ISSN
1859 - 0802

JOURNAL OF ANIMAL SCIENCE AND TECHNOLOGY
Vol. 153 (10/2025): 34 - 42

DOI: 10.70408/nias.i153-y2025-686

Methane yield and beef cattle productivity arising from the principal diets feed in the dry and rainy season of Vietnam

Chu Manh Thang[1], Dao Thi Binh An[1], Ngo Dinh Tan[1], Le Thi Thanh Huyen[1], Pham Doan Lan[1], Tran Thi Bich Ngoc[1], Nguyen Thanh Trung[1] and Roger Stephen Hegarty[2]

[1] National Institute of Animal Science (NIAS), Hanoi, Vietnam.

[2] University of New England, Armidale, NSW 2351, Australia

ABSTRACT

The effect of feeding principal beef cattle diets of Vietnam's dry and rainy seasons on feed intake, digestibility, rumen metabolic profile, performance and enteric methane production was assessed in an experiment. A survey was conducted to classify and document the principal diets of beef cattle in three regions of Vietnam, being North, Central and Southern Vietnam across the dry and rainy season. The animals were allocated in an experiment with three treatments feeding typical diets in two seasons. Fifteen growing male crossbred cattle (Wagyu x Zebu) with average initial BW (198.9kg ± 36.9) at 11.9-18.3 months of age were subjected to measurements of CH4 output using the GreenFeed system, nutrient intake and apparent digestibility in a RCBD experiment. Daily emissions of methane and CO2 were measured using a GreenFeed device (GF; C-Lock Inc. South Dakota USA) used in the "tie stall mode". In each season there was an effect of diet type for beef cattle on dry matter intake, animal performance and enteric CH4 emission using GF in two seasons of the year in Vietnam.

There is an effect of feeding different typical diets in the seasons for beef cattle on dry matter intake, animal performance and enteric CH4 emission using GreenFeed Emission Monitoring system (GEM; C-Lock Inc., Rapid City, South Dakota, USA). The Ym values of 5.3-5.4% with the treatment in dry season and 4.4-4.5% in rainy season. For CH4 emissions expressed in g/kg DMI (MY) were 27- 35 g CH4/kg DMI. The emission factor (EF) value (kg CH4/head/year) was around 53 to 69 found in the beef cattle fed the diets in dry season and 57 to 71 in those fed diets in rainy season.

Keywords: *Methane emission, dairy cow diets, regions, seasons, GreenFeed device*

Introduction

The recent increase in the sale price of beef cattle has raised considerable interest among farmers to switch from an extensive, pasture-based system to an intensive stall feeding production system in Vietnam. Methane is a potent greenhouse gas (GHG) whose atmospheric abundance has grown 2.5-fold over three centuries (Lassey, 2007), and which contributes proportionately 0.143 of all factors to present global warming (Johnson *et al.*, 2007). Within the agricultural sector, CH_4 emissions from enteric fermentation of ruminants are considered to be of primary concern (Johnson *et al.*, 2007).

There are many factors which influence unwanted CH_4 production by ruminants: level of feed intake, type and quality of feed, energy consumption, animal size, growth rate, level of production and environmental temperature (Broucek, 2014). Measurement of CH_4 emissions due to enteric fermentation must be taken into account under conditions as close as possible to typical as found in farming systems. The production of enteric CH_4 is a loss of feed

Corresponding author: Chu Manh Thang;
Tel: 0989126940; *Email: thangslu@gmail.com

energy from the diet and represents inefficient utilization of the feed (Chagunda *et al.*, 2009). In addition to environmental implications, ruminant methanogenesis represents a loss of 2% to 12% of the gross energy intake (Johnson and Johnson, 1995; Boadi and Wittenberg, 2002; Benchaar and Greathead, 2011).

The GreenFeed Emission Monitoring system (GEM; C-Lock Inc., Rapid City, South Dakota, USA) is the first time introduced to Vietnam as a system to estimate daily methane production (DMP) of cattle from repeated short-term measures of methane emission over a period of days, weeks or months. Velazco *et al.* (2016) showed that the DMP estimated by averaging multiple short-term breath measures of methane emission rate using GEM does not differ from these measures obtained from respiration champers.

