

# Methane abatement strategies for cattle: Lipid supplementation of diets

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Beauchemin, K. A., McGinn, S. M. and Petit, H. V. 2007. **Methane abatement strategies for cattle: Lipid supplementation of diets.** *Can. J. Anim. Sci.* **87**: 431–440. A study was conducted to investigate the impact of several lipid sources that supplied mainly long-chain fatty acids (FA), for their potential to reduce methane emissions from growing cattle. Sixteen Angus heifers (initial weight, 325 ± 41 kg) were used in the experiment, which was designed as a crossover with two groups, four 21-d periods, and four dietary treatments: control (no added lipid source), tallow, sunflower oil, and whole sunflower seeds. Lipid sources were added to supply 34 g fat kg<sup>-1</sup> of dietary dry matter (DM), bringing the total dietary fat content to about 59 g kg<sup>-1</sup> of DM. Adding tallow increased the dietary proportion of saturated FA (47 g 100 g<sup>-1</sup> of FA), whereas sunflower oil and seeds decreased the proportion (21 g 100 g<sup>-1</sup> of FA). The basal diets consisted of mainly whole-crop barley silage (650 g kg<sup>-1</sup> of DM). Compared with the control, ad libitum intake was reduced ( $P < 0.001$ ) with sunflower seeds, but not with tallow ( $P = 0.13$ ) or sunflower oil ( $P = 0.53$ ). About 14% less methane was emitted per animal when diets contained tallow or sunflower oil and 33% less methane was emitted when diets contained sunflower seeds ( $P < 0.001$ ), compared with the control diet (177.4 g d<sup>-1</sup>). Relative differences in methane emissions among lipid sources were maintained after correction for intake of DM or gross energy. The methane reduction caused by tallow and sunflower seeds was partly due to decreased diet digestibility. Digestibility of neutral detergent fiber in the total tract decreased ( $P < 0.05$ ) by 15% with tallow and by 20% with sunflower seeds compared with the control, with only a numerical reduction from control for sunflower oil (12%;  $P = 0.11$ ). Consequently, digestible energy intake was about 4% higher ( $P < 0.001$ ) for sunflower oil, but 3% lower ( $P = 0.02$ ) with tallow and 12% lower ( $P < 0.001$ ) with sunflower seeds, compared with the control. All lipid sources reduced methane emissions by an average of 17% when corrected for digestible energy intake (from 11.22 to 9.34 g methane Mcal<sup>-1</sup>;  $P = 0.01$ ). We concluded that adding about 3% lipid to high-forage diets in the form of saturated or unsaturated long-chain FA decreases methane emissions, and could have substantial effects on methane inventories if implemented commercially. All three lipid sources suppressed methane production, but sunflower oil has good potential for on-farm adoption because it had minimal effects of fiber digestibility, increased the intake of digestible energy and the rate of gain of cattle, and lowered methane production. Although tallow and sunflower seeds are usually cheaper sources of lipid than sunflower oil, their cost effectiveness as methane abatement strategies would also need to account for their potentially negative effects on digestible energy intake and performance of cattle fed high-forage diets.

**Key words:** Beef cattle, diet, fat, greenhouse gases, lipid; methane, oil

Beauchemin, K. A., McGinn, S. M. et Petit, H. V. 2007. **Strat  gies de r  duction des   manations de m  thane par les bovins : enrichissement de la ration avec des lipides.** *Can. J. Anim. Sci.* **87**: 431–440. Les auteurs ont tent   d'  valuer l'incidence de diverses sources de lipides fournissant surtout des acides gras (AG)    cha  ne longue sur la r  duction des d  gagements de m  thane par les bovins. L'exp  rience portait sur seize g  nisses Angus (poids initial de 325 ± 41 kg) et   tait structur  e en plan crois   de deux groupes, de quatre p  riodes de 21 jours et de quatre r  gimes : aucun apport de lipides (groupe t  moin), suif, huile de tournesol et graines de tournesol. Les lipides ont   t   ajout  s    la ration de fa  on    fournir 34 g de mati  re grasse par kg de mati  re s  che (MS), ce qui portait la concentration totale de la ration en lipides autour de 59 g par kg de MS. Le suif augmente la proportion d'AG satur  s dans la ration (47 g par 100 g d'AG) alors que l'huile et les graines de tournesol la diminuent (21 g par 100 g d'AG). La ration de base consistait essentiellement en ensilage d'orge entier (650 g par kg de MS). Comparativement au groupe t  moin, les graines de tournesol r  duisent l'ingestion quand la ration est servie    sati  t   ( $P < 0,001$ ), mais l'ingestion reste la m  me avec le suif ( $P = 0,13$ ) et l'huile de tournesol ( $P = 0,53$ ). Les sujets lib  rent environ 14 % moins de m  thane quand la ration contient du suif ou de l'huile de tournesol et 33 % moins quand on l'enrichit de graines de tournesol ( $P < 0,001$ ), comparativement    la ration t  moin (177,4 g par jour). La variation relative des   missions de m  thane d'une source de lipides    l'autre demeure, m  me apr  s correction pour l'ingestion de MS ou d'  nergie brute. La r  duction des d  gagements de m  thane attribuable au suif ou aux graines de tournesol r  sulte en partie de la moins grande digestibilit   de la ration. En effet, le suif diminue ( $P < 0,05$ ) la digestibilit   des fibres au d  tergent neutre dans l'ensemble du tube digestif de 15 %, et les graines de tournesol la r  duisent de 20 %, toujours par rapport    la ration t  moin, l'huile de tournesol n'entra  nant qu'une baisse num  rique (12 %;  $P = 0,11$ ). Par cons  quent, comparativement au groupe t  moin, on observe une hausse d'environ 4 % ( $P < 0,001$ ) de la quantit   d'  nergie digestible ing  r  e

