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Methane Emissions from Dairy Cows Measured Using the Sulfur Hexafluoride (SF_6) Tracer and Chamber Techniques

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ABSTRACT

Our study compared methane (CH_4) emissions from lactating dairy cows measured using the sulfur hexafluoride (SF_6) tracer and open-circuit respiration chamber techniques. The study was conducted using 16 lactating Holstein-Friesian cows. In each chamber, the cow was fitted with the SF_6 tracer apparatus to measure total CH_4 emissions, including emissions from the rectum. Fresh ryegrass pasture was harvested daily and fed ad libitum to each cow with a supplement of 5 kg of grain/d. The CH_4 emissions measured using the SF_6 tracer technique were similar to those using the chamber technique: 331 vs. 322 g of CH_4/d per cow. The accuracy of the SF_6 tracer technique was indicated by considering the ratio of the CH_4 emission measured using the SF_6 tracer to the emission measured using the chamber for each cow on each day. The calculated ratio of 102.3% (SE = 1.51) was not different from 100%. A higher variability within cow between days was found for the SF_6 tracer technique [coefficient of variation (CV) = 6.1%] than for the chamber technique (CV = 4.3%). The variability among cows was substantially higher than within cows, and was higher for the SF_6 technique (CV = 19.6%) than for the chamber technique (CV = 17.8%). Our CH_4 emission data were compared with whole-animal chamber studies conducted in Canada and Ireland. In the Canadian study the SF_6 technique did not measure CH_4 emissions from the rectum and emissions were 8% lower than those measured using the chamber, indicating that emissions from the rectum may be greater than previously measured (1%). The relationship between CH_4 emission and dry matter intake was examined for our data and for that reported in the Canadian study. There was a difference in the slopes of the regressions derived from our data and that from Canada; 17.1 vs. 20.8 g of CH_4/kg of dry matter intake. A difference between the 2 locations was expected based on the difference in diet composition for these 2 studies. The SF_6 tracer technique is reasonably accurate for inventory purposes and for evaluating the effects of mitigation strategies on CH_4 emissions.
Key words: chamber, dairy cattle, methane, sulfur hexafluoride

INTRODUCTION

There is a need to accurately measure enteric methane (CH_4) emissions from cattle because these emissions account for about 15% of global CH_4 emissions (Lassey et al., 1997). Methane is an important greenhouse gas having many times the global warming potential of CO_2 (IPCC, 2001). Methane emissions can be accurately measured by placing animals in sealed chambers with appropriate measures of gas flow and composition (Blaxter and Clapperton, 1965; Moe and Tyrrell, 1979); however, diets eaten by cows in chambers may differ from that selected by grazing animals (Clark, 2002). The majority of ruminants graze under extensive conditions, are free ranging, and select a variety of forage types. Their CH_4 production must be determined to calculate inventory. The sulfur hexafluoride (**SF_6**) tracer technique is often used to measure CH_4 emissions from grazing ruminants (Johnson et al., 1994; Lassey et al., 1997; Woodward et al., 2006), and although data appear to be defensible and repeatable, additional validation would provide a degree of certainty to CH_4 inventory.

Studies with beef cattle and sheep indicate that CH_4 estimated with the SF_6 tracer technique is 93 to 95% of that measured using whole-animal chambers (Johnson et al., 1994; Ulyatt et al., 1999; McGinn et al., 2006) and 105% of that measured using hood chambers (Boadi et al., 2002). The lower estimates using the SF_6 tracer technique are partly explained by the CH_4 released

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Table 1. Mean DMI, BW, BCS, and production of milk and milk composition for cows during the calorimetry period (n = 16)

Item	Mean	SD
DMI, kg/d		
Grain	4.4	0.53
Forage	13.8	2.71
BW, kg	496	57.5
BCS[1]	4.5	0.09
Milk yield, kg/d	22.3	3.78
Protein yield, kg/d	0.72	0.099
Protein, %	3.24	0.175
Fat yield, kg/d	0.91	0.146
Fat, %	4.08	0.451

[1]Based on an 8-point scale (Earle, 1976).

via the rectum (Murray et al., 1976). No comparisons between SF_6 and chamber techniques have been made with dairy cows at higher intakes and including rectal methane.

The objective of this study was to compare the SF_6 tracer gas technique to the chamber technique for measuring total enteric CH_4 emissions from lactating dairy cows. The use of the SF_6 tracer gas technique within the chambers enabled a direct comparison between the 2 techniques and included CH_4 both respired and released from the rectum.

MATERIALS AND METHODS

Cows and Experimental Design

Sixteen Holstein-Friesian cows were used to compare CH_4 emissions estimated using the respiration chamber and SF_6 tracer techniques. The cows were from the experimental herd at the Department of Primary Industries, Victoria, Ellinbank Research Centre (latitude 38°14′36.4″, longitude 145°56′09.5″) and were part of a larger study to evaluate the effects of monensin sodium on methanogenesis. They were normally pastured year round on a predominantly ryegrass sward and milked twice daily. Cows chosen for the experiment were of mixed age and had previously been trained to accept halters and headstall restraint. The experiment was conducted during November and December (late spring to early summer) 2005. Half of the cows had controlled-release monensin capsules placed into their rumens (Elanco Animal Health, Greenfield, IN) before starting the experiment. The effect of monensin on methane emissions will be reported elsewhere. Mean BW, BCS (according to the 8-point scale described by Earle, 1976), and milk yield and composition for cows used in the experiments are presented in Table 1.

Methane measurements were undertaken on pairs of cows (one with and one without a monensin slow-release capsule) over a 36-d period, with animals moved from the main herd to a metabolism facility and then placed in individual chambers. The cows were initially held in metabolism stalls for 4 d and each cow was fitted with a body harness and collection apparatus to enable separate collection of urine and feces. This adaptation period facilitated a smooth transition into the chambers where CH_4, intake, milk, feces, and urine production were measured over 3 d. There were 2 chambers and cows progressed through the experiment in pairs, 1 in each chamber. Every fourth day was reserved for cleaning and servicing the chambers. While one pair of cows was in the chambers, the next scheduled pair was brought into the metabolism stalls in readiness.

