



## Combination of cattle urine and dung patches synergistically increased nitrous oxide emissions from a temperate grassland under wet conditions

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### ARTICLE INFO

#### Keywords:

Dairy cattle  
Grazing cattle  
Emission factor  
Static chamber  
Methane  
Excreta

### ABSTRACT

During grazing, some of the nutrients ingested by cattle are returned to grassland as urine and dung patches and can be lost as greenhouse gases. Sites where cattle congregate are more likely to have overlapping excreta patches favouring enhanced nitrous oxide (N<sub>2</sub>O) emissions. However, there is no consensus about the magnitude of these or simultaneous methane (CH<sub>4</sub>) emissions or potential mitigation options. This study investigated the effect of combined cattle dung and urine depositions on N<sub>2</sub>O and CH<sub>4</sub> emissions, compared with emissions from separate depositions, under different weather conditions. Local emission factors (EFs) were then calculated for both gases. A quantitative assessment of published studies was also performed to search for N<sub>2</sub>O emissions drivers. Two field experiments were performed during two 98-day trials under dry and wet conditions in Tandil, Argentina. Treatments included fresh excreta patches of urine (0.75 L), dung (2.50 kg), dung + urine (2.50 kg + 0.75 L) from Holstein dairy cows, and a control (without excreta). Soil and excreta properties were analysed, and N<sub>2</sub>O and CH<sub>4</sub> fluxes from the patches were measured using the static chamber technique. Patches containing dung were shown to be localised CH<sub>4</sub> hotspots. Urine applied to soil, and the addition of urine to dung patches had a negligible effect on CH<sub>4</sub> fluxes. Urine, dung and combined patches were found to be localised N<sub>2</sub>O sources. Adding urine to dung patches under wet weather had a significant synergetic effect (threefold increase) on cumulative N<sub>2</sub>O emissions compared with the theoretical sum of separate excreta patches. Adding urine to dung patches under dry conditions gave an additive effect on N<sub>2</sub>O. These findings suggest that preventing overlapping excreta patches under wet conditions can help mitigate N<sub>2</sub>O emissions from temperate managed grazed pastures. The effect of combining excreta patches was also evident in the EF values obtained. That for CH<sub>4</sub> was consistent with the default IPCC value (0.75 g CH<sub>4</sub> kg<sup>-1</sup> VS), while N<sub>2</sub>O (EF = 0.03–0.39%) was lower than the updated IPCC 2019 value of 0.6%.

### 1. Introduction

Over 80% of large ruminants are produced in grazing systems which occupy about one-third of the Earth's ice-free land surface and account for 70% of global agricultural land (Steinfeld et al., 2006). In grassland ecosystems, livestock can help transfer plant biomass into highly nutritious animal protein foods while utilising resources that humans

cannot otherwise consume (Leip et al., 2019). However, large proportions of the nutrients ingested by livestock are returned to the soil through excreta depositions (urine and dung) (Haynes and Williams, 1993). Although the deposition rate of nutrients in excreta varies greatly due to diet composition and animal characteristics, the amount of nutrients in the relatively small excreta patches generally exceeds immediate plant requirements. Finally, this excess of nutrients is vulnerable to

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<https://doi.org/10.1016/j.agee.2022.108147>

Received 21 February 2022; Received in revised form 19 August 2022; Accepted 24 August 2022

Available online 6 September 2022

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losses via leaching and gaseous emissions (Cai et al., 2017).

Fresh excreta deposited by grazing cattle contain energy-rich and chemically reduced carbon (C) and nitrogen (N) (Oenema et al., 2008), which can result in hotspots of greenhouse gas (GHG) emissions from grassland (Cai et al., 2017). Freshly excreted dung has considerable potential to emit methane (CH<sub>4</sub>). It contains abundant methanogenic microorganisms from the digestive tract that convert readily available soluble C into CH<sub>4</sub> under the anaerobic conditions prevailing within the fresh dung patch (Hahn et al., 2018). In addition, since excreta contain significant amounts of organic N (Haynes and Williams, 1993), both urine and dung patches have the potential to act as a localised source of nitrous oxide (N<sub>2</sub>O), a potent GHG. Following deposition, a major fraction of the organic N in excreta is mineralised into ammonium (NH<sub>4</sub><sup>+</sup>) and later transformed into nitrate (NO<sub>3</sub><sup>-</sup>) through the activity of nitrifiers under partial aeration. The NO<sub>3</sub><sup>-</sup> produced can then be converted into dinitrogen (N<sub>2</sub>) by denitrifying bacteria under anoxic conditions. During both nitrification of NH<sub>4</sub><sup>+</sup> and denitrification of NO<sub>3</sub><sup>-</sup>, N<sub>2</sub>O gas may escape to the atmosphere as a by-product (Luo et al., 2017). The fraction of N emitted from excreta as N<sub>2</sub>O is highly influenced by environmental conditions, farm systems and manure management practices (Oenema et al., 2008).

Pasture-based dairy production systems are common in the temperate southern hemisphere. Cows rotationally graze in non-irrigated fenced paddocks with free access to water in stationary troughs (Luo et al., 2017). They frequently also have access to other preferred areas, like feeding bins, shaded areas, gateways and tracks within paddocks. These sites where animals congregate are subjected to more frequent excreta deposition and trampling, resulting in a high N load in compacted soil without vegetation, favouring higher N<sub>2</sub>O emissions (hotspots) (Saggar et al., 2022). The highly soluble N concentrations in urine and the available C and microorganisms in dung may have a synergistic effect, leading to higher GHG emissions than the theoretical sum from each separate excreta patch. Previous laboratory incubation studies indicate that combined dung and urine have a synergistic (super-additive) effect on N<sub>2</sub>O emissions, raising the level above the theoretical sum (Liao et al., 2018; van Groenigen et al., 2005a; Wu et al., 2020). However, it is unclear whether this synergistic effect also occurs under field conditions since there is no consensus in the literature on whether and to what extent combined cattle excreta patches enhance N<sub>2</sub>O emissions (Cardoso et al., 2019; Hyde et al., 2016; Li et al., 2016; Tully et al., 2017; van Groenigen et al., 2005b; Zhu et al., 2021). Studies examining CH<sub>4</sub> emissions found that overlapping excreta patches may have a negligible effect or even decrease CH<sub>4</sub> emissions compared to separate dung patches (Liao et al., 2018; Tully et al., 2017; Zhu et al., 2021). Data are particularly scarce for temperate grasslands, creating a need to identify grassland management practices that increase GHG emissions and formulate appropriate mitigation measures.

To better characterise GHG emissions from excreta on-farm and understand their contribution to total agricultural GHG in national inventories, the Intergovernmental Panel on Climate Change (IPCC) encourages the development of country-specific GHG emission factors (EFs). Estimating GHG emissions from excreta deposited during grazing includes CH<sub>4</sub> emissions from dung and N<sub>2</sub>O emissions from urine and dung (IPCC, 2019). It is also possible to develop individual EFs for N<sub>2</sub>O, partitioned into dung and urine, to produce disaggregated N<sub>2</sub>O EFs, which is widely recommended by multiple studies (Chadwick et al., 2018; Krol et al., 2016; Luo et al., 2019; van der Weerden et al., 2021, 2011). According to van der Weerden et al. (2021), this disaggregation can improve the accuracy of national inventories, but more data from poorly represented regions (e.g., Asia, Africa, South America) are needed.

Against this background, the aims of this study were i) to investigate the effect of combined cattle dung and urine patches on N<sub>2</sub>O and CH<sub>4</sub> emissions, compared with emissions from separate depositions, under different weather conditions and ii) to develop local disaggregated EFs for N<sub>2</sub>O and CH<sub>4</sub>. A quantitative assessment of literature data on the

effects of combined excreta patches was also performed to identify N<sub>2</sub>O emissions drivers. The starting hypotheses were that 1) separate urine and dung patches are localised N<sub>2</sub>O sources, and adding urine to dung patches results in a synergistic, rather than additive, increase in N<sub>2</sub>O emissions; 2) separate dung patches are localised CH<sub>4</sub> hotspots and adding urine to dung patches does not increase dung-based CH<sub>4</sub> emissions; and 3) the N<sub>2</sub>O EF for cattle urine is higher than for cattle dung patches, supporting disaggregation by excreta type.

