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Effect of concentrate feed level on methane emissions from grazing dairy cows

H. P. Jiao,* A. J. Dale,* A. F. Carson,* S. Murray,* A. W. Gordon,† and C. P. Ferris*[1]
*Agri-Food and Biosciences Institute, Hillsborough, Co. Down BT26 6DR, United Kingdom
†Agri-Food and Biosciences Institute, Belfast, Co. Antrim BT9 5PX, United Kingdom

ABSTRACT

Although the effect of nutrition on enteric methane (CH_4) emissions from confined dairy cattle has been extensively examined, less information is available on factors influencing CH_4 emissions from grazing dairy cattle. In the present experiment, 40 Holstein-Friesian dairy cows (12 primiparous and 28 multiparous) were used to examine the effect of concentrate feed level (2.0, 4.0, 6.0, and 8.0 kg/cow per day; fresh basis) on enteric CH_4 emissions from cows grazing perennial ryegrass-based swards (10 cows per treatment). Methane emissions were measured on 4 occasions during the grazing period (one 4-d measurement period and three 5-d measurement periods) using the sulfur hexafluoride technique. Milk yield, liveweight, and milk composition for each cow was recorded daily during each CH_4 measurement period, whereas daily herbage dry matter intake (DMI) was estimated for each cow from performance data, using the back-calculation approach. Total DMI, milk yield, and energy-corrected milk (ECM) yield increased with increasing concentrate feed level. Within each of the 4 measurement periods, daily CH_4 production (g/d) was unaffected by concentrate level, whereas CH_4/DMI decreased with increasing concentrate feed level in period 4, and CH_4/ECM yield decreased with increasing concentrate feed level in periods 2 and 4. When emissions data were combined across all 4 measurement periods, concentrate feed level (2.0, 4.0, 6.0, and 8.0 kg/d; fresh basis) had no effect on daily CH_4 emissions (287, 273, 272, and 277 g/d, respectively), whereas CH_4/DMI (20.0, 19.3, 17.7, and 18.1 g/kg, respectively) and CH_4-E/gross energy intake (0.059, 0.057, 0.053, and 0.054, respectively) decreased with increasing concentrate feed levels. A range of prediction equations for CH_4 emissions were developed using liveweight, DMI, ECM yield, and energy intake, with the strongest relationship found between ECM yield and CH_4/ECM yield (coefficient of determination = 0.50). These results demonstrate that offering concentrates to grazing dairy cows increased milk production per cow and decreased CH_4 emissions per unit of milk produced.

Key words: concentrate feed, dairy cattle, methane, grazing

INTRODUCTION

As concerns about climate change grow, international pressure to reduce greenhouse gas (**GHG**) emissions is increasing. For example, within the European Union (**EU**), legislation requires member states to reduce total GHG emissions by 20% (from 1990 levels) by 2020 (European Commission, 2010) and the United Kingdom (**UK**) Climate Change Act (UK Office of Public Sector Information, 2008) sets a target of an 80% reduction (from 1990 levels) by 2050.

Agriculture is known to be a significant source of GHG, with CH_4, N_2O, and CO_2 being the 3 main GHG emitted from the agricultural sector. In 2011, the EU agriculture sector produced 461,012 kt of CO_2 equivalents, representing approximately 10% of the total EU GHG emissions (European Environment Agency, 2012). With regard to CH_4, the global livestock sector is responsible for 37% of all human-induced CH_4 emissions, with 89% of these livestock-derived emissions arising from enteric fermentation (Steinfeld et al., 2006). It is important to have a clear understanding of factors influencing enteric CH_4 emissions from ruminant livestock if accurate GHG inventories and appropriate mitigation strategies are to be developed.

Although data and prediction equations describing CH_4 emissions from confined dairy cows have been extensively published (Ellis et al., 2007; Yan et al., 2010), much less information is available on CH_4 emissions from grazing cattle. This may reflect, in part, the challenges faced when measuring CH_4 emissions from grazing cattle. However, in many temperate regions dairy cows spend between 5 and 9 mo of the year grazing, and as such, emissions during this period represent a significant part of their annual emissions.

Evidence from cows offered confinement diets indicate that although total CH_4 emissions increase with

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[1]Corresponding author: conrad.ferris@afbini.gov.uk

increasing concentrate feed levels (Ferris et al., 1999b; Schils et al., 2006; Patel et al., 2011; Aguerre et al., 2011), emissions per liter of milk produced generally decrease (Yan et al., 2010). However, much less evidence exists concerning the effect of concentrate feed level on CH_4 emissions from grazing cows. In one of the few published studies, Lovett et al. (2005) measured CH_4 emissions from grazing cows offered either 1.0 or 6.0 kg/d of a fiber-based concentrate and found that whereas CH_4 production per kilogram of milk was unaffected by concentrate supplementation, CH_4 production per kilogram of FCM decreased with increasing concentrate feed level. In a more recent study involving 3 concentrate feed levels (2.0, 4.5, and 7.0 kg/cow per day), Young and Ferris (2011) observed that both daily CH_4 emissions and CH_4 production per liter of milk were unaffected by concentrate feed level. Nevertheless, this latter study was limited in scale and was undertaken over a short time period. No study appears to have investigated CH_4 emissions from grazing dairy cows offered a range of concentrate feed levels over a substantial period of time. Thus, the objective of the current study was to examine CH_4 emissions from grazing dairy cows offered a wider range of concentrate feed levels than in previous studies. In seeking to achieve this objective, we recognized that having sufficient cows on each concentrate level to provide adequate replication would be extremely difficult in view of the significant labor requirements associated with using the SF_6 technique (Johnson et al., 1994), especially in relation to the daily changing of canisters and gas analysis. To overcome this difficulty, this experiment adopted 4 measurement periods and the use of a repeated measures analysis to provide robust data capable of allowing the effects of concentrate level to be examined.