Animal Husbandry

Inside the chambers, cows were restrained by neck halters anchored to the floor. The harnessing device permitted feces and urine to be collected at the rear of the cow into separate collection vessels that were emptied each day. The apparatus allowed the cow to lie down on a padded mattress within the stall. Windows between the chambers enabled visual contact with the cow in the adjacent chamber and the surrounding environment.

Cows were milked twice daily using a portable milking apparatus in both the metabolism stalls and the chambers. Milking and feeding the cows in the chambers necessitated opening the chamber doors, thereby disrupting gas measurements. Milk weights were measured and subsampled into bronopol (0.5% wt/wt) preservative and analyzed for concentrations of fat and protein using a near-infrared milk analyzer (model 2000, Bentley Instruments, Chaska, MN).

The bulk of the cows' diet was fresh ryegrass pasture that was harvested daily. When cows were in the metabolism stalls or chambers, fresh pasture was placed in the feed bins twice daily to ensure ad libitum intake. When cows were in the chambers, the feed was provided while the chamber doors were open for milking. Cows also received 5 kg/d of cracked barley (as-fed basis) in 2 feedings. All feed offered and refused was weighed daily. Samples of feed and refusals were dried to determine DM content, and total daily DMI was calculated per cow (Table 1). Representative samples of the pasture and grain were collected daily and pooled to form 4 samples of each feed over the 32-d measurement period. The samples were oven dried and ground through a 0.5-mm sieve, then analyzed by near-infrared spectroscopy by a commercial laboratory (FeedTest, Hamilton, Victoria, Australia). The pasture contained 71.1 ± 1.2% apparently digestible DM, 16.4 ± 1.0% CP, and 54.4 ± 0.9% NDF, on a DM basis. The grain contained 85.8 ±

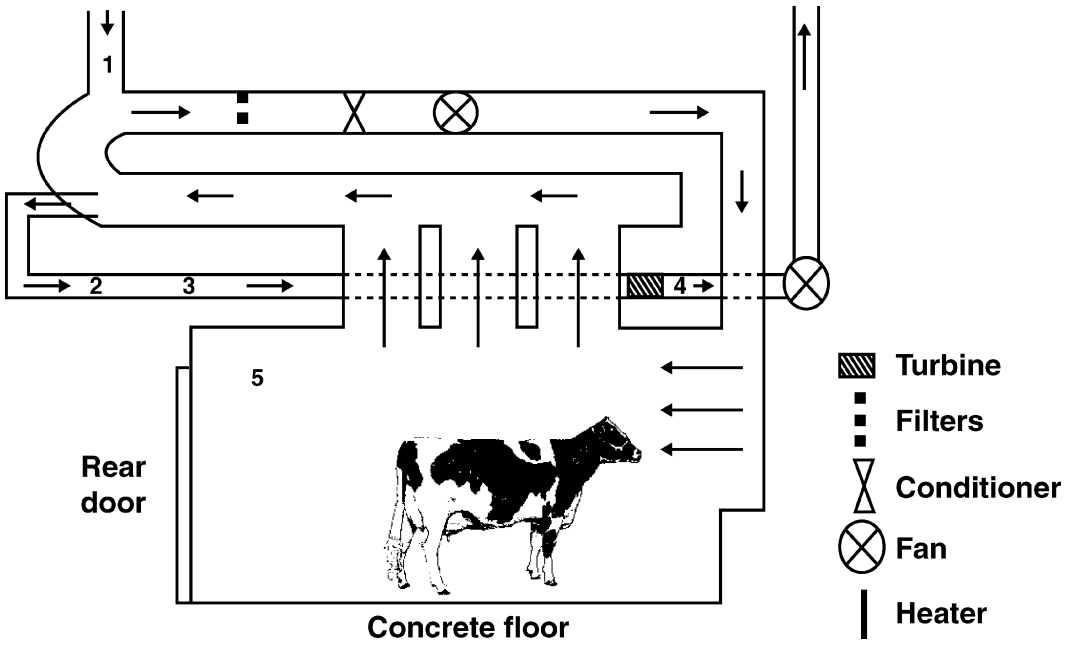

Figure 1. Schematic of the open-circuit respiration chambers located at the Department of Primary Industries, Ellinbank (Victoria, Australia) showing the airflow and conditioning, and release and sampling locations within the circulation system. Locations 1 and 2 are the intake and exhaust ducts sample points for noncalibration periods; location 3 is the injection point enabling the analytical system calibration; location 4 is the sample point for the system calibration; and location 5 denotes the chamber volume.

2.2% apparently digestible DM, 10.7 ± 0.3% CP, and 6.6 ± 0.8% ADF.

Chamber Design and Operation

Each chamber, located in an open barn, had a volume of 41.5 m^3, with inner surfaces of stainless steel, except for a concrete floor (Figure 1). The air within each chamber circulated independently of the other chamber at about 7.2 m^3/min. The circulated air was filtered and maintained at a set relative humidity (55% in this experiment), and temperature (20°C in this experiment). The condensed water from the dehumidifier was collected in an open container inside the chamber, allowing dissolved gases to vent back into the chamber. Air was recirculated and reentered the chamber at the front of the animal stall, with an exhaust rate of approximately 1 m^3/min, to the outside of the barn. The exhausted air stream first passed through straight polyvinyl chloride (**PVC**) duct (50 mm i.d.) to generate a laminar flow. Flow rate was measured using a turbine meter (SP2-CB-H7-A 4X, Sponsler Inc., Westminster, SC) and the data were recorded (DT800 data logger, Datataker Pty. Ltd., Rowville, Victoria, Australia) at 10-s intervals. The exhausted air was replaced with fresh air drawn in through a large-diameter PVC duct 40 m from the barn.

During the experiment, air was sampled continuously at the common intake duct and at each chamber's exhaust duct (locations 1 and 2 in Figure 1). The air samples were drawn using 3 dedicated diaphragm vacuum pumps with 16 L/min flow capacity (107CD18-198, Rietschle Thomas, Seven Hills, NSW, Australia). The sample streams were continuously dried in a dehumidifier (model LGD03-4, Tation, Frigematic Industries, Melbourne, Victoria, Australia) where the dew point was set to 2°C. The condensate was removed by providing a "controlled leak" with fine capillary tubing leading from the water traps.