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**Abbreviations:** ADF, acid detergent fiber; DM, dry matter; FA, fatty acids; GE, gross energy; NDF, neutral detergent fiber; VFA, volatile fatty acids

pour l'huile de tournesol, contre une baisse de 3 % ( $P = 0,02$ ) pour le suif et de 12 % ( $P < 0,001$ ) pour les graines de tournesol. Après correction pour l'ingestion d'énergie digestible (de 11,22 à 9,34 g de méthane par Mcal;  $P = 0,01$ ), les sources de lipides diminuent toutes les émissions de méthane d'en moyenne 17 %. On en conclut qu'ajouter environ 3 % de lipides sous forme d'AG à chaîne longue saturés ou insaturés à une ration riche en fourrages réduirait les émissions de méthane, avec les importantes conséquences que cela aurait sur l'inventaire de méthane si cette mesure était appliquée à la grandeur de l'industrie. Les trois sources de lipides freinent la production de méthane, mais l'huile de tournesol est celle qui pourrait être adoptée le plus facilement par les éleveurs car elle n'a que peu d'effets sur la digestibilité des fibres; elle augmente l'absorption d'énergie digestible et le taux de gain des animaux et elle réduit la production de méthane. Quoique le suif et les graines de tournesol soient des sources de lipides souvent moins onéreuses que l'huile de tournesol, l'évaluation de leur rentabilité en tant que mesure visant à réduire les émissions de méthane devraient prendre en compte leur incidence négative éventuelle sur l'absorption d'énergie digestible et le rendement des bovins surtout nourris de fourrages.

**Mots clés:** Bovins, régime, matière grasse, gaz à effet de serre, lipides, méthane, huile

Many governments have implemented policies to reduce greenhouse gas emissions because there is growing consensus that these emissions are linked to global warming and modern climate change (Karl and Trenberth 2003). Strategies that reduce enteric methane emissions from ruminants would help the agricultural sector meet these targeted reductions. Ruminant livestock are estimated to produce 15% of the global methane emissions (Lassey et al. 1997).

Enteric methane is produced by ruminants during the process of microbial digestion of feed. Typically 6 to 8%, but up to 12%, of the gross energy (GE) in feed is converted to methane gas in the rumen during this process (Johnson and Johnson 1995). The amount of feed consumed and the composition of the diet are the main factors affecting the amount of methane emitted (Johnson and Johnson 1995). Generally, as the intake of ruminally digestible carbohydrate increases, methane emissions increase.

Alterations in diet formulation have the potential to reduce enteric methane emissions (Monteny et al. 2006). In particular, it is well established that supplementation of diets with lipid sources reduces enteric methane emissions (Boadi et al. 2004). In our previous studies, sources of long-chain fatty acids (FA) such as sunflower oil and canola oil reduced methane emissions from cattle fed high-forage diets by up to 22% of GE intake when added at 45 g kg<sup>-1</sup> of dry matter (DM) (McGinn et al. 2004; Beauchemin and McGinn 2006a). Other sources of long-chain FA such as tallow and soybean oil have also been shown to reduce enteric methane (Johnson and Johnson 1995; Zinn and Plascencia 1996). Effectiveness of long-chain FA in suppressing methane is thought to be proportional to degree of unsaturation of the FA (Giger-Reverdin et al. 2003), although there is some uncertainty about this relationship (Johnson and Johnson 1995). Medium chain (i.e., C12:0 and C14:0) FA are also effective at reducing methane emissions (Machmüller 2006), but these lipid sources (e.g., coconut oil, genetically modified canola oil) are often cost prohibitive for livestock producers.

The potential for implementing lipid feeding on commercial farms as a methane mitigation strategy is high because lipids represent "natural" rather than chemical intervention. Furthermore, lipid sources such as oils, oilseeds, and animal fats are already used commercially by some Canadian cattle producers to increase energy density of the diet. Lipid

sources can have other benefits such as altering the FA composition of meat and milk, reducing the dustiness of feed, and increasing the absorption of fat-soluble nutrients [National Research Council (NRC) 2001].

Although adding lipid to the diet can reduce methane emissions, high supplementation rates can also reduce feed intake (Allen 2000) and fiber digestibility (Henderson 1973). Long-chain FA inhibit ruminal cellulolytic microbes (Maczulak et al. 1981), with degree of unsaturation and rate of release in the rumen positively associated with decreased ruminal fermentation (NRC 2001). Therefore, it is possible that feeding sources of saturated FA, such as tallow, could lower methane emissions without negative effects on fiber digestion. Alternatively, it is possible that the negative effects of feeding unsaturated long-chain FA on fiber digestion would be attenuated by feeding whole oilseeds that release lipids into the rumen more slowly than oil (Dhiman et al. 2000). For example, sunflower seeds contain over 40% fat and, even when fed in the unprocessed form, much of this fat is biohydrogenated in the rumen (Gibb et al. 2004). Therefore, feeding sunflower seeds may be a cost effective means of reducing methane emissions from cattle.

The purpose of our study was to investigate the impact of several sources of long-chain FA that are commercially available to Canadian cattle producers for their potential to reduce methane emissions. Our hypothesis was that enteric methane emissions of growing cattle fed a high-forage diet could be reduced with lipid supplementation and that the reduction in methane would be greater for lipid sources high in unsaturated FA (sunflower oil, sunflower seeds) than for sources high in saturated FA (tallow). A second objective was to compare the effects of sunflower oil and sunflower seeds, added to supply the same amount of lipid, on methane emissions. Sunflower seeds are typically a cheaper source of fat than sunflower oil because the cost of processing is eliminated. We hypothesized that these two lipid sources would be equally effective suppressants of methane, when added to supply the same amount of fat to the diet, but that the slow release of lipid from sunflower seeds would eliminate any potentially negative effects on fiber digestion.

## MATERIALS AND METHODS

### Experimental Design and Animals

The study was designed as a crossover design with two groups, four 21-d periods and four dietary treatments.