MATERIALS AND METHODS

This experiment was conducted at the Agri-Food and Biosciences Institute (Hillsborough, Co. Down, UK; 54°27′N; 06°04′W) during 2011.

Animals and Experimental Design

This study involved 40 Holstein-Friesian dairy cows [12 primiparous and 28 multiparous (mean parity, 2.4 ± 1.23 SD)], with cows having a mean PTA for milk yield and fat + protein yield of 238 (±136.6 SD) kg and 24.6 (±8.90 SD) kg, respectively, and a mean Profitable Lifetime Index (PLI) of £91 (±46.6 SD). Cows were selected from the Agri-Food and Biosciences Institute dairy herd, and were within the top 1% of the Holstein-Friesian population within the UK in terms of PLI. Cows had a mean pre-experimental milk yield of 31.6 (±6.53 SD) kg/d, and were a mean of 131 DIM (range = 30–240 d) at the start of the experiment.

Cows were allocated to 1 of 4 treatment groups 1 wk before the experiment start date (balanced for pre-experimental milk yield, calving date, and lactation number), with groups then randomly allocated to 1 of 4 treatments. Treatments comprised 4 concentrate feed levels (2.0, 4.0, 6.0, and 8.0 kg/cow per day; fresh basis). Concentrates were offered in the milking parlor during milking, with the daily allowance split between 2 equal feeds. The ingredient composition of the concentrate offered (g/kg, fresh basis) was as follows: soybean hulls: 187, maize: 160, wheat: 150, soybean meal: 125, rape meal: 90, molasses: 70; distillers grains: 60, wheat feed pellets: 57, citrus pulp: 40; Megalac (Volac Ltd., Orwell, UK): 20, limestone ($CaCO_3$): 11, calcined magnesite: 10, palm oil blend: 7.5, salt: 7.5, and trace minerals and vitamins: 5. The number of replicates per treatment was derived from a power calculation (for a power of 0.8), as the number of replicates required to identify a significant difference ($P = 0.05$) within any one period in CH_4/milk yield (g/kg; this being identified as one of the most important variables within the study), between treatments. This calculation was based on an assumed difference between the treatments of 3.5 g/kg, and an estimated variance within the data set of 8.0, the latter derived from the actual variance observed within data from a similar study by Vance et al. (2011). Although not accounted for within this calculation, the improved power associated with an experiment involving repeated measures was demonstrated by Vlaming (2008).

Grazing and Grassland Management

Cows grazed perennial ryegrass (*Lolium perenne*)-based swards throughout this experiment, with the 40 experimental cows part of a larger group of 88 cows. This group commenced grazing on April 13, 2011, and were grazing full time by April 19, with all cows offered 8.0 kg of concentrate/cow per day during this transition period. Over the following 14-d period, concentrate feed levels for all cows within each group were gradually adjusted to the designated treatment levels (2.0, 4.0, 6.0, or 8.0 kg/cow per day). Cows were split into their 4 experimental groups on April 27 (22 cows/group), with the experiment beginning on May 4. The grazing period continued until September 30, 2011.

Throughout the experiment each of the 4 treatment groups grazed separately, with the 4 groups grazing in close proximity to each other. A flexible rotational grazing system was adopted in which each treatment group was given access to fresh herbage daily, following the p.m. milking. A key grazing management target was that pre- and postgrazing sward heights were similar with all treatments (target of 5.5 cm), with this relatively high postgrazing sward height chosen so that herbage intakes would not be restricted with any treatment. This residual sward height was initially targeted by providing daily herbage allowances of approximately 16, 14, 12, and 10 kg of DM/cow per day for concentrate treatments 2.0, 4.0, 6.0, and 8.0 kg/d, respectively. However, these herbage allowances were reduced as the study progressed, and adjustments in daily herbage allowances were made between treatments, so as to maintain the target residual sward heights.

Target herbage allocations were achieved by making daily adjustments to the sizes of the areas grazed by each treatment group, based on pregrazing sward heights. Sward heights were measured daily (pre- and postgrazing) throughout the experimental period using a rising plate meter (Jenquip folding plate pasture meter; Jenquip, Feilding, New Zealand), with 20 sward-height measurements being taken at random in a "W" formation across the area designated for grazing within each treatment. The mean aboveground pre- and postgrazing herbage masses for each of the grazing areas were then estimated using the following linear equation:

$$\text{Herbage mass (kg of DM/ha)} = [\text{sward height (cm)} \times 316] + 330.$$

The target postgrazing residual sward height was 5.5 cm for all treatments, which, based on the equation above, represents a target residual herbage mass (above ground level) of 2,068 kg of DM/ha. Once the area of pasture necessary to achieve the required herbage allowance with each treatment group was determined, required paddock sizes for each treatment group were calculated and paddocks were established using temporary electrified fences. Fences were removed after each grazing, and reestablished before the next grazing, with paddock size determined by the grazing herbage mass before each grazing. Cows completed 6 grazing rotations during the course of the experiment, with approximately 35, 30, 30, 30, 25, 25, and 25 kg of N/ha (in the form of calcium ammonium nitrate) applied following each of these grazing cycles.

Measurements

Cow Performance. Throughout the study cows were milked twice daily, between 0600 and 0800 h, and between 1600 and 1800 h, with individual milk yields recorded automatically at each milking. During each CH_4 measurement period (described below), milk fat, protein, and lactose concentrations were determined at each milking using a MilkoScan milk analyzer (model FT 120; Foss UK Ltd., Warrington UK). Cow BW was recorded after every milking, with a mean BW calculated for each week. Condition score was assessed weekly using a 5-point scale as described by Edmonson et al. (1989; 1 = emaciated; 5 = extremely fat).

