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Residual Effects of Fertilization History Increase Nitrous Oxide Emissions from Zero-N Controls: Implications for Estimating Fertilizer-Induced Emission Factors

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Abstract

Agricultural N fertilization is the dominant driver of increasing atmospheric nitrous oxide (N_2O) concentrations over the past half-century, yet there is considerable uncertainty in estimates of N_2O emissions from agriculture. Such estimates are typically based on the amount of N applied and a fertilizer-induced emission factor (EF), which is calculated as the difference in emissions between a fertilized plot and a zero-N control plot divided by the amount of N applied. A fertilizer-induced EF of 1% is currently recognized by the Intergovernmental Panel on Climate Change (IPCC) based on several studies analyzing published field measurements of N_2O emissions. Although many zero-N control plots used in these measurements received historical N applications, the potential for a residual impact of these inputs on N_2O emissions has been largely ignored and remains poorly understood. To address this issue, we compared N_2O emissions under laboratory conditions from soils sampled within zero-N control plots that had historically received N inputs versus soils from plots that had no N inputs for 20 yr. Historical N fertilization of zero-N control plots increased initial N_2O emissions by roughly one order of magnitude on average relative to historically unfertilized control plots. Higher N_2O emissions were positively correlated with extractable N and potentially mineralizable N. This finding suggests that accounting for fertilization history may help reduce the uncertainty associated with the IPCC fertilizer-induced EF and more accurately estimate the contribution of fertilizer N to agricultural N_2O emissions, although further research to demonstrate this relationship in the field is needed.

Core Ideas

- The IPCC fertilizer-induced emission factor for N_2O is based on zero-N control plots.
- Many of these zero-N control plots received fertilizer N inputs 1 to 2 yr earlier.
- We compared soils with no N inputs (>20 yr) to soils with N inputs until 1 to 2 yr ago.
- Soils with historical N inputs had higher N_2O emissions during an incubation.
- Higher N_2O emissions were related to increased soil inorganic N and labile organic N.

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Increased inorganic N fertilizer use was integral to agricultural productivity gains during the past half-century (Pingali, 2012), and further increases in inorganic N use will likely be required to meet the expected doubling of food demand by 2050 (Mueller et al., 2012). However, the potential environmental consequences of inorganic N fertilizer use in agriculture are increasingly recognized, especially in the case of agricultural greenhouse gas (GHG) emissions and climate change (Foley et al., 2011). Agricultural production is responsible for approximately 5.2 to 5.8 Gt CO_2–eq yr^{-1}, which represents about 10 to 12% of all anthropogenic GHG emissions (Smith et al., 2014). Nitrous oxide (N_2O) emissions, which per molecule have a global warming potential that is 298 times that of carbon dioxide (CO_2), account for about half of agriculturally derived global warming potential worldwide (Myhre et al., 2013). In addition to the contribution of N_2O to climate change, it is thought to be the principal ozone-depleting substance currently being emitted (Ravishankara et al., 2009). Given the profound effects of N_2O emissions on the global environment, it is critical to accurately quantify anthropogenic N_2O emissions from agriculture and to understand how different agricultural management practices affect N_2O emissions.

The major agricultural sources of N_2O are inorganic fertilizers, manure management (including application to pasture and cropland), and crop residues (Smith et al., 2014). Early efforts to estimate agricultural N_2O emissions therefore focused on various forms of N fertilization and attempted to determine a general emission factor (EF) to describe the percent of N inputs directly lost as N_2O (Bouwman, 1996; Mosier et al., 1996; Mosier et al., 1998). A fertilizer-induced EF of 1% was adopted by the Intergovernmental Panel on Climate Change (IPCC) based on several studies analyzing published field measurements of N_2O emissions (Bouwman et al., 2002a, 2002b; Novoa and Tejeda, 2006; Stehfest and Bouwman, 2006); in particular, the

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Abbreviations: EF, emission factor; FA-N, fallow phase of the wheat–fallow rotation with historical N fertilization; FA-O, fallow phase of the wheat–fallow rotation with no historical N inputs for more than 20 yr; GHG, greenhouse gas; IPCC, Intergovernmental Panel on Climate Change; PMN, potentially mineralizable N; WFPS, water-filled pore space; WLCC, winter legume cover crop; WT-L, wheat–legume system going into the wheat phase of the rotation; WT-N, wheat phase of the wheat–fallow rotation with historical N fertilization; WT-O, wheat phase of the wheat–fallow rotation with no historical N inputs for more than 20 yr.