Throughout the experiment, an automated calibration of the gas analyzer was conducted every 4 h by directing ultrapure N_2 (zero) and a standard gas mixture (span) through the analyzer. The span gas had similar gas concentrations to that expected in the exhaust samples from the chambers. Each gas passed through the gas analyzer twice (4 min × 2) and the concentration recorded at 10-s intervals. The timing of the zero and span gas checks and all other analyses were controlled by the data logger. The difference between the analyzed zero and span gas concentrations and the actual zero and span concentrations were used to calibrate the gas analyzer. A primary standard gas (supplied by CSIRO Atmospheric Research Division, Aspendale, Victoria, Australia) was used before the ex-

periment to confirm the concentration of the standard gas mixture. Any change (drift) in the gas analyzer was corrected daily, but the correction was always <1% over the duration of the experiment.

The air streams were connected to a gas analyzer (Xentra 4100C1, Servomex, East Sussex, UK) containing an infrared CH_4 sensor with a range of 0 to 1,000 ppm. Sequencing of the sample streams to the analyzer was controlled by solenoids to ensure a quick response time after switching the air stream. The CH_4 concentration, exhaust airflow rates, and relative humidity and temperature of the exhaust air were recorded at 10-s intervals over repeated 4-min periods. In each 12-min period, the intake air (common to both chambers) was sampled for 4 min, followed by the exhaust from chamber 1 for 4 min and then the exhaust from chamber 2 for the final 4 min. This routine was repeated every 12 min.

Chamber Emission Calculations

The measurement day for each chamber was from the time the door was closed in the morning, following milking, feeding, and the exchange of canisters used in the SF_6 tracer technique, to the following morning when the door of each chamber was opened. In the afternoon, the doors were opened briefly for the second milking and feeding. For each measurement day, 3 modes were identified to facilitate chamber emission calculations. Mode 1 was the duration following door closure when the chamber air was transient; that is, CH_4 concentration was increasing inside the chamber. Mode 2 was the duration of the measurement day that the chamber air was in a steady-state condition, and mode 3 was designated as the duration the doors were opened (afternoon). In mode 3, the CH_4 emissions were assumed to be equal to the previous CH_4 emissions just before the door was opened. For the remaining modes 1 and 2 when the doors were closed, the CH_4 emission (F; g) was determined from a storage component and a flow-through component. The storage component accounted for the transient condition of the air in the chamber (mostly during mode 1 just after the door was closed), while the flow-through component accounted for the loss of CH_4 from the chamber. The storage component (first bracketed term in equation 1) was determined from the volume of the chamber (V; m^3), the difference between the CH_4 chamber concentrations at time t (C_t; g/m^3) and the previous measurement (C_{t-1}; g/m^3), and the duration between sequential measurements (D). The flow-through component (second bracketed term in equation 1) was determined from the difference between the exhaust and inlet CH_4 concentrations (ΔC; g/m^3), the exhaust airflow (v; m^3/min), and the duration between sequential measurements (D). The individual emissions from each sequential measurement (during mode 1, 2, or 3) were then summed over each measurement day (from door closing to door opening next morning) to give the total emission during each measurement day:

$$F = [V\,(C_t - C_{t-1})] + [v\;\Delta C\;D]. \qquad [1]$$

The accumulated CH_4 emission (ΣF; g) over each measurement day was corrected using calibration data collected automatically every 4 h. While the calibration of the gas analyzer was being conducted, the chamber gas measurements were interrupted. However, the emission during each calibration was accounted for by adding the calibration duration to the first calculation of emissions following the calibration.

The CH_4 concentration (C; g/m^3) was calculated from the mixing ratio (*Ratio;* μmol/mol) given by the gas analyzer, using the ideal gas law:

$$C = Ratio\;MW\;1000\;\frac{P}{RT} \qquad [2]$$

where MW is the molecular weight of CH_4 (16), 1,000 is the conversion for L/m^3, P is the air pressure (atm), R is the gas constant (0.08205 L atm mol^{-1} K^{-1}), and T is the temperature (K). In this calculation, only the mean of the last 12 of the available 24 data values from each 4 min cycle was used, which ensured the previously sampled air was purged from the system.

Calibration of the Chambers

The accuracy of the chambers was checked before starting each period by comparing a known release of ultrapure (>99.9%) CH_4 (Linde Gases Pty. Ltd., Thomastown, Victoria, Australia) to that calculated using equation 1, once steady-state conditions were reached. The rate of gas release was controlled using a mass flow controller (MFC series 100, Sierra Instruments Inc., Monterey, CA).

The chamber calibrations involved 3 stages. In stage 1, the chambers were operated with no source of gas (empty chamber and no gas release). This allowed evaluation of any bias error in the analytical system. Stage 1 tested that the magnitude of the calculated emission was indeed zero when there was no emission inside the chamber, and therefore that there was no significant offset (bias) in measurement system.

In stage 2, the mass flow controllers were used to release CH_4 into the exhaust duct at location 3 in Figure 1. The inlet sample line was moved from location 1 to location 2 and the sample line normally at 2 was moved

to location 4. In this manner, we could perform a near-instantaneous evaluation of the entire analytical system that included the turbines, sample lines, connections, solenoid valves, pumps, gas analyzer, data recorder, and the calculation of the flux rates. It was expected that near-perfect recovery should result from this test, because leakage of gas along the exhaust duct was unlikely.

In stage 3, all sampling lines were reverted back to their normal positions. The release of CH_4 was switched from location 3 to location 5 (inside the chamber). This configuration allowed the whole chamber to be included in the recovery test. With stage 3, the time required to reach a new equilibrium concentration with a constant injection rate from the mass flow controllers was 90 min. A total of 8 calibrations using stages 1, 2, and 3 were made for both chambers to characterize the recovery rates.

SF_6 Tracer Technique for Respiratory CH_4 Measurement

The SF_6 tracer technique has been used extensively to measure CH_4 production from sheep and cows in grazing and confined situations in New Zealand (Woodward et al., 2006) and elsewhere. The technique requires air sampled around the nostrils to be accumulated in an evacuated PVC canister placed around the cow's neck (Johnson et al., 1994). Air is drawn continuously over a 24-h period into the canister through a tube positioned near the nostrils of the cow. The continuous airflow rate is controlled by passage through a capillary tube so that approximately 0.8 mL/min is accumulated over 24 h, after which the canister is replaced.

