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Methane Emissions from Grazing Holstein-Friesian Heifers at Different Ages Estimated Using the Sulfur Hexafluoride Tracer Technique

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ABSTRACT

Although the effect of animal and diet factors on enteric methane (CH_4) emissions from confined cattle has been extensively examined, less data is available regarding CH_4 emissions from grazing young cattle. A study was undertaken to evaluate the effect of the physiological state of Holstein-Friesian heifers on their enteric CH_4 emissions while grazing a perennial ryegrass sward. Two experiments were conducted: Experiment 1 ran from May 2011 for 11 weeks and Experiment 2 ran from August 2011 for 10 weeks. In each experiment, Holstein-Friesian heifers were divided into three treatment groups (12 animals/group) consisting of calves, yearling heifers, and in-calf heifers (average ages: 8.5, 14.5, and 20.5 months, respectively). Methane emissions were estimated for each animal in the final week of each experiment using the sulfur hexafluoride tracer technique. Dry matter (DM) intake was estimated using the calculated metabolizable energy (ME) requirement divided by the ME concentration in the grazed grass. As expected, live weight increased with increasing animal age ($P < 0.001$); however, there was no difference in live weight gain among the three groups in Experiment 1, although in Experiment 2, this variable decreased with increasing animal age ($P < 0.001$). In Experiment 1, yearling heifers had the highest CH_4 emissions ($g{\cdot}d^{-1}$) and in-calf heifers produced more than calves ($P < 0.001$). When expressed as CH_4 emissions per unit of live weight, DM intake, and gross energy (GE) intake, yearling heifers had higher emission rates than calves and in-calf heifers ($P < 0.001$). However, the effects on CH_4 emissions were different in Experiment 2, in which CH_4 emissions ($g{\cdot}d^{-1}$) increased linearly with increasing animal age ($P < 0.001$), although the difference between yearling and in-calf heifers was not significant. The CH_4/live weight ratio was lower in in-calf heifers than in the other two groups ($P < 0.001$), while CH_4 energy output as a proportion of GE intake was lower in calves than in yearling and in-calf heifers ($P < 0.05$). All data were then pooled and used to develop prediction equations for CH_4 emissions. All relationships are significant ($P < 0.001$), with R^2 values ranging from 0.630 to 0.682. These models indicate that CH_4 emissions could be increased by 0.252 $g{\cdot}d^{-1}$ with an increase of 1 kg live weight or by 14.9 $g{\cdot}d^{-1}$ with an increase of 1 $kg{\cdot}d^{-1}$ of DM intake; or, the CH_4 energy output could be increased by 0.046 $MJ{\cdot}d^{-1}$ with an increase of 1 $MJ{\cdot}d^{-1}$ of GE intake. These results provide an alternative approach for estimating CH_4 emissions from grazing dairy heifers when actual CH_4 emission data are not available.

1. Introduction

Dairy production is a considerable source of greenhouse gas (GHG) emissions [1]. At a global scale, dairy production contributes 2.7% of total anthropogenic GHG emissions, while the total emissions attributed to dairy herds, including transport activities, meat production from old or young fattened stock, and draught power, are estimated to be about 4.0% of total anthropogenic GHG

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emissions [2]. Methane (CH_4) and nitrous oxide (N_2O) are the main GHGs emitted from the dairy sector, representing over 50% and 30%–40% of total anthropogenic GHG emissions, respectively [2]. Therefore, there is increasing interest in the development of more accurate data to predict total CH_4 emissions from different categories of dairy production systems. The literature contains many studies that evaluate the effects of animal and dietary factors on CH_4 emissions from adult dairy cattle around the world [3,4], but few studies on confined dairy heifers [5]. Furthermore, there is little information available on the quantification of enteric CH_4 emissions for grazing dairy heifers. As grazing dairy heifers are in different physiological states than confined adult cattle and are offered different diets, using the prediction models for adult cattle to predict CH_4 emissions for grazing young cattle could result in systematic errors. The lack of such information can impact the development of robust CH_4 emission inventories and appropriate mitigation strategies for dairy production systems.