## 2. Materials and methods

### 2.1. Site description

The study was performed at the campus of the National University of the Center of Buenos Aires Province (UNCPBA), Tandil, Argentina (37°19'07"S, 59°04'42"W, altitude 211 m asl), where the climate is warm temperate moist (following Fig. 10A.1 in IPCC, 2019). The mean annual temperature is 13.5 °C, and total annual precipitation is 880 mm (based on mean climate records 1981–2011). Data on air temperature at 2 m above the soil surface, and soil volumetric water content and soil temperature at 0.2 m depth during the study period were obtained from a meteorological station located 400 m from the experimental site. Precipitation was recorded with a rain gauge directly at the experimental site.

The soil at the experimental site is a Mollisol (typical Argiudoll, considered a fertile soil) (Panigatti, 2010). It comprises 37% sand, 37% silt and 26% clay. The topsoil (to 0.2 m depth) contains 2.7% organic C and 0.26% total N, its bulk density is 1080 kg m<sup>-3</sup> and the pH is 6.0 (Priano et al., 2017). During the study period, the soil was covered by naturalised pasture dominated by *Festuca arundinacea*, with presence of *Cirsium vulgare*, *Carduus acanthoides*, *Trifolium repens* and *Taraxacum* sp., which is typical of cattle pasture in the Pampa region. The experimental soil did not receive any fertiliser and had no grazing animals during the 14 years prior to the study (Faraminián et al., 2021). The grass was cut every month, between 0.1 and 0.25 m with a mowing machine, and removed from the area. Precisely, the pasture was manually cut within the collars containing the treatments when the height reached about 0.15 m to a final 0.05 m for a better measurement deployment.

### 2.2. Experimental design

Two separate 98-day trials were performed, the first in winter-spring (September 9 to December 17, 2019) and the second in summer-autumn (February 15 to May 25, 2020). As these experimental periods did not coincide precisely with any regular season in the region, they were named according to the predominant weather condition as "dry" and "wet" trials, respectively (since water content differed between the trials, but not mean temperature). More information on weather conditions is shown in the results (Section 3.1).

Treatments and the experimental design were similar in both trials (Fig. 1). Treatments consisted of the application of separate patches of fresh "urine", "dung", or a combination of dung + urine ("D+U"). A "control" treatment (without excreta) was included to determine background values and allow EF calculation for each excreta type. In both trials, the experimental site comprised a total area of 64 m<sup>2</sup> divided into four blocks (replicates), with four plots in each block (one per treatment). Within the respective plots, each treatment was applied in duplicate within circular collars (0.26 m in diameter) to simulate typical excreta patches on pasture. One of these duplicates was used for gas measurements and the other to monitor soil characteristics. The amount of fresh excreta applied to each chamber was based on the mean mass per defecation (2.5 kg) and mean volume per urination (0.75 L) defined during excreta collection. Naturally, urine and dung patches do not have "delimiting edges", and applying the excreta across the whole area of the chamber might affect N<sub>2</sub>O production. However, despite the limitations of the methodology, it was considered appropriate to comparatively

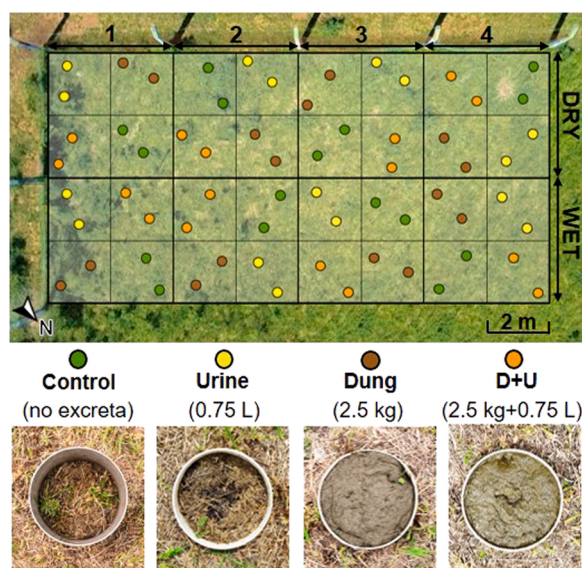


Fig. 1. Diagram of the field area used in the two trials (dry, wet), showing the experimental design with four treatments (control, dung, urine, D+U) and four replicates. Circles represent the location of each chamber (by duplicate within plots), colours indicate the treatment applied.

evaluate the  $N_2O$  emissions from overlapping excreta since this methodological approach has been previously used in published studies on excreta  $N_2O$  emissions that experts included in the IPCC database (Li et al., 2016; Luo et al., 2013; Thomas et al., 2017).

In the combined patch, 0.75 L of urine were applied overlapping the 2.5 kg of dung after its deposition. Both dung and urine were carefully applied from a height of 1 m directly to the centre of the collars, simulating the height of a natural excretion. The zone of influence included the entire collar as the patch-effective area. Fresh urine and dung were collected from the same lactating dairy cows (Argentinian Holstein,  $526 \pm 47$  kg live weight) between morning and afternoon milking the day before application in each trial (diets described below). Before the dry trial, 30 urinations were collected through manual stimulation under the vulva and poured together into a plastic can. Simultaneously, recently excreted dung was carefully collected from the naked soil surface of the individual feeding pens, weighed separately and then pooled and homogenised. In the wet trial, the same procedure was used for excreta collection (without quantification). The same urine and dung amounts were applied to a nearby field area to avoid the carry-over effect of treatments from the previous trial.

At collection in September (dry trial), the cows were grazing a pasture of *Lolium multiflorum* (ryegrass; intake  $7 \text{ kg DM animal}^{-1} \text{ d}^{-1}$ , 21% crude protein (CP) in DM basis), supplemented with a daily mixed ration of  $13 \text{ kg DM animal}^{-1}$  consisting of corn silage (5 kg DM, 6.6% CP) and energy concentrate (8 kg DM with 20% CP; 39% corn grain, 35% wheat bran, 22% soybean expeller, 4% minerals and vitamins, in DM basis). At collection in February (wet trial), the cows were grazing a pasture of *Medicago sativa* (alfalfa; intake  $13 \text{ kg DM animal}^{-1} \text{ d}^{-1}$ , 27% CP), supplemented with a daily concentrate ration of  $6 \text{ kg DM animal}^{-1}$  (with 27% CP).

### 2.3. $N_2O$ and $CH_4$ fluxes and emission factors

Greenhouse gas fluxes were measured using the static chamber technique. Sixteen cylindrical PVC collars (0.26 m internal diameter, 0.1 m height, area  $0.05 \text{ m}^2$ ) were used as the base of the static chambers. The chamber bases were inserted 0.05 m into the ground two days before excreta application to prevent soil disturbance influencing gas emissions and left in place for the entire trial period (98 days). The chamber bases were placed 1 m apart to prevent cross-contamination,

and the excreta doses were deposited inside. A lid of the same dimensions and material as the base (0.1 m height) was used during sampling to enclose the internal volume of the chamber. On each sampling occasion, the lid, equipped with a septum for gas sample collection, was fitted to the chamber base and sealed with an airtight rubber belt. Samples were collected using a 25 mL polypropylene syringe immediately after chamber closure (0 min) and after 10, 20 and 30 min to avoid saturation and ensure an adequate linear response in headspace gas concentrations ( $R^2 > 0.8$ ).

