1	Running head: Feed efficiency and methane emissions in beef cattle
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3	Methane and carbon dioxide emissions from yearling beef heifers and mature cows
4	classified for residual feed intake under drylot conditions
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22 Manafiazar, G., Baron, V.S., McKeown, L., Block, H., Ominski, K., Plastow, G., and Basarab

J.A. 2018. Methane and carbon dioxide emissions from yearling beef heifers and mature cows classified for residual feed intake under drylot conditions

This study quantified methane (CH_4) and carbon dioxide (CO_2) production from beef heifers and 25 cows classified for residual feed intake adjusted for off-test backfat thickness (RFI_{fat}), and reared 26 27 in drylot during cold winter temperatures. Individual performance, daily feed intake and RFI_{fat} were obtained for 1068 crossbred and purebred yearling heifers (eight trials) and 176 crossbred 28 mature cows (six trials) during the winters of 2015-2017 at two locations. A portion of these 29 30 heifers (147 high RFI_{fat}; 167 low RFI_{fat}) and cows (69 high RFI_{fat}; 70 low RFI_{fat}) were monitored for enteric CH₄ and CO₂ emissions using the Greenfeed Emissions Monitoring (GEM) system (c-31 32 lock Inc., Rapid City, SD, USA). Low RFI_{fat} cattle consumed less feed (heifers, 7.80 vs. 8.48 kg DM d⁻¹; cows, 11.64 vs. 13.16 kg DM d⁻¹; P<0.001) and emitted less daily CH₄ (2.5% for 33 heifers; 3.7 % for cows) and CO₂ (1.4% for heifers; 3.4% for cows) compared with high RFI_{fat} 34 cattle. However, low RFI_{fat} heifers and cows had higher CH₄ (6.2% for heifers; 9.9% for cows) 35 and CO₂ yield (7.3% for heifers; 9.8% for cows) per kg DMI compared with their high RFI_{fat} pen 36 mates. The GEM system performed at air temperatures between +20 and -30 °C. Feed intake of 37 38 heifers and mature cows were differently affected by ambient temperature reduction between +20 and -15° C, and similarly increased their feed intake at temperatures below -15° C. In 39 conclusion, low RFIfat animals emit less daily enteric CH4 and CO2, due mainly to lower feed 40 41 consumption at equal body weight, gain and fatness. These results further support the potential of selecting for low RFI as a strategy for reducing greenhouse gas emissions in beef cattle. 42 43 **Key words:** Beef cattle, methane, carbon dioxide, residual feed intake, drylot, cold temperature

- 44 Abbreviations: ADF, acid detergent fiber; ADG, average daily gain; BW, body weight; CH4,
- 45 methane; CO₂, carbon dioxide; DMI, dry matter intake; GEM, GreenFeed emission monitoring,
- 46 **KIN**, Roy Berg Kinsella Research Station; **LRDC**, Lacombe Research and Development Center;
- 47 **MMBW**, mid-test metabolic body weight; **NDF**, neutral detergent fiber; **RFI**, residual feed
- 48 intake; **RFI**_{fat}, residual feed intake adjusted for off-test back fat thickness; **SDMI**, standardized
- 49 dry matter intake; SF₆, sulphur hexafluoride; RC, respiratory chambers

51	Genetic selection for low residual feed intake (RFI) has been suggested as a permanent and
52	cumulative strategy for reducing methane (CH ₄) and greenhouse gas (GHG) emissions while
53	improving profitability of beef cattle production (Alford et al. 2006). Residual feed intake is
54	moderately heritable (0.29-0.46) and moderately repeatable (0.33-0.62) across animal types and
55	diets (Crews et al. 2003; Kelly et al. 2010 <i>a</i> ; Basarab et al. 2011; Durunna et al. 2013). However,
56	the actual GHG mitigation potential of selecting for low RFI (efficient) cattle remains uncertain
57	(Hegarty et al. 2007; Fitzsimons et al. 2013; McDonnell et al. 2016; Alemu et al. 2017) as CH ₄
58	emissions have rarely been measured under actual beef cattle production systems, particularly in
59	cold environments.
60	Respiratory chambers (RC) and sulphur hexafluoride (SF ₆) are widely used techniques to
61	measure individual cattle CH ₄ emissions (g d ⁻¹). Respiratory chambers are often referred to as the
62	"gold standard" for emission measurements, with individual animals studied for 1-3 d (Alemu et
63	al. 2017). However, inside the chamber the animal's feed intake is reduced, feeding behaviours
64	are altered, and short-term CH4 measurements are unlikely to reflect longer-term CH4 emissions
65	in production systems (Hegarty et al. 2007; Harper et al. 2011; Alemu et al. 2017). The SF_6
66	tracer technique can be used under grazing conditions but requires daily handling to exchange
67	collection chambers, as well as cleaning and maintenance of equipment between measurements
68	(Harper et al. 2011). This limits the number of animals that can be monitored by SF_6 , and it is
69	more appropriate for dairy cattle that are more accustomed to daily handling than beef cattle
70	(Harper et al. 2011). Alternatively, the GreenFeed Emissions Monitoring (GEM) system (c-lock
71	Inc., Rapid City, SD, USA) is a non-invasive method used to monitor CH ₄ emissions from
72	individual animals. The GEM system requires less animal training, and emissions can be
73	measured repeatedly in real-time over multiple 24-hr periods without the need to remove animals