A permeation tube containing SF_6 was placed into the rumen of each cow about 3 mo before the measurements reported here. The permeation tubes were manufactured in December 2004 by the National Institute of Water and Atmospheric Research, New Zealand, and were filled with about 2.3 g of SF_6. The release rate of SF_6 was predetermined over a 10-wk period by weighing each permeation tube weekly; the average release of SF_6 was 3.7 ± 0.7 mg/d.

The concentration of CH_4 and SF_6 in the canisters was analyzed by gas chromatography (Shimadzu 2010, Shimadzu Corp., Kyoto, Japan and Hewlett-Packard 5890, Hewlett-Packard Labs, Palo Alto, CA), fitted with an electron capture detector (350°C) to determine SF_6, and a flame-ionization detector (250°C) to determine CH_4 concentration. All samples were analyzed in duplicate except standards, which were analyzed in triplicate. The gas chromatograph was fitted with a 3.3-m molecular sieve column with an i.d. of 0.32 mm and film thickness of 300 μm (Alltech Associates, Auckland, New Zealand). The column and injector temperatures were both 85°C but baked out at 200°C daily. Nitrogen was used as the carrier gas at a flow rate of 40 mL/min.

Three standards prepared by the National Institute of Water and Atmosphere (Wellington, New Zealand) were used to calibrate both gas chromatographs. Standards were mixtures of SF_6 and CH_4 in low, medium, and high concentrations (range: 15 to 1,000 ppt for SF_6; 2 to 200 ppm for CH_4). The standards were run at the beginning and end of each day with the medium standard run every 10 samples throughout the day. Gas concentrations (SF_6 and CH_4) were determined from peak areas and identified from their different retention times relative to the known standards.

Calculation of CH_4 emissions requires measurement of background SF_6 and CH_4 concentrations to represent inspired air, usually upwind from grazing cows. In this experiment there was no difference in either CH_4 or SF_6 concentration when measured from a canister hung inside the chamber to measure chamber air and a canister positioned on the cow's neck while in the chamber. The lack of difference was due to the low exchange of air in the chamber (1 m^3/min) so there was a build-up of gas in the chamber, and also because of the circulation within the chamber (7.2 m^3/min). Because the CH_4 and SF_6 concentrations in the chambers and the canisters were similar, the chamber inlet concentration (location 1 in Figure 1) was used as a background concentration for calculating emissions from SF_6 in equation 3. In this manner, the SF_6 tracer technique effectively measured the same emissions as the chamber, including those respired, eructated, and released through the rectum. This was unlike the situation for chambers with a high-volume exchange of air (e.g., McGinn et al., 2006), which resulted in much lower concentrations of CH_4 and SF_6 in the chamber compared with the canister on the cow. In this latter case, the chamber concentrations are used as background values and the SF_6 tracer technique does not measure the flux from the rectum.

The CH_4 emission (Q_{CH4}; g/d) was calculated using the SF_6 and CH_4 mixing ratio (μmol/mol) sampled by the canisters (C_{SF6} and C_{CH4}, respectively) on the cows and inlet air streams (C^b_{SF6} and C^b_{CH4}, respectively), and the predetermined SF_6 release rate (Q_{SF6}; g/d) from the permeation tube (equation [3]) where MW is the molecular weight of the gases:

$$Q_{CH4} = \frac{C_{CH4} - C^b_{CH4}}{C_{SF6} - C^b_{SF6}} Q_{SF6} \frac{MW_{CH4}}{MW_{SF6}}. \quad [3]$$

Statistical Analyses

The ratio of the total daily CH_4 emission measured using the SF_6 technique to that measured using the

chamber was calculated for each cow as a direct measure of agreement between the 2 methods. These ratio data were analyzed using a linear mixed model that included fixed effects for chamber and treatment (monensin vs. control), and linear covariates for DMI, permeation tube SF_6 release rate, and actual CH_4 emissions (g/d). Random effects included in the model were day (corresponding to the 3 d of measurements for each pair of cows), cow, and cow by day. Fixed effects were tested for significance in the model using sequential Wald tests. Random effects (components of variance) were tested using the χ^2 change in scaled deviance for nested models. Nonsignificant fixed terms were then excluded from the model, as were nonpositive components of variance. Predicted mean ratios and corresponding SE were obtained using the simplified model. Histograms of residuals and graphs of residuals vs. fitted values were examined for nonnormality of distribution (skewness and outliers) and constant variance. Apparent outliers were further evaluated using robust Z-scores:

$$Z_i = \frac{1.349(r_i - q_{0.5})}{q_{0.75} - q_{0.25}},$$

where r_i is the ith random effect (residual), and q_p is the pth quantile of the distribution of random effects (Australian National Quality Assurance Program, 2006). The quantiles were estimated by a kernel density estimate (Silverman, 1986). This was done both for the residuals in the lowest stratum (repeated observations) and for the estimated random effects of cow. Units with large Z-scores (in this case >3.2) were removed from the analysis. All analysis was performed using GenStat (2006) software.

A mixed model was used to examine the components of variation for actual CH_4 emissions (g/d) estimated using both techniques. The mixed model included fixed effects for chamber, treatment (monensin vs. control), and a linear covariate for DMI. Random effects included in the model were cow and day within cow. The estimated components of variation were used to calculate, excluding data from outliers, the CV within cow and between cows.

A meta-analysis was developed to compare the ratios observed in the present study with the ratios obtained in studies conducted in Canada (McGinn et al., 2006) and Ireland (F. O'Mara, University College Dublin, Ireland; personal communication). In both these studies CH_4 emissions were estimated by chambers and by use of the SF_6 tracer technique within the chambers. The meta-analysis took the form of an elaborate mixed model for the combined ratio data. The model was defined by first developing an appropriate mixed model for each data set separately, and then incorporating these into a single model. Any nonpositive variance components identified in the initial separate analyses were not included in the extended model. The simplified model described above was used for the Australian data. The Canadian data were specified initially with factorial fixed effects for feed type by intake level, and random effects for period by chamber, and repeated measures nested within chamber by period. The random effects for period and chamber were excluded from the combined model. For the Irish data, the relationship between the ratio and intake level (fasted vs. fed), was first checked graphically. The combined mixed model included only a fixed effect for the mean, and a random effect for units. The combining of the 3 simplified models into a single analysis was achieved in GenStat by specifying missing values in each factor definition when the factor did not apply, adding the respective fixed and random effects models, and specifying an option in the GenStat code to retain all units in the analysis, whether factor levels were missing.

