



Effect of feeding practices and manure quality on CH₄ and N₂O emissions from uncovered cattle manure heaps in Kenya



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ABSTRACT

Countries in sub-Saharan Africa (SSA) rely on IPCC emission factors (EF) for GHG emission reporting. However, these were derived for industrialized livestock farms and do not represent conditions of smallholder farms (small, low-producing livestock breeds, poor feed quality, feed scarcity). Here, we present the first measurements of CH₄ and N₂O emissions from cattle-manure heaps representing feeding practices typical for smallholder farms in the highlands of East Africa: 1) cattle fed below maintenance energy requirements to represent feed scarcity, and 2) cattle fed tropical forage grasses (Napier, Rhodes, Brachiaria). Sub-maintenance feeding reduced cumulative manure N₂O emissions compared to cattle receiving sufficient feed but did not change EF_{N₂O}. Sub-maintenance feeding did not affect cumulative manure CH₄ emissions or EF_{CH₄}. When cattle were fed tropical forage grasses, cumulative manure N₂O emissions did not differ between diets, but manure EF_{N₂O} from Brachiaria and Rhodes diets were lower than the IPCC EF_{N₂O} for solid storage (1%, 2019 Refinement of IPCC Guidelines). Manure CH₄ emissions were lower in the Rhodes grass diet than when feeding Napier or Brachiaria, and manure EF_{CH₄} from all three grasses were lower than the IPCC default (4.4 g CH₄ kg⁻¹ VS, 2019 Refinement of IPCC Guidelines). Regression analysis revealed that manure N concentration and C:N were important drivers of N₂O emissions, with low N concentrations and high C:N reducing N₂O emissions. Our results show that IPCC EFs overestimate excreta GHG emissions, which calls for additional measurements to develop localized EFs for smallholder livestock systems in SSA.

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1. Introduction

Agriculture is responsible for ca. 9–14% of anthropogenic greenhouse gas (GHG) emissions globally, and without mitigation, these emissions could further increase (IPCC, 2015; Rosenzweig et al., 2020). The African continent is estimated to contribute 15% to global agricultural GHG emissions, a large share of which originates from livestock production systems (Tubiello et al., 2014). Globally, the livestock sector emits ca. 5.6–7.5 Gt CO₂-eq yr⁻¹ (considering the livestock supply chain, Herrero et al., 2016), most of which is attributed to enteric methane (CH₄) from ruminants (33%), while CH₄ and nitrous oxide (N₂O) emissions from manure management

account for another 10% of global agricultural emissions (Herrero et al., 2016; Owen and Silver, 2015; Steinfeld et al., 2006). Accurate GHG emission estimates from livestock production systems and their components (animals, manure, feed crops and forages) remain scarce for sub-Saharan Africa (SSA) due to a lack of *in situ* studies (Butterbach-Bahl et al., 2020; Goopy et al., 2018; Pelster et al., 2016; Zhu et al., 2018), but they are essential for national GHG reporting and for narrowing uncertainties regarding GHG emissions from agricultural sources. To reduce this knowledge gap, we here aim at quantifying GHG emissions from manure management in smallholder livestock production systems in the East African highlands.

Livestock production is essential for livelihoods (income and employment) of millions of people in SSA, where 45–80% of livestock production occurs in smallholder systems (Herrero et al.,

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2013a). Furthermore, livestock-derived food is critical to battle malnutrition, especially in children (Alonso et al., 2019; Frelat et al., 2016; Herrero et al., 2013b). With the growing population in SSA, the demand for livestock products is increasing (Herrero and Thornton, 2013). As a consequence, Africa is the continent with the highest annual growth rates of livestock numbers globally (2.3%, FAO, 2020). A follow-up effect is that the amount of livestock manure and subsequent GHG emissions will also rise.

Livestock manure is a source of methane (CH₄) and nitrous oxide (N₂O), two potent GHGs with a 100-year global-warming potential (GWP₁₀₀) 34 and 298 times more powerful than that of CO₂ (calculated on a per-mass basis and including climate-carbon feedbacks, IPCC, 2013, p. 714). Methane is produced by methanogenic archaea under anaerobic conditions (Conrad, 2009; Le Mer and Roger, 2001), for example when manure is stored in large manure piles that are not turned and contain a moist core (Amon et al., 2001). If oxygen (O₂) is available, for example in the outer crust of a manure pile, CH₄ is oxidized by methanotrophic bacteria, reducing net manure CH₄ emissions from manure heaps (Petersen et al., 2005). In addition to moisture and O₂ availability, CH₄ formation and consumption are controlled by temperature, with higher temperature promoting CH₄ production (Chadwick, 2005). Furthermore, the substrate C:N ratio affects CH₄ production from manure: in anaerobic digesters, methanogenesis was shown to be most effective at C:N ratios around 25–30 (Wang et al., 2012), whereas lower C:N ratios can lead to NH₄⁺ toxicity and suppress methanogenesis (Rajagopal et al., 2013; Yenigün and Demirel, 2013). Furthermore, high C:N ratios were shown to increase the diversity of methanogenic microbial communities in manure (Zhou et al., 2016). Nitrous oxide in manure is produced primarily via nitrification and denitrification (Chadwick et al., 2011). Therefore, N₂O emissions are usually highest under moist but not water-saturated conditions, when both aerobic (for nitrification) and anaerobic (for denitrification) microsites prevail (Butterbach-Bahl et al., 2013), while maintaining primarily anaerobic conditions during manure storage – for example via compaction or covering – was shown to reduce manure N₂O emissions (Chadwick, 2005). In addition to O₂ availability, N₂O emissions are controlled by the supply of N-containing substrates (Wallenstein et al., 2014) and the manure C:N ratio. Higher C:N ratios reduce N₂O emissions (Yamulki, 2006) because microorganisms compete for N resources, which leads to N immobilization and leaves less N available for nitrifying and denitrifying organisms (Chantigny et al., 2013).

While the processes leading to CH₄ and N₂O emissions from manure management are reasonably well understood and have been reviewed elsewhere (Chadwick et al., 2011; Petersen et al., 2013), the magnitude and temporal dynamics of these emissions are highly variable due to multiple drivers (e.g. climate, C and N availability, manure management system). While several studies have measured GHG emissions associated with manure management in developed countries, data from SSA remains scarce and is either limited to manure applications to pastures (Pelster et al., 2016; Zhu et al., 2018, 2020), or does not capture emissions from the entire heap due to the methodology (Ngwabie et al., 2019; Predotova et al., 2010). However, GHG fluxes are highly variable in space and time because manure heaps on smallholder farms are usually a heterogeneous mixture of manure of different age and/or animal species, often mixed with other materials such as bedding, crop residues, and kitchen waste (Ndambi et al., 2019). Therefore, to accurately measure GHG emissions from solid storage, the entire heap should be covered by the flux chamber, and measurements need to be done frequently and for the entire duration of the storage.