The effect of feeding different typical diets on both animal performance and CH_4 emission using measurements of GF system is not well documented. Understanding the reasons for the variability in enteric CH_4 production in relation to diet is essential to decreasing uncertainty in GHG emission inventories and to identifying viable GHG mitigation strategies. Thus, the objective of the present study was to determine the effect of feeding different diets on animal performance and enteric CH_4 emission in dry and rainy season of Vietnam.

Materials and methods

Materials

Fifteen growing male crossbred cattle (Wagyu × Zebu) with average initial BW (198.9 kg ± 36.9) at 11.9-18.3 months of age were subjected to measurements of CH_4 output using the GreenFeed system, nutrient intake and apparent digestibility in a RCBD experiment.

Experimental time and site

The experiment was conducted at a beef cattle farm belonging to the National Institute of Animal Science (NIAS) in Ba Vi district, Hanoi province, Northern part of Vietnam. The experiment was carried out in the dry season (April-May) and rainy season (August-September) of 2024 (Each gas collection experiment was conducted over 35 days per season).

Experimetal method

Experimental design

The experiment was carried out to evaluate the animal performance and enteric CH_4 emission by feeding different typical diets on dry and rainy season for beef cattle.

The experimental animals were allocated in an experiment RCBD design with three treatments feeding typical diets in two seasons. The diets were formulated based on the previous survey information of the typical ration for different dairy production in three regions (North-Central-South) of Vietnam. Typical seasonal diets on beef cattle farms were constructed based on a field survey. The feeding cattle diets are specific to each region (North, Central and South of Vietnam). This survey was conducted to collect information beef cattle diets in two season from August 2023 to May 2024 on 42 beef farms which were located in three typical cattle regions of Vietnam including a Northern region (10 farms in Bac Giang, Vietnam), a Central region (9 farms in Thanh Hoa; 9 farms in Binh Dinh, Vietnam), a Southern region (10 farms in Dong Nai and 4 farms in Ben Tre, Vietnam). The information of dietary ingredients and amounts of each that the farmers offer the cattle were recorded and took feed sample for analysis the nutrient compositions of each. Although the information from these survey was insufficient to allow the reliable analysis of nutrient concentrations in the diets, it suggests potential for diets in the regions and seasons (un-published).

The experimental diets for dry season and rainy is showed in Table 1. Experimental layout and measurement procedure were showed in Annex 1.

Table 1. The experimental diets (% on DM basis)

Items	Dry season			Rainy season		
	Diet 1	Diet 2	Diet 3	Diet 1	Diet 2	Diet 3
Maize stover silage (%)	-	-	53.17	-	-	54.02
Elephant silage	47.63	-	-	54.35	-	-
Ruzi grass				-	53.87	-
Rice straw	-	53.76	-	-	-	-
Pelleted Concentrate	7.40	15.13	7.60	5.78	5.84	5.82
Feed concentrate	44.97	31.11	39.23	39.87	40.29	40.16
Composition and nutrient value						
ME (MJ/kg DM)	4.82[b]	6.94[a]	6.85[a]	10.97[b]	9.94[c]	11.85[a]
OM (%DM)	93.3[b]	89.4[c]	95.2[a]	89.23[c]	90.26[b]	90.46[a]
CP (%DM)	12.44[b]	14.07[a]	12.80[b]	16.27[a]	14.28[c]	15.63[b]
EE (%DM)	3.24[a]	2.11[c]	2.75[b]	2.96[b]	2.75[c]	3.39[a]
CF (%DM)	19.76[b]	27.29[a]	17.99[c]	23.22[a]	23.39[a]	17.67[b]
NDF (%DM)	50.56[b]	59.02[a]	48.29[c]	26.11[a]	26.39[a]	21.69[b]
ADF (%DM)	29.31[c]	46.45[a]	32.14[b]	55.09[b]	56.49[a]	48.38[c]

Note: Diet 1, Diet 2 and Diet 3 were the typical ration for beef cattle in the North, Central and South of Vietnam, respectively.