Sampling was conducted between 10:00 and 12:00 h when the emission rate resembled the daily average determined by Priano et al. (2014) for the study area. Periodic gas sampling began the day before excreta application to soil and continued for 98 days, when the excreta gas fluxes equalled those of the control, with a total of 20 and 22 sampling occasions for the dry and wet trial, respectively. The sampling frequency varied throughout the 98 days of each trial. The first measurement occurred three hours after treatment application, and the following measurements were performed daily in the first week. The sampling frequency then decreased from three to two times per week during weeks 2–5, once per week during weeks 5–11, and finally once per month until the end of the trial (i.e. week 14). Within two days of collection, all air samples were analysed using a gas chromatograph (GC, Agilent 7890 A) equipped with a flame ionisation detector (FID) for  $CH_4$  and an electron capture detector (ECD) for  $N_2O$  (see Lombardi et al., 2021 for details).

Gas concentration was calculated using the ideal gas law, and gas fluxes were estimated by calculating the rate of change in concentration over time using linear regression, as described in Lombardi et al. (2021). Fluxes were corrected for the air temperature at sampling (measured on the soil surface inside the chambers with a data logger i-button DS1921G) and the atmospheric pressure at the site (obtained from the nearby meteorological station). All flux data were checked for linearity by examining the  $R^2$  value of concentration versus time, with replicates excluded from the dataset at  $R^2 < 0.8$ . The absence of flux in the chambers (i.e. flux = 0) was assumed when the rate of change in gas concentration was below the analytical precision limit of the GC device. This limit was determined by analysing several ambient air samples and then calculating the coefficient of variation ( $CV = 2.1\%$  for  $CH_4$ , 3.8% for  $N_2O$ ). Cumulative gas emissions ( $\text{mg m}^{-2}$ ) were estimated by integrating the linear interpolation of the daily fluxes over time (OriginLab 2018 Software). In addition, the theoretical sum of cumulative  $N_2O$  emissions from the separate dung and urine patches (named " $D+U_{\text{theoric}}$ ") was calculated to compare with the combined D+U treatment.

Emission factors for  $CH_4$  and  $N_2O$  were calculated using estimated cumulative gaseous emissions. The EF for  $CH_4$  ( $\text{g } CH_4 \text{ kg}^{-1} \text{ VS}$ ) was calculated only for treatments containing dung as described by IPCC (2019), i.e. as  $CH_4$  emitted directly by the excreta patch minus  $CH_4$  emissions from the corresponding control chamber, divided by units of volatile solids (VS) applied, i.e. organic matter. The EF for  $N_2O$ , expressed as a percentage of N emitted per unit of N applied, was calculated by subtracting cumulative  $N_2O$  emissions occurring in control from cumulative  $N_2O$  emissions lost directly from the excreta patch and then dividing this by the amount of N applied inside the chamber. The units of  $N_2O$  and N inputs needed to be the same for this calculation (e.g. cumulative  $N_2O$  emissions in  $\text{g } N_2O\text{-N m}^{-2}$  and N applied in  $\text{g N m}^{-2}$ ).

### 2.4. Soil and excreta parameters

Portions of fresh excreta were taken immediately after homogenisation in each trial and analysed for initial characterisation. Two 100 mL subsamples of urine were stored at  $-18^\circ \text{C}$  with 5 mL sulphuric acid (5%) to immobilise N. Two 100 g subsamples of dung were dried at  $65^\circ \text{C}$  to constant weight to determine DM content. Dung subsamples dried at  $105^\circ \text{C}$  were milled, and a single composite sample was analysed for VS content (through loss-on-ignition in a muffle furnace at  $550^\circ \text{C}$  for

four hours; AOAC, 1995, method 942.05). Total N concentrations in urine and dung were determined by the Kjeldahl method (AOAC, 1995, method 991.20). Dung organic C content was calculated from the known VS content using the formula developed by Iglesias Jiménez and Pérez García (1992). Characteristics of the urine and dung used in the trials are presented in Table 1.

Adjacent to each chamber where GHG fluxes were measured, the same treatment was applied using an extra chamber base as a mould. This area was used to evaluate soil water content, water filled pore space (WFPS) and exchangeable  $\text{NH}_4^+$  and  $\text{NO}_3^-$  concentrations in the top 0.1 m of soil. In addition, a 0.03 m diameter mould was used to sample the superficial dung and D+U patch for water content monitoring. Samples were collected on six occasions (days 3, 10, 22, 35, 49 and 98 in the dry trial; days 2, 10, 22, 41, 66 and 98 in the wet trial). Approximately 10 g of these samples were used to determine moisture gravimetrically by drying the soil and excreta samples at 105 °C until constant weight. The WFPS was calculated as Paul (2015) described using the soil water content (g  $\text{H}_2\text{O}$ /g dry soil) of each treatment and sampling day and the bulk density measured at the beginning of each trial. Soil particle density was assumed to be 2650  $\text{kg m}^{-3}$ . Separate moist samples, also 10 g, were subjected to extraction with 2 M KCl for determination of  $\text{NH}_4^+$  by the sodium salicylate-nitroprusside method and colourimetric reading at 667 nm (Borrero Tamayo et al., 2017) and direct determination of  $\text{NO}_3^-$  by salicylic acid nitrated in an alkaline solution and reading at 410 nm (Forster, 1995). For moisture correction, the gravimetric moisture content at 105 °C was applied.

## 2.5. Data analysis

Studies reporting field measurements of  $\text{N}_2\text{O}$  fluxes from excreta patches were compiled using Scopus® from Elsevier B.V. as the source of peer-reviewed papers to analyse the published data. Among the hits obtained, five studies best matching the characteristics of the present study were selected (Hyde et al., 2016; Li et al., 2016; Tully et al., 2017; van Groenigen et al., 2005b; Zhu et al., 2021). Including criteria were: i) field studies with  $\text{N}_2\text{O}$  measurements using the static chamber technique and GC; ii) treatments including separate cattle excreta patches (dung and urine), a combination of both (maintaining the amount of excreta applied in both separate patches) and a background (i.e. no excreta application); and iii) field monitoring for longer than 30 days. From these studies, data on mean cumulative  $\text{N}_2\text{O}$  emissions from each treatment were gathered by extracting from tables or the main text or, when these were not available, from diagrams using Data Thief III (<https://datathief.org/>). Within each study, each excreta type (control, urine (U), dung (D), and D+U) identified by the authors, divided into different weather conditions (dry and wet), was considered an observational unit. Also, specific study characteristics, such as climate zone, soil type, and amount of excreta applied, were gathered. Data were corrected for soil baseline  $\text{N}_2\text{O}$  emissions (by subtracting the control

**Table 1**  
Excreta composition and N application rates used in the dry and wet trials.

	Dry	Wet
<b>Excreta composition</b>		
<i>Urine</i>		
N ( $\text{g L}^{-1}$ )	5.0	8.9
<i>Dung</i>		
DM ( $\text{g kg}^{-1}$ )	126.8	123.3
VS ( $\text{g kg}^{-1}$ DM)	761.0	847.5
N ( $\text{g kg}^{-1}$ DM)	23.2	24.8
C ( $\text{g kg}^{-1}$ DM)	410.9	456.4
C:N ratio	17.7	18.4
<b>N application rates</b>		
<i>Urine</i> ( $\text{g N m}^{-2}$ )	72.1	126.9
<i>Dung</i> ( $\text{g N m}^{-2}$ )	140.6	145.1
<i>D+U</i> ( $\text{g N m}^{-2}$ )	212.7	272.0

DM: dry matter, VS: volatile solids, N: nitrogen, C: carbon.

value from each excreta result) to compare cumulative  $\text{N}_2\text{O}$  emissions across different experimental site characteristics (i.e. background properties). The theoretical sum of each observation ( $\text{D+U}_{\text{theoric}}$ ) was calculated by adding the separate values for dung and urine and dividing by the amount of N applied to avoid differences due to variations in excreta N input. The proportional increase in cumulative  $\text{N}_2\text{O}$  emissions was defined as  $\Delta = (\text{D+U}/\text{D+U}_{\text{theoric}})$ , i.e. comparing the combined D+U and  $\text{D+U}_{\text{theoric}}$ . With this definition, the effect of combining excreta is additive when the interaction term is equal to one and super-additive (synergetic) if the value shifts toward higher values ( $>1$ ). This approach helped interpret the available data on response ratios of  $\text{N}_2\text{O}$  emissions ( $N = 12$ , including our results). The statistical significance of differences in the proportional increase between dry and wet conditions was tested using ANOVA.

