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Does Plant Biomass Manipulation in Static Chambers Affect Nitrous Oxide Emissions from Soils?

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Abstract

One of the most widespread approaches for measurement of greenhouse gas emissions from soils involves the use of static chambers. This method is relatively inexpensive, is easily replicated, and is ideally suited to plot-based experimental systems. Among its limitations is the loss of detection sensitivity with increasing chamber height, which creates challenges for deployment in systems including tall vegetation. It is not always possible to avoid inclusion of plants within chambers or to extend chamber height to fully accommodate plant growth. Thus, in many systems, such as perennial forages and biomass crops, plants growing within static chambers must either be trimmed or folded during lid closure. Currently, data on how different types of biomass manipulation affect measured results is limited. Here, we compare the effects of cutting vs. folding of biomass on nitrous oxide measurements in switchgrass (*Panicum virgatum* L.) and alfalfa (*Medicago sativa* L.) systems. We report only limited evidence of treatment effects during discrete sampling events and little basis for concern that effects may intensify over time as biomass manipulation is repeatedly imposed. However, nonsignificant treatment effects that were consistently present amounted to significant overall trends in three out of the four systems studied. Such minor disparities in flux could amount to considerable quantities over time, suggesting that caution should be exercised when comparing cumulative emission values from studies using different biomass manipulation strategies.

Core Ideas

- Biomass manipulation infrequently affects nitrous oxide emission.
- Effects of biomass manipulation on emissions may vary by system.
- Effects of biomass manipulation on emissions do not appear to intensify with time.
- Considered collectively, minor treatment effects may amount to significant trends.
- Biomass presence has a small but significant effect on volume and flux estimation.

Measurement of greenhouse gas (GHG) flux from soils in cropping and biomass production systems is essential to understanding the impact of current and future management strategies on climate change. Nitrous oxide (N_2O), a potent GHG, accounts for approximately 7% of total global warming, with approximately 60% of anthropogenic N_2O arising from agricultural sources (Smith et al., 2007; Reay et al., 2012; Myhre et al., 2013). One of the most commonly used methods for measurement of GHGs including N_2O involves the use of static chambers, in which the headspace above a small (generally <0.5 m^2) area of ground is enclosed and buildup in gas concentration over time is used to calculate a rate of flux. This method has the advantage of being relatively inexpensive and easy to replicate (Parkin and Venterea, 2010; Clough et al., 2013; Collier et al., 2014). Additionally, because the area measured is small compared with that measured with other approaches (e.g., eddy covariance), it is ideally suited to plot-based experimental systems. The static chamber method has limitations as well. One of its limitations involves the handling of plant biomass within the measurement area, and this is the focus of this study.

Because increasing headspace volume relative to measurement area will result in lower concentrations for gasses emitted from the plant–soil system, maximum chamber height is limited by detection sensitivity and will typically be in the range of 15 to 40 cm depending on deployment time (i.e., the time over which gas concentration buildup is monitored) (Rochette and Eriksen-Hamel, 2008; Parkin and Venterea, 2010; Clough et al., 2013). The limited height range creates obvious challenges to conducting chamber-based gas measurements in systems with taller plants, and there are three common strategies for addressing the issue: (i) in row crops with wide spacing such as corn (*Zea mays* L.), chambers may be placed between rows and at varying proximity to plant shoots such that plants themselves are excluded from the measured area; (ii) plants growing within the measurement area may be removed, trimmed, or folded to fit within the chamber; and (iii) chambers may be extended to accommodate plant height, with longer deployment times used to compensate for increased volume. There are tradeoffs inherent to each of these strategies: omitting plant rows does not allow for

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Abbreviations: GHG, greenhouse gas; RMD, relative mean difference; VWC, volumetric water content.

fully representative sampling, manipulation of plants may alter the system being studied, and the height to which chambers can be extended is limited, with increasing volume also generating other issues related to homogeneity of gas concentrations within the headspace (Clough et al., 2013).

In perennial forage and biomass production systems, options (i) and (iii) above are often infeasible due to the nature of the system. For instance, row spacing in alfalfa may be as little as 20 cm, effectively excluding the possibility of placing chambers between rows. In switchgrass stands, where plant height may exceed 2 m, chamber extension becomes impractical. Similar issues are encountered in mixed plantings such as pasture or prairie systems. Thus, manipulation of biomass within the measurement area is a common choice for perennial crops, and it is important to consider how different types of manipulation may affect the area under study, particularly when manipulations are repeatedly applied over the course of a season or study with the potential to affect plant physiology and soil microclimate and/or ecology.