The sulfur hexafluoride (SF_6) tracer technique was developed by Johnson et al. [6] and is now widely used to estimate CH_4 emissions from grazing livestock. This technique uses the inert tracer gas SF_6 as a marker along with CH_4 concentration in an expired breath sample collected around the cattle's mouth and nostrils to calculate CH_4 emissions in a daily basis. Although this technique has been reported to have a range of limitations, such as possible reduction of the release rate of SF_6 from a permeation tube when placed in the rumen of cattle for a long time, its CH_4 emission estimates have been found to be comparable to direct measurements from respiration calorimeters [7,8]. Therefore, the objectives of the present study were to: ① use the SF_6 tracer technique to quantify CH_4 emissions from grazing replacement dairy heifers, ② evaluate the effects of age and physiological state of Holstein-Friesian heifers on CH_4 emissions, and ③ use these data to develop prediction equations for CH_4 emissions from young stock, based on animal and diet factors.

2. Materials and methods

This study was conducted at the Agri-Food and Biosciences Institute (AFBI) farm at Hillsborough, County Down, UK. It complied with the requirements of the UK Animals (Scientific Procedures) Act 1986 and was approved by the AFBI Hillsborough Ethical Review Group.

2.1. Animals, experimental design, and grazing management

Seventy-two Holstein-Friesian heifers, sourced from the AFBI Hillsborough dairy herd, were allocated to one of two grazing periods (Experiment 1 and Experiment 2), with 36 animals per period. Within each experiment, heifers were allocated, according to their age at the start of the study, to treatment groups reflecting one of three developmental stages (12 animals/group). The treatment groups were: calves (5–10 months), yearling heifers (12–17 months), and confirmed in-calf heifers (18–23 months). The in-calf heifers were made pregnant by artificial insemination and were predicted to calve by 24 months of age. In Experiment 1, all 36 animals grazed the same pasture of predominantly perennial ryegrass for 11 weeks in the early part of the 2011 grazing season (16 May to 29 July). A similar arrangement with grazing for 10 weeks was undertaken for the second group of 36 heifers in Experiment 2 (15 August to 21 October 2011). In each experiment, the grazing area was split into 16 paddocks of 1.6 hectares each. In Experiment 1, a leader-follower grazing system was used, with additional groups of non-experimental heifers (aged 6–11 months) used as the follower group for the calf treatment and as the leader group for the yearling or in-calf heifer treatment. In Experiment 2, an independent rotational paddock system was introduced due to less favorable weather and soil conditions. No supplementary feed was offered to any of the animals.

2.2. Pasture and animal measurements

Pasture height was measured daily pre- and post-grazing using a rising plate meter, with 40 random recordings taken across each paddock in a "W" formation [9]. Herbage was sampled daily through the "W" formation from pre-grazed areas of the pasture and deemed to be representative of what the animals were observed eating. Samples were collected using Gardena Accu 6 battery-powered shears (Kress and Kastner, Weiterstadt, Germany). Herbage was cut to a height that was considered to be representative of what was expected during grazing based on previous observations and consumption data. Fresh samples were analyzed for metabolizable energy (ME) concentration using near-infrared spectroscopy (NIRS) as described by Park et al. [10]. The remainder of each daily sample was chopped into 40–50 mm lengths, freeze-dried, and then hammer-milled before being composited for an analysis of neutral detergent fiber (NDF), acid detergent fiber (ADF), nitrogen (N), ash, and water-soluble carbohydrates (WSC) concentrations by wet chemistry methods. Gross energy (GE) was determined by isoperibol bomb calorimetry (Parr Instruments Co., Moline, Illinois, USA), according to the method described by Porter [11]. Nitrogen was determined by the Dumas combustion method using a nitrogen analyzer (Elementar Vario MAX CN; Elementar Analysensysteme GmbH, Hanau, Germany), and crude protein (CP) concentration was calculated as the Dumas N concentration multiplied by 6.25. Concentrations of NDF and ADF were determined as described by Cushnahan and Gordon [12], using a Fibertec M 1020 hot extractor and 1021 cold extractor (Tecator AB, Hoganas, Sweden). Ash concentration was obtained by burning samples in a muffle furnace at 550 °C for 10 h.

Animal live weight was recorded daily throughout each experiment using a calibrated electronic weighing scale (EziWeigh; Tru-Test Ltd., Auckland, New Zealand) with Bluetooth File Transfer of weights to a hand-held Psion data logger.

