ENVIRONMENTAL MANAGEMENT & CONSERVATION | RESEARCH ARTICLE

Carbon dioxide, nitrous oxide and methane emissions from the Waimate District (New Zealand) pasture soils as influenced by irrigation, effluent dispersal and earthworms

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Carbon dioxide, nitrous oxide and methane emissions from the Waimate District (New Zealand) pasture soils as influenced by irrigation, effluent dispersal and earthworms

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Abstract: Effects of wet/dry cycles in inducing greenhouse gas emissions are well documented. However, the effects of field drying and rewetting events remain poorly understood. This study investigated the impact of irrigation and effluent application on CO₂, N₂O and CH₄ in the Waimate District of New Zealand. Four soil management practices: (i) only added effluent, (ii) only added water, through irrigation, (iii) effluent and water added together, and (iv) neither water nor effluent added were sampled using static headspace chambers with a chamber diameter of 250 mm and height of 150 mm. All locations were sources of CO₂ and N₂O but net sinks of CH₄. Carbon dioxide fluxes ranged from 4.38 to 14.49 mg CO₂-C m⁻² hr⁻¹ while those for N₂O were between 0.007 and 0.012 mg N₂O-N m⁻² hr⁻¹. Wetting soils receiving effluent enhanced CO₂ production by 161%, suppressed N₂O fluxes by 17% but increased CH₄ uptake by 286%. When compared with control locations, effluent-only locations observed 50% less CO₂, yet highest N₂O emissions were observed on the same locations. Nitrous oxide emissions were positively correlated with CO₂ but...
negatively correlated with CH₄ emissions. Irrigation-only locations had 33% more earthworms than effluent locations. Maximum density and biomass occurred where both effluent and irrigation were applied. There was no evidence of relationships between earthworm measurements and gas fluxes.

**Subjects:** Agriculture & Environmental Sciences; Agriculture; Environmental Sciences; Soil Sciences; Microbiology; Ecology - Environment Studies

**Keywords:** earthworm density; earthworm biomass; effluent dispersal; greenhouse gas emissions; irrigation; Waimate District; New Zealand

1. Introduction

Although agriculture is a minor emitter of CO₂, it is a major emitter of N₂O and CH₄ (US Department of Agriculture, 2004). This is a particular problem in New Zealand (Saggar, Bolan, Bhandral, Hedley, & Luo, 2004) where agriculture contributes the largest proportion of the total greenhouse gas (GHG) emissions (Ministry for the Environment, 2012). The activities of soil decomposer organisms and roots lead to production of CO₂ (Schlesinger & Andrews, 2000), while three processes lead to N₂O release from soils: oxidation of ammonia to NO⁻ and NO₂⁻ (Kowalchuk & Stephen, 2001); reduction of NO₂⁻ to NO and N₂O under anoxic conditions (Knowles, 1982); and oxidation of ammonia to NO₂ which is then reduced to N₂O, NO and NO₂ (Wrage, Velthof, van Beusichem, & Oenema, 2001). On the other hand, CH₄ emissions result from anaerobic decomposition of organic matter (Thauer, 1998), while its oxidation by methanotrophs enhances the soil’s CH₄ sink capacity (Dalal, Allen, Livesley, & Richards, 2008).

The release and consumption of these gases depend on soil water content, temperature and substrate availability (Bollmann & Conrad, 1998; Philippot, Hallin, Börjesson, & Baggs, 2009) on an annual, seasonal or even daily basis (Du, Lu, & Wang, 2006). Therefore, irrigation and effluent (generated material containing cow excreta and urine diluted with wash down water after milking) application can directly or indirectly affect key variables that regulate these gas emissions from soils. Nevertheless, irrigation has received little investigation compared to contributions from other agricultural activities (King et al., 2009). Short-term emission pulses following soil drying and wetting are well described (Butterly, Bünemann, McNeill, Baldock, & Marschner, 2009; Linn & Doran, 1984; Priemé & Christensen, 2001). In general, these emissions recede in repeated drying and wetting events (Muhr, Goldberg, Borken, & Gebauer, 2008). The intensity and duration of the drying and wetting events could affect mineralisation and weathering (Borken & Matzner, 2009; Fierer & Schimel, 2002) and consequently lead to spatial and temporal emission fluctuations. Studies directly comparing GHG emissions from irrigated and un-irrigated soils report either increased emissions from irrigated soils (Horváth et al., 2010; Rochette et al., 2010), or no significant differences among them (Holst, Brüggemann, & Butterbach-Bahl, 2008).

Spreading animal waste onto land provides nutrients for pasture growth and supports rich microbial communities in soil (Elhottová et al., 2012). It also acts as a source of greenhouse gases (Liu & Greaver, 2009; Oenema et al., 2005) since they provide readily available substrates for microbial decomposition (Swift, Heal, & Anderson, 1979; Wardle et al., 2004). Soil management practices that affect earthworm communities stimulate microbial activity (McLean, Migge-Kleian, & Parkinson, 2006). Earthworm guts and associated structures (casts, burrows, middens) offer suitable microhabitats that support microbial communities (Marhan, Kandeler, & Scheu, 2007). Earthworms too have been shown to decompose more organic matter in wet soil than under dry conditions (Amador, Gørres, & Savin, 2005). It is therefore important to determine the influence of these earthworms on GHG emissions from soils.