Experimental animals and feeds

The animals were the same breed, sexual, and were individually identified by numbered ear tags (RFID). Cattle were allocated to treatments (n=3) using stratified randomisation, being ranked initially on months of age (Block), then on initial animal BW. Allocations were made so that there was one animal/treatment in each of 5 blocks. The re-growth of elephant grass (*Pennisetum purpureum)* at an age of 60-75 days cutting interval were harvested daily in the morning. It was chopped into 10-15 cm length before feeding.

Table 2. Chemical composition and nutrition value of experimental feeds (Mean, SD)

Feeds	DM (%)	CP (% DM)	EE (% DM)	CF (% DM)	NDF (% DM)	ADF (% DM)	Ash (% DM)
Elephant grasses	17.2 (1.83)	7.4 (1.67)	1.57 (0.08)	42.08 (2.58)	76.59 (1.53)	64.41 (3.41)	5.41 (0.90)
Elephant silage	17.4 (1.44)	7.57 (0.81)	1.88 (0.11)	42.17 (2.09)	79.79 (0.13)	70.87 (5.47)	9.91 (0.82)
Green bean husk	89.8 (0.41)	18.5 (0.59)	2.01 (0.29)	21.94 (1.04)	49.12 (4.37)	37.16 (0.16)	4.17 (0.16)
Maize meal	86.8 (0.29)	8.93 (0.69)	4.58 (0.06)	2.77 (1.19)	21.14 (11.3)	6.13 (0.19)	1.29 (0.02)
Maize stover silage	31.2 (1.89)	7.9 (0.09)	3.78 (1.81)	24.36 (0.05)	68.3 (22.7)	45.14 (5.18)	4.49 (0.08)
Pelleted Concentrate	89.4 (0.92)	18.76 (0.87)	4.99 (0.35)	9.88 (3.73)	37.9 (5.03)	23.93 (2.23)	7.63 (1.35)
Rice bran	90.4 (0.31)	7.12 (0.05)	6.20 (0.53)	33.56 (1.09)	55.22 (0.85)	47.90 (0.56)	12.82 (1.80)
Rice straw	91.5 (0.14)	7.735 (1.04)	2.03 (0.32)	38.29 (3.32)	74.37 (2.82)	64.78 (9.19)	12.1 0(.31)
Soyabean meal	87.3 (0.13)	49.36 (3.32)	1.61 (0.09)	4.66 (0.26)	31.77 (9.51)	8.84 (0.40)	6.80 (0.47)
Wheat bran	89.4 (0.39)	13.87 (0.59)	3.39 (0.13)	9.99 (0.90)	40.3 (2.84)	17.15 (1.09)	17.47 (1.17)

The silage elephant grass (*Pennisetum purpureum*) at an age of 60-75 days cutting interval were harvested in the morning. It was chopped into 10-15 cm length before silage. The maize stover ensilaged with 0.5% salt for 60 days before feeding.

Feeding and management

The feeds using in the experiment were based on diet in previous survey and was offered equally twice per day to individual animal in the morning (07h30) and afternoon (16h30). On each feeding occasion, the concentrate mixed with other grain feeds as designed in a separate bucket were supplied first to the animals, then grass, silage, and rice straw were offered.

Each animal was freely accessed to clean drinking water and a mineral lick block containing Ca 90 g, P 90 g, Na 150 g, Mg 5 g, Fe 10 g, Mn 6000 mg, Cu 800 mg, Co 400 mg, I 50 mg and Se 100 mg per 1 kg block.

The experiment period was lasted for 35 days including 7days of an adaptation period, 14 days for rumen stabilization and 14 days for diet restriction and data collection of feed intake, milk yield and the last period of data recording of CH_4 emission (2 x 3days), rumen fluid collection (1 day).

Cattle were also provided with a measured quantity of pelleted bait concentrate each time they accessed the GEM unit (89.4% DM, 18.8% CP, 37.9% NDF, 23.9% ADF, 4.9% fat)

Data collection

Feed intake: The daily feed consumption was recorded, and refusals were collected for individual animals in the morning of the next day. The intake of concentrate, supplemental feeds and forages were measured daily, based on the amount of feeds offered and refused. The total feed intake (DMI) was calculated as the sum of the intake of the feed components. Total gross energy (GE) intake was estimated from DMI and chemical components, using the equation suggested by Jentsch *et al.* (2007).

