Effect of nitrification inhibitor on nitrous oxide emissions in pasture soils

Jiafa Luo[A], Cecile de Klein[B], Mark Shepherd[A] and Stewart Ledgard[A]

[A]AgResearch Ruakura, Hamilton, New Zealand, Email Jiafa.luo@agresearch.co.nz, Mark.shepherd@agresearch.co.nz, Stewart.ledgard@agresearch.co.nz
[B]AgResearch Invermay, Mosgiel, New Zealand, Email Cecile.deklein@agresearch.co.nz

Abstract
In this paper we present findings from lysimeter studies that determined the effectiveness of the nitrification inhibitor dicyandiamide (DCD) on decreasing nitrous oxide (N_2O) emissions from dairy cow urine (loading 1000 kg N/ha) applied to three different soils in New Zealand under two rainfall regimes (annual average rainfall for the lysimeter site, 1100 mm, and twice the average, 2200 mm). Total N_2O emissions from the urine over the measurement period of 5 months varied significantly between the three soils, from 2.94 to 35.9 kg N_2O-N/ha. The DCD treatment reduced these N_2O emissions to between 2.45 and 19.5 kg N_2O-N/ha. The average N_2O emission factors of the urine-N (EF3) were 2.0% and 1.6% under the 1100 mm and 2200 mm rainfall regimes, respectively. These were decreased to 1.3% by the application of DCD under both rainfall regimes. The results indicate that under heavy rainfall DCD effectiveness may be reduced. The very wet (anaerobic) soil conditions may have resulted in the production of di-nitrogen gas (N_2), rather than N_2O gas. This could have influenced the effectiveness of DCD on reducing N_2O emissions.

Key Words
Nitrous oxide, grazed pasture, nitrification inhibitor, dicyandiamide, DCD.

Introduction
Nitrous oxide is produced in soils during the microbiological processes of nitrification and denitrification. N_2O production by nitrifying bacteria may arise either during NH_4^+ oxidation to nitrate (NO_2^-) or during dissimilatory NO_2^- reduction when oxygen supply is limited. During denitrification, N_2O is an intermediate in the dissimilatory reduction of nitrate (NO_3^-) and/or NO_2^- to N_2 under anaerobic conditions and is, therefore, both produced and consumed by denitrifying bacteria in soil (Bolan *et al.* 2004). Research in New Zealand (NZ) and overseas has shown that nitrogen (N) from urine patches is the major source of N_2O loss from grazed pastures (e.g. de Klein and Eckard 2008). One of the proposed options for reducing N_2O emissions is the use of nitrification inhibitor dicyandiamide (DCD; e.g. de Klein and Eckard 2008; Luo *et al.* 2010). DCD delays the bacterial oxidation of NH_4^+ to NO_3^- in the soil for a certain period by depressing the activity of *Nitrosomas* bacteria, therefore slowing nitrification and N_2O production. Research conducted largely in the South Island of NZ has shown that DCD can substantially reduce N_2O emissions from urine patches and grazed pastures with reduction potentials ranging from 60 to 80% (e.g. Di *et al.* 2007; Smith *et al.* 2008). Rainfall or irrigation is one of the key environmental drivers for N_2O emissions. In addition, rainfall/irrigation can displace DCD down the soil profile, away from the soil ammonium source, but very little is known about the impact of DCD displacement on its effectiveness for inhibiting nitrification. Therefore, there is need to examine the effectiveness of DCD on decreasing N_2O emissions from contrasting soils and under contrasting climatic conditions. In this paper we report results from a field lysimeter study which investigated the effect of DCD on N_2O emission on three pasture soils in the North Island of NZ under two rainfall regimes. A parallel study was also conducted on three soils in the South Island of NZ at the Lincoln university site (Di *et al.* 2010).

