^{-1}$ rainfall) lowland forest soil than the wet ($5.5\,m\,yr^{-1}$ rainfall) montane forest soil was not statistically significant because of the large spatial and temporal variations (Fig. 3). Before going into how the soil factors influence CH_4 fluxes, we want to point out the implications of correlations: a positive correlation between CH_4 fluxes and a soil variable indicates a decrease in CH_4 uptake rates with an increase in the soil parameter values whereas a negative correlation indicates an increase in CH_4 uptake rates with an increase in the soil parameter values. In the lowland forest, CH_4 fluxes were positively correlated with WFPS (Table 2). In the montane forest, CH_4 fluxes were negatively correlated with NH_4^+ concentrations and positively correlated with NO_3^- concentrations of the organic layer and mineral soil (Table 2). These opposing correlations of CH_4 fluxes with NH_4^+ and NO_3^- were because the temporal patterns of NH_4^+ and NO_3^- showed the opposite trend. The correlation between CH_4 fluxes and total soil mineral N ($NH_4^+ + NO_3^-$) concentrations (organic layer $R = -0.51$, $P = 0.01$, $n = 28$; mineral soil $R = -0.56$, $P = 0.00$, $n = 27$) followed that of NH_4^+ because NH_4^+ comprised the largest part of mineral N.

3.3 Effects of N addition on soil CH_4 fluxes

In the lowland forest, neither all CH_4 fluxes ($-24.22 \pm 1.64\,\mu g\,C\,m^{-2}\,h^{-1}$) nor the long-term CH_4 fluxes ($-26.14 \pm 2.00\,\mu g\,C\,m^{-2}\,h^{-1}$) from the N-addition plots differed from the CH_4 fluxes of the control plots (Fig. 3a, c; Table 1). The reason was the occasional CH_4 emissions from three of the four replicate plots of the control and N-addition treatment regardless of seasons (46 emission fluxes out of 373 plot-mean fluxes or 12 % of the observations, ranging from 0.4 to $210\,\mu g\,C\,m^{-2}\,h^{-1}$), resulting in the large spatial

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and temporal variations (i.e. large SE bars; Fig. 3a, c). For all CH_4 fluxes, we detected a positive correlation with WFPS and negative correlations with soil temperatures and NO_3^- concentrations (Table 2). The same soil factors showed similar trends of correlations with the long-term CH_4 fluxes (Table 2).

In the montane forest, despite the large mean CH_4 emissions from the N-addition plots (for all CH_4 fluxes $50.94 \pm 19.62\,\mu g\,C\,m^{-2}\,h^{-1}$; for long-term CH_4 fluxes $62.13 \pm 31.26\,\mu g\,C\,m^{-2}\,h^{-1}$), neither all CH_4 fluxes nor the long-term CH_4 fluxes differed from those of the control plots (Fig. 3b, d; Table 1). The reason was that frequent CH_4 emissions were observed from all eight plots (83 emission fluxes out of 351 plot-mean fluxes or 24 % of the observations, ranging from 0.2 to $2575\,\mu g\,C\,m^{-2}\,h^{-1}$). These CH_4 emissions were dominated by one pair of control and N-addition plots (49 emission fluxes out of 351 plot-mean fluxes), causing the large spatial and temporal variations (i.e. large SE bars; Fig. 3b, d). If we exclude this one pair of control and N-addition plots from the statistical analysis, there remained no difference between the N-addition and control plots, but the mean CH_4 fluxes showed net uptake instead of net emission (Table 1). Also, a few CH_4 emissions from N-addition plots were exceptionally high (21 out of 196 plot means with emissions $> 60\,\mu g\,CH_4\text{–}C\,m^{-2}\,h^{-1}$). Thus, we looked critically on how these few high CH_4 emissions influence the relationships between CH_4 fluxes and soil factors. We first analyzed the correlations between CH_4 fluxes and soil factors that include all emission fluxes and that exclude the exceptionally high CH_4 emissions of $> 60\,\mu g\,CH_4\text{–}C\,m^{-2}\,h^{-1}$. Considering all CH_4 fluxes, we observed a positive correlation with WFPS of the mineral soil and a negative correlation with NH_4^+ concentrations of the organic layer when the large emissions were included. When the large emissions were excluded, CH_4 fluxes remained negatively correlated with NH_4^+ concentrations of the organic layer (Table 2). Considering only the long-term CH_4 fluxes, we observed also a negative correlation with NH_4^+ concentrations of the organic layer both including and excluding the large emissions (Table 2).

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4 Discussion

4.1 CH_4 fluxes from control forests and comparison with published values

The mean annual CH_4 uptake rate in the control plots of the lowland forest was within the range of published values from (sub)tropical forests below 800 m elevation (Table 3). The few published CH_4 uptake rates that were lower than in our lowland forest soil were mainly from Amazon forest soils with low sand or high clay contents, and those with larger CH_4 uptake rates were mostly in sites with low clay content (Sousa Neto et al., 2011; Steudler et al., 1996). Indeed, from studies compiled in Table 3 the only significant correlation between annual CH_4 fluxes and site factors for the tropical forests below 800 m elevation was a positive correlation between annual soil CH_4 fluxes and clay contents ($R = 0.58$, $P = 0.02$, $n = 16$). A high content of clay decreases the contribution of coarse pores to the total porosity (Hillel, 1998). As coarse pores are especially important for gas diffusive transport, soil texture may be a good proxy variable for gas diffusion control on CH_4 uptake. Consistent with this correlation pattern, earlier studies have shown that CH_4 uptake is often limited by gas diffusion in the soil (Keller and Reiners, 1994). Also, the seasonal changes in CH_4 uptake of our lowland forest soil (Fig. 3a, c) were best explained by gas diffusion as was illustrated by the correlation of CH_4 fluxes with WFPS (Table 2); during the wet season when WFPS was high, CH_4 uptake was low because CH_4 diffusion from the atmosphere to this site's clayey soils was probably slowed down by the high soil water contents.