Extensive areas in New Zealand and all over the world are converting to farming principally supported by irrigation and effluent dispersal (FAQ, 2010; Manono, 2014). In the Waimate District, the average wetting frequency is every 17 days (Manono & Moller, 2015). This raises two questions on
how GHG fluxes will be affected over the 17-day drying–wetting cycles in soils kept under long-term irrigation and normal stock management. Will continuous rewetting result in reduction of microbial activity and subsequent emission rates? Or will potential pulses during rewetting events outweigh this reduction and lead to increased overall fluxes?

In regard to this, this research was conducted to investigate the effect of irrigation and effluent application on CO$_2$, N$_2$O and CH$_4$ emissions from pasture soils and how these emissions relate to earthworm measurements. The study compared irrigated and effluent-treated locations with control locations under similar management regimes. The study aimed at determining: (i) the effects of irrigation on GHG emissions, (ii) the effects of effluent dispersal on GHG emissions, and (iii) the relationship between these emissions and earthworm measurements.

2. Materials and methods

2.1. Study area

The study was conducted in five typical dairy farms under grass pastures in The Waimate District (44°38′–44°54′ S and 170°59′–171°08′ E) of the South Canterbury region, New Zealand. The district borders the Waitaki River to the south, Pareora River to the north and the Hakataramea Valley to the West. It supports productive pastoral and cropping farming that forms a typical New Zealand agro-ecosystem landscape with irrigation on the flat areas of the Waitaki Basin and a smaller number of sheep and beef farms with irrigation on the flanking hills. Irrigation water is conveyed onto paddocks under gravity by constructing slightly sloping bays separated by low borders that direct the flow (Figure 1) while effluent is added as slurry. The study farms had an average stocking rate of 3.3 Livestock Units per Hectare, had been under irrigation and effluent management regimes for ≥10 years and were located within 10 km of each other. Soils of the study region are characterised by slow permeability, limited rooting depth and a medium to high bulk density. In the New Zealand soil classification (Hewitt, 1998) the soils are classified as mottled fragic pallic soils, while in USDA soil taxonomy (Soil Survey Staff, 1998), the soils are udic Haplustepts. The climate in Glenavy is characterised by a mean annual rainfall of 500–600 mm (Tait, Henderson, Turner, & Zheng, 2006), common droughts in the summer months and an average annual temperature of 11.2°C.

2.2. Sampling location selection

Gas samples were collected from twelve paddocks (individual farm fields)—six receiving irrigation water and six receiving both irrigation water and effluent spread across five different farms (Table 1). Each study paddocks had sufficiently large fragments of “un-irrigated land” for comparisons. This ensured that other aspects of soil management (e.g. pasture renovation, drainage, fertilisation regimes, stocking rate and grazing rotations) remained similar between sampled locations. From each study paddock, five sampling locations, three from the irrigated and two from the dry patches, were...
randomly selected. To exclude confounding factors, such as areas where stock congregate and trample the soil, gas collection locations were always at least 30 m away from each other, trees, fences, gateways and water troughs.

2.3. Gas sampling and analysis

Gas fluxes were sampled over the 17-day irrigation cycle in the summer of 2013. Static headspace chambers with a chamber diameter of 250 mm and height of 150 mm were used for gas collection. The methodology described by de Klein, Barton, Sherlock, Li, and Littlejohn (2003) was followed. In order to minimise the inherent issues related to chamber-based gas measurements, the chambers had vented tubes and were covered with reflective insulators (Parkin, Mosier, Smith, et al., 2003). Short headspace time interval provides a more linear curve (de Klein et al., 2003) when compared with an extended time (Parkin & Venterea, 2010; Rochette, 2011). Therefore, a 30-min deployment time was considered appropriate for this study. Actual gas sampling corresponded with irrigation events and aligned with the 17-day irrigation schedule. The first gas samples were collected on the day after soil wetting and then every fourth day until the fifteenth day. Sampling was done between 11.00–15.00 h. In total, four gas samples were collected from each location over the experimental period.

Since fluxes were expected to be linear, two gas samples were collected, the first for time 0 \( t_0 \) taken immediately after placing the chamber and the second for time 1 \( t_1 \) after 30 min. A Polypropylene syringe (20 ml capacity) was used to draw gas samples through a three-way stopcock directly via double-wadded caps into glass vials (6 ml Labco Exetainers®, Labco Ltd, Lampeter, Ceredigion, UK www.labco.co.uk). The gas was flushed through at least three times to ensure that the collected gas in the vial was over-pressurised. Gas samples were stored at room temperature before transporting them to the laboratory for analysis. In the laboratory, the exetainers were equilibrated to atmospheric pressure and then \( CO_2 \), \( CH_4 \) and \( N_2O \) concentrations determined using gas chromatography (SRI Instruments, California, USA) Model 8610C attached to a Gilson 222XL Auto sampler. \( CH_4 \) was determined by a flame ionisation detector (FID) analyser, \( CO_2 \) was converted to \( CH_4 \) by a methaniser and then analysed while \( N_2O \) was determined by an electron capture detector (ECD). The gas chromatography was controlled by the SRI Peaksimple software for Windows (SRI Instruments Europe GmbH). Gas fluxes were quantified in parts per million (µl/l volume).

