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ORIGINAL ARTICLE

Strategies for mitigating N_2O and N_2 emissions from an intensive sugarcane cropping system

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Abstract In sugarcane cropping systems, high rates of N fertiliser are typically applied as sub-surface bands creating localised zones of high mineral N concentrations. This in combination with high levels of crop residue (trash) retention and a warm and humid climate creates conditions that are known to promote soil denitrification, resulting in high emissions of the potent greenhouse gas N_2O. These losses illustrate inefficient use of N fertilisers but total denitrification losses in the form of N_2 and N_2O remain largely unknown. We used the ^{15}N gas flux method to investigate the effect of cane trash removal and the use of the nitrification inhibitor 3,4-dimethylpyrazole phosphate (DMPP) on N_2 and N_2O emissions on a commercial sugarcane farm at Bundaberg, Australia. High gaseous N losses were observed under the standard grower practice where cane trash retention and N fertiliser application (145 kg N ha^{-1} as urea) resulted in N_2 and N_2O emissions (36.1 kg N ha^{-1}) from the subsurface N fertiliser band, with more than 50% of these losses emitted as N_2O. Cane trash removal reduced N_2 emission by 34% and N_2O emission by 51%, but had no effect on the $N_2O/(N_2+N_2O)$ ratio. The use of DMPP lowered N_2 and N_2O emission by 35% and 98%, respectively, reducing the percentage of these losses (N_2+N_2O) emitted as N_2O to only 4%. We conclude that the use of DMPP is an effective strategy to reduce N losses, minimise N_2O emissions, while keeping the benefits of cane trash retention in sugarcane cropping systems.

Keywords Denitrification · Climate change · Enhanced efficiency fertilisers · Nitrification inhibitors · Residue retention

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Introduction

Denitrification is the microbially facilitated process of reducing nitrate (NO_3^-) and nitrite. (NO_2^-) to gaseous forms of nitrogen (N), principally nitrous oxide (N_2O) as well as environmentally benign dinitrogen (N_2). Losses of N_2O and N_2 to the atmosphere in fertilised cropping systems can represent a substantial loss of applied fertiliser; possibly resulting in reduced plant

N uptake and reducing crop N use efficiency (NUE). Furthermore, losses of N_2O create environmental concerns; since N_2O is not only the third most important long-lived greenhouse gas (after CO_2 and CH_4), but also largest anthropogenic cause of stratospheric ozone depletion in the foreseeable future (Davidson and Kanter 2014; Ravishankara et al. 2009). Sugarcane is typically produced under conditions that are known to stimulate soil denitrification, i.e. high fertiliser inputs, a wet and warm climate, and N and C inputs due to the increasing adoption of cane trash retention. In contrast to burning, cane trash retention is known to prevent soil erosion (Valim et al. 2016), maintain soil moisture (Ng Cheong and Teeluck 2016; Sandhu et al. 2017), increase soil C (Canellas et al. 2010; Galdos et al. 2009; Thorburn et al. 2012), and provide a source of N (Fortes et al. 2013; Robertson and Thorburn 2007). However, increased soil moisture together with increased C and N substrate availability is conducive for denitrification, and cane retention has been shown to promote N_2O emissions (Wang et al. 2016c). Large N_2O emissions in sugarcane (Allen et al. 2010; Wang et al. 2016a) are indicative for high denitrification (N_2O+N_2) rates, and substantial N losses (Thorburn et al. 2017) from sugarcane systems are often attributed to denitrification. For Australian sugarcane cropping systems, only few studies report NUE using labelled ^{15}N fertiliser and suspect denitrification to be responsible for the loss of 25–60% of the applied fertiliser over one season (Chapman et al. 1994; Prasertsak et al. 2002; Takeda et al. 2021; Vallis et al. 1996). However, there is insufficient reliable data on N_2 losses from sugarcane soils based on field measurements since it is inherently challenging to measure N_2 emissions against the high atmospheric N_2 background. The only method that is suitable to directly measure N_2 emissions under field conditions is the ^{15}N gas flux (^{15}NGF) method where highly enriched ^{15}N fertiliser is applied to the soil, gas samples are taken using the static chamber method and analysed for their different isotopologues of N_2O and N_2 via isotope ratio mass spectrometry (IRMS) (Friedl et al. 2020a). One constraint of the ^{15}NGF method is that the ^{15}N label in the soil mineral NO_3^- pool is subject to dilution, as the nitrification of non-labelled N from the organic N pool leads to a gradual decrease of the ^{15}N label in the soil NO_3^- pool over time. Consequently, the use of the ^{15}NGF method is restricted to a limited time period following fertiliser (and hence ^{15}N tracer) application, and significant N_2 fluxes are often only found 1–4 weeks after tracer application. In sugarcane systems, only two studies exist that measured N_2 emissions in the field using the ^{15}NGF gas flux method. Weier (1996) found denitrification losses up to 3.6 kg N ha^{-1} day^{-1}, representing a loss of 40% of the applied fertiliser within 14 days after application from a commercial sugarcane farm in Australia. Warner et al. (2019) measured N_2 losses of up to 1.3 kg N ha^{-1} day^{-1} over a 7-day field campaign using a novel, field-based isotope ratio mass spectrometer system. These large losses highlight that denitrification can be a major loss mechanism from sugarcane soils with possibly significant economic cost to the industry, and strategies to mitigate denitrification losses by fertiliser and crop management are urgently needed.

In order to reduce N losses via denitrification from sugarcane systems, an improved nutrient management is required that better matches crop N demand with supply by adjusting N fertiliser inputs according to the seasonal yield potential, accounting for the ability of soil organic matter to supply N. Another proposed strategy to improve fertiliser NUE and reduce N_2O emissions in sugarcane systems is the use of nitrification inhibitors (NIs), delaying the conversion of ammonium (NH_4^+) to NO_3^- via nitrification, and thereby reducing substrate availability for denitrifying bacteria. Several studies have shown that the use of NIs can potentially increase crop NUE, reduce N leaching, and mitigate N_2O emissions from cropping soils (Abalos et al. 2014; Halvorson et al. 2014; Scheer et al. 2016; Wang et al. 2016b). In a meta-analysis, Akiyama et al. (2010) showed that NIs significantly reduce N_2O emissions by 38%, but so far research has focused on N_2O mitigation and the combined effect of NI on N_2 and N_2O emissions remains largely unexplored. Friedl et al. (2017) have shown that the use of the NI DMPP can reduce the N_2 losses in subtropical pasture by more than 70%, providing agronomic benefits that can offset the additional cost associated with the use of NIs. However, to date no such data exists for sugarcane cropping systems.

