Methane Oxidation in Forest, Successional, and No-till Agricultural Ecosystems: Effects of Nitrogen and Soil Disturbance

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ABSTRACT

Methane oxidation in well-aerated soils is a significant global sink for atmospheric methane. We examined the effects of soil disturbance (simulated tillage) and N-fertilizer additions on methane oxidation in old-growth forest, mid-successional, and no-till maize ecosystems in southwest Michigan, USA. We found highest oxidation rates in forest sites (about 30 μg CH_4–C m^{-2} h^{-1} on average), with average rates in successional and agricultural sites about 75 and 12% of this, respectively. In the forest and successional sites a one-time N-fertilizer addition (100 kg NH_4NO_3–N ha^{-1}) significantly suppressed oxidation for the several weeks that inorganic N pools were elevated. There was no effect of fertilizer addition in the agricultural site, where available N was already high and oxidation rates low. Soil disturbance by itself had no detectable effect on fluxes in any of the sites. Results confirm the overriding importance of elevated N for suppressing CH_4 oxidation in managed and unmanaged ecosystems, and suggest further that recovery of CH_4 suppression following agriculture is related to slow-changing soil properties such as soil organic matter composition or microbial community structure.

Upland soil is an important global sink for the greenhouse gas methane, consuming about 30 Tg CH_4 yr^{-1}, slightly more than the annual atmospheric loading rate of 22 Tg CH_4 yr^{-1} (IPCC, 2001). Soil methane uptake thus helps to keep the global atmospheric methane concentration in check, and increased uptake could help to mitigate increasing concentrations of methane in the atmosphere, now at 1745 ppb CH_4 (IPCC, 2001). Land use and in particular agriculture has a big impact on rates of soil CH_4 oxidation: a number of studies have shown that undisturbed forest and grassland soils consume substantially more methane than similar soils converted to agriculture (e.g., Ambus and Christensen, 1995; Willison et al., 1995a; Goulding et al., 1996; MacDonald et al., 1996; Prieme and Christensen, 1999; Robertson et al., 2000). In a variety of different studies various agricultural practices including fertilization, tillage, and the use of insecticides and herbicides have been demonstrated to inhibit soil methane uptake to different degrees (e.g., Mosier and Schimel, 1991; Goulding et al., 1995; Arif et al., 1996; Mosier et al., 1997a; Powlson et al., 1997; Topp et al., 1999; Hütsch, 2001).

Nitrogen fertilizer has been shown most often to reduce methane oxidation in forest, grassland, arable, and landfill soils, especially when applied in the ammonium form (Steudler et al., 1989; Mosier et al., 1991; Hansen et al., 1993; Hütsch et al., 1993; Bronson and Mosier, 1994; Crill et al., 1994; Hütsch et al., 1994; Castro et al., 1995; Willison et al., 1995b; Hütsch, 1996; Tlustos et al., 1998; Hilger et al., 2000). Tillage also has been shown to decrease methane oxidation in both natural and agricultural soils (Hütsch et al., 1994; Willison et al., 1995a; Cochran et al., 1997; Mosier et al., 1997b), however, it had no effect (Sanhueza et al., 1994; Mosier et al., 1998; Burke et al., 1999) and even increased (Kruse and Iversen, 1995) methane uptake in some soils. It is possible that tillage effects are in fact N effects, as tillage is known to increase N turnover in most soils (Robertson and Groffman, 2006), but it is also possible that tillage per se, as it affects soil structure, porosity, and other physical soil properties, inhibits CH_4 uptake. To date there have been no published studies of the effects of N and tillage either alone or interacting within different ecosystems.

In this study we examine both factors simultaneously by tilling and applying N fertilizer separately and in combination along a 3-point land-use gradient that includes old growth deciduous forests, mid-successional old fields equivalent to older Conservation Reserve Program sites, and no-till agricultural fields. In this way we can separate the effects of tillage on N availability, and thus CH_4 oxidation, separate from its effects on other soil properties. To the best of our knowledge, no prior studies have attempted to partition simultaneously the effects of N-fertilizer and tillage on soil CH_4 flux.

MATERIALS AND METHODS

Site Description

This study was conducted at the W.K. Kellogg Biological Station (KBS) Long-term Ecological Research (LTER) site at Hickory Corners, MI (42°24′ N Lat., 85°24′ W Long., elevation 288 m). Annual rainfall at KBS averages 890 mm yr^{-1} with about half falling as snow; potential evapotranspiration (PET) exceeds precipitation for about 4 mo of the year. Mean annual temperature is 9.7°C.

We measured methane fluxes at three replicated sites along a management intensity gradient: in mature deciduous forests (DF) never cut or cleared for agriculture, in mid-successional old fields (SF) abandoned from conventional agriculture 40 to 60 yr before this study, and in a no-till maize-soybean-wheat rotation (T2) established in 1988 on soil that had previously been plowed and farmed for >100 yr.

Dominant plants in the deciduous forest sites are red maple (*Acer rubrum* L.), sugar maple (*Acer saccharum* Marsh.), white oak (*Quercus alba* L.), northern red oak (*Quercus rubra* L.),

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Abbreviations: KBS, W.K. Kellogg Biological Station.

Table 1. Soil properties for the mature deciduous forest, mid-successional communities, and no-till agricultural field at the KBS LTER site. Values are means (±s.e.). Data are from http://lter.kbs.msu.edu.