Statistical analyses were performed using the software InfoStat (v. 2020), linked to the R programming environment (Di Rienzo et al., 2020). Daily  $\text{CH}_4$  and  $\text{N}_2\text{O}$  fluxes and soil parameters (variables with repeated measures) were analysed for each trial separately using a generalised linear mixed model (glmm, lmer package) with excreta treatment and sampling day as fixed effects and field plot as a random effect. Several models were developed with different link functions. The most appropriate fit was selected according to the lowest Akaike's information criterion (AIC) value for models that met the assumptions of residuals (homogeneity and normal distribution). Cumulative emissions and EFs were analysed for each trial separately with a glmm including excreta treatment and plot as fixed and random effect, respectively. Differences were determined using the LSD Fisher test at  $P < 0.05$  level. Correlations between soil/dung parameters and gas fluxes were determined using Spearman's correlation coefficient ( $\rho$ ).

## 3. Results

### 3.1. Experiment characteristics

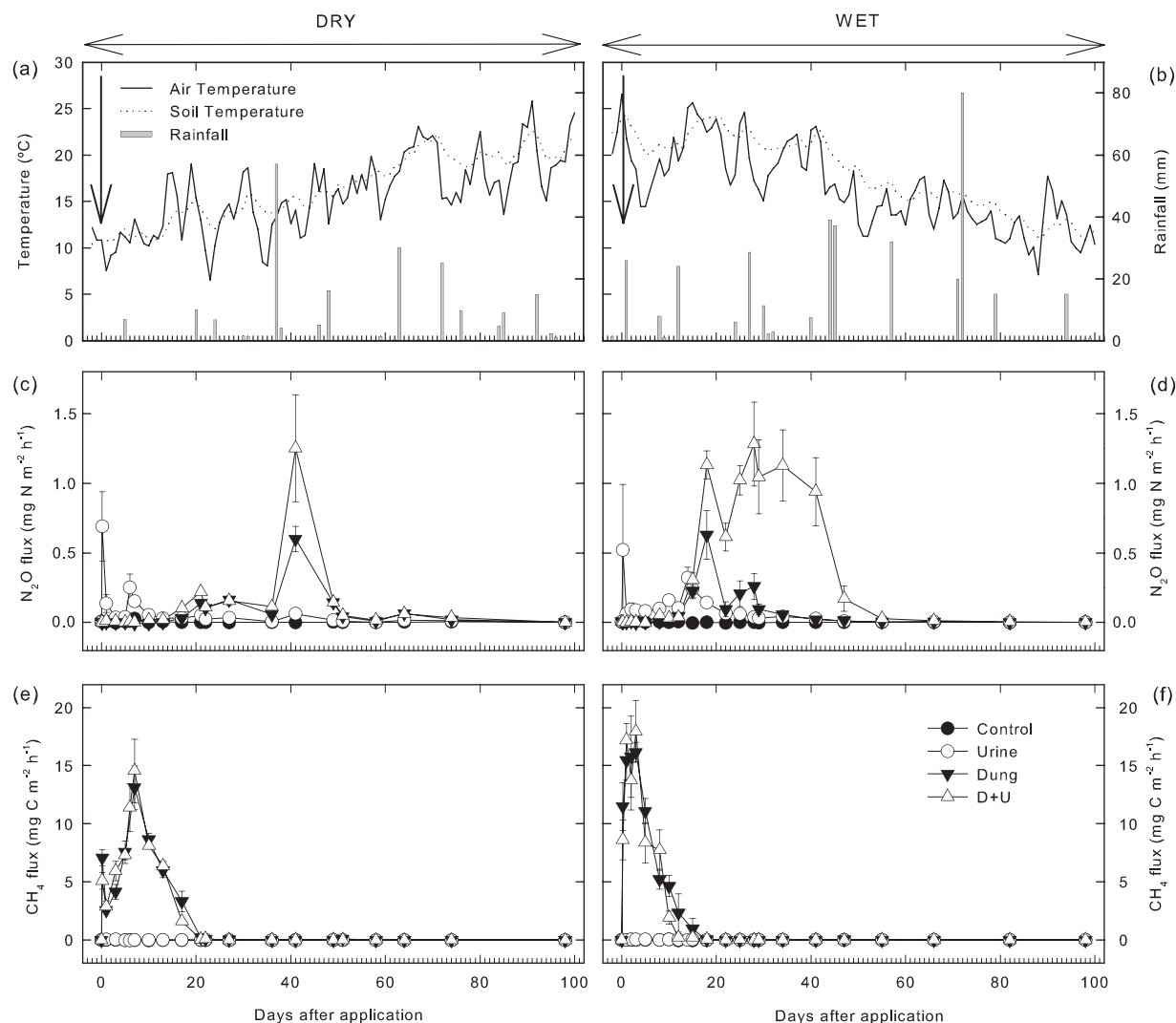
Overall mean air and soil temperature (16 °C and 18 °C) at the meteorological station were similar for the two 98-day trials (Fig. 2a, b). However, the dry trial had lower cumulative rainfall and soil volumetric water content (210 mm and 35%) than the wet trial (360 mm and 42%, respectively). During the initial ten days after excreta application, the air and soil temperature and rainfall were lower in the dry trial (11 °C, 11 °C and 7 mm, respectively) than in the wet trial (19 °C, 22 °C and 35 mm, respectively).

The chemical composition of the excreta across trials is shown in Table 1. Urine N content per volume in the dry trial was about half that in the wet trial. Therefore, the amount of total N applied inside each chamber containing urine, dung, and the D+U in the dry trial was 3.8, 7.4 and 11.1 g, respectively, while in the wet trial, it was 7.6, 6.6, and 14.2 g, respectively. These values converted to rates ( $\text{g N m}^{-2}$ ) are shown in Table 1. The organic C applied in dung patches (chambers) in the dry and wet trials was 130.2 and 139.6 g, respectively.

### 3.2. $\text{N}_2\text{O}$ and $\text{CH}_4$ fluxes

The deposition of the different excreta treatments showed differing emissions patterns depending on the type of excreta, producing localised GHG hotspots (Fig. 2). The data included several points on both the ascending and descending parts of the flux curves, suggesting that the gas sampling intervals were sufficient to capture the pulse of GHG emissions after excreta deposition.

Within each trial, the daily  $\text{N}_2\text{O}$  fluxes revealed a distinct pattern depending on the excreta treatment and time since application (Fig. 2c, d), resulting in a significant effect of excreta treatment, sampling day and their interaction ( $P < 0.001$  for all, in both trials). Soil  $\text{N}_2\text{O}$  fluxes (from control chambers) were low across both trials and sometimes even negative, also observed and explained in Lombardi et al. (2022). In urine patches,  $\text{N}_2\text{O}$  emissions increased immediately after urine application



**Fig. 2.** Dynamics of (a,b) rainfall and mean daily air and soil temperature and changes in (c,d) nitrous oxide (N<sub>2</sub>O-N) and (e,f) methane (CH<sub>4</sub>-C) fluxes over time (days) after application of cattle excreta (arrows) during the dry trial (left; from Sept 9 to Dec 17, 2019) and wet trial (right; from Feb 15 to May 25, 2020). Bars represent standard error of the mean (n = 4).

and then decreased to reach background levels at around day 49 and day 28 in the dry and wet trials, respectively. In contrast, N<sub>2</sub>O emissions from dung and D+U patches increased slowly until they reached a peak and then decreased. The N<sub>2</sub>O peak from the D+U patches was double the N<sub>2</sub>O peak from the dung only and lasted longer. During the dry trial, the dung and D+U patches took 74 and 98 days to reach similar fluxes as control, respectively, whereas in the wet trial, this took 41 and 55 days, respectively.