Therefore, the overall aims for this study were to: (i) quantify emissions of N_2O and N_2 from a subtropical sugarcane system following fertiliser application; (ii) assess the effect of cane trash retention on emissions of N_2O and N_2; and (iii) evaluate the efficacy of the NI DMPP to reduce N_2 and N_2O emissions with cane trash retained. As such, this study delivers an

improved quantitative process understanding for N_2 and N_2O emissions from sugarcane soils, testing a NI as a strategy to exploit the benefits of trash retention with minimal environmental offsets.

Materials and methods

Experimental design

The field experiment was conducted on a commercial sugarcane farm at Bundaberg, QLD (24°57′ S, 152°20′ E). The long-term (1959–2012) annual mean temperature in this subtropical region is 21.5 °C (Bundaberg Aero Station, the Bureau of Meteorology, Australia), with the lowest monthly mean temperature in July (16.1 °C) and the highest in January (25.8 °C). Mean annual rainfall is 1027 mm, where over half is received from December to March. The soil is classified as a redoxic Hydrosol (Isbell 2002) or Gleysol (WRB 2015) with loamy sand in the 0–0.3 m layer, underlain by sandy loam at about 0.3–0.6 m depth and sandy clay loam at about 0.6–1 m depth. In the 0–0.3 m layer, total organic C (TOC) and N ranged from 1.00 to 0.80% and from 0.07 to 0.05%, respectively, decreasing to 0.16% TOC and 0.02% N at 0.6−1 m depth. Soil pH ranged from 6.0 in the topsoil (0–0.3 m) to 5.7 at depth (0.6 − 1 m). Detailed information on selected soil physical and chemical properties is given in Table S1.

Sugarcane (CV Q238) was planted in the middle of raised beds (ca. 1.26 m wide) with a row spacing of 1.83 m in September 2013. The plant cane crops were fertilised at 145 kg N/ha, 28 kg P/ha, 100 kg K/ha and 25 kg S/ha by placing the fertilisers in bands (ca. 0.1 m under the surface) on both sides of the cane setts (ca. 0.05 m away). Following harvest of the plant cane in September 2014, three treatments were applied to compare DMPP-urea with conventional urea and assess the effect of the sugarcane trash blanket (cane residue retention):

(i) *DMPP*: Urea fertiliser with 0.6% DMPP solution (w/w) and sugarcane trash retained in the field.
(ii) *Trash retained (TRT)*: Standard Grower Practice, Urea fertiliser and sugarcane trash left in the field.
(iii) *Trash Removed (TRM)*: Urea fertiliser with sugarcane trash removed from the surface.

The treatments were arranged in a randomised block design with four replicates. The plots were 6.7 m along the row length and 9.2 m across five rows. A steel base (0.22 × 0.22 m) was installed on one side of the middle row in each plot for the manual chamber measurements. ^{15}N urea (60 atom %) was applied in solution corresponding to the recommended N fertiliser rate of 145 kg of N ha^{-1} as a band in the middle across the steel base, buried 0.1 m deep and ~0.05 m the sugarcane row. In the TRT and DMPP treatments a 0.05 m thick layer of sugarcane trash was placed on the soil surface, consistent with sugarcane trash management in the growing region.

Gas sampling

The ^{15}N gas flux method was used to measure N_2 and N_2O emissions by quantifying the increase in ^{15}N-labelled gases in the chamber headspace over time as described by Friedl et al. (2017). Gas samples were taken manually from the chambers once per week for 85 days after fertiliser application, except for the first week of the experiment when samples were taken twice. Polyethylene chambers with a headspace height of 0.314 m were placed on the steel frames, ensuring airtight conditions. Headspace gas samples (20 ml) were taken by connecting a syringe to a 2-way luer-lock tap installed in the lid of the chamber. Gas samples were then injected into a pre-evacuated 12 ml glass vial with a double wadded Teflon/silicon septa cap (Labco, UK). Headspace gas samples were collected at 0, 1 and 3 h after closure from each chamber. In addition, the ambient and soil temperature was taken once per replicate and hour and used to correct flux calculations for temperature.

Gas analysis and N_2O and N_2 flux calculations

All gas samples, taken 0, 1 and 3 h after closure, were analysed for N_2O by gas chromatography (GC) (Shimadzu GC-2014). Following GC analysis, gas samples taken at 0 and 3 h were analysed for the isotopologues of N_2 ($^{15}N^{14}N$, $^{15}N^{15}N$) and N_2O ([$^{14}N^{15}N^{16}O$ + $^{15}N^{14}N^{16}O$] and $^{15}N^{15}N^{16}O$) using an

automated isotope ratio mass spectrometer (Sercon, 20–20, UK) linked to a Sercon Cryoprep trace gas concentration system.

Flux rates of N_2O were calculated from the slope of the linear increase in gas concentration during the closure period. The coefficient of determination (R^2) was used as a quality check for linearity and flux rates were set to 0 if R^2 was < 0.80. Flux rates were corrected for temperature, air pressure and the ratio of cover volume to surface area as described by Scheer et al. (2014).