74	from their production environments (Manafiazar et al. 2017; Arthur et al. 2017). Numerous
75	studies have used the GEM, RC and SF ₆ techniques to quantify CH ₄ emission under various
76	management and nutritional strategies (Hammond et al. 2016; Manafiazar et al. 2017; Arthur et
77	al. 2017). Hegarty et al. (2007) monitored CH_4 emissions from 76 Angus steers using the SF_6
78	technique and concluded that a one kg reduction in breeding value for RFI decreased daily CH4
79	emission by 13.38 g d ⁻¹ , and low RFI heifers emitted 25% less CH ₄ daily than high RFI heifers.
80	Alemu et al. (2017) monitored 16 replacement heifers during May-June, previously classified for
81	RFI adjusted for off-test backfat thickness (RFI _{fat}), for 25 days using GEM and RC systems, and
82	reported lower CH ₄ emissions (10% by GEM; 5% by RC) in low RFI _{fat} compared with high
83	RFI_{fat} replacement heifers. However, CH_4 yields were similar between high and low RFI_{fat}
84	groups, respectively, at 27.7 and 28.5 g kg DMI ⁻¹ for GEM and 26.5 and 26.5 g kg DMI ⁻¹ for RC.
85	Fitzsimmons et al. (2013) tested 22 Simmental heifers using the SF ₆ method and reported that
86	low RFI animals emitted less CH ₄ compared to high RFI animals (260 vs. 297 g d^{-1}) and had
87	lower CH ₄ production (2.5 vs. 2.9 g kg BW ^{-0.75}). Overall, studies quantifying the mitigation
88	potential of RFI have had low animal number or used techniques that have a significant impact
89	on the animals' feeding behaviours. In addition, over 80% of Canadian beef cattle are located in
90	the western provinces and exposed to cold (>-20°C) winter temperatures and wind chill that
91	affect feed intake (Webster et al. 1970; National Research Council 2016). Thus, our objective
92	was to compare CH_4 and carbon dioxide (CO_2) emissions from beef cattle classified for RFI_{fat}
93	and reared in drylot during cold winter conditions using the GEM system.

94 MATERIALS AND METHODS

95 Animal care committees at the University of Alberta and Lacombe Research and Development

96 Center approved the study, and all animals were cared for in accordance with the guidelines of

97 the Canadian Council on Animal Care (Olfer. et al. 1993).

98 Data Acquisition and Animals

99 *Performance data*

This study consisted of eight trials using crossbred and purebred yearling beef heifers ranging in 100 age from 9.2-10.9 mo and six trials using crossbred beef cows ranging in age from 2.6-7.2 yr at 101 start of trial. Trials were conducted during the winters of 2015, 2016, and 2017 at the Roy Berg 102 103 Kinsella Research Station (KIN) and Lacombe Research and Development Center (LRDC), Alberta, Canada. In 13 of 14 trials a minimum of 21 d adaptation period was followed by a 72-86 104 d measurement period. In Trial 3 a 21-d adaptation period was followed by a 114 d feed intake 105 106 measurement period due to measuring three groups of heifers through the GEM system. Heifers at LRDC were crossbreds consisting of Aberdeen Angus × Hereford and Charolais × Red-107 Angus, while heifers at KIN were primarily crossbreds of Aberdeen Angus, Charolais, Hereford, 108 and Limousin and purebred Aberdeen Angus and Charolais. During each trial, animals were co-109 mingled in multiple drylot pens (46 m² per animal) fitted with 24 automated feeding stations 110 (GrowSafe Systems Ltd., Airdrie, AB) at LRDC and 40 stations at KIN (minimum of 29.3 111 m^2 per animal) resulting in feeding density of 6-8 animals per GrowSafe station. Daily feed 112 intake (kg DM d^{-1}) and feeding event behaviors (events d^{-1} ; duration, min d^{-1} ; head down time, 113 min d^{-1}) were measured as previously described by Basarab et al. (2003, 2011). Replacement 114 heifers were fed a high forage diet *ad libitum*, twice daily, while cows were fed a high forage-115 116 based diet once daily at KIN and LRDC (Table 1). Feed samples were collected weekly and 117 stored at -20°C until they were composited by month and analyzed for dry matter (DM), calcium,

118 phosphorus, crude protein, neutral detergent fibre (NDF) and acid detergent fibre (ADF). Dry matter was determined by drying a sample of the diet at 80 °C in a forced-air oven to a constant 119 weight. Calcium and phosphorus contents of the samples were determined by AOAC method 120 985.01 with the following modifications; 0.35 g of sample was ashed for 1 hr at 535 °C, digest in 121 open crucibles for 20 min in 15% nitric acid on hotplate, and samples were diluted to 50 ml and 122 123 analyzed on ICP (AOAC 2000). Nitrogen content was determined by Kjeldahl with crude protein calculated as $6.25 \times N\%$ (AOAC 2000). Neutral detergent fibre and ADF were determined 124 according to Van Soest method (Van Soest et al. 1991). Diet ingredients and chemical 125 126 compositions for each trial are presented in Table 1. Animals had ad libitum access to water, and pens were bedded with woodchips as needed. Animals were weighed prior to morning feeding 127 on two consecutive days at the start and end of the feed intake test period and at approximately 128 28-d intervals throughout. Each animal was measured for backfat thickness at the 12-13th rib 129 (mm) using an Aloka 500V diagnostic real-time ultrasound with a 17 cm 3.5 M Hz linear array 130 transducer (Brethour 1992) at the end of the trial. 131

132 *RFI*_{fat} calculation

Residual feed intake adjusted for off-test backfat thickness (RFI_{fat}) was calculated for each 133 134 animal within trial using performance and feed intake data. Details of RFI_{fat} calculation have been described by Basarab et al. (2007, 2011) and Manafiazar et al. (2015). Briefly, mean daily 135 feed intake as measured by the GrowSafe[®] Feed Intake system, and mean daily pellet intake as 136 137 measured by the GEM system were each multiplied by their respective DM content and then summed to give total mean daily dry matter intake (DMI). Standardized DMI (SDMI) was 138 139 calculated for each animal by multiplying total mean daily DMI by the metabolizable energy content of the diet, and then dividing by 10 to standardize the diet to 10 MJ ME kg DM⁻¹. Initial 140

141 on-test body weight, metabolic mid-weight (MIDWT) and average daily gain (ADG) were calculated from the linear regression of each animal's observed BW against day on test (Basarab 142 et al. 2003; Manafiazar et al. 2015) using PROC GLM (SAS 2016). The RFIfat was calculated as 143 the difference between SDMI and predicted SDMI for each animal using the following model: 144 Model 1: $Y_{ijk} = \beta_0 + PEN + \beta_1 ADG_i + \beta_2 Metabolic MIDWT_j + \beta_3 UBFEND_k + e_{ijk}$ 145 where, Y_{ijk} is the SDMI for animal *ijk*, β_0 is the regression intercept, *Pen* is the fixed effect of 146 pen, β_1 is the partial regression coefficient of SDMI on ADG (kg d⁻¹), β_2 is the partial regression 147 coefficient of SDMI on metabolic mid-weight (kg), β_3 is the partial regression coefficient of 148 149 SDMI on final ultrasound backfat thickness (UBFEND, mm), and e_{ijk} is the random error term. Animals were excluded from RFI_{fat} calculation if their linear growth curve had coefficients of 150 151 determination (\mathbf{R}^2) of less than 0.90.

