

Effects of grain supplementation on methane production of grazing steers using the sulphur (SF_6) tracer gas technique

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¹Department of Animal Science, University of Manitoba, Winnipeg, Manitoba, Canada R3T 2N2; ²Agriculture and Agri-Food Canada, P. O. Box 1000A, Brandon, Manitoba, Canada R7A 5Y3. Received 8 May 2001, accepted 22 February 2001.

Boadi, D. A., Wittenberg, K. M. and McCaughey, W. P. 2002. **Effects of grain supplementation on methane production of grazing steers using the sulphur (SF_6) tracer gas technique.** *Can. J. Anim. Sci.* **82**: 151–157. The objective of the study was to examine the effect of supplemental grain on methane (CH_4) production of grazing steers. Eight beef steers (344.6 ± 6.4 kg) were assigned to legume-grass pasture (C; $n = 4$) or legume-grass pasture plus a rolled barley supplement (S; $n = 4$). In a completely randomized design with repeated measures, CH_4 output was measured for two 24-h periods, using the SF_6 tracer gas technique as steers entered (IN) and exited (OUT) paddocks. Two, 4 and 4 kg of rolled-barley grain was fed daily to S steers during the EARLY, MID and LATE periods of the grazing season, respectively. Supplementation reduced forage dry matter intake (DMI) by 11% ($P = 0.03$) and increased total organic matter intake (TOMI) by 14% ($P = 0.001$). Daily CH_4 production was similar for C and S steers ($P > 0.05$). Methane production, increased ($P < 0.05$) from 256 L d^{-1} in the EARLY period to 364 L d^{-1} at the MID and 342 L d^{-1} at the LATE period. Energy lost as CH_4 , % total gross energy intake (TGEI) ranged from 4.7 to 8.4% (mean $6.5 \pm 0.3\%$) during the grazing season, and there was no difference between S ($6.4 \pm 0.6\%$) and C ($6.7 \pm 0.6\%$) steers ($P = 0.71$). Methane production declined with grazing on high-quality forages; steers on EARLY pastures had 44% and 29% lower ($P < 0.05$) energy loss as CH_4 than animals on MID and LATE pastures, respectively. There was also a 54% lower CH_4 loss when animals entered new paddocks relative to those exiting the paddocks ($P < 0.05$). It can be concluded that the effects of supplementation on CH_4 production were marginal in grazing steers. The study suggests that pasture quality plays a major role in the extent to which CH_4 production can be reduced with grain supplementation in grazing animals.

Key words: Methane, grazing steers, grain supplementation, pasture quality

Boadi, D. A., Wittenberg, K.A. et McCaughey, W. P. 2002. **Évaluation de l'incidence des suppléments de grain sur la production de méthane par les bouvillons en paissance au moyen de la technique de traçage à l'hexafluorure de soufre (SF_6).** *Can. J. Anim. Sci.* **82**: 151–157. L'étude devait établir l'incidence d'un supplément de grain sur la production de méthane (CH_4) par des bouvillons en paissance. Huit bouvillons de boucherie ($344,6 \pm 6,4$ kg) ont été placés dans un pré de graminées-légumineuses avec (S; $n = 4$) ou sans (C; $n = 4$) supplément de flocons d'orge. Dans le cadre d'une expérience entièrement randomisée à mesures répétées, on a déterminé la quantité de CH_4 produite au cours de deux périodes de 24 h par la technique de traçage au SF_6 , à l'entrée ou à la sortie des bouvillons de l'enclos. Les bouvillons S ont respectivement reçu chaque jour 2, 4 et 4 kg de flocons d'orge au début, au milieu et à la fin de la saison de paissance. Le supplément de grain diminue l'ingestion de matière sèche de 11 % ($P = 0,03$) et augmente la quantité totale de matière organique absorbée de 14 % ($P = 0,001$). Les bouvillons C et S libèrent la même quantité de CH_4 ($P > 0,05$). Le volume de méthane produit quotidiennement est passé ($P < 0,05$) de 256 litres au début de la période de paissance à 364 litres au milieu puis à 342 litres à la fin. L'énergie perdue sous forme de CH_4 se situait entre 4,7 et 8,4 % de la quantité totale d'énergie brute absorbée (moyenne de $6,5 \pm 0,3$ %) pendant la saison de paissance et on n'a observé aucune variation entre les bouvillons S ($6,4 \pm 0,6$ %) et C ($6,7 \pm 0,6$ %) ($P = 0,71$). Le volume de méthane produit diminue quand les animaux broutent des fourrages de haute qualité. Les bouvillons mis en paissance au début de la saison perdent respectivement 44 % et 29 % moins ($P < 0,05$) d'énergie sous forme de CH_4 que ceux placés au milieu ou à la fin de la saison. Les animaux qui entrent dans un nouvel enclos perdent aussi 54 % moins d'énergie en méthane que ceux sortant de leur enclos ($P < 0,05$). De l'étude on conclut que les suppléments de grain ont une incidence marginale sur le volume de méthane libéré par les bouvillons en paissance. Les résultats donnent à penser que la qualité du pâturage joue un rôle déterminant dans la capacité du supplément à réduire la quantité de méthane produite par les animaux en train de paître.