Material and methods
Lysimeter collection and installation
Intact soil monolith lysimeters (50 cm diameter by 70 cm deep) were collected from the Waikato, Rotorua and Northland regions in the North Island of NZ: free draining Horotiu silt loam (Typic Orthic Allophanic Soil), free draining Oropi sand (Buried Allophanic Orthic Pumice Soil) and slowly draining Waikare clay soil (Gleyed clay alluvial fulvi-appodic Soil) (Hewitt 1993). Measured soil properties are presented in Table 1. The pastures were a permanent mixed pasture of perennial ryegrass (*Lolium perenne* L.) and white clover (*Trifolium repens* L.) and had been under a cutting regime for 4 months prior to collection of the lysimeters to avoid the presence of fresh urine patches. The lysimeters were extracted by placing a metal cylinder casing with an internal cutting ring onto the soil surface, and carving around the cylinder edge while gradually pushing the cylinder down until the desired depth was reached. The soil monolith was then cut

1 – 6 August 2010, Brisbane, Australia. Published on DVD.

from the subsoil and removed from the collection site. The internal cutting ring created a small gap between the soil monolith and the cylinder wall, which was sealed using petroleum jelly to prevent edge-flow effects (Cameron *et al.* 1992). The lysimeters were transported and installed at ground level at the AgResearch Ruakura Research Centre lysimeter facility in Hamilton, NZ. In addition, 12 “mini-lysimeters” (25 cm deep and 4 for each soil type) were installed by the same method.

Table 1. Soil chemical (0-7.5 cm) and physical (0-70 cm) properties of the three soils.

Soil type	Waikato Horotiu silt loam	Rotorua Oropi sand soil	Northland Waikare clay
pH (H_2O)	5.9	5.7	5.9
Organic C (%)	6.5	10.7	9.6
Total N (%)	0.56	0.65	0.86
Olsen P (mg/kg)	16.0	53.0	56.0
CEC ($cmol_c$/kg)[A]	22.0	15.0	31.0
Base saturation (%)	53.0	41.0	78.0
Particle size (%)			
Sand	34.5	63.3	2.3
Silt	50.8	27.5	20.5
Clay	14.8	9.3	77.3

[A]cation exchange capacity

Treatments

Treatments included: Control (no urine and no DCD); Urine alone, applied on 15 May 2008 and Urine plus a nitrification inhibitor (DCD solution @10 kg DCD/ha), applied in May 2008, with a second application of DCD in July 2008. The urine was applied as fresh cow urine, adjusted for N concentration to achieve a loading of 1000 kg N/ha applied in a single application. The DCD was applied in solution sprayed onto the soil surface after urine addition (1 mm). Two nominal annual rainfall regimes (1,100 and 2,200 mm) were employed by using simulated rainfall/irrigation as required. Simulated rainfall was applied weekly to the appropriate lysimeters as spray irrigation to meet the targeted annual rainfall regimes. Total water input amounts (including natural rainfall and added water) were 756 and 1,374 mm over the period of the trial (5 months from May to Sept 2008) for the nominal annual rainfall regimes of 1,100 mm and 2,200 mm, respectively. There were 4 replicates of each treatment, except for the control, for which measurements of N_2O emissions were made from two mini lysimeters (25 cm diameter) for each soil type at each rainfall rate.

N_2O sampling and analysis

N_2O emission measurements were made using a soil cover techniques by fitting headspace chambers to the top of the lysimeters. The rim of each lysimeter was fitted with a channel filled with water to form a gas-tight seal when the headspace chambers were in place during measurement. The sampling procedure and N_2O emission calculation have previously been reported (Luo *et al.* 2008). Emission factors (EF3, N_2O-N emitted as % of N applied) were also calculated. Statistical analysis of the log-transformed data was performed using GenStat and least significant ratios (LSR) were calculated to compare the differences.

Results and discussion

Daily N_2O emissions

The N_2O fluxes from the control treatments (no urine and no DCD) remained low over the sampling period, confirming that most of the N_2O from grazed pastures is derived from animal urine. Dairy cow urine application sharply increased the initial N_2O fluxes on all three soils. The initial peaks occurred within a couple of days after urine application. The magnitude of the N_2O fluxes varied between soils. The N_2O fluxes from application of urine were lower from the Waikato Horotiu silt loam soil than from the Northland Waikare clay and Rotorua Oropi sand soils at most sampling times. At the 1,100 mm water regime, the DCD application significantly ($p<0.05$) reduced N_2O emissions on the Waikato soil and Rotorua soil, with lower N_2O fluxes from the soils treated with DCD than from the soils without DCD on most sampling days within a month after urine application. However, the DCD application had little effect on N_2O emissions on the Northland soil. At 2,200 mm water regime, the DCD application reduced N_2O emissions on the Rotorua soil, but had little effect on N_2O emissions on the Waikato and Northland soils.