estimation of the "mineral fertilizer and organic amendment" component of this EF is based on a dataset that includes over 1000 field measurements of cumulative N_2O emissions (IPCC, 2006; Stehfest and Bouwman, 2006). The fertilizer-induced EF is calculated as the difference in N_2O emissions between fertilized and zero-N control plots, expressed as a percentage of the fertilizer N applied (Bouwman, 1996; Stehfest and Bouwman, 2006). Although this procedure accounts for the effect of N fertilizer applied during the current field season, it ignores the fact that many of the zero-N control plots in the field trials used to estimate the fertilizer-induced EF received N inputs in the years preceding the experiments. "Zero-N control plots" are defined here as plots that receive 0 kg N ha^{-1} during the course of the experiment, which is frequently 1 yr, but that may or may not have received N fertilizer before the experiment; the interpretation used here is consistent with that used throughout much of the literature regarding the fertilizer-induced EF used by the IPCC (Bouwman, 1996; Bouwman et al., 2002a; IPCC, 2006; Mosier et al., 1996; Stehfest and Bouwman, 2006). A random sample of 10% of the measurements from areas under zero-N application in the dataset used by Stehfest and Bouwman (2006) revealed that, of those measurements for which fertilization history could reasonably be determined or inferred, 71% were historically fertilized (within 1–2 yr prior) and 29% had not received historical N inputs (for at least 20 yr). The approach used to calculate the fertilizer-induced EF thus overlooks the potential for a residual effect of previous N fertilizer applications on current N_2O emissions.

Despite early recognition that historical fertilization and management may be important factors in explaining variability in field N_2O measurements (Bouwman, 1996), few studies have directly evaluated the effect of historical inorganic N fertilization on N_2O emissions. Historical fertilization has been shown to increase soil denitrification capacity under supplemental N fertilization due to increases in total organic C and microbial biomass C (Drury et al., 1998). Furthermore, Graham et al. (2013) found that N_2O emissions from a historically manured soil were significantly higher than emissions from a nonmanured soil, even with no N application in the current year. Although this finding provides clear evidence of a residual effect from historical fertilization, it does not allow the effect of historical N inputs to be distinguished from that of historical C inputs. Our study specifically sought to assess the effect of historical N fertilization on potential N_2O emissions in the absence of external C inputs and supplemental N fertilization in the laboratory. To this end, we measured N_2O emissions under laboratory conditions using soils that differed only in inorganic N application over a 20-yr period. We hypothesized that soils collected from zero-N control plots with recent (1–2 yr prior) historical N fertilizer application would show residual effects from past N fertilization and have higher N_2O emissions than soils from zero-N control plots that received no historical N inputs for more than 20 yr.

Materials and Methods

Study Site and Treatment Structure

Soils were collected from the Century Experiment plots (formerly called Long-Term Research on Agricultural Systems) at the Russell Ranch Sustainable Agriculture Facility in Davis, CA (38°32′24″ N, 121°52′12″ W). Detailed information on the study site (including all main plot treatments for the Century Experiment), site management history before the initiation of the experiment, and historical yields for the rainfed wheat plots used in this study can be found in Agricultural Sustainability Institute (2016) and in Denison et al. (2004). The region is characterized by a Mediterranean climate, with over 90% of rainfall occurring from November to April and an average annual precipitation of around 500 mm (Denison et al., 2004). The lowest average monthly temperature is approximately 8°C in December, and the highest is approximately 23°C in July (Denison et al., 2004). The soils at the Century Experiment are a Yolo silt loam (fine-silty, mixed, nonacid, thermic Typic Xerorthent) and a Rincon silty clay loam (fine, montmorillonitic, thermic Mollic Haploxeralf). This research was conducted within a set of treatments consisting of rainfed wheat (*Triticum aestivum* L.) with alternating years of either a bare fallow or a winter legume cover crop (WLCC) mixture of vetch (*Vicia dasycarpa* Ten.) and pea (*Pisum sativum* L.). In total, five treatments representing different phases of these 2-yr crop rotations with distinct historical N management practices were considered (Fig. 1): (i) wheat phase of the wheat–fallow rotation with historical N fertilization (WT-N), (ii) fallow phase of the wheat–fallow rotation with historical N fertilization (FA-N), (iii) wheat phase of the wheat–fallow rotation with no historical N inputs (WT-O), (iv) fallow

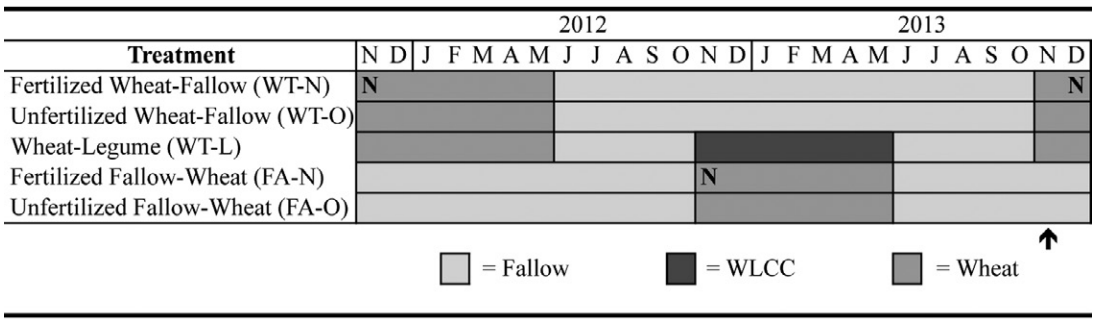

Fig. 1. Timeline of historical cropping and fertilization treatments in Century Experiment plots at the Russell Ranch Sustainable Agriculture Facility in Davis, CA, with each treatment represented in three replicated blocks. Soil samples (shown by arrow) were taken in November 2013 before inorganic N fertilization (shown by "N"). WLCC indicates a winter legume cover crop and was the source of organic N for the wheat–legume system going into the wheat phase of the rotation (WT-L), whereas other fertilized treatments received 84 kg N ha^{-1} as ammonium sulfate [$(NH_4)_2SO_4$].

phase of the wheat–fallow rotation with no historical N inputs (FA-O), and (v) a wheat–legume system going into the wheat phase of the rotation (WT-L).