The relationship between DMI and actual CH_4 emissions (g/d) was examined for the combined Australian and Canadian data sets (those studies in which DMI was measured) by fitting a similar combined model for the ratio of CH_4 (g/d) to DMI (kg/d).

RESULTS AND DISCUSSION

Chamber Performance

The chamber calibrations indicated that there was a maximum of 1% difference between the released and recovered CH_4. Thus, no correction factors were applied to the data.

Methane Emissions

The tracer technique failed on 6 out of 48 cow-days (88% successful measurements) due to breakages and blockages of air lines. Due to these missing values, mean CH_4 emissions for the experiment were calculated using only the paired data (i.e., when both techniques functioned).

The accuracy of the SF_6 tracer technique was investigated by considering the ratio of the CH_4 emission measured using the SF_6 tracer to the emission measured using the chamber for each cow on each day. In this analysis, data outliers were identified (Z-values >3.2) and removed from subsequent analysis of the ratios (Figure 2). Data outliers were found for 1 cow (over 3 d) having abnormally low chamber emissions and for a second cow (over 3 d) in which SF_6 tracer emissions were abnormally high. There were no obvious detectable physiological reasons for these abnormal emis-

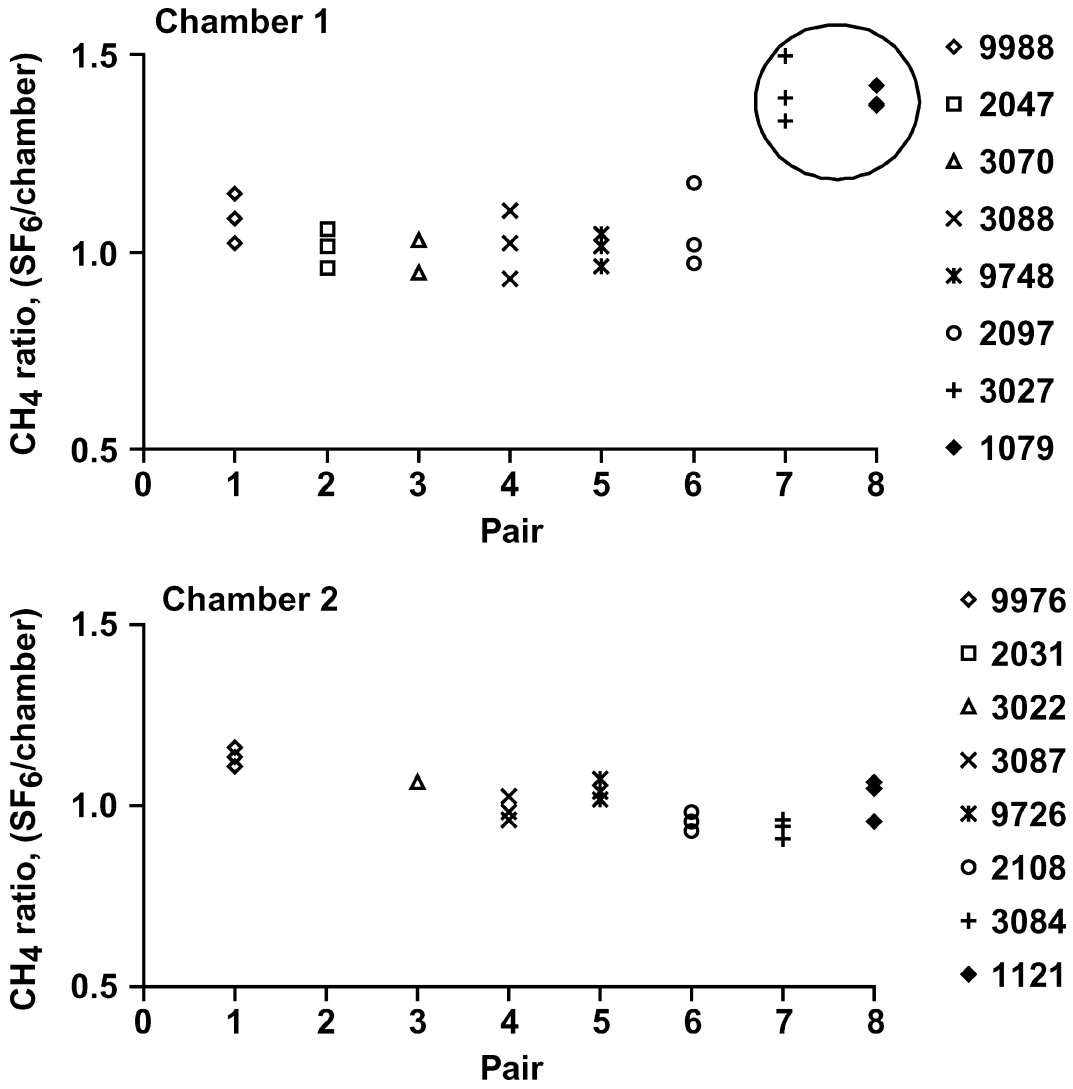

Figure 2. Ratio of the total daily CH_4 emissions measured using the SF_6 technique to that measured using the chamber for 8 pairs of individual cows (cow ID shown in the legend), by day and chamber. Data outliers are circled. Cows were paired with one cow placed in each chamber.

sions. For the chambers, CH_4 emissions averaged (mean ± SD) 322 ± 57.5 g/d (n = 36). For the SF_6 tracer technique, CH_4 emissions averaged (mean ± SD) 331 ± 74.6 g/d (n = 36).

Expressed as a percentage, with outliers removed, the ratio of CH_4 emissions from the SF_6 tracer technique to chamber emissions averaged 102.3% (SE = 1.51, not significantly different from 100%, $P = 0.14$). The ratio was not affected by chamber ($P = 0.72$) or monensin treatment ($P = 0.38$) and there was no effect ($P = 0.88$) of monensin on DMI (Table 2). There was also no effect of permeation tube release rate ($P = 0.90$) or actual CH_4 emission (g/d) measured in chambers ($P = 0.77$) on the ratio.