Sixteen Angus heifers were allocated to the two groups, with two cattle in each group fed one of four treatments. Animals were assigned to treatments to minimize crossover effects between periods and all animals received all diets by the end of the experiment. The groups were offset by 1 wk to facilitate measurements. All animals were cared for in accordance with the guidelines of the Canadian Council on Animal Care (1993).

At the start of the experiment, the heifers were about 1 yr old and weighed  $325 \pm 41$  kg (mean  $\pm$  SD). The heifers had been ovariectomized and used in a previous experiment (Beauchemin and McGinn 2006a); thus, they were conditioned to the facilities, including the environmental chambers. Animals were weighed at the start and end of each period to determine average daily gain. Weighings occurred 2 to 3 h after feed delivery and cattle were not fasted beforehand. From day 1 to 16 of each period, the heifers were housed individually in pens ( $4.9 \times 1.8$  m) equipped with a feeder and water bowl and bedded with straw. The pens were located in a sheltered, unheated barn. Daily feed intake and ruminal fermentation were measured in this facility. The cattle were then moved before the morning feeding on day 17 to four environmental chambers to measure enteric methane and total tract digestibility.

### Treatments and Diet

Four dietary treatments were evaluated (DM basis): control (no added lipid source), tallow, sunflower oil, and whole sunflower seeds. The lipid sources were added to supply  $34$  g fat  $\text{kg}^{-1}$  of DM, bringing the total dietary fat content to about  $59$  g  $\text{kg}^{-1}$  of DM. The basal diet consisted of mainly whole-crop barley silage ( $650$  g  $\text{kg}^{-1}$  of DM) supplemented with steam-rolled barley, and sources of protein, minerals, and vitamins (Table 1). The lipid sources replaced barley grain within the diet such that the forage to concentrate ratio was not changed. The diets were formulated to be iso-nitrogenous ( $127$  g  $\text{kg}^{-1}$  of DM) using soybean meal and canola meal as protein sources. The diets were also formulated to meet or exceed the mineral and vitamin requirements of cattle gaining  $1$  kg  $\text{d}^{-1}$  (NRC 1996).

A feed mixer (Data Ranger, American Calan, Inc., Northwood, NH) was used daily to prepare the total mixed rations. The sunflower seeds were incorporated into the ration using the feed mixer, but the sunflower oil and tallow rations were prepared using the feed mixer without the lipid sources, which were then mixed by hand into each animal's daily allocation of feed. The cattle were fed once daily at 0900 with feed offered for ad libitum intake (10% orts, DM basis). Quantities of feed offered and refused were recorded daily for each animal. Samples of diets and refusals were retained weekly for determination of DM content. The ad libitum DM intake (DMI) was calculated daily (DM offered minus the DM refused) for each heifer between days 7 and 16 inclusive.

### Ruminal Fermentation Measurements

Ruminal pH was measured 4 h after feeding on day 14 of each period using an oro-ruminal probe. A speculum was inserted through a lubricated rubber tube, which was insert-

ed into the esophagus and then the rumen via the mouth. Rumen contents (200 mL) were removed using an electric pump and samples were examined visually to ensure they were not contaminated with saliva. A pH meter (Accumet model 25, Denver Instrument Company, Arvada, CO) was used to measure pH immediately upon sampling. Afterwards, the ruminal fluid samples were kept frozen ( $-30^\circ\text{C}$ ) for analysis of volatile fatty acids (VFA) and ammonia. These samples were first prepared by squeezing whole ruminal contents through a layer of monofilament cloth with a pore size of  $250$   $\mu\text{m}$ . The filtrate (5 mL) was preserved with 1 mL of 25% (wt/vol)  $\text{HPO}_3$  for VFA analysis, with additional filtrate (5 mL) preserved with 1 mL of  $0.18$  M  $\text{H}_2\text{SO}_4$  for ammonia determination.

### Digestibility

Total tract digestibility of nutrients was determined using an external marker prepared from chromic oxide mixed with ground barley. Ten grams of marker, providing about 1.6 g of Cr, were added on top of each animal's feed allocation once daily during the last 10 d of each period. In all cases, the entire allotment of marker was consumed within the first few minutes of eating. Fecal samples (100 g wet weight) were collected twice daily at various times throughout the day to account for possible temporal fluctuations in Cr concentration. Sampling was from the rectum of each animal on the last 5 d of each period. The samples were pooled by day for each heifer within period, and immediately frozen. The samples were later dried at  $55^\circ\text{C}$  for 48 h in a forced-air oven, ground through a 1-mm screen (standard model 4, Arthur Thomas Co., Philadelphia, PA), and analyzed for DM, GE, neutral detergent fiber (aNDF), acid detergent fiber (ADF), crude protein (CP) and Cr. An additional fecal sample (100 g wet weight) was taken from each animal before dosing the marker each period. These samples were analyzed for DM and Cr. The Cr concentration in the feces taken pre-dosing was used to adjust for residual marker excretion, and in all cases this adjustment was very small. Chromium was assumed to be completely indigestible and the digestibility of DM was calculated as follows:

$$\text{DM digestibility} = 1 - (\text{Cr fed (mg d}^{-1}\text{)} / \text{DMI (kg d}^{-1}\text{)}) / \text{Cr in feces (mg kg}^{-1}\text{ of DM)}$$

where DMI was for the day on which the fecal samples were taken. Digestibility of GE, aNDF, ADF, and CP was calculated using the same approach. Intake of digestible DM and energy was calculated based on the intakes on the days of fecal sampling and the digestibility coefficients.