Soil property	Deciduous forest	Mid-successional communities	No-till maize field
Texture	Sandy loam	Sandy loam	Sandy loam
pH†	5.20 ± 0.12 a	5.66 ± 0.04 b	6.45 ± 0.09 c
Bulk density, g cm^{-3}‡	1.22 ± 0.08 a	1.40 ± 0.04 b	1.57 ± 0.06 bc
Total C, mg C g $soil^{-1}$§	1.52 ± 0.12 a	0.97 ± 0.03 ab	0.73 ± 0.09 b
Total N, mg N g $soil^{-1}$§	0.123 ± 0.09 a	0.093 ± 0.003 a	0.087 ± 0.009 b
Nitrate, μg N g $soil^{-1}$¶	1.93 ± 0.21 a	0.38 ± 0.05 b	2.79 ± 0.30 c
Ammonium, μg N g $soil^{-1}$¶	3.50 ± 0.37 ab	4.46 ± 0.58 a	2.08 ± 0.73 b

† Sampled 8 Mar. 2000 to a depth of 10 cm.
‡ Sampled 10 Apr. 1996 to a depth of 15 cm.
§ Sampled 7 Apr. 1999 to a depth of 25 cm.
¶ Average values from 2000 season to a depth of 25 cm.

flowering dogwood (*Cornus florida* L.), and sassafras [*Sassafras albidum* (Nutt.) Nees]. The mid-successional sites had been farmed for 50 to 100 yr before abandonment, mainly to maize and small grains such as wheat (*Triticum aestivum* L.), oat (*Avena sativa* L.), and barley (*Hordeum vulgare* L.) as per regional agronomic practice. Dominants in the mid-successional communities are Canada goldenrod (*Solidago canadensis* L.), quackgrass [*Elytrigia repens* (L.) Nevski], timothy (*Phleum pratense* L.), white hearth aster (*Aster pilosus* Willd.), Kentucky bluegrass (*Poa pratensis* L.), common yarrow (*Achillea millefolium* L.), Canada bluegrass (*Poa compressa* L.), autumn olive (*Elaeagnus umbellata* Thunb.), sassafras, gray goldenrod (*Solidago nemoralis* Ait.), smooth brome (*Bromus inermis* Leyss.), germander speedwell (*Veronica chamaedrys* L.), orchardgrass (*Dactylis glomerata* L.), flowering spurge (*Euphorbia corollata* L.), and honeysuckle (*Lonicera* spp.). The no-till system was planted to maize (*Zea mays* L.) during 2002, the year that this study was conducted. Before no-till establishment in 1988 the no-till sites had been moldboard plowed and planted mainly to corn and soybean (*Glycine max* L.). The use of fertilizers and pesticides before and during the present study followed best management practices. In the rotation before this study corn received 120 kg N ha^{-1}, wheat 60 kg N ha^{-1}, and soybeans no N fertilizer.

All sites were replicated within the larger landscape (n = 3 locations) and were on the same Kalamazoo/Oshtemo soil series (Austin, 1979). The soils at these sites are Typic Hapludalfs (fine or coarse-loamy, mixed, mesic soils) derived from glacial till about 12 000 yr ago (Crum and Collins, 1995). Soil surface horizon pH ranges from 5.2 in the forest to 6.5 in the no-till soils, and soil C from 1.52 to 0.73%C (Table 1).

Four 0.5 × 0.5 m plots separated by 1 m buffer strips were established in each replicate site and a 2 × 2 factorial design was imposed with N fertilizer and tillage as factors. To one plot was added 100 kg N ha^{-1} ammonium nitrate (NH_4NO_3), another plot was physically disturbed by hand shoveling to simulate soil tillage to 10-cm depth, another plot was both tilled and then fertilized, and a fourth plot served as control. All treatments were imposed within a single 2-h period. Ammonium nitrate was added as a 2000-mL solution sprinkled to simulate a 1-cm rainfall.

Gas Sampling

We measured in situ methane oxidation rates using a static chamber technique (Hutchinson and Livingston, 1993). Chambers were fashioned from a 25-cm diam. PVC pipe: bases (25 cm diameter × 10 cm high) were installed to the 3-cm depth in each plot and left in place except during agronomic operations. Immediately before sampling, a 4.5-cm high cap was placed on each base and sealed to the base with a latex skirt wrapped with an elastic band. At 10-min intervals, four 10-mL headspace samples were removed through a rubber septum in each cap using a syringe and put into 3-mL glass sample vials preflushed with headspace air. Within 3 d, vial contents were measured for CH_4 using a gas chromatograph (GC 5890 Series II, Hewlett-Packard, Palo Alto, CA) equipped with a flame ionization detector (FID), and for CO_2 using an infrared gas absorption (IRGA) analyzer (EGA CO_2 Analyzer, Analytical Development Co. LTD, Hoddesdon, England). Chambers were sampled 1 d before treatment and 1, 6, 16, 23, 52, 73, and 101 d after treatment.

Soil Analyses

Soil temperature was measured at the 0- to 5-cm depth at time of sampling using a soil temperature probe. Soil samples for other analyses were taken from the top 10 cm of soil using a 2.5-cm diam. soil probe. Fresh soils were passed through a 4-mm sieve and mixed by hand, and then subsamples were taken for moisture content and mineral N analysis. Before analysis, soils were stored in a refrigerator at 4°C. Soil moisture content was measured gravimetrically by drying the soil samples at 65°C for 3 d or until dry; further drying these soils at 105°C typically removes <0.8 g H_2O 100 g $soil^{-1}$ more moisture (data not shown). Mineral N measurements were obtained by extracting 20 g of dry soil with 100 mL 1 M KCl for 24 h then filtering through 2-μm pore size glass fiber filters. The filtrates were frozen before analysis for NH_4^+ and NO_3^- using an Alpkem continuous flow analyzer (Alpkem 3550, OI Analytical, College Station, TX) (Bundy and Meisinger, 1994).