Mots clés: Méthane, bouvillons en paissance, supplément de grain, qualité du pâturage

Cultivated and native pastures are primary resources for beef production in Western Canada. As forages mature, there is decreased digestibility, related to decreased nitrogen and increased fibre and lignin contents of the forage (Minson 1990). Reduced forage digestibility is accompa-

nied by decreased forage intake, and an increased acetate:propionate ratio, which favours increased methane (CH_4) production per unit of forage consumed (McAllister et al. 1996). Methane production from cattle constitutes 2–12% of gross energy loss, and is also a major contributor

Abbreviations: ADF, acid detergent fibre; CP, crude protein; DM, dry matter; DMI, dry matter intake; GE, gross energy; GEI, gross energy intake; IVDMD, in vitro dry matter digestibility; OM, organic matter; TGEI, total gross energy intake; TDMI, total dry matter intake; TOMI, total organic matter intake

to atmospheric greenhouse gas emissions (Johnson and Johnson 1995). As a result, strategies to reduce greenhouse gases from every sector of the animal industry have intensified in recent years.

The efficiency of gastrointestinal fermentation in ruminants dictates the ratio between the end products formed, i.e., efficiency is high when there is optimum microbial growth relative to volatile fatty acids produced (Van Soest 1982). Under such situations there is a high rate of digestion by microbes and lower CH₄ production (Leng 1993). Strategic supplementation can improve microbial efficiency by supplying the needed microbial growth factors (Leng 1993). Improvements in the efficiency of rumen fermentation resulting from urea and mineral supplementation have been shown to reduce the percentage of digested energy in low-quality feeds fermented to CH₄ and also to reduce CH₄ produced per kilogram of gain or milk (Hennessy and Williamson 1990; Ward et al. 1993).

In temperate regions, where legumes are often incorporated in pastures, the lack of energy (rather than protein) is the most limiting nutrient for production of meat or milk by grazing animals (Allden 1981). Supplementation of forages with grain, especially at high intake, can improve the efficiency of feed utilization, increase ruminal propionate concentration, and reduce CH₄ emissions by the provision of readily fermentable energy sources for microbes during fermentation (Reis et al. 2001).

Most of the above studies have been conducted with penned animals with limited studies on grazing animals. This can be attributed to difficulties in obtaining CH₄ measurements while animals are grazing. The recent development of the sulphur hexafluoride (SF₆) tracer gas technique allows direct measurement of individual grazing animals, without disturbance to their grazing behavior (Johnson et al. 1994). As a result, dietary strategies for reducing CH₄ production on pasture can be evaluated. McCaughey et al. (1997, 1999) have recently used the SF₆ tracer gas technique to measure the impact of grazing management on CH₄ production by steers and lactating beef cows. The objective of this study was to use the SF₆ tracer gas technique to measure the effects of grain supplementation on CH₄ production of grazing steers.

MATERIALS AND METHODS

Eight Red Angus yearling steers, averaging [344.6 ± 6.4 kg (mean ± SD)] were assigned to one of two dietary treatments to determine the effects of barley grain supplementation on CH₄ production during grazing. The selected steers were managed as part of a larger herd of 48 steers that were rotationally grazing on eight, 3.7-ha paddock pastures at the Brandon Research Station, Brandon, Manitoba, during the 1998 grazing season. Pastures consisted of alfalfa (*Medicago sativa* L.), and meadow brome grass (*Bromus biebersteinii* Roem and Schult.). The proportion of legume:grass was determined at the beginning and end of CH₄ collection periods.

Animals started grazing pastures on 5 May 1998. They were adapted to grazing and supplemental feeding for 6 wk

prior to CH₄ collection. During the adaptation period, the test steers were halter trained, in order to get them accustomed to wearing the CH₄ collection apparatus. Steers had free access to water and a commercially prepared mineral supplement. Shrunken body weights were taken at the start and end of trial to determine overall rate of gain. Animals were cared for in accordance to the guidelines of the Canadian Council on Animal Care.