Given the lack of reliable GHG emission data for manure storage in SSA there are high uncertainties in national GHG inventories

from African countries, which mostly rely on existing emission factors that were predominantly derived in very different livestock systems (Rufino et al., 2014). There are two main races of cattle in SSA: *Bos indicus* (cattle with humps) including the Boran, Sahiwal and Zebu cows (also called “indigenous” or “local” breeds), and *Bos taurus* (“exotic” or “imported” breeds) such as Holstein-Friesian, as well as hybrids between the two. At least 150 indigenous African cattle breeds have been described, but most African cattle remain largely uncharacterized (Mwai et al., 2015). Most indigenous cattle in SSA, including the Boran, are related to the Zebu cattle (Belemsaga et al., 2005). The Boran originate from Southern Ethiopia, Northern Kenya and Somalia, where they were presumably developed 1000 years ago. Boran and other Zebu cattle are adapted to hot climates and are today the most common indigenous cattle breeds in Africa (Kim et al., 2017; Mwacharo et al., 2006; Taye et al., 2017). Similar to other indigenous breeds, Boran are smaller (adults 400–500 kg) and less productive than high-yielding cattle breeds of developed countries, which are adapted to cooler climates and can reach up to 1000 kg. In addition, while cattle in developed countries are fed *ad libitum* on high-quality feeds and concentrates rich in N and micronutrients to maximize production, cattle in SSA largely rely on grazing, crop residues, and tropical forage grasses such as Napier (“Elephant grass”, *Pennisetum purpureum*) (Angassa and Beyene, 2003; Goopy et al., 2018; Mumba et al., 2018). While tropical grasses are not necessarily of poor-quality, the lack of pasture management can reduce feed quality, as pastures are often not fertilized and/or harvested after reaching maturity, resulting in high fiber and low protein content of the grass (Lukuyu et al., 2011; Muia et al., 2001). In addition, during some periods of the year (e.g. dry season) feed is scarce and cattle cannot meet their energy requirements, which reduces animal growth and thus production, and even leads to animal weight loss (Goopy et al., 2020; Ndung’u et al., 2018).

All of the above factors (cattle breed, feed quality and quantity) affect the chemical composition of cattle manure (Lupwayi et al., 2000; Sørensen et al., 2003). Nitrogen intake through feed determines the ratio at which N is excreted via either dung (partly and undigested feed and microbial biomass, mostly organic N) or urine (metabolized N, mostly urea and NH₄⁺) (Chadwick et al., 2018; Pereira et al., 2012). When cattle are fed a low-protein diet, a larger share of N is excreted via dung (fecal-N), whereas the share of N excreted via urine (urinary-N) decreases. Given the poor feeding conditions, the mean fecal-N:urinary-N ratio of cattle in SSA is 66:34 (Rufino et al., 2006), compared to ratios ranging from 50:50 to 25:75 in industrialized countries (Chadwick et al., 2018; van der Weerden et al., 2011). This, in turn, likely affects the rate of N₂O formation in the manure because organic N from dung first needs to be broken down before it is converted to N₂O, whereas urine-N has been shown to promote manure N₂O emissions rapidly after excretion (Zhu et al., 2018). It is therefore critical to obtain *in situ* measurements of manure GHG emissions representing local conditions to reduce uncertainty and improve national and regional GHG inventories in SSA (Rufino et al., 2014).

The specific aim of this study was to investigate how different animal diets affect GHG emissions from uncovered manure heaps as typically found in East Africa and elsewhere. To this end, we measured GHG emissions from manure collected during two animal feeding trials using native Boran cattle. The first trial compared manure CH₄ and N₂O emissions from animals fed below their maintenance energy requirements (MER, i.e. “hungry cows”) to animals fed at maintenance levels, while the second trial compared manure emissions from cattle fed different forage grasses. In both trials, the manure was stored as solid manure heaps; representing a common storage practice in SSA smallholder farming systems (Lekasi et al., 2003; Ndambi et al., 2019). We hypothesized (i) that

manure with low N concentrations and wide C:N ratios from cattle fed below MER and/or receiving low-protein tropical forage grasses would lead to reduced N₂O emissions compared to manure with high N concentration and narrow C:N, (ii) that low-N manure would result in N₂O emission factors (EF_{N₂O}, % manure-N emitted as N₂O-N) that are below the IPCC Tier 1 default EF_{N₂O} for solid manure storage, (iii) that manure with wide C:N ratios would lead to increased CH₄ emissions compared to manure with high N concentrations and a narrow C:N.

2. Material and methods

2.1. Site

The two experiments were conducted at the Mazingira Centre for Environmental Research and Education, hosted by the International Livestock Research Institute in Nairobi, Kenya (1°16'15.2" S 36°43'25.2" E, altitude 1860 m above sea level, mazingira.ilri.org). Nairobi has a subtropical highland climate (Cwb on the Köppen-Geiger Classification) (Climate-data.org, 2019). Mean annual temperature (MAT) is 19.0 °C and mean annual precipitation (MAP) is 869 mm, with 90% of the rainfall occurring in the two rainy seasons (long rains from Apr-Jun, short rains Nov-Dec).

2.2. Experimental setup

Animal manure (dung mixed with urine) was collected from two different animal feeding trials, which were conducted at the Mazingira Centre in 2016 (trial 1 – Sub-maintenance energy) and 2018 (trial 2 – Tropical forage grasses). During both trials, individual animals were housed in separate open pens (1.90 m × 2.87 m) with concrete floors and shaded with sailcloth roofing. Each pen had clean drinking water supplied from automatic water dispensers. Manure was collected from the pens in the early mornings, before animals rose to avoid cross-contamination or trampling of the manure. Each morning, manure was scraped from the ground, weighed, and heaped into concrete incubation chambers ([Supplementary Fig. S1](#) and description further below) until reaching a total of 100 kg fresh weight (FW) per heap. The base area of the fresh heap was 0.8 m × 0.7 m with a height of 0.7 m and a surface area of ca. 1.2 m². Similar manure heap sizes have been reported for smallholder farms in Ethiopia ([Minase et al., 2015](#)) and Niger ([Predotova et al., 2010](#)). This process to fill all manure incubation chambers took ca. 10 days. Manure from individual animals was not mixed but placed into separate incubation chambers, with each manure heap representing one replicate (n = 3). The incubation chambers were not shaded or covered from rainfall to represent a common manure management practice on mixed crop-livestock smallholder farms in SSA, where manure is stored in uncovered solid heaps ([Ndambi et al., 2019](#)). Manure incubation was conducted for 145 days, a storage period commonly found in SSA ([Ndambi et al., 2019](#); [Tittone et al., 2010](#)). Temperature inside the heaps was measured using temperature loggers (HOBO Pendant Temperature Logger, Onset, Australia) buried at ca. 45 cm depth. Moisture content of the manure heap was not measured. Precipitation and air temperature were measured with a climate station at the lab facility (Decagon Em50, METEK, Germany).

2.3. Trial 1 – Sub-maintenance energy

The sub-maintenance animal feeding trial (trial 1) is described in detail elsewhere ([Ali et al., 2019](#); [Goopy et al., 2020](#); [Wassie et al., 2019](#)). In brief, Boran yearling steers with a liveweight (LW) of 162.3 ± 3.8 kg were housed in the above-mentioned indi-

vidual pens. For the feeding experiment, a 4 × 4 latin square cross-over design with four levels of feeding at 100%, 80%, 60%, and 40% animal MER with a total sample size of 12 animals per treatment was employed. For the manure incubation experiment described here, manure was collected from the 100%, 80% and 40% MER feeding treatments from three animals per treatment, resulting in a total of nine manure heaps for this trial.

Animal diets consisted of locally-sourced Rhodes grass hay (*Chloris gayana* cv. Boma, DM = 875 g kg⁻¹, crude protein (CP) = 7.3% DM) for all treatments, plus a supplement for the 100% MER treatment to reach required energy levels (10% cotton seed meal [DM = 947 g kg⁻¹, CP = 4.6% DM] and 10% molasses [DM = 728 g kg⁻¹, CP = 4.6% DM]). Feeding period was 21 d of adaptation to the diets ([Machado et al., 2016](#)) plus 10 d during which manure was collected for the incubation. Animals were fed twice per day, in the mornings and early afternoons, via individual feeding troughs. To determine fecal-N:urinary-N ratio ([Wassie et al., 2019](#)), on six consecutive days total daily fecal production from individual animals was determined by collecting all feces manually from the floor as soon as steers defecated. Total daily urine was collected via harnesses fitted to the animals, from which urine was directed into individual closed buckets containing 100 ml 20% sulfuric acid to prevent volatile ammonia loss. All animal procedures followed international standards for animal care and scientific use, reviewed by the Institutional Animal Use and Care Committee (IACUC) of ILRI, permit No. IACUC-RC2016-11.