Statistical Analyses

The data were divided into two parts: before fertilization (Day 0) and after fertilization (Day 1, 6, 16, 23, 52, 73, and 101). For analysis of the first part, we used SPSS version 10.0.1 (SPSS Inc., 2001) for the analysis of variance (ANOVA), analysis of covariance (ANCOVA), and correlation analysis. We used Proc Mixed of SAS program version 8.0 (SAS Institute, 1999) for the ANOVA and ANCOVA for the postfertilization data, for which we treated site and treatment as fixed effects and day and site × treatment × day as random effects. Methane and CO_2 data were natural log transformed before ANOVA and ANCOVA to homogenize variances. We used untransformed data for correlation analysis.

RESULTS

Methane Oxidation in the Field

Methane oxidation rates were highest in the mature deciduous forest (Table 2), where average rates in the Control treatment were 32 (±3.2, n = 3 sites × 7 sample

Table 2. The effects of soil disturbance and N fertilizer on average methane oxidation, carbon dioxide flux, and soil properties in mature deciduous forest, mid-successional communities, and no-till agriculture at the KBS LTER site. Values are means ± standard error for three replicate sites. Within columns, values followed by different uppercase letters are significantly different ($p < 0.05$) among sites. Within rows, values followed by different lowercase letters are significantly different ($p < 0.05$) among treatments within a site.

	Control	Plowed	N-fertilized	N-fertilized + plowed
		CH_4 (μg CH_4–C m^{-2} h^{-1})		
Mature forest	31.6 ± 3.2 Aa	33.7 ± 3.6 Aa	12.7 ± 1.8 ABb	14.8 ± 2.1 Ab
Mid successional communities	23.7 ± 1.6 Aa	22.0 ± 2.3 Ba	14.8 ± 1.8 Ab	16.1 ± 1.7 Ab
No-till field	4.0 ± 0.7 B	5.0 ± 1.1 C	4.3 ± 1.1 B	2.3 ± 1.3 B
		CO_2 (mg CO_2–C m^{-2} h^{-1})		
Mature forest	108.9 ± 7.7 a	118.8 ± 9.6 a	135.9 ± 13.1 Ab	132.7 ± 12.7 Ab
Mid successional communities	124.9 ± 13.1 a	118.2 ± 14.8 a	153.8 ± 13.6 Ab	156.3 ± 15.5 Ab
No-till field	104.9 ± 9.2	95.6 ± 9.5	83.1 ± 7.4 B	91.5 ± 9.8 B
		Moisture (g H_2O·100g $soil^{-1}$)		
Mature forest	17.6 ± 1.5 A	17.9 ± 1.8 A	20.1 ± 1.6 A	18.5 ± 1.6 A
Mid successional communities	9.1 ± 0.9 B	9.4 ± 1.0 B	8.4 ± 0.8 B	7.9 ± 0.8 B
No-till field	9.7 ± 0.6 B	10.1 ± 0.5 B	10.8 ± 0.5 B	10.6 ± 0.6 B
		Temperature (°C)		
Mature forest	16.3 ± 0.6 AB	16.3 ± 0.6 AB	16.3 ± 0.6 AB	16.3 ± 0.6 AB
Mid successional communities	18.6 ± 0.5 AB	18.6 ± 0.5 AB	18.6 ± 0.5 AB	18.6 ± 0.5 AB
No-till field	20.4 ± 0.5 BC	20.4 ± 0.5 BC	20.4 ± 0.5 BC	20.4 ± 0.5 BC
		Nitrate (μg NO_3^-–N g $soil^{-1}$)		
Mature forest	4.1 ± 0.7 a	6.1 ± 1.1 a	105.2 ± 19.9 ABb	154.0 ± 33.7 b
Mid successional communities	0.5 ± 0.1 a	1.2 ± 0.3 a	77.3 ± 22.9 Aab	137.7 ± 41.6 b
No-till field	30.4 ± 7.4 a	19.7 ± 8.2 a	176.2 ± 25.6 Bb	181.5 ± 17.9 b
		Ammonium (μg NH_4^+-N g $soil^{-1}$)		
Mature forest	21.3 ± 2.8 a	22.0 ± 3.5 a	101.8 ± 20.1 ab	181.2 ± 38.4 b
Mid successional communities	24.9 ± 5.4 a	26.9 ± 4.3 a	98.2 ± 19.6 ab	180.3 ± 42.7 b
No-till field	20.3 ± 4.7 ab	27.2 ± 8.2 abc	124.4 ± 25.0 bcd	161.8 ± 28.0 cd

dates) μg CH_4–C m^{-2} h^{-1} and daily rates over all treatments ranged from 0 to 73 μg CH_4–C m^{-2} h^{-1}. Nitrogen added to forest soils reduced methane oxidation substantially (Fig. 1), with the effect most pronounced before Day 52 (Fig. 2). In contrast, plowing had no significant effect on methane oxidation in the mature forests nor was there a significant fertilizer × plowing interaction. Methane oxidation in both fertilized and plow × fertilized plots in the mature forests dropped sharply after treatment and started to increase after Day 52, when CH_4 uptake in the plow × fertilized plots began to increase faster than in the fertilizer only plots (Fig. 2a). In contrast, CH_4 oxidation in plowed plots was similar to control plots throughout the experiment.