To calculate the actual fluxes, gas concentration \( g(c) \) measurements were converted to mass units and corrected to field conditions by applying the Ideal Gas Law.

\[
g(c) = \frac{g(v) \times M \times P}{R \times T}
\]

where \( g(c) \) is the mass volume gas concentration, e.g. mg \( N_2O \) g/L enclosure; \( g(v) \) is the gas volume/ volume concentration (trace gas concentration expressed as parts per million by volume e.g. µl \( N_2O \) L\(^{-1}\) enclosure); \( M \) is the molar mass of the measured gas (g mol\(^{-1}\)); \( P \) is the barometric pressure; \( R \) is the universal gas constant (0.0820575 L atm K\(^{-1}\) mol\(^{-1}\)) and \( T \) is the air temperature at the time of sampling in °K (°K = °C + 273.15).

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**Table 1. Sampling plan showing the total number of gas sampling locations for each soil management treatment in each irrigation cycle. Gas was collected in two consecutive irrigation cycles**

| Paddock management | Irrigated | Effluent and irrigated |
|--------------------|-----------|------------------------|
| Number of paddocks | 6         | 6                      |
| Sampling location  | Control   | Irrigation only        |
|                    | Effluent only | Effluent and irrigation |
| Number of locations| 12        | 15                     |

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It was assumed that fluxes $g(f)$ were constant and that the concentration ($C$) increased/decreased linearly over the 30 min ($t$). The slope of the converted concentrations was used to calculate the gas fluxes using the equation:

$$g(f) = \frac{V \times C}{A}$$

where $g(f)$ is the gas flux as mass m$^{-2}$ hr$^{-1}$, e.g. mg N$_2$O-N m$^{-2}$ hr$^{-1}$, $V$ is the internal volume of the enclosure (chamber headspace) in m$^3$, $A$ is the soil area covered by the enclosure in m$^2$, and $C$ is the change in gas concentration over time in m$^{-3}$ hr$^{-1}$, e.g. mg CO$_2$-C m$^{-3}$ hr$^{-1}$.

Gas flux calculations followed the micrometeorological convention where positive fluxes are directed to the atmosphere and negative to the soil.

2.4. Earthworm measurements
Each time gas was collected, earthworms were also extracted from a 20 cm × 20 cm × 20 cm block of soil layer dug out using a spade so that the results can be expressed as density (numbers m$^{-2}$) and biomass (g m$^{-2}$). The soil extracted from the pit was placed onto a plastic sheet and searched for earthworms by sorting and crumbling the soil matrix by hand (Edwards & Lofty, 1977). Collected earthworms were identified to species level using external morphology keys and description provided in the literature. The collected earthworms were weighed using an electronic balance (accurate to 0.1 g). Please refer to Manono and Moller (2015) for further details.

2.5. Statistical analysis
All statistical analyses were performed using Genstat for Windows software (release 16). Emission differences between locations were analysed with a generalised linear mixed model fitted by restricted maximum likelihood (REML). Soil management levels and sampling days after soil wetting were assigned as fixed effects, and interactions between them incorporated when comparing the interaction effects. This meant that they were modelled separately as a function of treatment. To account for the lack of independence and hierarchical nature of the sampling, random effects were always nested as Farm/Paddock/Sampling location within the models. Orthogonal contrasts were calculated to quantify the effects of treatment separately as well as the differences on each sampling day. To test whether earthworms could best explain the variations in the observed emission differences, earthworm data were assigned as fixed effects. For possible relationships between the different gas emission rates, CH$_4$, CO$_2$ and N$_2$O fluxes were investigated by calculating Pearson’s correlation coefficients.

Preliminary models were constructed and residuals inspected to check for heteroscedasticity. The significance of predictor variables were assessed by Wald’s tests. In the modelling, N$_2$O and CH$_4$ fluxes were not normally distributed and transformation was necessary. Nitrous oxide fluxes were transformed to log$_{10}$, while CH$_4$ fluxes were natural log transformed. Since earthworm biomass measures are continuous rather than discrete counts (not normally distributed), arcsine transformations (Sokal & Rohlf, 1981) were used to normalise variances within REML models by incorporating the usual blocking structure. Predicted transformed means and confidence intervals were back-transformed for reporting, but the $p$-values reflect the tests done on transformed data. Results are presented as means ± 2 × SE (standard error of differences).

3. Results

3.1. Flux patterns as influenced by land treatment and day of sampling
In general, CO$_2$, N$_2$O and CH$_4$ fluxes showed significant differences between treatments (Figure 2). The highest CO$_2$ emission rates were observed in irrigated-only locations with least emissions from effluent-only locations. In contrast, highest N$_2$O fluxes were observed in effluent-only treated locations and the least from the control locations. Methane emission rates varied considerably showing huge differences between treatments.
Apart from CO₂ fluxes, temporal significant differences were observed for N₂O and CH₄ within the 15-day experimental period. N₂O emissions were highest on the sixth day after soil wetting and lowest on the fifteenth day. On the other hand, CH₄ uptake was highest on the sixth day and lowest on the second day. Nitrous oxide emissions increased immediately after soil wetting up to the sixth day before decreasing while CH₄ uptake increased immediately after soil wetting up to the sixth day before decreasing. The largest N₂O emissions were released from soils when CH₄ uptake was greatest. There was a negative correlation between CH₄ and N₂O ($R^2 = -0.342$, $p = 0.034$ (Table 2)).