2.4. Trial 2 – Tropical forage grasses

The tropical forage grass animal feeding trial (trial 2) is described in detail elsewhere ([Korir et al., in prep.](#)). In brief, Boran yearling steers (LW = of 216.3 ± 5.8 kg) were housed in the same individual pens as described above for the sub-maintenance trial. For the feeding experiment, a 6 × 4 Latin square cross-over design was employed with six levels of feeding with either Napier grass (*Pennisetum purpureum* cv. *Kakamega 1*), Brachiaria grass (*Brachiaria brizantha* cv. *xaraes*), or Rhodes grass (*Gloris gayana* cv. *Boma*) with or without legume intercropping (*Lablab purpureus*), with a total sample size of six animals per treatment. For the manure incubation experiment described here, manure was collected from the Brachiaria-only, Napier-only and Rhodes-only feeding treatments from three animals per treatment, resulting in a total of nine manure heaps from this trial. Animals were fed *ad libitum* on a diet consisting of either pure fresh Brachiaria grass (DM = 206 g kg⁻¹, CP = 8.3% DM), Napier grass (DM = 142 g kg⁻¹, CP = 9.6% DM), or Rhodes grass (DM = 233 g kg⁻¹, CP = 8.0% DM). Feeding period was 14 d of adaptation to the diets plus 10 d during which manure was collected for the incubation. Fecal-N:urinary-N ratio was determined via total collection of feces and urine as described for trial 1. Animals were fed three times per day, in the mornings, at noon and in the early afternoons, via individual feeding troughs. All animal procedures followed international standards for animal care and scientific use (permit No. IACUC-RC2017-15).

2.5. Methane and nitrous oxide concentration measurements and flux calculation

To determine GHG fluxes of the entire manure heaps, large non-flow-through non-steady-state gas flux chambers were constructed ([Supplementary Fig. S1](#)). Each manure incubation chamber consisted of two parts, a bottom part made of concrete (inner dimensions 0.8 m × 0.7 m × 0.3 m) with a drainage at the center, and a headspace chamber constructed from a PVC box (0.9 m × 0.8 m × 0.8 m). The wall of the concrete chamber was 15 cm thick and had a rain gutter inserted at its top at all four sides. Upon

chamber deployment, the rain gutter was filled with water and the PVC box was inserted into the rain gutter and submerged under water to provide an air-tight seal (see [Supplementary Fig. S1](#)). Each chamber lid was equipped with two handles, an electric fan (10 cm × 10 cm) for headspace air mixing, a rubber air-pressure equilibration tube (length 0.5 m, inner diameter 6 mm), a thermometer port, and a rubber septum for gas sample collection. For gas flux determination, chambers were closed, and 40 ml of headspace air sample were drawn at 0, 4, 8, 12 and 16 min and filled into pre-evacuated 20 ml gas vials to give a slight overpressure. Gas samples were collected daily for the first two months of incubation, then three times a week for the remaining four months until the end of the experiment. Additional gas samplings were done on days with rainfall. Samples were analyzed within one week on an SRI 8610C gas chromatograph equipped with two HayeSep D packed columns (3 m length, 1/8 in. diameter), one of which led to an electron-capture detector (ECD, temperature 350 °C) for N₂O detection, and the other one to a flame-ionization detector (FID, temperature 350 °C) for CH₄ detection (all parts supplied by SRI Instruments Europe GmbH, Bad Honnef, Germany). Carrier gas was pure N₂ at both FID and ECD lines at a flow rate of 25 ml min⁻¹, and the column oven temperature was set to 70 °C. For calibration, mixtures of N₂O and CH₄ in synthetic air were used at four concentrations ranging from 0.4–2.5 ppm for N₂O and 4–50 ppm for CH₄ (Air Liquide Middle East & Africa). Calibration gas was injected in quadruplicates for the lowest concentration and duplicates for the other three concentrations every 20 to 30 samples, and the relationship between peak areas and gas concentrations of calibration gases was used to calculate the gas concentrations of samples. Gas flux rates were calculated using the ideal gas law (Equ. 1),

$$\text{GHG flux} = \frac{d\text{Conc}}{dt} * \frac{P}{1013} * \frac{273}{T + 273} * \frac{MM}{22.41} * \frac{V}{DM} * 60 \quad (1)$$

using the change of gas concentration over time $d\text{Conc}/dt$ (ppm min⁻¹), air pressure P at the sampling location (hPa) corrected for air pressure at sea level (1013 hPa), chamber headspace temperature T (°C), atomic mass MM of the target compound (12 g mol⁻¹ for CH₄-C, 28 g mol⁻¹ for N₂O-N since there are two atoms of N in one molecule of N₂O), the ideal gas volume (22.41 L mol⁻¹), total volume of chamber lid plus base V (L) divided by manure heap dry matter at t_0 DM (kg) and multiplied by 60 to convert from minutes to hours, giving flux units of mg N₂O-N kg⁻¹ DM h⁻¹ and mg CH₄-C kg⁻¹ DM h⁻¹. For gas flux data cleaning, the R² of $d\text{Conc}/dt$ (linear slope) was calculated, and all fluxes with R² < 0.8 were discarded. This resulted in discarding of 3% of CH₄ fluxes and 11% of N₂O flux measurements.

Cumulative gas fluxes were calculated over the entire duration of the experiment (including days before the heap reached 100 kg FW to capture CH₄ emissions that are high in freshly excreted manure) by filling measurement gaps with a weighted running mean (gap window ±5 days, linear weighting), then multiplying the hourly flux rates of each day with 24 to derive the daily gas flux, and finally summing all daily gas fluxes to get the total cumulative flux (mg N₂O-N kg⁻¹ DM and g CH₄-C kg⁻¹ DM) over the experimental period (155 days). Emission factors for N₂O and CH₄ were calculated according to the 2019 Revision of the IPCC Guidelines and are given in units of g CH₄ kg⁻¹ volatile solid (VS = manure DM corrected for ash content) and % manure-N emitted as N₂O-N (IPCC, 2019).

2.6. Manure chemical composition

Manure chemistry was determined in fresh manure immediately after collection from the animal holding pens. Manure dry matter content was determined after drying at 105 °C until con-

stant weight. For manure organic C and total N concentration, additional samples were dried at 60 °C until constant weight, ground using a hammer mill (Retsch MM 400 mixer mill, Haan, Germany), and then analyzed on an automated elemental combustion analyzer (VarioMAX Cube, Elementar, Germany). Manure mass loss could not be measured in this experiment.

2.7. Statistical analysis

Effects of diet on manure chemical composition, cumulative GHG emissions and GHG EFs were evaluated using a one-way ANOVA (linear mixed effects model with diet as fixed factor and chamber as random effect) followed by Tukey's HSD *post hoc* test. One-sided *t*-test was employed to test if EFs were smaller than IPCC default values. Data were tested for normality using Shapiro-Wilk test and for homogeneity of variance using Bartlett's test, and data were log-transformed if necessary. To detect effects of manure chemistry on manure GHG emissions, Pearson's correlation analysis was conducted across the combined dataset of both trials. All statistical tests were performed using R version 3.5.3 (www.R-project.org, packages *stats*, *nlme*, *multcomp*, *Hmisc*, and *corrplot*).

3. Results

3.1. Manure chemical composition

Results of all ANOVA statistics are shown in [Supplementary Table 1](#). Differences between diets are only reported if they were statistically significant unless explicitly stated otherwise. Manure from the sub-maintenance feeding trial (trial 1) did not differ in DM or C content, but manure from cattle fed at 40% MER had a significantly lower N content (1.11 ± 0.08% DM) and a wider C:N ratio (35.6 ± 3.0) than animals that were fed at 100% MER (N content 1.78 ± 0.04% DM, C:N = 22.3 ± 0.6), with the 80% MER treatment in between the other two ([Table 1](#)). Furthermore, the fecal-N:urinary-N ratio decreased when cattle were fed at 80% (63:37) or 40% MER (52:48), indicating that relatively less N was excreted via feces than urine compared to animals fed at 100% MER (69:31). Ash content was higher in manure from cattle fed at 80% MER (18.8 ± 2.3% DM) and 40% MER (20.0 ± 3.5% DM) compared to 100% MER (15.0 ± 2.3% DM). Internal manure heap temperature ranged from 19.7 to 31.1 °C (mean 23.1 ± 1.8 °C) and did not differ between treatments ([Supplementary Fig. S2](#)). Animal and manure production for the sub-maintenance feeding trial are presented elsewhere ([Ali et al., 2021, 2019; Goopy et al., 2019; Wassie et al., 2019](#)).

