PHENOTYPIC ASSOCIATIONS BETWEEN METHANE PRODUCTION TRAITS, VOLATILE FATTY ACIDS AND ANIMAL BREEDING TRAITS

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SUMMARY

This paper reports results for 532 young Angus bulls and heifers measured for methane production in respiration chambers. The animals were tested on a roughage ration offered at 1.2-times maintenance, based on their pretest weight (WT). Rumen fluid was collected for analysis of volatile fatty acids (VFA). Mean WT was 410 ± 93 kg (sd), daily methane production (MP) 204 ± 31 L/day, MP per unit weight (MI) was 0.52 ± 0.10 L/kg WT, and methane produced per unit dry-matter intake (DMI; MY) was 29.9 ± 4.4 (L/kg DMI). Pearson correlations showed that WT and feed intake were moderately positively correlated with MP, and negatively correlated with MI and MY. Concentrations of the three most abundant VFA (acetate, propionate and butyrate), total VFA, and the molar proportions of propionate and butyrate were associated with variation in MI and MY, but less so with variation in MP. There were statistically-significant associations for MP and MI, but not MY, with standard BREEDPLAN weight and carcase weight EBV, but either no or weak associations with other carcase EBV. These preliminary results show that BREEDPLAN EBV could be used to reduce the intensity of cattle methane emissions, but not MY which appears to be independent of genetic variation in growth and carcase traits.

INTRODUCTION

Cattle and sheep emit methane, a potent greenhouse gas, as part of the fermentation process in their rumen. There is a strong positive relationship between feed intake and methane production. Direct selection for lower daily methane production (MP) may not be desirable because it could favour lower feed intake and/or lighter and slower growing animals. Methane intensity (MI) and methane yield (MY), being methane produced per unit of weight and per unit of feed intake respectively, measure methane production that is independent of size and feed intake.

MATERIALS AND METHODS

The Angus cattle measured were born in 2009 and 2011 in research herds at Agricultural Research Centres at Glen Innes and Trangie, NSW. The cattle were measured for methane production in 2011 and 2012 in the 10 open circuit respiration chambers at the University of New England campus, Armidale, NSW. The cattle were trucked to Armidale, with about 40 animals constituting a safe weight load for transport. Each year, within each herd and sex, cohorts of 40 head in 4 groups of 10 were formed and prepared for measurement. Progeny of individual sires were stratified across groups and cohorts. Before transport, the cohort of 40 animals was weighed and then fed in the groups of 10 an amount calculated to provide about 1.2 to 1.5-times their estimated energy requirement for maintenance, based on equation 1.21 of SCA (1990). The test ration was a commercial lucerne and oaten hay chaff (Manuka "Blue Ribbon" Chaff®), chosen to mimic good, dry pasture. Regular samples were taken for feed analysis over the 2 years and had an average content of 88% dry matter (DM), a crude protein content of 14% DM and metabolizable energy content of 9MJ/kg DM. After 10 days the animals were weighed again, with this weight used as their pretest weight (WT), and then transported to Armidale. There cattle were kept in their

groups of 10 and fed the same amount of the same chaff ration for a minimum of 4 days. Then the first of the 4 groups was moved into the animal house and each animal fed in an individual pen at 1.2-times maintenance based on its WT. Feeding a restricted daily allowance proportional to WT was done to avoid feed refusals, minimise day to day variation in daily MP and to avoid 'level of feeding' effects on MY.

Methane production was measured over 2 x 24h consecutive periods. Animals were placed in their chambers by 10.00, with their daily feed allowance in a feed bin and water available from a drinker inside the chamber. After 24 hours the chambers were briefly opened and the feed bin replaced with a clean bin and fresh feed. After 48 hours the animals were let out of the chambers, briefly restrained, and a sample of rumen fluid aspirated through a flexible stomach tube. The rumen fluid was preserved by acidification and then stored at -18^0C for subsequent analysis by gas liquid chromatography of VFAs, being products of the fermentation in the rumen. Most animals consumed their daily feed allowance within 8 to 12 hours, so for most animals these VFA concentrations represent levels following a short period of 12-or-so hours without fresh feed. The open circuit respiration chamber consists of an enclosed pen (1.8m x 3m) within a polycarbonate shell (3.6m x 2.4m x 2.4m), each with an individual mass flow meter and airflow subsample line connected by a multiplexer to a Servomex gas analyser.

