# **TECHNICAL REPORTS**

#### ATMOSPHERIC POLLUTANTS AND TRACE GASES

# Methane and Nitrous Oxide Emissions from Cattle Excreta on an East African Grassland

D. E. Pelster,\* B. Gisore, J. K. Koske, J. Goopy, D. Korir, M. C. Rufino, and K. Butterbach-Bahl

### **Abstract**

Greenhouse gas (GHG) emission measurements from livestock excreta in Africa are limited. We measured CH<sub>4</sub> and N<sub>3</sub>O emissions from excreta of six Boran (Bos indicus) and six Friesian (Bos taurus) steers near Nairobi, Kenya. The steers were fed one of three diets (T1 [chaffed wheat straw], T2 [T1 + Calliandra calothyrsus Meissner - 0.2% live weight per day], and T3 [T1 + calliandra -0.4% live weight every 2 d]). The T1 diet is similar in quality to typical diets in the region. Calliandra is a leguminous fodder tree promoted as a feed supplement. Fresh feces and urine were applied to grasslands and emissions measured using static chambers. Cumulative 28-d fecal emissions were 302  $\pm$  52.4 and  $95 \pm 13.8 \text{ mg CH}_{A}$ –C kg<sup>-1</sup> dry matter for Friesen and Boran steers, respectively, and 11.5  $\pm$  4.26 and 24.7  $\pm$  8.32 mg N<sub>2</sub>O-N kg<sup>-1</sup> dry matter for Friesian and Boran steers, respectively. For urine from Friesian steers, the N<sub>2</sub>O emissions were  $2.8 \pm 0.64$  mg N<sub>2</sub>O-N 100 mL urine $^{-1}$ . The CH $_{_{A}}$  emission factors (EFs) (246  $\pm$  49.5 and  $\bar{8}$ 7  $\pm$  12.7 g CH<sub>a</sub>-C yr<sup>-1</sup> animal<sup>-1</sup> for Friesan and Boran, respectively) were lower than the International Panel on Climate Change EFs (750 g CH<sub>4</sub>-C animal<sup>-1</sup> yr<sup>-1</sup>), whereas the N<sub>2</sub>O EFs (0.1 and 0.2% for the Friesian and Boran feces, respectively, and 1.2% for urine) were also lower than International Panel on Climate Change estimates. The low N content of the excreta likely caused the low emissions and indicates that current models probably overestimate CH<sub>4</sub> and N<sub>2</sub>O emissions from African livestock manure.

#### **Core Ideas**

- GHG emissions from African livestock excreta is lower than IPCC tier 1 emission factors.
- Low-quality feeds with low protein content result in low N content of excreta.
- Supplementation of cattle diet with calliandra reduced the methane emissions from cattle feces.
- The species of cattle causes differences in GHG emissions from feces.

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NCREASED ATMOSPHERIC CONCENTRATIONS of greenhouse gases (GHG) such as carbon dioxide ( $\rm CO_2$ ), methane ( $\rm CH_4$ ), and nitrous oxide ( $\rm N_2O$ ) over the last century are strongly correlated with global warming, and these gas concentrations continue to increase at an annual rate of approximately 0.4, 0.6, and 0.25%, respectively (Myhre et al., 2013). Although agricultural GHG sources account for approximately 12% of total anthropogenic emissions globally (Tubiello et al., 2015), in Africa GHG emissions from agriculture, forestry, and other land use accounts for approximately 61% of emissions (Valentini et al., 2014). Between 1990 and 2000, the agricultural emissions in Africa increased by about 99 MtCO<sub>2</sub>e (18%) (Vergé et al., 2007) and continued to increase by about 2% per year between 2001 and 2011 (Tubiello et al., 2014).

In sub-Saharan Africa, livestock comprise a large proportion of total agricultural emissions, most of which is from enteric CH<sub>4</sub> production in ruminants (Valentini et al., 2014). However, between 7 and 15% of agricultural GHG emissions are associated with livestock manure (Smith et al., 2014; Tubiello et al., 2014). However, these emission rates from livestock manure in Africa are estimated using emission factors (EFs) from the International Panel on Climate Change (IPCC) that have been derived using measurements primarily from states within the Organization for Economic Cooperation and Development. These regions have livestock species, breeds, diets, management systems, and climatic conditions that often differ from those in tropical Africa (IPCC, 2006).

In tropical and subtropical agricultural production systems, the climate is generally warmer than temperate systems, which could result in greater  $\rm N_2O$  and  $\rm CH_4$  emissions from excreta because emissions are often positively correlated with temperature (González-Avalos and Ruiz-Suárez, 2001; Rochette et al., 2014). However, the types of management systems used, the quality of the feeds, and the species of cattle raised may also affect emissions. The majority of African ruminants graze for much of their life (Schlecht et al., 2006), which results in over 40% of excreta

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Abbreviations: DM, dry matter; GHG, greenhouse gas; IPCC, International Panel on Climate Change; LW, live weight.

being deposited on rangelands and pastures (Rufino et al., 2006), much more than in temperate regions. Thus, approximately 80% of the emissions associated with excreta deposited on rangelands and pasture occurs in developing countries (Smith et al., 2014).