Total N_2O emissions

At 1,100 mm water regime, total N_2O emissions from the urine treatments over the measurement period of 5 months were 3.39, 23.4 and 35.9 kg N_2O-N/ha on the Waikato, Northland and Rotorua soils, respectively (Table 2). DCD application significantly ($p<0.05$) reduced the N_2O emissions from the Waikato and Rotorua soils. The EF3 values ranged from 0.3% to 3.5% for the urine treatment and 0.2 to 1.9% for the Urine + DCD treatment. The average reduction in the EF3 value due to DCD use was 35%.

Table 2. Total N_2O emissions and EF3 as affected by soil, water input, and urine-N and DCD. Bracketed values are standard errors of the mean (SEM).

Soil	Water input (mm/y)	Treatment	N_2O emissions (kg N/ha)	EF3 (%)	Reduction of EF3 by DCD (%)
Waikato, Horotiu	1,100	Control	0.550		
		Urine	3.39 (0.29)	0.3	
		Urine + DCD	2.45 (0.14)	0.2	33.3
Northland, Waikara	1,100	Control	0.299		
		Urine	23.4 (1.49)	2.3	
		Urine + DCD	19.1 (3.80)	1.9	17.4
Rotorua, Oropi	1,100	Control	0.582		
		Urine	35.9 (6.26)	3.5	
		Urine + DCD	19.5 (2.94)	1.9	45.7
Average under 1,100 mm rainfall regime					*35.0*
Waikato, Horotiu	2,200	Control	0.318		
		Urine	2.94 (0.33)	0.3	
		Urine + DCD	2.94 (0.30)	0.3	0
Northland, Waikara	2,200	Control	0.368		
		Urine	18.6 (1.12)	1.8	
		Urine + DCD	18.5 (1.76)	1.8	0
Rotorua, Oropi	2,200	Control	0.304		
		Urine	27.4 (1.71)	2.7	
		Urine + DCD	17.4 (4.54)	1.7	37.0
Average under 2,200 mm rainfall regime					*18.8*
Least significant ratios (LSR) for treatments (exclusive of control)			1.31	1.33	
Average under both rainfall regimes					27.8

At 2,200 mm water regime, total N_2O emissions from the urine treatments were 2.94, 18.6 and 27.4 kg N_2O-N/ha on the Waikato, Northland and the Rotorua soils, respectively (Table 2). Under the higher water regime, DCD significantly ($p<0.05$) reduced N_2O emissions only on the Rotorua soil. The EF3 values for the urine treatment ranged from 0.3% to 2.7%. DCD application reduced the EF3 value from the Rotorua soil by 37%. Under both rainfall regimes the average EF3 was 1.8% for the three soils and this was decreased to 1.3%, representing a 28% reduction.

The lower N_2O emissions observed from the Waikato soil are not unexpected as low emissions from this free-draining soil have been reported before (de Klein *et al.* 2003). Results from a concurrent study showed that N leaching losses of this soil were 40% higher than from the other soils (data not shown). The Rotorua soil is also classified as a free-draining soil but has a relatively high organic C content (Table 1), which could explain the high emissions observed from this soil. The high emissions from the Northland soil are likely to be due to prolonged wet soil conditions in this slow draining clay soil.

The effectiveness of DCD in reducing EF3 on the three North Island soils under both water input conditions was lower than that for the three South Island soils in a parallel study at the Lincoln site (Di *et al.* 2010). The average EF3 was 2.3% on the three South Island soils and this was decreased to 0.9%, representing a 61% reduction. The smaller DCD effectiveness on the three North Island soils may have been due to the above average rainfall during the measurement period, and possibly due to the milder North Island temperatures (data not shown), as both rainfall and temperature influence DCD movement and longevity. The results from this study indicate that under heavy winter rainfall DCD effectiveness may be reduced. This could either be due to DCD displacement done the profile or due to the very wet (anaerobic) soil conditions that may have resulted in the production of N_2, rather than N_2O gas from denitrification. Both these factor could have influenced the effectiveness of the DCD on reducing N_2O emissions.