The mean annual CH_4 uptake rate in the control plots of the montane forest was the lowest published so far for tropical forests above 800 m elevation (Table 3). This was caused by the frequent CH_4 emissions from our wet, montane forest soil (Fig. 3 b, d). From tropical forests above 800 m elevation (Table 3), we detected a positive correlation between annual CH_4 fluxes and rainfall ($R = 0.78$, $P = 0.04$, $n = 7$), which is in line with the gas diffusion control on soil CH_4 uptake as discussed above. Rainfall influences gas diffusion through its effects on soil moisture content. However, in contrast to the forests below 800 m elevation, we detected a negative correlation with clay contents

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($R = -0.68$, $P = 0.04$, $n = 9$). This can probably be explained by the occurrence of thick organic layers (Table 3) at the surface of some of these soils, which may interfere with gas exchange between soil and atmosphere. From an earlier study we conducted in montane forests of Ecuador, we found that contrary to common belief the deeper part of such organic layers may contribute to the CH_4-oxidation capacity of soils (Wolf et al., 2012). The thickness, bulk density and CH_4-oxidation capacity of these organic layers may influence CH_4 uptake stronger than the soil texture of the underlying mineral soil. We also detected a positive correlation between annual CH_4 fluxes and annual N deposition rates ($R = 0.96$, $P < 0.00$, $n = 6$) of tropical forests above 800 m elevation. This may suggest that CH_4 uptake is lower at sites with higher N deposition. However, this correlation is based on six sites that had N deposition rates of only $\leq 5.0\,\mathrm{kg\,N\,ha^{-1}\,yr^{-1}}$. At such low rates of N deposition, we think that inhibition of CH_4 oxidation by NH_4^+ is unlikely. Instead, we think that such correlation is only circumstantial because in these six sites annual N deposition was positively correlated with annual rainfall ($R = 0.89$, $P = 0.02$, $n = 6$), signifying that low CH_4 uptake was reported for sites with high rainfall and high N deposition. Thus, we think it is more likely that soil water content (which controls gas diffusion) as influenced by rainfall was the reason behind the observed correlation between annual CH_4 fluxes and annual N deposition rates.

For the control plots of the montane forest, we interpret the negative correlations of CH_4 fluxes with NH_4^+ and total mineral N concentrations as evidence that CH_4 consumption was N limited. We had similar findings of negative correlation between CH_4 fluxes and total mineral N concentrations in montane forest soils in Ecuador, suggesting N limitation on methanotrophic activity (Wolf et al., 2012). Although Bodelier and Laanbroek (2004) suggest that N limitation of methanotrophic bacteria is less likely at (sub)atmospheric CH_4 concentrations in the soil, we had ancillary measurements of the soil-air CH_4 concentrations in our montane forest soil that showed CH_4 concentrations in this forest soil were occasionally high. These measurements were conducted monthly from October 2008 to January 2010 in three control plots and three N-addition plots for various layers: 0.10 m above the soil surface, at the interface of the

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organic layer and mineral soil, at 0.05, 0.20, 0.40, 0.75 and 1.25 m depths in the mineral soil; we employed the same gas sampling methods described in our earlier works (Koehler et al., 2012). We found that 34 % of 421 observations had CH_4 concentrations in the mineral soil higher than the concentration at 0.10 m above the soil surface of 2.0 ± 0.1 ppm CH_4-C, particularly during periods of high rainfall and thus high soil water contents. Such high soil-air CH_4 concentrations in our montane forest may allow for population increases of methanotrophic bacteria which, in turn, may lead to N limitation on their activity (Bodelier and Laanbroek, 2004).

4.2 Response of soil CH_4 fluxes to N addition in the lowland and montane forests

In contrast to the findings from temperate forest soils (Steudler et al., 1989; Brumme and Borken, 1999), tropical pasture soil (Veldkamp et al., 2001) and subtropical forest soil (Zhang et al., 2008), CH_4 uptake in our lowland forest soil was not inhibited by chronic N addition. Instead, the negative correlation of CH_4 fluxes with NO_3^- concentrations in the N-addition plots suggests that increased NO_3^- levels in these plots had stimulated CH_4 consumption (Bodelier and Laanbroek, 2004) and/or had inhibited CH_4 production (Conrad, 1989). The latter is however unlikely because our ancillary measurements of CH_4 concentrations at various depths of the mineral soil (0.05, 0.20, 0.40, 0.75, 1.25 and 2 m depth) in this lowland forest during the same study years (May 2006–January 2009) showed that 11 % of the observations had higher soil-air CH_4 concentrations than the average soil-air CH_4 concentrations at a specific depth. These high soil-air CH_4 concentrations occurred in any depths of both N-addition and control plots regardless of seasons, indicating that inhibition by high NO_3^- levels in N addition plots on CH_4 production was unlikely (Koehler et al., 2012). Instead, there were other supporting indications that methanotrophic activity was N limited aside from the negative correlation of soil CH_4 fluxes with NO_3^- concentrations: soil-air CH_4 concentrations and contents (or the total amount of CH_4 in a soil-air volume) down to 0.4 m depth were 30 % lower in N-addition than in control plots, and the minimum CH_4 concentration of

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552 ± 42 ppb was reached at shallower depth (already at 0.40 m) in N-addition than in control plots (only at 1.25 m depth) (Koehler et al., 2012). It should be noted that these patterns were not influenced by WFPS because there were no differences in WFPS between control and N-addition plots at all depths. The reason why we did not detect significant differences in soil CH_4 fluxes despite stimulated CH_4 uptake by chronic N addition is first due to the large spatial and temporal variations of CH_4 fluxes (Fig. 3a, c). Similar large variability was reported for tropical lowland forest soils and was attributed to production of CH_4 by termites or in microsites of anaerobic conditions, and to temporal patterns of rainfall and soil moisture contents (Verchot et al., 2000; Davidson et al., 2004; Koehler et al., 2012). Second, CH_4 consumption was also largely limited by gas diffusion as shown by the positive correlation of CH_4 fluxes with WFPS (Table 2). Even if N addition stimulated methanotrophic activity, the supply of CH_4 as substrate from the atmosphere to the soil through diffusion did not change, and thus chronic N addition did not necessarily result in a larger CH_4 uptake rate. Stimulation of methanotrophic activity may be explained by a shift in N nutrition of type II methanotrophic bacteria from energy-demanding N_2 fixation to assimilation of soil mineral N (Koehler et al., 2012; Bodelier and Laanbroek, 2004) of which the NO_3^- concentrations had increased under chronic N addition (Fig. 2b).