The diet of African ruminants tends to be based on grasses and crop residues that are more fibrous than their counterparts from temperate regions, with lower digestibility and protein content (Schlecht et al., 2006). Dietary protein content, feed digestibility, and sugar content are known to influence the amounts and types of N and C voided in cattle excreta (Dijkstra et al., 2011; Merry et al., 2006; Rotz, 2004). Therefore, the lower-quality feeds likely result in excreta with reduced N concentrations and higher C/N ratios. To compensate for the low-protein diet, many agencies across eastern Africa promote the use of the legume fodder tree calliandra (Calliandra calothyrsus Meissner) as a feed supplement (Dawson et al., 2014). However, calliandra has high condensed tannin concentrations that cause higher recalcitrant N concentrations in cattle excreta (Delve et al., 2001). The increased C/N ratio and higher concentrations of recalcitrant N could result in lower N,O emissions from the feces (Chantigny et al., 2013) compared with temperate systems. Also, native African cattle (Bos indicus) tend to have better water-scavenging abilities than their counterparts from temperate regions (Bos taurus), resulting in different fecal dry matter (DM) content (Quarterman et al., 1957). This physical difference in the fresh feces may also affect N<sub>2</sub>O and CH<sub>4</sub> emissions from the excreta.

Agriculture in Kenya accounts for about 33% of the country's GHG emissions, and 90% of those emissions are from livestock (Stiebert, 2012). However, unlike temperate systems, much (roughly 60%) of the ruminant livestock in Kenya are kept by pastoralists in Kenya's arid and semiarid lands (Government of Kenya, 2010). Like most sub-Saharan countries, emission estimates in Kenya are estimated using the IPCC's Tier 1 approach. For development of proper emission reduction and mitigation strategies, actual measurements are necessary (Rosenstock et al., 2013; Rufino et al., 2014). Therefore, our objective was to estimate CH<sub>4</sub> and N<sub>2</sub>O emissions from the excreta of Boran and Friesian steers. Boran cattle are a common native breed, originally from the Borana plateau (Felius, 1995), whereas the Friesian is a European breed that is commonly promoted in eastern Africa to increase dairy production. We hypothesized that (i) there would be no significant differences in emissions from feces and urine between the species, (ii) the protein supplementation with calliandra would increase N2O emissions, and (iii) feeding patterns (i.e., oscillating protein supplementation; daily vs. bi-daily) of calliandra would not affect emissions.

# **Materials and Methods**

The study was performed on grassland at the campus of the International Livestock Research Institute (1°16′14″ S; 36°43′28″ E) having a mixture of kikuyu grass (*Pennisetum clandestinum* Hochst. ex Chiov.) and Rhodes grass (*Chloris gayana* Kunth), with grazing being simulated by cutting the grass approximately every 14 d. The mean annual precipitation for the area is 980 mm, and the mean annual temperature is 17.5°C. The driest month is July (mean precipitation, 15 mm), and the wettest month is April (mean, 219 mm). The warmest month is March (mean temperature, 19.1°C), and the coolest month is

July (mean temperature,  $15.1^{\circ}$ C) (AmbiWeb GmbH, 2015). The soils are well drained, deep humic nitisols (Jaetzold et al., 2006), with clay-textured (16% sand, 65.6% clay) topsoils (0–20 cm), a pH of 7.0, and a bulk density of 0.8 g cm<sup>-3</sup>.

Excreta samples were obtained from Friesian (n = 6; live weight [LW],  $183 \pm 5.8 \text{ kg}$ ) and Boran (n = 6; LW,  $126 \pm 3.4 \text{ kg}$ ) yearling steers forming the experimental cohort for a feeding trial. Experimental design and sampling protocol has been described in detail elsewhere (Korir et al., 2015). Briefly, steers were assigned initially to one of three dietary treatments: T1, ad libitum wheat (Triticum aestivum L.) straw; T2, T1 + daily calliandra supplementation (0.2% of LW); and T3, T1 + bi-daily calliandra supplementation (0.4% LW). The basal diet was similar to feed typically given to east-African cattle during the dry season. Each steer received all treatments in a  $3 \times 3 \times 2$  factorial crossover design (3 treatments by 3 periods by 2 breeds). Animals were housed individually with free access to water. Each treatment period lasted 4 wk. For additional information on the feed, feces, and urine collection and analysis, see Korir et al. (2015). On the final evening of each period, the stalls were cleaned, and the urine collection buckets were emptied and rinsed to remove any residual acid. The following morning (~12 h later), the fresh urine and feces were collected for application.

Two replicates of each excreta sample (i.e., 1.0 kg of fresh manure and 234 mL of urine) were surface applied to grazing plots. The plots were delineated by plastic frames ( $35 \times 25$  cm) inserted 8 cm into the soil. Plots were kept 30 cm apart to prevent cross contamination. The amount of feces and urine was approximately equivalent to an average deposition event as measured during the previous 4-wk period (Korir et al., 2015). There were 56 frames in total (2 blocks, each with 12 feces samples, 12 urine samples, and 4 controls), with treatment randomly assigned to each plot within each block. The frames remained fixed in the soil for 2 wk before and 4 wk after urine and feces application. Two weeks after application, we simulated a 20-mm rainfall.