Live weight: After the adaptation period the animals were weighed at 06.00 h before feeding on two consecutive days using RudWeight, and at 15 day intervals. Feed conversion ratio (FCR) was calculated as kg feed consumed /kg live weight gain (LWG).

Rumen fluid was collected from experimental animal using a rubber tube connected to a vacuum pump in the day of 35^{th}. The sample was analyzed the Protozoa counting and rumen VFAs profile composition at the National Center for Food Analysis and Assessment - Food industries research institute.

Digestibility was investigated using acid insoluble ash (AIA) as an internal marker and collection of manure samples directly from the rectum of animal.

Daily methane production (DMP): The CH_4 was measured using a GreenFeed devices (**GF**; C-Lock Inc.) the cow voluntarily placed their heads in a shroud and were detected by the RFID sensors. Eructated CH_4 was measured while cows consumed the pellet supplement. The GF devices were calibrated daily following the manufacturer's instructions. The pelleted concentrate was used to bait the cows and ensure the proper head position in the bin. On days when cows were being given access to the Greenfeed, the amount of pelleted concentrate being fed in the pen was reduced by an amount equal to the quantity of pellet provide through the Greenfeed. Methane emission energy (MEE, MJ/day) was converted from value of CH_4 production measuring from GF.

Methane conversion factor (Ym) as % of gross energy intake was calculated basal on MEE and total GE intake. Methane yield (MY) was calculated as g methane production per kg DM intake, daily. Methane intensity was calculated as g methane production per kg LWG, daily.

Chemical analysis

The feeds offered and individual feed refusals were sampled daily and pooled to a sample for each fifteen days. Samples of feeds, refusals were analysed for DM, CP, ether extract (EE), neutral detergent fibre (NDF), acid detergent fibre (ADF), ash and Acid insoluble ash (AIA).

The milk samples were analysed for DM, CP, fat and total solid. The DM (ID 930.15), CP (ID 976.05), ash (ID 942.05) and AIA (ID 941.12) were analysed according to the standard methods of AOAC 952.03 (2000). The EE was analyzed by ISO-6492 (1999) and NDF and ADF concentrations were determined according to the procedure of Van Soest *et al.* (1991).

Gas sampling and methane measurement

The GreenFeed system (C-lock inc. USA) was used to measure CH_4, CO_2, O_2, H_2 during three consecutive days. Each animal were measured on 8 occasions, for at least 5 minutes, with sampling times balanced across the 24h day.

Statistical analysis

The data were analysed statistically as a Randomized Completely Block Design (RCBD) by variance analysis (ANOVA) using the general linear model (GLM) procedure of Minitab software version 14.0 (Minitab, 2003). The treatment least square means showing significant differences at the probability level of P<0.05 were compared using Tukey's pairwise comparison procedure.

The statistical model used in the trial was $Y_{ij} = \mu + \alpha_i + \beta_j + \varepsilon_{ij}$ where Y_{ij} is the dependent variable, μ is the overall mean, α_i is effect of treatment i, β_j is effect of block and ε_{ij} is a random error.

Results and discussion

Feed intake, nutrient digestibility and performance

The data on feed intake and digestibility of experimental animal feeding the diets in dry and rainy is shown in Table 3. The difference in DMI (%BW) of animal fed these treatments was significant (P<0.05). The seasonal effect on DM intake and digestibility (CF, NDF and ADF) of cattle are significant (P<0.001) and LWG (P<0.05) of animal fed different diets.