In cropping systems, N_2O is predominantly emitted as a product of soil-based microbial nitrification and denitrification (Maag and Vinther, 1996; Mosier et al., 1998). There are also indications, however, that plants can facilitate N_2O emission from the soil via transpiration (Chang et al., 1998) and that under physical stresses, such as those associated with grazing or clipping, plants release root exudates that stimulate microbial conversion of N and concomitant N_2O emission (Jackson et al., 2008). In addition, there is some evidence of entirely plant-based production of N_2O (Smart and Bloom, 2001). Given these multiple modes of interaction between plants and N_2O fluxes to the atmosphere, it is important to understand how or whether plant biomass manipulation affects N_2O emissions estimated as part of chamber-based GHG measurements. Here, we examine the effects of biomass cutting vs. folding on N_2O emissions in perennial switchgrass and alfalfa cropping systems.

Materials and Methods

Site

The study was conducted at the Wisconsin Integrated Cropping System Trial within the University of Wisconsin–Madison's Arlington Agricultural Research Station (43°18′ N, 89°20′ W). Soil at the site is a highly productive Mollisol classified as Plano silt loam (fine-silty, mixed, superactive, mesic Typic Argiudoll). The soil developed under tallgrass prairie in loess over glacial till and is well drained with minimal slope (Sanford et al., 2012). Average annual temperature and precipitation for Arlington are 6.9°C and 869 mm, respectively (National Centers for Environmental Information, 2015).

Experimental Design

Switchgrass experiments were conducted during the 2011 growing season. Experimental plots were established in 2007 in a randomized complete block design consisting of three N application rate systems (0, 50, and 150 kg ha^{-1}) in six replicate blocks. Fertilizer was applied annually in spring as ammonium nitrate, with a 9 June application date in 2011. Plots measured 2.13 × 4.57 m. All plots were planted with the variety Forestburg and were harvested to 10 cm in November of each year. Two greenhouse gas measurement chambers were installed in each plot, at least 0.5 m away from the plot's edge, and each chamber was randomly assigned to either "Cut" or "Fold" treatments such that each plot contained one chamber under each biomass manipulation treatment.

Alfalfa experiments were conducted during the 2013 growing season in three replicate plots that were seeded with variety Pioneer 54Q32 in 2012. These plots were part of the long-term conventional dairy forage Wisconsin Integrated Cropping System Trial rotation consisting of 1 yr of corn followed by 3 yr of alfalfa. This rotation receives fertilizer in the form of liquid dairy manure in the fall after harvest of corn and third-year alfalfa phases. Thus, the study plots had not received fertilizer since the fall of 2011. Plots measured 155 m north-south × 18 m east-west. Two chambers were installed in each plot, centered east-west and at least 6 m from the edge north-south, and were randomly assigned to either Cut or Fold treatments such that each plot contained one chamber under each biomass manipulation treatment. Alfalfa was harvested to 10 cm on 19 June, 17 July, and 27 Aug. 2013.

N_2O Estimation and Biomass Manipulation

Nitrous oxide sample collection and analysis was performed essentially as described in Collier et al. (2014). In brief, fluxes were measured using vented static chambers (Livingston and Hutchinson, 1995) consisting of bases and lids constructed of high-density polyethylene (switchgrass) or galvanized steel (alfalfa). Chambers used in switchgrass had a diameter of 27 cm and were inserted 5 cm into the soil with 17 cm extending above the soil surface. Flat lids equipped with butyl-rubber gaskets along the contact edge were sealed on the tops of the chambers during measurement. Chambers used in alfalfa had a diameter of 40.5 cm. Bases were inserted 5 cm into the soil with 2.5 cm extending above the soil surface. Lids with a height of 10 cm were sealed on the tops of the bases at the time of measurement. For both chamber types, lids included a septum for sample collection and a vent with an interior coil of tygon tubing to allow for pressure equilibration and to minimize wind-induced changes in gas concentration within the chamber. Vent tubing had an inner diameter of 0.165 cm and length of 15 cm for chambers used in switchgrass and an inner diameter of 0.35 cm and length of 45 cm for chambers used in alfalfa. After lid closure, samples were collected by syringe at 0, 20, and 40 min for switchgrass and at 1, 12, 24, and 36 min for alfalfa. Two ambient samples were also averaged for approximation of initial concentration in alfalfa. Glass 5.9-mL Exetainer vials (Labco Limited) were overcharged with 10 mL of gas sample and transported to the University of Wisconsin–Madison for analysis by gas chromatography using an electron capture detector (micro-ECD, 7890A GC System, Agilent). Fluxes were calculated using both linear (Holland et al., 1999) and quadratic (Wagner et al., 1997) regression and with the revised Hutchinson/Mosier method (Pedersen et al., 2010). Because all three models yielded largely similar trends and levels of significance (data not shown), calculations based on the linear model were selected for presentation and further analysis throughout for the sake of consistency and to maximize the detection of relative differences between treatments (Venterea et al., 2009). In alfalfa, within-chamber biomass volume was approximated (see next section) and subtracted from total chamber volume during flux calculation.