The manure and urine were applied on the same day as collection. Thus, there were three different application periods, with the application date corresponding to the final date of each feeding period in the feed trial. The application dates were as follows: Period 1, 30 June 2014; Period 2, 28 July 2014; and Period 3, 25 Aug. 2014. Precipitation was measured continuously using an ECRN-100 high-resolution rain gauge. Soil moisture content and temperature were measured at the time of gas sampling using a GS3 soil moisture and temperature sensor (Decagon GS3). The soil was analyzed for pH using a Jenway (Staffordshire) 350 pH meter with an epoxy-bodied combination electrode in a 1:5 soil/ water solution. Soil bulk density was measured by dividing the oven-dried (12 h at 105°C) weight by the volume of the soil cores (5 cm diam., 20 cm length). Soil texture analysis was done at the Kenya Agricultural Research Institute using the hydrometer technique (Sheldrick and Wang, 1993).

# Sample Analysis

Fecal DM was determined by drying in a forced air oven at 105°C. Organic matter content was calculated by subtracting the ash content after total combustion in muffle furnace from the DM. Total N concentration in feed and urine were determined by micro Kjeldhal procedure (see Korir et al. 2015 for more information). The fecal C content was calculated from the

known organic matter using the formula by Jiménez and Pérez García (1992) for compost. Applied C and N were calculated by multiplying the amount of fresh feces applied by the total C or N content (mg C kg FW $^{-1}$  and mg N kg FW $^{-1}$ , respectively). The amount of urine-N applied was calculated by multiplying the volume of urine by the concentration of total Kjeldahl N in each sample.

The CO<sub>2</sub>, N<sub>2</sub>O<sub>3</sub> and CH<sub>4</sub> flux from the excreta and soil were measured using opaque polyvinyl chloride static chambers equipped with fans and vent tubes to allow continuous venting (Rochette and Bertrand, 2008). After the lids were clamped to the frames, a needle was inserted through the sampling port, and 60 mL of the headspace was sampled. The first 40 mL was used to flush out ambient air from an airtight 10-mL glass vial, after which the remaining 20 mL was injected to achieve overpressure of the vial to prevent contamination. Samples were taken immediately on closing of the chambers and again after 9, 18, and 27 min to ensure an adequate and linear response in headspace concentration. Once sampling was completed, the lids were removed. Sampling was done once a day for 2 d before application, once on the day of excreta application, daily for the after 4 d, and then two to three times per week for the subsequent 3 wk. On the application day, sampling occurred in the early afternoon, immediately after application; however, on all other days the sampling occurred in the morning, generally between 8:00 and 11:00 AM.

The gas samples from the chambers were analyzed within 1 wk for  $\rm CO_2$ ,  $\rm CH_{4^1}$  and  $\rm N_2O$  on a SRI 8610C gas chromatograph fitted with a flame ionization detector for  $\rm CO_2$  (after passing through a methanizer) and  $\rm CH_4$  and an electron capture detector for  $\rm N_2O$ . The carrier gas (pure  $\rm N_2$ ) had a flow rate of 20 mL min<sup>-1</sup>. Two calibration gases (800 ppm  $\rm CO_2$ , 7040 ppb  $\rm CH_4$ , and 823 ppb  $\rm N_2O$  and 400 ppm  $\rm CO_2$ , 4000 ppb  $\rm CH_4$ , and 360 ppb  $\rm N_2O$  in synthetic air; Air Liquide) were analyzed alongside the samples, and the relation between the peak areas of the nearest calibration gas and its concentration was used to calculate the concentrations within the samples assuming a linear response in peak area to concentration.

Gas fluxes were estimated by calculating the rate of change in concentration over time using a linear approach while correcting for air pressure and temperature using the ideal gas law. Data were validated by examining the CO, concentrations. In cases where the change in concentration was below the precision of the gas chromatograph, we assumed no flux (i.e., minimum detection limits of 5 μg CH<sub>4</sub>-C m<sup>-2</sup> h<sup>-1</sup>, 0.4 mg CO<sub>2</sub>–C m<sup>-2</sup> h<sup>-1</sup>, and 2  $\mu$ g N<sub>2</sub>O–N m<sup>-2</sup> h<sup>-1</sup>). The cumulative gas flux from the urine or feces application was estimated by summing the daily fluxes (using trapezoidal integration between sampling dates) and then subtracting the mean cumulative emissions from the control plots (i.e., those that received no application). The cumulative flux for the 2 d before excreta application was found to be similar for all plots, suggesting that differences between control plots and treatment plots were caused by the added excreta. Emission factors (the amount of CH<sub>4</sub> or N<sub>2</sub>O emitted per unit of C/N added) for CH<sub>4</sub> and N<sub>2</sub>O were calculated by dividing the amount of C or N lost as either CH<sub>4</sub>-C or N<sub>2</sub>O-N, respectively, by the amount of C or N added.

## **Statistical Analysis**

We used a random mixed model to analyze the feces and urine cumulative flux data (Imer package, R 3.0.3). All data were checked for normality using the Shapiro-Wilk test. We used ANOVA for CH<sub>4</sub> and CO, and Kruskal–Wallis for N<sub>2</sub>O (to account for the non-normal distribution) to compare chamber effects for the preapplication period. For fecal flux analysis, species and diet were used as fixed effects, and period and the animal were fitted as random effects. For urine, only the Friesians' data were analyzed. The Boran data were not collected because the collection caused swelling of the animals' urethra, forcing us to halt the urine collection. Consequently, only diet was used as a fixed effect in the model, with period and animal remaining as random effects. We tested for relations between the amount of C or N applied with the amount of CH<sub>4</sub>, CO<sub>2</sub>, and N<sub>2</sub>O emitted using a Pearson correlation. Annual emissions for a tropical livestock unit (one tropical livestock unit = one 250-kg steer) were calculated by multiplying the mean 1-mo emission (either mg CH<sub>4</sub> 28 d<sup>-1</sup> or mg N<sub>2</sub>O 28 d<sup>-1</sup>) per kg manure by the amount of manure (kg) produced by each animal, adjusting for differences in LW and extrapolating to 1 yr.