The ratio did, however, appear to be associated with DMI ($P < 0.001$). The estimated relationship between the ratio and DMI was:

$$\text{Ratio} = 102.4\ (\text{SE } 1.15) + 1.24\ (\text{SE } 0.342) \times (\text{DMI} - 18.39).$$

The ratio was not affected by level of CH_4 emission, but was affected by level of DMI. This seems contradictory because one would expect DMI to be correlated with level of CH_4 emission. However, estimates of CH_4 emissions varied with DMI depending upon the technique. The linear regression of CH_4 emission with DMI was 12.9 DMI (kg/d) + 72.3 ($R^2 = 0.39$) for the chamber

Table 2. Mean ratio of sulfur hexafluoride (SF_6) to chamber CH_4, expressed as a percentage, for the effect of chamber (1, 2) and monensin (control, treatment) and effect of monensin on DMI[1]

Item	Mean ratio (%)	SED[2]	DMI	SED
Chamber				
Chamber 1	102.8	3.14	—	—
Chamber 2	101.9	3.14	—	—
Monensin				
Control	103.5	3.12	18.1	1.57
Treatment	101.4	3.12	18.3	1.57

[1]Within a column, means for chambers and monensin differ, $P < 0.05$.

[2]Standard error of the difference between means.

technique and 18.5 DMI (kg/d) – 9.5 (R2 = 0.56) for the SF_6 tracer technique. On only 8 occasions (out of a total of 36 data observations) was DMI of cows above 20 kg/d; hence, more data would be required for cows with high intakes to establish greater confidence in the relationship reported above.

The ratio of 102.3% for the 2 techniques is different from that reported previously for beef cattle and sheep, in which the SF_6 technique was 5 to 7% lower than the chamber (Johnson et al., 1994; Ulyatt et al., 1999; McGinn et al., 2006). Differences between techniques in those studies were partially attributed to the release of CH_4 from the rectum, which was not measured by the SF_6 tracer technique in those studies, but was measured by the chambers. About 1% of the total enteric CH_4 emissions is released from the rectum in sheep (Murray et al., 1976), although no estimates are available for cattle. We expected that the 2 techniques in our study would give similar estimates of CH_4 emissions because our in-chamber use of the SF_6 tracer technique measured total enteric CH_4, including CH_4 both respired and released from the rectum.

Previous studies comparing the 2 techniques did not use animals emitting high quantities of CH_4 (Johnson et al., 1994; Ulyatt et al., 1999; McGinn et al., 2006). The results of our study support the hypothesis that the SF_6 tracer technique is accurate when used with dairy cows that have high DMI and high CH_4 emissions relative to beef cattle. A qualification to this statement is that at DMI above 20 kg/d, the SF_6 tracer technique may overestimate CH_4 emissions.

Variability in Estimates of Methane Emissions

One advantage of the chamber technique is that it provides information on the variability of emissions within a day. For an individual cow, the emission was typically highest after feeding, and peaking at about twice the lowest values found just before feeding, as shown in Figure 3.

Both techniques provide information on variability among cows and among days within cows. The coefficient of variation (**CV**) for CH_4 emissions in our experiment were calculated using the components of variance estimated for the random effects in the mixed model. The CV within cow (day-to-day) was 6.1% for the SF_6 tracer technique and 4.3% for the chamber technique. If we assume that the chamber measured real variation in CH_4 emissions from the cow, and the SF_6 technique measured variation in CH_4 emissions plus additional error associated with the SF_6 technique, then the additional imprecision due to the technique would be estimated by the difference in the corresponding SF_6 and chamber components of variance (day-to-day). This component implied a day-to-day CV for the SF_6 technique itself of 4.3%.

The higher variability between days using the SF_6 tracer technique indicates the need to replicate measurements over a greater number of days compared with chamber measurements to obtain the same level of precision as the chamber technique. This requirement would be even greater for studies conducted under grazing conditions where weather conditions are variable (Ulyatt et al., 1999). The variability among cows was substantially higher than within cows, and also showed a difference between techniques; CV of 19.6 and 17.8% for the SF_6 tracer and chamber techniques, respectively. These imply a CV of 8.2% measurement contribution from the SF_6 technique, excluding biological variation in actual CH_4.

Using chambers, Blaxter and Clapperton (1965) reported a 7.2% CV for day-to-day variation based on 989 24-h determinations of CH_4 for sheep and cattle. They also reported a CV between animals of 5.0 to 7.5% for sheep given a fixed amount of feed. The CV between animals, however, appears to be larger in chamber studies when intake is not restricted. For example, McCourt et al. (2005) reported a CV for 135 beef steers of 17.2% (DMI: 4.4 to 11 kg/d), Bruinenberg et al. (2002) reported a CV of 16.9% for 96 measurements from grass-fed dairy cows (DMI: 6.7 to 20.8 kg/d), and Yan et al. (1997) reported a CV of 18.2% for 221 dairy cows fed grass silage based-diets (DMI: 7.5 to 24.5 kg/d). Thus, the CV we report (approximately 18%) for animal variation was similar to that reported previously for cattle fed ad libitum.

Our study shows that when the SF_6 tracer technique is used on an animal within a chamber, the variability among animals is larger than for direct chamber measurements after accounting for effects of DMI. Boadi et al. (2002) also reported for a small number of animals that animal variability was greater for the SF_6 tracer

