

Soil type, bulk density and drainage effects on relative gas diffusivity and N₂O emissions

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Abstract. Nitrous oxide (N₂O), a greenhouse gas, contributes to stratospheric ozone depletion. Agricultural fertiliser use and animal excreta dominate anthropogenic N₂O emissions. Soil relative gas diffusivity (D_p/D_o) has been used to predict the likelihood of soil N₂O emissions, but limited information exists about how soil N₂O emissions vary with soil type in relation to D_p/D_o . It was hypothesised that, regardless of soil type, the N₂O emissions would peak at the previously reported D_p/D_o value of 0.006. Four pasture soils, sieved and repacked to three different bulk densities, were held at nine different soil matric potentials between near saturation and field capacity. Soil nitrate and dissolved organic matter concentrations were adequate for denitrification at all soil matric potentials. Increasing soil bulk density and soil matric potential caused D_p/D_o to decline. As D_p/D_o declined to a value of 0.006, the N₂O fluxes increased, peaking at $D_p/D_o \leq 0.006$. This study shows that the elevation of N₂O fluxes as a D_p/D_o threshold of 0.006 is approached, holds across soil types. However, the variability in the magnitude of the N₂O flux as D_p/D_o declines is not explained by D_p/D_o and is likely to be dependent on factors affecting the N₂O : (N₂O + N₂) ratio.

Keywords: agriculture, compaction, denitrification, gas diffusivity, greenhouse gas, matric potential, nitrous oxide, porosity.

Received 8 June 2020, accepted 24 August 2020, published online 25 September 2020

Introduction

Nitrous oxide (N₂O) is a potent greenhouse gas and currently the dominant ozone depleting substance (Ravishankara *et al.* 2009). Atmospheric concentrations of N₂O have increased since preindustrial times by 20%, from 271 to 332 ppb in 2020, due to land use and land use changes, especially in agriculture (Ciais *et al.* 2013; NOAA 2020). These agricultural emissions of N₂O have been predominately driven by the use of nitrogen (N) fertiliser and the deposition of animal excreta (Davidson 2009). Emissions of N₂O from agricultural soils arise from biotic and abiotic processes. Key biological pathways include nitrification, nitrifier-denitrification and denitrification (Zumft 1997; Wrage-Mönnig *et al.* 2018; Stein 2019). The relative dominance of a given biological pathway depends on substrate supply and the oxygen (O₂) status of the soil. In well oxygenated agricultural soils, fertiliser- or excreta-derived ammonia are oxidised, ultimately to nitrate. Under conditions of high ammonium supply, for example following urea fertiliser application, autotrophic ammonia oxidising bacteria (AOB) dominate nitrification (Hink *et al.* 2018) with N₂O emissions resulting

from biotic and abiotic reactions of the intermediary metabolites (Stein 2019). Hypoxic conditions stimulate AOB to perform nitrifier-denitrification (Stein 2019). If the soil becomes anaerobic, heterotrophic denitrification becomes the dominant process producing N₂O (Butterbach-Bahl *et al.* 2013; Zhu *et al.* 2013). Denitrification requires both a carbon (C) source and nitrate, or one of the obligate intermediary N compounds in the denitrification sequence, as substrates. Thus, production of N₂O from an agricultural soil is highly dependent on the soil's O₂ status, N substrate availability and in the case of denitrification also the C supply.

Because the diffusion of a gas through water is $\sim 1 \times 10^4$ times slower than in air, the effective diffusion coefficient for O₂ in soil is proportional to the volume fraction of soil that is water-filled (Farquharson and Baldock 2008). Consequently, measures of soil water content, such as water-filled pore space (WFPS), have often been used as a predictor for determining the occurrence of denitrification. Farquharson and Baldock (2008) queried the use of WFPS as a predictor for N₂O emissions because WFPS is a normalised dimensionless value that fails to quantify the fraction of the entire soil

volume that is filled with water, or air, and thus it is not directly proportional to the diffusion of gases. Hence, while adequate for comparing processes in a single soil with a constant soil bulk density (ρ_b) it becomes problematic when comparing soils with varying soil ρ_b . This was also demonstrated by Balaine *et al.* (2013), who showed that soil repacked to varying soil ρ_b , and maintained at different moisture contents, resulted in peak N₂O emissions occurring across a relatively wide range of WFPS. Above an upper limit of WFPS, strongly anaerobic conditions also induce full denitrification, with N₂O reduced to dinitrogen (N₂). Farquharson and Baldock (2008) went on to suggest that measures of soil water content should be linked to structural parameter(s) to better describe gas diffusion in soils. Relative soil gas diffusivity (D_p/D_o) accounts for pore connectivity and continuity of the functional gas pore phase: where D_p is the gas diffusion constant in the soil (m³ soil air m⁻¹ soil s⁻¹) and D_o is the gas diffusion coefficient of the same gas in free air (m² air s⁻¹). Accordingly, Balaine *et al.* (2013) were able to show that peak N₂O emissions were poorly explained by WFPS but a strong linear relationship ($P < 0.01$, $r^2 = 0.82$) with D_p/D_o was observed. Moreover, these N₂O peaks emissions from a soil repacked to varying soil ρ_b and held over a range of soil moisture contents aligned with a threshold value of D_p/D_o , equal to 0.006. Furthermore, Balaine *et al.* (2016) found that when examining cumulative N₂O emissions over 35 days there was an increase in N₂O emissions at $D_p/D_o < 0.006$. Stepniewski (1981) reported that soils became anaerobic for plant roots at $D_p/D_o < 0.002$. Friedl *et al.* (2018) confirmed a D_p/D_o threshold value (0.006) for denitrification-derived N₂O after applying ammonium nitrate to subtropical pasture soils with maximum N₂O emissions at $D_p/D_o = 0.006$ on day 1 of the study. However, on day 2 the N₂O emissions reached their maximum at $D_p/D_o = 0.0068$ with the shift thought to result from residual O₂ at day one and increasing anaerobic conditions on day 2, resulting in the enhanced reduction of N₂O to N₂ or entrapment of denitrified N₂O in the soil. Thus, the relationship between D_p/D_o and N₂O flux potentially alters with biological O₂ demand. After incorporating high C residues into a cropping system, high N₂O fluxes were observed at calculated $D_p/D_o > 0.02$, a value considered as a threshold for anaerobiosis (Stepniewski 1981), which was postulated to be due to the high C inputs increasing O₂ demand and denitrification activity (Petersen *et al.* 2013). In grazed

pastures, C inputs include root exudation and mineralisation of soil organic matter and these will vary with climate, soil fertility, management and soil type. Hence, the laboratory-defined threshold for peak N₂O emissions, recorded by Balaine *et al.* (2013) for only one soil, may shift due to increased O₂ demand. Interestingly, however, Owens *et al.* (2016) found that after applying ruminant urine to a pasture soil *in situ*, that the N₂O emissions only increased substantially when D_p/D_o declined to ~ 0.006 , consistent with the laboratory observations of Balaine *et al.* (2013, 2016) and Friedl *et al.* (2018). Further evaluation of N₂O emissions in relation to D_p/D_o , under controlled conditions for a range of soils, is still required to better understand the robustness of this threshold.