3.2. Effect of the interaction between management and day of sampling on gas fluxes

The temporal heterogeneity was small and almost constant over the time of the experiment for the control and effluent-only locations (Figure 2 (A) and (B)). As the experiment was carried out during the dry summer months without rainfall, the soil moisture content was relatively uniform. Carbon dioxide and N₂O fluxes were mostly positive while soils generally acted as sinks for CH₄ throughout the measurement period. Only N₂O fluxes exhibited a significant interaction effect between soil

| Table 2. Pearson’s correlation coefficients between CH₄, CO₂, and N₂O emissions |
|---------------------------------------------------------------|
| CH₄ (mg CH₄-C m⁻² hr⁻¹) | CO₂ (mg CO₂-C m⁻² hr⁻¹) | N₂O (mg N₂O-N m⁻² hr⁻¹) |
|--------------------------|--------------------------|--------------------------|
| CH₄ (mg CH₄-C m⁻² hr⁻¹) | 1 | -0.267 | -0.342 |
| | | $p = 0.012$ | $p = 0.034$ |
| CO₂ (mg CO₂-C m⁻² hr⁻¹) | | 1 | 0.165 |
| | | | $p = 0.014$ |
| N₂O (mg N₂O-N m⁻² hr⁻¹) | | | 1 |

Note: The coefficients are significant at $p < 0.05$
management and day of sampling during the experimental period (Figure 2 (C) and (D)). While no obvious trends were observed for CO$_2$, soil wetting enhanced the soil’s sink capacity for CH$_4$ (Figure 2 (C) and (D)). Nevertheless, these flux changes were not large enough to exhibit interaction effects for these gases.

3.3. Earthworm density and biomass as influenced by soil management

The control locations had the least earthworm density and biomasses, followed by effluent-only and then irrigation-only locations, while effluent and irrigated locations had the highest earthworm measurements. The same trend was exhibited for *Aporrectodea caliginosa* (Savigny, 1826) and *Lumbricus rubellus* (Hoffmeister, 1843) species. On the other hand, the day of sampling after soil wetting did not show significant differences for any of the earthworm measurements. The interaction between soil management and day of sampling after soil wetting did not produce any significant interaction effects for the earthworm measurements (Figure 3(A)–(D)).

3.4. Are emissions higher where there are more earthworms?

More complex Generalised Linear Mixed models were built to explore the same soil response variables for GHG emissions but with earthworms, management, days after soil wetting and the interaction between management and days after soil wetting as fixed effects. The Generalised Linear Mixed Models were of the form:

\[
\text{Gas flux} \rightarrow \text{soil management} + \text{days after soil wetting} + \text{management.days after wetting} + \text{Earthworms}
\]

This choice of model allowed the same blocking structure as incorporated in the REML models. Whereas the preceding models simply tested for independent responses for gas fluxes, these were designed to test whether gas fluxes varied for a given level of earthworms in the treatments. The full set of model parameters and constants are given in Table 3. Overall, the analysis showed no evidence that the gas flux changes were associated with a change in earthworm measures.
4. Discussion

4.1. Overall changes in soil gas fluxes

As reported in other studies, soils were net sources of CO₂ and N₂O emissions (Bhandral, Bolan, Saggar, & Hedley, 2007; Bolan et al., 2004; de Klein et al., 2003; Rochette et al., 2010). The exhibited N₂O pulses after soil wetting suggests that the rewetting and drying cycles did not have a

Table 3. Predictor parameter values in generalised linear mixed models to predict gas fluxes from the interaction of soil treatment and day of sampling after the irrigation event

| Treatment                        | Soil property | CO₂ (mg CO₂-C m⁻² hr⁻¹) | N₂O (mg N₂O-N m⁻² hr⁻¹) | CH₄ (mg CH₄-C m⁻² hr⁻¹) |
|----------------------------------|---------------|--------------------------|--------------------------|--------------------------|
|                                  | Transformation used | Untransformed | Logₑₓ | Natural log |
|                                | Constant       | 4.423 | 0.012 | -0.014 |
|                                | se             | 1.761 | 0.007 | 0.025 |
| Soil treatment                   | Effluent only  | 0.000 | 0.000 | 0.000 |
|                                  | Irrigation only| 7.902*** | -0.161* | -0.020*** |
|                                  | Effluent and irrigation | 7.860*** | -0.140* | -0.020*** |
|                                | se             | 1.922 | 0.155 | 0.027 |
| Sampling day after soil wetting  | 2nd            | 0.000 | 0.000 | 0.000 |
|                                  | 6th            | 1.415 | -1.077** | -0.001* |
|                                  | 10th           | 0.118 | -0.176** | -0.007* |
|                                  | 15th           | -0.004 | -0.134** | -0.009* |
| Land treatment and day of sampling interaction | Effluent only | 2nd | 0.000 | 0.000 |
|                                  | 6th            | 0.000 | 0.000 | 0.000 |
|                                  | 10th           | 0.000 | 0.000 | 0.000 |
|                                  | 15th           | 0.000 | 0.000 | 0.000 |
|                                  | Irrigation only| 2nd | 0.000 | 0.000 |
|                                  | 6th            | -0.378 | 0.124* | -0.006 |
|                                  | 10th           | -0.593 | 0.022* | 0.012 |
|                                  | 15th           | -0.026 | 0.188* | 0.024 |
|                                  | Effluent and irrigation | 2nd | 0.000 | 0.000 |
|                                  | 6th            | -0.691 | 0.278* | -0.057 |
|                                  | 10th           | -0.691 | 0.166* | -0.032 |
|                                  | 15th           | 4.665 | -0.218* | -0.019 |
|                                  | se             | 2.628 | 0.173 | 0.035 |
| Earthworms                       | A. caliginosa  | -0.001 | 0.000 | 0.000 |
|                                  | se             | 0.002 | 0.000 | 0.000 |
|                                  | L. rubellus    | 0.002 | 0.000 | -0.000 |
|                                  | se             | 0.004 | 0.001 | 0.000 |