### Methane Measurements

Enteric methane emissions were measured the last week of each period using four whole animal chambers, as described by Beauchemin and McGinn (2005). The chambers measured  $4.4$  m wide  $\times$   $3.7$  m deep  $\times$   $3.9$  m tall ( $63.5$   $\text{m}^3$ ; Conviron Inc., model C1330, Winnipeg, MB). Two animals receiving the same diet were placed in each chamber, with the pairing of cattle within the chambers the same throughout the experiment. Each animal was housed within the

**Table 1. Ingredient composition (g kg<sup>-1</sup>, DM basis) of the diets**

Ingredient	Control	Tallow	Sunflower oil	Sunflower seeds
Barley silage <sup>z</sup>	650.0	650.0	650.0	650.0
Barley grain, steam-rolled	312.6	268.0	268.0	247.3
Sunflower seeds	0	0	0	89.3
Sunflower oil	0	0	34.0	0
Tallow	0	34.0	0	0
Canola meal	12.0	17.5	17.5	0
Soybean meal	12.0	17.5	17.5	0
Limestone	6.7	6.3	6.3	6.7
Salt	6.1	6.1	6.1	6.1
Vitamin and mineral premix	0.6	0.6	0.6	0.6

<sup>z</sup>The whole-crop barley silage contained (mean  $\pm$  SD) 4.16  $\pm$  0.027 Mcal kg<sup>-1</sup> of DM of gross energy, 111  $\pm$  4.9 g kg<sup>-1</sup> of DM of crude protein, 423  $\pm$  14.2 g kg<sup>-1</sup> of DM of neutral detergent fiber, and 258  $\pm$  9.8 g kg<sup>-1</sup> of DM of acid detergent fiber.

chamber in a stall equipped with a feeder. At least the first 12 h that animals were in the chambers was considered an adjustment period, and then methane emissions from each chamber were recorded for 3 consecutive 24-h days each period.

Air temperature within the chambers was maintained at 10°C. Each chamber was vented by a fresh-air intake and exhaust duct with dedicated fans on each duct. The flow rates of the intake and exhaust ducts generated a small positive pressure (< 2 Pa) inside the chamber, which prevented in-flow of gases into the chamber. The air volume within the chamber was exchanged every 5 min. The methane concentrations in the intake and exhaust air streams were monitored using a methane analyzer (Siemens Inc., model Ultramat 5E, Karlsruhe, Germany). The difference between the incoming and outgoing mass of methane was assumed to be the amount of methane generated in each chamber by the two animals.

The chamber doors were opened twice daily for feeding, cleaning, and fecal sampling and the corresponding fluxes were omitted. These daily interruptions had little impact on the daily emissions because the time constant (i.e., time required for gas concentrations to reach steady-state) for the chambers was 5 to 10 min. Below and to the rear of each stall (two per chamber) was a hole in the floor leading to the manure removal track. The floor of the chamber was cleaned each morning to remove the accumulated manure. A flexible rubber mat hanging vertically effectively sealed the chamber from the manure removal tract.

The chambers were calibrated before the experiment by releasing a known amount of methane into each chamber and calculating the recovered amount by the chamber. At this time, the released methane was matched with the recovered methane, by adjusting the air flow through the chamber by 1.224 to 1.764 times. This adjustment was needed because it is difficult to measure the mean air flow using a point measurement in large ducts. The air flows of the intake and exhaust ducts of each chamber were continually monitored throughout the experiment to ensure there was no change in air flow.

### Chemical Analyses

Chemical analyses were performed on each sample in duplicate, and where the coefficient of variation for the replicate

analysis was > 5%, the analysis was repeated. Samples of feed, orts and feces were dried in a forced air oven at 55°C for 48 h and then ground (1-mm screen, Wiley mill, Arthur Hill Thomas Co., Philadelphia, PA). The DM was determined by drying the samples at 135°C for 2 h, followed by hot weighing [Association of Official Analytical Chemists (AOAC) 2002; Method 930.15]. The OM content was then calculated as the difference between 100 and the percentage ash (AOAC 2002; Method 942.05). Gross energy was determined using an adiabatic calorimeter (model 1241, Parr, Moline, IL). The aNDF was determined as described by Van Soest et al. (1991) using heat stable  $\alpha$ -amylase and sodium sulfite, and ADF was determined according to AOAC (2002; Method 973.18). For the measurement of CP (N  $\times$  6.25), samples were ground using a ball mill (Mixer Mill MM2000, Retsch, Haan, Germany) to a fine powder. Nitrogen was quantified by flash combustion with gas chromatography and thermal conductivity detection (Carlo Erba Instruments, Milan, Italy). Crude fat was determined by extraction with ether using a Soxhlet system HT6 apparatus (Tecator, Fisher Scientific, Montreal, QC) according to the Method 920.39 (AOAC 2002). Fatty acid methyl ester profiles were measured by gas-liquid chromatography on a Hewlett-Packard 6890 chromatograph (Hewlett-Packard Ltd., Montreal, QC) with a G1315A autosampler equipped with a flame-ionization detector and a split-splitless injector, as described by Delbecchi et al. (2001). Chromium was determined by inductively coupled plasma emission spectrometry (SpectroCiros<sup>CCD</sup>, Spectro Analytical Instruments, GmbH & Co., Kleve, KG, Germany) after dry ashing and extraction. The VFA were quantified by gas chromatography (model 5890, Hewlett Packard, Little Falls, DE) with a capillary column (30 m  $\times$  0.32 mm i.d., 1- $\mu$ m phase thickness, bonded polyethylene glycol; Supelco Nukol, Sigma-Aldrich, Oakville, ON), and flame-ionization detection. Ammonia content of ruminal samples was determined using the method described by Weatherburn (1967) modified to use a plate reader.

### Calculations and Statistical Analysis

Cumulative daily methane emissions from each chamber were divided by two to obtain the average emission per animal. The daily methane emission was also expressed per kilogram of DMI and as a percentage of GE and digestible

energy intake, based on the intake of the cattle within the chamber on the same days that methane was measured. Data for digestibility were averaged over days for each animal within period.

The data were analyzed using a mixed model procedure (SAS Institute, Inc. 2002). The individual animal was the experimental unit for intake, average daily gain, digestibility, and rumen fermentation variables, because these data were obtained from individually penned animals. For these variables, the model included the fixed effect of diet. Animal nested within group and period nested within group were considered as random effects. The restricted maximum likelihood method was used for estimating the variance components, and the Kenward-Roger's option was used to adjust the degrees of freedom. Day (1 to 16) was considered a repeated effect for ad libitum DMI with animal within period within group as the subject.