The wheat–legume (WT-L) treatment only received N inputs associated with N_2 fixation by the vetch and pea cover crop mixture, which was incorporated into the soil each spring in alternating years over the 20-yr duration of the experiment. The total N content of the cover crop biomass has previously been measured at 134 to 202 kg N ha^{-1}, which includes both biologically fixed N_2 and N uptake from the soil (McGuire et al., 1998). The wheat–fallow rotation with historical N fertilization (WT-N, FA-N) received 84 kg N ha^{-1} as ammonium sulfate [$(NH_4)_2SO_4$] every other year before sowing of wheat. The wheat–fallow rotation with no historical N inputs (WT-O, FA-O) did not receive N fertilizer over the 20-yr duration of the experiment. Immediately before the initiation of the long-term experiment, the site was cropped to unfertilized sudangrass (*Sorghum vulgare* Pers.) for 2 yr and before that commercial alfalfa (*Medicago sativa* L.) was grown with an unknown fertilization history (Denison et al., 2004). The historically fertilized wheat phase of the wheat–fallow treatment (WT-N) last received inorganic N fertilizer 2 yr before soil sampling, and the historically fertilized fallow phase of the wheat–fallow rotation (FA-N) last received N fertilizer 1 yr before sampling (Fig. 1). Fertilization history was treated as a categorical variable ("historically fertilized" [i.e., received N inputs until 1–2 yr before sampling] or "historically unfertilized" with no N inputs for more than 20 yr) rather than as a continuous variable of time since fertilization, given that each phase of the crop rotation had only two different times since fertilization. The size of each experimental unit is 0.4 ha, replicated three times across a 40-ha field site.

Soil Sampling and Nutrient Availability Assessment

Soil samples (0–15 cm) were collected separately from each field plot in November 2013 after initial bed preparation (tillage) but before N fertilization and planting. Nine soil cores were taken per field plot (135 cores total). Samples were thoroughly mixed, air-dried, and passed through a 2-mm sieve to remove organic matter debris and gravel and to homogenize soil structure. Three laboratory replicates were used for determination of extractable N and potentially mineralizable N (PMN) in soil collected from each of the 15 field plots. For extractable N, subsamples (10 g) of the 2-mm sieved soil were added to 100 mL of 2 mol L^{-1} KCl and mixed for 1 h on a mechanical shaker. The solution was filtered through Whatman Grade 42 filter paper (GE Healthcare UK Limited) and stored at 4°C before colorimetric analysis for NH_4^+ and NO_3^- on a spectrophotometer (Keeney and Nelson, 1982). Soil samples were analyzed for PMN according to methods adapted from Powers (1980). In brief, 5 g dry soil was incubated anaerobically in 10 mL of deionized water at 40°C for 7 d. Before capping the vials, N_2 gas was bubbled through the solution to facilitate the removal of oxygen and induce anaerobic conditions (Fonte and Six, 2010). Samples were mixed with 50 mL of 2 mol L^{-1} KCl after 7 d, shaken for 1 h, and analyzed for NH_4^+ as described above. Soil samples were analyzed for total N and C at the UC Davis Stable Isotope Facility using dry combustion at 1000°C and an Elementar Vario EL Cube or Micro Cube elemental analyzer (Elementar Analysensysteme GmbH).

Incubation Experiment

Subsamples of 200 g each from individual main treatment plots (15 total) were repacked into specimen cups with perforated bases to a bulk density of 1.25 g cm^{-3}. These specimen cups were initially wetted through capillary rise by placing cups in Petri dishes with deionized water (Haney and Haney, 2010). The cups were weighed, and the remaining water was added to the upper portion of the soil with a syringe to bring the samples up to 80% water-filled pore space (WFPS), a level chosen based on the results of preliminary incubations at differing levels of WFPS. A total of three laboratory replicates was used for each of the soils being incubated (45 cores total). Three control samples were prepared in the same manner but without the addition of soil. Samples were placed in 1.89-L Mason jars and incubated in the dark at 25°C for 2 wk (the duration of the gas sampling period) with the lids off to allow for air circulation and soil drying. After 1 wk, samples were rewetted to 80% WFPS using the same approach to simulate wet–dry cycles occurring in the field.