In the montane forest soil, there was also an indication that methanotrophic activity was stimulated by chronic N addition as shown by the negative correlations between CH_4 fluxes and NH_4^+ concentrations of the organic layer. However, this N-stimulated methanotrophic activity was masked by the frequent CH_4 emissions. The frequent CH_4 emissions in this wet montane forest soil indicated the regulation of WFPS of the mineral soil on CH_4 fluxes, as was shown by their positive correlation when all CH_4 fluxes are included in the statistical analysis (Table 2). The regulation by WFPS suggests not only through diffusive limitation of CH_4 as substrate for methanotrophs but also through occurrence of anaerobic condition for CH_4 production. Indeed, the WFPS of this montane forest was high throughout the year (Fig. 1b) and our ancillary measurements of WFPS at various depths in the mineral soil of these plots, conducted monthly during

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October 2008 to January 2010, showed WFPS between 96 ± 1 % and 88 ± 1 % from 0.20 m down to 1.25 m depth. Such high WFPS may have favoured CH_4 production and thus the frequent CH_4 emissions from all eight plots. This was probably the principal reason why we were not able to detect potential differences in CH_4 uptake rates between control and N-addition plots despite an indication of N limitation on CH_4 consumption. Exclusion of one pair of control and N-addition plots that strongly dominated the CH_4 emissions during our four-year measurements did not change the statistical trend even though the mean CH_4 uptake rates in the N-addition plots were seemingly larger than the control plots in all years (Table 1).

4.3 Consequences of chronic N deposition on soil CH_4 fluxes from tropical forests

Nine to twelve years of N addition to a lowland forest and one to four years of N addition to a montane forest did not affect soil CH_4 fluxes, although we found indications that CH_4 consumption may have been N limited at both sites. We proposed the following reasons why such N-stimulated CH_4 consumption did not lead to statistically larger CH_4 uptake: (1) for the moist, lowland forest soil, this was caused by limitation of CH_4 diffusion from the atmosphere into the clayey soils particularly during the wet season when WFPS was high; (2) for the wet, montane forest soil, this was due to the high WFPS in the mineral soil throughout the year, which may not only limit CH_4 diffusion from the atmosphere into the soil but also favours CH_4 production; and (3) both forest soils showed large spatial and temporal variations of CH_4 fluxes. The lowland forest soil showed occasional but low CH_4 emissions whereas the montane forest soil showed more frequent CH_4 emissions with few exceptional large emissions (Fig. 3); such high CH_4 concentrations in the soil provide high amounts of substrate for methanotrophy and favour N limitation on methanotrophic bacteria (Bodelier and Laanbroek, 2004).

Our results contrast with the only published study about N-addition effects on soil CH_4 fluxes from (sub)tropical forests, which was conducted in China, where increasing N addition rates resulted in decreasing CH_4 uptake rates. These results were attributed

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to several possible causes (high N status, low pH values and Al toxicity) (Zhang et al., 2008, 2011). Although our lowland forest soil also had a high N status (Corre et al., 2010) and our montane forest soil also had low pH and high exchangeable Al (see Sect. 2.2), the differences in site conditions between our sites and this forest in China are that this China site had suffered decades of high N deposition (Table 3) leading to soil pH values below 4.0, exchangeable Al of > 400 mg Al kg^{-1} even in the control plots, and never emitted CH_4 during the 1 yr measurements. Sub-atmospheric CH_4 concentrations are possibly prevalent in this China site and in such conditions methanotropic activity is less likely to be N limited (Bodelier and Laanbroek, 2004).

If our explanation for the contrasting effects of N additions between our study sites and that of Zhang et al. (2008) holds up throughout the tropics, it is unlikely that elevated N deposition on tropical forests will lead to widespread inhibition of CH_4 uptake. We expect that in tropical montane forests, which typically have low N availability, N deposition may stimulate CH_4 uptake in sites where occasional CH_4 emissions occur or will cause no change in CH_4 uptake in sites where no CH_4 emissions occur. In tropical lowland forests, which often have a high N availability, N deposition only appears to inhibit CH_4 uptake if soil pH values have become so low that considerable Al toxicity occurs. In other situations, it seems more likely that N deposition will not affect CH_4 fluxes or may even stimulate CH_4 uptake. Whether N additions to tropical forests with N-limited methanotrophic activity can indeed stimulate soil CH_4 uptake remains to be seen. The most likely time when CH_4 uptake may be stimulated by N additions is during dry periods/seasons when CH_4 supply from the atmosphere is not or less limited by gas diffusion. The most likely place where CH_4 uptake may be stimulated by N addition is in forests with a strong seasonal rainfall where occasional CH_4 emissions occur during the rainy season and strong uptake occurs during the dry season.

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References

Adamek, M., Corre, M. D., and Hoelscher, D.: Early effect of elevated nitrogen input on aboveground net primary production of a lower montane rain forest, Panama, J. Trop. Ecol., 25, 637–647, doi:10.1017/S0266467409990253, 2009.