For methane emission data, the chamber was considered as the experimental unit. The model included the fixed effects of treatment and the random effects of chamber nested within group and period nested within group. Day (1 to 3) was treated as a repeated measure with chamber by period by group as the subject. The variance-covariance error structure that was used was compound symmetry, unstructured, or first-order autoregressive as determined by the fit statistics. Differences among means were tested using a protected ( $P < 0.05$ ) LSD test. Treatment effects were declared significant at  $P < 0.05$  and trends were discussed at  $P < 0.10$ .

## RESULTS

Adding lipid sources to the diet increased dietary crude fat content (from 24.6 to 58.5 g kg<sup>-1</sup> of DM; Table 2). As expected, tallow increased the proportion of saturated FA in the diet (47 g 100 g<sup>-1</sup> of FA), whereas sunflower oil and seeds decreased (21 g 100 g<sup>-1</sup> of FA) the proportion compared with the control diet (32 g 100 g<sup>-1</sup> of FA). The combined effects of higher dietary fat content and changes in FA profiles resulted in the cattle fed the various diets consuming different amounts of saturated and unsaturated fats. Specifically, intake of saturated FA was about 3.4-times higher for cattle fed tallow compared with control (244 vs. 72 g d<sup>-1</sup>), but only 1.6- or 1.3-times higher for cattle consuming sunflower oil or sunflower seeds. Similarly, intake of unsaturated FA was 2.7- and 2.5-times higher for cattle consuming sunflower oil and sunflower seeds compared with control (418 and 383 vs. 154 g d<sup>-1</sup>) and only 1.8-times higher for cattle consuming tallow (276 g d<sup>-1</sup>).

Although lipid supplementation increased the energy density of the diets, GE intake was not statistically increased for lipid treatments compared with the control (Table 3). Cattle fed sunflower seeds had lower ( $P < 0.05$ ) DMI relative to the control, which led to reduced GE, CP, and aNDF intakes. A small, non-significant ( $P = 0.13$ ) drop in DMI occurred for cattle fed tallow compared with the control such that GE intake was similar, but aNDF and ADF intakes were reduced ( $P < 0.05$ ) by feeding tallow. There was a trend ( $P = 0.08$ ) for higher average daily gain for cattle fed sunflower oil ( $P = 0.02$ ) when compared with control or sunflower seeds ( $P = 0.03$ ).

Feeding sunflower seeds reduced the apparent total tract digestibility of DM and GE compared with the control ( $P < 0.05$ ) because of a substantial reduction ( $P < 0.05$ ) in fiber digestibility (29% reduction for aNDF and 25% reduction for ADF; Table 4). Tallow also reduced ( $P < 0.05$ ) aNDF digestibility by 15% and ADF digestibility by 20%. However, sunflower oil caused only a 12% numerical ( $P = 0.11$ ) reduction in aNDF digestibility compared with the control, with no effect ( $P = 0.47$ ) on ADF digestibility.

Reductions in fiber digestibility for cattle fed tallow or sunflower seeds led to decreases in digestible DMI. Compared with the control diet, digestible DMI was reduced ( $P < 0.05$ ) by 4% with tallow and by 17% with sunflower seeds, but not with sunflower oil ( $P = 0.33$ ). In fact, intake of digestible energy was about 4% higher for sunflower oil ( $P < 0.001$ ), but 3% lower with tallow ( $P = 0.02$ ) and 12% lower with sunflower seeds ( $P < 0.001$ ), compared with the control. Digestibility of CP was higher ( $P < 0.05$ ) for all diets containing lipid.

The only major effect of lipid feeding on ruminal fermentation was a reduction in ruminal ammonia ( $P < 0.001$ ), with no difference among lipid sources (Table 5). There was no effect of treatment on ruminal pH ( $P = 0.13$ ), total VFA ( $P = 0.15$ ), or individual proportions of VFA ( $P > 0.22$ ).

Dry matter intake on the days of methane measurements followed a similar trend as the ad libitum intakes, but the intakes of cattle in the chambers were about 2 to 4% lower than their intakes outside the chambers (Table 6). Compared with the control diet, methane emitted per animal was about 14% lower for diets containing tallow or sunflower oil and 33% lower for diets containing sunflower seeds ( $P < 0.001$ ). Relative differences in methane emissions among lipid sources were maintained after correction for intake. When compared on the basis of DMI, diets containing tallow or sunflower oil were 11% lower than the control, while the diet containing sunflower seeds was 23% lower than the control ( $P < 0.001$ ). Methane emissions as a percentage of GE intake were also reduced by lipid feeding with an average reduction of 15% for tallow or sunflower oil, and 25% reduction for sunflower seeds ( $P < 0.001$ ). The greater reduction in methane emissions for sunflower seeds compared with tallow and sunflower oil was mostly due to its lower digestibility. Expressed on the basis of digestible energy intake, all lipid sources reduced methane by 17% ( $P = 0.01$ ), with no differences among lipid sources.

## DISCUSSION

This study examined the potential of reducing enteric methane emissions from beef cattle by adding commercially available lipid sources to the diet. The lipid sources chosen provided mainly long-chain FA ranging in degree of saturation (tallow vs. sunflower) and rate of release into the rumen (sunflower oil vs. seeds). All lipid sources were added to the diets to provide about 33 g kg<sup>-1</sup> of DM, bringing the total fat content of the diet to about 59 g kg<sup>-1</sup> of DM. The sunflowers used in this experiment were fed whole because there is little advantage of rolling sunflower seeds in terms of cattle growth performance (Gibb et al. 2004), and processing adds to the cost. Generally, it is recom-