Sampling was conducted in June, August, September, and October for switchgrass (10 sampling events total) and in August through October for alfalfa (nine sampling events total). In alfalfa, chamber bases were removed before harvest and reinstalled approximately 3 m away after harvest. In Fold treatments for both crops, plants whose height exceeded that of the chamber base and lid were gently folded to fit inside during measurement and were unfolded once the lid was removed. Before each sampling event in the switchgrass Cut treatment, any plants growing inside the chamber whose height exceeded that of the chamber were trimmed to the height of the chamber walls before attachment of the lid. In the alfalfa Cut treatment, plants were trimmed to a height of 10 cm (similar to harvest) whenever their height inside the chamber before sampling exceeded 19 cm, the height above which plants were visibly damaged by folding to fit into chambers. Alfalfa Cut treatments received four such induced trimmings during the study period on 1 August, 23 August, 13 September, and 7 October.

Biomass and Soil Measurement

A 0.6- × 2.1-m patch adjacent to each alfalfa chamber was maintained with the same trimming treatments as within the chamber. At each sampling event, three measurements of alfalfa standing height were made at random points within the plot or the trimmed patch for the Fold and Cut treatments, respectively. Within-chamber biomass volume was also approximated on each alfalfa sampling date by placing an extra chamber base within the trimmed or untrimmed patch adjacent to the chamber (according to treatment) and harvesting all of the interior biomass down to a stubble of <1 cm. Harvested biomass volume was measured by water displacement. Soil temperature and volumetric water content (VWC) were also measured during the time in which gas samples were collected. In switchgrass, soil temperature was measured using data loggers (Pendant Temperature Data Logger, Onset Computer Corp.) buried approximately 10 cm under the chambers, and VWC was measured immediately adjacent to the chambers using a time domain reflectometer (FieldScout 300, Spectrum Technologies, Inc.). In alfalfa, soil temperature was measured using a 15-cm probe (Checktemp 1C, Hanna Instruments), and soil VWC was measured by time domain reflectometry within 2 m in the adjacent trimmed or untrimmed patch. The average of two measurements for soil probe–based temperature and three measurements for VWC was recorded.

Statistical Analysis

The effect of biomass manipulation on daily fluxes and ancillary soil and biomass values was analyzed using a paired *t* test. Cumulative emissions over the course of the study period were estimated by linear interpolation and analyzed with a paired *t* test. Additionally, the relative mean difference (RMD) in flux between biomass manipulation treatments was calculated for each combination of date × system as $[2\times(\overline{x}_{dC}-\overline{x}_{dF})]/(\overline{x}_{dC}+\overline{x}_{dF})$, where $\overline{x}_{dC}$ represents the daily average flux of Cut, and $\overline{x}_{dF}$ represents the daily average flux of Fold. Daily RMD values were analyzed collectively for each system by calculating the mean and 95% confidence interval of the distribution based on a two-tailed *t* test to evaluate overall trends. The Wilcoxon signed-rank test was used to determine the statistically significant difference of the mean RMD from zero, with each system treated as a population of RMD values, each corresponding to a different sampling date. A significance threshold of $\alpha = 0.1$ was used throughout to minimize the likelihood of Type II errors. All statistical analyses were performed in JMP v.11 (SAS Institute, 2013).

Results and Discussion

N_2O Flux

Out of the 39 sampling events across the four systems studied, there were five instances in which biomass manipulation had a statistically significant effect on N_2O flux (Fig. 1), only slightly