152 Enteric CH₄ and CO₂ measurements

Simultaneously with the RFI tests, one GEM system was located in a pen within each location 153 (LRDC and KIN), and different groups of animals rotated through the GEM pen to be monitored 154 for enteric CH₄ and CO₂ emissions. Animals voluntarily visited the GEM system, which 155 156 recorded beginning and end time of each visit, visit time and number of visits per day. Animals were accustomed to visit and consumed pellets from the GEM system two weeks before each test 157 period. The GEM systems were programmed such that each animal was allowed to receive a 158 159 maximum of six pellet drops from the unit every four hr and could have up to six visits per d for a maximum of 36 pellet drops per d. Average pellet drop weight was 31.1 g (SD=0.2), 30.2 g 160 (SD=0.3) and 32.7 (SD=0.2) in 2015, 2016 and 2017, respectively, for the unit located in LRDC. 161 Average pellet drop weight was 40.8 g (SD=0.2), 39.8 g (SD=0.1) and 39.3 g (SD=0.2) in 2015, 162 2016 and 2017, respectively, for the unit located in KIN. Pellets consisted of barley, corn 163

distillers grain screenings, soy meal, wheat millrun, beef vitamin/trace, mineral pre-mix, CaCO3,
chelated zinc, manganous oxide, and sodium chloride (Masterfeeds, AB, Canada). Average pellet
chemical compositions were 79.7% TDN, 14.7% CP, 5.5% ADF, 14.2% NDF, 2.04% Ca, and
0.47% P.

Methane emissions were measured from the GEM system as described by Manafiazar et 168 169 al. (2017). In brief, continuous and negative air flow from a system fan draws air past the animal's nose and mouth when it enters the shroud, thus mixing air with respired and/or 170 eructated CH₄ and CO₂. This mixture is drawn up a collection pipe, remixed, sampled and 171 172 analyzed by a nondispersive infrared analyzer. The unit also continuously collects samples from background CH₄ and CO₂ concentrations in a similar manner without presence of animals. The 173 174 units were calibrated weekly for CH₄ and CO₂ using a zero (semi-pure nitrogen) and span gas $(CO_2 = 5054 \text{ ppm for KIN unit, and } CO_2 = 5144 \text{ ppm for LRDC unit, and } CH_4 = 487 \text{ ppm and the}$ 175 balance gas of nitrogen for both units), and dilution rate was determined by releasing a small 176 amount (~10 ml) of propane every 5 hr. The CO₂ recovery test was performed at the beginning 177 of each trial and every month throughout. The recovery rate was 99% \pm 5.5% for 3 min of CO₂ 178 release into the GEM system. Raw data included time of visits, CH₄ and CO₂ concentration from 179 180 background, sum of animal respiration and eructation, dilution factor, and calibration information were uploaded to c-lock Inc., where it was stored, processed and then downloaded 181 182 through the internet for further analysis. More details on data processing from GEM and 183 operative procedure are described by Manafiazar et al. (2017).

184 Climate Data

185 Climate condition data was acquired from Alberta Agriculture online weather data site

186 (http://www.agric.gov.ab.ca/acis/alberta-weather-data-viewer.jsp). Climatic data for LRDC were

187 from a weather station located one km east of the outdoor feed intake pens, while KIN climatic

- 188 data was collected from a weather station located 5 km straight north of the outdoor feed intake
- pens. These data included year, month, day, daily precipitation (mm), air temperature (°C),

190 minimum and maximum of air temperature ($^{\circ}$ C), relative humidity (%), wind speed (km h⁻¹) and

- 191 wind direction ($0-360^\circ$ with 0 being North). A wind chill index was calculated as:
- 192 Wind chill index, $^{\circ}C = 13.12 + 0.6215T 11.37V^{0.16} + 0.3965T \times V^{0.16}$, where T=ambient
- 193 temperature in degrees C and V = wind speed in km hr^{-1} .

194 Data preparation and statistical analysis

195 Data preparation

Raw data generated by the GEM system and uploaded to c-lock Inc. website were stored in files 196 197 named COMMAND, DATA, FEED, and RFID. The COMMAND file includes date and time of commands for calibration, heater on or off, feeder on or off, and fan on or off. The DATA file 198 has time (MM/DD/yyyy HH:mm:ss), released pellets associated with time, and drop setting. The 199 FEED file contains time, animal identification and received drops associated with the time, and 200 drop setting. The RFID file contains animal identification, the time when an animal was detected 201 and when the animal left the unit. The raw data was downloaded to a local PC, and pre-202 203 developed EXCEL worksheets from c-lock Inc. were used to compile the raw data and generate CH₄ and CO₂ emissions, visit frequency, total valid visit duration, and visit time for each visit 204 (MM/DD/yyyy HH:mm:ss). Total valid visit duration was defined as the amount of time spent in 205 206 the GEM system where the animal's head was continuously in the shroud within 20 cm of the proximity sensor for a minimum of two min. Total number of drops per day for each animal was 207 208 also extracted from the c-lock Inc. webpage. Visit data were then converted to daily emission 209 data using SAS software program (SAS 2016) by rolling average for CH_4 and CO_2 and by

210 summing total valid visit duration and visits frequency per day for further analysis. Daily data

generated by GEM were matched to daily feed intake data using animal identification and date. 211

Enteric CH₄ and CO₂ production are expressed as emission (g d^{-1}) and yield (g kg⁻¹ DMI). 212

Descriptive statistics were attained using PROC MEANS in SAS statistical software package 213