Gas Sampling and Analysis

Gas sampling was performed at 4, 28, 52, 76, 100, and 172 h after the initial wetting and again at 4, 28, 52, and 76 h after the rewetting. At each sampling event, the Mason jars were removed from the 25°C temperature-controlled room, ventilated thoroughly with a fan, returned to the room, and sealed with airtight lids containing brass ports and silicon septa. For logistical considerations, sampling events were conducted in two sampling blocks, with each of the 15 field plots (historical treatment and field block combination) represented in both blocks. Gas samples (25 mL) were removed with a syringe from each Mason jar at 20, 40, and 60 min and injected into 12.5-mL evacuated glass vials (Labco Ltd.). Ambient gas samples were taken at the time of lid placement. All samples were analyzed for N_2O peak area on a GC-2014 gas chromatograph equipped with a ^{63}Ni electron capture detector set at 325°C for N_2O concentrations (Shimadzu Scientific). Nitrous oxide was separated by a stainless steel column packed with Hayesep D, 80/100 mesh at 75°C. A 1-mL headspace gas was injected into the GC inlet port using an auto-sampler (Bandolero, XYZTEK).

Flux Calculations

The peak area for each gas sample was converted to a concentration using linear regression ($R^2 > 0.99$) against three standards with known gas concentrations (1, 3.05, and 9.95 ppm N_2O). Fluxes of N_2O were estimated from the linear increase of gas concentration over time based on $R^2 \geq 0.9$ (Linquist et al., 2015). Gas concentrations were converted to mass per unit weight ($\mu g\ N_2O\ kg^{-1}$) using the Ideal Gas Law with the room air temperature measured during each sampling event and an atmospheric pressure of 0.101 MPa. Fluxes of N_2O were computed as:

$$F = \frac{\Delta C}{\Delta t} \times \frac{V}{A} \times \propto \quad [1]$$

where F is gas flux rate for N_2O ($\mu g\ N_2O$–N kg $soil^{-1}$ h^{-1}), $\Delta C/\Delta t$ denotes the increase or decrease of gas concentration in the chamber (g L^{-1} h^{-1}), V is the Mason jar volume (L), A is weight of the soil (kg), and α is a conversion coefficient for

elemental N. A significance test was made to determine if the gas flux was close or equal to zero at $P < 0.05$. Gas fluxes that failed the linearity test were not included in the data analysis, whereas gas fluxes that failed the significance and detection tests were set to zero flux. A complete discussion of the flux methodology is described in Adviento-Borbe et al. (2015).

Data Analysis

Linear interpolation was used to integrate the N_2O fluxes over the incubation period across all sampling time points for each individual subsample. We used the PROC MIXED model for variance analysis on the cumulative N_2O fluxes with the historical treatment as a fixed effect, blocking variables as random effects (the aforementioned sampling blocks in the laboratory and long-term field blocks), and the subsamples (i.e., laboratory replicates) nested within the historical treatment and field block combinations (SAS Institute Inc., 2011). All possible two- and three-way interactions were tested, and interactions were included in the model to the degree that it minimized the corrected Akaike Information Criteria, which resulted in the inclusion of the two-way treatment interactions with both the field block and sample block as random effects; the interactions were treated as random effects because they both included at least one random effect, in accordance with SAS Institute Inc. (2011). An optimized power-transformation was performed on the data ($N_2O–N^{0.211}$) to ensure that the assumptions of normality, homogeneity of variance, and nonmultiplicative treatment effects were met. Because an exploratory model showed no significant interaction between fertilization history and the crop rotation phase, orthogonal contrasts were used to compare the main effect of historical inorganic N fertilization (WT-N, FA-N vs. WT-O, FA-O) and the main effect of the phase of the wheat–fallow rotation (WT-N, WT-O vs. FA-N, FA-O). Soil nutrient data met the ANOVA assumptions without transformation and were analyzed using PROC MIXED and an adjusted Tukey's for separation of treatment means. Simple linear regression (PROC GLM) was used to analyze relationships between selected soil parameters (extractable N, PMN, total N, and total C) and cumulative N_2O emissions (SAS Institute Inc., 2011).

Results

N_2O Fluxes

Maximum N_2O emissions, which ranged from approximately 0.8 to 13.6 μg $N_2O–N$ kg $soil^{-1}$ h^{-1}, occurred for all treatments between 4 and 52 h after the initial wetting and decreased to almost zero before the rewetting 1 wk later (Fig. 2). Emissions were lowest for soils from the fallow phase of the historically unfertilized wheat–fallow rotation (FA-O) and highest for soils from the wheat phase of the historically fertilized wheat–fallow rotation (WT-N). Much lower emissions (averaging 3% of the observed maximum for each treatment) were found in all treatments after rewetting at 7 d, and the N_2O flux again declined to approximately zero within 52 h. The cumulative N_2O flux was highest (523 μg $N_2O–N$ kg^{-1}) in soils entering the wheat phase of the historically fertilized wheat–fallow rotation (WT-N) and lowest (16.6 μg $N_2O–N$ kg^{-1}) in soils entering the fallow phase of the historically unfertilized wheat–fallow rotation (FA-O) (Fig. 3). The wheat phase of the historically fertilized wheat–fallow rotation (WT-N) had approximately 4.4 times higher cumulative N_2O emissions than the wheat phase of the historically unfertilized wheat–fallow rotation (WT-O), and the fallow phase of the historically fertilized wheat–fallow rotation (FA-N) had approximately 18 times higher cumulative N_2O emissions than the fallow phase of the historically unfertilized wheat–fallow rotation (FA-O). On average, cumulative N_2O emissions during the incubation (which consisted of two wetting