For fecal DM, C, and N content, we used a two-factor ANOVA with species and diet as fixed factors. For the urine-N content, we used a single-factor ANOVA with diet as the single, fixed factor. Homoscedasticity was confirmed through visual observations of residual plots.

# Results

The mean LW of the Friesian steers was approximately 50% greater than that of the Boran steers (P < 0.001) (Table 1). The Friesian steers also produced about 0.5 kg d<sup>-1</sup> more feces than the Boran steers (P < 0.001). Steers receiving the bi-daily supplementation produced 1.0 kg more feces (Table 1) than steers on the basal diet (P = 0.02). The Boran steers' feces had a higher DM content than the Friesian steers' feces (P < 0.001) (Table 1), whereas the feces C content was similar among all treatments. Feces from animals fed the basal diet contained 29% less N than both the supplemented diets (P < 0.001) (Table 1). Because we applied the feces on a fresh weight basis, the application rates were 98.6 g C and 2.5 g N from the Friesian steer feces, compared with 118.7 g C and 3.1 g N with the Boran feces. Approximately 1 wk after application, we noticed an influx of termites in some of the plots, and by the end of the second week all of the feces plots had evidence of termites. The urine-N concentration and application rates were similar among the different diets (Table 2)

Cumulative CH<sub>4</sub> emissions from the urine plots were similar to control plots (i.e., plots with nothing added) (P=0.42) (Table 3), indicating, as expected, that urine application did not affect CH<sub>4</sub> emissions. However, application of feces did cause greater CH<sub>4</sub> emissions compared with the controls (P=0.001). The differences between control and application was mainly due to a short-term pulse of CH<sub>4</sub> immediately after application. During this pulse, the CH<sub>4</sub> emission rates increased from around 20 to over  $600 \, \mu \mathrm{g \ m^{-2} \ h^{-1}}$  and then decreased exponentially to near background within about 1 wk (Fig. 1). Cumulative CH<sub>4</sub> fluxes (Table 3) differed by breed and diet, with the feces of the Friesian steers releasing more CH<sub>4</sub> than feces from the Boran steers (P=0.032) and with both the diets with calliandra

supplementation resulting in higher feces  $\mathrm{CH_4}$  emissions than the basal diet (P=0.03 and 0.01 for bi-daily and daily supplementation diets, respectively). The interaction between diet and breed had no detectable effect on the  $\mathrm{CH_4}$  emissions from the feces application (P=0.56). The presence of termites within the chambers, which was generally noticed 1 to 2 wk after application, did not cause a noticeable increase in  $\mathrm{CH_4}$  emissions.

Cumulative 28-d CO<sub>2</sub> fluxes from the urine plots were approximately 34.0 g m<sup>-2</sup> greater than emissions from the control plots (P=0.002), whereas feces application resulted in 42.5 g m<sup>-2</sup> more CO<sub>2</sub> than the control plots (P=0.02) over the 28-d monitoring period (Table 3). Neither the diet nor the species affected CO<sub>2</sub> emissions from the feces and the urine applications (P>0.10 for all). The CO<sub>2</sub> emissions from both the urine and feces applications increased immediately after application and then returned to baseline values after about 7 d (Fig. 1 and 2). Addition of water (or rainfall) increased emissions; however,

emissions in the control plots also increased to a similar degree (Fig. 1 and 2), suggesting that this increase was related to the rewetting rather than the excreta.

Both the feces and urine application resulted in greater cumulative  $\rm N_2O$  emissions over the 28-d period than the controls (P=0.007 and 0.013 for feces and urine application, respectively) (Table 3). However, even though the amount of N applied as urine was approximately 25% of the amount of N applied as feces (Tables 1 and 2), the  $\rm N_2O$  emissions from the urine were higher than emissions from feces, likely because the N in urine is more available than the N in feces. In general, the applications did not cause immediate increases in  $\rm N_2O$  emissions; however, the addition of water 2 wk after the application caused a spike in emissions, particularly in Period 3 (Fig. 1 and 2). The  $\rm N_2O$  emission rates were highest during the Period 3, when emission rates in the urine plots exceeded 250  $\rm \mu g~m^{-2}~h^{-1}$  and emissions from the feces plots exceeded 200  $\rm \mu g~m^{-2}~h^{-1}$ . This spike in emissions

Table 1. Animal live weight, mean daily deposition, percent dry matter (DM), percent carbon (dry matter basis), total Kjedahl nitrogen (dry matter basis) content, and C/N ratio of 1 kg of fresh cattle manure added to grassland at the campus of the International Livestock Research Institute, Nairobi, Kenya (n = 34).