Experimental Layout

The dietary treatments consisted of four steers [342.8 ± 6.4 kg (mean ± SD)], which grazed alfalfa and meadow brome grass pastures (C), and four steers [346.5 ± 6.4 kg (mean ± SD)], which grazed the alfalfa-meadow brome grass pastures and were supplemented with steamed-rolled barley grain (S). Three CH₄ sampling periods: EARLY (17–25 June 1998); MID (20–29 July 1998) and LATE (12–21 August 1998) were selected in the grazing season to reflect changes in forage quality as the grazing season progressed. Methane production was measured from steers over two 24-h sampling times per sampling period: as they entered (IN) and exited (OUT) paddocks to reflect the changing pasture quality within paddocks during a sampling period. Animals were rotated to new paddocks every 9 d. The experiment was conducted as a completely randomized design with repeated measures.

Grain Supplementation

Steers assigned as S received 2, 4 and 4 kg d⁻¹ of steamed-rolled barley grain in the EARLY, MID and LATE grazing season, respectively. The increase from 2 to 4 kg d⁻¹ grain supplementation occurred on 9 July, 11 d prior to the MID sampling period measurements. Animals were individually fed once a day (in the afternoon), using individual feeders located close to the pastures for the entire grazing season. Grain and ort samples were weighed and subsampled daily in each sampling period. Feed and ort samples were composited by period for future analyses.

Forage Availability and Botanical Composition

Forage availability was determined at the beginning and end of each sampling period. Forage was clipped with an electric grass shear to a height of 50 mm within 0.25-m² quadrats. Ten randomly placed quadrats were sampled on each occasion. Dead material was separated from the forage, to ensure accurate estimates of current availability. Forage samples were separated by hand into legume and grass portions and dried at 50°C for 48 h. The mean dry weights of grass and legumes were used in calculating herbage mass (kg ha⁻¹), and ratios of legume:grass.

Forage Quality

In each of the three sampling periods, three oesophageal-fistulated steers were used to collect grazed forage samples for forage quality analyses. Fistulated steers were fasted overnight and allowed to graze in the sampling paddock for 20 min at both IN and OUT. Extrusa samples were obtained in canvas collection bags fitted around the cannulated areas.

Extrusa samples were squeezed through two layers of cheesecloth and the saliva fraction sampled and frozen immediately after separation. The saliva fraction was used to correct for organic matter losses of solid samples (Cohen 1979). The solid fractions of samples were immediately dried in a forced-draught oven at 50°C for 48 h. Dried samples were ground using a Wiley mill fitted with a 1-mm screen, and later analyzed for dry matter (DM), organic matter (OM), crude protein (CP), acid detergent fibre (ADF), neutral detergent fibre, gross energy (GE), *in vitro* dry matter digestibility (IVDMD).

Total and Forage Intake

Total DMI (TDMI) was determined by the following calculation (Burns et al. 1989):

$$\text{DMI (g d}^{-1}\text{)} = \text{faecal DM output (g d}^{-1}\text{)} / 1 - (\text{IVDMD}/100)$$

Faecal output was estimated using Cr₂O₃ controlled release capsules (Captec Ltd, Auckland, NZ), as described by Barlow et al. (1988). Each steer was dosed a week prior to the sampling period to establish a constant release of Cr₂O₃ in the gut. Faecal grab samples were collected for 3 d each at IN and OUT sampling times. This ensured that intakes correspond accurately with CH₄ estimates taken at IN and OUT. Forage intake of S steers was estimated by subtracting DMI of barley grain from TDMI. Faecal samples were packed with ice after collection, and frozen immediately. Faecal DM output was calculated as the ratio of Cr₂O₃ released (1.72 g d⁻¹):Cr₂O₃ concentration in faeces (g g⁻¹DM).

Methane Gas Sampling and Analyses

Methane gas was sampled using the SF₆ tracer gas technique (Johnson et al. 1994). Stainless-steel permeation tubes (12.5 mm × 40 mm), with known release rates for SF₆ were placed in the rumen, per os, a week prior to the start of gas sampling. The rate of SF₆ was controlled by a permeable Teflon membrane held in place by a stainless-steel Swagelok nut. Each tube was charged with 260–300 mg of SF₆ and incubated at 39°C. Release rates of SF₆ were determined by measuring the weight loss of tubes for 8 wk to establish a steady pre-determined rate. Release rates of the permeation tubes used in this study ranged from 350 to 700 ng min⁻¹.