(SAS 2016). 214

A total of 1133 animals were tested for daily feed intake, but 65 animals were excluded 215 from RFI_{fat} calculation as their linear growth curve had coefficients of determination (R²) of less 216 than 0.90. The number of animals in each feed efficiency test, start date and number of animals 217 that voluntarily visited the GEM system are presented in Table 1. Heifers averaged 2.75 visits d⁻¹ 218 and cows averaged 2.40 visits d⁻¹ to GEM system and were monitored on average 43 d (ranged 219 from 20 to 63 d) within trial. A total of 28 heifers and eight cows were excluded from analysis 220 221 since they had lower than 20 spot visits to the GEM system during the monitoring times (Manafiazar et al. 2017), leaving 314 heifers (147 high and 167 low RFI_{fat} heifers in eight trials) 222 and 139 cows (69 high and 70 low RFI_{fat} cows in six trials) for further analysis (Table 2). 223 Response variables included CH₄ and CO₂ emission (g d⁻¹), yield (g kg DMI⁻¹), and ratio (g CH₄ 224 g CO₂⁻¹), and number of visits to GEM (visits d⁻¹), pellet intake (kg d⁻¹), DMI (kg d⁻¹), average 225 daily gain (ADG, g d⁻¹), and ultrasound off-test back fat thickness (mm). 226 227

Statistical analysis

Enteric gas and performance traits were subjected to analysis of covariance using PROC MIXED 228

229 in SAS statistical software package (SAS 2016). Mid-point weight, ADG, RFI, and off-test back

fat thickness were subjected to analysis using Model I that included RFIfat group, trial, and 230

interaction of RFI_{fat} group by trial as fixed effects and animals nested in days of test and error as 231

232 random effects.

- 233 Model I: $Y_{ijkl} = \mu + RFI_i + Trial_j + RFI \times Trial_{ij} + Animal (Day)_{kl} + e_{ijkl}$
- Enteric CH₄ and CO₂ traits were subjected to analysis using Model II with fixed and random
- effects as described for Model 1, as well as number of visits over the test period (Nvisits) and
- total of valid visit duration time during the test period (Tvisits) as covariates.
- 237 Model II: $Y_{ijklmn} = \mu + RFI_i + Trial_j + RFI_x Trial_{ij} + Animal (Day)_{kl} + \beta_1 Nvisits_m + \beta_2 Tvisits_n$
- $_{238}$ + e_{ijklmn}
- 239 The PROC SGSCATTER was used to generate graphs for the relationships between ambient
- temperature and traits of interest, while PROC REG was used to quantify associations of wind
- chill index with DMI, CH₄ and CO₂ production.

242 **RESULTS AND DISCUSSION**

243 Diurnal pattern of enteric CH₄ and CO₂ emissions

Diurnal patterns of enteric CH₄ and CO₂ emissions were generated by averaging daily CH₄ and 244 CO₂ across animals and trials by hour of day for heifers (Figure 1) and cows (Figure 2). These 245 figures illustrate that CH₄ and CO₂ emissions began to increase rapidly for heifers and cows from 246 0900 to 1000 hr, which coincided with feed delivery between 0830 to 0900 hr. Peak emissions 247 for cows occurred between 1300-1800 hr and then started to decline after 1800 hr as they were 248 249 fed once in the morning. However, emissions for the heifers peaked between 1800-2000 hr due to a second feed delivery at approximately 1530-1630 hr. Maximum CH₄ and CO₂ emissions 250 (peak point) over the pattern were divided by their minimum emissions (nadir point) to estimate 251 252 maximum variability of diurnal pattern of CH₄ and CO₂ emissions over 24 hr. The variability for CH₄ emissions were 1.48 and 1.85 from replacement heifers and cows, respectively. The 253 254 variability for CO₂ emissions were 1.43 and 2.1 for replacement heifers and cows, respectively. 255 Cows had a more variable CH₄ and CO₂ diurnal pattern compared to heifers, likely due to

256	increased variability in age (2.6-7.2 yr for cows; 9-11 mon of age for heifers), body weight (532-
257	769 kg for cows; 226-370 kg for heifers) and feed intake of cows compared with heifers.
258	Diurnal pattern of CO ₂ emission had lower variation compared to CH ₄ in both cows and
259	heifers. Both enteric CH ₄ and CO ₂ emissions and yields are proportional to DM intake (Grainger
260	et al. 2007). In addition, CO ₂ is proportional to energy expenditure, and at a given level of
261	energy intake, CO ₂ output is much less variable than enteric CH ₄ (Hegarty 2013). Methane
262	emissions were found to be most affected by feed intake, diet composition, and feeding
263	frequency (Hegarty 2013; Knapp et al. 2014; Manafiazar et al. 2017). Crompton et al. (2010) fed
264	lactating dairy cows at different intervals with 1, 2, or 4 times equal feeding daily. Higher
265	variability (2.5 nadir point) was reported when animals were fed once daily, and the lowest
266	variability (1.6) was observed when animals were fed 4 times daily (Crompton et al. 2010). This
267	also further explains higher variability of cows in our study, which were fed once compared to
268	heifers that were fed twice daily. Zimmerman et al. (2013) reviewed variability of CH ₄ diurnal
269	pattern using the GEM system and reported a range of 1.2 to 1.6. In all studies, lowest CH4 and
270	CO ₂ emissions occurred just before feeding in the morning and highest emissions occurred in the
271	afternoon after second feeding. Our results of CH4 emission pattern are comparable with
272	previous studies.

273 Performance, feeding behaviour, feed intake and CH₄ and CO₂ production

Heifer and cow performance during the feed efficiency test periods at LDRC and KIN are
presented in Table 2. High and low RFI_{fat} heifers and cows were similar in on-test age, test midpoint weight, ADG, and off-test back fat thickness, but differed in RFI_{fat} by 0.78 kg DMI d⁻¹ for
all heifers and by 1.11 kg DMI d⁻¹ for all cows tested. Among animals that visited the GEM
system, high and low RFI_{fat} heifers and cows were also similar in on-test age, test mid-point

weight, ADG, off-test back fat thickness, and in magnitude of difference for RFIfat (0.73 kg DMI 279 d⁻¹ for heifers; 1.21 kg DMI d⁻¹ for cows). These results are consistent with previous results 280 reported by other researchers (Kelly et al. 2010b; Durunna et al. 2013; Basarab et al. 2007, 281 2011). In our study, 79 cows were also feed efficiency tested as heifers, and showed moderate 282 positive (P<0.001) correlations for RFI_{fat} ($r_p=0.36$), DMI ($r_p=0.33$), final back fat thickness ($r_p=$ 283 284 (0.63) and MIDWT ($r_p = 0.66$) of heifers and cows. These results were expected as positive and moderate to high phenotypic and genetic correlations have been reported by (Archer. et al. 2002) 285 between heifer post-weaning and mature cow RFI ($r_p = 0.40$; $r_g = 0.98$), DMI ($r_p = 0.51$; $r_g = 0.94$), 286 287 ADG ($r_p = 0.28$; $r_g = 0.72$), and MIDWT ($r_p = 0.70$; $r_g = 0.82$).