Thus, the objectives of this experiment were to further evaluate soil D_p/D_o in relation to the occurrence of N₂O emissions, under controlled conditions, across a wider range of soils, under a range of soil ρ_b and moistures in order to better validate the results obtained by Balaine *et al.* (2013, 2016). It was hypothesised that (i) the interactive effects of soil ρ_b and water content on D_p/D_o would result in elevated emissions of N₂O when D_p/D_o declined to a threshold close to 0.006, (ii) the robustness of D_p/D_o as an indicator for N₂O emissions would be consistent across different soils and (iii) D_p/D_o would indicate the onset of elevated N₂O emissions better than WFPS.

Materials and methods

Soil collection and experimental design

Four pasture soils were sampled (0–15 cm depth) in spring 2017: a Wakanui silt loam (Mottled Immature Pallic Soil) from the dairy farm at Lincoln University (43°38'41.3"S, 172°26'34.6"E); a Waipara loam soil (Mottled-argillic Fragic Pallic Soil) from a hill country farm at Limeworks Road, North Canterbury (42°58'2.28"S, 172°38'19.68"E); a Temuka silty loam (Typic Orthic Gley Soil) from a dairy farm near Lincoln (43°39'11.88"S, 172°29'22.92"E); and a well-drained Otorohanga loam (Typic Orthic Allophanic Soil) collected at Ruakura, AgResearch, Hamilton (37°46'44.9"S, 175°18'47.6"E). Soil classifications are as defined by Lilburne *et al.* (2012). Soils were air-dried and sieved to ≤ 2 mm and the gravimetric water content was determined (Blakemore *et al.* 1987). Soil particle densities and particle sizes (Table 1) were analysed using recognised methods (Hao *et al.* 2008; Kroetsch

Table 1. Soil (0–10 cm) texture, particle density and carbon contents

IUSS, International Union of Soil Science: clay 0–2 mm, silt 2–20 mm and sand 20–2000 mm. USDA, United States Department of Agriculture: clay 0–2 mm, silt 2–63 mm and sand 63–2000 mm

	Wakanui		Waipara		Temuka		Otorohanga	
	IUSS	USDA	IUSS	USDA	IUSS	USDA	IUSS	USDA
Clay (%) ^A	22.3	22.3	9.9	9.9	14.5	14.5	11.3	11.3
Silt (%)	53.4	72.1	22.2	46.8	34	55.7	39.7	66.6
Sand (%)	24.3	5.6	67.9	43.3	51.5	29.8	48.9	22.1
Particle density (g cm ⁻³) ^B	2.59 ± 0.01		2.65 ± 0.02		2.61 ± 0.02		2.46 ± 0.02	
Carbon (%) ^C	1.57 ± 0.06		1.17 ± 0.15		1.85 ± 0.10		2.21 ± 0.12	

^ATexture analyses were performed using a laser diffraction particle analyser (Mastersizer 3000, Malvern Panalytical, UK).

^BHao *et al.* (2008).

^CBased on loss on ignition (Blakemore *et al.* 1987).

and Wang 2008). Soil organic C contents (Table 1) were determined by loss on ignition (Blakemore *et al.* 1987). Repacked soil cores were constructed by compacting sieved soil to a depth of 5 cm into stainless steel (SS) rings (7.3 cm internal diameter, 7.4 cm deep) at bulk densities designated by treatment. The SS-rings had fine nylon mesh placed over the bottom of the ring to prevent soil egress.

The experimental design consisted of four soils, three levels of soil ρ_b for each soil and nine levels of matric potential (−0.5, −1.0, −2.0, −3.0, −4.0, −5.0, −6.0, −8.0 and −10.0 kPa), replicated four times. Soil ρ_b treatments for the Wakanui, Waipara and Temuka soils were set at 1.0, 1.1 or 1.2 Mg m^{−3}. However, due to the allophanic nature of the Otorohanga soil and the relatively high organic matter content it could not be packed at 1.2 Mg m^{−3} and so the soil ρ_b treatments for this soil were set at 0.9, 1.0 or 1.1 Mg m^{−3}.

So that the soil cores had excess NO₃-N substrate available for denitrification the soil cores were presoaked in a KNO₃ solution (1800 µg mL^{−1} NO₃-N) for 2 days before being placed on the tension tables. The aim of this was to better enable observations of the effects of soil characteristics (bulk density, matric potential and organic matter content) on N₂O emissions. Tension tables were prepared as described by Romano *et al.* (2002) and soil cores were placed on the tables to equilibrate for 4 days. Before placing soils cores on the tension tables, 10 mL of the KNO₃ solution were poured evenly across the tension tables to provide a good connection between soil cores and the tension table. Soil cores were weighed daily to determine when the equilibrium at the desired matric potential was achieved. It was physically impossible to run all soil cores simultaneously. Thus, a total of 108 soil cores (three levels of soil ρ_b × nine levels of soil matric potential × four types of soil) were on the tension tables at any given time (one replicate), with subsequent replicates run in batches (Balaine *et al.* 2013). Using a new set of cores, with air-dried repacked soil for each replicate, ensured that the initial soil NO₃[−] concentration and soil conditions were the same for each replicate. The tension tables were sited in a room where the temperature fluctuations were negligible (20 ± 1°C).