Table 3. The feed intake, nutrient digestibility and animal performance

Items	Dry season			Rainy season			Effects	
	Diet 1	Diet 2	Diet 3	Diet 1	Diet 2	Diet 3	Diets	Seasons
Feed intake								
Total DM intake (kg/day)	4.44^{e}	7.39^{a}	5.15^{d}	7.23^{a}	6.12^{b}	5.71^{c}	***	***
DM intake (%BW)	2.04^{c}	3.28^{a}	2.35^{b}	2.44^{b}	2.15^{c}	2.05^{c}	***	***
DM intake (kg $BW^{0.75}$)	78.3^{e}	126.8^{a}	90.3^{c}	100.9^{b}	88.3^{cd}	83.6^{de}	***	***
GE intake (MJ/day)	82.2^{d}	131^{a}	96.8^{c}	130^{a}	110^{b}	104^{bc}	***	***
Digestibility (%)								
DM	79.71^{a}	52.2b^{b}	77.95^{a}	65.42^{ab}	54.56^{b}	66.14^{ab}	**	*
OM	80.53^{a}	55.18^{b}	78.95^{a}	64.75^{ab}	54.18^{b}	65.90^{ab}	**	*
CP	80.63^{a}	54.52^{bc}	76.34^{a}	63.99^{ab}	42.65^{c}	62.87^{abc}	***	**
EE	85.40^{a}	56.96^{b}	84.19^{a}	79.15^{a}	62.03^{b}	86.62^{a}	***	NS
CF	75.16^{a}	56.32^{bc}	74.44^{ab}	58.62^{abc}	48.49^{c}	48.99^{c}	**	***
NDF	76.99^{a}	51.44^{b}	75.79^{a}	55.59^{b}	45.70^{b}	51.04^{b}	**	***
ADF	67.76^{ab}	50.70^{abc}	72.37^{a}	48.41^{bc}	37.25^{c}	40.33^{c}	*	***
Animal performance								
LWG (g/day)	792.3	896.1	688.5	431.6	515.8	636.8	NS	*

*Note: [a,b,c]Mean within rows with different superscripts are significantly different (P<0.05); Diet 1, diet 2 and diet 3 were the typical ration for beef cattle in the North, Central and South of Vietnam, respectively. * significantly different level at P<0.05, **significantly different level at P<0.01, *** significantly different level at P<0.001, NS: none significantly different (P>0.05)*

In the present study, the different intake and digestibility of animal fed beef cattle diets could be due to the structure dietary fiber among the regions and seasons, which facilitate easier attack by microorganisms in the rumen and solubility characteristics. For this reason, the increased intake in the diets is likely due to the combined effects of diets in the regions and seasons.

The and protozoa and rumen VFAs profile are showed in Table 4. There was no significant difference of protozoa ($x10^5$) and rumen VFAs profile among these treatments (P>0.05). There are an effect of season on the acetate and propionate (as % of total VFAs) (P<0.01).

Table 4. Protozoa and rumen VFAs profile

Items	Dry season			Rainy season			Effects	
	Diet 1	Diet 2	Diet 3	Diet 1	Diet 2	Diet 3	Diets	Seasons
Protozoa (x 10^5 cell/ml) (post 04h feeding)	2.40	3.09	4.10	4.28	3.48	4.37	NS	NS
Acetate (% total VFAs)	66.8	67.7	67.2	65.8	64.4	64.4	NS	**
Propionate (% total VFAs)	17.5	17.5	17.8	18.7	20.0	19.7	NS	**
Butyrate (% total VFAs)	15.7	14.8	15.0	15.5	15.6	15.9	NS	NS

*Note: ^{a,b,c}Mean within rows with different superscripts are significantly different (P<0.05); Diet 1, diet 2 and diet 3 were the typical ration for beef cattle in the North, Central and South of Vietnam, respectively. * significantly different level at P<0.05, **significantly different level at P<0.01, *** significantly different level at P<0.001, NS: none significantly different (P>0.05)*

Methane emisson

The GreenFeed Emission Monitoring system (GEM; C-Lock Inc., Rapid City, South Dakota, USA) is a commercial system developed to estimate daily methane production (DMP) of cattle from repeated short-term measures of methane emission over a period of days, weeks or months. Velazco *et al.* (2016) showed that the DMP estimated by averaging multiple short-term breath measures of methane emission rate using GEM does not differ from these measures obtained from respiration champers.