2.3. Methane measurements

Methane emissions from individual heifers were estimated over a four-day period in the final week of each experiment, using minor modifications [8] of the SF_6 tracer technique from Johnson et al. [6]. To summarize, a permeation tube containing SF_6 was placed in the rumen of each animal seven days and four days, respectively, prior to commencing CH_4 measurements in Experiments 1 and 2. The preparation, calibration, and use of permeation tubes was as described by Muñoz et al. [8] and the allocation of permeation tubes to heifers was randomized. The SF_6 release rate and projected expiry date of each permeation tube were known prior to placement in the rumen. The measured SF_6 release rates of the permeation tubes ranged from 3.99 $mg \cdot d^{-1}$ to 6.09 $mg \cdot d^{-1}$ in Experiment 1 and from 4.15 $mg \cdot d^{-1}$ to 6.37 $mg \cdot d^{-1}$ in Experiment 2. Expired breath samples, taken at a point just above the animal's nostrils, were collected in vacuum canisters with a volume of 1.7 L for calves and 2.5 L for yearling and in-calf heifers; the canisters were evacuated to over 900 mbar (1 mbar = 100 Pa) prior to use. The sample flow rate was adjusted (reduced) by crimping a short length of stainless steel tube and including it within the approximately 1 m length of Teflon/PVC/silicone sampling tubing. Measured flow rates were between 0.25 $mL \cdot min^{-1}$ and 0.35 $mL \cdot min^{-1}$ for calves and between 0.35 $mL \cdot min^{-1}$ and 0.45 $mL \cdot min^{-1}$ for yearling and in-calf heifers. Canisters were removed after 24 h and pressurized to ~500 mbar with N_2 gas, prior to gas chromatography (GC) (Varian 3600 GC; Varian Inc., Palo Alto, California, USA) analysis of CH_4 and SF_6 concentrations in the breath samples, which was performed as described by Muñoz et al. [8]. Concentrations of SF_6 and CH_4 in the ambient air were determined daily in samples captured by a canister that was placed close to, but upwind of, each experimental paddock.

These values were taken into account when calculating CH_4 emissions from each animal. Methane emissions from all animals were estimated in four successive 24 h collection periods in the final week of each experiment.

2.4. Calculation of grazed grass intake

Grazed grass dry matter (DM) intake ($kg\cdot d^{-1}$) for individual animals was estimated using the calculated ME intake ($MJ\cdot d^{-1}$) divided by the ME concentration in grazed grass, as determined by near-infrared reflectance analysis [10]. The ME intake was calculated as the sum of ME requirements for maintenance (ME_m) and growth (ME_g), the activity allowance for grazing and, where appropriate, the ME requirement for pregnancy (ME_p). The ME_m was derived using equations developed by Jiao et al. [5]. The activity allowance for grazing was estimated using equations from the Agricultural and Food Research Council (AFRC) [13]. The ME_g was calculated using the net energy requirement for growth (NE_g, $MJ\cdot d^{-1}$; Eq. (1)) and the efficiency of ME use for growth (k_g; Eq. (2)) from the AFRC [13].

$$NE_g = \frac{\left(1.15\times\left(4.1+0.0332\times LW-0.000009\times LW^2\right)\right)}{\left(1-0.1475\times\Delta LW\right)} \tag{1}$$

$$k_g = 0.78\times\left(ME/GE\right)+0.006 \tag{2}$$

where LW is live weight, ΔLW is daily live weight gain ($kg\cdot d^{-1}$) calculated from a linear regression of live weight against time, and ME and GE are the ME and GE concentrations ($MJ\cdot kg^{-1}$ DM), respectively, in grazed grass.

The energy used for pregnancy was calculated using Eqs. (3) and (4), along with the efficiency of ME use for pregnancy (0.133), as recommended by the AFRC [13].

$$\log E_t = 151.665-151.64\exp\left(-0.0000576t\right) \tag{3}$$

$$E_c = 0.025\times W_c\times\left(E_t\times 0.0201\exp\left(-0.0000576t\right)\right) \tag{4}$$

where E_c is the energy retention for pregnancy ($MJ\cdot d^{-1}$), E_t is the total energy retention (MJ), W_c is the calf birth weight [13], and t is the number of days from conception.