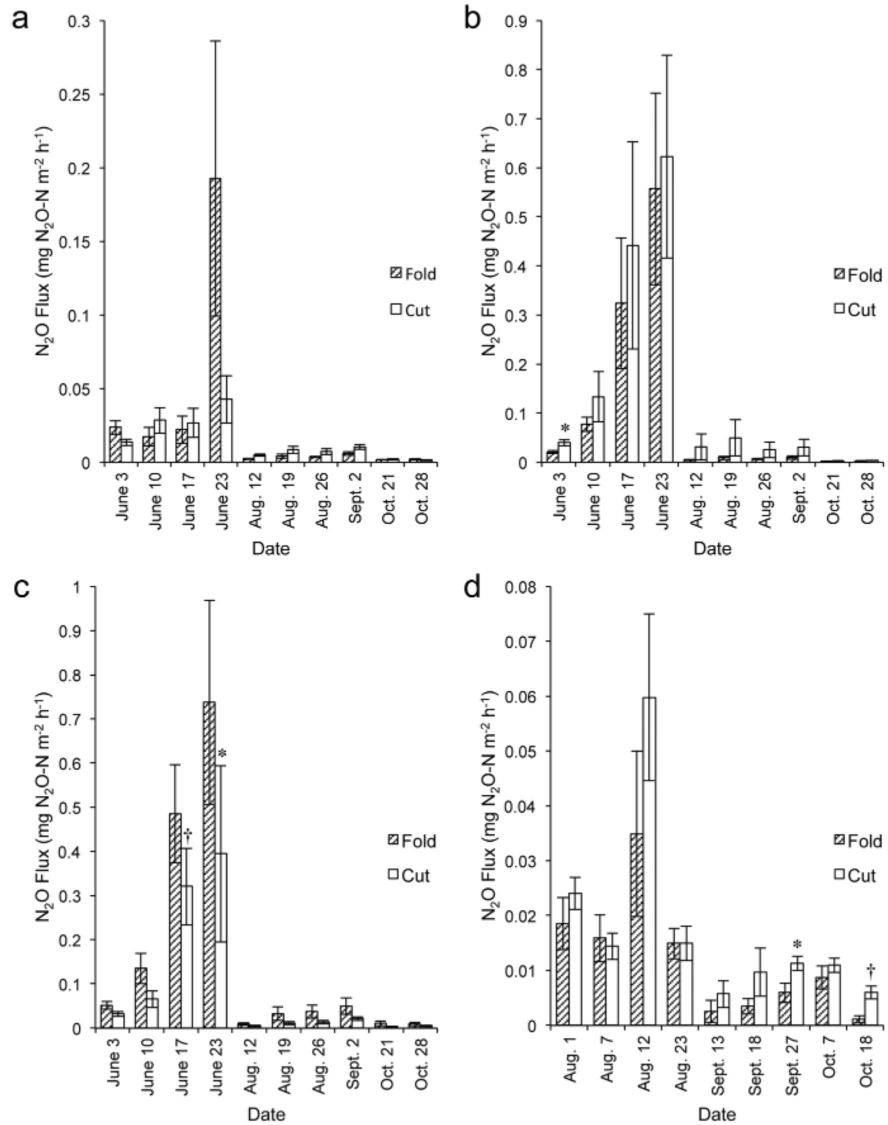

Fig. 1. Impact of biomass manipulation by date on N_2O flux in (a) switchgrass receiving 0 kg N ha^{-1}, (b) switchgrass receiving 50 kg N ha^{-1}, (c) switchgrass receiving 150 kg N ha^{-1}, and (d) alfalfa in a dairy forage rotation. Switchgrass dates are in 2011; alfalfa dates are in 2013. Error bars represent SE (switchgrass, n = 6; alfalfa, n = 3). *Significant difference from the Cut treatment for the a = 0.05 level. †Significant difference from the Cut treatment for the a = 0.1 level.

higher than the 3.9 instances of Type 1 error that would be expected at $\alpha = 0.1$. In three of the five cases, flux in the Cut treatment was higher than in the Fold treatment, with the opposite relationship found in the remaining two cases. These treatment effects were not observed in any particular part of the growing season. There was little evidence, therefore, of accumulating impact of biomass manipulation over time. In alfalfa, where a significant treatment effect was observed at the end of the season (18 October) (Fig. 1d), chambers had been removed and reinstalled in different locations after the 27 August harvest, so any accumulated treatment impact would have been from only the preceding 35 d. The same phenomenon was not observed for the set of sampling events before the 27 August harvest. Intriguingly, 18 October was the only date on which plant height appeared to decline somewhat in the Fold treatment, possibly a result of minor lodging and/or senescence (Fig. 2a). This is in contrast to the active growth observed in the Cut treatment at this time and raises the possibility that enhanced or prolonged plant growth, stimulated by the Cut treatment, may play a role in the higher rates of N_2O flux periodically observed in Cut treatments. Plant growth also appears to have been stimulated by the Cut treatment in switchgrass: during the early summer period of active vegetative growth before reaching standing peak, plant height was less in the Cut than in the Fold treatment (averaging 34 and 40 cm, respectively) (Fig. 3a), but the Cut treatment included weekly trimmings back to 17 cm, and the heights therefore indicate vigorous regrowth. Defoliation is also known to stimulate short-term root C exudation, which in turn stimulates rhizospheric N mineralization (Hamilton et al., 2008). Long-term grazing has been found to stimulate both nitrification and denitrification (Le Roux et al., 2003), whereas Jackson et al. (2015) found that rotational grazing stimulated N_2O emissions for several days after a grazing treatment. Thus, it is not unusual that biomass cutting should, at least in some cases, result in increased N_2O flux.