The ion currents (I) at *m/z* 44, 45, and 46 enabled the molecular ratios ^{45}R ($^{45}I/^{44}I$) and ^{46}R ($^{46}I/^{44}I$) to be calculated for N_2O, and I at *m/z* 28, 29 and 30 enabled ^{29}R ($^{28}I/^{29}I$) and ^{30}R ($^{28}I/^{30}I$) to be calculated for N_2. Fluxes of N_2 were calculated using the equations given by Spott et al. (2006), calculating the enrichment of the NO_3^- pool undergoing denitrification a_p, and the fraction of N_2 derived from this pool (f_p):

$$f_p = \frac{a_m - a_{bgd}}{a_p - a_{bgd}} \quad (1)$$

where a_{bgd} is the ^{15}N abundance of the atmospheric background and a_m is the measured ^{15}N abundance of N_2 from headspace gas samples taken 0 and 180 min after closure, respectively. Both a_{bgd} and a_m are calculated as

$$a_i = \frac{^{29}R + 2{*}^{30}R}{2{*}(1 + ^{29}R + ^{30}R)} \quad (2)$$

The ^{15}N enrichment of the soil NO_3^- pool undergoing denitrification a_p is calculated for N_2 and N_2O (a_p N_2 and a_p N_2O) as

$$a_p = \frac{^{30}x_m - a_{bgd} * a_m}{a_m - a_{bgd}} \quad (3)$$

The measured fraction of m/z 30 in N_2 $^{30}x_m$ is calculated as:

$$^{30}x_m = \frac{^{30}R}{(1 + ^{29}R + ^{30}R)} \quad (4)$$

To calculate a_p N_2O, ^{45}R and ^{46}R were converted to ^{29}R and ^{30}R by correcting for the naturally occurring O_2 isotopes:

$$^{29}R = ^{45}R - ^{17}R \quad (5)$$

$$^{30}R = ^{46}R - ^{29}R * ^{17}R - ^{18}R \quad (6)$$

Using the value of 0.00038 for ^{17}R and 0.002079 for ^{18}R (Arah 1997). If only ^{29}R was > the detection limit (DL), f_p was calculated as

$$f_p = \frac{1}{1 - \frac{^{29}R(1-a_p)^2 - 2a_p(1-a_p)}{^{29}R(1-a_{bgd})^2 - 2a_{bgd}(1-a_{bgd})}} \quad (7)$$

using a_p derived from N_2O assuming N_2 and N_2O evolving from the same NO_3^- pool undergoing denitrification. The headspace N_2 concentrations, corrected for air pressure and temperature, were multiplied by the respective f_p values giving N_2 fluxes expressed in g N_2–N emitted g^{-1} soil day^{-1}. Cumulative fluxes of N_2O and N_2 were calculated by linear interpolation between sampling events.

The between batch precision of the IRMS for N_2 based on the standard deviation of atmospheric air samples (n = 60) at 95% confidence intervals (Friedl et al. 2020a) was 1.46×10^{-6} and 1.36×10^{-6} for ^{29}R and ^{30}R, respectively. The corresponding method detection limit (DL) ranged from 5.5 mg N_2–N m^{-2} day^{-1} with a_p assumed at 60 atom % to 16.6 mg N_2–N m^{-2} day^{-1} with a_p assumed at 20 atom %. If N_2 fluxes were below the DL, fluxes were set to 50% of the calculated DL.

The fraction of fertiliser derived N_2O and N_2 (*ndff*) was calculated as the ratio of ^{15}N atom excess % of N_2O or N_2 and the ^{15}N atom excess % of the N fertiliser applied, with the soil derived fraction calculated as the difference of 1 and *ndff*. The measured ^{15}N atom fraction in N_2O (a'*s*) was used for the ^{15}N recovery in N_2O, accounting for N_2O from nitrification and denitrification mediated pathways. For N_2, a_p N_2 was used for the ^{15}N recovery in N_2. If only R29 > DL, a_p derived from N_2O was assumed as the enrichment of the NO_3^- pool undergoing denitrification and used to calculate ^{15}N in N_2.

Auxiliary measurements

Soil water content in the topsoil (0–0.1 m) was recorded using *in-situ* moisture probes and a data logger (ThetaProbe, Delta-T Devices Ltd, UK). Soil samples (0–0.1 m and 0.1–0.3 m) for soil mineral N analysis were taken directly from the fertiliser band

next to crop row 2 or 4, twice during the experimental period, 9 and 59 days after fertilisation. Three soil samples were extracted for NO_3^- and NH_4^+ with 2 M KCl (1:5 w:v) for 1 h on a rotary shaker, filtered through Whatman no. 40 filter papers and analysed colorimetrically (Rayment and Lyons 2011).

Statistical analysis

Statistical analyses were conducted with SPSS 27.0 (SPSS Inc.,2020). Treatment effects on N_2, N_2O, and soil mineral N concentrations were examined by analysis of variance (ANOVA) ($p < 0.05$). Normal distribution of the data was assessed by the Shapiro-Wilk test for normality. Homogeneity of variance was verified by Levene's test for equality of variances. Differences between treatments were assessed using the Ryan-Einot-Gabriel-Welsh test. Effects of treatments on the temporal pattern of a'*s* were examined using linear mixed effect models accounting for repeated measurements by specifying chambers as subjects with repeated measures over time. Values in the figures represent means ± standard error of the mean.

Results

Emissions of N_2 and N_2O

Of 168 N_2 fluxes, 15% were discarded due to analytical problems, and 78% exceeded the DL, of which 36% were calculated using a_p derived from N_2 and 64% were calculated using a_p derived from N_2O.

Significant emissions of N_2 could be measured in all treatments and ranged from 33.4 to 73.4 mg m^{-2} day^{-1}. In the week before N fertilisation, 93 mm of rain fell at the site. The temporal pattern of N_2 emissions showed elevated N_2 emissions in all treatments on day 1 after fertiliser application followed by 42 mm rain within 2 days, and a declining trend over the first 3 weeks of the experiment with little rainfall and declining soil moisture levels (Fig. 1). After a period of heavy rainfall and increasing soil moisture levels, a second peak of N_2 emissions was observed on day 44 of the experiment in all treatments. From day 50 onwards, only small fluxes (< 10 mg m^{-2} day^{-1}) were observed in the DMPP and TRM treatments, with slightly higher N_2 emissions in the TRT treatment. Over the 85 days observation period cumulative N_2 losses from the fertiliser band amounted to 1703.44 ± 212.74 mg N_2–N m^{-2} in the TRT treatment with significantly lower losses (< 1200 mg N_2–N m^{-2}) in the DMPP and TRM treatments (Table 1).