**Table 2. Chemical composition of the diets (mean ± standard deviation)<sup>z</sup>**

Item	Control	Tallow	Sunflower oil	Sunflower seeds
Dry matter (DM) (g kg <sup>-1</sup> )	487 ± 23.9	478 ± 26.0	477 ± 21.7	484 ± 23.3
Organic matter (g kg <sup>-1</sup> DM)	922 ± 3.1	908 ± 6.9	908 ± 6.9	926 ± 6.4
Gross energy (Mcal kg <sup>-1</sup> DM)	4.41 ± 0.061	4.53 ± 0.108	4.64 ± 0.119	4.67 ± 0.024
Crude protein (g kg <sup>-1</sup> DM)	124 ± 3.9	127 ± 4.1	127 ± 3.1	128 ± 1.1
aNDF (g kg <sup>-1</sup> DM)	329 ± 12.5	316 ± 11.4	322 ± 11.7	343 ± 13.9
ADF (g kg <sup>-1</sup> DM)	187 ± 9.7	175 ± 8.2	192 ± 5.5	210 ± 6.4
Fat (g kg <sup>-1</sup> DM)				
Crude fat	24.6 ± 0.73	58.6 <sup>y</sup>	58.6 <sup>y</sup>	58.4 ± 1.44
Saturated <sup>x</sup>	7.8	27.5	12.4	11.8
Unsaturated <sup>w</sup>	16.8	31.1	46.2	46.6
FA (g 100 g <sup>-1</sup> detected FA) <sup>v</sup>				
C16:0	25.78	26.84	14.24	13.78
C16:1	0.22	2.76	0.31	0.12
C18:0	1.65	14.52	4.34	3.95
C18:1 <sub>n9trans</sub>	0.07	2.25	0.22	0.06
C18:1 <sub>n9cis</sub>	13.37	29.24	22.81	17.35
C18:1 <sub>n7cis</sub>	1.27	1.70	1.20	0.85
C18:2 <sub>n6cis</sub>	43.79	12.34	49.97	57.62
C18:3 <sub>n3</sub>	7.91	2.08	3.01	2.71
Others	5.94	8.27	3.90	3.56
Saturated <sup>x</sup>	31.85	46.90	21.21	20.21
Unsaturated <sup>w</sup>	68.15	53.10	78.79	79.79
Polyunsaturated <sup>u</sup>	52.20	15.16	53.49	60.69

ADF, acid detergent fiber; aNDF, neutral detergent fiber.

<sup>z</sup>Based on weekly samples for dry matter, a pooled sample for the experiment for fatty acids, and period samples for the remaining analyses.

<sup>y</sup>Values are calculated based on the amount of fat manually added to the diet daily.

<sup>x</sup>Saturated = C8:0 + C10:0 + C12:0 + C13:0 + C14:0 + C15:0 + C16:0 + C17:0 + C18:0 + C20:0 + C21:0 + C22:0 + C23:0 + C24:0.

<sup>w</sup>Unsaturated = 100 - saturated.

<sup>v</sup>Only fatty acids with > 1 g 100 g<sup>-1</sup> are listed individually.

<sup>u</sup>Polyunsaturated = C18:2 + C18:3 + C20:2 + C20:3 + C20:4 + C20:5 + C22:2 + C22:4 + C22:5 + C22:6.

**Table 3. Ad libitum nutrient intakes and average daily gain of cattle fed a high-forage diet with various added lipid sources (n = 8)**

Item	Treatments				SEM	P value
	Control	Tallow	Sunflower oil	Sunflower seeds		
Intakes						
Dry matter (kg d <sup>-1</sup> )	9.18 <sup>a</sup>	8.86 <sup>a</sup>	9.05 <sup>a</sup>	8.22 <sup>b</sup>	0.322	< 0.001
Gross energy <sup>z</sup> (Mcal d <sup>-1</sup> )	40.5 <sup>a</sup>	40.0 <sup>ab</sup>	42.0 <sup>a</sup>	38.5 <sup>b</sup>	1.42	0.01
aNDF (kg d <sup>-1</sup> )	3.02 <sup>a</sup>	2.79 <sup>b</sup>	2.90 <sup>ab</sup>	2.79 <sup>b</sup>	0.102	0.03
ADF (kg d <sup>-1</sup> )	1.74 <sup>a</sup>	1.56 <sup>b</sup>	1.73 <sup>a</sup>	1.73 <sup>a</sup>	0.061	0.003
Crude protein (kg d <sup>-1</sup> )	1.14 <sup>a</sup>	1.12 <sup>a</sup>	1.15 <sup>a</sup>	1.06 <sup>b</sup>	0.043	0.006
Initial body weight (kg)	324	324	324	324	32.8	1.00
Average daily gain <sup>y</sup> (kg d <sup>-1</sup> )	0.91	0.97	1.10	0.93	0.066	0.08

ADF, acid detergent fiber; aNDF, neutral detergent fiber; SEM, standard error of the mean.

<sup>z</sup>Sunflower oil vs. control ( $P = 0.08$ ).

<sup>y</sup>Tallow vs. sunflower oil ( $P = 0.06$ ).

*a, b* Means in the same row without a common letter differ ( $P < 0.05$ ).

mended that total fat should not exceed 6 to 7% of the dietary DM otherwise a depression in feed intake can occur, negating the advantages of increased energy density of the diet (NRC 2001). The diets in our study were just below this upper threshold.

All three lipid sources evaluated in this study decreased methane emissions per day, with larger reductions for sunflower seeds. Cattle fed sunflower seeds produced less methane per day than cattle fed the other lipid sources because they ate less feed and the feed was less digestible. Sunflower seed was the only lipid source to decrease feed intake. The decrease in intake was likely due to the 30% lower digestibility of aNDF for the sunflower diet, which would have contributed to rumen fill. Intake of cattle fed

high-forage diets is limited by slow rate of digestion and slow rate of passage from the rumen (Allen 1996). In high-concentrate diets, adding sunflower seeds increases DMI because of the additional effective fiber and improvement in rumen function (Gibb et al. 2004).

Although sunflower seeds tended to suppress methane to a greater extent than tallow or sunflower oil per kilogram of DMI or GE intake, this trend was negated when digestible energy intake was considered. Thus, all lipid sources were equally effective in suppressing methane when differences in intake and fiber digestion were accounted for.