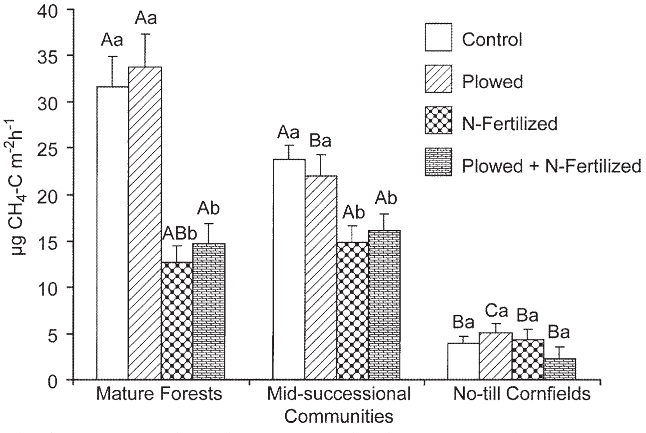

Fig. 1. The reduction of methane oxidation due to soil disturbance and ammonium nitrate fertilizer (100 kg N ha^{-1}) in mature forests, mid-successional communities, and no-till corn (*Zea mays* L.) fields at the W.K. Kellogg Biological Station Long-term Ecological Research (KBS LTER) site. Vertical bars are standard errors of mean (s.e., $n = 3$ sites × 7 sample dates). Different higher and lowercase letters represent significant differences ($P < 0.05$) of treatments among sites and within site, respectively.

Methane oxidation rates in the mid-successional sites were, on average, about 75% of rates in the forest (Table 2): average oxidation rates in the control plots were 24 (±1.6) μg CH_4–C m^{-2} h^{-1} and rates over all plots ranged from 2.4 to 50 μg CH_4–C m^{-2} h^{-1}. Both N fertilizer and N fertilizer plus plowing reduced methane oxidation in these plots significantly ($p < 0.05$), from 24 to 15 μg CH_4–C m^{-2} h^{-1} on average (Fig. 1). In contrast, plowing alone had no detectable effect on oxidation. Similar to fluxes in mature forests, CH_4 oxidation in fertilized and in plowed × fertilized plots exponentially decreased after treatment but started to recover within several weeks (Fig. 2b); in contrast to forest soils, by Day 52 fertilized and plowed × fertilized effects were nil.

Methane oxidation was lowest in the no-till sites, about 12% of rates on average in the forest (Table 2); the average no-till control plot rate was 4.0 (±0.7) μg CH_4–C m^{-2} h^{-1}. Rates across all no-till site treatments ranged from –8 to 17 μg CH_4–C m^{-2} h^{-1} (negative oxidation rates indicate methane production). Neither added N nor plowing significantly affected soil methane uptake in this site (Fig. 1), and rates of oxidation stayed low throughout the experiment (Fig. 2c).

Carbon Dioxide Fluxes

In situ CO_2 production ranged from 20 to 335 mg CO_2–C m^{-2} h^{-1} among the different sites and treatments. On average, CO_2 fluxes did not significantly differ among sites; average control treatment fluxes were 105–125 mg CO_2–C m^2 h^{-1} (Table 2). Nitrogen-fertilizer addition stimulated CO_2 fluxes in the forest and succes-

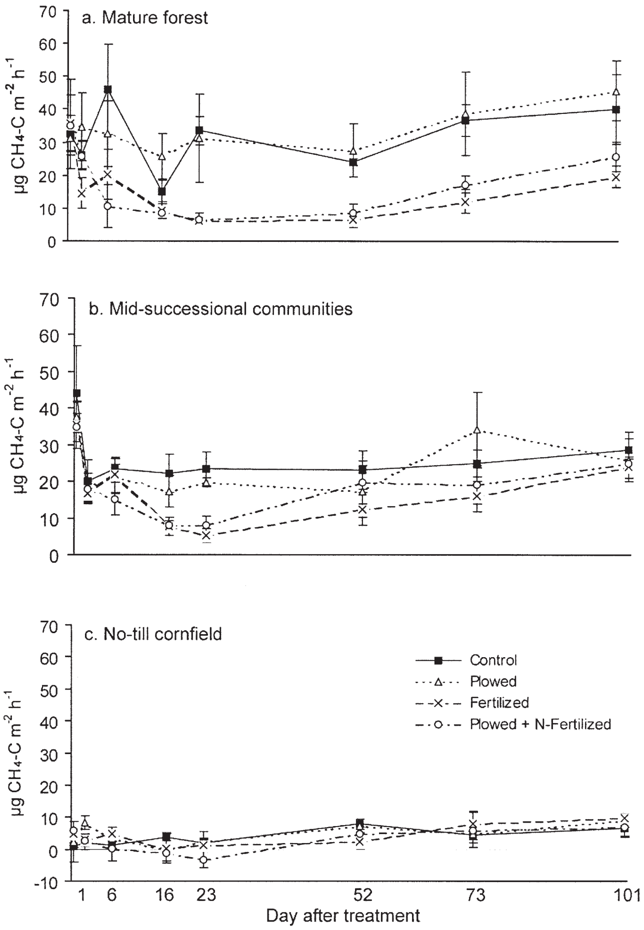

Fig. 2. Average daily methane oxidation as affected by soil disturbance and N-fertilizer: (a) mature deciduous forests, (b) mid-successional communities, (c) no-till corn (*Zea mays* L.) fields, with standard error bars ($n = 3$ sites) per ecosystem type.

sional sites about 20% (Table 2, Fig. 3); there were no detectable fertilizer effects on soil CO_2 flux in the no-till field nor of simulated tillage in any sites except on a few specific sampling dates (Fig. 4).