In the tropical forage grass feeding trial (trial 2), manure from animals fed with Napier grass had a lower C content (38.2 ± 0.2% DM) than when animals were fed with Rhodes (41.2 ± 0.1% DM) or Brachiaria (40.8 ± 0.1% DM), while N content was lower in manure from cattle fed with Rhodes (1.04 ± 0.02% DM) compared to Brachiaria (1.21 ± 0.04% DM) and Napier (1.20 ± 0.05% DM). Manure from cattle fed with Brachiaria had a narrower C:N ratio (34.9 ± 0.2) than when animals were fed with Rhodes (39.1 ± 0.7) or Napier (39.6 ± 0.3). Dry matter content did not differ between treatments. The fecal-N:urinary-N ratio did not differ between treatments and was on average 55:45. Ash content was slightly lower in manure from the Napier diet (16.3 ± 0.5% DM) compared to Brachiaria (17.8 ± 0.2% DM) and Rhodes (21.4 ± 1.3% DM). Internal temperature in the heap ranged from 16.6 to 25.3 °C (mean 21.1 ± 1.6 °C) and did not differ between treatments ([Supplementary Fig. S2](#)).

Table 1

Chemical composition of manure from two cattle feeding trials: In the sub-maintenance feeding trial, cattle were fed a diet supplying either 100%, 80%, or 40% of the animal's metabolic energy requirement (MER); in the tropical forage grass feeding trial, cattle were fed either with Brachiaria, Rhodes or Napier grass. Lowercase letters denote differences between diets (one-way ANOVA, $p < 0.05$). Data are means \pm SE (n = 3).

	Manure C content (% DM)	Manure N content (% DM)	Manure C:N	Manure dry matter (% FM)	Manure ash content (% DM)*	Fecal-N: urinary-N ratio*
<i>Sub-maintenance feeding trial</i>						
100% MER	39.6 \pm 0.3	1.78 \pm 0.04 ^b	22.3 \pm 0.6 ^a	22.7 \pm 0.9	15.0 \pm 2.3 ^a	69:31 ^c
80% MER	38.9 \pm 0.2	1.35 \pm 0.16 ^{ab}	30.2 \pm 3.7 ^{ab}	21.3 \pm 0.7	18.8 \pm 2.3 ^b	63:37 ^b
40% MER	38.7 \pm 0.8	1.11 \pm 0.08 ^a	35.6 \pm 3.0 ^b	22.6 \pm 0.8	20.0 \pm 3.5 ^b	52:48 ^a
<i>Tropical forage grass feeding trial</i>						
Brachiaria	40.8 \pm 0.1 ^b	1.21 \pm 0.04 ^b	34.9 \pm 0.2 ^a	18.0 \pm 1.2	17.8 \pm 0.2 ^b	53:47
Rhodes	41.2 \pm 0.1 ^b	1.04 \pm 0.02 ^a	39.1 \pm 0.7 ^b	19.1 \pm 1.2	21.4 \pm 1.3 ^b	61:39
Napier	38.2 \pm 0.2 ^a	1.20 \pm 0.05 ^b	39.6 \pm 0.3 ^b	15.4 \pm 1.4	16.3 \pm 0.6 ^a	51:49

* from Wassie et al. (Wassie et al., 2019) for the sub-maintenance feeding trial, and Korir et al. (in prep.) for the tropical forage grass feeding trial.

3.2. Manure CH₄ and N₂O emissions

In the sub-maintenance feeding trial, CH₄ emissions were high in fresh manure immediately after collecting and piling in the heap (Fig. 1), with maximum CH₄ flux rates ranging from 3.51 \pm 1.10 mg CH₄-C kg⁻¹ DM h⁻¹ (40% MER) to 7.63 \pm 4.11 mg CH₄-C kg⁻¹ DM h⁻¹ (100% MER). Within three weeks, CH₄ emissions

from all treatments decreased to flux rates ranging from 0.016 \pm 0.015 to 1.36 \pm 0.29 mg CH₄-C kg⁻¹ DM h⁻¹ and stayed in that range over the remaining duration of the experiment. In contrast, N₂O flux rates were low immediately after collection (Fig. 1), ranging from 0.005 \pm 0.08 (100% MER) to 0.084 \pm 0.049 mg N₂O-N kg⁻¹ DM h⁻¹ (80% MER) in the first week after heaping. Maximum N₂O flux rates ranging from 0.062 \pm 0.031 mg N₂O-N kg⁻¹ DM h⁻¹ (40%

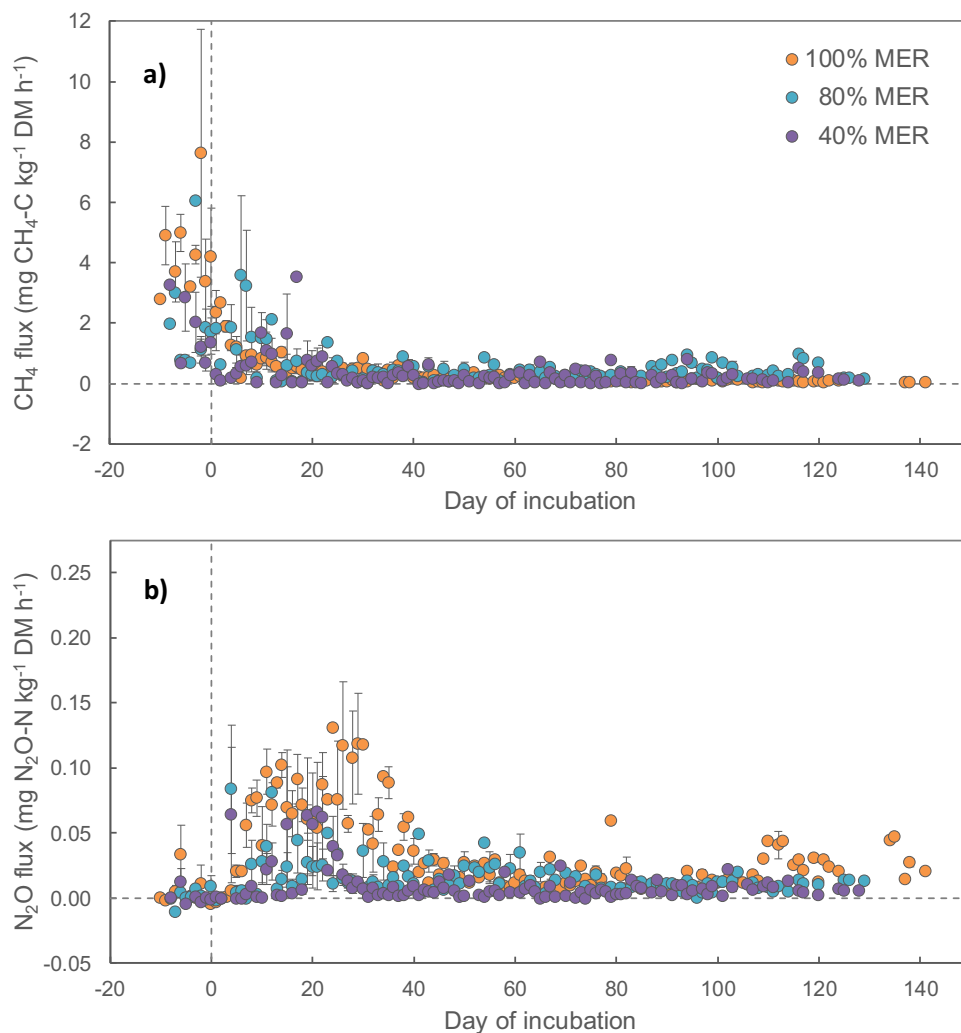


Fig. 1. Flux rates of methane (panel a) and nitrous oxide emissions (panel b) from solid manure heaps of cattle fed a diet meeting 100%, 80% or 60% of the animals' maintenance energy requirement (MER). The x-axis shows Day of incubation of the manure heap, with zero denoting the day on which the manure heap reached 100 kg fresh weight. Data points are means \pm SE (n = 3).