During the GEM monitoring period, low RFIfat females consumed less feed (heifers 288 289 8.7%; cows 13.1%), spent less time in the feeding bunk (heifer 6.5%: cows 5.5%) and had less head down time (heifers 19.7%; cows 3.6%) with fewer visits to the bunk (heifers 18.1%; cows 290 6.8%) compared with high RFI_{fat} females (Table 3). These results are consistent with those of 291 292 others who compared feed intake and feeding behaviour between different groups of beef yearling heifers classified for RFIfat (Kelly et al. 2010b; Durunna et al. 2013; Kayser and Hill 293 2013). High RFI_{fat} heifers visited the GEM system more frequently and spent more time at each 294 295 visit and received more pellets from the unit compared with low RFI_{fat} heifers, while high and low RFI_{fat} cows were similar in visit frequency and visit time to GEM. This latter result may 296 have been due to the familiarity of 79 cows with the GEM system when they were exposed to it 297 298 as heifers.

The main effect of RFI_{fat} group across trial was significant for CH_4 and CO_2 emission and yield for both heifers and cows (Table 3). Low RFI_{fat} heifers emitted 2.5% less CH_4 and 1.4% less CO_2 (P<0.001) compared with high RFI_{fat} heifers, while low RFI_{fat} cows emitted 3.7% less

CH ₄ and 3.4% less CO ₂ (P<0.001) compared with high RFI _{fat} cows. However, low RFI _{fat} heifers
and cows had higher CH_4 and CO_2 yield (P<0.001) per kg of DMI but were similar for CO_2/CH_4
ratio. Other researchers using different techniques such as SF ₆ (Hegarty et al. 2007; Fitzsimons et
al. 2013), RC and GEM system (Alemu et al. 2017) also reported that low RFI _{fat} heifers had
lower feed intake and emitted less daily CH4 compared with high RFIfat heifers. Recent studies
(Hegarty et al. 2007; Fitzsimons et al. 2013; Alemu et al. 2017) are inconsistent in CH ₄ yield
depending on the method of measurement, diet composition and definition of methane yield.
Contradictory to our results, Fitzsimons et al. (2013) reported higher CH ₄ yield for high RFI
Simmental heifers when CH ₄ yield was expressed as g CH ₄ kg BW ^{-0.75} (2.9 vs. 2.5; n=28).
Whereas, Alemu et al. (2017) reported no difference in CH4 yield for low RFI _{fat} compared with
high RFI_{fat} heifers when CH_4 yield was expressed as g CH_4 kg DMI^{-1} using GEM and RC
(GEM, 27.7 vs. 28.5, n=16, P > 0.25; RC, 26.5 vs. 26.5; n=16, P > 0.99). Hagerty et al. (2007)
reported lower CH ₄ yield for low RFI steers (n=20) such that low RFI cattle emitted 41.2 g less
CH ₄ per unit of ADG.
Lower CH ₄ and CO ₂ emission (g d^{-1}) in low RFI _{fat} heifers and cows appears to be
partially due to lower feed intake compared with their high RFI_{fat} pen mates and is reflected by
the high positive phenotypic correlations ($r_p = 0.85$ to 0.89; $P < 0.001$) between CH ₄ and CO ₂
emission and DMI (Herd et al. 2014; Manafiazar et al. 2017), and moderate negative phenotypic
correlations (r_p = -0.36 and -0.77, P < 0.001) between CH ₄ and CO ₂ yield and DMI (Manafiazar
et al. 2017). Potts et al. (2017) recently reported that 9 to 31% of the variation in RFI, when dairy
cows are fed low starch diet, can be explained due to the animal's digestive ability, suggesting
that low RFI animals had higher digestibility capacity. Bonilha et al. (2017) also reported that

These associations between RFI and DMI, and higher digestibility in low RFI animals could explain our results such that low RFI_{fat} animals are more efficient in utilizing feed by having longer rumen retention time, increased DM and protein digestibility, resulting in more substrate hydrogen ions available for methanogenesis and consequently higher CH₄ and CO₂ yield, but lower daily CH₄ emissions. In addition, lower DMI and changing rumen retention time may also impact the proportions of microbial populations present with the protozoal populations that support the methanogenes being relatively more sensitive to higher passage rates.

332 Climate Variables and DMI, CH4, CO2, and Visit Frequency

333 Ten of 14 trials were performed where average daily ambient temperature was below 0°C, and animals experienced temperatures below -20°C in 12 trials (Figures 3 and 4). Heat loss from 334 animals is higher when lower air temperature is accompanied by wind, and is referred to as wind 335 chill (Tarr 2015). Average daily wind chill during the CH₄ and CO₂ measurement periods by trial 336 at the KIN and LRDC are illustrated in Figures 3 and 4, respectively, for heifers and cows. 337 338 Lower critical temperature (LCT) is defined as the temperature below which animals must increase their metabolic rate from the basal point to maintain homeostasis and core body 339 temperature. The LCT for beef cows is variable and depends on cow's age, hair depth, thickness 340 341 and condition, hide thickness, backfat thickness, feed type and availability of shelter (National Research Council 2016). Lower critical temperature of -8°C was reported for mature cows with 342 343 dry and heavy winter coats, whereas a LCT of -34.1°C was reported for steers with about 1 kg of 344 ADG (Block et al. 2001). Scatter plots between wind chill and DMI (a), CH₄ emission (b), CO₂ emission (c), and visit frequency to GEM (d) for heifers and cows are presented in Figures 5 and 345 346 6, respectively. Scatter plots between ambient temperature and these same variables showed the 347 same pattern as with wind chill index and are not shown. Heifers and cows had different DMI