Soil Physical and Chemical Factors

Average soil moisture (0- to 10-cm depth) was significantly higher ($p < .05$) in mature forests (180 g H_2O kg dry soil^{-1}) than in mid-successional communities (91 g H_2O kg soil^{-1}) and no-till fields (97 g H_2O kg soil^{-1}) (Table 2). Soil moisture was not much affected by treatment and within sites was relatively stable across sample dates (Fig. 5).

Soil temperature was, on average, 2 to 4°C cooler in the mature forests (16°C average) than in the mid-successional communities (18°C) and no-till field (20°C; Table 2). Soil temperature dropped somewhat over the course of the experiment, but the relative ranking of the sites remained unchanged (Fig. 6). There were no significant treatment effects on soil temperature.

Soil nitrate in control plots differed significantly among sites (Table 2). Nitrate was highest in the no-till fields (30.4 ± 7.4 μg NO_3^-–N g soil^{-1}), followed by mature forests (4.1 ± 0.7 μg NO_3^-–N g soil^{-1}) and mid-successional communities (0.5 ± 0.1 μg NO_3^-–N g soil^{-1}) (Table 2). N-fertilizer addition substantially increased soil nitrate in all sites as intended (Table 2); levels of soil nitrate dramatically increased after fertilizer application in every site and then declined rapidly until reaching prefertilization levels at Day 73 for mature forests and Day 52 for mid-successional communities (Fig. 7). In contrast, nitrate in no-till field soils remained high during the sampling period.

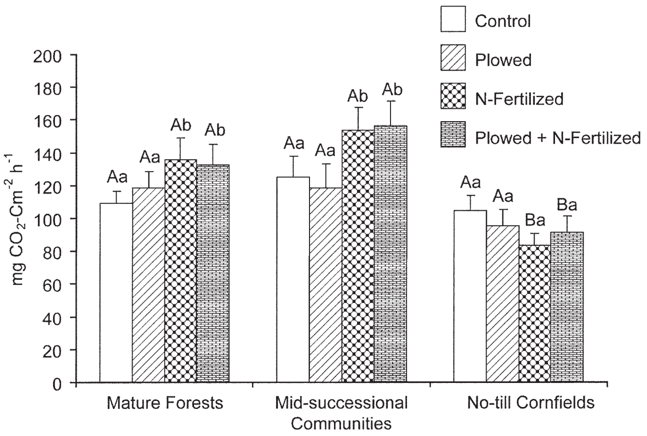

Fig. 3. Net soil CO_2 fluxes as affected by soil disturbance and ammonium nitrate fertilizer (100 kg N ha^{-1}) in mature forests, mid-successional communities, and no-till corn (*Zea mays* L.) fields at the KBS LTER site. Vertical bars are standard errors of mean (s.e., $n = 3$ sites × 7 sample dates). Different higher and lowercase letters represent significant differences ($P < 0.05$) for treatments among sites and among treatments within the same site, respectively.

Unlike nitrate, soil ammonium did not differ significantly among sites; average soil ammonium levels in control plots ranged from 20 to 25 μg NH_4^+–N g soil^{-1} (Table 2). Treatment effects were similar to those for nitrate, with fertilized treatments having an order of magnitude more N than control and plowed treatments, which did not significantly differ. The temporal patterns of soil ammonium were also similar to those for nitrate (Fig. 8).

Controls on Methane and Carbon Dioxide Fluxes

Before treatment, methane oxidation was strongly associated with CO_2 flux ($r = 0.70$, $p < 0.01$) and soil moisture ($r = -0.40$, $p < 0.01$), and weakly associated with soil nitrate levels ($r = -0.40$, $p < 0.05$) (Table 3). Carbon dioxide flux, in turn, was strongly associated with soil temperature ($r = -0.83$, $p < 0.01$) and more weakly associated with moisture, ammonium, and nitrate ($r = 0.34$–0.54, $p < 0.05$) (Table 3).

After treatment, CH_4 fluxes were moderately and approximately equally associated with soil ammonium, nitrate, and temperature ($r = 0.31$, $p < 0.01$). Carbon dioxide fluxes after treatment were most associated with moisture ($r = 0.31$, $p < 0.01$) and ammonium ($r = 0.17$, $p < 0.01$) levels (Table 3).

DISCUSSION

Methane Oxidation

Soil CH_4 oxidation was highest in mature forest, followed by mid-successional communities and no-till agri-