Lipid supplementation of diets reduces methane emissions by decreasing the amount of organic matter fermented in the rumen, the activity of methanogenic bacteria, and the

**Table 4. Apparent digestibility in the total tract and intake of digestible nutrients for cattle fed a high-forage diet with various added lipid sources (n = 8)**

Item	Treatments				SEM	P value
	Control	Tallow	Sunflower oil	Sunflower seeds		
Digestibility (%)						
Dry matter	60.2a	59.5ab	60.6a	56.2b	1.64	0.08
Gross energy	60.3	58.7	60.7	56.0	1.65	0.08
aNDF	38.6a	32.8b	34.1ab	27.4b	2.63	0.003
ADF	32.7a	26.3b	30.7ab	24.4b	2.66	0.02
Crude protein	52.7b	58.7a	59.2a	59.7a	1.92	< 0.001
Intake						
Digestible dry matter (kg d <sup>-1</sup> )	5.48a	5.26b	5.42a	4.55c	0.180	< 0.001
Digestible energy (Mcal d <sup>-1</sup> )	24.30b	23.55c	25.39a	21.42d	0.757	< 0.001

ADF, acid detergent fiber; aNDF, neutral detergent fiber; SEM, standard error of the mean.  
*a-d* Means in the same row without a common letter differ (*P* < 0.05).

**Table 5. Ruminal fermentation variables for cattle fed a high-forage diet with various added lipid sources (n = 8)**

Item	Treatments				SEM	P value
	Control	Tallow	Sunflower oil	Sunflower seeds		
Ruminal pH	6.90	6.80	6.79	6.96	0.059	0.13
Total VFA (mM)	91.3	83.7	85.9	83.6	2.66	0.15
VFA (mol 100 mol <sup>-1</sup> )						
Acetate (A)	62.2	61.0	61.4	60.6	1.00	0.22
Propionate (P)	22.3	23.4	23.4	24.1	1.06	0.24
Butyrate	10.7	10.9	10.4	10.2	0.35	0.41
A:P	2.88	2.70	2.71	2.58	0.156	0.15
Ammonia (mM)	4.07a	2.33b	1.80b	1.92b	0.376	< 0.001

SEM, standard error of the mean; VFA, volatile fatty acids.  
*a, b* Means in the same row without a common letter differ (*P* < 0.05).

**Table 6. Daily methane emissions and corresponding dry matter intake (DMI) for cattle housed in chambers and fed a high-forage diet with various added lipid sources (n = 4)<sup>z</sup>**

Item	Treatments				SEM	P value
	Control	Tallow	Sunflower oil	Sunflower seeds		
DMI (kg d <sup>-1</sup> )	8.86a	8.66a	8.64a	7.85b	0.274	0.002
Methane						
g heifer <sup>-1</sup>	177.4a	153.4b	152.7b	119.6c	7.20	< 0.001
g kg <sup>-1</sup> of DMI	20.0a	17.8b	17.7b	15.4b	0.73	0.002
% of GE intake <sup>y</sup>	6.67a	5.72b	5.60bc	4.97c	0.250	< 0.001
% of DE intake	11.22a	9.81b	9.33b	8.87b	0.544	0.01

GE, gross energy; DE, digestible energy.  
<sup>z</sup>All variables were measured over a 3-d period.  
<sup>y</sup>Sunflower oil vs. sunflower seeds (*P* = 0.06).

*a-c* Means in the same row without a common letter differ (*P* < 0.05).

number of ruminal protozoa, and through the use of hydrogen during the biohydrogenation process (Johnson and Johnson 1995). In our study, adding lipids to the diet decreased the amount of organic matter fermented in the rumen because the lipid sources replaced barley grain (starch). For the tallow and sunflower seed diets, a decline in the total tract digestibility of aNDF and ADF also reduced the amount of organic matter fermented. Furthermore, in the case of sunflower seeds, an additional decrease in the amount of organic matter fermented in the rumen occurred due to the drop in intake. It must be acknowledged that the DM digestibility of the control diet in this study was about 2 to 4% units lower than expected based on the range of 60.9

to 64.0% reported for diets containing 70 to 75% whole-crop barley silage (McGinn et al. 2004; Beauchemin and McGinn 2005; Beauchemin and McGinn 2006a; Beauchemin et al. 2007). The lower than expected digestibility was likely attributed to slightly lower-quality barley silage used in the present study (estimated TDN of barley silage was 57% vs. 58 to 63% in the previous studies).

Expressing methane emissions per unit of digestible energy is useful in that it eliminates the confounding effects of intake and digestibility. The fact that the three lipid sources tested were equally effective when differences in intake and fiber digestion were accounted for indicates that the biohydrogenation process may play only a minor role in reducing

methane emissions, as suggested by Johnson and Johnson (1995). This conclusion contrasts to the findings of Giger-Reverdin et al. (2003) that depression in methane per unit of DMI is proportional to degree of unsaturation of the FA. It is possible that the positive relationship between methane per unit of DMI and degree of unsaturation of FA reported previously by Giger-Reverdin et al. (2003) was confounded by negative effects of the unsaturated FA on digestibility and intake. Alternatively, it is possible that the differences in intake of saturated and unsaturated FA by the cattle fed the various diets were too small to create detectable differences in methane production due to biohydrogenation of FA.