2.5. Statistical analysis

Data were analyzed as a one-way analysis of variance, with animal groups as the treatment factor in each experiment. For each case, if this analysis proved to be significant, Fisher's least significant difference test was used to assess pair-wise differences between the different treatments (age groups). Linear regressions were also performed to develop relationships between CH_4 emissions and DM intake or live weight, or between CH_4 energy output (CH_4-E) and GE intake or ME intake, using all data from Experiments 1 and 2, and with grazing season as a random effect.

3. Results

3.1. Nutritive value of the grazed grass

Table 1 presents the chemical composition of the fresh grass that was available to the grazing cattle in Experiments 1 and 2. The quality of the grazed grass that was obtained in the present study was typical of that commonly observed in dairy farms in Northern Ireland. The CP concentrations ranged from 160 $g\cdot kg^{-1}$ DM to 235 $g\cdot kg^{-1}$ DM in Experiment 1, and a slightly larger range was observed in Experiment 2. The mean, minimum, and maximum values of WSC concentration in Experiment 1 (early-mid grazing season) were higher than those in Experiment 2 (mid-late grazing season). However, the mean ADF and NDF concentrations were similar in the two experiments, although the minimum values in Experiment 1 were smaller than those in Experiment 2.

Table 1
Chemical composition of fresh grass.

	Mean	SD	Minimum	Maximum
Experiment 1				
Dry matter ($g\cdot kg^{-1}$)	161	31.8	110	219
Ash ($g\cdot kg^{-1}$ DM)	93	6.4	84	102
Gross energy ($MJ\cdot kg^{-1}$ DM)	18.5	0.30	18.1	19.0
Crude protein ($g\cdot kg^{-1}$ DM)	179	27.0	160	235
Acid detergent fiber ($g\cdot kg^{-1}$ DM)	233	21.8	199	266
Neutral detergent fiber ($g\cdot kg^{-1}$ DM)	490	38.8	417	544
Lipid ($g\cdot kg^{-1}$ DM)	42	6.8	35	52
Water-soluble carbohydrates ($g\cdot kg^{-1}$ DM)	169	26.2	139	213
Experiment 2				
Dry matter ($g\cdot kg^{-1}$)	145	22.0	112	215
Ash ($g\cdot kg^{-1}$ DM)	108	19.7	81	137
Gross energy ($MJ\cdot kg^{-1}$ DM)	18.5	0.30	18.0	19.0
Crude protein ($g\cdot kg^{-1}$ DM)	207	36.7	151	249
Acid detergent fiber ($g\cdot kg^{-1}$ DM)	247	8.0	237	261
Neutral detergent fiber ($g\cdot kg^{-1}$ DM)	492	26.9	454	549
Lipid ($g\cdot kg^{-1}$ DM)	35	4.2	31	40
Water-soluble carbohydrates ($g\cdot kg^{-1}$ DM)	117	34.2	60	162

3.2. Effects on live weight and feed intake

Live weight and feed intake data from Experiments 1 and 2 are presented in Table 2. As expected, live weight increased as heifer age increased ($P < 0.001$) in both experiments. Although live weight gain was similar among the three treatment groups in Experiment 1, this variable reduced linearly with increasing age of animals in Experiment 2 ($P < 0.001$). As a result, calculated feed intake (DM, GE, and ME) increased significantly with increasing age of animals ($P < 0.001$) in both experiments, although the differences between yearling and in-calf heifers in Experiment 2 did not reach the significant level.

3.3. Effects on enteric methane emissions

Methane emission data from Experiments 1 and 2 are presented in Table 3. In Experiment 1, yearling heifers had the highest CH_4 emissions ($g\cdot d^{-1}$) and in-calf heifers produced more CH_4 than calves ($P < 0.001$). When expressed as CH_4 emissions per unit of live weight or DM intake, or CH_4-E as a proportion of GE or ME intake, yearling heifers had higher emission rates than calves and in-calf heifers ($P < 0.001$). In Experiment 2, CH_4 emissions ($g\cdot d^{-1}$) increased linearly with increasing animal age ($P < 0.001$), although the difference between yearling and in-calf heifers was not significant. The CH_4/live weight ratio was lower in in-calf heifers than in the other two groups ($P < 0.001$), while CH_4-E as a proportion of GE or ME intake was lower in calves than in yearling and in-calf heifers ($P < 0.05$).