Examining all sampling events collectively, there was a moderate trend toward higher N_2O emissions in Cut than in Fold treatments in the switchgrass 50N (i.e., 50 kg N ha^{-1}) and alfalfa systems ($p < 0.01$ and $p < 0.05$, respectively), with an opposite trend observed in the switchgrass 150N system ($p < 0.01$) and no significant treatment effect in the switchgrass 0N system (Fig. 4). Even where significant differences in flux were not detected for discrete sampling events, the significant trends observed in three out of the four systems studied suggest a consistent, if minor, effect of biomass manipulation, which could become relevant particularly in cases where measurements over the course of a study or season are used to calculate total emissions, as in Table 1. Although this study was not designed for calculation of cumulative emissions, estimates for the study period can be seen to diverge widely based on biomass manipulation treatment in some cases. The lack of a consistent trend among systems, however, suggests that there may not be a single overarching effect of biomass manipulation on N_2O flux and that multiple factors are more likely involved.

It is striking that the three switchgrass systems each responded differently. This may indicate an interaction between biomass manipulation response and N availability, similar to the N-responsive plant-mediated flux observed by Chen et al. (1999). It is also possible that a high N fertilization rate led to

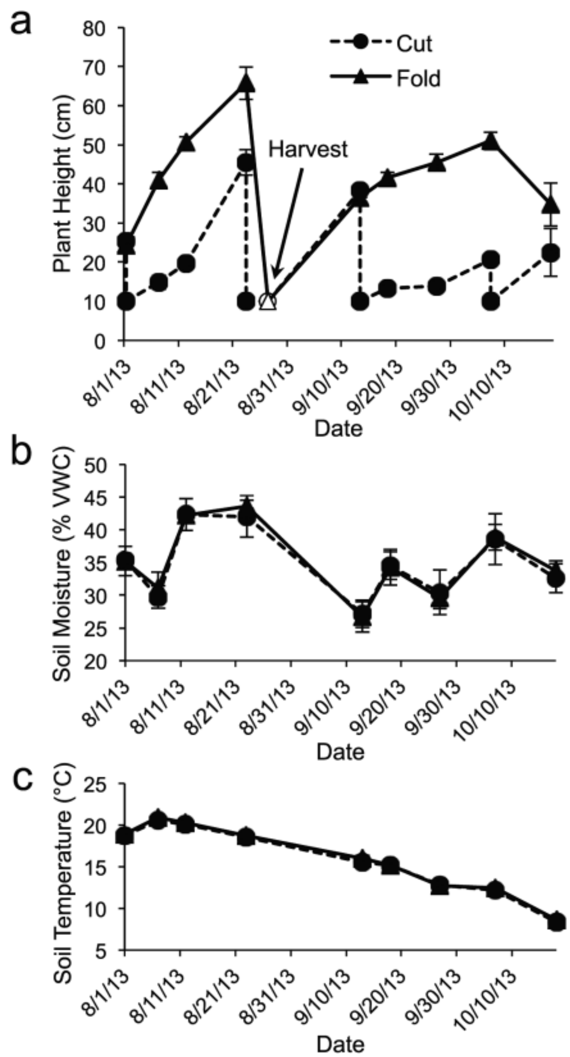

Fig. 2. Impact of biomass manipulation on (a) plant height, (b) soil volumetric water content (VWC), and (c) soil temperature in alfalfa. Unfilled points in (a) represent height estimated based on reported harvest height rather than measured values. Error bars represent SE (n = 3).

denser foliage, which would reduce the effective chamber headspace volume and elevate gas concentrations. Such a phenomenon would skew flux calculations and could at least partially account for the trend of higher flux in the Fold compared with the Cut treatment observed in the 150N system. Although switchgrass biomass volume was not measured as a part of this study, the effect of plant biomass on flux calculations was found to be non-negligible in the alfalfa system (see next section) and is thus a reasonable consideration.