Adamek, M., Corre, M. D., and Hoelscher, D.: Responses of fine roots to experimental nitrogen addition in a tropical lower montane rain forest, Panama, J. Trop. Ecol., 27, 73–81, doi:10.1017/S0266467410000507, 2011.

Arnold, J., Corre, M. D., and Veldkamp, E.: Cold storage and laboratory incubation of intact soil cores do not reflect in-situ nitrogen cycling rates of tropical forest soils, Soil Biol. Biochem., 40, 2480–2483, doi:10.1016/j.soilbio.2008.06.001, 2008.

Bender, M. and Conrad, R.: Effect of CH_4 concentrations and soil conditions on the induction of CH_4 oxidation activity, Soil Biol. Biochem., 27, 1517–1527, doi:10.1016/0038-0717(95)00104-M, 1995.

Bodelier, P. L. E., and Laanbroek, H. J.: Nitrogen as a regulatory factor of methane oxidation in soils and sediments, FEMS Microbiol. Ecol., 47, 265–277, doi:10.1016/s0168-6496(03)00304-0, 2004.

Breuer, L., Kiese, R., and Butterbach-Bahl, K.: Temperature and moisture effects on nitrification rates in tropical rain-forest soils, Soil Sci. Soc. Am. J., 66, 834–844, 2002.

Brumme, R. and Borken, W.: Site variation in methane oxidation as affected by atmospheric deposition and type of temperate forest ecosystem, Global Biogeochem. Cy., 13, 493–501, doi:10.1029/1998GB900017, 1999.

Conrad, R.: Control of methane production in terrestrial ecosystems, in: Exchange of Trace Gases between Terrestrial Ecosystems and the Atmosphere, edited by: Andreae, M. O. and Schimel, D. S., Wiley, Chichester, UK, 39–58, 1989.

Conrad, R.: Soil microorganisms as controllers of atmospheric trace gases (H_2, CO, CH_4, OCS, N_2O, and NO), Microbiol. Rev., 60, 609–640, 1996.

Conrad, R.: Microbial ecology of methanogens and methanotrophs, Adv. Agr., 96, 1–63, doi:10.1016/S0065-2113(07)96005-8, 2007.

Corre, M. D., Veldkamp, E., Arnold, J., and Wright, S. J.: Impact of elevated N input on soil N cycling and losses in old-growth lowland and montane forests in Panama, Ecology, 91, 1715–1729, doi:10.1890/09-0274.1, 2010.

Crawley, M. J.: The R Book, John Wiley & Sons Ltd, Chichester, UK, 2009.

Crill, P. M., Martikainen, P. J., Nykanen, H., and Silvola, J.: Temperature and N fertilization effects on methane oxidation in a drained peatland soil, Soil Biol. Biochem., 26, 1331–1339, doi:10.1016/0038-0717(94)90214-3, 1994.

Davidson, E. A., Ishida, F. Y., and Nepstad, D. C.: Effects of an experimental drought on soil emissions of carbon dioxide, methane, nitrous oxide, and nitric oxide in a moist tropical forest, Glob. Change Biol., 10, 718–730, doi:10.1111/j.1365-2486.2004.00762.x, 2004.

Denman, K. L., Brasseur, G., Chidthaisong, A., Ciais, P., Cox, P. M., Dickinson, R. E., Hauglustaine, D., Heinze, C., Holland, E., Jacob, D., Lohmann, U., Ramachandran, S., Dias, P. L. D. S., Wofsy, S. C., and Zhang, X.: Couplings between changes in the climate system and biogeochemistry, in: Climate Change 2007: The Physical Science Basis. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change, edited by: Solomon, S., Qin, D., Manning, M., Chen, Z., Marquis, M., Averyt, K. B., Tignor, M., and Miller, H. L., Cambridge University Press, Cambridge, UK and New York, NY, USA, 2007.

Dutaur, L. and Verchot, L. V.: A global inventory of the soil CH_4 sink, Global Biogeochem. Cy., 21, GB4013, doi:10.1029/2006gb002734, 2007.

Fang, H. J., Yu, G. R., Cheng, S. L., Zhu, T. H., Wang, Y. S., Yan, J. H., Wang, M., Cao, M., and Zhou, M.: Effects of multiple environmental factors on CO_2 emission and CH_4 uptake from old-growth forest soils, Biogeosciences, 7, 395–407, doi:10.5194/bg-7-395-2010, 2010.

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10, 6007–6037, 2013

Frankenberg, C., Bergamaschi, P., Butz, A., Houweling, S., Meirink, J. F., Notholt, J., Petersen, A. K., Schrijver, H., Warneke, T., and Aben, I.: Tropical methane emissions: a revised view from SCIAMACHY onboard ENVISAT, Geophys. Res. Lett., 35, L15811, doi:10.1029/2008gl034300, 2008.

Galloway, J. N., Townsend, A. R., Erisman, J. W., Bekunda, M., Cai, Z., Freney, J. R., Martinelli, L. A., Seitzinger, S. P., and Sutton, M. A.: Transformation of the nitrogen cycle: recent trends, questions, and potential solutions, Science, 320, 889–892, doi:10.1126/science.1136674, 2008.

Hietz, P., Turner, B. L., Wanek, W., Richter, A., Nock, C. A., and Wright, S. J.: Long-term change in the nitrogen cycle of tropical forests, Science, 334, 664–666, doi:10.1126/science.1211979, 2011.

Hillel, D.: Environmental Soil Physics, Academic Press, San Diego, California, USA, 771 pp., 1998.

Keller, M. and Matson, P. A.: Biosphere–atmosphere exchange of trace gases in the tropics: evaluating the effects of land use changes, in: Global Atmospheric-Biospheric Chemistry, edited by: Prinn, R. G., Plenum Press, New York, 103–117, 1994.

Keller, M. and Reiners, W. A.: Soil-atmosphere exchange of nitrous oxide, nitric oxide and methane under secondary succession of pasture to forest in the Atlantic lowlands of Costa Rica, Global Biogeochem. Cy., 8, 399–409, doi:10.1029/94GB01660, 1994.