Туре	Diet	Live weight	Deposition	DM	Total C	Kjeldahl N	C/N ratio
		kg	kg DM d⁻¹	%	——— g kg <sup>-1</sup> FW ———		
Friesian	Basal diet (wheat straw only)	181.5 ± 10.3†	$1.11 \pm 0.03$	20.6 ± 0.41a‡	97.6 ± 2.23a	1.72 ± 0.026a	56.9 ± 1.64a
	Daily supplement (calliandra) + basal diet	185.5 ± 10.9	$1.39 \pm 0.05$	$20.3 \pm 0.58a$	96.5 ± 2.41a	2.56 ± 0.181b	38.5 ± 2.33b
	Bi-daily supplement (calliandra) + basal diet	181.7 ± 11.0	$1.47 \pm 0.05$	$20.4 \pm 0.57a$	96.8 ± 2.79a	2.46 ± 0.121b	39.7 ± 2.15b
	Mean	$183.0 \pm 5.8$	$1.31 \pm 0.03$	$20.4 \pm 0.28$	$97.0 \pm 1.33$	$2.23 \pm 0.118$	45.3 ± 2.41
Boran	Basal diet (wheat straw only)	$126.9 \pm 5.4$	$0.76 \pm 0.04$	22.7 ± 1.73b	107.2 ± 8.68b	1.98 ± 0.106a	53.8 ± 2.27a
	Daily supplement (calliandra) + basal diet	$121.8 \pm 7.7$	$0.93 \pm 0.07$	$25.7 \pm 0.65$ b	121.1 ± 2.41b	$3.00 \pm 0.267$ b	41.5 ± 3.39b
	Bi-daily supplement (calliandra) + basal diet	$128.4 \pm 6.0$	$0.95 \pm 0.02$	$25.9 \pm 0.75$ b	122.8 ± 2.79b	$3.08 \pm 0.215$ b	40.6 ± 2.20b
	Mean	125.7 ± 3.5	$0.88 \pm 0.03$	24.7 ± 0.75	116.8 ± 3.59	2.67 ± 0.167	45.5 ± 2.07

 $<sup>\</sup>dagger$  Values are mean  $\pm$  SEM.

Table 2. Urine nitrogen concentration and amount of nitrogen applied from the application of 234 mL of fresh cattle urine to grassland at the campus of the International Livestock Research Institute, Nairobi, Kenya (n = 15).

Туре	Diet	Urine N concentration	Urine N application rate		
		g L <sup>-1</sup>	mg N plot⁻¹		
Friesian	Basal diet (wheat straw only)	$2.59 \pm 0.201 \dagger$	$605 \pm 47.1$		
	Daily supplement (calliandra) + basal diet	$2.60 \pm 0.136$	609 ± 31.9		
	Bi-daily supplement (calliandra) + basal diet	$2.66 \pm 0.173$	$622 \pm 40.5$		

<sup>†</sup> Values are mean  $\pm$  SEM.

Table 3. Cumulative  $CH_{a'}$   $CO_{2'}$  and  $N_2O$  emissions over 28 d from the application of 1 kg of fresh cattle manure or 234 mL of fresh urine to grassland at the campus of the International Livestock Research Institute, Nairobi Kenya.

	Diet	Emissions†					
Туре		Dung application			Urine application		
		CH₄-C	CO <sub>2</sub> -C	N <sub>2</sub> O-N	CH₄-C	CO <sub>2</sub> -C	N <sub>2</sub> O-N
		mg CH₄−C kg manure⁻¹	g CO <sub>2</sub> –C kg manure <sup>–1</sup>	mg N <sub>2</sub> O–N kg manure <sup>–1</sup>	mg CH <sub>4</sub> –C 234 mL urine <sup>-1</sup>	g CO <sub>2</sub> -C 234 mL urine <sup>-1</sup>	$mg N_2O-N$ 234 mL urine <sup>-1</sup>
Friesian	basal diet (wheat straw only)	34.4 ± 9.7ab‡	$40.5 \pm 5.3$	1.92 ± 1.19	$-10.4 \pm 4.29$	$36.2 \pm 6.18$	$8.4 \pm 2.09$
	daily supplement (calliandra) + basal diet	75.2 ± 15.5c	$42.7 \pm 5.4$	$4.01 \pm 1.85$	$-5.5 \pm 4.24$	$35.5 \pm 7.25$	$7.3 \pm 2.98$
	bi-daily supplement (calliandra) + basal diet	71.9 ± 24.7c	$31.3 \pm 9.4$	0.93 ± 1.18	$-6.3 \pm 6.39$	30.3 ± 12.32	$4.0 \pm 1.18$
Boran	basal diet (wheat straw only)	11.6 ± 3.9a	$43.1 \pm 7.0$	$7.04 \pm 4.55$	ND§	ND	ND
	daily supplement (calliandra) + basal diet	$24.6 \pm 3.8b$	$46.4 \pm 8.2$	$7.82 \pm 2.74$	ND	ND	ND
	bi-daily supplement (calliandra) + basal diet	$34.7 \pm 7.8b$	$53.0 \pm 8.5$	2.94 ± 1.11	ND	ND	ND

<sup>†</sup> Emissions shown were calculated by subtracting the emissions from the control (no addition) plots from the total emissions from plots with the added feces; mean cumulative (28 d) emissions from control plots:  $12.0 \pm 5.54$  mg  $CH_a-C$  m<sup>-2</sup>,  $99.0 \pm 6.28$  g  $CO_2-C$  m<sup>-2</sup>, and  $5.1 \pm 1.56$  mg  $N_2O-N$  m<sup>-2</sup> (n=34).

<sup>‡</sup> Different letters indicate significant differences within columns.

<sup>‡</sup> Values are mean ± SEM. Different letters indicate significant differences within columns.

<sup>§</sup> ND, not determined.