Emissions of N_2O showed a distinctly different temporal pattern than the N_2 fluxes (Fig. 1). On day 1 after fertiliser application only very small fluxes were observed in all treatments. From day 4 onwards significantly elevated N_2O emissions were observed with highest emissions in the TRT treatment (116.9 mg N_2O–N m^{-2} day^{-1} on day 8). N_2O emissions stayed elevated for approximately 4 weeks and returned to very small fluxes (< 0.5 mg N_2O–N m^{-2} day^{-1}) from day 36 onwards in all treatments. Over the 85 days, 1913.0 mg N_2O–N m^{-2} and 936.4 mg N_2O–N m^{-2} were lost in the TRT and TRM treatments, respectively, with significantly reduced losses of 48.7 mg N_2O–N m^{-2} in the DMPP treatment.

Changes in the corresponding $N_2O/(N_2+N_2O)$ product ratio are shown in Fig. 1. Values across all treatments showed a high variation and ranged from 0 to 0.89. In the TRT and TRM treatments the $N_2O/(N_2+N_2O)$ ratio showed a clear peak from day 4 to day 36 after fertiliser application. During this period the N_2O emissions accounted for 67% and 60% of the total N_2 and N_2O losses, in the TRT and TRM treatments, respectively. Only DMPP treatment showed constantly low $N_2O/(N_2+N_2O)$ ratios (ranging from 0 to 0.16), due to very small N_2O fluxes coinciding with elevated N_2 emissions. Over the 85 days experimental period N_2O losses accounted for 46–48% of the total N_2 and N_2O losses in the TRT and TRM treatment, with a significantly lower $N_2O/(N_2+N_2O)$ ratio of 4% in the DMPP treatment (Table 1).

Total denitrification losses (N_2+N_2O) were highest in TRT (36164.1 mg m^{-2}), followed by TRM (20608.8 mg N m^{-2}) and DMPP (11548.7 mg N m^{-2}) (Table 1), with 75–81% of the N_2 losses and 23–51% of the N_2O losses derived from fertiliser (Table 2). This represents a loss of 7%, 4% and 2% of the fertiliser N applied as a band from the TRT, TRM and DMPP treatment respectively.

The temporal variation of the ^{15}N fraction in N_2O differed between treatments, showing values around 0.5 for the ^{15}N atom fraction in N_2O for TRT and TRM in the beginning of the experiment, decreasing sharply 36 days after fertilisation to less than 0.005 for the rest of the experiment (Fig. 2). The

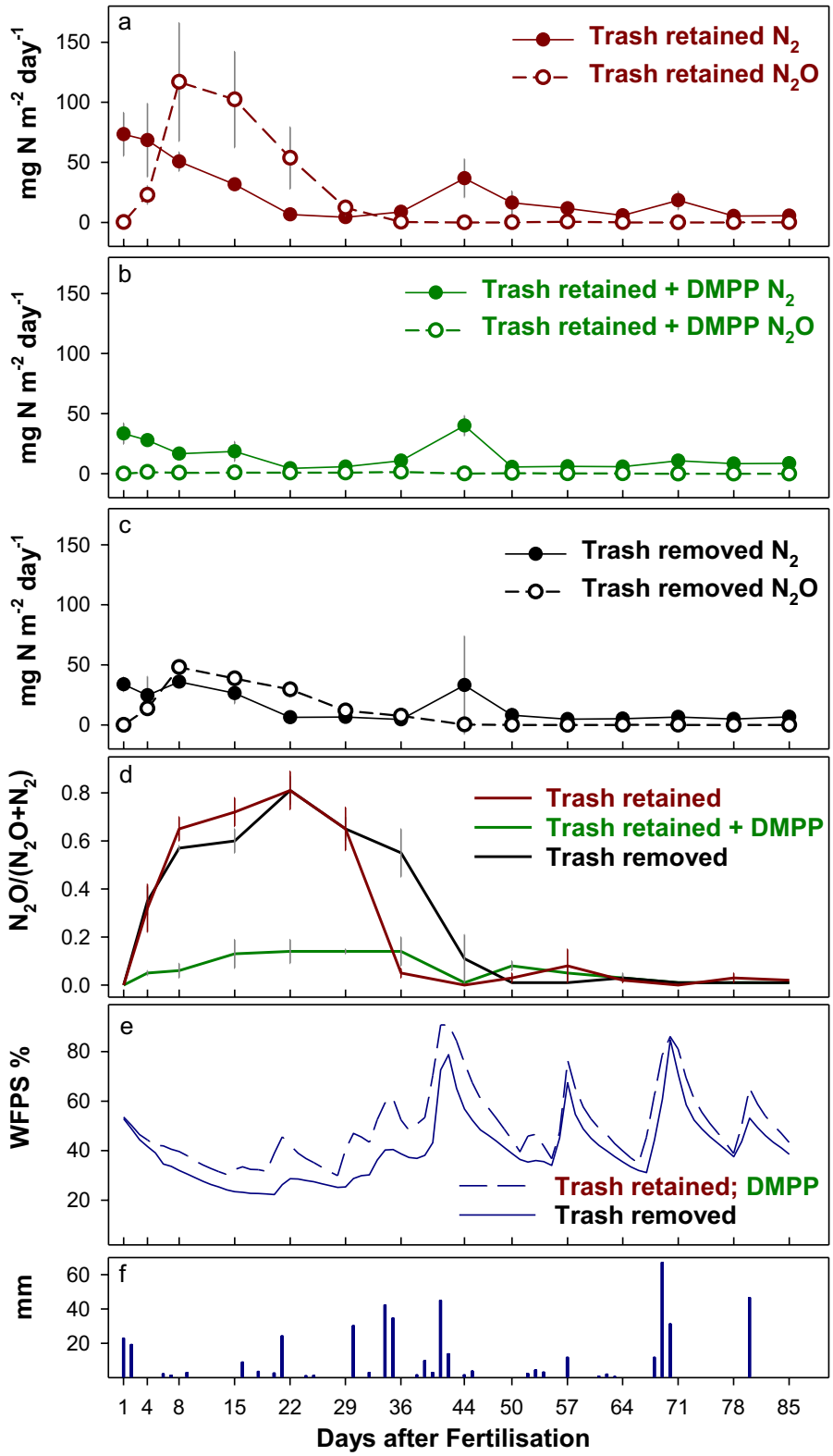

Fig. 1 Average daily N_2 and N_2O fluxes for for the different treatments **a**, **b** and **c** with the corresponding product ratio $N_2O/(N_2+N_2O)$ (**d**), soil water filled pore space (WFPS) 0-10 cm depht, (**e**) and daily precipitation (**f**)