A heavy and intensive rainfall event (total rainfall for 24 hours was 48 mm, of which 21 mm occurred over a 4 hour period) about a month after treatments were applied on the three soils caused large amounts of NO_3^- to be leached out of the soils (up to 29 kg leached N as a result of this rainfall; Shepherd *et al.* 2009). Consequently, N_2O fluxes rapidly declined after the rainfall. These results from North Island soils suggest that rainfall patterns (not just total annual rainfall) can be an important driver that determines DCD effectiveness for reducing N_2O emissions.

Conclusion

There were variations in the effectiveness of DCD to reduce N_2O emissions from animal urine. DCD was most effective in the Rotorua sand that exhibited the highest N_2O emission during the measurement period. Rainfall also altered DCD effectiveness with a lower reduction being found under the higher rainfall regime. The results also suggest that rainfall patterns (not just total amount of rainfall) can be an important driver that determines DCD effectiveness for reducing N_2O emissions. Overall, the DCD was not as effective in reducing N_2O emissions from the three soils at the Ruakura site as it was found to be in the soils at the South Island site (Di *et al.* 2010).

Acknowledgments

This study was funded by Ministry of Agriculture and Forestry, and through the P21 Environment programme (C10X0603) funded by Foundation for Research Science & Technology, DairyNZ, Fonterra and Meat & Wool NZ.

References

Bolan NS, Saggar S, Luo J, Bhandral R, Singh J (2004) Gaseous emissions of nitrogen from grazed pastures: processes, measurements and modelling, environmental implications, and mitigation. *Advances in Agronomy* **84,** 37-120.

Cameron KC, Smith NP, McLay CDA, Fraser PM, McPherson RJ, Harrison DF, Harbottle P (1992) Lysimeters without edge-flow: an improved design and sampling procedure. *Soil Science Society of America Journal* **56,** 1625-1628.

de Klein CAM, Eckard RJ (2008) Targeted Technologies for nitrous oxide abatement from Animal Agriculture. *Journal of Experimental Agriculture* **48,** 14-20.

de Klein CAM, Barton L, Sherlock RR, Li Z, Littlejohn RP (2003) Estimating a nitrous oxide emission factor for animal urine from some New Zealand pastoral soils. *Australian Journal of Soil Research* **41,** 381-399.

Di HJ, Cameron KC, Sherlock RR (2007) Comparison of the effectiveness of a nitrification inhibitor dicyandiamide, in reducing nitrous oxide emissions in four different soils under different climatic and management conditions. *Soil Use and Management* **23,** 1-9.

Di HJ, Cameron KC, Sherlock RR, Shen JP, He JZ, Winefield CS (2010) Nitrous oxide emissions related to bacteria and not to archaea populations in nitrogen rich grassland soils. *Soil Biology and Biochemistry* (submitted).

Hewitt AE (1993) 'Methods and Rationale of the New Zealand Soil Classification'. Landcare Research Science Series No. 2.

Luo J, Lindsey S, Ledgard S (2008) Nitrous oxide emissions from animal urine application on a New Zealand pasture. *Biology and Fertility of Soils* **44,** 463-470.

Luo J, de Klein CAM, Ledgard SF, Saggar S. (2010) Management options to reduce nitrous oxide emissions from intensively grazed pastures: a review. *Agriculture, Ecosystems and Environment* (submitted).

Shepherd M, Welten B, Ledgard SF (2009) Effectiveness of dicyandiamide in reducing nitrogen leaching losses from two contrasting soil types under two rainfall regimes 0 a lysimeter study. In 'Nutrient management in a rapidly changing world'. (Eds LD Currie, CL Lindsay) pp 177-184. (FLRC: Massey University, Palmerston North, NZ).

Smith LC, de Klein CAM, Catto WD (2008) Effect of dicyandiamide applied in a granular form on nitrous oxide emissions from a grazed dairy pasture in Southland, New Zealand. *New Zealand Journal of Agricultural Research* **51,** 387-396.