Data on 218 animals of approximately 2-years of age tested in 2011 and 314 yearling-age animals in 2012 was used. The magnitude of phenotypic associations between methane production over the second-24hr period and VFA were determined by calculating Pearson correlation coefficients. Variation in methane traits associated with genetic variation in cattle breeding traits, as measured by standard BREEDPLAN® EBV, was assessed by the calculating regression coefficients for the traits against EBV in a general linear model. These EBV are described in BREEDPLAN (2010). Only EBV reported for at least 525 of the 532 animals are analysed. Fixed effects fitted were year, herd and sex, and their 2-way interactions. The interactions of EBV with the fixed effects (that is, differences in the slopes of the relationships) were mostly non-significant and are not reported. Pearson correlations and regression coefficients that differed from zero ($P<0.05$) were taken as evidence for a statistically-significant association. BREEDPLAN EBV for weight and carcass traits, extracted in May 2013, were used.

RESULTS

Summary statistics are presented in Table 1. There was substantial variation in the methane production traits measured. Pretest weight and DMI were strongly positively correlated with MP, and negatively correlated with MI and MY (Table 2). Significant correlations for the three most abundant VFA (acetate, propionate and butyrate), total VFA, and the molar proportions of propionate and butyrate, showed that variation in VFA production was associated with variation in MI and MY, but less so for variation in MP.

Fitting the fixed effects of year of test, then herd and sex each explained part of the variation in MP (3%, 17%, 8% respectively), MI (71%, 0.1%, 3%) and MY (55%, 1%, 4%), and together with their interactions explained 32%, 76% and 63% of the variation in MP, MI and MY, respectively. Variation in MP had statistically-significant associations with weight EBV and the EBV for carcase_wt and fat (Table 2). Variation in MI had statistically-significant associations with weight EBV, EBV_carcase_wt and EBV_ema, whereas MY was not associated with any EBV except EBV_ema.

DISCUSSION

These preliminary results show large natural variation between animals in MP, MI and MY. Heavier animals were offered more feed and had a greater MP, which might be considered undesirable, but had lower MI and MY, which is desirable in terms of greenhouse gas emissions.

Should these correlations hold at a genetic level, then selection for lower MP could reduce DMI and animal performance, and increase both MI and MY.

Table 1. Summary statistics for n=532 young Angus bulls and heifers tested for methane production in 2011 and 2012

Trait	Mean	SD	Maximum	Minimum
Pretest weight (WT kg)	410	93	670	229
Dry-matter intake DMI; kg/d)	6.9	1.1	9.5	4.6
Methane production (L/d)	204	31	350	115
Methane intensity (L/kg WT)	0.52	0.10	0.68	0.24
Methane yield (L/kg DMI)	29.9	4.4	41.2	15.0
Acetate (mmoles/L)	38.6	10.9	82.5	6.4
Propionate (mmoles/L)	8.1	2.9	36.2	1.1
Isobutyrate (mmoles/L)	0.6	0.2	1.4	0.1
Butyrate (mmoles/L)	4.2	1.5	10.5	0.7
Isovalerate (mmoles/L)	1.1	0.3	2.4	0.2
Valerate (mmoles/L)	0.4	0.2	1.9	0.0
Total VFA (mmoles/L)	53.0	15.2	131	8.6
Acetate%	73.0	1.6	77.3	63.1
Propionate%	15.1	1.7	27.7	12.4
Butyrate%	7.9	1.0	13.5	3.2
EBV_BWT (kg)	3.9	1.9	10.2	-0.7
EBV_200d_wt (kg)	23.8	7.0	42.0	3.0
EBV_400d_wt (kg)	45.8	12.2	78.0	4.0
EBV_600d_wt (kg)	56.6	16.1	104	9.0
EBV_carcase_wt (kg)	28.3	9.6	57.0	-4.0
EBV_ema (cm^2)	0.9	1.4	5.4	-2.8
EBV_rib_fat (mm)	0.8	1.7	6.6	-4.0
EBV_rump_fat (mm)	0.6	1.8	6.8	-3.4
EBV_rby (%)	-0.1	0.9	2.0	-3.1