MER) to 0.117 ± 0.049 mg N₂O–N kg⁻¹ DM h⁻¹ (100% MER) were observed from day seven until day 40. After that, N₂O flux rates decreased again to the same range as before the emission peak, apart from days after rainfall (Supplementary Fig. S3), where flux rates showed small increases (up to 0.041 ± 0.010 mg N₂O–N kg⁻¹ DM h⁻¹) in the 100% MER treatment only. Total rainfall was 144 mm while the mean air temperature was 16.7 ± 1.5 °C (range 13.3 to 19.9 °C) during the experiment (Supplementary Fig. S3).

Cumulative CH₄ fluxes in the sub-maintenance feeding trial did not differ between diets (Fig. 2), with a mean flux of 1.80 ± 0.23 g CH₄–C kg⁻¹ DM. Similarly, EF_{CH₄} did not differ between treatments, with a mean value of 2.93 ± 0.37 g CH₄ kg⁻¹ VS, and they were all similar to the IPCC default value for solid storage of manure from non-dairy cattle in low productivity systems in tropical montane climates, which is 4.4 g CH₄ kg⁻¹ VS (IPCC, 2019, p. 10.64). Cumulative N₂O emissions (Fig. 2) of the two sub-maintenance diets (mean of 41.3 ± 8.6 mg N₂O–N kg⁻¹ DM) were lower than emissions of the 100% MER diet (103.2 ± 16.4 mg N₂O–N kg⁻¹ DM). EF_{N₂O}, however, did not differ between diets (mean value of $0.41 \pm 0.08\%$ manure–N emitted as N₂O–N), but the EF_{N₂O} from all diets were significantly lower than the IPCC default value

for direct N₂O emissions from solid storage, which is 1% (IPCC, 2019, p. 10.100).

In the forage grass feeding trial, patterns of CH₄ and N₂O emissions were similar to those observed in the sub-maintenance trial: CH₄ emissions were highest in the first four weeks of incubation (Fig. 3), with maximum flux rates ranging from 1.62 ± 0.76 mg CH₄–C kg⁻¹ DM h⁻¹ (Rhodes) to 4.22 ± 1.21 mg CH₄–C kg⁻¹ DM h⁻¹ (Napier). After that, CH₄ emissions decreased in all treatments to flux rates ranging from 0.012 ± 0.009 to 0.91 ± 0.14 mg CH₄–C kg⁻¹ h⁻¹ DM and remained in that range until the end of the measurements. Nitrous oxide emissions were low in fresh manure but increased after approx. one week (Fig. 3), with maximum N₂O flux rates ranging from 0.122 ± 0.050 mg N₂O–N kg⁻¹ DM h⁻¹ (Brachiaria) to 0.150 ± 0.020 mg N₂O–N kg⁻¹ DM h⁻¹ (Napier) between day eight and day 35. After that, N₂O emissions decreased again to flux rates close to zero, except for days after rainfall events (Supplementary Fig. S3), where emissions increased to fluxes up to 0.045 ± 0.016 mg N₂O–N kg⁻¹ DM h⁻¹. Total rainfall was 377 mm, while the mean air temperature was 16.9 ± 1.7 °C (range 13.2 to 19.9 °C) during the experiment (Supplementary Fig. S3).

Cumulative CH₄ emissions in the forage grass feeding trial (Fig. 4) were lower in manure from cattle fed with Rhodes grass

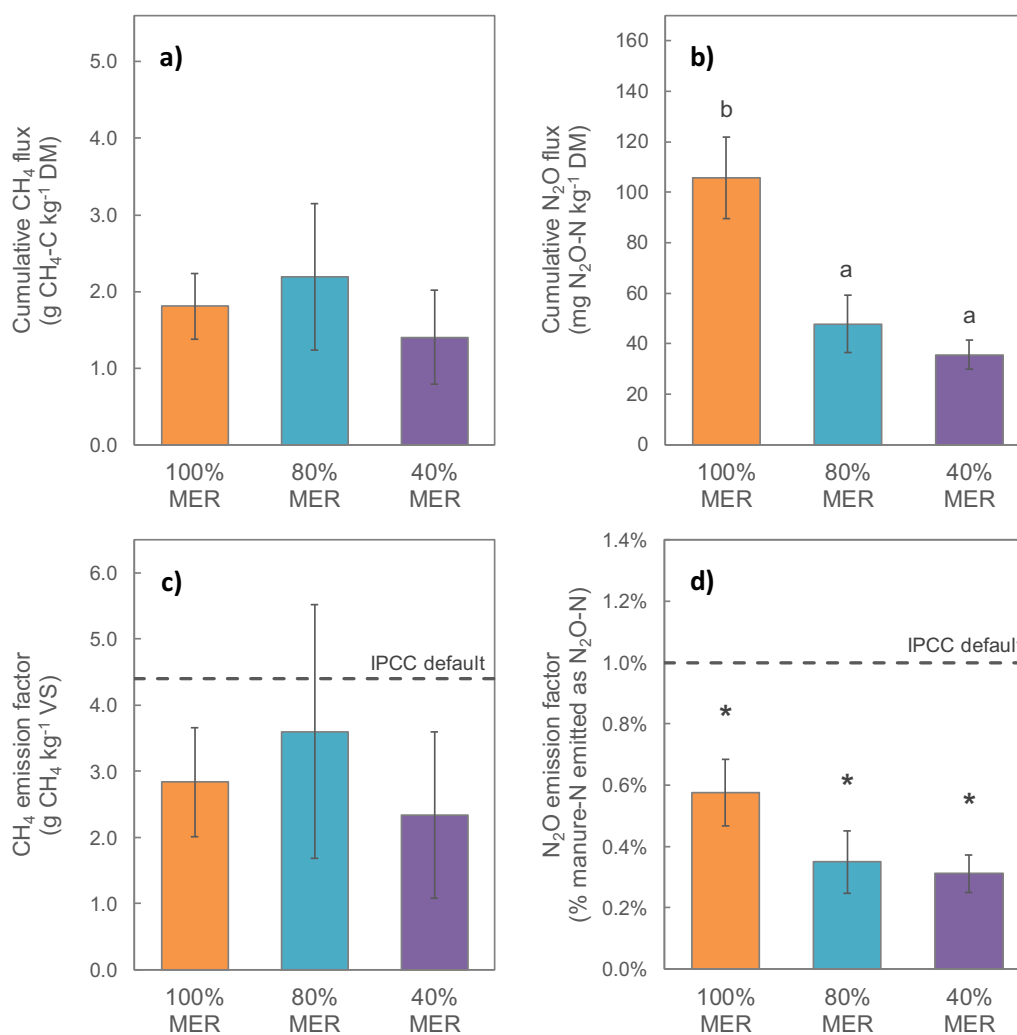


Fig. 2. Upper row: cumulative emissions from manure heaps for methane (panel a) and nitrous oxide (panel b) for cattle diets meeting either 100%, 80%, or 40% animal maintenance energy requirement (MER). Bottom row: emission factors (EF) for methane (panel c) and nitrous oxide (panel d). Lowercase letters denote significant differences between diets (one-way ANOVA, $p < 0.05$), asterisks denote significant differences (one-sided t -test, $p < 0.05$) between measured values and IPCC default values (2019 Refinement) for solid manure storage (dashed lines, IPCC default EF_{CH₄} = 4.4 g CH₄ kg⁻¹ VS, IPCC default EF_{N₂O} = 1.0% manure–N emitted as N₂O–N). Bars represent means \pm SE ($n = 3$).