CH_4. Enteric CH_4 emissions were recorded during 4 separate measurement periods using the SF_6 technique (Johnson et al., 1994). During period 1 (June 6 to 9), emissions were recorded during 4 consecutive 24-h periods, whereas during each of periods 2 to 4 (June 26 to 30, July 31 to August 4, and September 4 to 8, respectively) emissions were recorded during 5 consecutive 24-h periods. The permeation tubes used within this experiment contained approximately 2.4 g of SF_6 gas at filling (March 2011) and were incubated at 39°C until they were orally administered to cows on May 27 (10 d before the first experimental period). Prior to this, the release rate of SF_6 from each permeation tube was determined via weekly gravimetric weighing over an 8-wk period to produce an 8-point regression curve ($R^2 > 0.999$). The mean release rate of SF_6 from the permeation tubes at the start of the experiment was 5.40 (±0.82 SD) mg/d. To correct for the known decline in the rate of release of SF_6 from the permeation tubes within the rumen during the course of the experiment, 10 surveillance tubes, with similar release rates as the tubes used in the experiment, were maintained at 39°C in an incubator and monitored weekly until 6 wk after the completion of the final measurement period. The release rate of SF_6 from the surveillance tubes was found to decline by 0.15% per day, and this value was subsequently used to adjust the release rates for the experimental tubes during periods 2, 3, and 4, as described by Lassey et al. (2001).

On d 1 of each of the 4 measurement periods, each cow was restrained within a head-locking gate, at approximately 1400 h (before the p.m. milking), and fitted with a halter (to support the nose piece and CH_4 sampling line) and a polyvinyl chloride (2.5-L) collection canister. The equipment used was described by Johnson et al. (2007), with the exception of the capillary tubing, which was modified as described below. Prior to exiting the gate, the sample line was connected to the collection canister (which had previously been evacuated to over 90 kPa) using a quick-connect fitting (Swagelok Co., Solon, OH), thus allowing a gas sample to be drawn up into the evacuated canister at a rate of between 0.6 to 0.7 mL/min (approximately). This flow rate was regulated via a length (approximately 5.0 cm) of capillary tubing (0.102 mm i.d.; Alltech Associates Applied Science Ltd., Lancashire, UK) that had been crimped at several points. Crimping was part of the calibration process necessary to achieve the correct flow rate, with flow rate measured using a digital flow meter (Cole-Parmer Instrument Co., Vernon Hills, IL). The gas sample drawn into the evacuated canister was collected from the area around the cow's nostrils, and contained eructated gas (a mixture of normal atmospheric gases, SF_6, and CH_4).

At approximately 1400 h on the following day, cows were returned to the head-locking gate and the sample line was removed from the canister. Used canisters were replaced with new canisters on d 2 to 5 (d 2 to 4 for period 1) of each measurement period, whereas on d 6 (d 5 for period 1), both the halter and canister were removed. On each day, used canisters were subsequently charged with N (a carrier gas) to a pressure of approximately 50 kPa, and analyzed for concentrations of SF_6 and CH_4 via gas chromatography, as described by Johnson et al. (2007), using a Varian 3600 gas chromatograph (Varian Inc., Palo Alto, CA). Background (ambient) concentrations of gases were measured during each 24-h period using 4 evacuated canisters attached to sample lines. Two of these were placed at each end of the block of paddocks being grazed by the experimental cows, with the open end of the sample line located approximately 40 cm above ground level.

Feed Sampling and Analysis

During each CH_4 measurement period, herbage pluck samples were taken daily from within the areas being grazed by each of the experimental groups (at 20 random locations, at a height of approximately 5.0 cm above ground level). One subsample from the grazing area for each treatment was analyzed for ME content by near-infrared reflectance spectroscopy using the methodology described by Park et al. (1998) for grass silage, but using a calibration equation developed for fresh grass. The remainder of the sample was dried for 48 h at 60°C for DM determination and the daily dried samples subsequently bulked for each period. Bulked samples were analyzed for concentrations of N, ADF, NDF, water-soluble carbohydrates (**WSC**), ash, and gross energy (**GE**). The concentrate offered during the study was sampled weekly, with weekly samples bulked for each 4-wk period. Concentrate samples were subsequently analyzed for DM, N, ADF, NDF, GE, and ash concentrations. The feedstuffs offered were analyzed as described by Ferris et al. (1999a), with the exception of N, which was analyzed using the Dumas method (Jung et al., 2003).

Herbage Intake

During each CH_4 measurement period, herbage DMI for each cow was estimated from performance data using the back-calculation approach. This was deemed appropriate, as the work was funded to provide data to help improve the UK GHG inventory, and the ability to be able to relate emissions to the calculated energy requirement of the national herd was recognized as a possible strategy by which to scale up emissions to a national level. During each measurement period milk energy content was determined from daily milk composition data using the equation of Tyrrell and Reid (1965), whereas mean daily BW change during each measurement period was determined by linear regression of weekly BW data for a 5-wk period (2 wk before measurement, the week of measurement, and 2 wk after measurement). Total energy required for maintenance, production, tissue change, pregnancy (where appropriate), and activity was determined using the equations contained within "Feed into Milk," the UK dairy cow feed rationing system (Agnew et al., 2004). The ME content of the concentrates offered was calculated as 13.0 MJ/kg of DM [based on published values for individual ingredients according to "Feed into Milk" (Agnew et al., 2004) as implemented in FeedByte software (Scotland's Rural College, Edinburgh, UK)], whereas the ME content of the herbage grazed was determined using near-infrared reflectance spectroscopy, as already described.