Table 1 Cumulative emissions of N_2 and N_2O from the fertiliser band in a subtropical sugarcane system over 85 days and their respective soil and fertiliser derived fractions

	N_2O+N_2	$P=0.035$	N_2 –N mg m^{-2}	$P=0.027$	N_2O –N mg m^{-2}	$P=0.056$	$N_2O/(N_2+N_2O)$	$P<0.001$
Trash retained	36164.1 ± 961.0	A	1703.4 ± 212.7	A	1913.0 ± 802.1	A	0.48 ± 0.07	A
DMPP	11548.7 ± 474.1	B	1107.2 ± 51.8	B	47.6 ± 5.4	B	0.04 ± 0.01	B
Trash removed	20608.8 ± 1048.7	AB	1124.5 ± 121.5	B	936.4 ± 63.9	AB	0.46 ± 0.04	A

Table 2 Soil and fertiliser derived fractions of cumulative N_2 and N_2O emissions from the fertiliser band in a subtropical sugarcane system over 85 days

N_2O	Fertiliser derived %	$P<0.001$	Soil derived %	$P<0.001$
Trash retained	51.2 ± 0.6	A	48.8 ± 0.6	B
DMPP	23.0 ± 4.2	B	77.0 ± 4.2	A
Trash removed	49.9 ± 0.6	A	50.1 ± 0.6	B
N_2	Fertiliser derived %		Soil derived %	
Trash retained	80.0 ± 3.9	A	20.0 ± 3.9	A
DMPP	74.4 ± 4.2	A	25.6 ± 4.2	A
Trash removed	81.5 ± 3.5	A	18.5 ± 3.5	A

initial ^{15}N fraction of N_2O in the DMPP treatment was below 0.3, decreasing to < 0.005 until the end of the experiment. Values for a_p N_2O and a_p N_2 ranged from 0.24 to 0.56, and 0.27 to 0.59, respectively and agreed largely over the time of the experiment (Figure S1).

Soil mineral N content

Nine days after fertilisation soil NO_3^- levels ranged from 19.1 to 47.1 NO_3^- mg kg^{-1} soil in the 0-0.1 m layer, and 18.4 to 44.0 NO_3^- mg kg^{-1} soil in the 0.1–0.3 m layers with lowest concentration in the DMPP treatment, although with no significant difference between the treatments due to a high variability between the replicates (Table 3). Soil NH_4^+ concentration 9 days after fertilisation ranged from 53.2 to 97.2 NH_4^+ mg kg^{-1} soil in the 0-0.1 m layer, with highest average values in the DMPP treatment, but no significant treatment effect. Soil NO_3^- and NH_4^+ levels dropped to almost zero in all treatments 59 days after fertiliser application, with slightly elevated levels in the DMPP treatment in the 0-0.1 m layer.

Discussion

Novel fertiliser management strategies are needed to reduce environmental pollution, mitigate climate change, and increase profitability in sugarcane farming systems. Such strategies need to target denitrification, as wet and warm climatic conditions in combination with high fertiliser inputs and increasing crop residue retention are especially conducive for high losses of fertiliser N via this pathway. The first field study to investigate the effect of the NI DMPP and sugarcane trash retention on N_2 and N_2O losses from a commercial sugarcane farm demonstrates (a) an increase of denitrification by sugarcane trash retention, (b) high losses of N_2 and N_2O from banded N fertiliser, (c) a substantial reduction of total denitrification losses (N_2+N_2O) from the fertiliser band by DMPP and (d) a shift in the N_2O:N_2 ratio towards N_2 by DMPP.

Temporal variability of N_2O and N_2 emissions across treatments suggests that NO_3^- concentration around the N fertiliser band determines the $N_2O/(N_2+N_2O)$ ratio

Fertiliser N application followed by 42 mm rain resulted in highly elevated N_2 emissions at day one