Gases exhaled from the nose and mouth were drawn into pre-evacuated (30 mm Hg) stainless-steel collection spheres (130-mm diameter), through a 900-mm capillary tubing (128 µm i.d) with an in-line 15 µm filter and flexible nose piece (McCaughy et al. 1999). The gas collection apparatus was fitted to a halter. The collection spheres were suspended by a neck strap attached to the halter apparatus with a quick connect fitting (McCaughy et al. 1997). The collection system was designed to deliver half its volume during a 24-h collection, ensuring a uniform collection rate.

At each sampling time, two consecutive 24-h gas samples were collected from each animal. Steers were restrained in a chute to remove collection spheres prior to feeding of barley grain to prevent blockages of capillary lines with feed particles. New canisters were replaced after feeding and animals were then returned to pasture. Background air samples were collected each time animals were sampled. The

background gas concentrations were used to correct expired gas concentrations. Collected spheres were checked for pressure to identify blocked or leaking capillary systems to ensure data used represented a complete 24-h period. Spheres were then pressurized to 110 kPa with pure N₂, to prevent sample contamination prior to analysis, and to allow injection of gas samples into the sample loop of the gas chromatograph.

A gas chromatograph (Star 3600, Varian, Mississauga, ON) fitted with electron capture and flame ionization detectors was used to determine SF₆ and CH₄, respectively. The gas chromatograph was fitted with a Molecular Sieve 0.5 nm (1800 mm) column and a Poropak QS (1800 mm) column for SF₆ and CH₄, respectively. The column oven temperature was 35°C and nitrogen was used as the carrier gas with a flow rate of 30 mL min⁻¹. Samples were analyzed in duplicates. Prepared standards were used to standardize the gas chromatograph for SF₆ (20 ppt; Scott-Marrin Inc., Riverside, CA) and CH₄ (100 ppm; Supelco, Mississauga, ON) prior to sample analysis. Daily CH₄ emissions were calculated as follows:

$$\text{CH}_4 \text{ (L min}^{-1}\text{)} = \frac{\text{permeation tube SF}_6 \text{ release rate (L min}^{-1}\text{)} \times [\text{CH}_4]/[\text{SF}_6]}{\text{where } [\text{CH}_4] \text{ and } [\text{SF}_6] \text{ are the concentrations of CH}_4 \text{ and SF}_6 \text{ in samples after background concentrations have been deducted.}}$$

where [CH₄] and [SF₆] are the concentrations of CH₄ and SF₆ in samples after background concentrations have been deducted.

Chemical Analyses

Barley grain and ort samples were dried for 48 h in a forced-draught oven at 60°C for DM determination. Faecal and saliva samples were freeze dried at -35°C (Genesis 25LE freeze dryer, Gardiner NY). Dried barley, forage and faecal samples were ground using a Wiley mill fitted with a 1-mm screen. Samples were analyzed for CP, using a Kjeltec 1030 auto analyzer [Tecator Inc., Herndon, VI; (Association of Official Analytical Chemists 1990)] method no. 984.13 and ash using method no. 942.05 (Association of Official Analytical Chemists 1990). Acid detergent fibre and neutral detergent fibre of grain and forage samples were determined using an ANKOM 200 fibre analyzer (Fairport NY), with procedures described by Komarek (1993).

Gross energy was determined using a Parr 1241 adiabatic bomb calorimeter. *In vitro* dry matter digestibility (IVDMD) of forage and grain samples were determined by the method of Tilley and Terry (1963) using bovine inoculum. Chromium concentration in faecal samples was determined by atomic absorption spectrophotometry (Model IL 551 AA/AE spectrophotometer), using air and acetylene flame (Williams et al. 1962).

Statistical Analyses

Diet quality, intake and methane data were analyzed by least square analyses of variance using GLM in SAS Institute, Inc. (1990) in the model:

$$Y_{ijkl} = \mu + T_i + A_{j(i)} + P_k + (TP)_{ik} + (AP)_{j(i)k} + M_l + (TM)_{il} + (PM)_{kl} + (TPM)_{ikl} + \epsilon_{ijkl}$$

Table 1. Effect of time of grazing (Period) and length of time in a paddock (Time) on the nutrient analyses (DM basis) of oesophageal masticates from alfalfa-meadow bromegrass pastures (LS means \pm SE)