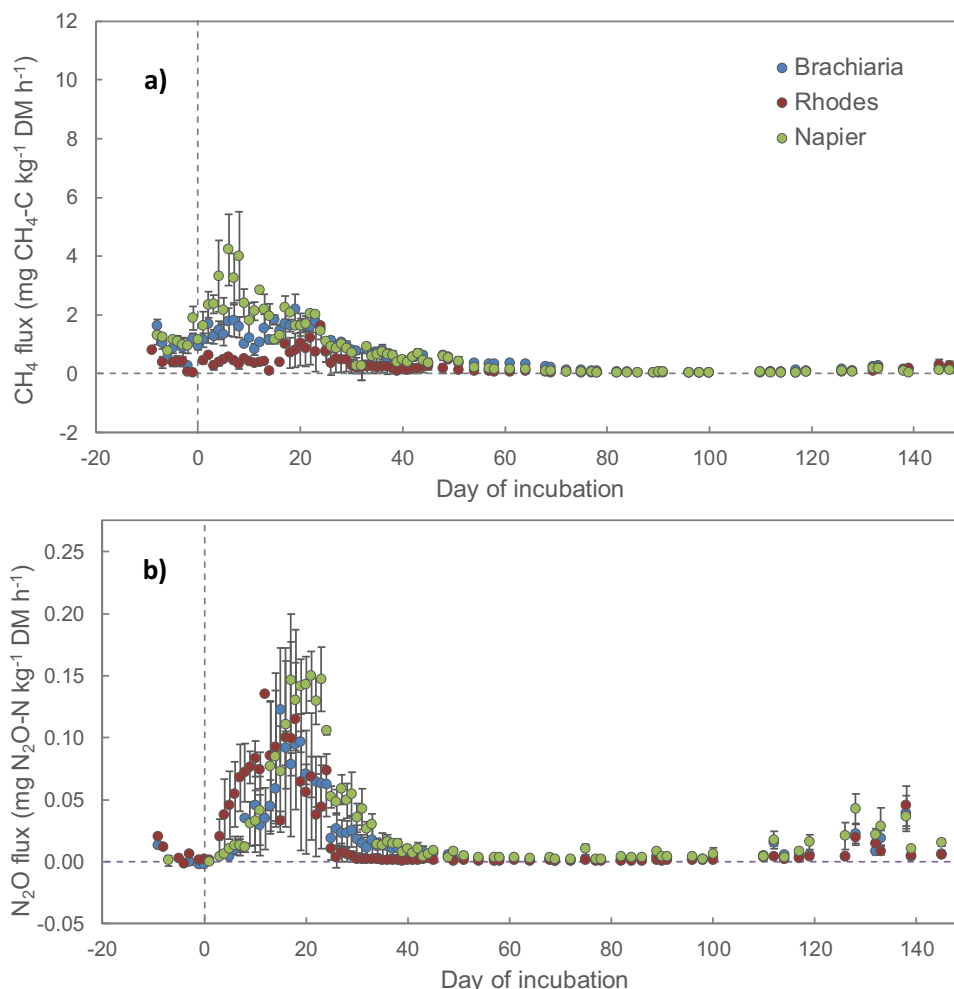


Fig. 3. Flux rates of methane (panel a) and nitrous oxide (panel b) emissions from manure heaps of cattle fed a diet of either Brachiaria grass, Rhodes grass, or Napier grass. The x-axis shows Day of incubation of the manure heap, with zero denoting the day on which the manure heap reached 100 kg fresh weight. Data points are means \pm SE ($n = 3$).

(0.55 ± 0.34 g CH₄-C kg⁻¹ DM) compared to the other two tropical grasses (mean of 1.72 ± 0.15 g CH₄-C kg⁻¹ DM). EF_{CH_4} was also lower in manure from the Rhodes-grass diet (0.94 ± 0.57 g CH₄-kg⁻¹ VS) compared to the other two diets (mean of 2.77 ± 0.26 g CH₄ kg⁻¹ VS), and EF_{CH_4} from all three grass diets were significantly lower than the IPCC default value (2019 Refinement) of 4.4 g CH₄-kg⁻¹ VS. Cumulative N₂O emissions as well as EF_{N_2O} did not differ between grass diets (Fig. 4). However, EF_{N_2O} of the Brachiaria diet ($0.38 \pm 0.004\%$) and the Rhodes diet ($0.44 \pm 0.13\%$) were significantly lower than the IPCC default value (2019 Refinement) of 1.0%.

3.3. Correlations between GHG emissions and manure chemistry

Results from the correlation analysis are shown in Supplementary Table 2. Across both trials, manure N played a crucial role in controlling manure N₂O emissions: cumulative N₂O emissions were positively correlated with manure N concentration ($r = 0.574$, $p = 0.013$) and with the fecal-N:urinary-N ratio ($r = 0.498$, $p = 0.036$), and negatively correlated with the manure C:N ratio ($r = -0.533$, $p = 0.023$). EF_{N_2O} was not correlated with any of the measured manure chemistry parameters. Manure C:N ratio was negatively correlated with manure N concentration ($r = -0.960$, $p < 0.001$) and with the fecal-N:urinary-N ratio ($r = -0.621$, $p = 0.006$), and positively correlated with manure C concentration ($r = 0.501$, $p = 0.034$). Manure CH₄ emissions were

not correlated with any of the measured manure chemistry parameters. In addition, cumulative CH₄ and N₂O emissions as well as EF_{CH_4} and EF_{N_2O} were positively correlated with each other (cumulative emissions, $r = 0.507$, $p = 0.032$; EFs, $r = 0.502$, $p < 0.034$).

4. Discussion

The aim of the present study was to generate local GHG emission data from solid manure storage that consider small local cattle breeds as well as feeding conditions of poor quality and insufficient quantity often experienced by livestock in East Africa. Manure heap N₂O flux rates in our study ranged from 0.03 to 0.15 mg N₂O-N kg⁻¹ DM h⁻¹. This is in line with other studies measuring emissions over solid cattle manure heaps, for example Parkinson et al. (2004) who measured flux rates of 1–14 g N₂O-N t⁻¹ DM d⁻¹, which corresponds to 0.04–0.58 mg N₂O-N kg⁻¹ DM h⁻¹, or Chadwick (2005) who measured 0.00–0.35 mg N₂O-N kg⁻¹ DM h⁻¹. Peak N₂O flux rates in our study were 25% of the flux rates reported in Parkinson et al. (2004) and 43% of the flux rates reported in Chadwick (2005), probably due to narrower C:N ratios in these studies (C:N = 23.8 in Chadwick, 2005; C:N = 20.6 in Parkinson et al., 2004) compared to ours (mean C:N = 33.6 \pm 6.5). This in turn might be explained by the different cattle breeds (local indigenous compared to high-producing cattle) and lower feed quality in our study. As hypothesized (hypothesis i), feed scarcity