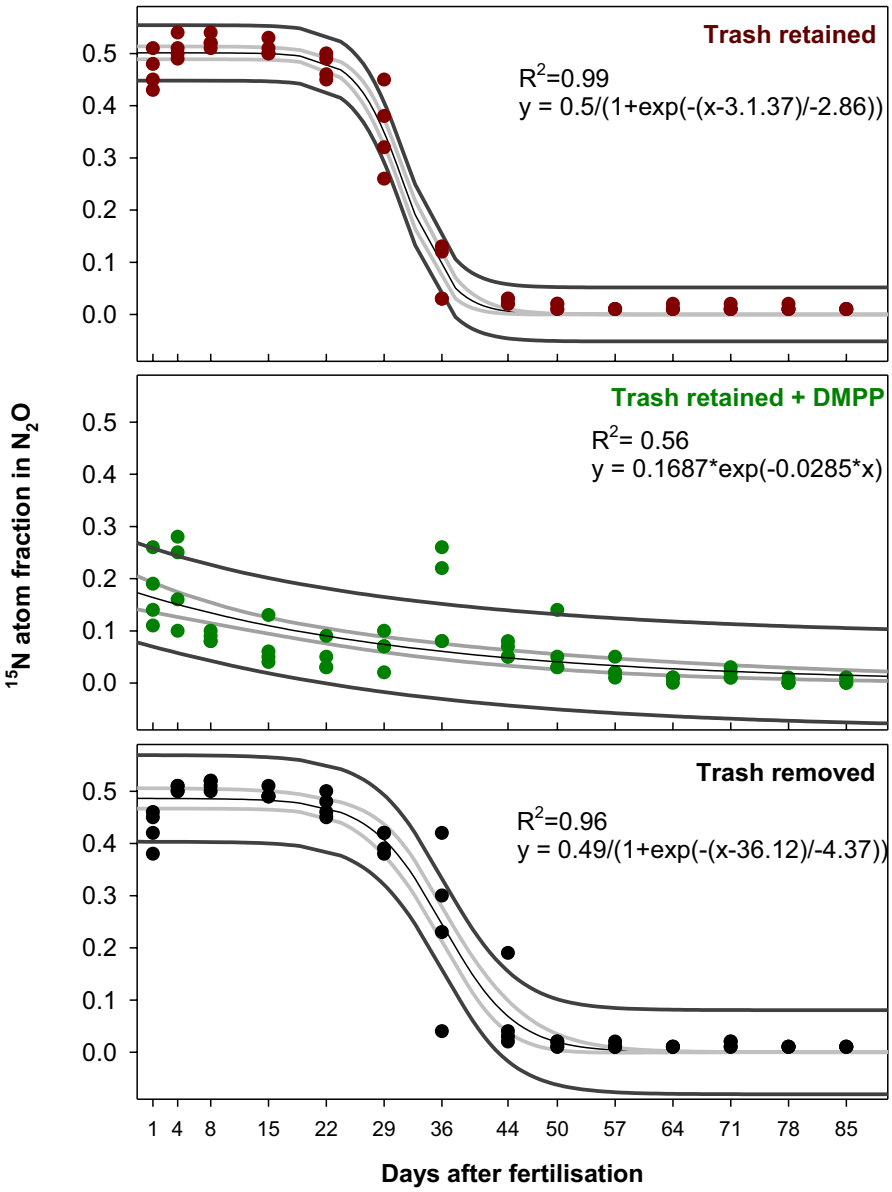

Fig. 2 The ^{15}N atom fraction in N_2O (a'*s*) for the different treatments

from all treatments. These emissions (33.4–73.4 mg m^{-2} day^{-1}) are within the range of N_2 emissions previously reported from sugarcane systems after the application of N fertiliser (Warner et al. 2019; Weier 1996) and show that the fertiliser applied as urea was rapidly converted to NO_3^- within the soil and available for denitrification to take place. Interestingly, there were only very small N_2O emission on day 1, indicating that the NO_3^- was completely reduced to N_2 during the denitrification process. This is in contrast to other studies that investigated N_2 and N_2O fluxes from fertilised cropping system in the field where often a burst of N_2O emission on day one and a shift towards full denitrification to N_2 over time is reported (Buchen et al. 2016; Friedl et al. 2017; Warner et al. 2019). This pattern has commonly been explained by increasing anaerobic condition due to increased O_2 consumption in the soil profile (Meyer et al. 2010; Rohe et al. 2021; Yang et al. 2021) and/or the time required for the activation of the N_2O reductase activity responsible for the reduction of N_2O to N_2 (Morley and Baggs 2010; Zheng and Doskey 2016). The different pattern in our study can most likely be explained by > 90 mm rainfall in the week

Table 3 Soil mineral N (NH_4^+ and NO_3^-) at 0–0.1 m and 0.1–0.3 m depth in the fertiliser bands 9 and 59 days after fertilisation for the different treatments including Urea, DMPP and Trash removed

NH_4^+ mg kg^{-1} soil		9 days after fertilisation	59 days after fertilisation
Trash retained	0–0.1 m	53.2 ± 24.8	1.5 ± 0.2
DMPP	0–0.1 m	97.2 ± 29.1	2.1 ± 0.3
Trash removed	0–0.1 m	85.2 ± 18.4	1.6 ± 0.1
Trash retained	0.1–0.3 m	25.4 ± 17.4	0.4 ± 0.4
DMPP	0.1–0.3 m	47.5 ± 13.7	1.1 ± 0.2
Trash removed	0.1–0.3 m	48.6 ± 22.5	1.2 ± 0.2
NO_3^- mg kg^{-1} soil		9 days after fertilisation	59 days after fertilisation
Trash retained	0–0.1 m	47.1 ± 13.3	1.5 ± 0.1
DMPP	0–0.1 m	19.1 ± 8.0	5.1 ± 3.6
Trash removed	0–0.1 m	41.7 ± 5. 7	1.0 ± 0.1
Trash retained	0.1–0.3 m	32.0 ± 11.0	1.5 ± 0.1
DMPP	0.1–0.3 m	18.4 ± 3.5	1.8 ± 0.3
Trash removed	0.1–0.3 m	44.0 ± 18.0	1.0 ± 0.5

prior to the experiment and its effect on denitrification enzyme activity, together with the dynamics of N substrate supply from the urea band. The high N_2 emissions at day one clearly show that reduction enzymes involved in the denitrification process including the N_2O reductase were already activated, likely due to anoxia in the soil caused by prior rainfall (Uchida et al. 2014), promoting a complete reduction of NO_3^- and high emissions of N_2 in all treatments. However, it seems that increasing concentrations of NO_3^- around the fertiliser band inhibited either *de novo* synthesis or activity of the N_2O reductase increasing N_2O emission and the corresponding $N_2O/(N_2+N_2O)$ product ratio. The low N_2O emission and $N_2O/(N_2+N_2O)$ product ratios in the DMPP treatment however indicate inhibition of nitrification, preventing a build-up of NO_3^- and thus promoting the reduction of N_2O to N_2. This line of argument agrees with the reported suppression of the N_2O reductase gene *nosZ* by high NO_3^- concentrations, and the attenuation of this effect by DMPP, leading to an increase in *nosZ* abundance and a reduction in N_2O emissions (Friedl et al. 2020b). The decrease of N_2O and N_2 emissions in all treatments between day 8 and day 29 can be attributed to a gradual decrease of mineral N concentration in the fertiliser band. It seems that in particular the period of heavy rainfall between day 29 and day 50 after fertiliser application resulted in a depletion of soil mineral N via plant uptake, leaching and denitrification. The simultaneous decrease in the ^{15}N enrichment of N_2O (a'*s)* in the TRT and TRM treatments (Fig. 2) is consistent with consumption of NO_3- via these pathways, as co-occurring and/or following nitrification leads to a dilution of the ^{15}N label in the soil NO_3- pool.