N₂O and relative gas diffusivity measurements

Nitrous oxide fluxes were measured after soil cores had attained equilibrium on the tension table (4 days). Each soil core was placed in a 1-L Mason jar, which was then sealed with an air-tight lid equipped with a septum. Gas samples (10 mL) were taken at 0, 15 and 30 min after sealing using a 25G hypodermic needle attached to a three-way stopcock, which was connected to a 20-mL glass syringe. Collected gas samples were injected into pre-evacuated 6-mL Exetainer® vials. Immediately before analysis, the gas samples were brought to ambient pressure and then analysed for N₂O on a gas chromatograph (Clough *et al.* 2009). Reference gases, N₂O in N₂ (0.2 ± 0.004, 1.0 ± 0.01, 2.0 ± 0.04 and 5.0 ± 0.1, supplied by BOC Gas New Zealand) were used for constructing standard curves. The change in N₂O concentration over time was used to calculate N₂O fluxes (Hutchinson and Mosier 1981).

Measurements of D_p/D_o were performed after N₂O emission measurement using the method described by Rolston and

Moldrup (2002). In brief, repacked soil cores were positioned, isolated, above a chamber that was flushed with an O₂-free gas mixture (90% Ar and 10% N₂) until the chamber was O₂ free. Then, the soil core base was connected to the chamber, allowing ambient air to diffuse through the soil core into the chamber. A precalibrated sensor (KE-12, Figaro Inc.) recorded the increase in the O₂ concentration in the chamber. Subsequently, the method of Rolston and Moldrup (2002) was used to calculate D_p/D_o . The natural logarithm of the relative concentration of O₂ in the chamber (ln C_r) was calculated, and C_r was calculated as follows:

$$C_r = \frac{C_g - C_s}{C_0 - C_s}$$

where C_g is the concentration of O₂ in the chamber at a time t , C_0 is the concentration of O₂ in the chamber at the beginning of the experiment ($t = 0$) and C_s is the O₂ concentration (20.9%) above the soil core. The linear slope of the plot of ln C_r vs t was determined and is equal to:

$$- D_p \alpha_1^2 / \varepsilon$$

Then D_p was determined using the calculated value of ε and the value of α_1 taken from table 46–1 in Rolston and Moldrup (2002). The soil gas diffusion coefficient in air (D_o) was calculated according to Currie (1960).

Soil analyses

After taking N₂O samples and measuring D_p/D_o the soil cores were extruded into a Ziploc® plastic bag. The soil was well mixed before taking a 10-g subsample to determine gravimetric water content at 105°C for 24 h. A calibrated flat-surface pH electrode was used to measure the pH of the extruded-mixed soil (Broadley James Corp., Irvine, CA, USA). The equivalent of 4 g of dry soil was extracted with 40 mL of 2 M KCl for 1 h to determine inorganic-N concentrations. After filtering (Whatman 42) the extracts were analysed for NO₃[−]-N and NH₄⁺-N on a flow injection analyser (Blakemore *et al.* 1987). Similarly, dissolved organic carbon (DOC) was extracted using 5-g equivalent of dry soil and 30 mL of deionised water shaken for 30 min before centrifugation (2280 × g for 20 min, at 25°C, model Kubota 8420) and filtration (Whatman 42), with analyses performed on a Shimadzu TOC analyser (Shimadzu Oceania Ltd, Sydney, Australia).

Statistical analyses

Statistical analyses were performed using R studio (version 3.4.3). Data were tested for normality, residual repartition and the homoscedasticity. A log-transformation was applied to the N₂O data where conditions for normality were not met. A repeated-measures analysis, using two-way ANOVA, with matric potential and soil ρ_b as factors, was used to test for overall treatment differences between measured variables, with Tukey's *post-hoc* test used to determine specific differences between means. Comparisons were made between soil ρ_b treatments within soils and across matric potential, and where common soil ρ_b occurred comparisons were made across soils.

Results

Soil chemical and physical properties

Soil pH varied with soil type ($P < 0.05$): averaged across soil matric potential, the Waipara, Otorohonga, Temuka and Wakanui soils had soil pH of 5.15, 5.28, 5.73 and 6.00 respectively. As drainage increased (more negative soil matric potential) the soil pH within each soil also decreased ($P < 0.05$; Fig. 1). The most significant decrease in soil pH was for the Wakanui soil: at -0.5 kPa, soil pH range was 6.34–6.41 and declined to 5.67–5.70 at -10 kPa. For each soil type, increasing soil ρ_b generally resulted in an increase in soil pH at all levels of soil matric potential ($P < 0.05$; Fig. 1).

Soil DOC concentrations varied with soil type ($P < 0.05$): averaged across soil matric potential, the Waipara, Otorohonga, Temuka and Wakanui soils had DOC concentrations of 75, 114, 162 and 208 $\mu\text{g g}^{-1}$ soil respectively. These soil DOC concentrations either remained relatively stable (Temuka and Waipara soils) or tended to decline (Wakanui and Otorohonga soils) as soil matric potential became more negative (Fig. 2); however, there was no significant interaction between soil ρ_b and soil matric potential on DOC concentrations within a given soil ($P > 0.05$). Neither soil ρ_b nor soil matric potential caused significant changes to DOC concentration for the Waipara or Temuka soils ($P > 0.1$). However, in the Wakanui and Otorohonga soil, DOC concentrations varied with soil ρ_b ($P < 0.05$): DOC concentrations were higher for the highest

soil ρ_b (1.2 and 1.1 Mg m^{-3}) than for the lowest soil ρ_b (1 and 0.9 Mg m^{-3}). In these same soils, the soil DOC concentrations were significantly lower at -8 and -10 kPa than at -0.5 and -1 kPa ($P < 0.05$, Fig. 2).

There was no consistent effect of soil matric potential on soil NO_3^- -N concentrations (Fig. 3). However, soil ρ_b did affect soil NO_3^- -N concentrations in the Wakanui soil when averaged across soil matric potential: at 1.0 Mg m^{-3} these were higher ($P < 0.05$) than at 1.1 and 1.2 Mg m^{-3} . In the Temuka soil, the NO_3^- -N concentrations also declined ($P < 0.05$) as soil ρ_b increased – when averaged across soil matric potential, equalling 741, 644 and 555 mg NO_3^- -N kg^{-1} of soil at soil ρ_b of 1.0, 1.1 and 1.2 Mg m^{-3} respectively. Similarly, in the Otorohonga soil at soil ρ_b of 0.9, 1.0 and 1.1 Mg m^{-3} average NO_3^- -N concentrations were 751, 688 and 604 NO_3^- -N kg^{-1} respectively ($P < 0.05$). The Waipara soil was the only soil where NO_3^- -N concentrations did not vary with soil ρ_b .