3.4. Relationship between CH_4 emissions and live weight and feed intake

Data obtained for calves, yearling heifers, and in-calf heifers in Experiments 1 and 2 were pooled and used to explore relationships between CH_4 emissions and live weight, and feed and energy intakes. A series of prediction equations (Eqs. (5)–(8) in Table 4) were developed using pooled data from the two experiments for all three groups of heifers ($n = 72$). Fig. 1 and Fig. 2 illustrate the relationships

Table 2
Effects of heifer age groups on live weight and feed intake in Experiments 1 and 2.

	Heifer age group			SE	P
	Calves (5–10 months)	Yearlings (12–17 months)	In-calf (18–23 months)		
Experiment 1					
Live weight (kg)	217^a	404^b	514^c	12.61	< 0.001
Live weight gain ($kg\cdot d^{-1}$)	1.05	1.11	0.99	0.064	0.445
DM intake ($kg\cdot d^{-1}$)	5.37^a	8.79^b	10.21^c	0.394	< 0.001
GE intake ($MJ\cdot d^{-1}$)	101^a	161^b	186^c	7.3	< 0.001
ME intake ($MJ\cdot d^{-1}$)	62^a	99^b	114^c	4.5	< 0.001
Experiment 2					
Live weight (kg)	246^a	411^b	520^c	17.12	< 0.001
Live weight gain ($kg\cdot d^{-1}$)	0.82^c	0.59^b	0.38^a	0.053	< 0.001
DM intake ($kg\cdot d^{-1}$)	5.34^a	6.95^b	7.91^b	0.370	< 0.001
GE intake ($MJ\cdot d^{-1}$)	100^a	127^b	145^b	6.8	< 0.001
ME intake ($MJ\cdot d^{-1}$)	60^a	77^b	88^b	4.1	< 0.001

[a,b,c] indicate that values in the same row with different superscripts are significantly different ($P < 0.05$).

Table 3
Effect of heifer age groups on enteric methane emissions in Experiments 1 and 2.

	Heifer age groups			SE	P
	Calves (5–10 months)	Yearlings (12–17 months)	In-calf (18–23 months)		
Experiment 1					
CH_4 emissions ($g\cdot d^{-1}$)	98^a	189^c	172^b	5.6	< 0.001
CH_4/live weight ($g\cdot kg^{-0.75}$)	1.71^a	2.10^b	1.60^a	0.054	< 0.001
CH_4/DM intake ($g\cdot kg^{-1}$)	18.5^a	21.7^b	17.1^a	0.74	< 0.001
CH_4-E/GE intake ($MJ\cdot MJ^{-1}$)	0.055^a	0.066^b	0.052^a	0.0022	< 0.001
CH_4-E/ME intake ($MJ\cdot MJ^{-1}$)	0.089^a	0.107^b	0.084^a	0.0037	< 0.001
Experiment 2					
CH_4 emissions ($g\cdot d^{-1}$)	106^a	155^b	169^b	5.3	< 0.001
CH_4/live weight ($g\cdot kg^{-0.75}$)	1.72^b	1.72^b	1.56^a	0.029	< 0.001
CH_4/DM intake ($g\cdot kg^{-1}$)	19.9	22.8	21.8	0.81	0.052
CH_4-E/GE intake ($MJ\cdot MJ^{-1}$)	0.059^a	0.069^b	0.066^b	0.0025	0.016
CH_4-E/ME intake ($MJ\cdot MJ^{-1}$)	0.098^a	0.114^b	0.109^{ab}	0.0041	0.025

[a,b,c] indicate that values in the same row with different superscripts are significantly different ($P < 0.05$).

Table 4
Prediction equations for methane emissions of Holstein-Friesian heifers.