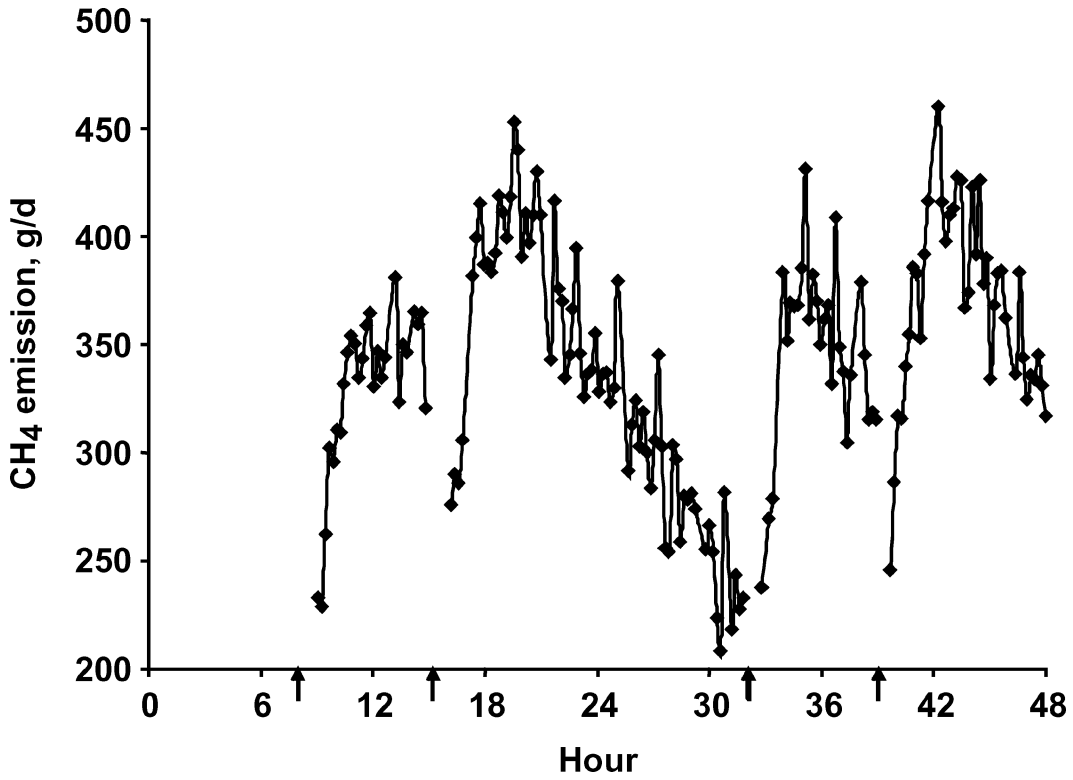

Figure 3. Example of CH_4 emissions from a single cow over 2 d beginning the morning of November 27 using the chamber technique. The arrows indicate feeding and milking times.

technique than for chambers, even though the animals had similar intakes. Vlaming et al. (2005) summarized a number of studies that used the SF_6 tracer technique with grazing dairy cows and sheep, and reported CV for animal variation of 31.1 and 35.8%, respectively. However, DMI was variable in that study and was not accounted for in the CV calculations. We did adjust for intake in our analysis. Our CV estimates increase to 19.2% if we do not adjust for DMI.

Meta-Analysis

Combining the data from the present experiment (Australian) with the Canadian and Irish data resulted in a data set characterized by a wide range of CH_4 emissions due to the range in DMI, diet quality, and animal type (Figure 4). In the Canadian study, the chambers measured emissions from the whole animal, whereas the SF_6 tracer technique only measured respired CH_4. The Canadian data set (n = 46) was for growing beef cattle fed diets containing 30 to 70% concentrate (DM basis) fed at 65 or 100% of ad libitum intake. One outlier with a ratio of 153% was excluded from subsequent analysis. In the Irish study, both the chambers and the SF_6 tracer technique measured emissions from the whole animal. The Irish data set was for nonlactating Holstein-Friesian cows fed a diet of 67% concentrate and 33% straw (DM basis, ranging from maintenance level of energy intake or fasted over a period of 6 d).

The accuracy of the SF_6 tracer technique, as indicated by the mean ratios of SF_6 to chamber CH_4 for the individual data sets used in the meta-analysis, differed (P = 0.004) among countries (Table 3). As expected, the ratio was highest for the Australian data set because both techniques measured whole-animal CH_4 emissions, whereas that was not the case in the Canadian study. The lower ratio obtained for the Irish data set may be due to the very low emissions of some cows caused by fasting. With very low emissions, a difference of only a few grams per day could cause a large change in the ratio. The meta-analysis demonstrated that over a range of conditions (Australian, Canadian, and Irish studies), the SF_6 tracer technique gave CH_4 emission values that were about 8% lower than complete recovery of whole-animal emissions. However, this difference is also explainable in terms of the different DMI and the relationship between the ratio of SF_6 to chamber CH_4 and DMI, observed both within and between studies.

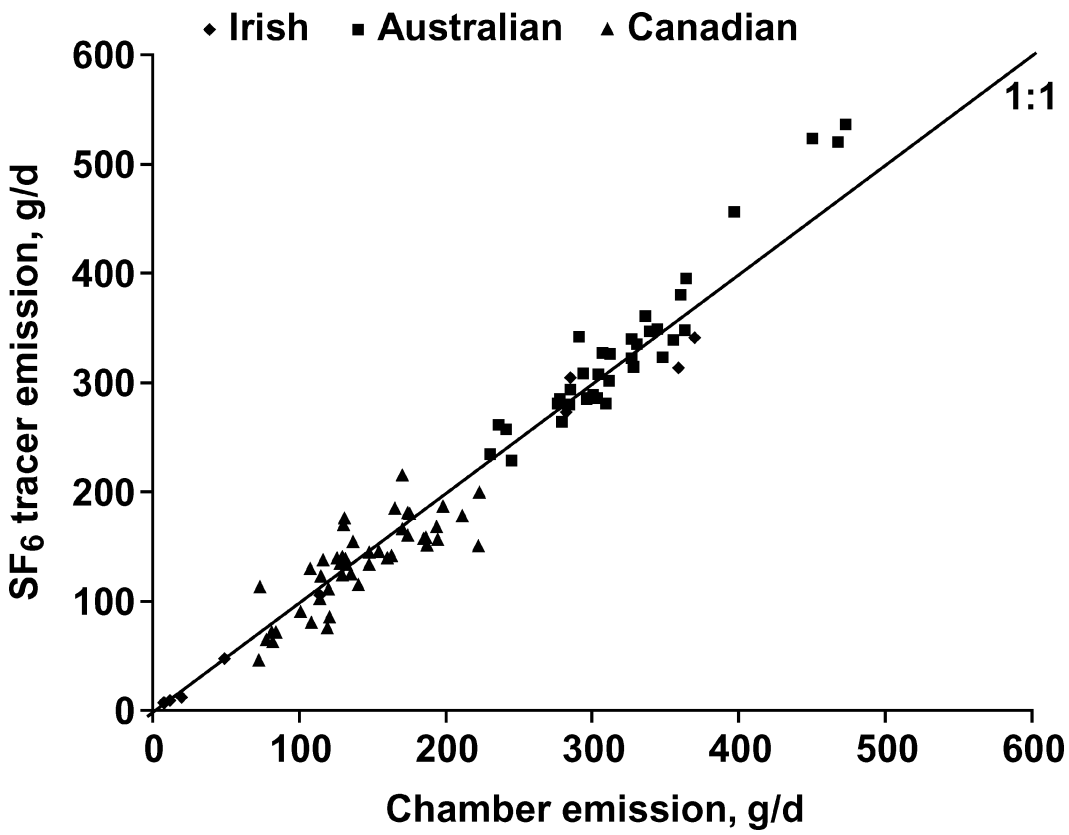

Figure 4. Methane emissions measured using the chamber and SF_6 techniques from Australia (this study, excluding data from 2 outliers), Canada (McGinn et al., 2006), and Ireland (F. O'Mara, University College Dublin, Ireland; unpublished data).