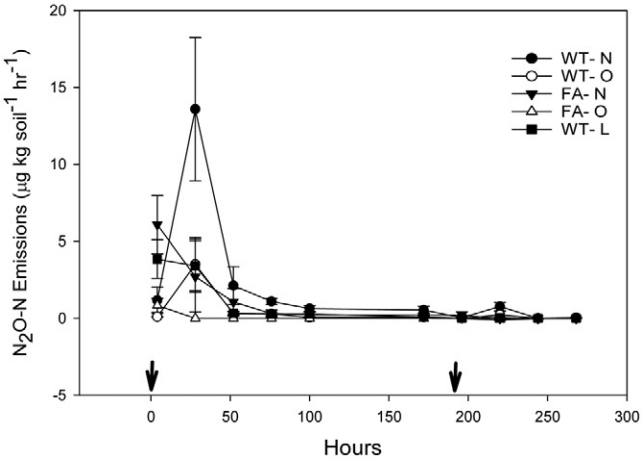

Fig. 2. Average N_2O fluxes over the 10-d incubation in soils from the Century Experiment plots at the Russell Ranch Sustainable Agriculture Facility in Davis, CA, with different historical N management treatments: fertilized (WT-N) and unfertilized (WT-O) plots entering the wheat phase of the rotation, fertilized (FA-N) and unfertilized (FA-O) plots entering the fallow phase of the rotation, and plots with a leguminous cover crop in alternating years entering the wheat phase of the rotation (WT-L). The arrows indicate the initial wetting of soils to 80% WFPS and a subsequent rewetting. Error bars represent the SEM.

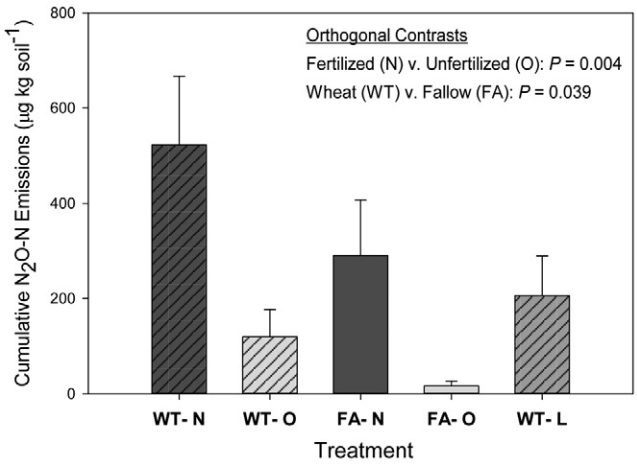

Fig. 3. Average cumulative N_2O emissions during a laboratory incubation using soils from the Century Experiment plots at the Russell Ranch Sustainable Agriculture Facility in Davis, CA, with different historical N management treatments: fertilized (WT-N) and unfertilized (WT-O) plots entering the wheat phase of the rotation, fertilized (FA-N) and unfertilized (FA-O) plots entering the fallow phase of the rotation, and plots with a leguminous cover crop in alternating years entering the wheat phase of the rotation (WT-L). Hatched and clear patterns indicate treatments going into wheat and fallow, respectively; bar colors indicate the type of N fertilization (dark gray = inorganic N, light gray = leguminous cover crop, and white = none). Orthogonal contrasts between treatments are shown with the corresponding *P* value: historically fertilized versus historically unfertilized treatments (WT-N, FA-N vs. WT-O, FA-O) and treatments going into the wheat versus fallow phases of the wheat–fallow rotation (WT-N, WT-O vs. FA-N, FA-O). Error bars represent the SEM.

events) were approximately one order of magnitude greater for the historically fertilized wheat–fallow rotation than for the historically unfertilized wheat–fallow rotation ($P = 0.004$) (Fig. 3). The treatments entering the fallow phase of the wheat–fallow rotation showed lower cumulative N_2O emissions than those entering the wheat phase of the wheat–fallow rotation for both the historically fertilized (WT-N > FA-N) and unfertilized treatments (WT-O > FA-O); the average emissions from the treatments entering the fallow phase of the wheat–fallow rotation were significantly lower than those from the treatments going into the wheat phase of the wheat–fallow rotation ($P = 0.039$) (Fig. 3). The cumulative N_2O emissions from the treatment entering wheat with a WLCC in alternating years (WT-L) were intermediate between the historically fertilized (WT-N) and historically unfertilized (WT-O) treatments entering the wheat phase of the wheat–fallow rotation, although this difference could not be tested with orthogonal contrasts.

Soil N and C

Similar to trends observed for N_2O fluxes, total extractable soil N (combined NH_4^+–N and NO_3^-–N) was highest for the wheat phase of the historically fertilized wheat–fallow rotation (WT-N) and lowest for the fallow phase of the historically unfertilized wheat–fallow rotation (FA-O) (Table 1). The same general trend was observed for PMN, with treatments receiving organic or inorganic N (WT-L, WT-N, FA-N) having 66% higher PMN on average than treatments receiving no N inputs (WT-O, FA-O). Extractable N levels were positively correlated with N_2O production ($P = 0.001$) and explained approximately 56% of the variation in cumulative N_2O emissions (Fig. 4). The PMN was also positively correlated with N_2O emissions (Table 1). Total N and C varied from 0.86 to 1.33 g kg^{-1} dry soil and from 7.35 to 12.05 g kg^{-1} dry soil, respectively. Although treatments with historical N inputs (inorganic N or WLCC) tended to have higher total N and C contents, there were no significant differences in total soil C among treatments, and the only difference in total soil N was between the wheat–legume system going into the wheat phase of the rotation (WT-L) and the wheat phase of the historically unfertilized wheat–fallow rotation (WT-O) (Table 1). No correlation was observed between cumulative N_2O emissions and either total N or total C (Table 1).