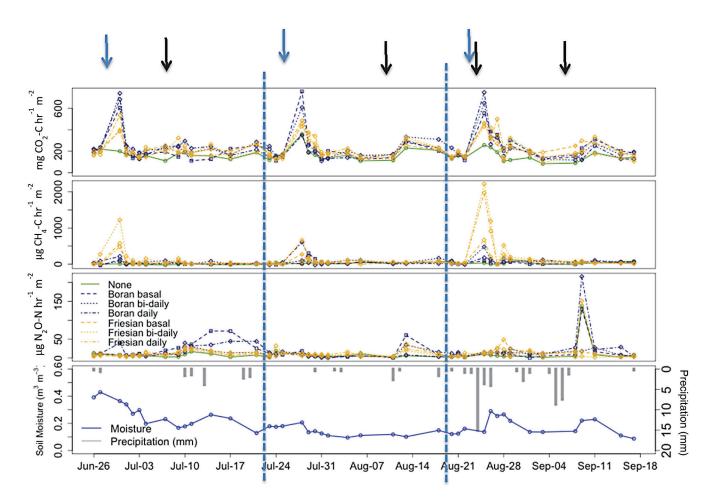


Fig. 1. Mean  $CO_2$  (mg  $CO_2$ –C  $m^{-2}$   $h^{-1}$ ),  $CH_4$  ( $\mu$ g  $CH_4$ –C  $m^{-2}$   $h^{-1}$ ), and  $N_2O$  ( $\mu$ g  $N_2O$ –N  $m^{-2}$   $h^{-1}$ ) flux rates from 1 kg fresh feces applications to grassland, along with soil moisture (0.05 m depth) and precipitation. Blue arrows indicate application of excreta, black arrows indicate water application, and blue dashed lines differentiate three application periods. The different treatments are as follows: basal, basal diet of chaffed wheat straw; bi-daily, basal diet + bi-daily supplement of calliandra (2% live weight); Borana, feces from a borana steer (*Bos indicus*); daily, basal diet + daily supplement of calliandra (1% LW); Friesian, feces from a Friesian steer (*Bos taurus*); none, no feces application.

coincided with a rain event that deposited over 22 mm of rain over 3 d and a corresponding increase in soil moisture from 0.20 to 0.26% v/v (Fig. 2). For the feces application, the  $\rm N_2O$  emissions were greater from the Boran than from the Friesian steers (P=0.09) (Table 3), whereas diet had no effect on  $\rm N_2O$  emissions (P=0.19). The urine application increased  $\rm N_2O$  emissions compared with the controls; however, diet had no detectable effect on emissions (P=0.37). The interaction between diet and breed had no detectable effect on  $\rm N_2O$  emissions from the feces applications (P=0.77).

There were no correlations between the C applied and  $CO_2$  emissions (P=0.34) or between the N applied and  $N_2O$  emissions (P=0.86). However, there was a significant negative correlation between the C applied and cumulative  $CH_4$  emissions (P=0.008; R=-0.32). However, the range of applied C and N was very narrow (see Table 1), and the correlations could change if a wider range is used.

# **Discussion**

The amount of feces voided is determined by feed intake and digestibility. In turn, feed intake is related mainly to LW (CSIRO, 2007). Although all the steers were yearlings, the Friesian steers were heavier than the Borans (Table 1), which was reflected in differences in feed intake and the amount of feces

voided. Breeds of *Bos indicus* adapted to harsh climatic conditions, such as Borans, have higher water reabsorption efficiencies compared with *Bos taurus* breeds like Friesians (Quarterman et al., 1957), which likely caused the higher DM content in the Boran feces. Because we applied manure on a fresh-weight basis, the higher DM content in the Boran feces resulted in more C being added to those plots. However, even with the greater amount of C applied, the CH<sub>4</sub> emissions from the Boran feces were less (per kg of fresh feces) than the emissions from the Friesian feces. Because most of the CH<sub>4</sub> was produced very rapidly after application, it is likely that the higher moisture content of the Friesian feces formed a more favorable environment for CH<sub>4</sub> production (Lodman et al., 1993) for a longer period of time, allowing for additional methanogenesis (Conrad, 1996).

With the influx of termites, we expected to see a corresponding increase in  $\mathrm{CH_4}$  emissions. However, this did not occur, suggesting that the termites we saw produced little to no  $\mathrm{CH_4}$  or that they transferred the feces to another location, producing  $\mathrm{CH_4}$  somewhere else. Further study to properly evaluate the role of termites in fecal  $\mathrm{CH_4}$  emissions is required.

The greater amounts of feces deposited by the Friesians (Table 1) would produce cumulative CH<sub>4</sub> fluxes that were approximately five times greater per steer than cumulative CH<sub>4</sub> fluxes from the feces of a Boran steer. Even when accounting for