Ancillary Measures

Our treatments did not significantly alter soil temperature or moisture in any of the systems studied (Fig. 2b, 2c, 3b, 3c), which was not surprising considering that soil measurements were taken immediately after any trimming was imposed and thus would have largely reflected the soil's prior condition. Because sampling dates were spaced at approximately weekly intervals, the soil was always covered with vigorous growth or regrowth leading up to sampling and measurement events. The lack of difference in soil temperature and moisture between treatments suggests that

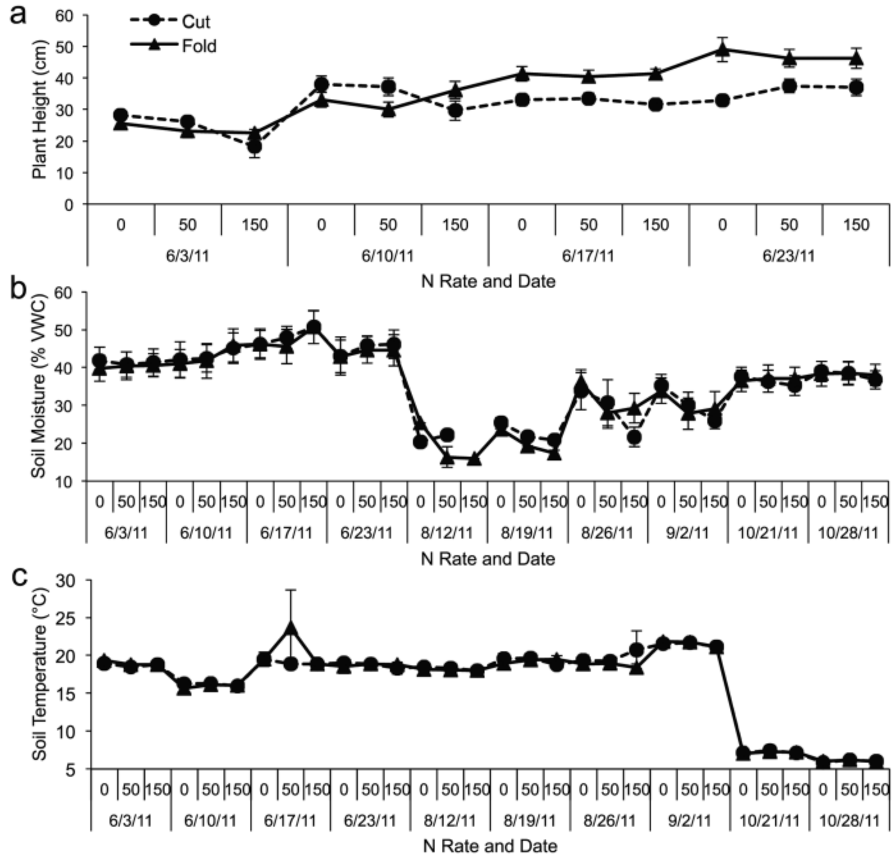

Fig. 3. Impact of biomass manipulation on (a) plant height, (a) soil volumetric water content (VWC), and (c) soil temperature in switchgrass receiving three different N rates (0, 50, and 150 kg N ha^{-1}). Error bars represent SE (*n* = 6).

where treatment effects were observed, they were more likely the result of alterations in soil nutrient balance or biological activity rather than changes in soil physical parameters.

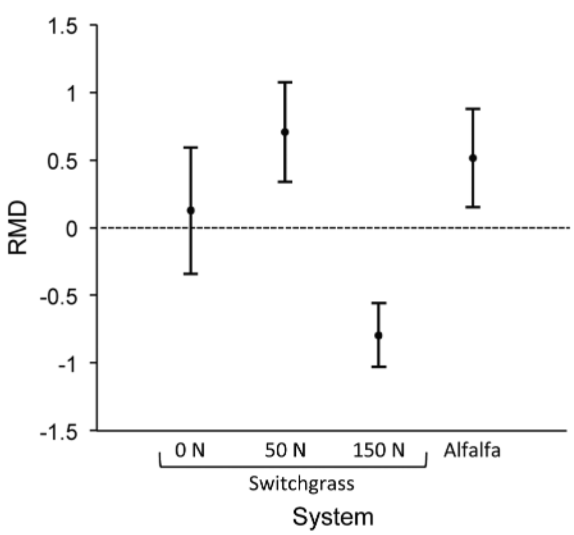

Fig. 4. Relative mean difference (RMD) between Cut and Fold treatments expressed as a fraction of the combined average. All sampling events for each system were included in the analysis (switchgrass systems, *n* = 10; alfalfa, *n* = 9). The dashed line marks an RMD of 0. Positive values represent higher flux under the Cut treatment; negative values represent higher flux under the Fold treatment. Error bars depict the 95% confidence interval of the mean. 0 N, 50 N, and 150 N refer to 0, 50, and 150 kg N ha^{-1}, respectively.