Factors	NDF (%)	ADF (%)	CP (%)	ASH (%)	IVDMD (%)	GE (kJ g ⁻¹)
<i>Period (P)^z</i>						
EARLY	50.5 \pm 1.1	33.4 \pm 0.7b	19.2 \pm 0.4	10.2 \pm 0.3b	75.5 \pm 0.8a	18.4 \pm 0.02a
MID	54.2 \pm 1.5	36.5 \pm 1.0a	17.9 \pm 0.5	11.7 \pm 0.4a	70.5 \pm 1.1b	17.9 \pm 0.02b
LATE	52.9 \pm 1.4	36.6 \pm 0.9a	18.2 \pm 0.5	11.1 \pm 0.4a	71.7 \pm 0.9b	17.9 \pm 0.02b
<i>P</i> -value	0.15	0.03	0.92	0.03	<0.01	0.01
<i>Time (T)^y</i>						
IN	36.4 \pm 1.1b	26.9 \pm 0.7b	25.4 \pm 0.4a	10.6 \pm 0.3	81.0 \pm 0.7a	18.8 \pm 0.02a
OUT	68.6 \pm 1.1a	44.0 \pm 0.8a	11.4 \pm 0.4b	11.4 \pm 0.3	64.1 \pm 0.8b	17.6 \pm 0.02b
<i>P</i> -value	<0.01	<0.01	<0.01	0.09	<0.01	<0.01
<i>(P) \times (T)</i>						
<i>P</i> -value	0.79	0.89	0.55	0.01	0.56	0.01

^zPeriod (P) = EARLY (17–25 June); MID (20–29 July) and LATE (12–21 Aug.) period of a grazing season.

^yTime (T) = IN (entry) and OUT (exit) of paddocks.

a, b Means within each factor followed by different letters differ ($P < 0.05$).

Where Y_{ijkl} is the trait under consideration; μ is the overall mean; T_i is the dietary treatment ($i = 1, 2$); $A_{j(i)}$ is the animals within treatments, which was used as the error term to test for dietary treatment effect; P_k is the sampling period in a grazing season ($k = 1 \dots 3$); $(TP)_{ik}$ is the treatment \times sampling period interaction; $(AP)_{j(i)k}$ is the animals within treatment \times sampling period interaction, which was used as an error term to test for sampling period and treatment \times sampling period interaction; M_l is the sampling time (IN and OUT) of paddocks ($l = 1, 2$); $(TM)_{il}$ is the treatment \times sampling time interaction; $(PM)_{kl}$ is the sampling period \times sampling time interaction; $(TPM)_{ikl}$ is the treatment \times sampling period \times sampling time interactions; and ϵ_{ijkl} is the experimental error term. Means were separated at the 5% level of significance using the probability of differences (PDIFF) option.

RESULTS AND DISCUSSION

Forage Availability and Quality

The available forage on offer during the EARLY sampling period was 2932 kg DM ha⁻¹ at IN and 1308 kg DM ha⁻¹ at OUT. The MID period herbage mass was 3388 kg DM ha⁻¹ at IN and 1945 kg DM ha⁻¹ at OUT. The LATE period herbage mass was 3232 kg DM ha⁻¹ at IN and 1830 kg DM ha⁻¹ at OUT. The alfalfa:meadow bromegrass ratio, DM basis, of pasture forage during the EARLY, MID and LATE sampling periods were 20.0:80.0; 33.1:66.9; and 37.6:62.4, respectively.

The ADF content of forages consumed by oesophageal-fistulated steers at the EARLY period was 3% lower ($P < 0.05$) than the MID and the LATE periods, while the CP content remained uniform ($P > 0.05$) during the course of the grazing season (Table 1). This suggests that the CP content of forages, which ranged from 19.2 to 18% in the grazing season, would not have been a limiting factor to fermentation efficiency in that rumen microbes would be supplied with adequate protein for growth and digestion (Leng 1993).

Forage IVDMD on the other hand declined ($P < 0.05$) by 5% from EARLY to the MID and the LATE periods. Forage quality changes during the grazing season were due to a small shift in species composition and physiological matu-

ration of the plant. These results are within the range of values reported by McCaughey et al. (1997) for nutrient analyses of alfalfa-meadow bromegrass pastures. A more dramatic shift in forage quality was observed during the period of paddock occupation. There was a 17% increase ($P < 0.05$) in ADF content, a 14 and 17% decline in CP and IVDMD, respectively ($P < 0.05$), from the time animals entered to the time they exited paddocks (Table 1).