Notes: se is the statistical error that is the amount by which the observed value differs from its expected value. Parameter values are significance at *p < 0.05, **p < 0.01, ***p < 0.001 from REML Genstat model.
permanent effect on soil N₂O emissions. However, the regular wetting and drying may have suppressed CO₂ and CH₄ fluxes as opposed to emission pulses observed in initial soil wetting after long dry spells (Unger, Mágua, Pereira, David, & Werner, 2010; Xu, Baldocchi, & Tang, 2004). Moreover, constantly moist soil do not exhibit these flushes (Chowdhury, Marschner, & Burns, 2011). This observation is in agreement with other studies where fluxes recede in subsequent soil wetting events (Muhr et al., 2008). The enhanced fluxes in irrigated and irrigated and effluent-dispersed treatments may have resulted from increases in C and N mineralisation. This observation corroborates other emission studies in a Sonoran desert ecosystem (Sponseller, 2007); a fertilised grassland (Kim, Mishurov, & Kiely, 2010) and in a semi-arid grassland (Wu et al., 2010). On the other hand, the observation in control treatment may have been influenced by lack of water addition. Decreased soil water content lowers respiration, reduces diffusion of substrates and results in soil microbes reallocating resources potentially affecting nutrient cycling; thus, moisture limitations limit soil microbes in dry soils (Manzoni, Taylor, Richter, Porporato, & Ågren, 2012; Schimel, Balser, & Wallenstein, 2007; Stark & Firestone, 1995).

The significant flux differences observed between treatments illustrate the importance of management practices as a factor in regulating soil microbial activity in managed agro-ecosystems. However, unlike for earthworms where the effects of adding water and effluent together appear to be additive (Manono & Moller, 2015), each gas flux differed according to land treatment. For example effluent-only treated locations recorded the lowest CO₂ emissions, while the same locations recorded the highest N₂O emissions. This suggests that there is no interdependent effect of having both effluent and water added together for the three GHG emissions.

4.2. Changes in CO₂ fluxes
Carbon dioxide fluxes appeared to be associated with the level of organic additions, e.g. lower emissions from locations where effluent was applied. Unfortunately, these locations had the highest N₂O emission rates, an indication that effluent application may contribute to CO₂ mitigation but simultaneously enhance N₂O emissions. Lack of CO₂ emission differences for sampled days suggests that CO₂ fluxes are determined by long-term soil management practices rather than short-term events.

4.3. Changes in N₂O fluxes
Higher N₂O emissions from effluent-only locations corroborate other New Zealand N₂O emission studies (Barton & Schipper, 2001; Bhandral et al., 2007; Bolan, Saggar, Luo, Bhandral, & Singh, 2004; Saggar et al., 2004). One possible cause of these enhanced emissions is the exacerbation of anaerobic conditions resulting from microbial metabolism of the higher amounts of organic carbon in effluent (e.g. in one study, in New Zealand pastures, one cubic metre of effluent contained 7,400 g of total solids, 2,247 g of total carbon, 246 g of total nitrogen and 55 g of total phosphorus (Di, Cameron, Silva, Russell, & Barnett, 2002)) and the higher oxygen demand in polluted water (EPA, 1990). Since effluent is added as slurry, the increased soil water content reduces soil aeration, while increased microbial activity reduces soil oxygen availability, thus alteration of both nitrification and denitrification processes; the main processes that drive N₂O formation (Amha & Bohne, 2011; Beare, Gregorich, & St-Georges, 2009; Horváth et al., 2010). Similarly, continuous changes between wetting and drying may increase water stable aggregates, thus not easily decomposed but stored for longer time and consequently intensifying soil microbial activity (Degens & Sparling, 1995; Pulleman, Six, Uyl, Marinissen, & Jongmans, 2005). Further, mineral nitrogen contents can be higher in effluent than non-effluent locations.

4.4. Changes in CH₄ fluxes
Apart from the control location, soils consistently acted as a sink for CH₄ which is typical for temperate grassland soils (Liebig, Gross, Kronberg, & Phillips, 2010; Mosier, Delgado, Cochrane, Valentine, & Parton, 1997). The lack of evidence of CH₄ flush shifts after soil wetting contributed to the
contrasting views with respect to CH₄ emission studies (Davidson, Ishida, & Nepstad, 2004). High soil water content immediately after soil wetting may have restricted oxygen diffusion into the soil which is necessary to obligate aerobic methanotrophs for oxidation to occur. This caused the dip in CH₄ uptake following soil wetting. Reduced CH₄ uptake in un-irrigated locations may have resulted from reduced metabolic activity of methanotrophs due to moisture stress. Indeed, this has been reported to occur when soil moisture is low (Kammann, Grünhage, Jäger, & Wachinger, 2001; Liu et al., 2007). Continuous moisture availability and carbon-rich effluent dispersed onto paddocks allowed for maximum CH₄ uptake.