As expected, the soil WFPS decreased with progressively more negative soil matric potentials ($P < 0.05$) but the rate of decrease varied with soil type: averaged across soil matric potential and soil ρ_b , the Wakanui, Otorohonga, Waipara and Temuka soils had WFPS values of 85.4%, 85.7%, 86.3% and 90.6% respectively, with corresponding ranges of 60.0–97%, 55.1–100%, 61.4–100% and 68.7–100%. Soil WFPS also varied with soil ρ_b ($P < 0.05$), with the lowest WFPS observed at the lowest soil ρ_b ; 1 Mg m^{-3} for the Wakanui, Waipara and Temuka soils and 0.9 Mg m^{-3} for the Otorohonga soil. An interaction between soil ρ_b and matric potential resulted

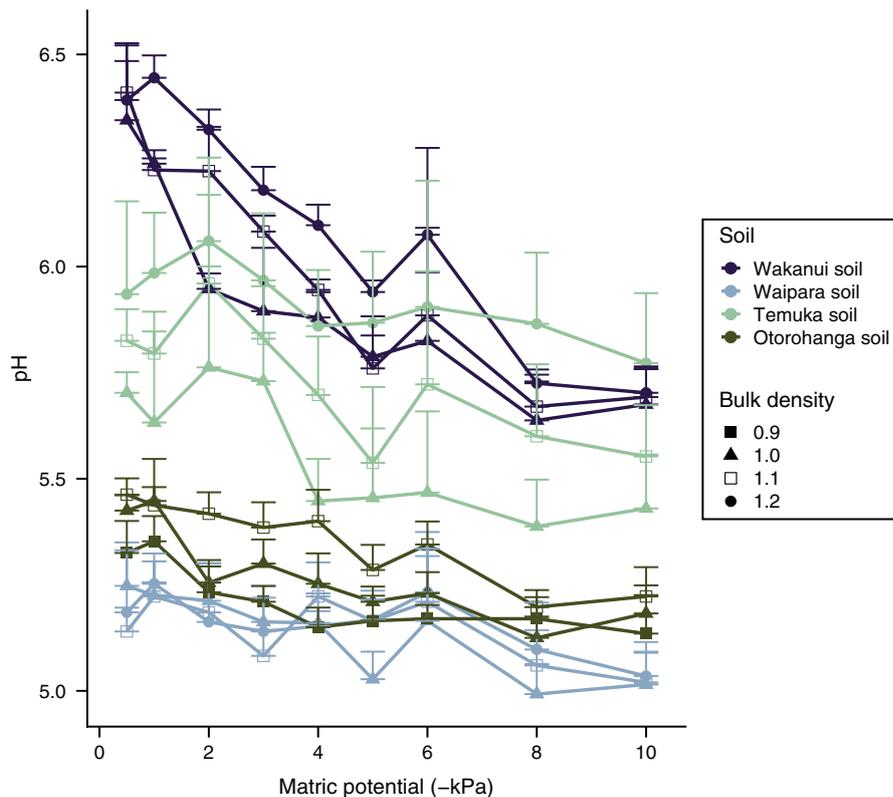


Fig. 1. Mean soil pH versus soil matric potential. Numerals in the legend indicate soil bulk density treatments applied (Mg m^{-3}). Error bars = s.e.m., $n = 4$.

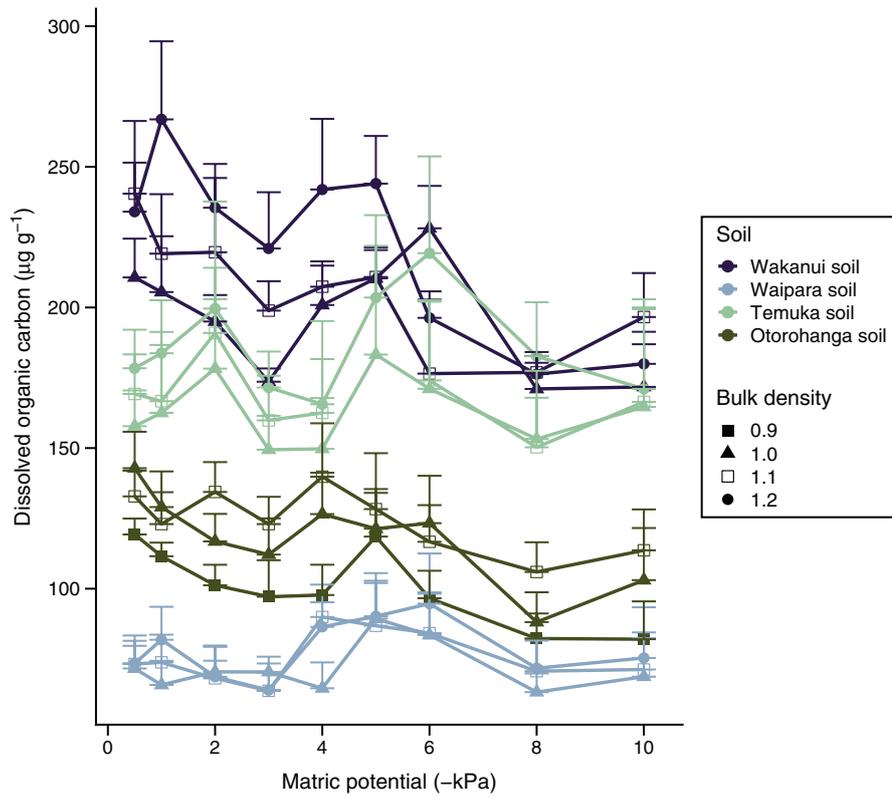


Fig. 2. Mean dissolved organic carbon concentration ($\mu\text{g g}^{-1}$) versus soil matric potential. Numerals in the legend indicate soil bulk density treatments applied (Mg m^{-3}). Error bars = s.e.m., $n = 4$.