Keller, M., Varner, R., Dias, J. D., Silva, H., Crill, P., and de Oliveira, R. C.: Soil-atmosphere exchange of nitrous oxide, nitric oxide, methane, and carbon dioxide in logged and undisturbed forest in the Tapajos National Forest, Brazil, Earth Interact., 9, 1–28, doi:10.1175/EI125.1, 2005.

Kiese, R., Hewett, B., Graham, A., and Butterbach-Bahl, K.: Seasonal variability of N_2O and CH_4 uptake by tropical rainforest soils of Queensland, Australia, Global Biogeochem. Cy., 17, 1043, doi:10.1029/2002GB002014, 2003.

Kiese, R., Wochele, S., and Butterbach-Bahl, K.: Site specific and regional estimates of methane uptake by tropical rainforest soils in north eastern Australia, Plant Soil, 309, 211–226, doi:10.1007/s11104-008-9545-0, 2008.

Koehler, B., Corre, M. D., Veldkamp, E., Wullaert, H., and Wright, S. J.: Immediate and long-term nitrogen oxide emissions from tropical forest soils exposed to elevated nitrogen input, Glob. Change Biol., 15, 2049–2066, doi:10.1111/j.1365-2486.2008.01826.x, 2009.

BGD
10, 6007–6037, 2013

Koehler, B., Corre, M. D., Steger, K., Well, R., Zehe, E., Sueta, J. P., and Veldkamp, E.: An in-depth look into a tropical lowland forest soil: nitrogen-addition effects on the contents of N_2O, CO_2 and CH_4 and N_2O isotopic signatures down to 2 m depth, Biogeochemistry, 111, 695–713, doi:10.1007/s10533-012-9711-6, 2012.

Linn, D., and Doran, J.: Effect of water-filled pore space on carbon dioxide and nitrous oxide production in tilled and nontilled soils, Soil Sci. Soc. Am. J., 48, 1267–1272, 1984.

Livingston, G. P., Hutchinson, G. L., and Spartalian, K.: Trace gas emission in chambers, Soil Sci. Soc. Am. J., 70, 1459–1469, doi:10.2136/sssaj2005.0322, 2006.

Loftfield, N., Flessa, H., Augustin, J., and Beese, F.: Automated gas chromatographic system for rapid analysis of the atmospheric trace gases methane, carbon dioxide, and nitrous oxide, J. Environ. Qual., 26, 560–564, 1997.

Martinson, G. O., Werner, F. A., Scherber, C., Conrad, R., Corre, M. D., Flessa, H., Wolf, K., Klose, M., Gradstein, S. R., and Veldkamp, E.: Methane emissions from tank bromeliads in neotropical forests, Nat. Geosci., 3, 766–769, doi:10.1038/Ngeo980, 2010.

Mosier, A. R., Parton, W. J., Valentine, D. W., Ojima, D. S., Schimel, D. S., and Delgado, J. A.: CH_4 and N_2O fluxes in the Colorado shortgrass steppe: 1. Impact of landscape and nitrogen addition, Global Biogeochem. Cy., 10, 387–399, doi:10.1029/96GB01454, 1996.

Nesbit, S. P. and Breitenbeck, G. A.: A laboratory study of factors influencing methane uptake by soils, Agr. Ecosys. Environ., 41, 39–54, doi:10.1016/0167-8809(92)90178-E, 1992.

Purbopuspito, J., Veldkamp, E., Brumme, R., and Murdiyarso, D.: Trace gas fluxes and nitrogen cycling along an elevation sequence of tropical montane forests in Central Sulawesi, Indonesia, Global Biogeochem. Cy., 20, GB3010, doi:10.1029/2005GB002516, 2006.

R Development Core Team: R: A Language and Environment for Statistical Computing, R Foundation for Statistical Computing, Vienna, Austria, 2011.

Sousa Neto, E., Carmo, J. B., Keller, M., Martins, S. C., Alves, L. F., Vieira, S. A., Piccolo, M. C., Camargo, P., Couto, H. T. Z., Joly, C. A., and Martinelli, L. A.: Soil-atmosphere exchange of nitrous oxide, methane and carbon dioxide in a gradient of elevation in the coastal Brazilian Atlantic forest, Biogeosciences, 8, 733–742, doi:10.5194/bg-8-733-2011, 2011.

Steudler, P. A., Bowden, R. D., Melillo, J. M., and Aber, J. D.: Influence of nitrogen fertilization on methane uptake in temperate forest soils, Nature, 341, 314–316, doi:10.1038/341314a0, 1989.

BGD
10, 6007–6037, 2013

Steudler, P. A., Melillo, J. M., Feigl, B. J., Neill, C., Piccolo, M. C., and Cerri., C. C.: Consequences of forest-to-pasture conversion on CH_4 fluxes in the Brazilian Amazon Basin, J. Geophys. Res.-Atmos., 101, 18547–18554, doi:10.1029/96JD01551, 1996.

Veldkamp, E., Weitz, A. M., and Keller, M.: Management effects on methane fluxes in humid tropical pasture soils, Soil Biol. Biochem., 33, 1493–1499, doi:10.1016/S0038-0717(01)00060-8, 2001.

Verchot, L. V., Davidson, E. A., Cattanio, J. H., and Ackerman, I. L.: Land-use change and biogeochemical controls of methane fluxes in soils of eastern Amazonia, Ecosystems, 3, 41–56, doi:10.1007/s100210000009, 2000.

Vitousek, P. M., Aber, J. D., Howarth, R. W., Likens, G. E., Matson, P. A., Schindler, D. W., Schlesinger, W. H., and Tilman, D. G.: Human alteration of the global nitrogen cycle – sources and consequences [review], Ecol. Appl., 7, 737–750, doi:10.1890/1051-0761(1997)007[0737:HAOTGN]2.0.CO;2, 1997.