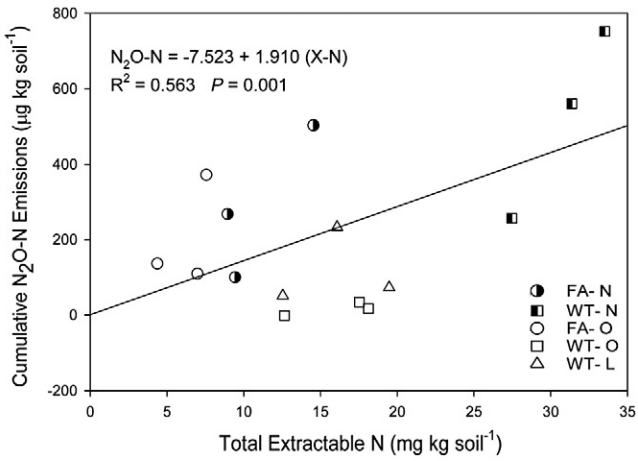

Fig. 4. Correlation between total extractable N in soil samples before a laboratory incubation using soils from the Century Experiment plots at the Russell Ranch Sustainable Agriculture Facility in Davis, CA, and cumulative N_2O emissions during the incubation. Historical management and N fertilization treatments are indicated by distinct symbols: fertilized (WT-N) and unfertilized (WT-O) plots entering the wheat phase of the rotation, fertilized (FA-N) and unfertilized (FA-O) plots entering the fallow phase of the rotation, and plots with a leguminous cover crop in alternating years entering the wheat phase of the rotation (WT-L).

Discussion

Historical N Application as a Driver of N_2O Emissions

In this laboratory incubation, N_2O flux dynamics were comparable to the general temporal trends found in similar studies (e.g., Beare et al., 2009; Zhu et al., 2013), with the greatest emissions observed between 4 and 52 h after the initial wetting (Fig. 2). Peak daily flux estimates were approximately 0.02 to 0.33 mg N_2O–N kg $soil^{-1}$ d^{-1}, and, although a much smaller peak was observed on rewetting, the relative treatment differences remained consistent (Fig. 2). Differences in cumulative N_2O emissions among treatments were principally driven by peak emissions in the first few days after wetting (one sampling event in particular), and, given the resultant uncertainty surrounding the cumulative N_2O emissions, the results and associated correlations reported here should be interpreted with caution; nonetheless, this is consistent with the results of a preliminary incubation

Table 1. Correlation of average total extractable N, potentially mineralizable N, total N, and total C values with cumulative N_2O emissions for soils collected from five rainfed winter wheat treatments in the Century Experiment at the Russell Ranch Sustainable Agricultural Facility in Davis, CA.

Treatment†	Total extractable N	Potentially mineralizable N	Total N	Total C
	mg kg^{-1} dry soil		g kg^{-1} dry soil	
WT-N	30.8 a‡	40.6 ab	1.13 ab	10.1 a
WT-O	16.1 b	31.0 b	0.993 b	8.75 a
WT-L	16.0 b	46.7 a	1.23 a	10.9 a
FA-N	11.0 bc	48.1 a	1.17 ab	10.4 a
FA-O	6.30 c	28.7 b	1.03 ab	9.56 a
R^2§	0.563	0.407	0.204	0.116
P value	0.001**	0.011*	0.091	0.213

* Significant at the 0.05 probability level.

** Significant at the 0.01 probability level.

† Treatments are fertilized (WT-N) and unfertilized (WT-O) plots entering the wheat phase of the rotation, fertilized (FA-N) and unfertilized (FA-O) plots entering the fallow phase of the rotation, and plots with a leguminous cover crop in alternating years entering the wheat phase of the rotation (WT-L).

‡ Values followed by the same letter are not significantly different at $P < 0.05$.

§ The *P* values and R^2 values are for simple linear regression of each parameter with cumulative N_2O emissions.

and the influence of N_2O emission spikes reported for both field and laboratory experiments (Clayton et al., 1997; Harrison-Kirk et al., 2013; Ryden, 1983). The highest emissions from the historically fertilized treatments (WT-N, FA-N) were of the same order of magnitude as those reported elsewhere for historically fertilized samples that received no N inputs during the experiment (Guo et al., 2011) but were approximately one order of magnitude lower than those reported for historically unfertilized samples supplemented with nitrate (Drury et al., 1998) (Fig. 2).