Statistical Analysis

Three cows in period 1 (one from each of the 4.0, 6.0, and 8.0 kg of concentrate treatments), 1 cow in period 2 (from the 6.0 kg of concentrate treatment), and 1 cow in period 3 (from the 6.0 kg of concentrate treatment) were excluded from the statistical analysis due to missing CH_4 data. Seven cows (2, 2, and 3 from the 2.0, 6.0, and 8.0 kg of concentrate treatments, respectively) were dried off before the period-4 measurement period, and were excluded from statistical analysis.

Methane data [CH_4, CH_4/DMI, CH_4/ECM, and CH_4/ME intake (**MEI**)] within each period were initially analyzed using ANOVA. Data on animal performance, feed intake, milk production, and CH_4 emissions across periods 1 to 4 were then combined and analyzed using REML analysis. The mixed model used included the following terms as fixed effects: constant + lactation number + DIM + period + concentrate level + period × concentrate level, whereas cow numbers within period were fitted as random effect. Correlations between time points were modeled using a power model.

Regression relationships between CH_4 and BW, DMI, MEI, and GE intake (**GEI**), and ECM were developed using REML analysis based on individual cow data within all periods. In these models, lactation number and DIM were fitted as fixed effects, and cow number was fitted as a random effect. The coefficient of determination values were estimated from pseudo coefficient of determination values using the square of the correlation between fitted values and observed values. Data were analyzed using GenStat 14.2 (Lawes Agricultural Trust, Rothamsted Research, Harpenden, UK). Differences between treatments were tested at $P < 0.05$ using least significant difference.

RESULTS

Chemical Composition of Feedstuffs and Sward Heights

The concentrate offered had a CP, NDF, ADF, and ash concentration of 206 (±2.4 SD), 302 (±39.8 SD), 174 (±13.3 SD), and 90 (±1.9 SD) g/kg of DM, respectively; a GE content of 18.1 (±0.1 SD) MJ/kg of DM; and a calculated ME content of 13.0 MJ/kg of DM. The chemical composition (mean of the 4 CH_4 measurement periods) of the herbage offered with the 2.0, 4.0, 6.0, and 8.0 kg/d concentrate treatments was as follows: DM: 154 (±13.9 SD), 156 (±15.1 SD), 151 (±11.6 SD), and 154 (±12.8 SD) g/kg, respectively; CP: 229 (±10.5 SD), 237 (±21.7 SD), 248 (±20.4 SD), and 220 (±20.2 SD) g/kg of DM, respectively; ME: 11.8 (±0.26 SD), 11.8 (±0.31 SD), 11.7 (±0.30 SD), and 11.8 (±0.25 SD) MJ/kg of DM, respectively; NDF: 436 (±12.9 SD), 427 (±26.1 SD), 435 (±23.2 SD), and 436 (±25.3 SD) g/kg of DM, respectively; ADF: 215 (±10.7 SD), 206 (±14.7 SD), 206 (±8.4 SD), and 209 (±9.8 SD) g/kg of DM, respectively; WSC: 133 (±28.9 SD), 129 (±21.3 SD), 128 (±29.6 SD), and 127 (±31.3 SD) g/kg of DM, respectively. Averaged across the 4 CH_4 measurement periods, mean pre- and postgrazing sward heights were 12.0 (±1.70 SD) and 6.0 (±0.77 SD) cm, 11.8 (±1.36 SD) and 6.0 (±0.65 SD) cm, 11.9 (±2.07 SD) and 5.6 (±0.57 SD) cm, and 11.9 (±1.33 SD) and 5.3 (±0.61 SD) cm, for the 2.0, 4.0, 6.0, and 8.0 kg of concentrate treatments, respectively.

Effect of Concentrate Feed Level on BW, Feed Intake, and Milk Production

Whereas BCS ($P = 0.14$) and milk fat content ($P = 0.70$) were unaffected by period, milk protein content increased ($P = 0.049$). In contrast, grass DMI, total DMI, MEI, milk yield, ECM yield, fat-plus-protein yield, milk energy output ($P < 0.001$), BW ($P = 0.018$), and BW change ($P = 0.003$) all decreased with period.

With the exception of BW change ($P = 0.032$), no period × concentrate level interactions were detected for any of the cow performance measures presented in Table 1 ($P > 0.1$), and consequently only the main effects of concentrate level are presented. Concentrate feed level had no significant effect on BW ($P = 0.21$), BW change ($P = 0.085$), and BCS ($P = 0.48$; Table 1). Concentrate feed level had a significant effect on grass DMI ($P < 0.001$) and total DMI ($P = 0.007$), with cows offered 2.0 kg of concentrate consuming 4.4 kg/d additional herbage DM, on average, compared with those on the 8.0 kg of concentrate treatment. Total MEI was affected by treatment ($P < 0.001$), with cows on the 2.0 and 4.0 kg of concentrate treatments having significantly lower MEI than those on the 6.0 and 8.0 kg of concentrate treatments ($P < 0.05$). Milk yield, milk fat + protein yield, ECM yield, and milk energy output increased ($P < 0.001$) with increasing concentrate feed level (Table 1). Both milk fat ($P < 0.001$) and milk protein concentrations ($P = 0.003$) decreased with increasing concentrate feed levels. The ECM/DMI ($P < 0.001$) and milk energy yield/MEI ($P < 0.001$) were lowest with cows offered the 2.0 kg of concentrate treatment. Values for BW change (kg/d) for the 2.0, 4.0, 6.0, and 8.0 kg/d concentrate treatments were −0.27, −0.54, −0.07, and −0.19 (period 1); 0.25, 0.12, 0.03, and −0.17 (Period 2); 0.12, −0.19, 0.03, and −0.29 (period 3); and −0.30, −0.43, −0.22, and 0.12 (period 4), respectively.