348 response to wind chill index. For example, heifers decreased their DMI with decreasing wind chill index between +20 and -15°C, but increased their DMI when wind chill decreased further. 349 Regression analysis of heifer data revealed that with 1 unit decrease in wind chill index above -350 15°C, the amount of DMI decreased 43 g ($\beta_1 = 43$ (9) g, $R^2 = 0.38$, P < 0.001). Whereas, the 351 relationship is changed below -15°C such that 1 unit decrease in wind chill index increased DMI 352 by 32 g ($\beta_1 = 32$ (29) g, $\mathbb{R}^2 = 0.08$, $\mathbb{P} > 0.28$) though the linear relationship was not significant. 353 However, cows generally increased their DMI with decreasing wind chill index (Figure 6a). 354 Regression analysis showed that in cows a 1 unit decrease in wind chill index increased DMI by 355 24 g ($\beta_1 = 24$ (14) g, $\mathbb{R}^2 = 0.09$, $\mathbb{P} > 0.09$). Further regression analysis revealed that the 356 interaction between RFI_{fat} group and wind chill index were not significant for heifers (P=0.08) 357 and for cows (P=0.62), thus indicating that low and high RFI_{fat} heifers and cows had similar DMI 358 359 responses to wind chill index. Methane and CO_2 emission declined as wind chill index decreased in both heifers and cows 360 (Figures 5b, 5c, 6b and 6c). Regression analysis revealed that with 1 unit decrease in wind chill 361 index the amount of CH₄ decreased 1.03 g ($\beta_1 = 1.0$ (0.2) g, $R^2 = 0.48$, P < 0.001) in heifers and 362 2.3 g ($\beta_1 = 2.3$ (0.3) g, $R^2 = 0.62$, P < 0.001) in cows. High and low RFI_{fat} mature cows had the 363 same (P > 0.62) response in CH₄ reduction per one unit decrease in wind chill index, but high 364 RFIfat heifers emitted more CH4 compared to low RFIfat heifers in response to one unit decrease 365 in wind chill index (1.37 g vs. 0.87 g, P<0.024). Regression analysis also showed that with 1 unit 366 decrease in wind chill index the amount of CO₂ decreased 37 g ($\beta_1 = 37$ (5) g, R² = 0.53, P < 367 0.001) in heifers and 59 g ($\beta_1 = 59$ (12) g, $R^2 = 0.42$, P < 0.001) in cows. High and low RFI_{fat} 368 mature cows had the same (P > 0.62) response of CO_2 reduction to 1 unit decrease in wind chill 369 370 index. However, high RFI_{fat} heifer had a two-fold reduction in CO₂ production in response to 1

°C decrease in wind chill index (52.2 g vs. 26.8 g, P < 0.021). Visit frequency to GEM system
was also affected by wind chill index such that peak of visit occurred at about 0 °C and -10 °C of
wind chill index for heifers and cows, respectively (Figures 5 and 6, d).

Generally, voluntary DMI increases as air temperature decreases (National Research Council 374 2016), and negative association between ambient temperature and DMI (-0.19) has been reported 375 376 in steers (Milligan and Christison 1974). We observed the same trend in mature cows consuming a triticale silage diet at LRDC and a barley silage diet at KIN. However, the opposite effect was 377 observed in heifers above -15°C consuming a barley silage:barley grain diet (90:10% as fed). 378 379 This could be due to a chronic versus acute exposure such that intake drops in response to temperature and encouraged heifers to look for shelter then increase their DMI. It appears that 380 cows had experienced the cold temperature before and were able to better manage the cold stress. 381 Factors other than ambient temperature may also affect the animals' performance such as feed 382 type and shelter accessibility. Webster et al. (1970) reported that ad libitum fed cattle are not 383 384 affected by Canadian prairie winter temperature when dry and sheltered from wind. It is also reported that digestibility of feedstuffs is reduced at lower environmental temperature along with 385 reduced voluntary feed intake (Milligan and Christison 1974). Methane and carbon dioxide 386 387 emissions are proportional to DMI, and it is expected that CH₄ and CO₂ emissions will increase with increasing DMI. In this study, we observed reduction in CH₄ and CO₂ emissions despite 388 389 increasing DMI for cows and heifers at wind chill indexes below -15°C. It is generally assumed 390 that colder air temperature increases DMI, ruminal passage rate and this may reduce daily CH₄ emissions and yields (McAllister et al. 1996). Low RFI_{fat} heifers had lower reduction of CH₄ and 391 392 CO₂ in response to 1 °C decrease in wind chill index compared to high RFI_{fat} without significant