4.5. Relationship between CO₂, N₂O and CH₄ fluxes
Soils were sources of CO₂ and N₂O and the two gases were positively correlated. This positive linkage may have resulted from shared available substrates, labile carbon and nitrogen (Bollmann & Conrad, 1998), and other common controlling factors such as water and oxygen. Similarly, shared microbial processes may have produced similar impacts on microbial processes that generate these two gases (Firestone & Davidson, 1989). The larger N₂O emissions coinciding with peak CH₄ uptake under irrigated locations that were negatively correlated indicate the occurrence of spatially separated soil microsites with contrasting environments and processes. It suggests possible shifts of N₂O along with changes in CH₄ emissions that can offer trade-off opportunities for reduced net GHG emissions and warrant further studies for this determination.

4.6. Relationship between cumulative fluxes and earthworms
Many complex and mutual interdependent relationships between earthworms and soil microbes have been observed in many studies (Drake & Horn, 2006; Lubbers, Brussaard, Otten, & Van Groenigen, 2011; Lubbers et al., 2013). However, there was no evidence of a predictive relationship between earthworm measurements and gas fluxes in the present study. This contrasted with other studies where positive correlations between earthworms and gas flux emissions have been observed. For example, those observed between CO₂ fluxes and earthworms (Caravaca, Pera, Masciandaro, Ceccanti, & Roldán, 2005) and N₂O fluxes and earthworms (Borken, Gründel, & Beese, 2000). Interactions between earthworms and other soil biotic and abiotic components may have developed several feedback disruptions that limited the impact of earthworms in this study. Earthworm presence in all treatments may have posed problems for detecting differences in gas emissions. Extreme treatments, thus; experiments with and without earthworms or microcosm experiments containing a known number of individual earthworms or known biomasses would be ideal in such a comparison.

4.7. Methodological constraints
Caution is needed when interpreting these results since the experiment was not replicated over several wetting and drying cycles or seasons. Follow-up work through several such cycles is necessary to provide greater insight into these relations. In addition, fluxes may have varied from farm to farm depending on individual farmer management. An attempt was made to minimise this disruption from the above effects by using a hierarchical and nested sampling design.