Table 5. Methane emission and EF value

Items	Dry season			Rainy season			Effects	
	Diet 1	Diet 2	Diet 3	Diet 1	Diet 2	Diet 3	Diets	Seasons
Total CH_4, g/day	153.8^e	189.7^b	146.2^f	194.3^a	163.7^c	156.8^d	**	**
MY (g CH_4/kg DMI)	35.4^a	26.8^c	29.9^b	28.3^{bc}	26.9^c	27.8^c	*	*
Methane intensity (g CH_4/kg meat produced)	230^d	217^f	224^e	409^b	413^a	362^c	**	***
EF value (kg CH_4/head/year)	56.1^e	69.2^b	53.4^f	70.9^a	59.7^c	57.2^d	*	**
Ym (%)	5.29^b	5.39^a	5.26^c	4.41^e	4.45^{de}	4.49^d	***	***

Note: ^{a,b}Mean within rows with different superscripts are significantly different (P<0.05); MY: Methane yield (g CH_4 per kg DM intake); FPCM: fat protein corrected milk, EF: emission factor value; Diet 1, diet 2 and diet 3 were the typical ration for beef cattle in the North, Central and South of Vietnam, respectively

In the present study, the average daily CH_4, EF value and Ym in different regions and seasons is showed in Table 5. The greater value of MY but lower value of methane intensity (g CH_4/kg kg FCM or g CH_4/kg kg FPCM) was observed in animal fed the diets in dry season comparing to those fed in rainy season. The value methane production is variable among the regions and season. The Ym values of 5.3-5.4% with the treatment in dry season and 4.4-4.5% in rainy season. For CH_4 emissions expressed in g/kg DMI (MY) were 27- 35 g CH_4/kg DMI in current study. This is in agreement with prior data recommended by IPCC (2019) in dairy cattle in different production levels ranged by 19-21.4.

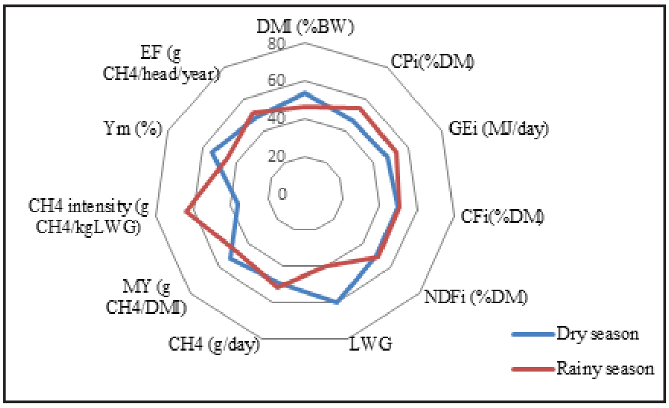

Figure 1. Effects of different seasons on feed intake, animal performance and CH_4 emission

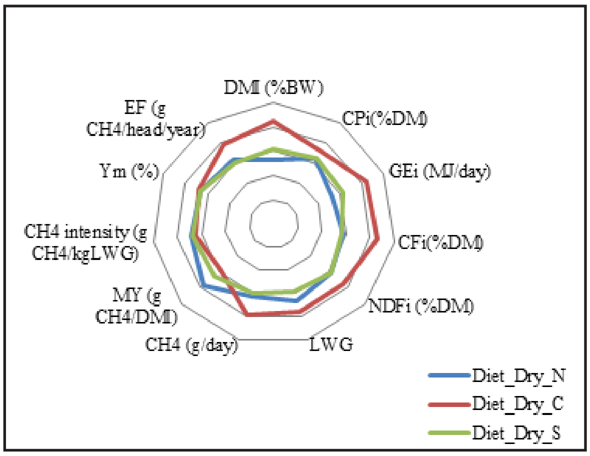

Figure 2. Effects of different typical diets of beef cattle on feed intake, animal performance and CH_4 emission in dry season