Intake

Concentrate feeding influenced the degree of change in forage DMI between IN and OUT sampling times ($P < 0.05$, Table 2). Forage intake was similar for S and C steers when they entered a paddock; however at OUT, supplementation reduced ($P < 0.05$) forage intake by 31%. For every kilogram DM increase in barley grain intake, forage intake declined by 0.8 kg DM at OUT. This is in contrast to the general findings of higher substitution effects with high-quality forages compared to lower quality forages. Meijis and Hoekstra (1984) observed that herbage intake declined with supplementation when herbage availability was low, while at a high forage availability, the decline in herbage OM intake was higher. In their study, forage quality was similar at both levels of herbage offered, while quality changes occurred as animals grazed in our trial. Average forage DMI declined ($P < 0.05$) by 11% in S steers. This is similar to the reduction in forage intake with concentrate supplementation observed by others (Young et al. 1980; Faverdin et al. 1991).

Forage DMI was observed to be lowest ($P < 0.05$) at the MID sampling period. Increasing concentrate offered and consumed at the MID sampling period to 4 kg d⁻¹ appears to be the cause for the decline in forage intake. This observation is consistent with other studies where a decline in forage intake with increasing level of concentrate supplementation has been observed (Meijis and Hoekstra 1984; Faverdin et al. 1991). Forage quality and quantity changes from the time animals entered a paddock to the time they left it affected forage DMI the most, with a 51% decline in intake at OUT ($P < 0.05$). On average there was a

Table 2. Effect of grain supplementation (Diet), time of grazing (Period) and time in a paddock (Time) on forage intake of steers (LS means \pm SE)

Factors	DMI (kg d ⁻¹)	DMI (% BW)	OMI (kg d ⁻¹)
<i>Diet^a</i>			
C	11.1 \pm 0.3a	2.9 \pm 0.1a	10.5 \pm 0.3a
S	9.9 \pm 0.3b	2.5 \pm 0.1b	9.2 \pm 0.3b
<i>P</i> value	0.03	<0.01	0.05
<i>Period^b</i>			
EARLY	12.1 \pm 0.4a	3.3 \pm 0.1a	11.4 \pm 0.4a
MID	8.5 \pm 0.4b	2.1 \pm 0.1c	7.9 \pm 0.4c
LATE	11.0 \pm 0.4a	2.6 \pm 0.1b	10.2 \pm 0.4b
<i>P</i> value	<0.01	<0.01	<0.01
<i>Time^c</i>			
IN	14.1 \pm 0.3a	3.6 \pm 0.1a	13.3 \pm 0.3a
OUT	6.9 \pm 0.3b	1.7 \pm 0.1b	6.3 \pm 0.3b
<i>P</i> value	<0.01	<0.01	<0.01
<i>Diet \times Time</i>			
C \times IN	14.0 \pm 0.4a	3.7 \pm 0.1a	13.3 \pm 0.4a
C \times OUT	8.1 \pm 0.5b	2.1 \pm 0.1b	7.6 \pm 0.5b
S \times IN	14.2 \pm 0.4a	3.6 \pm 0.1a	13.3 \pm 0.4a
S \times OUT	5.6 \pm 0.4c	1.4 \pm 0.1c	5.1 \pm 0.4c
<i>P</i> value	<0.01	0.02	<0.01

^aDiet: C = (alfalfa/grass pasture); S = (barley grain + alfalfa/grass pasture).

^bPeriod (P) = EARLY (17–25 June), MID (20–29 July) and LATE (12–21 Aug.) period of a grazing season.

^cTime (T) = IN (entry) and OUT (exit) of paddocks.

a–c Means within each factor followed by different letters differ ($P < 0.05$).

21% drop in digestibility of forages and a 39% increase in fibre content of forages contributing to the reduced DMI. Minson (1990) indicated that as plants mature there is a reduction in the concentration of digestible nutrients and an increase in structural content, which increases gut fill and reduces intake.

It was observed that TDMI of S steers was higher ($P < 0.05$) than that of C steers at IN (Table 3). However, at OUT, there were no differences ($P > 0.05$) in TDMI between S and C steers. This may be because forage DMI did not drop at IN with supplementation. Also, it is possible that animals accustomed to rotational grazing are willing to compromise DMI on the last days in a grazed paddock, because they can compensate the first few days after entering a new paddock. Supplemental grain increased ($P < 0.05$) TOMI by 14% in S steers (Table 3). Similar observation have been made in other studies (Young et al. 1980; Reis et al. 2001).