Cumulative N<sub>2</sub>O emissions were significantly affected by excreta application (Table 2). The combination of D+U gave the highest cumulative N<sub>2</sub>O emissions in both trials. Although urine-based cumulative N<sub>2</sub>O emissions were at least ten times greater than control values in both trials, there were no statistical differences between those treatments. There were also no statistical differences in cumulative N<sub>2</sub>O emissions between dung, urine and control during the wet trial. In addition, cumulative N<sub>2</sub>O emissions from the D+U patches were 1.2 and 3.1 times greater than the theoretical sum (D+U<sub>theoretic</sub>) of the separate excreta patches, with total D+U<sub>theoretic</sub> emissions of 265.3 and 236.6 mg N<sub>2</sub>O-N m<sup>-2</sup> in the dry and wet trials, respectively. These values were significantly different in the wet trial (P = 0.007) but were not significant in the dry trial (P = 0.357).

The quantitative assessment comparing combined D+U and D+U<sub>theoretic</sub> on N<sub>2</sub>O emissions from this and the five available studies

(Hyde et al., 2016; Li et al., 2016; Tully et al., 2017; van Groenigen et al., 2005b; Zhu et al., 2021) showed a mean proportional increase of  $\Delta = 1.44$  (SEM 0.18). Only three observations (from N = 12) were below the additive effect ( $0.94 < \Delta < 1$ ), while half the values were within the range  $0.94 < \Delta < 1.27$ , and the rest  $\Delta > 1.27$ . A trend for an additive effect ( $\Delta = 1.15$ ) was observed under dry weather conditions and for a synergetic effect ( $\Delta = 1.73$ ) under wet conditions (SEM 0.23, significant at 10% level) (Fig. 3). No specific study characteristics, such as climate zone, soil type or amount of excreta applied, coincided with the explanation of any variation in the data.

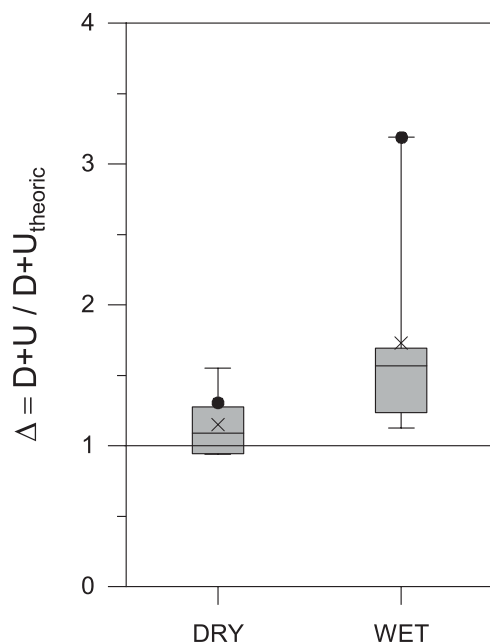
The EF for N<sub>2</sub>O was not affected by the different excreta treatments during the dry trial but under wetter conditions (Table 2). Thus, the combined D+U differed from the separate urine and dung patches during the wet trial, with a significantly greater N<sub>2</sub>O EF. Furthermore, there were no significant differences in N<sub>2</sub>O EF when analysing by disaggregation among separate excreta types (urine vs dung) in both trials (P = 0.18 and P = 0.68 for dry and wet, respectively). There was also no seasonal variation within separate dung or urine patches (P = 0.16 and P = 0.64, respectively). Thus, the mean N<sub>2</sub>O EF values obtained for separate dung and urine patches in this study were 0.11 and  $0.09 \pm 0.01\%$ , respectively, without significant differences (P = 0.19). Considering all three types of excreta applied, the mean N<sub>2</sub>O EF was  $0.14 \pm 0.08\%$ .

**Table 2**

Cumulative emissions and emission factors (EFs) for nitrous oxide (N<sub>2</sub>O) and methane (CH<sub>4</sub>) as affected by the type of excreta deposited on grassland during the dry and wet trials.

	Control	Urine	Dung	D+U	SE	P-value
<b>Dry trial</b>						
<i>Cumulative emissions</i>						
N <sub>2</sub> O (mg N m <sup>-2</sup> )	6.9 a	72.6 a	192.7 b	327.7c	30.1	< 0.001
CH <sub>4</sub> (mg C m <sup>-2</sup> )	-56.2 a	-53.3 a	2546.9 b	2670.1 b	218.5	< 0.001
<i>Emission factor, EF</i>						
N <sub>2</sub> O (%)	n.d.	0.09 a	0.13 a	0.15 a	0.02	0.194
CH <sub>4</sub> (g kg <sup>-1</sup> VS)	n.d.	n.d.	0.75 a	0.79 a	0.09	0.785
<b>Wet trial</b>						
<i>Cumulative emissions</i>						
N <sub>2</sub> O (mg N m <sup>-2</sup> )	3.4 a	103.2 a	133.4 a	736.9 b	61.9	< 0.001
CH <sub>4</sub> (mg C m <sup>-2</sup> )	-31.9 a	-17.7 a	2538.9 b	2822.7 b	174.4	< 0.001
<i>Emission factor, EF</i>						
N <sub>2</sub> O (%)	n.d.	0.08 a	0.09 a	0.27 b	0.03	0.002
CH <sub>4</sub> (g kg <sup>-1</sup> VS)	n.d.	n.d.	0.77 a	0.69 a	0.07	0.447

N: nitrogen, C: carbon, VS: volatile solids. Values are the mean and SE standard error (n = 4). Different lowercase letters within rows indicate significant differences between excreta treatments (p < 0.05).



**Fig. 3.** Proportional increase ( $\Delta$ ) in cumulative N<sub>2</sub>O emissions from the combined D+U treatment compared to the theoretical sum D+U<sub>theoretic</sub> of the individual patches, separated by weather conditions (dry and wet). If the relation is  $\Delta = 1$ , then the effect of combining urine and dung on N<sub>2</sub>O emissions is additive. If the  $\Delta > 1$ , combining urine and dung gives a positive synergetic effect on N<sub>2</sub>O production, more than the sum of the individual emissions. Data included the present and previously published studies (N = 12), and calculation details are in "Data analysis" section. Crosses represent the average data values; black circles are this study's result. The range of whiskers is minimum and maximum values.

The daily CH<sub>4</sub> fluxes were significantly affected by sampling day, type of excreta treatment and their interaction (P < 0.001 for all, in both trials) (Fig. 2e, f). Control and urine chambers were a sink for atmospheric CH<sub>4</sub> during the two trials (mean flux of -0.02 mg CH<sub>4</sub>-C m<sup>-2</sup> h<sup>-1</sup>), although transient CH<sub>4</sub> emissions of about 0.03 mg CH<sub>4</sub>-C m<sup>-2</sup> h<sup>-1</sup> were detected shortly after urine application. The CH<sub>4</sub> fluxes from the dung and D+U patches were observed immediately after excreta application (faster in the wet trial), which decreased to near background levels. From days 21 and 14 onwards in the dry and wet trials, respectively, the CH<sub>4</sub> fluxes from all treatments were statistically similar. Cumulative CH<sub>4</sub> emissions were significantly affected by the excreta treatments applied (Table 2). Those from urine patches were similar to control, whereas patches containing dung acted as a CH<sub>4</sub> source without being significantly affected by urine addition in both trials. The CH<sub>4</sub> EF for dung (with and without urine) averaged 0.75 ± 0.14 g CH<sub>4</sub> kg<sup>-1</sup> VS (range 0.52–1.01 g CH<sub>4</sub> kg<sup>-1</sup> VS), with no effect of urine addition.