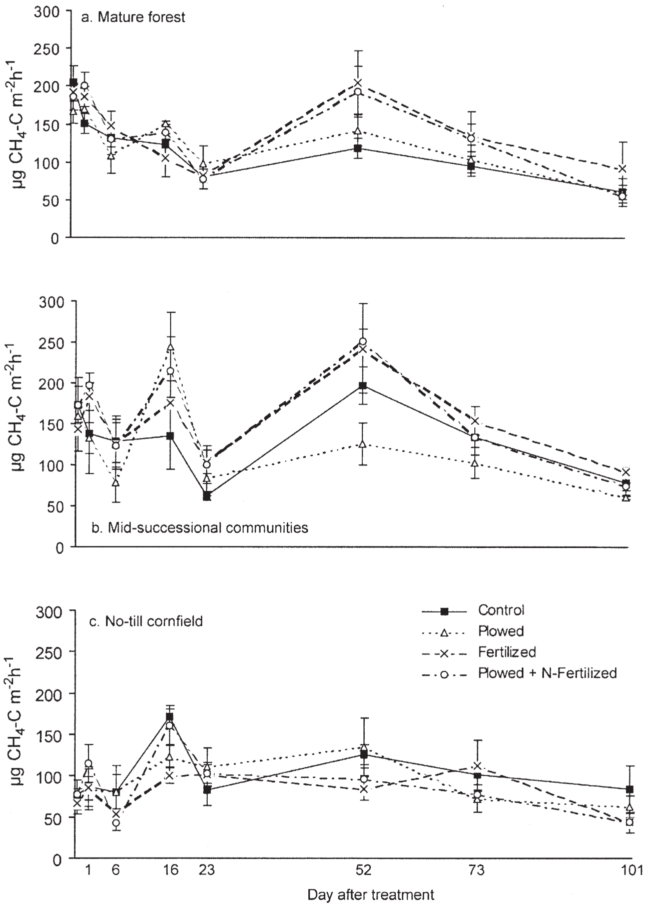

Fig. 4. Average daily carbon dioxide fluxes as affected by soil disturbance and N-fertilizer: (a) mature deciduous forests, (b) mid-successional communities, (c) no-till corn (*Zea mays* L.) fields, with standard error bars ($n = 3$ sites).

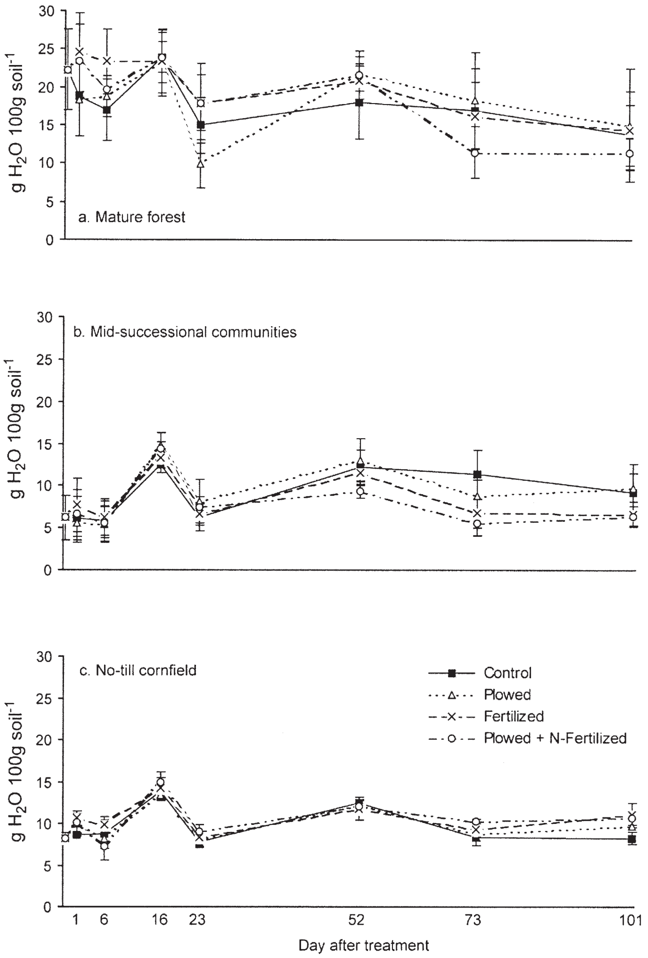

Fig. 5. Average daily soil moisture in the study sites as affected by soil disturbance and N-fertilizer: (a) mature deciduous forests, (b) mid-successional communities, (c) no-till corn (*Zea mays* L.) fields, with standard error bars ($n = 3$ sites).

culture, respectively (Table 2 and Fig. 1). Agricultural fluxes were on average <12% of those in the forests and successional sites. This pattern is similar to that observed in prior agriculture and natural vegetation comparisons (e.g., Mosier et al., 1991; Goulding et al., 1995; Arif et al., 1996; Powlson et al., 1997; Robertson et al., 2000; Hütsch, 2001).

Before treatment, soil temperature appeared to be the most significant factor controlling differences in CH_4 oxidation among sites. Mature forest soils have significantly higher levels of total C, total N, and ammonium as compared with the no-till field soils, which had higher soil nitrate levels and greater bulk density (Table 1). The lower soil bulk density in the forest implies more gas diffusion, which has been shown to increase soil CH_4 uptake by methanotrophs in soil crumbs and soil aggregates (Ball et al., 1997a; Ball et al., 1997b). Mature deciduous forest soils were also acidic, which suggests acid-adapted CH_4 oxidizing bacteria in this forest. These trends among sites also persisted after treatment: across all treatments, mature forest soil still had more CH_4 oxidation than the no-till field soils, while mid-successional communities soils were intermediate (Fig. 1).

Nitrogen-fertilizer (100 kg N ha^{-1}) markedly inhibited soil CH_4 consumption in our forest and mid successional sites, by 60 and 40% respectively (Table 2), similar to patterns found elsewhere (e.g., Bender and Conrad, 1994; King and Schnell, 1994; Hütsch, 1996; Gulledge et al., 1997; King and Schnell, 1998). However, added N did not further suppress CH_4 oxidation in our no-till fields as found in some studies (e.g., Mosier and Schimel, 1991; Bronson and Mosier, 1993; Tate and Striegl, 1993; Mosier et al., 1998), probably due to the already high soil nitrate and ammonium levels in these soils.