A second peak of N_2 emissions was observed in all treatments on day 44 of the experiment. This peak occurred after a series of heavy rainfall events leading to increased soil moisture, saturation and thus promoting denitrification in the soil profile. In contrast to N_2, N_2O fluxes showed no significant response to this rainfall event, resulting in almost complete denitrification to N_2 with $N_2O/(N_2+N_2O)$ product ratios ranging from 0.001 to 0.003. This can be most likely attributed to the combination of limited O_2 availability and increased entrapment of N_2O in the soil matrix after rainfall, together with depleted levels of soil NO_3^- more than 6 weeks after fertilisation. The low N_2 and N_2O fluxes after day 50, despite the occurrence of several heavy rainfall events show, that denitrification was limited by N substrate availability, which is also reflected in the low mineral N values measured at day 59 in all treatments.

DMPP reduces magnitude and $N_2O/(N_2+N_2O)$ ratio of N_2O and N_2 emissions from the fertiliser band

The use of DMPP lowered N_2 emission by 35% and N_2O emission by 98% compared to the standard TRT treatment, respectively. This is among the highest reported N_2O reductions efficiency of an NI. In a recent meta-analysis Ruser and Schulz (2015) summarised the results from 140 field studies on the effect of different NIs on N_2O emissions from agricultural soils and found a mean reduction potential of approx. 35%, but a wide range of inhibitory effects has been reported by numerous studies depending on soil type, climate and management system. The high N_2O reduction agrees well with an inhibitory effect of DMPP ranging from 60 to 83% in sub-tropical summer cereal cropping system in Australia where the fertiliser was applied as a band besides the crop rows (De Antoni Migliorati et al. 2016; Scheer et al. 2016), indicating that NI might be especially effective in reducing N_2O emissions from fertiliser bands. Banding N-fertilisers creates a biochemical environment in which different N transformations will occur, compared to when the same product is broadcast or mixed into soil which has implications for the effective utilisation of NIs when applied in sub-surface fertiliser bands (Janke et al. 2020). A sub-surface fertiliser N band will create localised zones of high NO_3^- concentration that are particularly vulnerable to denitrification. Our results indicate that DMPP is very efficient at inhibiting the conversion of urea into NO_3^- from the fertiliser band leading to the high reduction in N_2O emissions. This in contrast to observations by Janke et al. (2019) that did not find enhanced inhibition of nitrification with DMPP in a highly concentrated urea band in an soil incubation essay, highlighting the difficulties to transfer results from small scale soil incubations to the field level. As incubation containers restrict diffusion of N from the simulated band, high concentrations of ammonia following urea hydrolysis can exert inhibitory effects on the second step of nitrification, i.e. the oxidation of NO_2^- to NO_3^- (Breuillin-Sessoms et al. 2017). These conditions may be representative for the urea band itself, but not for zone around the band, where diffusion supplies N that is readily nitrified. This assumption is consistent with the immediate onset of fertiliser derived N_2O and N_2 emissions in the study presented here, demonstrating rapid supply of NO_3^- shortly after urea application, effectively limited by the use of DMPP.

The effectiveness of DMPP in our study is further supported by the significant reduction of N_2 emission in the DMPP treatment and the observed shift of denitrification losses towards N_2. It is known that during denitrification, NO_3^- competes with N_2O as terminal electron acceptor, leading to high $N_2O/(N_2+N_2O)$ ratios under high NO_3^- concentrations, and DMPP has been shown to mitigate this effect (Friedl et al. 2020b). However, there is hardly any field data available that investigated the effect of DMPP on both N_2 and N_2O emission under field conditions. Friedl et al. (2017) reported that DMPP reduced N_2 losses from a sub-tropical pasture by more than 70%, providing the first field-based evidence that DMPP can substantially reduce N_2 emissions, but found no effect on N_2O emissions. The contrasting finding that in our study DMPP had the strongest reduction effect on N_2O can be explained by the different fertiliser management and soil N cycling in sugarcane systems. In our study, a high rate of N fertiliser was applied in a band, leading to a build-up of NO_3^- in the soil, while at the pasture sites a low rate of fertiliser was broadcasted at the surface which, combined with tight N cycling between the soil microbial biomass and plants in pasture soils, limited NO_3^- concentrations resulting in complete denitrification to N_2. Overall, our study suggests that the use of DMPP in sugarcane systems with banded fertiliser does not only offer environmental benefits by reducing N_2O emissions but also substantially reduces overall denitrification losses, supporting findings from a modelling study highlighting DMPP as an economically viable strategy to improve NUE and reduce N_2O emissions for Australian sugarcane industry (De Antoni Migliorati et al. 2021).