Equations[a]	R^2	P	Eq. No.
Using data from calves, yearling heifers, and in-calf heifers in both Experiments 1 and 2 ($n = 72$)			
$CH_4 = 0.252_{(0.020)}\ LW + 50.92_{(9.96)}$	0.682	< 0.001	5
$CH_4 = 14.94_{(1.28)}\ DM\ intake + 36.77_{(11.08)}$	0.651	< 0.001	6
$CH_4\text{-}E = 0.046_{(0.004)}\ GE\ intake + 1.93_{(0.63)}$	0.639	< 0.001	7
$CH_4\text{-}E = 0.075_{(0.007)}\ ME\ intake + 1.93_{(0.66)}$	0.630	< 0.001	8
Using data from calves only in both Experiments 1 and 2 ($n = 24$)			
$CH_4 = 0.340_{(0.023)}\ LW + 23.23_{(5.37)}$	0.910	< 0.001	9
$CH_4 = 13.80_{(1.31)}\ DM\ intake + 27.89_{(8.31)}$	0.780	< 0.001	10
$CH_4\text{-}E = 0.041_{(0.004)}\ GE\ intake + 1.54_{(0.46)}$	0.783	< 0.001	11
$CH_4\text{-}E = 0.066_{(0.006)}\ ME\ intake + 1.57_{(0.49)}$	0.743	< 0.001	12
Using data from yearling heifers only in both Experiments 1 and 2 ($n = 24$)			
$CH_4 = 0.244_{(0.054)}\ LW + 72.61_{(28.068)}$	0.253	< 0.001	13
$CH_4 = 10.40_{(2.467)}\ DM\ intake + 89.51_{(21.028)}$	0.579	< 0.001	14
$CH_4\text{-}E = 0.032_{(0.007)}\ GE\ intake + 4.94_{(1.157)}$	0.582	< 0.001	15
$CH_4\text{-}E = 0.052_{(0.012)}\ ME\ intake + 4.92_{(1.15)}$	0.585	< 0.001	16

[a] Values in parentheses are the SE of the coefficients or constants; units are $g\cdot d^{-1}$ for CH_4 (methane emissions), kg for LW (live weight), $kg\cdot d^{-1}$ for DM intake, and $MJ\cdot d^{-1}$ for CH_4-E, GE, and ME intake.

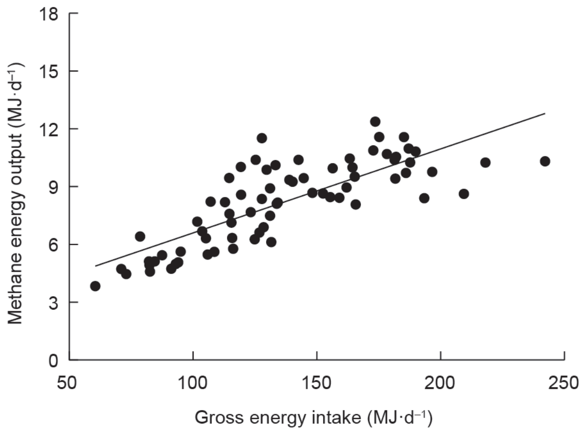

Fig. 1. The relationship between gross energy intake and CH_4 energy output for all three groups of Holstein-Friesian heifers.

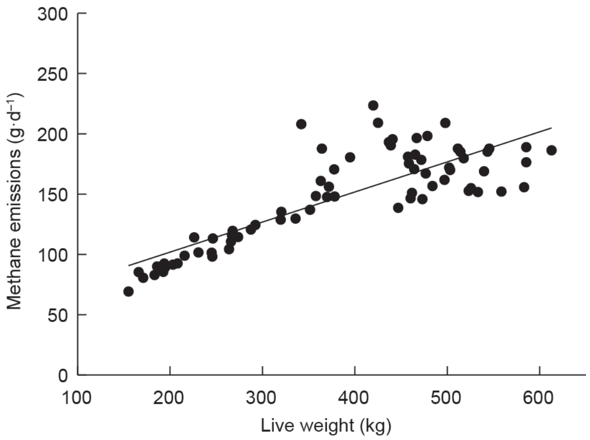

Fig. 2. The relationship between live weight and CH_4 emissions for all three groups of Holstein-Friesian heifers.

between CH_4-E and GE intake, and between CH_4 emission and live weight, respectively. All relationships were significant ($P < 0.001$), with R^2 from 0.630 to 0.682. These equations indicate that CH_4 emissions could be increased by 0.252 $g{\cdot}d^{-1}$ with an increase of 1 kg live weight or by 14.94 $g{\cdot}d^{-1}$ with an increase of 1 $kg{\cdot}d^{-1}$ of DM intake. They also indicate that CH_4-E could be increased by 0.046 $MJ{\cdot}d^{-1}$ or 0.075 $MJ{\cdot}d^{-1}$, respectively, with an increase of 1 $MJ{\cdot}d^{-1}$ of GE or ME intake.