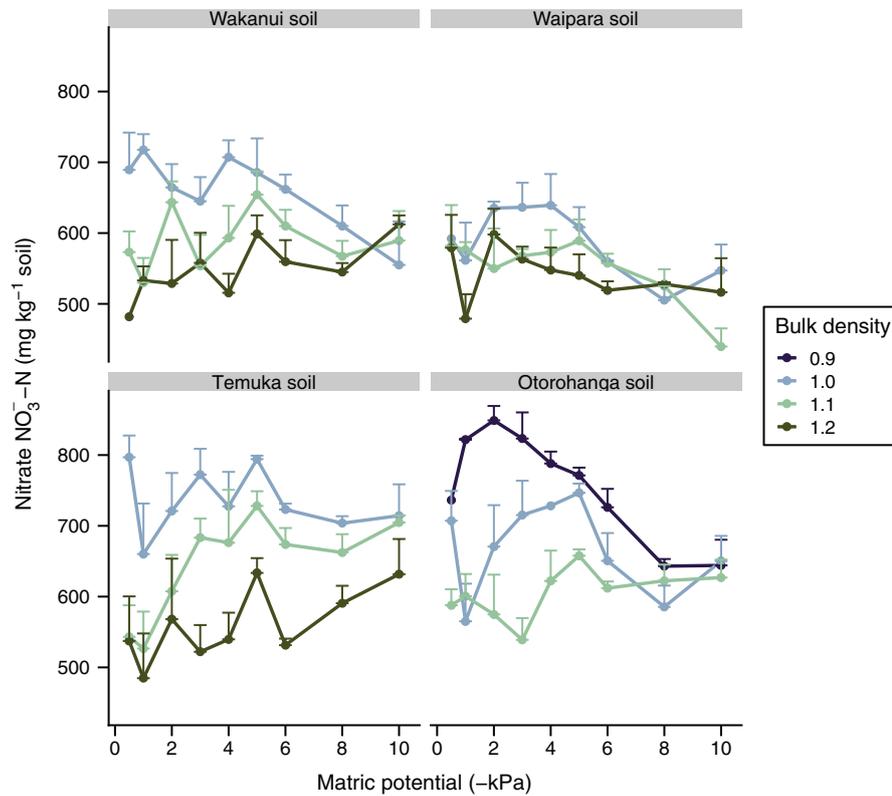


Fig. 3. Soil nitrate concentrations versus soil matric potential for the four soils. Numerals in the legend indicate soil bulk density treatments applied (Mg m^{-3}). Error bars = s.e.m., $n = 4$.

in WFPS declining at a faster rate at lower soil ρ_b values for all soils, except the Waipara soil (Fig. 4).

The range in measured mean D_p/D_o across the soil matrix potential treatments, when averaged across all soil ρ_b treatments, was 0.003–0.014 for the Wakanui, 0.003–0.015 for the Waipara, 0.004–0.007 for the Temuka soil and 0.005–0.017 for the Otorohanga soil. An interaction between soil ρ_b and soil matrix potential affected D_p/D_o in the Wakanui soil, where D_p/D_o at 1.2 Mg m⁻³ was lower ($P < 0.05$) than at 1.0 Mg m⁻³ for soil matrix potentials below -2 kPa (Fig. 5). The same was observed in the Temuka soil, except that D_p/D_o was significantly lower at 1.2 Mg m⁻³ compared with at 1.0 Mg m⁻³, for soil matrix potentials below -3 kPa (Fig. 5). Similarly, D_p/D_o values for 1.0 and 1.1 Mg m⁻³ were lower for the 0.9 Mg m⁻³ treatment in the Otorohanga soil below -2 kPa (Fig. 5). However, in the Waipara soil there was no effect of soil ρ_b on D_p/D_o (Fig. 5). A plot of D_p/D_o vs soil air-filled porosity showed D_p/D_o values over a range of air-filled porosity of 0–0.3 cm³ cm⁻³ (Fig. 6).

Soil N₂O fluxes and relationships with soil physical parameters

For each soil, the N₂O-N fluxes were highest at the lowest soil matrix potential (-0.5 kPa), with N₂O-N fluxes decreasing as soil matrix potential decreased (Fig. S1, available as Supplementary material). The range in the N₂O-N fluxes

varied with soil type ($P < 0.05$). The highest fluxes occurred in the Otorohanga soil (0.12–691 mg m⁻² h⁻¹), closely followed by the Wakanui soil (0.08–660 mg m⁻² h⁻¹). The Temuka soil N₂O-N fluxes ranged within 0.22–408 mg m⁻² h⁻¹ and the lowest N₂O-N fluxes occurred in the Waipara soil with 0.07–124 mg m⁻² h⁻¹. Soil N₂O-N fluxes ($P < 0.05$) were higher with increasing soil ρ_b , and in the Temuka and Otorohanga soils where soil ρ_b and soil matrix potential interacted ($P < 0.05$): soil N₂O-N fluxes decreased more slowly at the highest soil ρ_b as soil matrix potential became more negative.

As D_p/D_o declined, the N₂O-N flux increased with peak N₂O fluxes at a D_p/D_o value close to or less than 0.006 (Fig. 7). Plotting N₂O-N fluxes vs WFPS (Fig. 8) or volumetric soil water content (Fig. S2, available as Supplementary material) showed no clear relationship across soils or soil ρ_b ; but N₂O-N fluxes increased with increasing soil moisture, peak N₂O-N fluxes occurred at varying WFPS or volumetric soil water content values depending on soil type and soil ρ_b (Fig. 8, Fig. S2).

Discussion

Soil chemical and physical characteristics

Minimum soil concentrations of NO₃⁻ and DOC, required to support denitrification in soil, have previously been reported to be ≥ 5 mg N kg⁻¹ soil and 40 mg C kg⁻¹ soil respectively (Beauchamp *et al.* 1980; Ryden 1983). High concentrations of