393	differences in DMI response. This may be indicative of low RFI _{fat} heifers having a lower passage
394	rate and higher digestibility than high RFI _{fat} heifers, which is worthy of further investigation.
395	CONCLUSION
396	Efficient (low RFI_{fat}) beef heifers and cows emitted less CH_4 and CO_2 (g d ⁻¹) but had higher CH_4
397	and CO ₂ yields (g kg ⁻¹ DMI) compared with their high RFI _{fat} pen mates. These results further
398	confirm that selecting and breeding animals for low RFI_{fat} has the potential to decrease
399	greenhouse gas emissions from beef cattle production. These results also confirm that the
400	mitigation potential is feasible in cold environments.
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				Yearling	heifers						Matu	re cows		
Location ^a	LRDC			KIN					LRDC			KIN		
Trial	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Start year of test	2015	2016	2017	2015	2015	2016	2016	2017	2015	2016	2017	2015	2016	2017
Start date	19 Feb	22 Dec	18 Jan	5 Jan	15 Dec	10 Mar	7 Dec	13 Mar	17 Nov	19 Oct	18 Oct	17 Nov	22 Nov	15 Nov
Days on test	75	72	114	78	76	75	84	81	86	84	84	77	79	82
Number of animals	86	103	114	225	145	150	149	163	20	20	21	40	40	40
Groups in GEM	1	1	3	1	1	1	1	2	1	1	1	1	1	1
Animals monitored	by GEM													
High RFI	14	8	45	17	16	16	15	16	6	6	7	16	16	18
Low RFI	12	5	38	31	13	20	13	35	5	8	7	15	18	17
Diet ingredient com	position, as	s fed basis												
Barley silage, %	90.0	100.0	90.0	55.0	55.0	55.0	78.0	88.2				95.0	85.0	85.0
Barley grain, %	10.0	0.0	10.0										10.0	10.0
Protein supp., %				5.0	5.0	5.0	4.0	2.4				5.0	5.0	5.0
Whole Oat, %				27.0	27.0	27.0	6.0							
Canola Meal, %				13.0	13.0	13.0								
Triticale silage, %									100.0	100.0	100.0			
Corn DDGS, %							12.0	9.4						
Nutrient composition	n ^b , % DM													
TDN, %	63.3	63.5	62.5	67.1	70.1	67.2	69.3	68.9	57.5	59.5	59.5	60.6	61.8	66.7
Crude protein, %	12.5	11.4	13.3	19.0	19.4	15.9	17.6	17.5	9.9	11.9	11.9	13.9	14.4	14.3
ADF, %	29.1	30.9	32.9	24.2	22.6	26.8	26.3	27.9	37.1	35.3	35.3	31.4	28.4	25.9
NDF, %	44.5	46.6	49.5	35.8	35.1	41.3	42.0	40.9	52.0	49.8	49.8	46.1	43.7	37.8
Calcium, %	0.60	0.46	0.69	1.2	0.93	0.85	0.86	1.08	0.34	0.32	0.32	1.23	1.23	1.35
Phosphorous, %	0.34	0.29	0.31	0.51	0.51	0.39	0.47	0.48	0.25	0.27	0.27	0.31	0.35	0.32

Table 1. Number of animals, diet ingredient and nutrient composition by animal type, location and trial for animals tested for residual feed intake (RFI) and measured for enteric methane and carbon dioxide emissions using the Greenfeed Emissions Monitoring (GEM) system.

^a LRDC refers to the Lacombe Research and Development Centre, while KIN refers to the Roy Berg Kinsella Research Station.

^b TDN, ADF and NDF refers to total digestible nutrients, acid detergent fibre and neutral detergent fibre, respectively.

Performance traits	Overall	Mean	Residu of	al feed in f-test bac	take, ad kfat (RI	justed for FI _{fat})	P values for main effect and interaction				
	n	(SD)	High	Low	SE ^a	P value	Trial	Trial x RFI _{fat}			
All heifer during feed test periods (75	-114 d)										
Number of heifers	•		540	528							
On-test age, d	1068	278 (45)	283	282	1.1	0.807	< 0.044	0.513			
Test mid-point weight, kg	1068	352.9 (45.0)	357.3	357.6	1.4	0.869	< 0.001	0.967			
Test ADG, kg d ⁻¹	1068	1.05 (0.25)	1.02	1.02	0.00	0.575	< 0.001	0.693			
Off-test backfat, mm	1068	5.5 (2.2)	5.8	5.7	0.08	0.686	< 0.001	0.934			
Test RFI _{fat} , kg DM d ⁻¹	1068	0.00 (0.52)	0.38	-0.40	0.01	< 0.001	0.356	< 0.001			
Heifers that visited the Greenfeed Emissions Monitoring system during feed test periods											
Number of heifers			147	167							
On-test age, d	314	291 (40)	287	287	1.3	0.910	< 0.001	0.009			
Test mid-point weight, kg	314	269.8 (43.3)	368.7	368.4	2.4	0.928	< 0.001	0.970			
Test ADG, kg d ⁻¹	314	1.01 (0.21)	1.02	1.01	0.01	0.771	< 0.001	0.427			
Off-test backfat, mm	314	5.8 (2.3)	6.1	5.8	0.2	0.201	< 0.001	0.019			
Test RFI _{fat} , kg DM d ⁻¹	314	-0.05	0.34	-0.39	0.03	< 0.001	0.963	0.742			
		(0.48)									
Cow during feed test periods (77-86 d)										
Number of cows			84	92							
On-test age, yr	176	4.0 (2.2)	4.6	4.6	0.1	0.899	< 0.001	0.447			
Test mid-point weight, kg	176	651 (92.5)	678.6	674.7	7.8	0.628	< 0.001	0.852			
Test ADG, kg d ⁻¹	176	0.95 (0.29)	0.89	0.88	0.03	0.848	< 0.001	0.989			
Off-test backfat, mm	176	9.8 (4.5)	10.9	10.7	0.05	0.640	< 0.001	0.327			
Test RFI _{fat} , kg DM d ⁻¹	176	0.00 (0.67)	0.59	-0.52	0.06	< 0.001	0.777	0.003			
Cows that visited the Greenfeed Emissions Monitoring system during feed test periods											
Number of cows			69	70							
On-test age, yr	139	3.5 (2.1)	4.1	4.3	0.2	0.443	< 0.001	0.913			
Test mid-point weight, kg	139	638 (86)	678.1	669.2	9.7	0.358	< 0.001	0.917			
Test ADG, kg d ⁻¹	139	0.99 (0.26)	0.92	0.90	0.04	0.613	< 0.001	0.884			
Off-test backfat, mm	139	9.2 (3.9)	10.5	10.5	0.6	0.986	< 0.001	0.997			
Test RFI _{fat} , kg DM d ⁻¹	139	0.00 (0.71)	0.63	-0.58	0.08	< 0.001	0.963	0.004			

Table 2. Least squares means for performance traits in replacement beef heifers (8 trials) and mature cows (6 trials) during feed intake testing at Lacombe Research and Development Centre and the Roy Berg Kinsella Research Station.