5. Conclusion
Effluent application especially on irrigated land has significant effects on GHG emissions. The environmental impact of the moderate increases on CO₂ and N₂O with irrigation is of concern, especially since increases in irrigation are expected throughout the world as part of ongoing intensification of agriculture. Lack of relationships between earthworms and GHG emissions in this study suggest the importance of examining whole systems with a longer term perspective to establish how earthworm–microbial interactions influence GHG emissions from agricultural soils.
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References
Armador, J. A., Görres, J. H., & Savin, M. C. (2005). Role of soil water content in the carbon and nitrogen dynamics of Lumbricus terrestris L. burrow soil. Applied Soil Ecology, 28, 15–22. http://dx.doi.org/10.1016/j.apsoil.2004.06.009
Amho, Y., & Bohne, H. (2013). Denitrification from the horticultural peats: Effects of pH, nitrogen, carbon, and moisture contents. Biology and Fertility of Soils, 49, 293–302. http://dx.doi.org/10.1007/s00374-010-0536-y
Barton, L., & Schipper, L. (1998). Influence of O2 availability on N2O production and nitrous oxide emissions from various types of farm effluents. Nutrient Cycling in Agroecosystems, 51, 193–208. http://dx.doi.org/10.1023/A:10057075-007-9107-5
Bolan, N., Soggar, 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 (Vol. 84, pp. 37–120). Amsterdam: Academic Press.
Bollmann, A., & Conrad, R. (1998). Influence of O2 availability on NO and N2O release by nitrification and denitrification in soils. Global Change Biology, 4, 387–396. http://dx.doi.org/10.1046/j.1365-2486.1998.00161.x
Borken, W., Gründel, S., & Beece, F. (2000). Potential contribution of Lumbricus terrestris L. to carbon dioxide, methane and nitrous oxide fluxes from a forest soil. Biology and Fertility of Soils, 32, 142–148. http://dx.doi.org/10.1007/s003740000228
Borken, W., & Matzner, E. (2009). Reappraisal of drying and rewetting effects on C and N mineralization and fluxes in soils. Global Change Biology, 15, 808–824. http://dx.doi.org/10.1111/gcb.2009.15.issue-4
Butterly, C., Bünemann, E., McNell, A. M., Baldock, J. A., & Marschner, P. (2009). Carbon pulses but not phosphorous pulses are related to decreases in microbial biomass during repeated drying and rewetting of soils. Soil Biology and Biochemistry, 41, 1406–1416. http://dx.doi.org/10.1016/j.soilbiobiochem.2009.03.018
Caravaca, F., Pena, A., Maccanndaro, G., Ceccanti, B., & Boldrin, A. (2005). A micromac approach to assessing the effects of earthworm inoculation and oat cover cropping on CO2, CH4, and N2O fluxes and biological properties in an amended semiarid soil. Chemosphere, 59, 1625–1631. http://dx.doi.org/10.1016/j.chemosphere.2004.12.032
Chowdhury, N., Marschner, P., & Burns, R. G. (2013). Soil microbial activity and community composition: Impact of changes in matric and osmotic potential. Soil Biology and Biochemistry, 43, 1229–1236. http://dx.doi.org/10.1016/j.soilbiobiochem.2011.02.012
Dalal, R. C., Allen, D. E., Livesley, S. J., & Richards, G. (2008). Magnitude and biophysical regulators of methane emission and consumption in the Australian agricultural, forest, and submerged landscapes: A review. Plant and Soil, 309, 43–76. http://dx.doi.org/10.1007/s11104-007-9466-7
Davidson, E. A., Ishido, F. Y., & Nepstad, D. C. (2004). Effects of an experimental drought on soil emissions of carbon dioxide, methane, nitrogen oxide, and nitric oxide in a moist tropical forest. Global Change Biology, 10, 718–730. http://dx.doi.org/10.1016/j.gcbio.2004.10.005
de Klein, C. A., Barton, L., Sherlock, R. R., Li, Z., & Littlejohn, R. P. (2003). Estimating a nitrous oxide emission factor for animal urine from some New Zealand pastoral soils. Australian Journal of Soil Research, 41, 381–399. http://dx.doi.org/10.1071/S000758202128
Degens, B., & Sparling, G. (1993). Repeated wet-dry cycles do not accelerate the mineralization of organic C involved in the macro-aggregation of a sandy loam soil. Plant and Soil, 175, 197–203. http://dx.doi.org/10.1023/A:10040011355
Di, H., Cameron, K., Silva, R., Russell, J., & Borrett, J. (2002). A lysimeter study of the fate of 15 N-labelled nitrogen in cow urine with or without farm dairy effluent in a grazed dairy pasture soil under flood irrigation. New Zealand Journal of Agricultural Research, 45, 235–244. http://dx.doi.org/10.1080/00288233.2002.9513514
Drake, H., & Horn, M. (2006). Earthworms as a transient heaven for terresterial denitrifying microbes: A review. Engineering in Life Sciences, 6, 261–265. http://dx.doi.org/10.1016/1055-6286(98)00118-6
Du, R., Lu, D., & Wang, G. (2006). Diurnal, seasonal, and inter-annual variations of N2O fluxes from native semi-arid grassland soils of Inner Mongolia. Soil Biology and Biochemistry, 38, 3474–3482. http://dx.doi.org/10.1016/j.soilbio.2006.06.012
Edwards, C. A., & Lofty, J. R. (1977). Methane emissions and opportunities for control. Workshop results of Interprograminal Panel on climate change (EPA/400/9-90/007). Washington, DC: US EPA.
FAO. (1990). Yearbook 2010: Fishery and agriculture statistics. Rome: Food and Agricultural Organisation of the United Nations.
Fierer, N., & Schimel, J. P. (2002). Effects of drying-rewetting frequency on soil carbon and nitrogen transformations. Soil Biology and Biochemistry, 34, 777–787. http://dx.doi.org/10.1016/S0038-0717(02)00007-X

Firestone, M. K., & Davidson, E. A. (1989). Microbiological basis of NO and N2O production and consumption in soil. In Schimel, D. S. & Andreea, M. O. (Eds.), Exchange of trace gases between terrestrial ecosystems and the atmosphere (pp. 7–21). Chichester: Wiley.

Hewitt, A. E. (1998). New Zealand Soil Classification. Lincoln: Manoaki Whenua press.

Holst, J., Brüggemann, N., & Butterbach-Bahl, K. (2008). Effects of irrigation on nitrous oxide, methane and carbon dioxide fluxes in an Inner Mongolian steppe. Advances in Atmospheric Sciences, 25, 748–756.

Horváth, L., Grosz, B., Machon, A., Tuba, Z., Nagy, Z., Czóbel, S., Z., ... Weilinger, T. (2010). Estimation of nitrous oxide emission from Hungarian semi-arid sandy and loess grasslands; effect of soil parameters, grazing, irrigation and use of fertilizer. Agriculture, Ecosystems and Environment, 139, 255–263. http://dx.doi.org/10.1016/j.agee.2010.08.011

Kammann, C., Grünhage, L., Jäger, H. J., & Wachinger, G. (2011). Earthworm invasions of ecosystems devoid of earthworms: Effects on soil microbes. Biological Invasions, 8, 1257–1273. http://dx.doi.org/10.1007/s10530-006-9020-x

King, A., Evatt, K., Six, J., Poch, R., Rolston, D., & Hopmans, J. (2001). Methane fluxes from differentially managed grassland study plots: The important role of CH4 oxidation in grassland with a high potential for CH4 production. Environmental Pollution, 115, 261–273. http://dx.doi.org/10.1016/S0269-7491(01)00140-7

Liu, L., & Greaver, T. L. (2009). A review of nitrogen enrichment effects on three biogenic GHGs: The CO2 sink may be largely offset by stimulated N2O and CH4 emission. Ecology Letters, 12, 1103–1117. http://dx.doi.org/10.1111/j.1461-0248.2009.01313.x

Lubbers, I. M., Brussaard, L., Otten, W., & Van Groenigen, J. W. (2011). Earthworm-induced N mineralization in fertilized grassland increases both N2O emission and crop-N uptake. European Journal of Soil Science, 62, 152–161. doi:10.1111/j.1365-2389.2010.01313.x

Lubbers, I. M., van Groenigen, K. J., Fonte, S. J., Six, J., Brussaard, L., & van Groenigen, J. W. (2013). Greenhouse-gas emissions from soils increased by earthworms. Nature Climate Change, 3, 187–194.