Effect of Concentrate Feed Level on CH_4 Emissions

The effect of concentrate level within period is presented in Figure 1, with CH_4 production (g/d) unaffected by concentrate level in any of periods 1 to 4, whereas CH_4/DMI decreased with increasing concentrate feed level in period 4 ($P = 0.010$) and CH_4/ECM decreased ($P = 0.048$ and $P = 0.062$) with increasing concentrate feed level in periods 2 and 4, respectively. Methane energy output (**CH_4-E**), as a proportion of MEI (CH_4-E/MEI), was unaffected by treatment in any of periods 1 to 3 ($P > 0.05$), but decreased with concentrate feeding in period 4 (Figure 1). Nevertheless, comparing the effect of period on CH_4 production was not a primary objective of this study due to the relatively small number of cows per treatment, and period effects being confounded by stage of lactation, herbage quality, and weather conditions during each measurement period. Nevertheless, CH_4 production, CH_4/DMI, CH_4/GEI ($P < 0.001$), and CH_4/MEI ($P = 0.012$) tended to decrease with period, whereas CH_4/milk yield ($P = 0.49$) and CH_4/ECM yield ($P = 0.54$) did not change with period. However, no period × concentrate level interactions existed for any of the CH_4 data examined ($P > 0.1$); the main effects of concentrate level are presented in Table 2. Concentrate feed level had no effect ($P = 0.52$) on daily CH_4 emissions (g/d), whereas CH_4/DMI ($P = 0.005$), CH_4-E/MEI ($P < 0.001$), and CH_4-E/GEI

Table 1. Effect of concentrate feed level on BW, BCS, feed intake, and milk production of grazing dairy cows (mean of the 4 CH_4 measurement periods)

	Concentrate level (kg/d)					
Item	2.0	4.0	6.0	8.0	SED[1]	P-value
BW (kg)	577	552	565	570	12.4	0.21
BW change[2] (kg/d)	−0.05	−0.26	−0.06	−0.13	0.097	0.085
BCS	2.4	2.5	2.4	2.5	0.08	0.48
Grass DMI (kg/d)	12.8^a	10.8^b	10.3^b	8.4^c	0.42	<0.001
Total DMI (kg/d)	14.5^a	14.2^a	15.5^b	15.4^b	0.42	0.007
Forage proportion (kg/kg of DM)	0.88^d	0.75^c	0.66^b	0.54^a	0.011	<0.001
ME intake (MJ/d)	174^a	173^a	189^b	190^b	5.01	<0.001
Milk yield (kg/d)	19.6^a	22.4^b	25.9^c	26.5^c	0.87	<0.001
Milk fat (g/kg)	45.5^c	41.5^b	36.8^a	36.5^a	1.42	<0.001
Milk protein (g/kg)	35.6^b	34.0^a	34.0^a	33.6^a	0.56	0.003
Milk lactose (g/kg)	43.7	44.4	44.0	44.5	0.30	0.051
Milk fat + protein yield (kg/d)	1.55^a	1.75^b	2.02^c	2.06^c	0.066	<0.001
ECM yield (kg/d)	21.1^a	22.8^b	24.8^b	25.1^c	0.75	<0.001
Milk energy output (MJ/d)	64.2^a	69.5^b	75.3^c	76.5^c	2.27	<0.001
Milk yield/DMI (kg/kg)	1.35^a	1.57^b	1.66^c	1.71^c	0.043	<0.001
ECM yield/DMI (kg/kg)	1.46^a	1.59^b	1.59^b	1.63^b	0.035	<0.001
Milk energy yield/ME intake (MJ/MJ)	0.37^a	0.40^b	0.40^b	0.40^b	0.0086	<0.001

[a–d] Means with different superscript letters within a row are different ($P < 0.05$).

[1] SED = SE of the difference.

[2] Estimated by linear regression of weekly BW data for a 5-wk period (2 wk before measurement, the week of measurement, and 2 wk after measurement).

($P = 0.015$) decreased with increasing concentrate feed levels.

Regression Relationships on CH_4 Emissions

Regression relationships describing CH_4 emissions are presented in Table 3. All relationships were significant ($P < 0.001$), with coefficient of determination values ranging from 0.15 to 0.50. Methane emissions based on these relationships were 18.5 g/kg of DMI and 11.5 g/kg of ECM, when the constant was omitted from the relationship. The strongest relationship was identified between ECM and CH_4/ECM (equation 6 in Table 3; $R^2 = 0.50$).

DISCUSSION

It has been comprehensively documented that when grazing cows are offered concentrate supplements, pasture DMI usually decreases, whereas total DMI and milk yield increases (Bargo et al., 2003). Whereas the primary objective of the current study was not to examine the milk yield response to concentrate supplementation (due to the relatively small number of cows per treatment), the responses observed were largely as described by Bargo et al. (2003). Moving from the 2.0 to the 8.0 kg of concentrate treatments, herbage DMI decreased by 4.4 kg/d (a mean herbage substitution rate of 0.84 kg/kg of concentrate DMI), whereas total DMI increased by 0.9 kg/d. Similarly, milk yield increased with concentrate feeding, with the mean daily response between the 2 extreme concentrate levels being 6.9 kg of milk and 4.0 kg of ECM. The latter represents a milk yield response of 0.67 kg of milk/kg of concentrate, with this within the range of published responses (Journet and Demarquilly, 1979; Bargo et al., 2003). The decreasing marginal milk yield response to concentrate supplementation was again in agreement with the findings of studies summarized by Stockdale et al. (1987) and Bargo et al. (2003), and reflects the decreasing marginal MEI response with increasing concentrate feed levels.