Wagner, S. W., Reicosky, D. C., and Alessi, R. S.: Regression models for calculating gas fluxes measured with a closed chamber, Agronom. J., 89, 279–284, 1997.

Werner, C., Zheng, X., Tang, J., Xie, B., Liu, C., Kiese, R., and Butterbach-Bahl, K.: N_2O, CH_4 and CO_2 emissions from seasonal tropical rainforests and a rubber plantation in Southwest China, Plant Soil, 289, 335–353, doi:10.1007/s11104-006-9143-y, 2006.

Werner, C., Kiese, R., and Butterbach-Bahl, K.: Soil-atmosphere exchange of N_2O, CH_4, and CO_2 and controlling environmental factors for tropical rain forest sites in western Kenya, J. Geophys. Res., 112, D03308, doi:10.1029/2006jd007388, 2007.

Wolf, K., Flessa, H., and Veldkamp, E.: Atmospheric methane uptake by tropical montane forest soils and the contribution of organic layers, Biogeochemistry, 111, 469–483, doi:10.1007/s10533-011-9681-0, 2012.

Wright, S. J., Yavitt, J. B., Wurzburger, N., Turner, B. L., Tanner, E. V. J., Sayer, E. J., Santiago, L. S., Kaspari, M., Hedin, L. O., Harms, K. E., Garcia, M. N., and Corre, M. D.: Potassium, phosphorus or nitrogen limit root allocation, tree growth and litter production in a lowland tropical forest., Ecology, 92, 1616–1625, doi:10.1890/10-1558.1, 2011.

Zhang, T., Zhu, W., Mo, J., Liu, L., and Dong, S.: Increased phosphorus availability mitigates the inhibition of nitrogen deposition on CH_4 uptake in an old-growth tropical forest, southern China, Biogeosciences, 8, 2805-2813, doi:10.5194/bg-8-2805-2011, 2011.

BGD
10, 6007–6037, 2013

Zhang, W., Mo, J., Zhou, G., Gundersen, P., Fang, Y., Lu, X., Zhang, T., and Dong, S.: Methane uptake responses to nitrogen deposition in three tropical forests in southern China, J. Geophys. Res., 113, D11116, doi:10.1029/2007jd009195, 2008.

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Table 1. Annual soil CH_4-C fluxes ($kg\,C\,ha^{-1}\,yr^{-1}$, mean ±SE, $n = 4$) from the control and N-addition plots, separated into all and long-term fluxes, with the latter including only the fluxes measured at least six weeks after a N application. For the montane forest, values in brackets are estimates that excluded one pair of plots (control and N addition) which dominated CH_4 emissions (49 emission fluxes out of 351 plot-mean fluxes).

Site	Treatment	2006	2007	2008	2009
Montane	Control	−1.69 ± 0.36 (−1.83 ± 0.48)	−1.18 ± 0.41 (−1.15 ± 0.58)	−0.53 ± 0.50 (−0.93 ± 0.42)	1.91 ± 2.49 (−0.56 ± 0.57)
	1–4 yr N addition, all fluxes	−1.86 ± 0.57 * (−2.37 ± 0.35)	7.64 ± 9.40 (−1.75 ± 0.30)	4.42 ± 5.86 (−1.42 ± 0.59)	8.99 ± 10.41 (−1.42 ± 0.38)
	1–4 yr N addition, long-term fluxes	−2.19 ± 0.76 * (−2.91 ± 0.37)	8.34 ± 9.97 (−1.63 ± 0.30)	5.36 ± 6.82 (−1.44 ± 0.63)	8.56 ± 9.89 (−1.33 ± 0.43)
Lowland	Control	−1.93 ± 0.24	−1.82 ± 0.51	−2.38 ± 0.54	−1.60 ± 0.45
	9–12 yr N addition, all fluxes	−2.33 ± 0.85	−2.22 ± 0.60	−1.94 ± 0.98	−2.20 ± 0.50
	9–12 yr N addition, long-term fluxes	−2.09 ± 0.92	−2.42 ± 0.68	−2.15 ± 0.51	−2.16 ± 0.51

* The two pre-treatment measurements from January and February 2006 were not included in the calculation.

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Table 2. Pearson correlation coefficients between soil CH_4-C fluxes ($\mu g C m^{-2} h^{-1}$) and soil variables, using the mean values of each treatment on each sampling day, measured from 2006 to 2009. For the montane forest N-addition plots, coefficients in brackets are from analyses that include the few events of large CH_4 emissions (please see Sect. 2.5).

Site and Treatment	$n \geq$	Water-filled pore space (%)	NH_4^+ ($mg N kg^{-1}$)	NO_3^- ($mg N kg^{-1}$)	Soil temperature (°C)
Montane	Organic layer				
Control	28	−0.31	$-0.58^{b,c}$	$0.62^{b,c}$	–
1–4 yr N addition, including all fluxes	27	−0.05 (−0.18)	$-0.43^{a,c}$ ($-0.38^{a,c}$)	-0.12^{c} (0.34^{c})	–
1–4 yr N addition, long-term fluxes (i.e. measured ≥6 weeks after N addition)	24	−0.13 (0.06)	$-0.48^{a,c}$ ($-0.44^{a,c}$)	0.11^{c} $(0.29)^{c}$	–
Montane	0–0.05 m mineral soil				
Control	27	0.26	-0.56^{b}	$0.54^{b,c}$	−0.17
1–4 yr N addition, including all fluxes	26	0.14 (0.37^{b})	−0.25 (−0.31)	-0.35^{c} (0.16^{c})	−0.16 (0.09)
1–4 yr N addition, long-term fluxes	23	−0.09 (0.30)	−0.33 (−0.36)	-0.01^{c} $(0.08)^{c}$	0.02 (0.14)
Lowland	0–0.05 m mineral soil				
Control	32	0.57^{b}	-0.03^{c}	-0.14^{c}	−0.02
9–12 yr N addition, including all fluxes	33	0.49^{b}	0.16^{c}	$-0.40^{a,c}$	-0.34^{a}
9–12 yr N addition, long-term fluxes	28	0.54^{b}	0.27^{c}	$-0.37^{a,c}$	-0.39^{a}

[a,b] $P \leq 0.05$, and $P \leq 0.01$, respectively.
[c] Data were logarithmically transformed before analysis (please see Sect. 2.5).