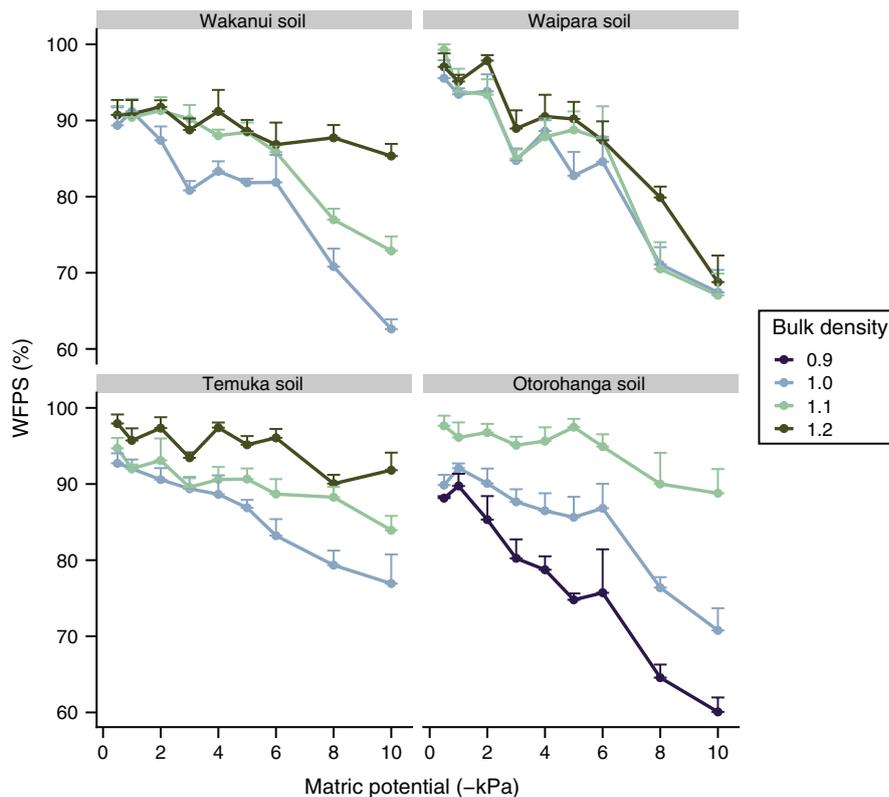


Fig. 4. Water-filled pore space (WFPS, %) versus soil matrix potential for the four soils. Numerals in the legend indicate soil bulk density treatments applied (Mg m⁻³). Error bars = s.e.m., $n = 4$.

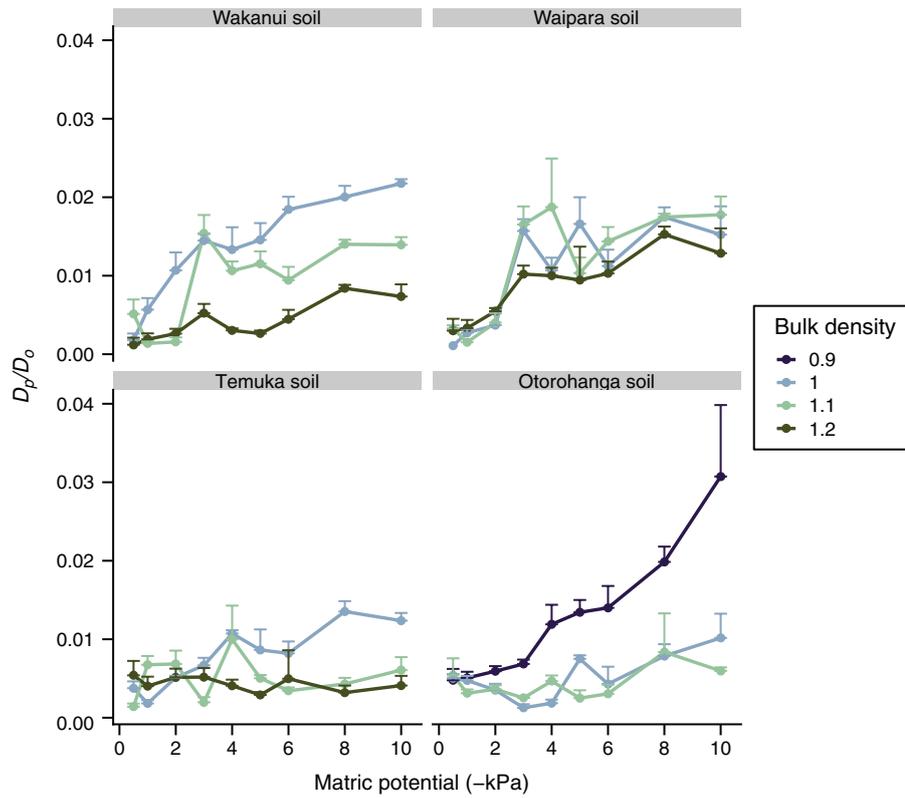


Fig. 5. Relative gas diffusivity (D_p/D_o) at varying soil bulk density and soil matric potential. Numerals in the legend indicate soil bulk density treatments applied ($Mg\ m^{-3}$). Error bars = s.e.m., $n = 4$.

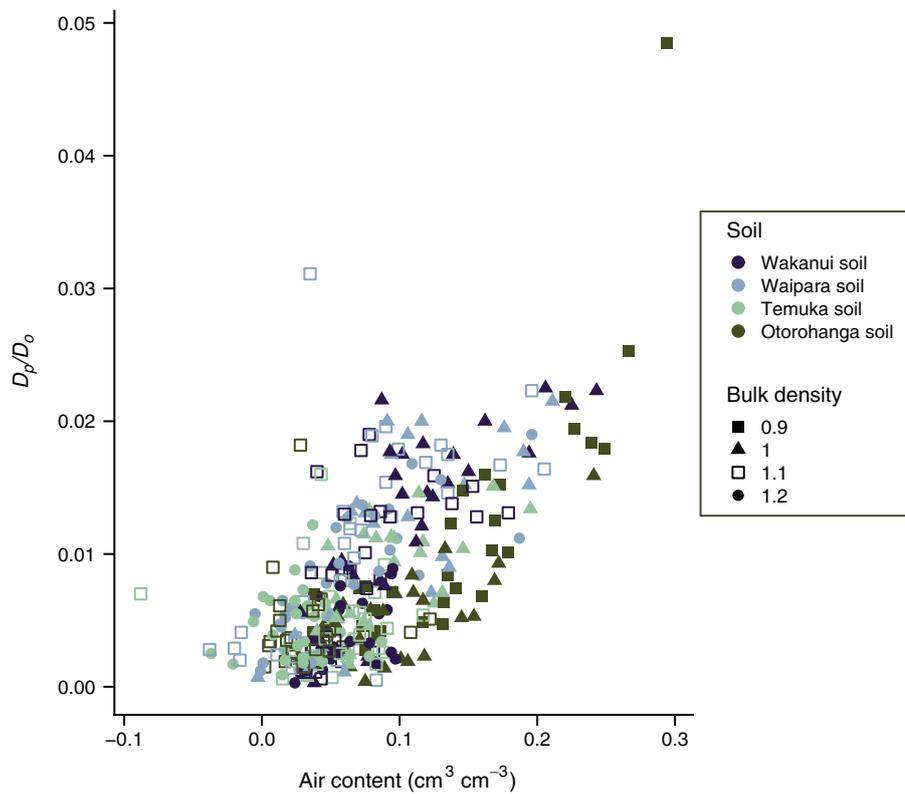


Fig. 6. Soil relative gas diffusivity (D_p/D_o) versus soil air-filled porosity ($cm^3\ cm^{-3}$).