Manono, B. O. (2014). Denitrification. Microbiological Reviews, 49, 43–49. http://dx.doi.org/10.1016/j.micrev.2014.02.002

McLean, M. A., Migge-Kleian, S., & Parkinson, D. (2006). Earthworm invasions of ecosystems devoid of earthworms: Effects on soil microbes. Biological Invasions, 8, 71–83. http://dx.doi.org/10.1007/s10530-006-9020-x

Miceli, G., & Tripodi, G. (2010). Repeated drying-rewetting cycles and their effects on the emission of CO2, NO, NOX, and CH4 in a forest soil. Journal of Plant Nutrition and Soil Science, 171, 719–728. http://dx.doi.org/10.1007/s10010-009-9342-y
Rochette, P. (2011). Towards a standard non-steady-state chamber methodology for measuring soil N$_2$O emissions. Animal Feed Science and Technology, 166–167, 141–146.

Soggar, S., Bolan, N. S., Bhendral, R., Hedley, C., & Luo, J. (2004). A review of emissions of methane, ammonia, and nitrous oxide from animal excreta deposition and farm effluent application in grazed pastures. New Zealand Journal of Agricultural Research, 47, 513–544.

Schimel, J., Bolser, T. C., & Wallenstein, M. (2007). Microbial stress-response physiology and its implications for ecosystem function. Ecology, 88, 1386–1394. doi:10.1890/06-0219

Schlesinger, W., & Andrews, J. (2000). Soil respiration and the global carbon cycle. Biogeochemistry, 48, 7–20. doi:10.1023/A:1006247623877

Soil Survey Staff. (1998). Keys to soil taxonomy. Washington, DC: United States Department of Agriculture.

Sokal, R., & Rohlf, F. (1981). Biometry: The principles and practice of statistics in biological research. New York, NY: WH Freeman.

Sponseller, R. A. (2007). Precipitation pulses and soil CO$_2$ flux in a Sonoran Desert ecosystem. Global Change Biology, 13, 426–436. doi:10.1111/j.1365-2486.2006.01307.x

Stark, J. M., & Firestone, M. K. (1995). Mechanisms for soil moisture effects on activity of nitrifying bacteria. Applied and Environmental Microbiology, 61, 218–221.

Swift, M. J., Heal, O. W., & Anderson, J. M. (1979). Decomposition In terrestrial ecosystems (Vol. 5). Oakland, CA: University of California Press.

Tait, A., Henderson, R. D., Turner, R., & Zheng, X. (2006). Thin plate smoothing spline interpolation of daily rainfall for New Zealand using a climatological rainfall surface. International Journal of Climatology, 26, 2097–2115. http://dx.doi.org/10.1002/(ISSN)1097-0088

Thauer, R. K. (1998). Biochemistry of methanogenesis: A tribute to Marjory Stephenson: 1998 Marjory Stephenson prize lecture. Microbiology, 144, 2377–2406. http://dx.doi.org/10.1099/00221287-144-9-2377

Unger, S., Mágus, C., Pereiro, J. S., David, T. S., & Werner, C. (2010). The influence of precipitation pulses on soil respiration—Assessing the “Birch effect” by stable carbon isotopes. Soil Biology and Biochemistry, 42, 1800–1810. http://dx.doi.org/10.1016/j.soilbio.2010.06.019

US Department of Agriculture (USDA). (2004). US Agriculture and Forestry greenhouse gas inventory: 1990–2001 (Tech Bull No. 1907, 164 pp.). Washington, DC: Global change program Office, Office of the Chief Economist.

Wardle, D. A., Bardgett, R. D., Kilonomos, J. N., Setalía, H., van der Putten, W. H., & Wall, D. H. (2004). Ecological linkages between aboveground and belowground biota. Science, 304, 1623–1633. doi:10.1126/science.1094875

Wrase, N., Velthof, G. L., van Beusichem, M. L., & Oenema, O. (2001). Role of nitrifier denitrification in the production of nitrous oxide. Soil Biology and Biochemistry, 33, 1723–1732. http://dx.doi.org/10.1016/S0038-0717(01)00096-7

Wu, X., Yao, Z., Brüggemann, N., Shen, Z. Y., Wolf, B., Dannemann, M., & Zheng, X. (2010). Effects of soil moisture and temperature on CO$_2$ and CH$_4$ soil–atmosphere exchange of various land use/cover types in a semi-arid grassland in Inner Mongolia, China. Soil Biology and Biochemistry, 42, 773–787. doi:10.1016/j.soilbio.2010.01.013

Xu, L., Baldocchi, D. D., & Tang, J. (2004). How soil moisture, rain pulses, and growth alter the response of ecosystem respiration to temperature. Global Biogeochemical Cycles, 18(4).