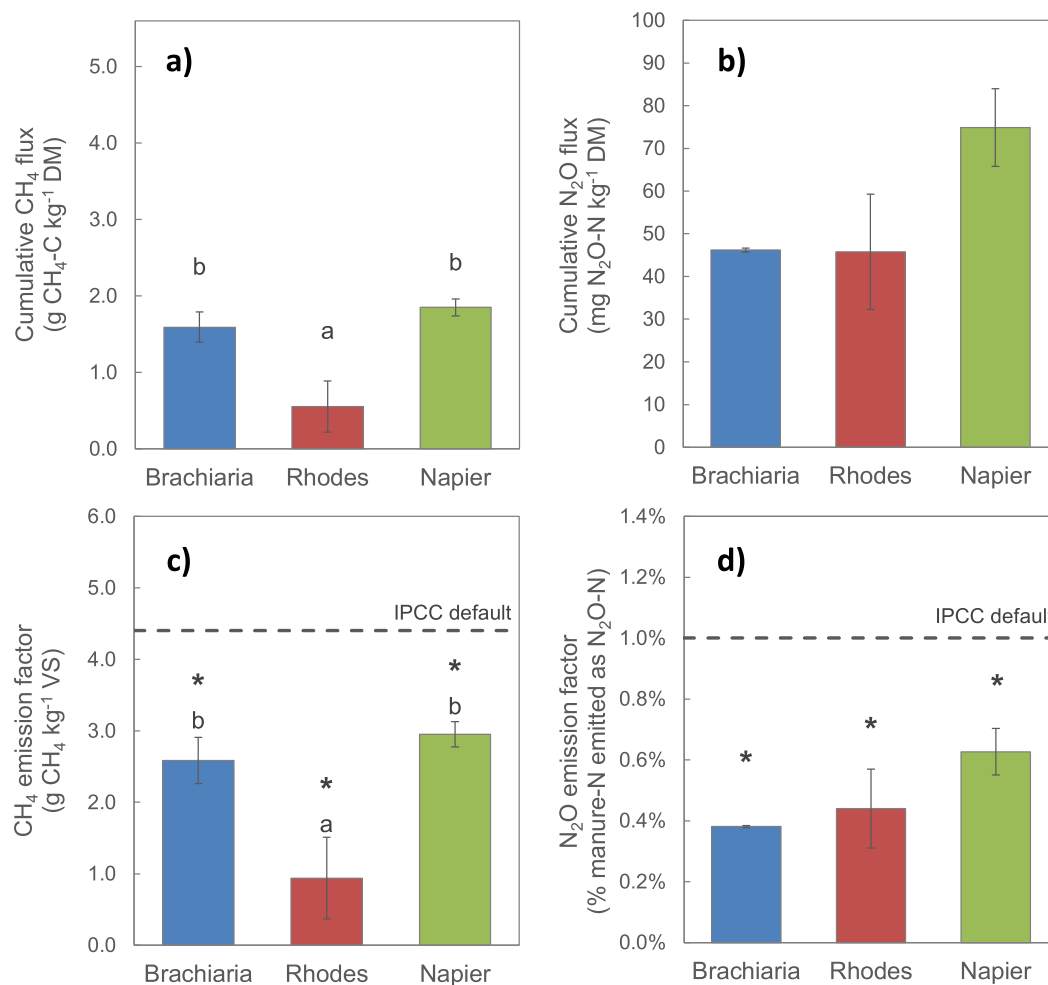


Fig. 4. Upper row: cumulative emissions from manure heaps for methane (panel a) and nitrous oxide (panel b) of cattle fed with either Brachiaria grass, Rhodes grass, or Napier grass. Bottom row: emission factors (EF) for methane (panel c) and nitrous oxide (panel d). Lowercase letters denote significant differences between diets (one-way ANOVA, $p < 0.05$); asterisks denote significant differences (one-sided t -test, $p < 0.05$) between measured values and IPCC default values (2019 Refinement) for solid manure storage (dashed lines, IPCC default $EF_{CH_4} = 4.4 \text{ g CH}_4 \text{ kg}^{-1} \text{ VS}$, IPCC default $EF_{N_2O} = 1.0\% \text{ manure-N emitted as N}_2\text{O-N}$). Bars represent means \pm SE ($n = 3$).

reduced manure N concentrations which then led to lower cumulative N₂O emissions from manure heaps in the two sub-maintenance diets (40% and 80% MER) compared to the 100% MER treatment. This was supported by a positive correlation between manure N concentration and N₂O emissions across all diets. Effects of cattle diet on manure GHG emissions have been reported previously (e.g. Dijkstra et al., 2013; Külling et al., 2008; Mathot et al., 2012), yet to our knowledge this is the first study to directly link feed scarcity and manure quality to CH₄ and N₂O emissions from manure heaps in SSA.

In contrast to cumulative N₂O emissions, the EF_{N_2O} expressed as percentage of excreted N was not affected by feed scarcity in the present study, indicating that the manure N in the heaps was as likely to be converted to N₂O in cattle fed below or at maintenance levels. This was surprising because feed limitation and low N intake usually reduce the fraction of N that is metabolized by the animals and then excreted via urine (Dijkstra et al., 2013; Külling et al., 2008). Urine-N, however, has been shown to be more effective in promoting N₂O emissions than fecal-N in tropical pastures (Chadwick et al., 2018; López-Aizpún et al., 2020; Pelster et al., 2016; Zhu et al., 2020). Therefore, a decrease in urinary-N and a shift towards fecal-N would be expected to reduce the proportion of total manure N that is converted to N₂O (i.e., the EF_{N_2O}). However, in contrast to our expectations, the fecal-N:urinary-N ratio

actually *decreased* in cattle fed below maintenance, mainly because the amount of fecal-N expelled decreased with reduced feed intake, while urine volume and urine N concentrations remained constant (Ali et al., 2019; Wassie et al., 2019). This was explained by weight loss and mobilization of body protein in response to sub-maintenance feeding, and an increased excretion of endogenous N via urine (Wassie et al., 2019). Nevertheless, the observed shift from fecal-N to urinary-N due to restricted feed intake did not affect manure heap EF_{N_2O} in the present study. It is however possible that with prolonged feed scarcity beyond the duration simulated in our trial (31 days), as might occur during extended drought periods, the partitioning of fecal-N:urinary-N might change and then affect manure N₂O emissions or the manure EF_{N_2O} .

Contrastingly to feed scarcity, forage grass species did not influence manure N₂O emissions in our study. Both the cumulative N₂O emissions and the EF_{N_2O} from the three grass diets (Brachiaria, Rhodes, Napier) were similar. This might be because the chemical composition of the grasses fed was similar across the three species, with CP ranging from 7.4 to 9.6% DM and fiber contents ranging from 648 to 710 g kg⁻¹ DM for neutral detergent fiber (NDF) and from 344 to 442 g kg⁻¹ DM for acid detergent fiber (ADF) (Korir et al., in prep.). Overall, tropical perennial forage grasses are known to have low CP and high fiber contents compared to temperate pas-

tures (Lukuyu et al., 2011; Mutimura et al., 2018), often limiting cattle productivity and reducing N excretion rates in SSA. In addition, in our study the forage grasses were not fertilized. Therefore, the low manure N concentration and high manure C:N ratios in trial 2 (C:N range 34.9–39.6) we observed were expected (hypothesis i) and in the range of what others have reported for cattle fed solely on unfertilized tropical forage grasses (e.g., manure C:N of 36.6 for cattle fed on Napier, Muinga et al., 2007), which likely limited N₂O formation in the manure in this trial.