Although our primary objective was to compare the 2 techniques for measuring CH_4 emissions, the study also allowed us to examine the relationship between CH_4 emission and DMI. This relationship is of interest because CH_4 is expressed on the basis of DMI for inventory purposes in some countries (e.g., New Zealand). The relationship between CH_4 emissions measured in chambers and DMI was examined using only the Australian and Canadian data sets, because intakes were not available for the Irish data. In both studies, CH_4 emissions estimated using the chambers were proportional to DMI (Figure 5). There was a difference ($P < 0.001$) between the slope of the Australian and Canadian data sets (17.1 vs. 20.8; SED = 0.93) when CH_4 (g/d) was plotted against DMI (kg/d) with the intercept = 0. A difference between the 2 locations was expected based on the difference in diet composition for these 2 studies. Our value of 17.1 g of CH_4/kg of DMI for dairy cows was slightly lower than the mean values of 18.4 to 19.8 g of CH_4/kg of DMI reported by Bruinenberg et al. (2002) for dairy cows fed mostly grass in chambers, possibly due to the higher proportion of concentrate (26 vs. 10% of DMI) fed in our study.

Table 3. Mean ratio of sulfur hexafluoride (SF_6) to chamber CH_4, expressed as a percentage, for Australian, Canadian (McGinn et al., 2006), and Irish (F. O'Mara, University College Dublin, Ireland; unpublished data) data sets

Source of data	Animal type	Mean ratio (%)	SE
Australia	Lactating dairy cows	102.3[a]	1.51
Canada	Growing beef cattle	94.1[b]	3.75
Ireland	Nonlactating dairy cows	89.3[b]	4.22

[a,b]Within a column, means for countries without a common superscript differ, $P < 0.05$.

CONCLUSIONS

Total CH_4 emissions were similar (322 and 331 g of CH_4/d), when measured using chambers and the SF_6 tracer technique. A meta-analysis performed on data sets from 3 locations indicated that the SF_6 tracer technique resulted in CH_4 emission values that were about 8% lower than those measured by the chamber. This probably occurred because the SF_6 technique does not normally measure emissions from the rectum. These

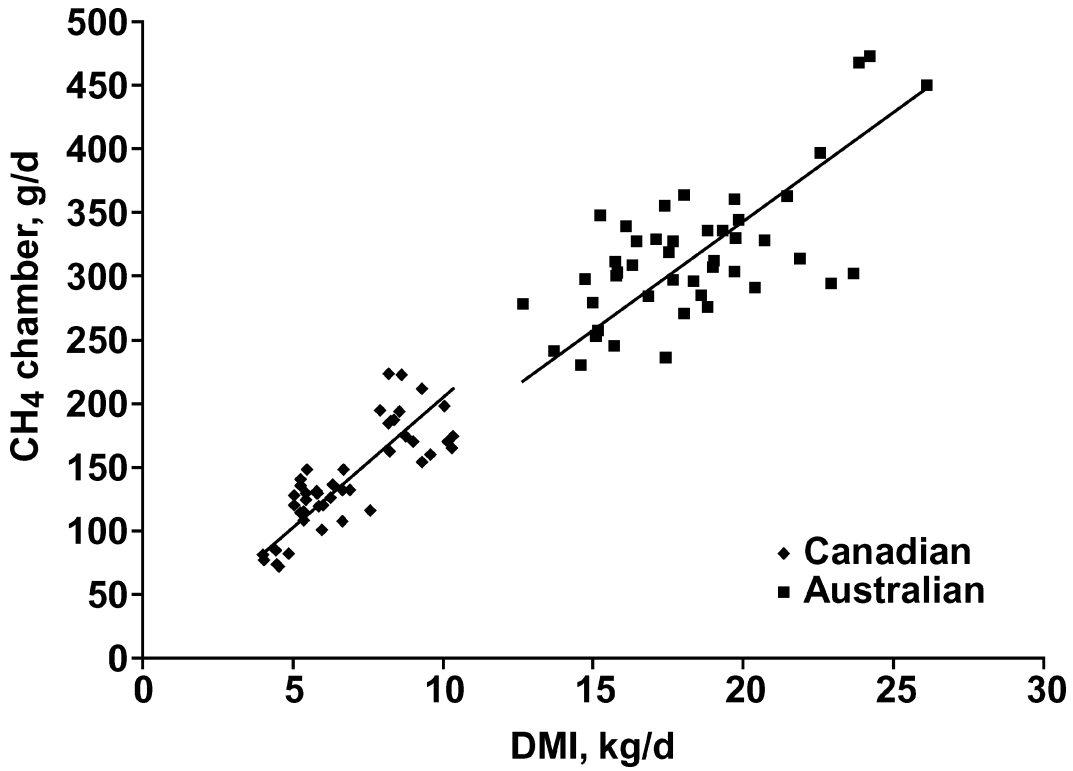

Figure 5. Relationship between CH_4 emission determined in chambers and DMI for Australian (excluding outlier data from 1 cow) and Canadian (McGinn et al., 2006) data. Lines are through the origin and have slope estimates of 17.06 for the Australian data and 20.79 for the Canadian data ($P < 0.001$; SED = 0.928).

emissions may be higher than previously reported (i.e., 1%), and if known, could be factored into the estimation of CH_4 emissions using the SF_6 tracer technique. We conclude that the SF_6 tracer technique can be used with a reasonable degree of accuracy for inventory purposes and for evaluating the effects of mitigation strategies on CH_4 emissions.

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