We were surprised that soil tillage alone did not show any significant effect on soil CH_4 uptake in our sites. Although the depth of tillage in this study was about 10 cm less than the 20-cm depth of normal tillage, and thus had a less destructive effect on soil structure than normal agricultural tillage, it nevertheless represents a strong soil disturbance to a portion of the soil horizon that is responsible for >50% of soil CH_4 uptake (Suwanwaree and Robertson, unpublished data, 2005). Others working in tropical savanna (Sanhueza et al., 1994), Piedmont floodplain maize fields (Burke et al., 1999), and an acid oxisol site in Puerto Rico (Mosier et al., 1998) have also failed to find a tillage effect.

Although not statistically significant, soil tillage slightly alleviated the inhibition effect of N fertilizer in mature and mid-successional communities, especially after Day 52 (Fig. 2) when soil nitrate and ammonium had already

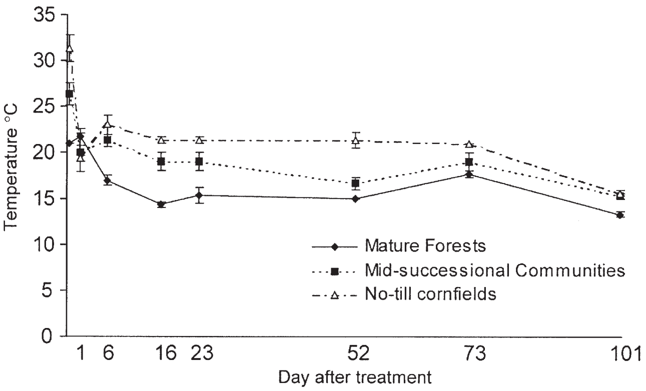

Fig. 6. Average daily soil temperature in three W.K. Kellogg Biological Station (KBS) study sites: (a) mature deciduous forests, (b) mid-successional communities, (c) no-till maize fields, with standard error bars (n = 3 sites). There were no differences among experimental treatments.

declined markedly (Fig. 7 and 8). Tillage may have increased soil aeration in these plots before the onset of compaction, allowing a greater flow of CH_4 into soil microsites and thereby providing CH_4 oxidizing bacteria more access to the gas.

Long-term Recovery of Methane Oxidation

That our successional fields had rates of CH_4 oxidation only midway between those of the no-till and deciduous forest sites suggests a recovery period of well over half a century for methane uptake following the cessation of agricultural activities in these soils. In a 1999 study on these same soils Robertson et al. (2000) had found that soils <10 yr post-abandonment had uptake rates only about 10% greater than those still farmed, suggesting further that recovery starts quickly but is slow. These rates of recovery—decades to century—are similar to those found in Denmark and Scotland (Prieme et al., 1997), in North American grassland sites (Ojima et al., 1993), and in heath soils (Kruse and Iverson, 1995).

Since oxidation rates in our no till plots are no different from tilled plots in the earlier study, and since plowing in none of our plots further inhibited oxidation (Fig. 1), it seems unlikely that the slow recovery of CH_4 oxidation is related to the recovery of soil structure per se. Likewise, since the short-term recovery of suppressed oxidation following N-fertilizer addition was relatively rapid in both the forest and mid-successional communities (Fig. 2), it seems unlikely that the slow recovery is related to persistent N saturation. Rather, long-term recovery is likely related to slow-changing soil properties not related directly to soil structure, such as soil organic matter composition or quantity or microbial community structure.

There may thus be a two-tiered mechanism affecting the suppression of CH_4 oxidation in these soils. In the short-term, suppression appears related principally to short-term enzyme inhibition associated with ammonium availability and its effects on CH_4–oxidizing nitrifiers or heterotrophs. In the long term, following years of fertilization or otherwise elevated N availability, suppression may be related additionally to changes in soil microbial community structure or available substrate. Recovery of CH_4 oxidation may thus depend on both the cessation of chronic N addition and recovery of the soil microbial community, likely also related to long-term changes in soil organic matter.

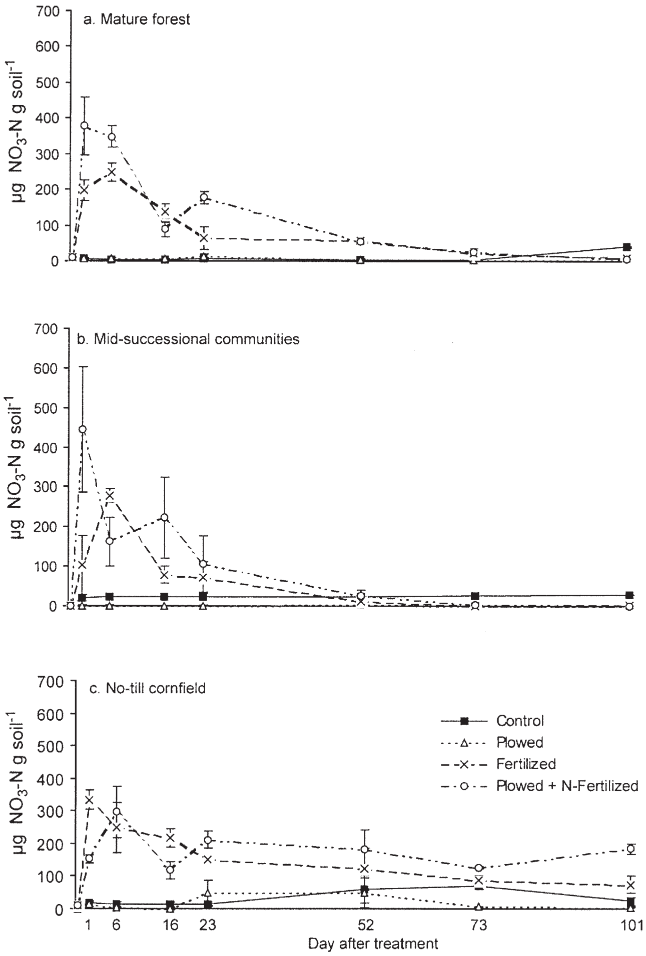

Fig. 7. Average daily soil nitrate in the study sites as affected by soil disturbance and N-fertilizer: (a) mature deciduous forests, (b) mid-successional communities, (c) no-till corn (*Zea mays* L.) fields, with standard error bars (n = 3 sites).