This study demonstrates that supplementing beef cattle diets with dietary lipids reduces methane emissions, which could reduce national greenhouse gas inventories. Inventories of enteric methane emissions are calculated by many countries using the Intergovernmental Panel on Climate Change (IPCC) guidelines (IPCC 1996). The IPCC Tier 1 method involves multiplying the number of cattle in each livestock class by a fixed emission factor per animal. The emission value ( $\text{g d}^{-1}$ ) observed in our study for the control diet is about 38% higher than the Tier 1 default methane emission value for non dairy cattle ( $129 \text{ g d}^{-1}$ ; cool climate countries). It is well known that emission values per animal are associated with a high degree ( $\pm 50\%$ ) of uncertainty, and that the IPCC default values underestimate emissions from cattle fed ad libitum (Beauchemin and McGinn 2006b). Thus, countries are encouraged to use the IPCC Tier 2 method, which accounts for intake of the cattle. The Tier 2 method calculates emissions from the number of cattle in each livestock class, GE intake, and methane conversion rates (% of GE intake) for specific diets. A default methane conversion rate of 6% is used for all diets, except feedlot diets containing more than 90% concentrate (DM basis). Thus, the IPCC default conversion rate of 6% is actually 10% lower than the conversion rate of 6.67% observed for the control diet in this study. The reductions in methane conversion rate (% of GE intake), ranging from 15 to 25%, observed in this study with lipid supplementation of diets would have significant impact on methane inventories when calculated using the Tier 2 approach.

Use of supplementary lipids can increase the energy intake of cattle and the resulting average daily gain if the negative effects on fiber digestion and intake outweigh the increase in energy density of the diet. Such was the case with sunflower oil where the increase in digestible energy intake corresponded to higher average daily gain compared with control cattle. These results contrast with those for cattle fed tallow or sunflower seeds, which had lower or similar digestible energy intake compared to the control cattle. In commercial cattle feeding, lipids are typically added to high-concentrate diets fed to finishing cattle (Krehbiel et al. 1995). Thus, the potential harmful effects on fiber digestion are minimized by feeding a high-concentrate diet low in fiber.

It is well known that feeding lipids can inhibit fiber digestion in the rumen (Maczulak et al. 1981; Jenkins 1993). Degree of unsaturation of the FA and rate of release in the

rumen are thought to be positively associated with a decrease in ruminal fermentation (NRC 2001). However, our results do not concur with this general trend. The depression in fiber digestion was statistically similar for sunflower oil and seeds, and numerically greater for sunflower seeds, giving no support to the theory that feeding sunflower seed limits the negative effect of oil on fiber digestion by slowing the release of lipid into the rumen (Dhiman et al. 2000). Furthermore, there was little difference in the negative effect of tallow and sunflower oil on fiber digestion, thus unsaturated FA were not more harmful to fiber digestion. It is possible that the harmful effects of feeding unsaturated FA that are not slowly released in the rumen would have been greater had a higher proportion of added lipid been used in the study. For example, in a previous study, we observed a 33% decrease in aNDF digestibility and a 29% decrease in ADF digestibility when 4.5% (DM basis) sunflower oil was added to a high-forage diet similar to the one used in the present study (McGinn et al. 2004). It appears that lowering the proportion of added fat from 45 to 33  $\text{g kg}^{-1}$  of DM, as was the case in the present study, reduced the negative effects of sunflower oil on fiber digestion.

Despite a decreased amount of organic matter fermented in the rumen for lipid-supplemented diets, there was surprisingly little change in ruminal fermentation variables, other than ammonia. It is possible that the lack of treatment effect on total VFA and proportions of individual VFA was due to the limited sampling schedule for ruminal fluid. Also, samples taken from the rumen do not reflect VFA production because they do not account for differential absorption of VFA from the rumen.

Feeding lipid is known to reduce protozoal numbers in the rumen (Ivan et al. 2004). The net result is decreased intra-ruminal N recycling, increased microbial protein synthesis, and increased flow of metabolisable protein to the duodenum (Clark et al. 1992). The substantial decrease in ruminal ammonia concentrations observed for the lipid-supplemented diets is evidence in support of reduced protozoal numbers (Ivan et al. 2000, 2004). Ammonia concentrations decrease because of the greater uptake of ammonia resulting from greater microbial growth. Increased microbial protein synthesis would be expected to increase the quality of the metabolisable protein flowing to the duodenum (Clark et al. 1992). This effect could account for the higher N digestibility in the total tract observed for lipid-supplemented diets. Increased protein digestibility would be an additional benefit to cattle producers implementing lipid feeding, as it could reduce the cost of feeding supplementary feed protein.

In summary, adding 33  $\text{g fat kg}^{-1}$  of DM to high-forage diets in the form of saturated or unsaturated long-chain FA decreased methane emissions as a percentage of GE intake by 15 to 25%. Reduced methane conversion rate would have substantial effects on national methane inventories when calculated using the IPCC Tier 2 method [an example of application of the Tier 2 method in calculating inventories is given by Basarab et al. (2005)]. When differences in digestible energy intake were accounted for, all three lipid sources were equally effective suppressants of methane

regardless of their FA composition. Thus, the hypothesis that the reduction in methane is proportional to degree of unsaturation of the FA was rejected. Furthermore, there was no evidence that the slow release of oil from sunflower seeds eliminates the negative effects of lipids on fiber digestion.

### CONCLUSIONS

This study demonstrates that several commercially available lipid sources have the potential to reduce enteric methane emissions from cattle, when added to the diet to supply 3.3% added fat. Specifically, tallow and sunflower oil reduced methane production as a percentage of GE intake by 15%, with a reduction of 25% for sunflower seeds. When differences in digestible energy intake were accounted for, all three lipid sources reduced methane emissions by 15%. Mitigating methane losses from cattle will have long-term environmental benefits in terms of reducing agriculture's contribution to greenhouse gas emissions. However, these ingredients may increase the cost of feeding, thus use of lipid feeding on commercial beef cattle farms for methane abatement will depend on the economic benefits determined by their effects on animal performance. Based on this short-term feeding study, feeding sunflower oil appears to have good potential for on-farm adoption because it increased the rate of gain of cattle in addition to lowering methane. Although tallow and sunflower seeds are usually cheaper sources of lipid than sunflower oil, their cost effectiveness as methane abatement strategies would also need to account for their negative effects on intake of digestible energy, which could reduce cattle performance. Further work to incorporate a life cycle analysis of feeding oil to cattle should follow to fully understand the impact on the entire greenhouse gas emissions.

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