Although many calorimeter studies have examined CH_4 emissions from confined cows offered conserved forage-based diets, fewer studies have examined emissions from grazing dairy cows. Methane emissions from grazing cattle are normally measured using the SF_6 technique, with a recent study by Muñoz et al. (2012) demonstrating that the technique can provide estimates of CH_4 emissions that are comparable with direct measurements undertaken within respiration calorimeters. Nevertheless, several possible sources of error associated with technique are recognized. These include the nonlinear decline in SF_6 release from permeation tubes (corrected for in this study), effects of permeation tube calibration temperature and animal temperature (Deighton et al., 2014), and accurate measurement of background gas concentrations (Williams et al., 2011).

A numbers of studies have used the SF_6 technique to measure CH_4 emissions from grazing dairy cows during

Table 2. Effect of concentrate feed level on CH_4 emissions from grazing dairy cows (mean of the 4 CH_4 measurement periods)

	Concentrate level (kg/d)					
Item[1]	2.0	4.0	6.0	8.0	SED[2]	*P*-value
CH_4 (g/d)	287	273	272	277	11.1	0.52
CH_4 energy (MJ/d)	16.0	15.2	15.2	15.4	0.62	0.52
CH_4/DMI (g/kg)	20.0^{c}	19.3^{bc}	17.7^{a}	18.1^{ab}	0.72	0.005
CH_4/milk yield (g/kg)	15.4^{c}	12.9^{b}	11.2^{a}	10.8^{a}	0.66	<0.001
CH_4/ECM (g/kg)	14.1^{c}	12.5^{b}	11.4^{ab}	11.1^{a}	0.56	<0.001
CH_4-E/GE intake (MJ/MJ)	0.059^{c}	0.057^{bc}	0.053^{a}	0.054^{ab}	0.0021	0.015
CH_4-E/ME intake (MJ/MJ)	0.093^{b}	0.089^{b}	0.081^{a}	0.082^{a}	0.0031	<0.001

a–cMeans with different superscript letters within a row are different ($P < 0.05$).

[1] CH_4-E = CH_4 energy; GE = gross energy.

[2] SED = SE of the difference.

the last decade, with Robertson and Waghorn (2002), Ulyatt et al. (2002), Lovett et al. (2005), Cavanagh et al. (2008), McCourt et al. (2008), O'Neill et al. (2010), and Wims et al. (2010) recording average emissions of 18.5, 13.1, 19.4, 23.9, 17.9, 12.9, and 15.8 g of CH_4/kg of milk produced, respectively, and 21.3, 16.1, 18.7, 18.2, 36.6, 18.1, and 19.3 g of CH_4/kg of DMI, respectively. Across the 4 concentrate levels used in the current study, mean CH_4 emissions per kilogram of milk and per kilogram of DMI were 12.6 and 18.8 g, respectively. Although these