Methane Production

Although S steers gained 57 kg more over 142 d of grazing than C steers ($P < 0.05$), CH₄ production (L d⁻¹, L kg⁻¹ body weight) was not different between the two treatments ($P > 0.05$; Table 4) as was expected. Methane production (L kg ADG⁻¹) was 297.7 \pm 32.8 and 373.5 \pm 34.6 for S and C steers, respectively ($P = 0.16$). This could suggest that the 14% increase in TDMI with grain supplementation is not sufficient to cause a significant change in CH₄ production.

Daily CH₄ production ranged from 256 L d⁻¹ in the EARLY period to 342 L d⁻¹ in the LATE period. These val-

Table 3. Effect of grain supplementation (Diet), time of grazing (Period) and time in a paddock (Time) on total intake^a of steers (LS means \pm SE)

Factors	T DMI (kg d ⁻¹)	T DMI (% BW)	T OMI (kg d ⁻¹)
<i>Diet^a</i>			
C	11.1 \pm 0.3b	2.9 \pm 0.1b	10.5 \pm 0.3b
S	12.9 \pm 0.3a	3.2 \pm 0.1a	12.2 \pm 0.3a
<i>P</i> value	0.01	0.02	<0.01
<i>Period^b</i>			
EARLY	13.0 \pm 0.4a	3.6 \pm 0.1a	12.4 \pm 0.4a
MID	10.2 \pm 0.4b	2.6 \pm 0.1c	9.6 \pm 0.4c
LATE	12.7 \pm 0.4a	3.0 \pm 0.1b	12.0 \pm 0.4b
<i>P</i> value	<0.01	<0.01	<0.01
<i>Time^c</i>			
IN	15.6 \pm 0.3a	4.0 \pm 0.1a	14.8 \pm 0.3a
OUT	8.4 \pm 0.3b	2.1 \pm 0.1b	7.8 \pm 0.3b
<i>P</i> value	<0.01	<0.01	<0.01
<i>Diet \times Time</i>			
C \times IN	14.0 \pm 0.4b	3.7 \pm 0.1b	13.3 \pm 0.4b
C \times OUT	8.1 \pm 0.5c	2.1 \pm 0.1c	7.6 \pm 0.5c
S \times IN	17.2 \pm 0.4a	4.3 \pm 0.1a	16.3 \pm 0.4a
S \times OUT	8.6 \pm 0.4c	2.1 \pm 0.1c	8.1 \pm 0.4c
<i>P</i> value	<0.01	0.02	<0.01

^aTotal intake = forage intake + barley grain intake.

^bDiet: Con = (legume-grass pasture); Sup = (barley grain + legume-grass pasture).

^cPeriod (P) = EARLY (17–25 June), MID (20–29 July) and LATE (12–21 Aug.) period of grazing season.

^dTime (T) = IN (entry) and OUT (exit) of paddocks.

a–c Means within each factor followed by different letters differ ($P < 0.05$).

ues were comparable to those observed by McCaughey et al. (1997), who reported daily CH₄ production values between 242.1 and 306.7 L d⁻¹ from steers grazing pastures using the SF₆ tracer gas technique. Methane production varied between sampling periods, as a result of changes in forage quality and intake. McCaughey et al. (1999) also observed differences in CH₄ production between sampling periods from lactating beef cows grazing alfalfa-meadow bromegrass pastures. Variations in CH₄ production observed by McCaughey et al. (1999) were, however, attributed to changes in forage quality rather than voluntary intake, which was similar between sampling periods.

There were only marginal effects ($P > 0.05$) of supplementation on CH₄ production expressed as L CH₄ kg⁻¹ TOMI or CH₄, % of TGEI by grazing steers (Table 4). Similarly, Visser et al. (1998) showed that when silage diets were supplemented with three levels of flaked corn starch (0, 2 and 4 kg), the proportion of propionic acid increased (less CH₄) for cows fed diets containing early cut grass silage; however, no effects were found for cows fed late cut grass silage. There were no differences ($P < 0.05$) in energy losses as CH₄ when goats eating Italian ryegrass pellets (*Lolium multiflorum* Lam.) were supplemented with corn at 50% of the ration, although the efficiency of utilization of the forage was improved (Islam et al. 2000). Reis et al. (2001), however, reported a reduction in the acetate to propionate ratio and increased ruminal propionate concentration when 10 kg DM of a corn-based concentrate was

Table 4. Effects of grain supplementation on CH₄ production of grazing steers (LS means ± SE)