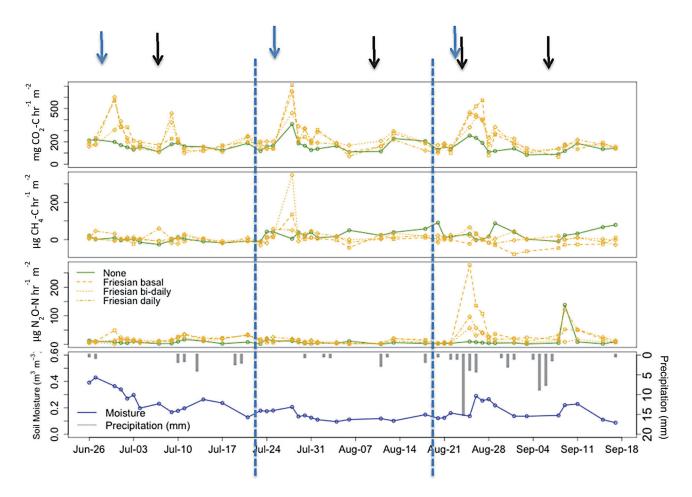


Fig. 2. Mean CO $_2$  (mg CO $_2$ –C m $^{-2}$  h $^{-1}$ ), CH $_4$  ( $\mu$ g CH $_4$ –C m $^{-2}$  h $^{-1}$ ), and N $_2$ O ( $\mu$ g N $_2$ O–N m $^{-2}$  h $^{-1}$ ) flux rates from 234-mL urine applications to grassland along with soil moisture and precipitation. Blue arrows indicate application of urine, black arrows indicate water application, and blue dashed lines differentiate the three periods. The different treatments are as follows: basal, basal diet of chaffed wheat straw; bi-daily, basal diet + bi-daily supplement of calliandra (2% live weight); daily, basal diet + daily supplement of calliandra (1% live weight); Friesian, urine from a Friesian steer (Bos taurus); none, no feces application.

differences in LW, fecal  $\mathrm{CH_4}$  emissions (per kg LW) from the Friesian steers were still 3.5 times higher than from the Boran steers. The different water contents of the feces therefore appear to result in different emission potentials as well. Although it is known that feces properties differ between the two species, to our knowledge this is the first study to report differences in  $\mathrm{CH_4}$  emissions between feces from *Bos indicus* and *Bos taurus*.

The measured  $\mathrm{CH_4}$  emissions, which ranged from 11.6 to 75.2 mg  $\mathrm{CH_4-C}$  kg feces over a 28-d period (Table 3), were much lower than those in a previous study (Jarvis et al., 1995), which found that  $\mathrm{CH_4}$  losses from 1 kg of feces in the United Kingdom ranged from 300 to 2040 mg  $\mathrm{CH_4-C}$  kg feces over a 10- to 15-d period. However,  $\mathrm{CH_4}$  losses measured in our study were much more consistent with two other UK studies that measured  $\mathrm{CH_4}$  losses of between 20 and 90 mg  $\mathrm{CH_4-C}$  kg feces over study periods of between 15 and 60 d (Holter, 1997; Yamulki et al., 1999) as well as a study in Brazil that measured emission rates between 10 and 60 mg  $\mathrm{CH_4-C}$  kg feces over 30 d (Mazzetto et al., 2014).

Dietary composition is known to affect the N concentration, soluble organic C, readily fermentable carbohydrates (Boadi et al., 2004), and C/N ratio of feces, which consequently affects CH $_4$  emissions (Cardenas et al., 2007). The steers used in this study were fed a basal diet with a low crude protein content (2%),

which is known to increase N use efficiency, reduce the excreta N concentrations, and increase the excreta C/N ratio (Korir et al., 2015). The C/N ratio of the feces was much higher than previous studies (e.g., Jarvis et al., 1995; Qian and Schoenau, 2002; Yamulki et al., 1999) where the ratio typically ranged between 10 and 15 and is likely why these emissions were so low when compared with those from Jarvis et al. (1995). However, these results were consistent with Mazzetto et al. (2014), who also used low-quality tropical fodders. Fecal CH $_4$  emissions were found to be negatively correlated to the C/N ratio of the feces (Table 1), consistent with previous studies (Jarvis et al., 1995). The C/N ratios in our study ranged from 38 to 54, all of which were greater than the C/N ratios (13–28) found in Jarvis et al. (1995), indicating that the effect of C/N ratio on CH $_4$  emissions extends across a much wider range than previously thought.

Assuming that emission rates remain the same throughout the rest of the year and that the deposition rates and feces properties remain consistent throughout the year, the annual fecal CH $_4$  emissions per tropical livestock unit would be approximately 117  $\pm$  17.0 g CH $_4$  yr $^{-1}$  animal $^{-1}$  for the Boran and 328  $\pm$  68.1 g CH $_4$  yr $^{-1}$  animal $^{-1}$  for the Friesan breeds (after adjustment for differences in LW). These results are between 9 and 25% of what the IPCC uses as the CH $_4$  emission factor for feces from African cattle (IPCC, 2006). However, emission

rates, deposition rates, and feces properties are likely to change during the different seasons, and it is important to measure the effects of these seasonal changes as well. Even though the increased temperature typical of the tropics should increase emissions compared with temperate regions, it appears that the effect of the lower-quality fodder and consequently the higher C/N ratio of the feces has a stronger effect on controlling emissions in these systems.

Because the feed trial took place during the dry season, we were limited in our supply of fresh manure and were only able to measure emissions during the dry season. We did add 20 mm of precipitation 2 wk after application to mimic a rain event, which resulted in an increase in emission rates. This, along with changes to fodder quality and therefore manure quality as well, suggests that the emission factors may change if measurements are made throughout the year rather than during just one season. A previous study found higher emissions during the "summer," when temperatures were higher than during a cooler "winter" period in Brazil (Mazzetto et al., 2014). Also, deposition rates and feces properties are not constant throughout the year (Rufino et al., 2006; Schlecht et al., 2006). Given that the ad libitum diet with additional protein supplementation was a better quality diet than many African cattle receive during the dry season, it is likely that annual field emissions are lower than what we suggest here, although additional investigation is required to verify these assumptions. Excreta emissions on pasturelands, however, are typically very low in comparison to enteric CH<sub>4</sub> emissions (Jarvis et al., 1995; Mazzetto et al., 2014), although they are still important for emission reporting (e.g., IPCC, 2006; Tubiello et al., 2015).