Compared to the IPCC default values, the EF_{N₂O} in our study was more than 50% lower (mean $0.45 \pm 0.05\%$) than the IPCC EF_{N₂O} of 1% (2019 Refinement of IPCC Guidelines), confirming our hypothesis ii. One reason for the low EF_{N₂O} in our study could be the wide C:N ratio of the manure, indicated by a negative correlation of manure C:N with N₂O emissions. Our values are in line with C:N values of farm-yard manure (FYM) from cattle in other studies from SSA, e.g. 23.1 ± 9.7 for Kenya (Lekasi et al., 2003), 21.7 for Kenya (Tiftonnell et al., 2010) and 29.1 ± 9.1 for South Africa (Mkhabela and Materechera, 2003). For comparison, studies from Europe have measured FYM C:N values for cattle of 14 (Amon et al., 2001), 17–24 (Chadwick, 2005) and 15 (Yamulki, 2006), and all of these studies reported EF_{N₂O} values that were higher than in our study (0.6% in Amon et al., 2001; 1.3–2.3% in Chadwick, 2005; 0.7% in Yamulki, 2006). In addition, a review of manure fertilizer quality across SSA found that cattle manure had a mean C:N of 23.3 ± 1.6 and an N:P of 3.6 ± 0.3 , indicating N-limitation and not P-limitation of manure decomposition (Sileshi et al., 2017). Nitrogen limitation reduces nitrification and denitrification rates and thereby also gaseous N losses via N₂O (Butterbach-Bahl et al., 2013; Firestone and Davidson, 1989). Our findings are in line with a growing number of studies reporting that N₂O emissions and EF_{N₂O} from cattle excreta in Africa are lower than the IPCC default values (Pelster et al., 2016; Zhu et al., 2020, 2018), even after the recent refinement of the IPCC guidelines in 2019. Given the fact that many cattle are exposed to at least seasonal feed scarcity and are regularly fed with poor-quality forages, this introduces bias in livestock GHG budgets and emission reporting for SSA and calls for incentives to promote the development of regionally-specific EFs for livestock systems in SSA.

In this study, our aim was to represent local conditions of smallholder farms in the East African highlands. More specifically, we refer to small, low-producing livestock breeds, such as the Boran cattle used in the study, which represent two thirds of cattle in Kenya (Rege et al., 2001), as well as feed scarcity and poor quality of diets consisting mostly of unfertilized pasture or tropical forage grasses without the addition of protein or energy supplements. This is a common situation on East African smallholder farms that cannot afford to purchase supplements or produce good-quality hay or silage. It should be noted that particularly in cooler and more humid climates across SSA, other cattle breeds and conditions might prevail, for example the dairy production zones of Western Kenya or Uganda sometimes use higher-producing imported or cross-breeds as well as feed rations with a higher energy and protein content (Staal et al., 2001). This might lead to higher N excretion rates and a narrower C:N ratio of the manure and might thus result in EF_{N₂O} that are closer to what has been reported for developed countries. Also, these systems might use other manure management practices such as biogas digesters or slurry pits, although to our knowledge this remains the exception rather than the norm for African smallholder farms (Mwirigi et al., 2014; Ndambi et al., 2019).

In addition to animal breed and manure management system, climate is a crucial driver of GHG emissions from manure. The experiments in the present study were conducted in Nairobi in the Kenyan highlands at 1860 m asl, where the climate is moderate and relatively humid compared to the drier and hotter lowlands.

Manure moisture content was not measured in this experiment, but we saw an effect of rainfall on N₂O emissions, with small emission peaks after rainfall events. Other authors have reported increased N₂O emissions after rainfall from manure on croplands (Sänger et al., 2010), in feedlots (Parker et al., 2017) and from solid manure storage (Maltais-Landry et al., 2018), whereas others have reported slightly lower N₂O emissions from uncovered manure heaps due to increased leaching of NO₃⁻, albeit after very heavy rainfall events (Chadwick, 2005). The magnitude and duration of N₂O emission responses to rainfall will depend on many factors, including but not limited to N concentration and chemical form (organic or inorganic), heap size, addition of bedding or other organic material to the heap, temperature, radiation, wind speed, and magnitude of the rainfall event. Including all these variables was beyond the scope of our experiments; additional studies are therefore needed to assess effects of climate on N₂O emissions from solid manure storage in other climatic zones of SSA.

In contrast to our expectations (hypothesis iii), manure CH₄ emissions were not correlated with manure C:N ratio. However, cumulative CH₄ emissions were ca. 75% lower in manure from the Rhodes grass diet compared to all other treatments. Manure from the Rhodes diet had the lowest N concentrations of all treatments ($1.04 \pm 0.02\%$ DM), which was even slightly (but not significantly) lower than the N concentration of manure in the 40% MER diet ($1.11 \pm 0.08\%$ DM). Nitrogen availability is known to affect methanogenesis in multiple ways (Bodelier and Laanbroek, 2004; Sterling et al., 2001). At low environmental N concentrations, studies from wetlands found positive correlations between soil N and methanogen numbers, indicating N limitation of methanogens (Bodelier, 2011). Furthermore, addition of mineral N can enable methanogens to switch from energetically costly N fixation and divert more energy to growth, thereby increasing CH₄ emissions (Liu et al., 2011). It is possible that the low N availability in manure from the Rhodes grass diet suppressed activity of methanogens and thereby reduced manure heap CH₄ emissions compared to the other diets, but since we did not measure methanogenic activity or abundance, we cannot prove this.

The manure CH₄ emissions in our study (ranging from 0.003 to $7.63 \text{ mg CH}_4\text{-C kg}^{-1} \text{ DM h}^{-1}$) are in the same range as reported elsewhere for manure heaps, e.g. by Chadwick (2005) who measured CH₄ flux rates ranging from 0.26 to $3.68 \text{ mg CH}_4\text{-C kg}^{-1} \text{ DM h}^{-1}$. Compared to the IPCC default EF_{CH₄} for solid manure storage (2019 Refinement), the mean EF_{CH₄} in our study was similar or lower (mean $1.73 \pm 0.49 \text{ g CH}_4 \text{ kg}^{-1} \text{ VS}$), pointing again towards a potential bias in national GHG inventories that purely rely on the existing EFs. This also indicates that the manure chemistry resulting from poor-quality and low-quantity diets (high fiber content, high ash content, low N content) restricted methanogenesis in our study.

In this study, manure was stored for 155 days to reflect common practice on smallholder farms in SSA where manure is rarely stored for longer than one dry season (ca. 4–5 months) (Ndambi et al., 2019; Tiftonnell et al., 2010). However, different practices of solid manure storage might exist on some farms (e.g. manure is dumped in a heap and then not used as fertilizer but left to decompose over a longer time period; manure is mixed with bedding material, organic waste, other waste; manure of different livestock species is mixed), which are not represented by our incubations. We therefore caution the reader to keep this in mind when using the emission factors presented here for inventory purposes.

In order to follow the IPCC Tier 2 GHG inventory approach, comprehensive data such as animal numbers and herd composition, feed intake, N excretion rates, N leaching rates, etc. are needed on a regional or national level. Despite the importance of livestock in SSA such data are not readily available, which is why most countries rely on default values, which are likely not representative for

smallholder systems and can lead to emission overestimation. It was beyond the aim of our study to build a complete dataset for true Tier 2 GHG emission estimation. However, a critical part of developing Tier 2 estimates is the development of localized emission factors, as we did here.

5. Conclusions

This is the first study reporting direct CH₄ and N₂O emissions from entire cattle excreta heaps, with manure quality being affected by feed scarcity and poor-quality diets; a common situation in SSA. Our findings corroborate the growing body of research showing that livestock excreta in smallholder systems in SSA have lower GHG emissions than those reported from industrialized systems, and that the local conditions in SSA are still not sufficiently represented in the IPCC default values even after the 2019 refinement. Given the growing livestock numbers in SSA, this has implications for national and global agricultural GHG budgets. Furthermore, since many developing countries have signed and ratified the 2015 Paris Agreement (UNFCCC, 2015) and are striving to achieve sustainable intensification and low-emissions development of their livestock production systems, data that represent local breeds and feeds and take into account local manure and feed management practices are essential to inform on GHG emission baselines and to develop meaningful climate-smart interventions. To reduce uncertainties in national livestock GHG inventories in SSA, further knowledge of livestock production (rates and practices) is necessary jointly with reliable GHG emission measurements of individual GHG emission sources (animal, manure, soil, etc.). Finally, studies investigating other pathways of nutrient losses from manure (e.g. leaching, volatilization) and that consider both direct and indirect GHG emissions across the entire production system and their mitigation in SSA are urgently needed.

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.

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Appendix A. Supplementary material

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.wasman.2021.03.014>.

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