^a Standard error of mean differences

trials) during feed intake te	esting at Lac	combe Research	and Deve	lopment	Centre a	and the Roy I	Berg Kinsella	Research Sta	ation			
Traits	Overall n	Mean (SD)	Residual feed intake, adjusted for off-test backfat (RFI _{fat})			P values for other main effects, interactions and covariates						
			High	Low	SE ^a	P value	Trial	Trial x RFI _{fat}	Day	Visit	Visit time	
Heifer daily observations	during me	ethane measurer	nent (24-	-62 d)								
Feed intake, kg DM d ⁻¹	6789	8.17 (1.82)	8.48	7.80	0.05	< 0.001	< 0.001	0.045	< 0.001			
Feeding duration, min d ⁻¹	8456	140.6 (45.7)	131.9	123.8	1.2	< 0.001	< 0.001	< 0.001	< 0.001			
Feeding headdown, min d ⁻¹	8456	97.4 (38.3)	92.4	77.2	1.1	< 0.001	< 0.001	< 0.001	< 0.001			
Feeding frequency, events d ⁻¹	8456	66.5 (39.8)	65.3	55.3	0.75	< 0.001	< 0.001	< 0.001	< 0.001			
GEM visits, visits d ⁻¹	9099	2.75 (1.53)	2.88	2.58	0.03	< 0.001	< 0.001	< 0.001	< 0.001			
GEM visit time, min d ⁻¹	9099	12.36 (7.3)	13.3	12.1	0.2	< 0.001	< 0.001	< 0.001	< 0.001			
GEM pellet intake, kg d ⁻¹	9095	0.52 (0.21)	0.49	0.45	0.07	< 0.001	< 0.001	< 0.001	< 0.001			
CH ₄ emission, g d ⁻¹	9099	178.9 (47.2)	184.1	179.7	1.2	0.001	< 0.001	< 0.001	< 0.001	0.221	0.059	
CO ₂ emission, g d ⁻¹	9099	6105 (1364)	6317	6230	28	0.001	< 0.001	< 0.001	< 0.001	0.319	0.006	
CH ₄ yield, g kg ⁻¹ DMI	6789	22.8 (7.2)	22.7	24.1	0.20	< 0.001	< 0.001	< 0.001	< 0.001	0.369	0.169	
CO ₂ yield, g kg ⁻¹ DMI	6789	771 (211)	785	842	6.3	< 0.001	< 0.001	< 0.001	< 0.001	0.715	0.237	
CO ₂ /CH ₄ ratio, g d ⁻¹	9099	35.5 (8.8)	35.7	36.0	0.18	0.159	< 0.001	< 0.001	< 0.001	0.997	0.205	
Cow daily observations d	uring meth	nane measureme	ent (33-5-	4 d)								
Feed intake, kg DM d ⁻¹	4069	12.69 (2.65)	13.16	11.64	0.08	< 0.001	< 0.001	< 0.001	< 0.001			
Feeding duration, min d ⁻¹	4073	171.6 (74.1)	147.8	140.1	1.9	< 0.001	< 0.001	< 0.001	< 0.001			
Feeding headdown, min d ⁻¹	4073	104.8 (49.3)	104.6	100.9	1.6	0.02	< 0.001	< 0.001	< 0.001			
Feeding frequency, events d ⁻¹	40731	76.0 (30.4)	75.4	70.6	0.8	< 0.001	< 0.001	< 0.001	< 0.001			
GEM visits, visits d ⁻¹	4694	2.40 (1.23)	2.28	2.21	0.04	0.135	< 0.001	0.011	< 0.001			
GEM visit time, min d ⁻¹	4702	10.12 (5.50)	9.63	9.32	0.17	0.116	< 0.001	0.002	< 0.001			
GEM pellet intake, kg d ⁻¹	2810	0.50 (0.23)	0.45	0.42	0.09	0.003	< 0.001	0.002	< 0.001			
CH_4 emission, g d ⁻¹	4694	237.5 (69.4)	241.2	232.7	2.3	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	
CO_2 emission, g d ⁻¹	4694	8417 (1836)	8145	7875	51	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	
CH ₄ yield, g kg ⁻¹ DMI	4061	19.52 (7.2)	19.2	21.1	0.2	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	

Table 3. Least squares means of feeding behaviour, and methane and carbon dioxide production traits in replacement heifers (8 trials) and mature cows (6 trials) during feed intake testing at Lacombe Research and Development Centre and the Roy Berg Kinsella Research Station

CO ₂ yield, g kg ⁻¹ DMI	4061	685 (208)	640.8	703.6	6.5	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
CO ₂ /CH ₄ ratio, g d ⁻¹	4694	37.1 (8.6)	35.5	36.0	0.3	0.045	< 0.001	< 0.001	< 0.001	0.031	< 0.001

^a Standard error of mean differences





535 Figure 1. Diurnal distribution of enteric methane and carbon dioxide emissions from replacement beef

beifers fed a high forage diet in drylot as measured by the GreenFeed Emission Monitoring system.





539 Figure 2. Diurnal distribution of enteric methane and carbon dioxide emissions from cows fed a

540 forage diet in winter drylot as measured by the GreenFeed Emission Monitoring system.



543 Figure 3. Average daily wind chill during GreenFeed Emission Monitoring system measurements by trial

at the Roy Berg Kinsella Research Station during the winters of 2015 to 2017. Black solid lines are

heifers while blue solid lines are cows. There were 14, 12, 1, 0, 19, 23, 1 and 9 d where daily wind chill

averaged below -20 °C for 2015 KIN heifers at KIN, 2015-16 cows KIN, 2016 KC heifers, 2016 Angus/

547 Charolaise heifers, 2016-17 KC cows, 2017 KC heifers, 2017 AN/CH heifers and 2017-18 KC cows,

548 respectively.



Figure 4. Average daily wind chill during Greenfeed Emission Monitoring system measurements by trial at the Lacombe Research and Development Centre during the winters of 2015 to 2017. Black solid lines are heifers while blue solid lines are cows. There were 0, 11, 3, 21, 5, 0, 0, and 1 d where daily wind chill averaged below -20 °C for 2015 heifers, 2015-16 cows, 2016 heifers, 2016-17 cows, 2017 pen 2 heifers,

555 2017 pen 3 heifers, 2017 pen 1 heifers and 2017-18 cows, respectively.

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Figure 5. Scatter plot of wind chill index versus average daily dry matter intake (a), average daily
methane (CH4) emission (b), average daily carbon dioxide (CO2) emission (c), and number of visits per

564 day in replacement heifers. Each point is the mean across animals and days for a particular wind chill

565 index.



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570 Figure 6. Scatter plot of wind chill index versus average daily dry matter intake (a), average daily



572 day in mature cows.