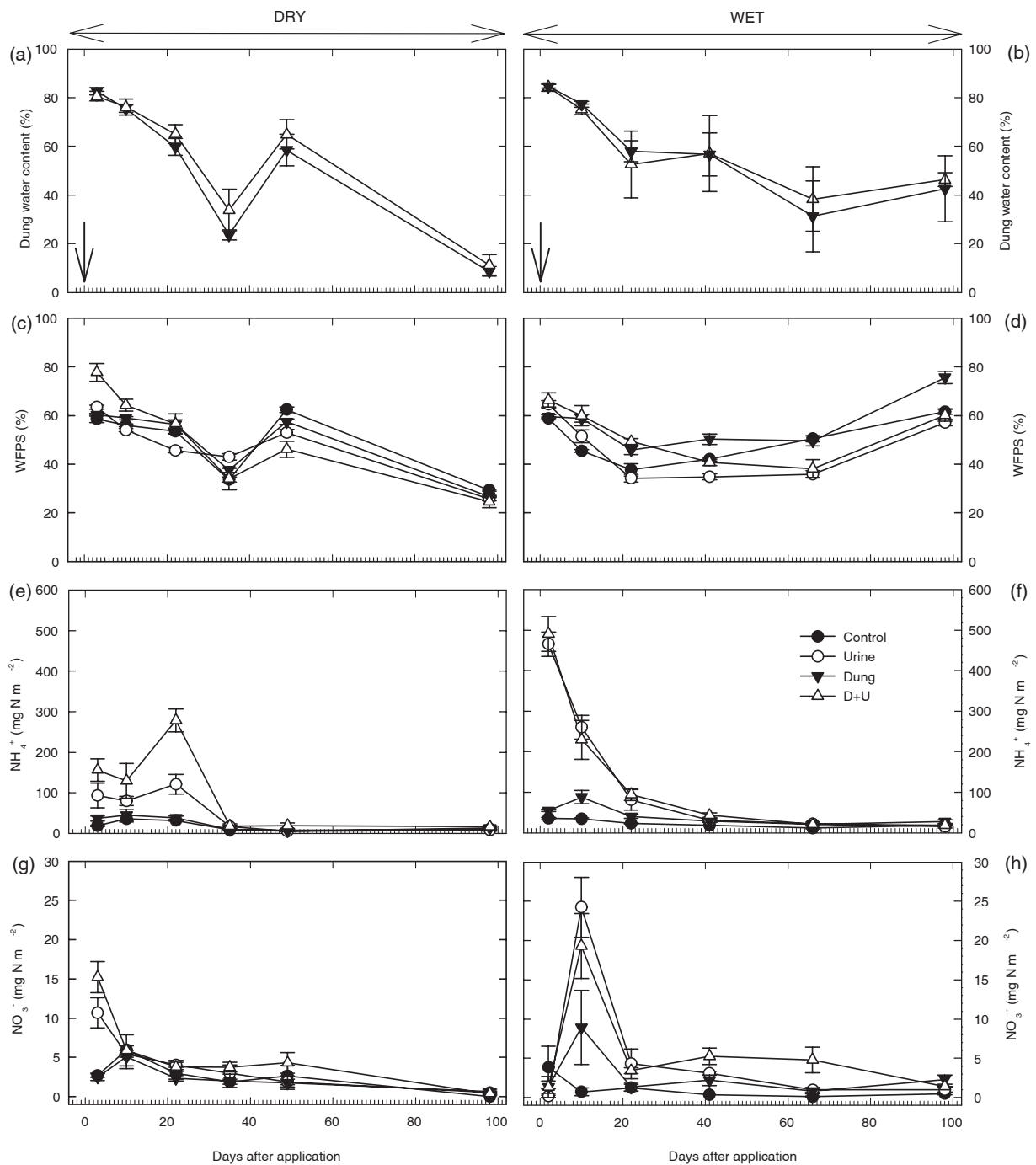
### 3.3. Changes in soil and excreta parameters

Dung water content decreased over time after application in both trials, starting from an initial average moisture of 88% (Fig. 4a, b). Dung moisture reached lower values during the dry trial, which was significantly affected by addition of urine, sampling day and their interaction (P = 0.029, P < 0.001 and P = 0.013, respectively). Under wet conditions, dung moisture was not significantly affected by addition of urine (P = 0.834). Dung moisture content was significantly and positively correlated with dung-based CH<sub>4</sub> flux in the dry ( $\rho = 0.85$  and  $\rho = 0.80$  with and without urine addition, respectively) and in the wet trial ( $\rho = 0.72$  and  $\rho = 0.70$ , respectively). The dung patches began to show a thin crust layer of approximately 0.05 m due to drying the first week after deposition in the dry trial and the second week in the wet trial. Large numbers of ants and woodlice were observed under the dung patches 20–30 days after application.

The mean value of WFPS, including all treatments, was slightly lower in the dry trial (49.2 ± 15.3%) than in the wet trial (51.5 ± 12.8%) but without significant differences (P = 0.27). Within each trial, the soil WFPS revealed a distinct pattern depending on the excreta treatment and time since application (Fig. 4c, d), with a significant interaction in both trials (P < 0.001). For instance, soil with urine addition had the highest WFPS values (59.0–78.0%) during the first ten days after deposition, but it had lower values than soil beneath dung the rest of the days. The soil beneath dung had the highest mean WFPS values over the entire trials (49.6–56.7%). The mean WFPS in the control soil was 48.9% and 49.8% for the dry and wet trials, respectively. WFPS was significantly correlated with N<sub>2</sub>O fluxes from the urine patches in the dry trial (r = 0.43, P < 0.04) and with control soil in the wet trial (r = 0.52, P < 0.02).

The dominant mineral N form in the soil was exchangeable NH<sub>4</sub><sup>+</sup>, while the NO<sub>3</sub> content remained lower, representing about 10% of the NH<sub>4</sub><sup>+</sup> content within each treatment throughout both trials. The NH<sub>4</sub><sup>+</sup> concentration in the soil beneath excreta patches increased following excreta application, especially in urine. It was affected by the type of excreta applied (P < 0.001), with significant variation also between sampling days (P < 0.001) (Fig. 4e, f). Higher exchangeable NH<sub>4</sub><sup>+</sup> concentrations were observed in the soil beneath patches containing urine during the wet trial than during the dry trial. Similarly, the NO<sub>3</sub> content increased following urine application (Fig. 4g, h). The NO<sub>3</sub> concentration in soil was also affected by excreta treatment (P < 0.02) and sampling day (P < 0.001). Application of dung without urine did not significantly increase the NH<sub>4</sub><sup>+</sup> and NO<sub>3</sub> content in the soil beneath patches compared with the control.

During the dry trial, the NO<sub>3</sub> content in the soil under urine patches was significantly and strongly correlated with N<sub>2</sub>O flux (r = 0.57, P < 0.04). In contrast, during the wet trial, the exchangeable NH<sub>4</sub><sup>+</sup> content in the soil under urine patches was significantly and strongly correlated with N<sub>2</sub>O fluxes (r = 0.77, P < 0.001). The other treatments



**Fig. 4.** Change in (a,b) dung water content and (c,d) water full pore space (WFPS), (e,f) ammonia ( $\text{NH}_4^+$ ) and (g,h) nitrate ( $\text{NO}_3^-$ ) content in soil beneath excreta treatments over time (days) after application of cattle excreta (arrows) in the dry trial (left; from Sept 9 to Dec 17, 2019) and wet trial (right; from Feb 15 to May 25, 2020). Bars represent standard error of the mean (n = 4).

showed no significant correlation between  $\text{NH}_4^+$  or  $\text{NO}_3^-$  content in soil and  $\text{N}_2\text{O}$  flux. Probably, with a shallower soil sampling depth, more differences in soil parameters and better correlation with GHG fluxes could be observed. None of the treatments had significant amounts of  $\text{NH}_4^+$  or  $\text{NO}_3^-$  remaining in the soil at the end of the trial periods (day 98).