Carbon Dioxide

Carbon dioxide fluxes differed among sites only after treatment. And although treatments did not significantly affect CO_2 fluxes, the fertilizer and fertilized × plowed combination plots in mid-successional and mature forests had 25% higher CO_2 production than control and plowed-only plots (Table 2 and Fig. 3). This suggests a modest but not statistically significant effect of added N on microbial activity. In the no-till field, plowed and N fertilizer treatments had no noteworthy effect on soil CO_2 emission. Soil moisture was the most important positive factor affecting soil CO_2 flux (Table 3).

CONCLUSIONS

Mature forest soils had the highest overall methane oxidation rates, followed by mid-successional and agricultural systems, respectively.

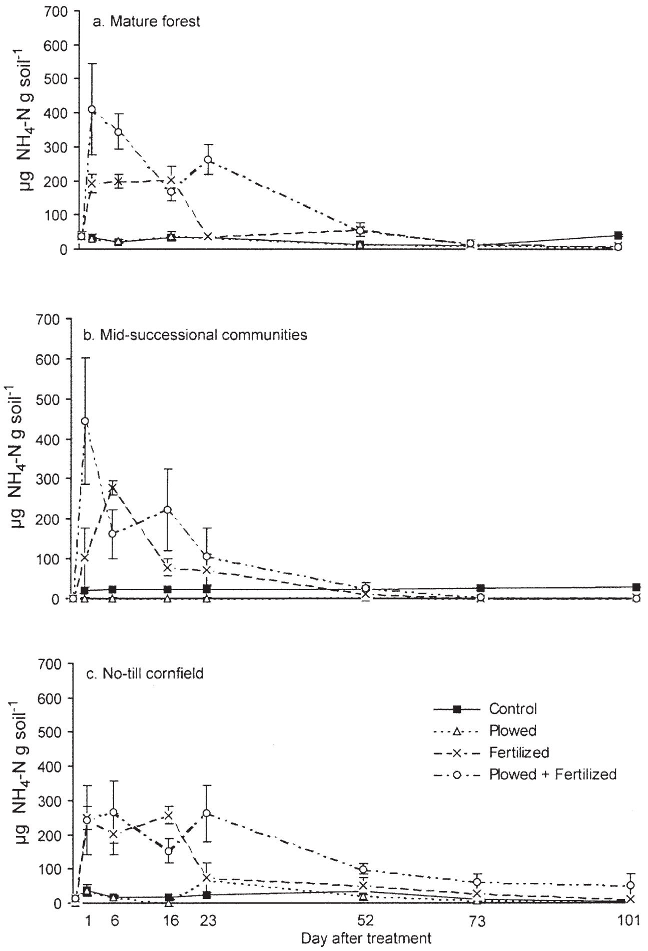

Fig. 8. Average daily soil ammonium as affected by soil disturbance and N-fertilizer: (a) mature deciduous forests, (b) mid-successional communities, (c) no-till corn (*Zea mays* L.) fields, with standard error bars ($n = 3$ sites).

Nitrogen added to the forest and mid-successional sites substantially reduced methane oxidation within 2 wk of N addition. In the no-till site, where oxidation rates were already low and soil N levels already high, additional N had no effect on soil CH_4 uptake. Nitrogen suppression of CH_4 uptake persisted for 8 wk in the successional sites and >15 wk in the forest; recovery was associated with the return of soil inorganic N pools to background levels.

Table 3. Relationships among methane oxidation, carbon dioxide, and other soil properties before and after experimental treatment in mature deciduous forest, the mid-successional community, and the no-till agricultural sites. Values are Pearson correlation coefficients (*r*).

	CO_2	Moisture	Temperature	Nitrate	Ammonium
			Before treatment†		
CH_4	0.701§	−0.678§	−0.168	−0.401¶	0.179
CO_2	1.00	0.536¶	−0.831§	−0.336¶	0.512§
			After treatment‡		
CH_4	0.054	0.094	−0.339§	−0.279§	−0.216§
CO_2	1.000	0.306§	0.009	0.055	0.170§

† $n = 3$ sites × 3 replications × 4 sample plots.
‡ $n = 3$ sites × 3 replications × 4 treatments × 7 sample dates.
§ Correlation is significant at the 0.01 level (2-tailed).
¶ Correlation is significant at the 0.05 level (2-tailed).

Plowing had no detectable effect on methane oxidation in any of the three sites. The effects of plowing + N-fertilizer were no greater than the effects of fertilizer alone.

The impact of agriculture on methane oxidation is thus likely due primarily to greater N availability via N fertilization rather than to the disruption of soil structure or other effects of plowing. Substantially increasing the N inputs to mid-successional and mature ecosystems reduces rates of oxidation that would otherwise be relatively high.

Recovery of CH_4 oxidation rates from long-term suppression appears related to the recovery of microbial community structure or soil organic matter composition following the cessation of elevated N inputs.

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