Higher concentrations of VFA were associated with higher MI and MY, but not with variation in MP. The molar proportions of the major VFA have been previously shown to be related to MY in cattle, with propionate being negatively, and butyrate positively, related to MY (Whitelaw *et al.* 1984), as found in this experiment. A high propionate pattern is a desirable as hydrogen from rumen fermentation is used in its formation, rather than being used for CH_4 formation. These relationships open the possibility of using VFA as indicator traits for methane emissions. However, the strength of their phenotypic association, with correlation coefficients of 0.4 or lower, are too low to be either an accurate screening test for high or low emitting cattle, or for genetic improvement based on indirect selection, conclusions also reached for sheep by Robinson *et al.* (2010). In this experiment, the VFA concentrations for most animals represent levels following a short period of 12-or-so hours without fresh feed, and may have been different had rumen fluid been sampled during peak fermentation that occurs immediately following feed consumption. More careful scrutiny of VFA as markers for methane emissions is still warranted.

The statistically-significant associations for MP and MI, but not MY, with standard BREEDPLAN weight EBV provides evidence that MP and MI could be altered by appropriate emphasis on these EBV. Increase in weight EBV was associated with greater MP (usually undesirable), lower MI (desirable) and no change in MY. The carcase EBV, except carcase_wt,

had little or no association with variation in methane emission traits. Preliminary estimates of genetic correlations are reported in Donoghue *et al.* (2013).

These results show that substantial natural variation in methane emissions is present in these Angus cattle, and that in BREEDPLAN the beef industry has a system which could be used to reduce the intensity of methane greenhouse gas emissions, but perhaps not MY which appears to be independent of genetic variation in the recorded growth and carcase composition traits.

Table 2. Pearson correlations for methane production (MP), methane intensity (MI) and methane yield (MY) with weight, feed intake and volatile fatty acids (VFA), and regression coefficients (±se) with BREEDPLAN EBV

	MP (L/day)	MI (L/kg WT)	MY (L/kg DMI)
Pretest weight (WT; kg)	0.53***	-0.79***	-0.59***
Dry-matter intake (DMI; g/d)	0.55***	-0.76***	-0.59***
Acetate (mmoles/L)	-0.07	0.33***	0.29***
Propionate (mmoles/L)	-0.05	0.16***	0.13**
Butyrate (mmoles/L)	-0.09*	0.40***	0.35***
Total VFA (mmoles/L)	-0.07	0.32***	0.28***
Acetate %	0.00	0.06	0.07
Propionate %	0.05	-0.41***	-0.39***
Butyrate %	-0.14**	0.44***	0.38***
EBV_BWT (kg)	3.5 ± 0.6***	-2.6 ± 1.2*[+]	0.07 ± 0.06
EBV_200d_wt (kg)	1.6 ± 0.2***	-0.9 ± 0.3**[+]	0.03 ± 0.02
EBV_400d_wt (kg)	.89 ± 0.08***	-0.5 ± 0.2**[+]	0.02 ± 0.01
EBV_600d_wt (kg)	.69 ± 0.06***	-0.5 ± 0.1***[+]	0.01 ± 0.01
EBV_carcase_wt (kg)	1.1 ± 0.1***	-0.6 ± 0.2**[+]	0.02 ± 0.01
EBV_ema (cm^2)	0.3 ± 0.8	-5.0 ± 1.5***[+]	-0.19 ± 0.09*
EBV_rib_fat (mm)	-1.6 ± 0.7*	-1.1 ± 1.2[+]	-0.11 ± 0.07
EBV_rump_fat (mm)	-1.5 ± 0.6*	-1.3 ± 1.2[+]	-0.11 ± 0.07
EBV_rby (%)	1.2 ± 1.3	-0.6 ± 2.3[+]	0.01 ± 0.13

*P<0.05; **P<0.01; ***P<0.001. [+]These coefficients and se have been multiplied by 1,000.

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