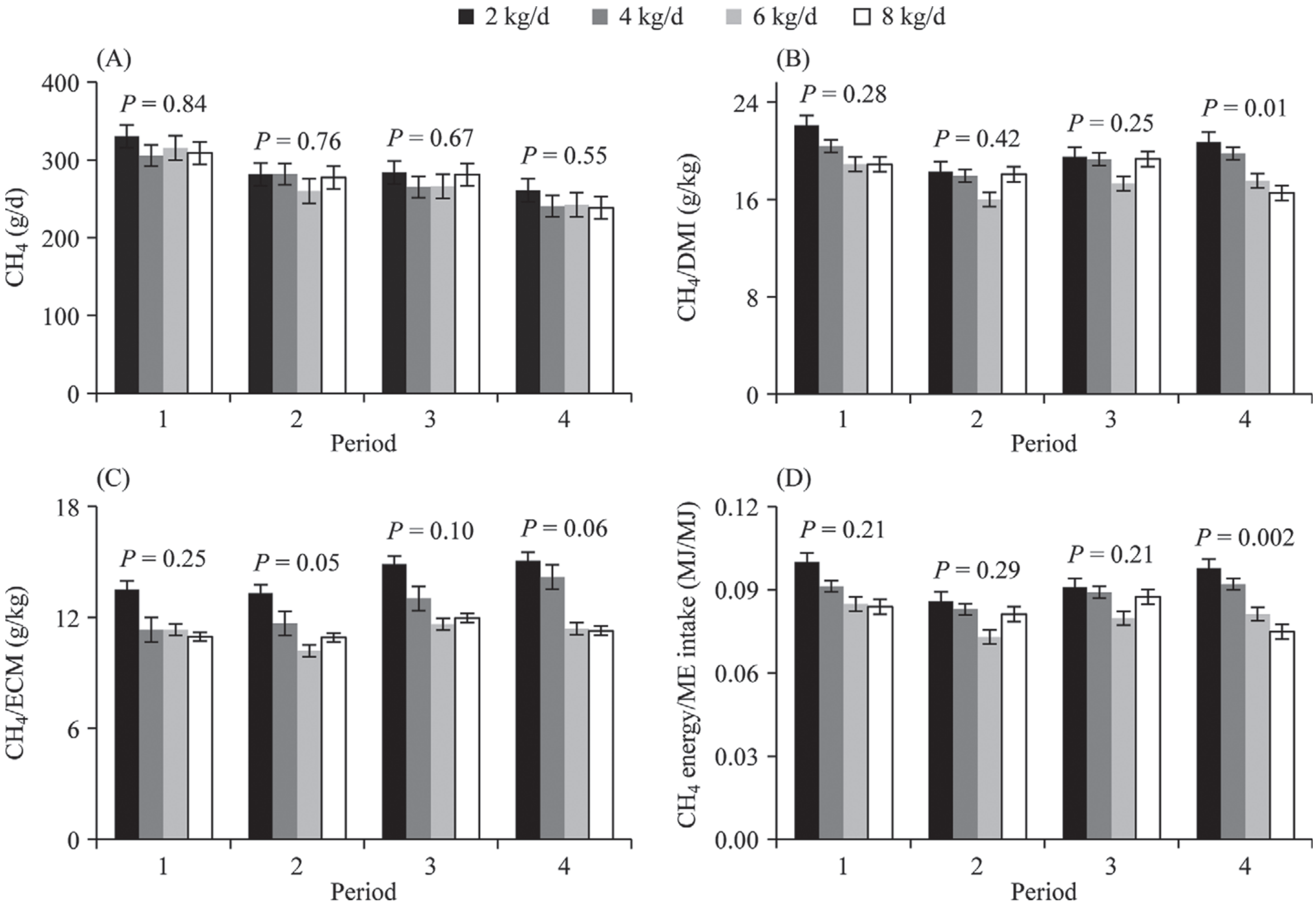

Figure 1. Effect of concentrate feed level on daily CH_4 emissions (A), CH_4/DMI (B), CH_4/ECM (C), and CH_4 energy/ME intake (D) within each of the 4 CH_4 measurement periods. Error bars represent standard errors.

Table 3. Regression relationships describing CH_4 emissions from grazing dairy cows (based on combined data from all 4 CH_4 measurement periods)[1]

Equation no.	Equation[2]	R^2
1	CH_4 (g/d) = $0.348_{(0.1095)}$BW (kg) + $163.0_{(53.73)}$	0.15
2	CH_4 (g/d) = $9.51_{(2.039)}$DMI (kg/d) + $136.3_{(42.49)}$	0.23
3	CH_4 (g/d) = $6.02_{(1.076)}$ECM (kg/d) + $103.0_{(43.79)}$	0.21
4	CH_4-E (MJ/d) = $0.028_{(0.0061)}$GE intake (MJ/MJ) +$7.7_{(2.36)}$	0.23
5	CH_4-E (MJ/d) = $0.048_{(0.0091)}$ME intake (MJ/MJ) + $6.7_{(2.36)}$	0.25
6	CH_4/ECM (g/kg) = $-0.314_{(0.0512)}$ECM (kg/d) + $17.8_{(2.08)}$	0.50

[1] Values in subscript parentheses are SE data.

[2] CH_4-E = CH_4 energy; GE = gross energy.

values largely agree with many of the values presented above, the studies cited involved cows at different lactation stages, producing a wide range of milk yields, and grazing swards of very different compositions. All of these factors can influence CH_4 emissions.

Although most published studies have been designed to provide basic CH_4 emission values from grazing cattle, few have examined the effect of different management strategies on CH_4 emissions from dairy cows grazing temperate pastures. For example, relatively little is known about the effect of concentrate feed level on CH_4 emissions from grazing dairy cattle. However, in one recent study, Lovett et al. (2005) measured CH_4 emissions from grazing cows offered either 1.0 or 6.0 kg/d of a fiber-based concentrate, and found that total CH_4 production (346 and 399 g/cow per day, respectively) increased with increasing concentrate supplementation, CH_4 production per kilogram of milk produced (21.0 and 17.7 g/kg, respectively) was unaffected (although tending to decrease), and CH_4 production per kilogram of FCM decreased (19.3 and 16.0 g/cow per day, respectively). In another study involving 3 concentrate feed levels (2.0, 4.5, and 7.0 kg/cow per day), Young and Ferris (2011) observed that neither daily CH_4 production (252, 263, and 262 g/cow) nor CH_4 production per kilogram of milk produced (20.1, 17.9, and 16.0 kg/kg) were affected by concentrate feed level, although the latter tended to decrease.

In the present study, which involved a wider range of concentrate levels, a larger number of cows, and 4 measurement periods, daily CH_4 emissions ranged from 272 to 287 g/d, and in common with the findings of Young and Ferris (2011), were unaffected by concentrate supplementation. However, as ample evidence exists that DMI is a key driver of CH_4 emissions from animals offered confined diets (Ellis et al., 2007), concentrate feeding, which normally results in an increase in total DMI (as observed in the current study) might have been expected to increase daily CH_4 emissions. That this did not occur is likely to reflect the decreasing forage proportion in the diet as concentrate feed level increased. Enteric CH_4 production is associated primarily with production of acetic acid and butyric acid and, in general, the fermentation of predominantly forage diets results in a higher molar proportion of acetic acid than occurs with concentrate-based diets (Ørskov and Ryle, 1990). Conversely, concentrate-based diets normally contain greater proportions of more readily fermentable components that favor propionate production during rumen fermentation, with a consequent reduction in CH_4 production per unit of fermentable OM in the rumen (Johnson and Johnson 1995). In addition, the more rapid fermentation associated with concentrate-based diets tends to result in a lower rumen pH, and this will also inhibit the growth of methanogens and protozoa (Hegarty, 1999). These different rumen fermentation patterns in the current study were clearly reflected in the decrease in milk fat levels with increasing concentrate supplementation, with rumen acetate production in the lower concentrate treatments being a precursor of milk fat (Cozma et al., 2013). Thus, although intakes increased with increasing concentrate feed levels (with higher CH_4 emissions expected), this was offset by the increasing quality of the diet offered (and associated lower emissions), with the overall result being no effect of diet on total daily emissions.