Over the course of the study, alfalfa biomass volume ranged from 0.6 to 1.1% and from 1.4 to 2.2% of chamber volume in Cut and Fold treatments, respectively. Accounting for within-chamber biomass volume led to an increase in the calculated flux rate (compared with calculations based on unadjusted chamber volumes) averaging 0.7 and 1.7% for Cut and Fold treatments, respectively. This small but significant ($p < 0.0001$) difference between biomass manipulation treatments highlights the importance of using consistent approaches to handling and accounting for plant biomass within static chambers. Measurement of plant biomass is time consuming, and in practice it may not be feasible to take such measurements each day of a GHG sampling campaign, particularly where plot sizes are small and the area available for biomass harvesting is limited. To facilitate accurate comparisons between studies, however, it would be prudent to at least indicate whether or not biomass is present within chambers and to allow for rough estimation based on a height-to-volume comparison. More extensive investigation of the effect of plant biomass on effective chamber volume and other aspects of N_2O dynamics within chambers also seems warranted.

At the conclusion of the study, alfalfa dry mass within the Fold treatment chambers was found to be 78% that of unmanipulated portions of the plot ($p < 0.05$). This finding serves as a reminder that although the impacts of biomass trimming appear generally to be more severe, folding also has the potential to affect plant growth.

Conclusions

Overall, we found only limited evidence of a strong day-to-day effect of biomass manipulation on N_2O flux in switchgrass and alfalfa and little basis for concern related to an intensifying effect over time. However, minor but consistent day-to-day differences between treatments amounted to significant trends in three out of four systems. Taken collectively over a long sampling period, such minor differences could amount to considerable

Table 1. Cumulative emissions.

System	Treatment		*p* value
	Fold	Cut	
	—— kg N_2O–N ha^{-1}† ——		
Switchgrass			
0 kg N ha^{-1}	0.24	0.18	0.5
50 kg N ha^{-1}	1.14	1.73	0.2
150 kg N ha^{-1}	2.05	0.98	0.02
Alfalfa	0.21	0.30	0.3

† Cumulative emissions are calculated for the study period, which for switchgrass is defined as 3–23 June plus 2 Sept.–28 Oct. 2011 and for alfalfa as 1 Aug.–18 Oct. 2013.

quantities, and caution should therefore be exercised in comparing cumulative emission values between studies using different biomass manipulation strategies. Although the majority of systems studied suggest some tendency toward increased N_2O flux with biomass trimming, not all were in agreement. Response to biomass trimming may vary between species and may also have been influenced by fertilizer regimes. Examination of additional species and mixed-species systems under varying conditions would help to clarify the sources of differential response to biomass manipulation. Finally, we noted a small but significant effect on calculated flux based on adjusting for within-chamber plant volume. Because of this observation, it is suggested that the presence or absence of biomass within chambers should be consistently reported, and biomass volume should be estimated whenever possible.

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References

Chang, C., H.H. Janzen, E.M. Nakonechny, and C.M. Cho. 1998. Nitrous oxide emission through plants. Soil Sci. Soc. Am. J. 62:35–38. doi:10.2136/sssaj1998.03615995006200010005x

Chen, X., P. Boeckx, S. Shen, and O. Van Cleemput. 1999. Emission of N_2O from rye grass (*Lolium perenne* L.). Biol. Fertil. Soils 28:393–396. doi:10.1007/s003740050510

Clough, T.J., P. Rochette, S.M. Thomas, M. Pihlatie, J.R. Christiansen, and R.E. Thorman. 2013. Chamber design. In: C. de Klein and M. Harvey, editors, Nitrous oxide chamber methodology guidelines. Ministry for Primary Industries, Wellington, UK. p. 19–33.

Collier, S.M., M.D. Ruark, L.G. Oates, W.E. Jokela, and C.J. Dell. 2014. Measurement of greenhouse gas flux from agricultural soils using static chambers. J. Vis. Exp. 90:E52110. doi:10.3791/52110

Hamilton, E.W., III, D.A. Frank, P.M. Hinchey, and T.R. Murray. 2008. Defoliation induces root exudation and triggers positive rhizospheric feedbacks in a temperate grassland. Soil Biol. Biochem. 40:2865–2873. doi:10.1016/j.soilbio.2008.08.007

Holland, E.A., G.P. Robertson, J. Greenberg, P.M. Groffman, R.D. Boone, and J.R. Gosz. 1999. Soil CO2, N2O, and CH4 exchange. In: G.P. Robertson, D.C. Coleman, C.S. Bledsoe, and P. Sollins, editors, Standard soil methods for long-term ecological research. Oxford Univ. Press, New York. p. 185–201.