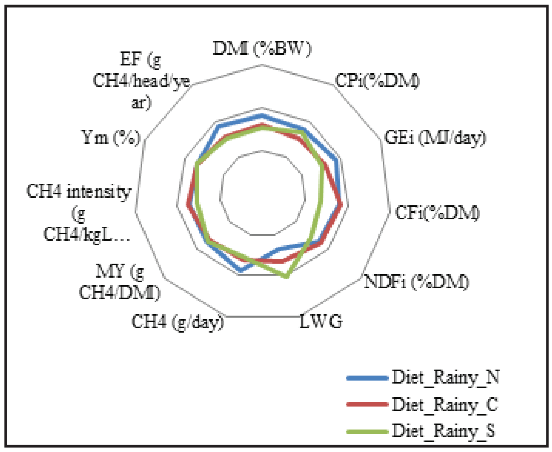

Figure 3. Effects of different typical diets of beef cattle on feed intake, animal performance and CH_4 emission in rainy season

The emission factor (EF) value (kg CH_4/head/year) was around 53 to 69 in the animal fed the diets in dry season and 57 to 71 in those fed diets in rainy season, in the current study. There were regional and seasonal effects on the EF value from beef cattle in Vietnam. In dry season, the effects on methane emission factor and intensity are obviously clear among the diets (Figure 2). The EF value in present study was slightly higher than the value recommended by IPCC default value (43-56 kg/head/year) for other cattle in the production system in Asia (IPCC, 2019).

Energy lost as enteric CH_4 from mature cattle ranges from 2.2 to nearly 12% of gross energy (GE) intake (Johnson *et al.*, 1995). This range largely depends on the level of feed intake and the composition of the diet (Moss *et al.*, 2000). Canesin *et al.* (2014) has been indicated that the feed system is based on tropical forages with the diet consists in low nutritional value forage, resulted in the low performance of animals and the increased production of greenhouse gases (GHGs), mainly enteric methane. In dry season, this occurred as a result of the chemical variation in the forage materials which could be the primary cause of differences in dry matter, protein and NDF intake may effect on the animal performance and CH_4 production in current study. Demarchi *et al.* (2016) had reported that a direct relationship among forage quality, DM intake and consequently CH_4 emissions among seasons. In the study, higher feed intake in Treat. 2 resulted in lower value of MY while the greater DMI in Treat. 3 showed the higher MY comparing to the value in Treat. 1. In the ruminants with high feed intakes, reductions in enteric CH_4 emissions per unit of intake with increased digestibility of feeds have been reported (Hristov *et al.*, 2013). In dry season, it is probably due to the animals in treatment 3 fed the diet with higher structural carbohydrates comparing to other diets. Janssen (2010) have been reported that the animal fed the diet with high structural carbohydrates be more methanogenic than soluble carbohydrates.

Conclusions

There is an effect of feeding different typical diets in the seasons for beef cattle on dry matter intake, animal performance and enteric CH_4 emission using GreenFeed Emission Monitoring system (GEM; C-Lock Inc., Rapid City, South Dakota, USA). The Ym values of 5.3-5.4% with the treatment in dry season and 4.4-4.5% in rainy season. For CH_4 emissions expressed in g/kg DMI (MY) were 27- 35 g CH_4/kg DMI. The emission factor (EF) value (kg CH_4/head/year) was around 53 to 69 found in the beef cattle fed the diets in dry season and 57 to 71 in those fed diets in rainy season, in the study.

Acknowledgements

The authors acknowledge the support from the New Zealand Agricultural Greenhouse Gas Research Centre (NZAGRC) for the budget.

Conflict of interest

The authors declare that they have no conflict of interest.

Authors contribution

All authors contributed to the work, discussed the results and contributed to the final manuscript.

Annex

Annex 1. Experimental layout

Items	Diet 1	Diet 2	Diet 3
N° of animal (head)	5	5	5
Adaptation time (days)		21	
Experimental time (days)		35	
		Activities	
Day 1st -7th	Adaptation period		
Day 8-21st	Cow fed experimental diets, weighing animal BW The cows were trained to familiar with the Greenfeed system		
Day 22th - 28th	Measurements: daily feed intake, milk yield Also collect bulked manure and feed samples from day 21- 35		
Day 29, 30, 31st	Measurement CH_4, CO_2, O_2 using GreenFeed (C-Lock inc.)		
Day 32th	Recovery day for cattle		
Day 33,34,35th	Measurement CH_4, CO_2, O_2 using GreenFeed (C-Lock inc.)		
Day 35th	Rumen fluid sample (pH, Protozoa)		

References

AOAC 952.03. 2000. Official method of analytical chemists, 17th. M. Gaithersburg, ed. Association of Analytical Chemists, Washington, DC.