#### 4. Discussion

##### 4.1. Effect of combined dung and urine patches on $\text{N}_2\text{O}$ fluxes

The key finding in this study was that adding urine to dung patches under wet conditions gave a significant threefold increase (synergetic

effect) in cumulative  $\text{N}_2\text{O}$  emissions compared with the theoretical sum  $\text{D}+\text{U}_{\text{theoric}}$  of the separate patches. This confirms the starting hypothesis (1). However, under dry conditions, the cumulative  $\text{N}_2\text{O}$  emissions from the combined  $\text{D}+\text{U}$  showed an additive effect compared with the  $\text{D}+\text{U}_{\text{theoric}}$  value. Assessment of the proportional increase in  $\text{N}_2\text{O}$  emissions from available data (Fig. 3) also indicated, although with weak evidence, a synergetic effect in wet conditions and an additive effect during dry weather. Three of the studies reviewed provided strong evidence of such an effect in temperate and tropical climates (Hyde et al., 2016; Li et al., 2016; Tully et al., 2017), whereas another study found weak evidence (van Groenigen et al., 2005b). And Zhu et al. (2021) found no significant differences pointing to an additive effect regardless

of weather conditions. Thus, there is still a gap in understanding the interaction between dung and urine and the main factors affecting the  $N_2O$  emissions.

Direct  $N_2O$  emissions from excreta depositions (separate or combined) were related, to some degree, to changes in moisture, soil WFPS, and soil N content. This was confirmed in the present study by the strong correlation between soil WFPS, mineral N and urine-based  $N_2O$  emissions. It is well documented that urine deposition unavoidably increases denitrification, at least in the short-term, increasing  $N_2O$  production (de Klein et al., 2003). Denitrification activity is also expected to increase after dung depositions, promoted by the anaerobic conditions and high levels of soil  $NO_3^-$  and readily available C (Oenema et al., 2008). However, no significant correlation was found between soil properties and  $N_2O$  fluxes from patches containing dung. The soil beneath D+U patches initially accumulated  $NH_4^+$  due to the rewetting with urine (Fig. 4e, f) but did not immediately increase  $N_2O$  emissions. It began to emit 15 days after application, similar to separate dung. It showed significant  $N_2O$  emissions when dung water content was between 50% and 70%, soil WFPS between 40% and 60% and soil mineral N content higher than control. This indicates that N transformation in D+U results from an interrelation of factors controlling excreta degradation, including what occurs within the patch.

Higher soil  $NH_4^+$  and  $NO_3^-$  contents were observed beneath excreta patches in the wet trial. It seems that the wetter conditions promoted further N mobilisation. According to Haynes and Williams (1993), the weather immediately after dung depositions and the initial consistency of dung affect the initial release of nutrients. When the wetter conditions followed excreta application in this study, the crust took longer to form and more rain could penetrate and rewet the patch (supported by the higher dung moisture values, Fig. 4a, b). As denitrification is a microbially facilitated process, it is strongly controlled by moisture and temperature (de Klein et al., 2003). Thus, the synergetic increase in D+U under wet conditions might have been due to the formation of more anaerobic microsites for  $N_2O$  production. High  $N_2O$  emissions were also observed after rainfall events (>10 mm); for instance, after the 57 mm rain event in the dry trial, which probably indicates that low moisture limits denitrification. Similarly, van Groenigen et al. (2005b) observed increased  $N_2O$  emissions from combined dung and urine patches during wet summer conditions (mean temperature >20 °C, WFPS ~60%), while Tully et al. (2017) found elevated  $N_2O$  emissions coincident with rainfall events. However, Zhu et al. (2021) did not observe such effect, possibly because of the little rain registered during the rainy seasons that did not increase soil moisture enough to promote denitrification (maximum WFPS < 44% in rainy seasons). Therefore, the wetter the weather, the greater the denitrification and  $N_2O$  stimulation.

Moreover, mineral N concentrations in the soil beneath D+U were higher during the wet trial, likely due to higher urine N content derived from higher dietary N intake. In ruminants, it is well documented that the dietary N use efficiency decreases as the N intake increases, being the urine the main via of N excretion (Dijkstra et al., 2013; Whitehead, 2000). This increase in urine N content during the wet trial likely reduced the C:N ratio of the combined D+U patch. This ratio was negatively correlated with N mineralisation and  $N_2O$  emissions in several studies (Pelster et al., 2016; Simon et al., 2018). The lower C:N ratio of D+U in the wet trial would probably be responsible for the higher  $N_2O$  emissions. Thus, when excreta are combined, moisture and C:N ratio can play an important role in N transformations and  $N_2O$  emissions.

Results from this study indicate that management practices that reduce the load of N derived from overlapped excreta depositions could mitigate  $N_2O$  emissions from grasslands under wet conditions in temperate climates. Rotating livestock accumulation areas in the field could cycle nutrients more efficiently, reduce losses and lower the fertiliser use, resulting in a more sustainable system (Luo et al., 2017; Saggari et al., 2022). Strip grazing and movable troughs for water and supplementation could help make the distribution of excreta patches as

uniform as possible (Whitehead, 2000). The success of applying these strategies may also be seasonal, by moving water points when stressing summer conditions are present (White et al., 2001) or by reducing grazing time during wet months of the year (de Klein et al., 2006). However, animal housing periods may create other hotspots, meaning that GHG emissions should be investigated using a life cycle assessment approach.

#### 4.2. Urine addition did not affect $CH_4$ fluxes

The addition of urine to dung patches had a negligible effect on dung-based  $CH_4$  emissions, agreeing with the hypothesis (2). Similar findings have been made in other studies examining  $CH_4$  emissions from combined excreta patches (Liao et al., 2018; Zhu et al., 2021). However, another study found that urine addition to dung patches can significantly decrease  $CH_4$  emissions compared with the dung patch alone (Tully et al., 2017). They attributed the decrease in  $CH_4$  to the redox potential of the different treatments. In the present study, urine addition most likely simultaneously stimulated and inhibited  $CH_4$  production, giving no overall significant effect. Similarly, soil  $CH_4$  fluxes were not significantly affected by urine addition. Other studies have found similar results for temperate and tropical grasslands (Nichols et al., 2016; Pelster et al., 2016; Zhu et al., 2021), where both control and urine depositions acted as sinks for atmospheric  $CH_4$ , regardless of the type of pasture. According to Cai et al. (2017), urine deposition does not influence  $CH_4$  fluxes from different soils, regardless of the type of grazing animal involved.

Patches containing dung acted as localised  $CH_4$  hotspots and depended strongly on the age of the dung (supported by the significant effect of time after deposition). Several studies have observed similar  $CH_4$  emission patterns in temperate climates (Kelly et al., 2016; Nichols et al., 2016; Priano et al., 2014). Dung  $CH_4$  emissions are attributed to entrained enteric  $CH_4$  from freshly voided dung and new  $CH_4$  produced within the dung patch (Cai et al., 2017). The latter is often considered the main origin of  $CH_4$  from dung patches, explained by the simultaneous abundance of methanogens, readily available soluble C and anaerobic conditions (Hahn et al., 2018). In the present study, dung moisture content appeared to explain the dung and D+U based  $CH_4$  emissions since high  $CH_4$  emissions were observed when moisture was > 65%, i.e. when anoxic conditions were present, supported by the strong positive correlation between these parameters.