Similar relationships were also developed using pooled data from the two experiments for groups of calves ($n = 24$) and yearling heifers ($n = 24$), respectively. These results are presented in Table 4. In general, the R^2 values of the relationships for the calf group (Eqs. (9)–(12)) are greater than those obtained when using the whole dataset of all three groups (Eqs. (5)–(8)), while the corresponding values for the yearling heifer group (Eqs. (13)–(16)) are smaller than those obtained for all three groups or for the calf group. There was no significant relationship between CH_4 emissions and live weight, DM intake, GE intake, or ME intake for the in-calf heifer group, so those results are not presented here.

4. Discussion

4.1. Effects of age and physiological state of heifers on enteric methane emissions

Enteric CH_4 emissions from UK dairy cattle are currently estimated using the Tiers 1 and 2 emission factors from the Intergovernmental Panel on Climate Change (IPCC) [14,15]. It is important to note that values for the proportion of energy lost as CH_4 from adult dairy cows are currently used when calculating CH_4 emissions from young stock. Because young stock are estimated to produce approximately 20% of the total enteric CH_4 emissions from the UK dairy sector [16], improving the accuracy of evaluating the dietary energy that is lost as CH_4 from adult dairy cows at specific developmental stages is desirable, as is the development of prediction equations for CH_4 emissions based on GE and DM intakes.

Until relatively recently, the significance of CH_4 emissions in ruminant livestock agriculture was considered largely in terms of the wasteful loss of dietary energy and the associated impact on the efficiency of dietary energy utilization, with a mean value of 0.065 as the CH_4-E/GE intake proposed by the IPCC [14]. In the current study, the average treatment mean ratios of CH_4-E/GE intake across all heifer ages ranged between 0.052 and 0.069. These values are similar to those for confined lactating dairy cows, as measured using calorimeter chambers [3,17], and to those for confined heifers and steers aged 6–22 months [5]. Furthermore, the CH_4 data from the present study are comparable to data that have been obtained recently from grazing cattle using the SF_6 tracer technique. For example, Jiao et al. [18] reported that CH_4-E/GE intake decreased from 0.059 to 0.053 for grazing Holstein dairy cows that were offered concentrates at 2–8 $kg_{DM}{\cdot}d^{-1}$—a range that was only marginally lower than that found in the present study. Cavanagh et al. [19] reviewed CH_4/DM intake ratios from 698 grazing Jersey-Friesian dairy cows and obtained a mean value of 18.2 $g{\cdot}kg^{-1}$, which lies within the range (17.1–22.8 $g{\cdot}kg^{-1}$) obtained in the present study. For beef heifers grazing on high and low herbage masses, Boland et al. [20] found that herbage mass had no significant effect on CH_4/DM intake ratios (19.3 $g{\cdot}kg^{-1}$ vs. 21.1 $g{\cdot}kg^{-1}$), for which the average value (20.2 $g{\cdot}kg^{-1}$) was very close to the mean value of 20.3 $g{\cdot}kg^{-1}$ obtained in the present study. However, SF_6 estimates of CH_4 data should be interpreted with caution as there is evidence that the SF_6 tracer technique is more likely to produce errors. Such errors include the risk of a non-linear decline in the SF_6 release rate from permeation tubes, the effects of permeation tube calibration temperature and recipient animal intra-ruminal temperature [21], and the need to accurately measure ambient background gas concentrations [22].

In the current study, the calculated GE intake increased progressively from calves through yearling heifers to in-calf heifers in both experiments. The in-calf heifers ate more DM but produced lower ratios of CH_4/DM intake, CH_4-E/GE intake, and CH_4-E/ME intake than the yearling heifers, although the difference was significant only in Experiment 1. Changes in rumen function and in the kinetics of the passage of diet components through the rumen help to explain some of the differences in CH_4 emissions that were observed between young, juvenile, and adult dairy stock. Johnson KA and Johnson DE [23] suggested that the rate of digesta flow affected the amounts of CH_4 generated, with higher feed intake promoting an increased rumen passage rate and, subsequently, a lower CH_4-E/GE intake. Bannink et al. [24] argued that the prediction of CH_4 emissions should not solely focus on accommodating the effects of nutrition on overall digestion and apparent feed utilization by cows, but should also consider the effects of nutrition on intra-ruminal fermentation conditions and, consequently, on the formation of volatile fatty acids and on the rumen hydrogen generation-utilization balance.