Factors	CH ₄ (L d ⁻¹)	CH ₄ (L kg BW ⁻¹)	CH ₄ (L kg ⁻¹ TDMI)	CH ₄ (L kg ⁻¹ TOMI)	CH ₄ (% TGEI)
<i>Diet^a</i>					
C	310.5 ± 25.9	0.80 ± 0.06	30.8 ± 2.9	32.8 ± 3.1	6.7 ± 0.63
S	331.2 ± 24.6	0.81 ± 0.06	29.4 ± 2.7	31.3 ± 2.9	6.4 ± 0.59
<i>P</i> -value	0.58	0.91	0.73	0.73	0.71
<i>Period^b</i>					
EARLY	256.3 ± 12.8 ^b	0.70 ± 0.03 ^b	21.7 ± 1.5 ^c	22.8 ± 1.6 ^c	4.7 ± 0.33 ^c
MID	363.9 ± 11.8 ^a	0.91 ± 0.03 ^a	38.2 ± 1.7 ^a	40.9 ± 1.8 ^a	8.4 ± 0.36 ^a
LATE	342.4 ± 11.8 ^a	0.81 ± 0.03 ^a	30.5 ± 1.5 ^b	32.4 ± 1.6 ^b	6.6 ± 0.33 ^b
<i>P</i> -value	<0.01	<0.01	<0.01	<0.01	<0.01
<i>Time^x</i>					
IN	325.5 ± 13.7	0.82 ± 0.04	22.1 ± 1.7 ^b	23.2 ± 1.8 ^b	4.6 ± 0.37 ^b
OUT	316.2 ± 13.0	0.80 ± 0.04	38.2 ± 1.8 ^a	40.9 ± 1.9 ^a	8.5 ± 0.38 ^a
<i>P</i> -value	0.63	0.46	<0.01	<0.01	<0.01

^aDiet: C = (alfalfa/grass pasture); S = (barley grain + alfalfa/grass pasture).

^bPeriod (P) = EARLY (17–25 June), MID (20–29 July) and LATE (12–21 Aug.) period of a grazing season.

^xTime (T) = IN (entry) and OUT (exit) of paddocks.

a–c Means within each factor followed by different letters differ ($P < 0.05$)

supplemented to lactating cows fed direct-cut grass-legume forages. The response of grain supplementation in reducing CH₄ has been shown to depend on the type of forage in the basal diet, and the level and type of supplementation (Farverdin 1991).

Sampling period and sampling time affected CH₄ production ($P < 0.01$), with the lowest CH₄ loss [% gross energy intake (GEI)] observed during the EARLY period and at entry into paddocks ($P < 0.01$; Table 4). The higher proportion of digestible nutrients and increased total digestible nutrient intake probably created a more rapid passage rate in the rumen and a shift in fermentation patterns towards less methanogenesis. McCaughey et al. (1999) observed that with higher intakes on alfalfa-grass pastures compared to grass-only pastures, energy loss through CH₄ production by lactating beef cows was 7.1 vs. 9.5% of GEI, respectively ($P < 0.05$). Energy lost as CH₄, %TGEI ranged from 4.7 to 8.4% during the study with an overall mean loss of 6.5 ± 0.3%, which is comparable to values from steers grazing alfalfa-meadow brome pastures (4.1–5.2%; McCaughey et al. 1997) and forage-fed heifers using the SF₆ tracer gas technique (7.2%) and respiratory calorimetry (7.1%; Johnson et al. 1994). Pasture quality, as dictated by the time of grazing and time in a paddock, influenced CH₄ production ($P < 0.05$) more than grain supplementation ($P > 0.05$) using the SF₆ tracer gas technique.

CONCLUSIONS

The SF₆ tracer gas technique was used to measure CH₄ production in grazing steers, under two management regimes. There were marginal effects of grain supplementation on CH₄ production in the study. Methane production was lowest at EARLY grazing and at entry into paddocks when forage quality was highest using the SF₆ tracer gas technique. The study implies that pasture quality plays a major role in the extent to which CH₄ production can be reduced with grain supplementation in grazing animals. Response to grain supplementation in grazing animals as a strategy for reduc-

ing greenhouse gases in the beef industry may be dependent on pasture quality and quantity.

ACKNOWLEDGEMENTS

The authors appreciate the statistical assistance of Dr. G. H. Crow. The field and technical assistance of Mr. Clayton Robins and staff of the Brandon Research Station, Agriculture and Agri-Food Canada, is much appreciated. The authors thank Ms. Janice Haines for her help with laboratory analyses. This trial was financially supported by the Agriculture and Agri-Food Canada Greenhouse Gas Initiative.

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