Our results show that soils from a wheat–fallow rotation with historical inorganic N inputs over the past 20 yr (WT-N, FA-N) had significantly higher cumulative N_2O emissions during a laboratory incubation than soils from the wheat–fallow rotation with no historical N fertilization for more than 20 yr (WT-O, FA-O), regardless of the phase of the rotation (Fig. 3). Because none of the treatments received N fertilizer in the year before our experiment (equivalent to zero-N control plots), this difference in emissions provides clear evidence of a residual effect of historical fertilizer N inputs and corroborates our original hypothesis. Similar to our findings, Graham et al. (2013) showed that a soil with historical inputs of manure had higher N_2O emissions than a soil without historical manure application. However, the simultaneous addition of N and organic C associated with manure inputs in their study made it difficult to evaluate the effect of N fertilization alone on N_2O emissions.

Our findings suggest that historical N inputs alone (in the absence of external C inputs) can play an important role in driving N_2O emissions possibly due to their effect on both soil inorganic N and organic N pools. For example, the low extractable N levels in the wheat–legume (WT-L) treatment were similar to the wheat phase of the historically unfertilized wheat–fallow rotation (WT-O), but the relatively high PMN levels in the WT-L treatment were on par with the wheat phase of the historically fertilized wheat–fallow rotation (WT-N); this resulted in cumulative N_2O emissions that were intermediate between the historically fertilized and historically unfertilized treatments (Fig. 3; Table 1). Cumulative N_2O emissions showed a stronger positive correlation with extractable N levels ($R^2 = 0.563$; $P = 0.001$) than with PMN levels ($R^2 = 0.407$; $P = 0.011$), although this was principally due to differences between phases of the crop rotation (Fig. 4; Table 1). Despite receiving the same historical fertilization (Fig. 1), soil extractable N levels and N_2O emissions were lower in the treatments entering the fallow phase than in those entering the wheat phase (Fig. 3; Table 1), possibly due to immobilization of available soil N from the incorporation of C-rich wheat straw residues before the fallow phase (e.g., Rizhiya et al., 2011). Available inorganic N is an important control on N_2O emissions and may explain why the two treatments entering the fallow phase of the rotation (FA-N and FA-O) both had lower N_2O emissions than their respective counterparts entering the wheat phase (WT-N and WT-O) (Fig. 3).

Whereas extractable N levels were related to the cropping cycle phase, higher PMN levels and the trend toward higher total soil N under the WT-N, FA-N, and WT-L treatments corresponded better to historical N inputs (Table 1). Drury et al. (1998) found that increased organic C and N from long-term N fertilization can result in enhanced denitrification capacity, although their experiment included supplemental N fertilization in all treatments and thus studied the indirect effects of long-term N fertilization on N_2O emissions rather than the direct effect of increased background soil organic and inorganic N. The treatments with historical N inputs (WT-N, FA-N, WT-L) generally had higher total soil C and N levels (although not significant) than the historically unfertilized treatments (WT-O, FA-O). Furthermore, historical N inputs and the resulting higher biomass inputs under the WT-N, FA-N, and WT-L treatments corresponded to higher PMN levels (Table 1). Although cumulative N_2O emissions were not significantly correlated with either total C or N, there was a significant positive correlation with PMN levels (Table 1). The higher PMN levels may explain why the fallow phase of the historically fertilized wheat–fallow rotation (FA-N) had higher cumulative N_2O emissions than the wheat phase of the historically unfertilized wheat–fallow rotation (WT-O) despite lower extractable N levels (Fig. 3; Table 1). Historical fertilization may lead to higher cumulative N_2O emissions by altering a variety of soil properties; the composition of microbial communities, for example, has been shown to change with historical fertilization (Enwall et al., 2005; Parham et al., 2003). However, the correlations presented here suggest that both soil inorganic N and labile organic N, as evidenced by PMN, may play a role in the observed residual effect of historical fertilization on N_2O emissions.

Implications of N Fertilization History for Fertilizer-Induced Emissions

The current standard for estimating N_2O emissions at the country-scale is the IPCC Tier 1 approach, which accounts for all N inputs to agriculture and uses the 1% fertilizer-induced EF discussed previously (IPCC, 2006). During the early stages of development for the IPCC Tier 1 approach, it was suggested that global emissions estimates must include N_2O emissions from all possible N sources: mineralization from land use change, atmospheric deposition, subsurface aquifers, crop residues, current fertilization, and fertilization in past years (Mosier et al., 1996). Although there have been questions about the relative magnitude of emissions from each of these N sources (e.g., indirect emissions in Turner et al. [2015]), the IPCC Tier 1 approach at least nominally accounts for most of these N sources as direct and indirect N_2O emissions (IPCC, 2006). However, we suggest that it fails to address N inputs from fertilization in past years and only partially accounts for historical N fertilization by incorporating N from crop residue (which would be higher with historical N fertilization).