In the present study, all animals grazed on pasture with no concentrate supplements offered; however, enteric CH_4 emissions were different when analyzed independently for developmental age of heifer. This finding suggests that for animals on similar grazed grass, enteric emissions can be influenced by additional factors such as animal age or other, as yet unclear, sources of individual animal variation. Boadi et al. [25] and Grainger et al. [7] each observed considerable within-animal and between-animal variations in CH_4 production from cattle (dairy cows and beef cows, respectively) that, in each case, received the same or similar type of diet. Although the CH_4-E/GE intake ratios that were obtained in the present two experiments were within the levels published by the IPCC [14], 404 trials with Holstein cows in the United States [26] generated a wider range of CH_4-E/GE intake ratios (from 0.016 to 0.099), while Yan et al. [17], working at this institute, reported accumulated values from 0.037 to 0.101 for 247 UK Holstein-Friesian cows. These data further highlight the large variation that exists in CH_4-E/GE intake ratios, and suggest a need for further investigation of the effects of age and stage of development of the target animal, its physiological states, and the impact of diet characteristics, when evaluating CH_4 emissions.

4.2. Prediction of enteric methane emission

Data from the current study were used to develop a range of prediction equations for CH_4 emission by young dairy (Holstein-Friesian) stock. In line with previous studies [27–29], the current data supports a strong relationship between CH_4 emissions and DM intake, which may be linked to an increase in the availability of fermentable substrate [30]. Yan et al. [31] and Mills et al. [32] generated prediction equations for dairy cows using models that correlated nutrient intake and CH_4 emissions. Yan et al. [31] developed a supplementary equation using live weight and milk yield as CH_4 co-predictors in adult dairy cows; this equation has facilitated CH_4 emission predictions on commercial farms where intake data is not readily available.

The equations derived from the current study provide a means to estimate CH_4 emissions by heifers at different developmental stages and ages. In essence, the linear relationship between live weight and CH_4 emission reflects an increase of 0.252 $g \cdot d^{-1}$ CH_4 for an increase of 1 kg of heifer live weight. Zhao et al. [33] also found a significant relationship between live weight and CH_4 emissions in sheep that were offered fresh grass-only diets.

Holstein-Friesian replacement heifer-rearing programs in the United Kingdom and Northern Ireland typically aim to achieve a near-constant rate of growth, with heifers reaching a target first-calving weight of 540–560 kg at 24 months of age [34]. This strategy presumes a breeding age of 13.5–15 months [34]. The current study provides approaches to predict CH_4 emissions from grazing heifers using the animal's live weight as the determinant. These equations, together with the relationships that have been established between CH_4 emissions, feed DM, and energy intake, add useful information to the scientific literature; they can be used to estimate CH_4 emissions for grazing heifers, and thereby help to improve the accuracy of the national CH_4 emission inventories for cattle production systems.

5. Conclusions

The current study found that the CH_4-E from grazing dairy herd replacement heifers, when expressed as a proportion of GE intake, ranges from 0.052 $MJ \cdot MJ^{-1}$ to 0.066 $MJ \cdot MJ^{-1}$ in the early grazing season and from 0.059 $MJ \cdot MJ^{-1}$ to 0.069 $MJ \cdot MJ^{-1}$ in the late grazing season. This result compares with the single value of 0.065 $MJ \cdot MJ^{-1}$ that is recommended by the IPCC [14]. Relying only on the fixed IPCC [14] value to calculate CH_4 emissions from grazing young dairy stock may introduce significant error when assimilating data for strategic and policy considerations. The present data were used to develop a range of prediction equations for CH_4 emissions, which provide an alternative approach to estimate CH_4 emissions for grazing dairy heifers where actual CH_4 emission data are not available.

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Compliance with ethics guidelines

Steven J. Morrison, Judith McBride, Alan W. Gordon, Alastair R. G. Wylie, and Tianhai Yan declare that they have no conflict of interest or financial conflicts to disclose.

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