Jackson, L.E., M. Burger, and T.R. Cavagnaro. 2008. Roots, nitrogen transformations, and ecosystem services. Annu. Rev. Plant Biol. 59:341–363. doi:10.1146/annurev.arplant.59.032607.092932

Jackson, R.D., L.G. Oates, W.H. Schacht, T.J. Klopfenstein, D.J. Undersander, M.A. Greenquist, M.M. Bell, and C. Gratton. 2015. Nitrous oxide emissions from cool-season pastures under managed grazing. Nutr. Cycling Agroecosyst. 101:365–376. doi:10.1007/s10705-015-9685-6

Le Roux, X., M. Bardy, P. Loiseau, and F. Louault. 2003. Stimulation of soil nitrification and denitrification by grazing in grasslands: Do changes in plant species composition matter? Oecologia 137:417–425. doi:10.1007/s00442-003-1367-4

Livingston, G.P., and G.L. Hutchinson. 1995. Enclosure-based measurement of trace gas exchange: Applications and sources of error. In: P.A. Matson and R.C. Harris, editors, Biogenic trace gases: Measuring emissions from soil and water. Blackwell Science Ltd., Oxford, UK. p. 14–51.

Maag, M., and F.P. Vinther. 1996. Nitrous oxide emission by nitrification and denitrification in different soil types and at different soil moisture contents and temperatures. Appl. Soil Ecol. 4:5–14. doi:10.1016/0929-1393(96)00106-0

Mosier, A., C. Kroeze, C. Nevison, and O. Oenema. 1998. Closing the global N_2O budget: Nitrous oxide emissions through the agricultural nitrogen cycle. Nutr. Cycling Agroecosyst. 52:225–248. doi:10.1023/A:1009740530221

Myhre, G., D. Shindell, F.-M. Bréon, W. Collins, J. Fuglestvedt, J. Haung, et al. 2013. Anthropogenic and natural radiative forcing. In: T.F. Stocker, D. Qin, G.-K. Plattner, M. Tignor, S.K. Allen, J. Boschung, et al., editors, Climate change 2013: The physical science basis. Cambridge Univ. Press, Cambridge, UK and New York. p. 659–740.

National Centers for Environmental Information. 2015. Climate data online. NOAA. www.ncdc.noaa.gov/cdo-web/datatools/ (accessed 9 June 2015).

Parkin, T.B., and R.T. Venterea. 2010. Chamber-based trace gas flux measurements. In: R.F. Follett, editor, Sampling protocols. www.ars.usda.gov/research/GRACEnet (accessed 5 July 2015).

Pedersen, A.R., S.O. Petersen, and K. Schelde. 2010. A comprehensive approach to soil-atmosphere trace-gas flux estimation with static chambers. Eur. J. Soil Sci. 61:888–902. doi:10.1111/j.1365-2389.2010.01291.x

Reay, D.S., E.A. Davidson, K.A. Smith, P. Smith, J.M. Melillo, F. Dentener, and P.J. Crutzen. 2012. Global agriculture and nitrous oxide emissions. Nat. Clim. Change 2:410–416. doi:10.1038/nclimate1458

Rochette, P., and N.S. Eriksen-Hamel. 2008. Chamber measurements of soil nitrous oxide flux: Are absolute values reliable? Soil Sci. Soc. Am. J. 72:331–342. doi:10.2136/sssaj2007.0215

Sanford, G.R., J.L. Posner, R.D. Jackson, C.J. Kucharik, J.L. Hedtcke, and T.-L. Lin. 2012. Soil carbon lost from Mollisols of the north central U.S.A. with 20 years of agricultural best management practices. Agric. Ecosyst. Environ. 162:68–76. doi:10.1016/j.agee.2012.08.011

SAS Institute. 2013. JMP version 11. SAS Inst., Cary, NC.

Smart, D.R., and A.J. Bloom. 2001. Wheat leaves emit nitrous oxide during nitrate assimilation. Proc. Natl. Acad. Sci. USA 98:7875–7878. doi:10.1073/pnas.131572798

Smith, P., D. Martino, Z. Cai, D. Gwary, H. Janzen, P. Kumar, et al. 2007. Agriculture. In: B. Metz, O.R. Davidson, P.R. Bosch, R. Dave, and L.A. Meyer, editors, Climate change 2007: Mitigation. Cambridge Univ. Press, Cambridge, UK and New York. p. 497–540.

Venterea, R.T., K.A. Spokas, and J.M. Baker. 2009. Accuracy and precision analysis of chamber-based nitrous oxide gas flux estimates. Soil Sci. Soc. Am. J. 73:1087–1093. doi:10.2136/sssaj2008.0307

Wagner, S.W., D.C. Reicosky, and R.S. Alessi. 1997. Regression models for calculating gas fluxes measured with a closed chamber. Agron. J. 89:279–284. doi:10.2134/agronj1997.00021962008900020021x