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Table 3. Compilation of CH_4-C fluxes ($kg\,CH_4-C\,ha^{-1}\,yr^{-1}$) from soils of old-growth (sub)tropical forests, sorted from smallest to largest uptake rates within each elevation category.

Country	Elevation (m)	Annual CH_4-C flux	Annual rainfall (mm)	Clay content (%)	Sand content (%)	Organic layer thickness* (cm)	N deposition ($kg\,N\,ha^{-1}\,yr^{-1}$)	Reference
Sites < 800 m elevation								
Brazil	120	−0.55	2000	80	18	0	not reported	Keller et al. (2005)
Brazil	120	−0.83	2000	75	20	0	not reported	Davidson et al. (2004)
China	720	−0.93	1557	54	17	0	18.0	Fang et al. (2010)
Brazil	100	−1.57	1850	80*	15*	0	not reported	Verchot et al. (2000)
Panama	43	−1.93	2715	69	7	0	9.0	Present study
China	300	−1.93	1564	22	20	0	38.0	Fang et al. (2010)
Australia	50	−2.35	4395	30*	60*	0	not reported	Kiese et al. (2008)
Australia	800	−2.41	1594	30*	60*	0	not reported	Kiese et al. (2008)
China	770	−2.58	1493	18	59	0	18.0	Werner et al. (2006)
Brazil	120	−2.60	2000	38	60	0	not reported	Keller et al. (2005)
Brazil	100	−2.74	3050	32	60	0	8.0	Sousa Neto et al. (2011)
Australia	50	−2.94	3609	60*	20*	0	not reported	Kiese et al. (2008)
Costa Rica	60	−3.45	4200	76	20	0	9.6	Keller and Reiners (1994)
Brazil	124	−3.50	2200	20*	75*	0	not reported	Steudler et al. (1996)
China	300	−3.60	1927	29	38	0	36.0	Zhang et al. (2008)
Brazil	400	−4.90	3050	16	67	0	8.0	Sousa Neto et al. (2011)
Sites > 800 m elevation								
Panama	1200	−0.37	5461	13	61	10	5.0	Present study
Ecuador	3000	−1.06	4500	17	30	14	4.4	Wolf et al. (2012)
Indonesia	2470	−1.45	not measured	17	59	15	not measured	Purbopuspito et al. (2006)
Indonesia	1190	−2.45	1590	12	64	0	2.6	Purbopuspito et al. (2006)
Ecuador	2000	−3.10	1950	18	25	13	2.9	Wolf et al. (2012)
Indonesia	1800	−3.32	not measured	32	51	20	not measured	Purbopuspito et al. (2006)
Brazil	1000	−4.40	2300	20	57	0	2.1	Sousa Neto et al. (2011)
Kenya	1600	−4.94	1662	34	43	0	not reported	Werner et al. (2007)
Ecuador	1000	−5.60	2230	25	41	4	1.5	Wolf et al. (2012)

* Percentages of clay and sand were estimated from the reported soil texture class. If no organic layer was mentioned, we assumed that it was absent (i.e. thickness of 0 cm).

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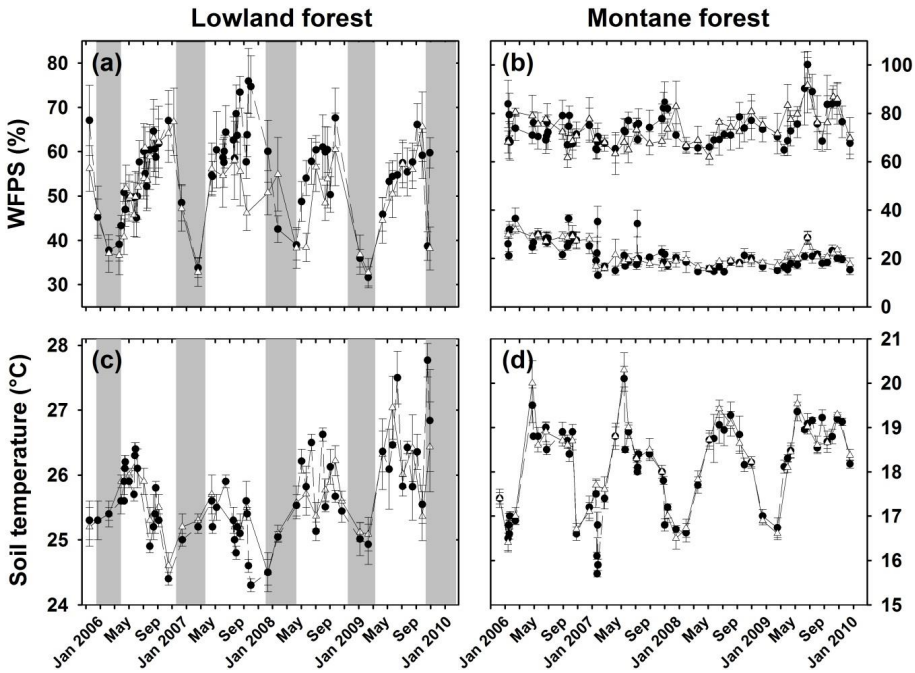

Fig. 1. Mean (±SE, $n = 4$) soil water-filled pore space (WFPS) and temperature at 0–0.05 m mineral soil in the control (Δ) and N-addition (•) plots of the lowland forest (**a** and **c**) with 9–12 yr of treatment and of the montane forest (**b** and **d**) with 1–4 yr of treatment. For WFPS in the montane forest, the upper and lower values are for the 0–0.05 m mineral soil and organic layer, respectively. Grey shadings in **(a)** and **(c)** mark the dry seasons.