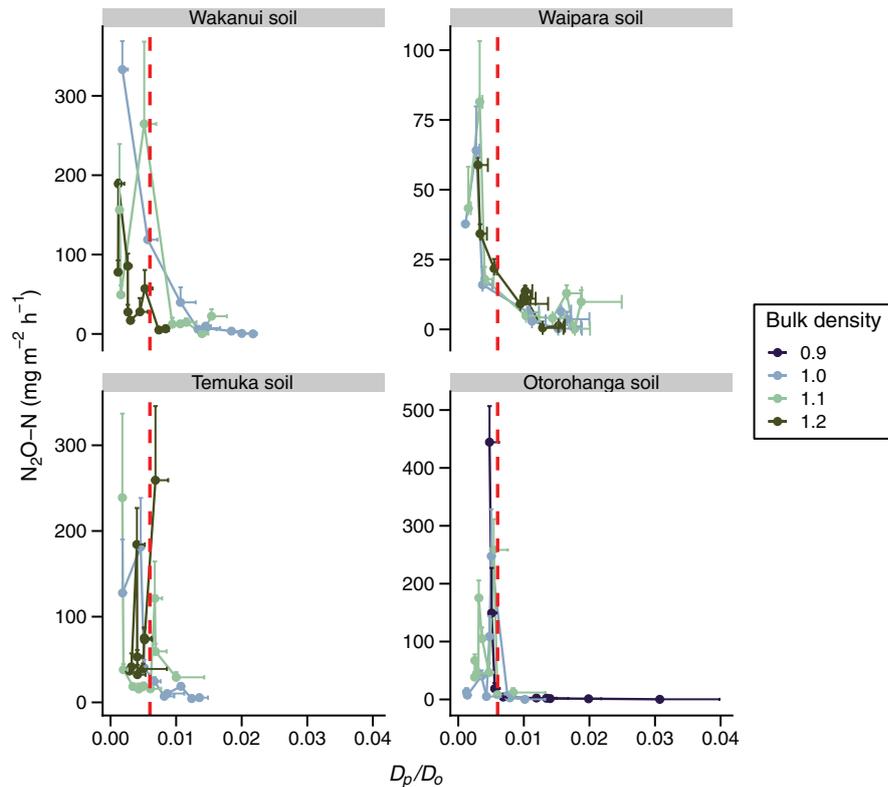


Fig. 7. Relationship between measured N₂O-N fluxes and measured D_p/D_o for each soil separately and at varying soil bulk density. Numerals in the legend indicate soil bulk density treatments applied (Mg m^{-3}). Error bars = s.e.m., $n = 4$.

NO_3^- increase the denitrification rate in the presence of a C substrate (Weier *et al.* 1993) because the DOC assumes the role of the electron donor during NO_3^- reduction under denitrifying conditions. Thus, based on the results of this experiment, it can be assumed that the soil NO_3^- -N and DOC concentrations, which were comparable in magnitude to those previously observed by Balaine *et al.* (2013), were not limiting for denitrification regardless of soil type or treatment.

Elevated soil pH values, observed in the higher soil ρ_b treatments, are consistent with such conditions creating more anaerobic conditions (lower D_p/D_o and higher WFPS), suitable for denitrification which results in a net release of OH^- ions (Wrage *et al.* 2001). Declines in soil pH with increasing soil drainage were likely the result of reduced denitrification rates or increased rates of mineralisation with ensuing nitrification and subsequent soil acidification as soils were drained.

Soil WFPS decreased as soil matric potential became more negative, with the rate of decrease lower at higher soil ρ_b because increasing soil ρ_b (compaction) not only decreases total porosity but also creates a shift in pore size distribution, observed as a reduction in macroporosity and an increase in microporosity (Chamindu Deepagoda *et al.* 2019b). This results in an increase in the air-entry pressure and a decrease in air-permeability making the soils relatively more anaerobic as soil ρ_b increases at a given soil matric potential. Soil texture also affects pore size distribution and the reason that the Waipara soil WFPS was less affected by

increasing bulk density was most likely the higher sand content of this soil, facilitating the retention of a higher fraction of pore space with macroporosity.

Values of D_p/D_o decreased when both soil ρ_b and soil matric potential increased, resulting in a concurrent decline in air-filled porosity and increasing soil moisture, which in turn enhanced tortuosity of the soil pore network (Chamindu Deepagoda *et al.* 2019a). Soil D_p/D_o values were within the range previously observed for repacked soil cores held over a similar range of soil ρ_b and moisture (Balaine *et al.* 2013).

Soil N₂O fluxes and relationships with soil physical parameters

Based on the prior studies of Balaine *et al.* (2013, 2016), it was hypothesised that soil N₂O emissions would become elevated as D_p/D_o decreased toward 0.006. Using only one soil type, Balaine *et al.* (2013) found that soil N₂O fluxes peaked at D_p/D_o of 0.006, regardless of soil ρ_b , and declined as D_p/D_o decreased further. The increase in N₂O fluxes occurred as the increasingly anaerobic conditions, as defined by a decline in D_p/D_o , created an environment suitable for N₂O production. The decline in N₂O fluxes, as D_p/D_o decreases further, has been shown to be due to N₂O being denitrified to N₂ (Klefoth *et al.* 2014; Balaine *et al.* 2016). In the current study, the interactive effects of soil ρ_b and soil matric potential generally resulted in enhanced N₂O fluxes at D_p/D_o close to 0.006, or