The increase in  $\rm CO_2$  from the feces application was expected because we were adding approximately between 97 and 117 g C to the plots. However, there was very little C applied with the urine application, which also resulted in a similar increase in  $\rm CO_2$  emissions. The addition of the urea-N likely caused increased soil organic matter decomposition, also known as priming (Kuzyakov et al., 2000), which resulted in the large increase in  $\rm CO_2$  emissions.

Urine application increases N<sub>2</sub>O emissions compared with control plots (Table 3) likely because the greater N availability provided additional substrate for denitrification. The lack of rain during the first two periods may have limited denitrification; however, the precipitation at the start of the third period likely mobilized additional C and N substrate for denitrification (Birch, 1960; Ruser et al., 2006) while providing anaerobic sites. These peaks in N2O emissions tend to be the result of denitrification, which requires the correct air-water balance, and the degree of gas diffusivity (Balaine et al., 2013; Butterbach-Bahl et al., 2013; Singurindy et al., 2009). Period 3 emissions were twice as high as Period 1 emissions and at least eight times higher than Period 2 emissions (Fig. 2). Because the amounts of applied N applied were almost equal, it seems that the greater precipitation received in period 3 (45.6 mm) compared with Period 1 (11.6 mm) and Period 2 (6.4 mm; see Fig. 2) caused the higher flux in Period 3. The burst of N<sub>2</sub>O after a precipitation event and urine/feces additions to grassland has been observed in other trials as well (Boon et al.. 2014; de Klein et al., 2003; Yamulki et al., 1999).

Opposite to the CH<sub>4</sub> flux, the feces of Borans produced more N<sub>2</sub>O than that of the Friesians. It is possible that the

higher moisture content of the Friesian feces favored further reduction of  $\rm N_2O$  to  $\rm N_2$ . Calliandra addition to the diet was expected to increase both fecal N concentration and  $\rm N_2O$  emissions. However, although supplementation resulted in greater N excreted in the feces, it did not cause measurable differences in  $\rm N_2O$  emissions. However, we measured only total C and N, whereas it is the labile portion of both that are substrates for denitrification. The high tannin content of calliandra (3.2% for this study) (Korir et al., 2015) has been found to result in greater concentrations of recalcitrant N in the feces (Delve et al., 2001) that may have limited N availability for denitrifiers.

The higher  $N_2O$  emissions from urine compared with feces were likely because the N in urine is more easily available (Sordi et al., 2014). This is also suggested by the delay in  $N_2O$  emissions from the feces application after the simulated rainfall (Fig. 1 and 2), which did not occur in the urine applications. It is likely that the fecal N needed to be mineralized before denitrification could occur. In addition, the high amounts of C in the feces and the high C/N ratio likely caused rapid N immobilization, resulting in less available  $NO_3$  and further reduction of  $N_2O$  to  $N_2$  (Blackmer and Bremner, 1978; Senbayram et al., 2012).

The EFs for the feces in this study (0.2% from the Boran and 0.1% from Friesians) and from the urine (1.2% from the Friesian only) were lower than the 2% estimated by the IPCC (2006), although they were similar to a Brazilian study that also measured low N<sub>2</sub>O emissions from cattle excreta on tropical rangelands (Mazzetto et al., 2015). Because fecal C/N ratios were found to be negatively correlated with N mineralization (Qian and Schoenau, 2002) and N<sub>2</sub>O emissions (Chantigny et al., 2013), it is likely that the high C/N ratio of the feces in this tropical system was responsible for the low emissions.

However, because our measurements took place during the dry season, it is possible that emissions during the rainy season may be higher than what was measured here. Although it is likely that the increased water availability during the rainy season may cause increased GHG emissions, the improvement in fodder and therefore manure quality (Rufino et al., 2006; Schlecht et al., 2006) will likely have a larger effect on emission rates. With the exception of Period 3, there was only a small increase in N<sub>2</sub>O emissions with a simulated 20-mm rainfall event, suggesting that the lack of rainfall alone may not be causing the low EF. Rather, the low N<sub>2</sub>O emissions are more likely related to the low N concentration and high C/N ratio typical of tropical cattle manure (Rufino et al., 2006), which was related to the poor-quality fodder (Korir et al., 2015).

#### **Conclusions**

As indicated, the IPCC (2006) EF are at least two times higher for fecal CH<sub>4</sub>, 10 to 20 times higher for fecal N<sub>2</sub>O, and about two times higher for urine N<sub>2</sub>O. The low EFs in this study were likely due to the poor-quality diet (low crude protein) and the subsequent low excreta N. Also, the native cattle breeds (i.e., Bos indicus) have better water retention than imported breeds, which caused decreased CH<sub>4</sub> but increased N<sub>2</sub>O emissions. The diets used in this study were consistent with those frequently used in smallholder farms in the region and similar in digestible energy to the low-quality fodder category used by the IPCC to estimate livestock emissions, suggesting that emission factors

used for GHG inventories in this region may need to be revised. However, additional studies performed under different climatic seasons, linked with measurements of enteric fermentation and with measurements performed over extended periods, are required to verify these findings.

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