Cane trash management has no effect on the $N_2O/(N_2+N_2O)$ ratio

Cane trash removal reduced N_2 emission by 34% and N_2O emission by 51%, respectively. Numerous studies have reported a stimulation of N_2O emissions in response to crop residue retention and a recent meta-analysis found an average increase in soil N_2O emissions by 43% with crop residue incorporation compared to residue removal (Abalos et al. 2022). This average increase

agrees well with the results of our study, but it needs to be noted that the effect of crop residue retention on N_2O emissions depends on multiple factors including the C:N ratio and the amount of residues returned, mineral N content of the soil, and the increase of soil moisture due to residue retention. Overlapping effects of these factors can lead to both increases and reductions in N_2O emissions due to residue retention (Nguyen et al. 2015). In sugarcane systems overall a stimulation of N_2O emissions with cane trash retention has been reported (Wang et al. 2016c). The study presented here shows for the first time an increase of both N_2O and N_2 emissions in response to cane trash retention. Residues may supply N for the production of N_2O and N_2 despite their high C:N ratio, yet absolute amounts in comparison to those from the urea band are likely to be negligible. The temporal pattern of soil water content suggests slightly higher WFPS in the trash retained treatments (TRT, DMPP), which may explain at least partially increased emissions of N_2 and N_2O as compared to trash removed. The trash blanket is also likely to release labile C compounds upon wetting, which is consistent with previous research that reported increased rates of soil denitrification in the presence of easily degradable C compounds (Senbayram et al. 2012; Weier et al. 1993), linked to the creation of anoxic micro-sites in the soil profile (through increased oxygen consumption via soil respiration) and the supply of an energy source to denitrifying microorganisms in the soil. However, the effect of available organic C in soils on the $N_2O/(N_2+N_2O)$ product ratio is still not fully understood, but a shift in the N_2O:N_2 ratio towards N_2 is generally assumed (Giles et al. 2012; Li et al. 2021). In our study, there was no significant effect of trash management on the $N_2O/(N_2+N_2O)$ product ratio with almost identical ratios with or without trash retention (TRT, TRM) over the entire experimental period. These findings, together with the reduction of the $N_2O/(N_2O+N_2)$ ratio by DMPP indicate that the ratio was mainly controlled by the high NO_3^- availability around the fertiliser band, confirming results from a previous study that found substantially lower $N_2O/(N_2O+N_2)$ product ratio after the application of labile C only in soil with low NO_3^- contents (Senbayram et al. 2012).

Sources of N_2O and N_2

The enrichment of the NO_3^- pool undergoing denitrification a_p was calculated from N_2O, and whenever possible, from N_2 data. Comparing a_p derived from N_2O and N_2 shows a good agreement between both, supporting the assumption of a relatively uniform ^{15}N labelled NO_3^- pool (Friedl et al. 2020a). Interestingly, the DMPP treatment had no effect on a_p, despite the decrease in N_2O emissions. This suggests that DMPP reduced the nitrification of both fertiliser as well as soil N, which likely led to a reduced NO_3^- pool for denitrification, but had no effect of the proportion of ^{15}N labelled (fertiliser derived) and non-^{15}N labelled (soil derived) N, resulting in similar a_p values as compared to TRT and TRM. This assumption is supported by previous studies demonstrating reduced nitrification of soil derived N (Friedl et al. 2017), as rainfall and/or irrigation after N fertilisation promote the diffusion of both N fertiliser and NIs, leading to a decoupling of N fertiliser and NIs. Denitrification is assumed as the sole source of N_2, and consequently DMPP showed no effect on the soil and fertiliser derived fraction of N_2 emissions (Table 2). Even though a*p* showed no treatment effect, the overall ^{15}N fraction in N_2O a'*s* was lower in the DMPP treatment (Fig. 2) compared to TRT and TRM. Furthermore, DMPP increased the soil derived fraction of emitted N_2O. The NI DMPP targets the ammonia monooxygenase (AMO), the enzyme catalysing the first step of nitrification, the conversion of NH_4^+ to hydroxylamine (NH_2OH). However, not all nitrifiers oxidise NH_4^+ via AMO (Martikainen 2022; Wood 1990), and N_2O production pathways including heterotrophic nitrification by fungi may not be affected by the use of DMPP. The lower a'*s* values in the DMPP treatment suggest a larger relative contribution of N_2O production pathways bypassing AMO when classic autotrophic nitrification is inhibited, consistent with the larger contribution of soil derived N to N_2O emissions in the DMPP treatment. The significant reduction of N_2O by DMPP suggests that nitrification in this sugarcane soil is dominated by nitrifiers which possess AMO. Nevertheless, research under controlled conditions is needed to evaluate N_2O source partitioning in response to NIs from sugarcane systems, as the relative contribution of heterotrophic/fungal N_2O production pathways may be of significance for overall N_2O emissions in these agroecosystems with residue retention.

Conclusion

The first field study investigating the effect of the NI DMPP and sugarcane trash retention on N_2O and

N_2 losses from a sugarcane system demonstrates significant losses of N_2O and N_2 from a subsurface N fertiliser band, with more than 50% of these gaseous N losses emitted as N_2O. The large amount of N_2O lost highlights that high N substrate availability close to the fertiliser band is conducive for denitrification losses, and that ensuing high NO_3^- concentrations shift the N_2O:N_2 ratio towards N_2O. Emissions of N_2O and N_2 in this study were measured from the N fertiliser band, likely accounting for a large fraction of overall N_2O and N_2 loss. Nevertheless, further method development is needed to measure/upscale N_2O and N_2 emissions from both fertilised (band) and non-fertilised (furrow) areas, as current analytical methods are limited to fertilised areas within a field. Trash retention increased the magnitude of N_2O and N_2 emissions likely reflecting overlapping effects of increased soil water content and labile C supply from residues. The lack of response of the $N_2O/(N_2+N_2O)$ ratio to trash management however suggests that soil NO_3^- concentrations predominantly control the $N_2O/(N_2+N_2O)$ ratio in banded systems with trash retention. The NI DMPP was not only extremely effective in reducing overall N_2O and N_2 losses but also in promoting complete denitrification of N_2O to environmentally benign N_2, with only 4% of total N_2O and N_2 losses emitted as N_2O. This shows that DMPP might be especially effective in reducing N_2O emissions from banded fertiliser were localised zones of high NO_3^- concentration around the fertiliser band can be expected. Consequently, the use of DMPP appears to be an effective strategy to minimise N losses, while keeping the benefits of cane trash retention. Assessing DMPPs effect on other N loss pathways and sugarcane yield will show if the reduction in overall N losses allows for lower N fertiliser rates. Combining NIs with reduced N rates compensates for the price premium of DMPP fertilisers, making it an economically viable strategy to improve NUE while reducing the greenhouse gas-footprint of the sugarcane industry.

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Data Availability The datasets analysed during the current study are available from the corresponding author on reasonable request.

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