Historical management has been shown to have a significant impact on N_2O emissions in long-term studies (e.g., Adviento-Borbe et al., 2010; Drury et al., 2014). Our results, in combination with the results discussed previously from other laboratory studies based on long-term experiments (e.g., Drury et al., 1998; Graham et al., 2013), emphasize that N fertilization history can significantly affect N_2O emissions from agricultural soils independent of current N fertilization practices. The higher levels of both extractable N and PMN in the historically fertilized systems imply that larger N pools persisted for at least 2 yr after N fertilization, and the correlation with higher N_2O emissions suggests that these larger pools of available N and readily mineralizable N can lead to higher emissions (Fig. 4; Table 1). Although it is not clear if the results from our study and the other laboratory experiments

mentioned above translate to a field setting, a recent field experiment by Wilson et al. (2015) found that historical fertilization at high levels can have a residual effect on N_2O emissions, such that a single year with no N inputs does not reduce emissions to background levels. This residual effect of past N fertilization may in part explain two notable issues regarding the fertilizer-induced EF: (i) the uncertainty associated with this EF is considerable, with a range of −70 to +200% (Crutzen et al., 2008; IPCC, 2006), and (ii) initial bottom-up estimates of N_2O emissions did not agree well with top-down global N budget methods based on the stratospheric N_2O sink and the accumulation of N_2O in the atmosphere (Crutzen et al., 2008; Mosier et al., 1998).

Numerous studies have suggested methods to improve the accuracy of N_2O emissions estimates and reduce the corresponding uncertainty (Leip et al., 2011; Lesschen et al., 2011; Li et al., 2001; Philibert et al., 2012). Historical N fertilization has been recognized as an important source of variability in N_2O emissions (Bouwman, 1996), and our results confirm that, in a laboratory setting at least, historical inorganic N inputs can have a significant effect on N_2O emissions. Nonetheless, it is now clear that the studies on which the fertilizer-induced EF is based used a mix of historically fertilized and historically unfertilized zero-N control plots (Bouwman, 1996; Bouwman et al., 2002a, 2002b; Stehfest and Bouwman, 2006). As previously mentioned, we attempted to determine the fertilization history for a random sample of 10% of the published field measurements of N_2O emissions under zero-N application in the Stehfest and Bouwman (2006) dataset; based on this analysis, we concluded that, for sites whose fertilization history was explicitly described or could be reasonably inferred, over two thirds had in fact received fertilizer N inputs 1 to 2 yr before the experiments used to determine the fertilizer-induced EF. Attempts to reduce the uncertainty associated with the fertilizer-induced EF have not, to our knowledge, addressed this variability in fertilization history.

Another potential issue with the fertilizer-induced EF used in the IPCC Tier 1 approach is that initial bottom-up estimates of N_2O emissions appeared to be underestimated when compared to top-down estimates (Mosier et al., 1998; Crutzen et al., 2008). Various studies have improved agreement by incorporating previously neglected N_2O sources (Crutzen et al., 2008; Davidson, 2009; Smith et al., 2012), and, given the large uncertainty range for the fertilizer-induced EF, these two methods are generally comparable (Del Grosso et al., 2008). However, indirect N_2O emissions from soils still appear to be underestimated by the IPCC bottom-up approach (Smith et al., 2012). Turner et al. (2015) suggested that an underestimation of indirect riverine emissions may account for this discrepancy. They point out that estimates of direct N_2O emissions from soil are well constrained (Turner et al., 2015), but these estimates only account for N_2O emissions from fertilizer N during the first year after application (Mosier et al., 1998). As such, a residual effect of past N fertilization may offer a complementary explanation for the underestimation of N_2O emissions from agricultural soils; indeed, this type of underestimation was predicted early on in the development of the fertilizer-induced EF (Mosier et al., 1996). As more research on the residual effect of past N fertilization on N_2O emissions emerges, those findings may need to be considered in the calculation of the fertilizer-induced EF. Given the potential for such a residual effect on N_2O emissions and the variable N fertilization histories used to calculate the EF (Stehfest and Bouwman, 2006), time since fertilization could be an important covariate in studies used to calculate the fertilizer-induced EF, although analyzing this continuous relationship was beyond the scope of our study. Furthermore, accounting for N fertilization history may help reduce the uncertainty associated with the fertilizer-induced EF, improve agreement of different methods for estimating N_2O emissions, and more accurately assess the contribution of agriculture to anthropogenic GHG emissions.

Conclusions and Recommendations for Future Research

Soils with a 20-yr N fertilization history had substantially higher cumulative N_2O emissions during a 10-d incubation than soils from the same site with no historical N inputs for more than 20 yr. This increase in N_2O emissions appears to be related to differences in available and readily mineralizable N that are independent of the N applied during the current growing season, resulting in a residual effect of past N fertilization on N_2O emissions. Although this finding is important as a proof of concept, these laboratory results need to be verified under field settings. These results can lay the groundwork for research in long-term field experiments to evaluate the residual effects of historical fertilization on background N_2O emissions. The main objective of our research was to evaluate the effect of historical N fertilization on the baseline emissions from the zero-N control plots used in the calculation of fertilizer-induced emissions. Further field research will be critical to determine how fertilization history affects fertilizer-induced emissions themselves and to evaluate the relative contributions of historical and current season N fertilization to total fertilizer-induced N_2O emissions. Lastly, given that our initial analysis revealed variable fertilization histories in the studies behind the IPCC fertilizer-induced EF, we believe that an in-depth literature review or a meta-analysis analyzing the effect of time since fertilization on N_2O emissions would provide valuable insight into the relative importance of this variable for the estimation of agricultural N_2O emissions.

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