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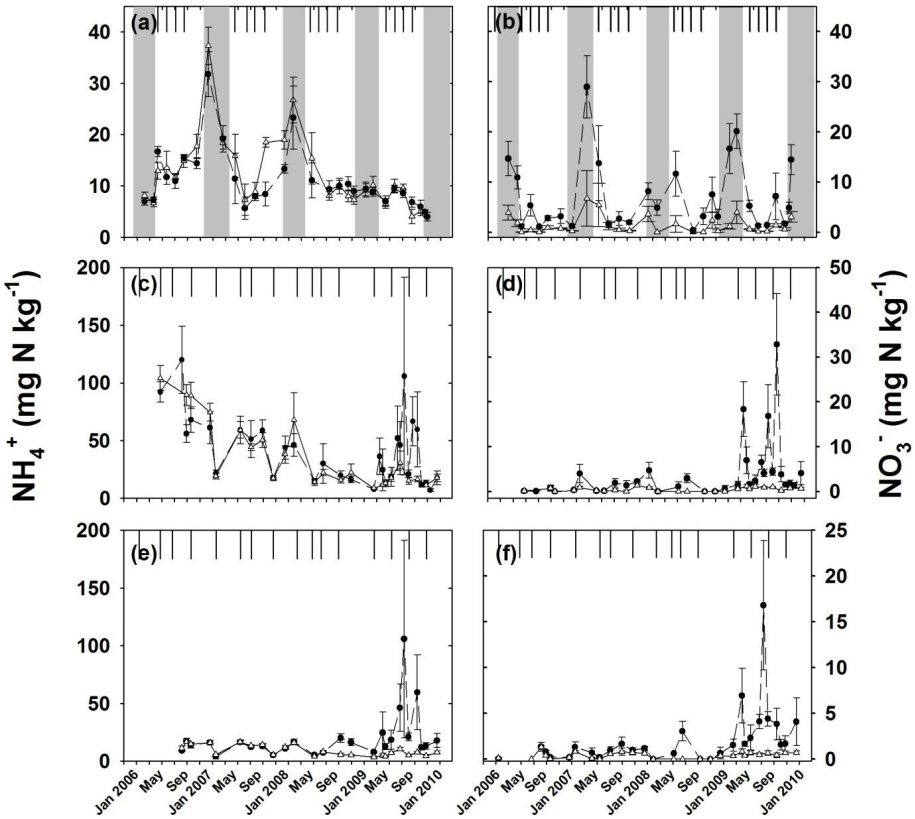

Fig. 2. Mean (±SE, n = 4) soil-extractable ammonium (NH_4^+, left panels) and nitrate (NO_3^-, right panels) at 0–0.05 m mineral soil in the control (Δ) and N-addition (•) plots of the lowland forest (**a** and **b**) and montane forest (**c** and **d** for organic layer, e and f for 0–0.05 m mineral soil). The black vertical lines indicate dates of N addition during 9–12 yr of treatment in the lowland forest and 1–4 yr of treatment in the montane forest. Grey shadings in **(a)** and **(b)** mark the dry seasons.

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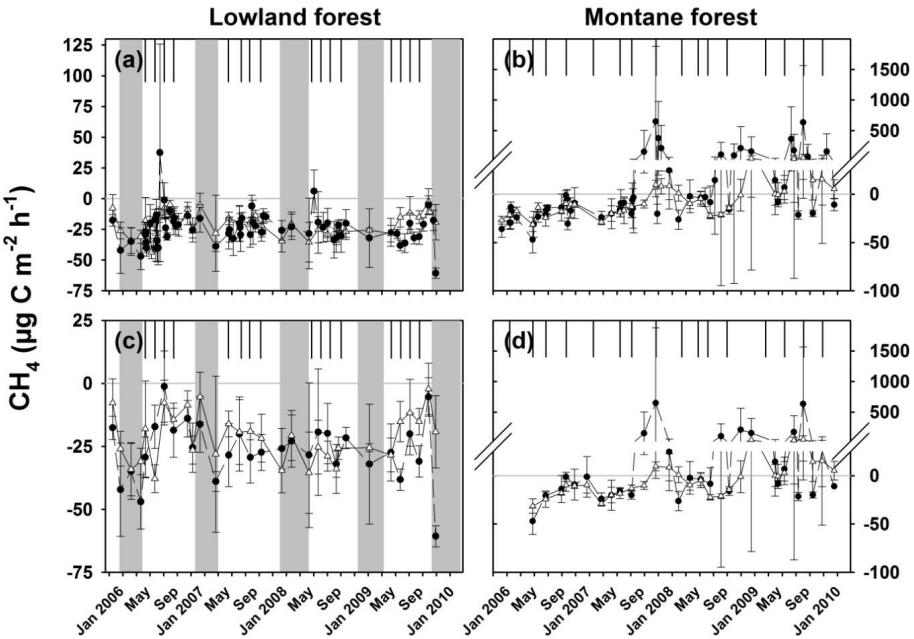

Fig. 3. Mean (±SE, n = 4) soil CH_4-C fluxes from the control (Δ) and N-addition (•) plots of the lowland forest (**a** and **c**) and montane forest (**b** and **d**). The black vertical lines indicate dates of N addition during 9–12 yr of treatment in the lowland forest and 1–4 yr of treatment in the montane forest; the grey horizontal lines mark the zero flux. The upper panels include all fluxes whereas the lower panels show only the long-term fluxes, which were measured at least six weeks after a N addition. Grey shadings in **(a)** and **(c)** mark the dry seasons.

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