In studies involving confined cows, ample evidence exists that CH_4 emissions per kilogram of milk produced declines with increasing concentrate feed levels. For example, in a meta-analysis of 986 dietary treatments, Huhtanen and Hetta (2012) reported a highly significant and positive relationship between dietary concentrate intake and production of milk and ECM. This effect is likely due to a reduction in emissions per kilogram of DMI associated with improved diet quality, combined with a dilution (due to higher milk yields) of CH_4 emissions associated with DMI required to maintain the cow. Similarly, in the current study, CH_4/milk yield decreased with the increasing levels of concentrate supplementation (15.4, 12.9, 11.2, and 10.8 g of CH_4/kg of milk, with concentrate levels of 2.0, 4.0, 6.0, and 8.0 kg/cow per day, respectively); this is in agreement with the findings of Lovett et al. (2005) for FCM. The higher emissions per kilogram of milk in the latter study is likely to reflect the fact that cows were in late lactation (an average of 251 DIM at the start of CH_4 measurements), whereas in the current study, cows were, on average, 160, 182, 217, and 238 DIM at the start of each CH_4 measurement period, respectively. The effect of this on CH_4 emissions per kilogram of milk produced is 2-fold: first, average milk yield in the current study was 17.4% higher than in the study by Lovett et al. (2005) and, as such, in their study, a greater proportion of total CH_4 emissions was derived from food consumed to meet the cows maintenance requirements, thus giving an higher apparent CH_4 emission per kilogram of milk produced. In addition, data for BW change in the current study indicates that, on average, across all treatments, cows were losing BW, suggesting mobilization of body tissues for milk production. Energy derived from mobilized body tissue can replace dietary energy in supporting milk production, without associated CH_4 production, thus reducing apparent CH_4 emissions per kilogram of milk produced. In contrast, although BW change data were not presented by Lovett et al. (2005), the late-lactation cows in that study were likely to have been using some of the food consumed for body tissue gain, and this would have resulted in higher apparent CH_4 emission value per kilogram of milk produced.

The decline in CH_4-E/GEI with decreasing forage proportion has been long established. For example, Flatt et al. (1969) observed a linear decrease in CH_4-E/GEI (0.054, 0.044, and 0.038) when lactating dairy cows were offered diets containing decreasing proportions of alfalfa (0.60, 0.40, and 0.20, respectively), whereas Tyrrell and Moe (1972) observed that CH_4-E/GEI was reduced from 0.064 to 0.051 when the concentrate proportion in the diet was increased from 0.31 to 0.59. More recently, in a study involving lactating dairy cows offered grass silage-based diets, Ferris et al. (1999b) observed a linear reduction in CH_4-E/GEI (from 0.071 to 0.062) when the concentrate proportion was increased from 0.37 to 0.70, whereas in a review of published data for confined cows, Yan et al. (2000) observed that a 0.10 increase in the forage proportion of the diet increased CH_4-E/GEI by 0.0025 (average CH_4-E/GEI = 0.068). In agreement, in the present study, CH_4-E/GEI decreased from 0.059 to 0.054 when the forage proportion decreased from 0.88 to 0.54, although the actual values for CH_4-E/GEI in the current study were slightly lower than those reported by Ferris et al. (1999b) and Yan et al. (2000). This difference is likely due in part to the fact that the current study involved fresh grass-based diets, whereas these latter studies involved grass silage-based diets. Grass silage generally has higher fiber content (Roca-Fernández et al., 2012) and lower WSC content (O'Kiely et al., 1988; Narasimhalu et al. 1989) than fresh grass, which results in a significantly higher acetate:propionate ratio compared with the grass diets. In addition, the SF_6 technique does not include CH_4 emissions from the rectum, and this may also contribute to the lower values for CH_4-E/GEI compared with measurements undertaken within respiratory chambers. In a recent study, Muñoz et al. (2012) reported the SF_6 measurements for grazing cattle should be increased by 3% to take account of CH_4 emissions from the rectum.

Although CH_4 production can be measured using the SF_6 technique within a research environment, the technique is time consuming and expensive. One of the ultimate objectives of experiments such as the current one will be to provide data that will allow the development of equations by which CH_4 emissions can be predicted for grazing cows. To allow the accurate development of such equations, a meta-analysis of data from a range of studies will be required. However, in the current study, a range of prediction equations for CH_4 emissions were developed, with these based on BW, feed intake, ECM yield, and energy intake. The poorest relationship obtained was for BW ($R^2 = 0.15$), with previous studies involving confined cattle also indicating that BW was a poor predictor of CH_4 emissions (Holter and Young, 1992; Yan et al., 2009; Jiao et al., 2014). Slightly improved relationships were obtained for intake parameters (DM, GE, and ME: $R^2 = 0.23$–0.25), although these intake measurements were based on performance data, rather than direct measurements of grass intake. A strong negative regression relationship ($R^2 = 0.50$; equation 6 in Table 3) was established between CH_4/ECM and ECM, with this demonstrating that CH_4 emissions per unit of milk decreased with increasing milk production.

The results of the current study demonstrate that although total CH_4 emissions were unaffected by increasing concentrate feed levels, CH_4 emissions per kilogram of milk produced declined with concentrate feeding. Thus, provided it is accompanied by an increase in milk production, offering increasing concentrate levels to grazing cows provides a strategy to reduce emissions per kilogram of milk produced. However, from a climate change perspective, the impact of concentrate feeding on total GHG (CO_2, CH_4, and N_2O) emissions associated with the entire milk production system must be considered. This includes GHG emissions arising from growing, processing, and transporting the individual concentrate ingredients; the effect of higher milk yields on herd fertility and health, and their effects on herd survival; and possible savings in GHG emissions that may arise due to lower forage intakes (and associated low inputs for grass production) associated with increased concentrate feeding. These factors demonstrate the need to examine the effect of any change in a single component of a system, not just in terms of emissions of a single GHG at the local farm level, but via a full life-cycle-analysis approach.

CONCLUSIONS

The present study obtained CH_4 emission data for grazing dairy cows offered a range of concentrate levels. Whereas daily CH_4 emissions were unaffected by concentrate feeding, CH_4 emissions per kilogram of DMI and per kilogram of ECM decreased with increasing concentrate level. Thus, provided cows are able to produce a reasonable milk yield response to additional concentrates offered, concentrate supplementation can reduce CH_4 emissions per unit of milk produced. However, from a climate change perspective, the impact of concentrate feeding on total GHG emissions associated with the entire milk production system should be considered.

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