Benchaar, C. and H. Greathead. 2011. Essential oils and opportunities to mitigate enteric methane emissions from ruminants. Animal Feed Science and Technology 166, pp. 338-355.

Boadi, D. and Wittenberg, K. 2002. Methane production from dairy and beef heifers fed forages differing in nutrient density using the sulphur hexafluoride (SF_6) tracer gas technique. Canadian Journal of Animal Science 82(2), pp. 201-206.

Broucek, J. 2014. Production of Methane Emissions from Ruminant Husbandry: A Review. Journal of Environmental Protection Vol.05No.15:12.

Canesin, R.C., Berchielli, T.T., Messana, J.D., Baldi, F., Pires, A.V., Frighetto, R.T.S., Fiorentini, G., and Reis, R.A. 2014. Effects of supplementation frequency on the ruminal fermentation and enteric methane production of beef cattle grazing in tropical pastures. Revista Brasileira de zootecnia 43:590-600.

Chagunda, M., Römer, D., and Roberts, D. 2009. Effect of genotype and feeding regime on enteric methane, non-milk nitrogen and performance of dairy cows during the winter feeding period. Livestock Science 122(2-3), pp. 323-332.

Demarchi, J.d.A., Manella, M., PRIMAVESI, O., Frighetto, R., Romero, L., Berndt, A., and de LIMA, M. 2016. Effect of seasons on enteric methane emissions from cattle grazing Urochloa brizantha.

Hristov, A., Oh, J., Firkins, J., Dijkstra, J., Kebreab, E., Waghorn, G., Makkar, H., Adesogan, A., Yang, W., and Lee, C. 2013. Special topics-Mitigation of methane and nitrous oxide emissions from animal operations: I. A review of enteric methane mitigation options. Journal of animal science 91(11), pp. 5045-5069.

IPCC. 2019. Refinement to the 2006 IPCC Guidelines for National Greenhouse Gas Inventories. E. Calvo Buendia, Tanabe, K., Kranjc, A., Baasansuren, J., Fukuda, M., Ngarize, S., Osako, A., Pyrozhenko, Y., Shermanau, P. and Federici, S. (eds)., ed, IPCC, Switzerland. Volume 4, Chapter 10. https://www.ipcc.ch/report/2019-refinement-to-the-2006-ipcc-guidelines-for-national-greenhouse-gas-inventories/.

ISO-6492. 1999. International organization of standardization. Animal feeding stuffs. Determination of Fat content.. D. f. s. a. quality, ed, Hanoi, Vietnam.

Janssen, P.H. 2010. Influence of hydrogen on rumen methane formation and fermentation balances through microbial growth kinetics and fermentation thermodynamics. Animal Feed Science and Technology 160(1-2), pp. 1-22.

Johnson, K.A., and Johnson, D.E. 1995. Methane emissions from cattle. Journal of animal science 73(8):2483-2492.

Johnson, J.M.F., Franzluebbers, A.J., Sharon Lachnicht Weyers, and Donald C. Reicosky. 2007. Agricultural opportunities to mitigate greenhouse gas emissions. Environmental Pollution. Volume 150, Issue 1, November 2007, pp. 107-124

Lassey, K.R. 2007. Livestock methane emission: From the individual grazing animal through national inventories to the global methane cycle. Agricultural and Forest Meteorology 142 (2007) 120-132

Moss, A.R., Jounany, J.P., and Neevbold, J. 2000. Methane production by ruminants: its contribution to global warming. Ann Zootech 49, pp. 231-253

Minitab. 2003. Minitab statistical software, release 14 for Window. Minitab Inc., State College, PA.

Van Soest, P. v., Robertson, J., and Lewis, B. 1991. Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. Journal of dairy science 74(10), pp. 3583-3597.

Velazco, J., Mayer, D., Zimmerman, S., and Hegarty, R. 2016. Use of short-term breath measures to estimate daily methane production by cattle. animal 10(1), pp. 25-33.

Opponent: Dr. Pham Kim Cuong