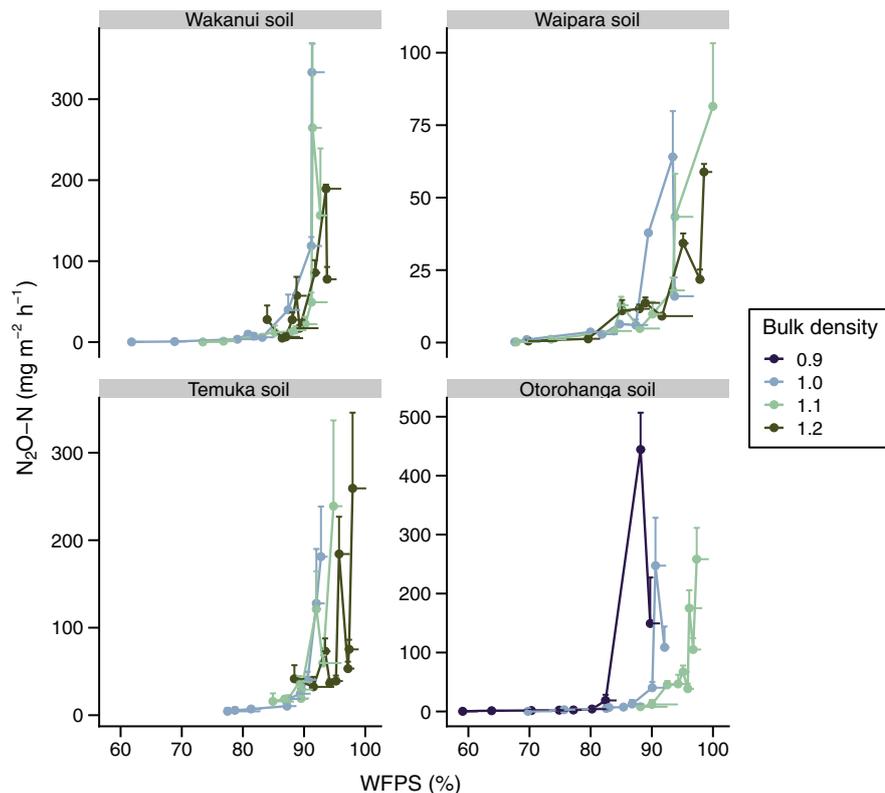


Fig. 8. Relationship between measured $\text{N}_2\text{O-N}$ fluxes and WFPS (%) for each soil at varying soil bulk density. Numerals in the legend indicate soil bulk density treatments applied (Mg m^{-3}). Error bars = s.e.m., $n = 4$.

less, but not necessarily peaking at 0.006. Thus, under the conditions of this study, the variation in the soil C content, postulated to potentially alter the level of O_2 consumption, did not cause a significant shift in the D_p/D_o value where N_2O emissions readily increased. Following a freeze–thaw event, the presence of labile C inducing an increased O_2 demand was postulated by Petersen *et al.* (2013) to be a reason for denitrification activity at $D_p/D_o > 0.02$, a threshold normally considered to indicate the onset of anaerobiosis (Stepniewski 1981). However, Owens *et al.* (2017) found that in ruminant urine-affected soil, *in situ*, N_2O fluxes also increased when the soil D_p/D_o was ~ 0.006 , despite labile C being supplied via plant root exudation. Chamindu Deepagoda *et al.* (2019a) found that N_2O emissions peaked in intact soil cores taken from pastures over a D_p/D_o range of 0.005–0.01. Friedl *et al.* (2018) also found N_2O and N_2 fluxes from subtropical dairy pasture soils increased exponentially as D_p/D_o declined, with maximum N_2O fluxes at D_p/D_o of 0.0068. Hence, these current results examining a wider number of soils sit well with previous work and support the use of D_p/D_o as an interpretive tool for understanding the occurrence of N_2O emissions or for predicting the potential for N_2O emissions given soil ρ_b and soil moisture data are available for calculating (e.g. Moldrup *et al.* 2013). The potential onset of soil N_2O emissions has often been considered by determining the degree of WFPS; but, as Farquharson and Baldock (2008) explain, for a given WFPS the volume fraction

of air varies depending on the soil ρ_b . The variable D_p/D_o is an integrated measure of the interactive effects of soil ρ_b and WFPS on air-filled porosity and thus gas diffusion. Hence, the onset of N_2O emissions occurs over a relatively wide range of WFPS. This was most noticeable in the Otorohanga soil.

However, the magnitude of the N_2O fluxes, and the anticipated decline in the N_2O fluxes occurring for $D_p/D_o < 0.006$, was not consistent across soils. In the Otorohanga soil, where the highest N_2O fluxes occurred, N_2O emissions peaked at D_p/D_o of 0.006 regardless of soil ρ_b , and then declined; in the Waipara soil where the lowest N_2O fluxes occurred, N_2O emissions peaked at a similar D_p/D_o , but < 0.006 , regardless of soil ρ_b before declining. This variation in the magnitude and the decline in the N_2O fluxes for $D_p/D_o < 0.006$ may be due to other factors affecting the denitrification rate and the $\text{N}_2\text{O}:(\text{N}_2\text{O} + \text{N}_2)$ ratio. Soil O_2 supply acts as the primary determinant of denitrification commencing. Friedl *et al.* (2018) found a shift in the N_2O response to D_p/D_o between days 1 and 2 after rewetting of subtropical pasture soils and proposed the presence of residual O_2 on day 1 and increasing anaerobic conditions on day 2 to explain the proposed effect of D_p/D_o on a relative shift in $\text{N}_2:\text{N}_2\text{O}$ partitioning. However, other soil variables, soil pH and anoxic C mineralisation, have also been shown to influence both the denitrification rate and the $\text{N}_2\text{O}:(\text{N}_2\text{O} + \text{N}_2)$ ratio in pasture soils, with liming and enhanced C supply promoting N_2O reduction to N_2 and

denitrification rates (Samad *et al.* 2016; Senbayram *et al.* 2019). Conversely, elevated soil NO₃⁻ concentrations may prevent N₂O reduction, thus enhancing N₂O:(N₂O + N₂). Recently, this effect of soil NO₃⁻ concentration was shown to override the effect of liming with respect to N₂O:(N₂O + N₂) in a sandy cropping soil; high concentrations of NO₃⁻ (45 mg N kg⁻¹ soil) almost completely inhibited N₂O reduction (Senbayram *et al.* 2019). Hence, the variation in the four soils used in the current study, in terms of the delayed or lack of N₂O reduction, may have been a partial consequence of the relatively high soil NO₃⁻ concentrations enhancing N₂O:(N₂O + N₂). Future research is needed to examine the role of these factors in altering N₂O:(N₂O + N₂) with respect to the soil D_p/D_o value. In conclusion, this study shows that across a range of soils, varying in texture and C content, the soil D_p/D_o value is a valuable, and theoretically robust, tool for determining the onset of N₂O emissions when denitrification substrates are present.

Conflicts of interest

The authors declare no conflicts of interest.

Acknowledgements

The authors gratefully acknowledge funding (SOW14-GPLER-SP198-LIN) from the New Zealand Fund for Global Partnerships in Livestock Emissions Research (GPLER), an international research fund set up by the New Zealand Government in support of the Global Research Alliance on Agricultural Greenhouse Gases (GRA). This paper forms part of the PhD thesis of Camille Rousset (2020).

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Handling Editor: Iris Vogeler