#### 4.3. Emission factors

Dung-based  $N_2O$  EF values in this study ranged from 0.05% to 0.16% and were within the 0.01–0.53% range reported in other studies performed under temperate climates (Table 3). Moreover, the urine-based  $N_2O$  EFs showed lower values (range 0.03–0.14%) than those gathered in Table 3 (range 0.05–2.93%). There were no significant differences in  $N_2O$  EF between separate urine and dung patches. This contradicts hypothesis (3) and previous findings recommending disaggregation for excreta type with higher EFs from urine patches (Bell et al., 2015; Chadwick et al., 2018; Krol et al., 2016; Luo et al., 2019; van der Weerden et al., 2011, 2020, 2021). The low  $N_2O$  emissions from urine could be explained by high N uptake by plants and soil heterotrophs of long-term unfertilised grassland, which are strongly N-limited and can outcompete nitrifiers (Liu et al., 2016). The low soil fertility at our study site (SOC 2.7%, total N 0.26%, pH 6.0) could also mean that, if the N input is below a certain threshold, there might not be enough substrate (e.g.  $NO_3^-$ ) for  $N_2O$  production, as observed by van der Weerden et al. (2020). In line with this, a review by Cai and Akiyama (2016) found that when specific soil conditions were present (SOC <4%, total N < 0.4, pH >5.5), coinciding with the soil characteristics mentioned above, there were no significant differences between dung and urine  $N_2O$  EF.

Combined D+U under wet field conditions resulted in threefold higher  $N_2O$  EF than the separate depositions, whereas there were no

**Table 3**

Field studies in temperate climates measuring N<sub>2</sub>O from excreta patches are shown. Data included location, measuring period (days), and mean N<sub>2</sub>O EF (%) range for urine and dung patches.

Reference	Location	Days	Urine EF (%)	Dung EF (%)
Bell et al. (2015)*	United Kingdom	365	0.20–1.07	0.10–0.20
Cardenas et al. (2016)*	United Kingdom	350	0.11–2.96	0.10–0.39
Chadwick et al. (2018)*	United Kingdom	365	0.05–2.93	0.04–0.53
Hyde et al. (2016)*	Ireland	180	0.11	0.01
Li et al. (2016)*	New Zealand	271	1.72	0.48
Lombardi et al. (2021)	Argentina	125	n.d.	0.06–0.13
Luo et al. (2013)*	New Zealand	120–365	0.07–1.35	0.02–0.40
Luo et al. (2019)	New Zealand	90–180	0.10–0.72	0.05–0.27
Nichols et al. (2016)*	United States	365	0.11–0.13	0.10
Rochette et al. (2014)*	Canada	120–365	0.11–2.63	0.04–0.28
Thomas et al. (2017)*	Canada	365	1.32	0.03
van der Weerden et al. (2011)*	New Zealand	135–180	0.05–0.94	0.00–0.17
This study	Argentina	98	0.03–0.14	0.05–0.16
IPCC (2006)	Global	365	2.00	2.00
IPCC (2019)	Wet climate locations	365	0.77 (0.03–3.82)	0.13 (0.00–0.53)

\* Studies used by IPCC (2019) for estimation of default N<sub>2</sub>O EF.

significant differences under dry conditions (Table 2). These results were consistent with van Groenigen et al. (2005a), who found that dung addition to urine patches led to an eightfold increase in N<sub>2</sub>O EF and pointed out a need for realistic EF to consider the sites where cattle congregate. However, this N<sub>2</sub>O EF (0.10–0.39%) were below the updated 0.6% default EF estimated by IPCC for cattle N in wet climates (IPCC, 2019). Besides, when disaggregating for excreta type, mean urine-based N<sub>2</sub>O EF (0.09%) was lower than the urine-EF value (0.77%) of IPCC (2019), whilst mean dung-based N<sub>2</sub>O EF (0.11%) was closer to the dung-EF value (0.13%) of IPCC (2019). Therefore, the more recent IPCC values seem more accurate for the Pampa region than the previous IPCC default value (2%; IPCC, 2006), which overestimated N<sub>2</sub>O emissions from cattle excreta patches.

The mean CH<sub>4</sub> EF for patches containing dung, regardless of urine addition, was 0.75 ± 0.14 g CH<sub>4</sub> kg<sup>-1</sup> VS. This value was within the range between 0.3 and 1.9 g CH<sub>4</sub> kg<sup>-1</sup> VS of other studies performed in Argentina, Australia and Colombia (Lombardi et al., 2021, 2022; Kelly et al., 2016). Besides, it was within the range of the updated IPCC value (0.6 ± 0.2 g CH<sub>4</sub> kg<sup>-1</sup> VS) for dung deposition on rangelands (IPCC, 2019). As few studies have examined CH<sub>4</sub> EF from dung patches and expressed it based on VS, more region-specific studies are necessary.

## 5. Conclusions

This study showed that cattle urine and dung deposition affect CH<sub>4</sub> and N<sub>2</sub>O emissions from grassland soils in different ways. Separate dung patches were found to be a localised CH<sub>4</sub> hotspot while adding urine to dung patches had a negligible effect on CH<sub>4</sub> emissions. In addition, separate and combined urine and dung depositions were also localised N<sub>2</sub>O sources. The results indicated a synergistic (super-additive) effect on cumulative N<sub>2</sub>O emissions from overlapping dung and urine patches, compared with individual depositions, under wet conditions. This suggests that avoiding combined dung and urine patches under wet conditions, e.g., encouraging a more uniform distribution of grazing animals, could be part of an alternative mitigation strategy for N<sub>2</sub>O emissions in temperate managed grazed pastures.

The mean dung-based CH<sub>4</sub> EF (0.75 g CH<sub>4</sub> kg<sup>-1</sup> VS) found here,

representing pasture systems common in the Pampa region of Argentina, was within the range of the updated IPCC (2019) value (0.6 g CH<sub>4</sub> kg<sup>-1</sup> VS) for dung deposition on pasture. The mean direct N<sub>2</sub>O emissions (0.14%) were well below the EF of 2% established by IPCC 2006 for cattle N in excreta. Thus, the revised dung (0.13%) and urine (0.77%) EF for wet climates in IPCC (2019) are much closer to the value found in this study for dung (0.11%), but higher than the value found for urine (0.09%). This suggests a need to reduce dependence on default Tier 1 EF values and invest in studies to develop more region-specific Tier 2 EFs for the IPCC Guidelines for National Greenhouse Gas Inventories. Additional studies are required to verify these findings under different cattle production systems and environmental conditions to develop robust EFs from excreta depositions on pastures.

## Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## Data Availability

Research Data from combined an separate cattle dung and urine depositions on N<sub>2</sub>O and CH<sub>4</sub> emissions under different weather conditions are included: <https://data.mendeley.com/datasets/tmhyptg3xy>.

## Acknowledgements

The authors would like to thank the group “Fisicoquímica Ambiental” (GFQA-IFAS-UNCPBA) for its support during sampling, “Fundación Instituto de la Leche” (FIL) for supplying cattle excreta, Claudia Faverin (EEA INTA Balcarce) for lending the static chambers, and “Instituto de Hidrología de Llanuras” (IHLLA-UNCPBA) for providing meteorological data. Banira Lombardi also gratefully acknowledges Gabriel Gatica for statistics guidance, Eduardo Vázquez for writing assistance and the Environment and Health Applied Sciences Doctoral Program (DCAAS, UNCPBA, Argentina), within which she is performing her doctoral studies. Financial support was provided by two grants: PICT 2015-2540 of the Agencia Nacional de Promoción Científica y Tecnológica (ANPCyT) of the Ministerio de Ciencia, Tecnología e Innovación (MINCyT), Argentina and 03-PIO-23H of the Secretaría de Ciencia, Arte y Tecnología (SECAT), UNCPBA, Argentina. Gabriel Viduzzi was awarded an undergraduate scholarship to promote scientific vocation EVC-CIN 2020 (project 19402) from the Consejo Interuniversitario Nacional (CIN), Argentina.

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