Heritability estimates of methane emissions from sheep

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The objective of this study was to determine the genetic parameters of methane (CH4) emissions and their genetic correlations with key production traits. The trial measured the CH4 emissions, at 5-min intervals, from 1225 sheep placed in respiration chambers for 2 days, with repeat measurements 2 weeks later for another 2 days. They were fed in the chambers, based on live weight, a pelleted lucerne ration at 2.0 times estimated maintenance requirements. Methane outputs were calculated for g CH4/day and g CH4/kg dry matter intake (DMI) for each of the 4 days. Single trait models were used to obtain estimates of heritability and repeatability. Heritability of g CH4/day was 0.29 ± 0.05, and for g CH4/kg DMI 0.13 ± 0.03. Repeatability between measurements 14 days apart were 0.55 ± 0.02 and 0.26 ± 0.02, for the two traits. The genetic and phenotypic correlations of CH4 outputs with various production traits (weaning weight, live weight at 8 months of age, dag score, muscle depth and fleece weight at 12 months of age) measured in the first year of life, were estimated using bivariate models. With the exception of fleece weight, correlations were weak and not significantly different from zero for the g CH4/kg DMI trait. For fleece weight the phenotypic and genetic correlation estimates were −0.08 ± 0.03 and −0.32 ± 0.11 suggesting a low economically favourable relationship. These results indicate that there is genetic variation between animals for CH4 emission traits even after adjustment for intake. The portion of the variation that was independent of intake showed no unfavourable genetic relationships with the production traits that were measured in these animals. These results suggest that it may be feasible to breed ruminants with lower methane emissions.

Keywords: sheep, methane, heritability, genetic correlation, global warming

Implications

To breed grazing ruminants with reduced methane emissions, we first need to demonstrate that there is repeatable individual variation in this trait and that a portion of this variation is genetically inherited. This study has provided evidence for both even after adjustment for intake. The portion of the variation that was independent of intake showed no unfavourable genetic relationships with the production traits that were measured in these animals. These results suggest that it may be feasible to breed ruminants with lower methane emissions.

Introduction

Rumen methanogenesis results in the loss of 6% to 10% of gross energy intake and globally is the single most significant source of anthropogenic methane (CH4) emissions. This source represents a major share of New Zealand’s total greenhouse gas emissions (Cottle et al., 2011; Pinares-Patino et al., 2011c; Clark, 2013). One option to mitigate these emissions is to genetically select for ruminants that emit less CH4. There is currently limited evidence about the genetics of this trait and its relationship with other production traits including feed intake. Recent extensive reviews of rumen CH4 formation by Janssen (2010) and Clark (2013) indicated that altering rumen hydrogen (H2) concentrations affects CH4 emissions, and there is limited evidence showing that emissions are positively correlated with lower rumen flow rates and higher acetate/propionate ratios (Pinares-Patino et al., 2003).

Pinares-Patino et al. (2011a and 2011c) presented results that showed that individual CH4 emissions expressed as g CH4/kg dry matter intake (DMI) is a repeatable trait across both ages and diets and that it is also accompanied by changes in rumen outflow rates. Similarly, preliminary studies
have shown a repeatable, and in some a significant genetic, component to CH$_4$ emissions, albeit often compromised by lack of intake measurements (Hegarty and McEwan, 2010; Hegarty et al., 2010; Robinson et al., 2010 and 2011; Goopy et al., 2011; Garnsworthy et al., 2012). Specifically, the available parameter estimates are very limited in their nature, have low accuracy and are often compromised by factors intrinsic to the measurement technique.

Several modelling studies have also been undertaken using assumed parameters which suggest that high implicit carbon costs (CO$_2$ equivalent or (CO$_2$-e)) would be required to stabilise CH$_4$ emissions while increasing productivity even when CH$_4$ emissions and feed intake are measured (Cottle et al., 2011; Cottle and Connington, 2012; Ludemann et al., 2012). These authors also uniformly suggested that improvement of this trait is best expressed to end users as emission per unit of animal product. The objective of this study was to accurately determine the genetic parameters of CH$_4$ emissions and their genetic correlations with key production traits in sheep using respiration chambers where feed intake was measured and CH$_4$ emissions monitored accurately over several days, while a standard diet was fed. It is part of a larger initiative to create divergent selection lines for this trait so as to understand the anatomical, physiological and microbiological as well as productivity changes that accompany selection for this trait.

Material and methods

All experimentation conducted was approved by AgResearch's Invermay and Palmerston North Animal Ethics committees. These included but were not restricted to applications 11930, 11975, 12206, 12233, 12241, 12324, 12414. The experiment was conducted at the Ulyatt-Reid Large Animal Facility of AgResearch Grasslands Research Centre in Palmerston North, New Zealand and CH$_4$ was measured in respiration chambers with a 10 to 15-day interval in-between in the pens. Methane emissions were measured for 2 or 3 days consecutively in each round. Individual animal DMI was measured in metabolic crates during each round (4 days) and then in respiration chambers (2 days); feeding level was based on LW and was estimated as 2.0 times maintenance requirements. Animals were fed in two equal-sized meals at 9 am (hour 0) and 3 pm (hour 6) daily. Both in R1 and R2, individuals were randomly allocated to groups (4; each of 24 animals) and respiration chambers and typically 10 progeny per sire were randomly selected to be measured for CH$_4$ emission. Only complete records were used and if a chamber seal was broken or less than 95% of the offered feed was eaten on the day of measurement then the record was discarded.

The CH$_4$ measurements have been previously described by Pinares-Patiño et al. (2011a). The chambers and brief measurement protocol used are outlined in Figure 1. Animals were acclimatised in pens for 21 days to a standard lucerne pellet diet (19% CP, 43% NDF and 10 MJ ME/kg dry matter). This was followed by two measurement rounds (R1 and R2) in respiration chambers with a 10 to 15-day interval in-between. Methane emissions were measured for 2 or 3 days consecutively in each round. Individual animal DMI was measured in metabolic crates during each round (4 days) and then in respiration chambers (2 days); feeding level was based on LW and was estimated as 2.0 times maintenance requirements. Animals were fed in two equal-sized meals at 9 am (hour 0) and 3 pm (hour 6) daily. Both in R1 and R2, individuals were randomly allocated to groups (4; each of 24 animals) and respiration chambers and typically 10 progeny per sire were randomly selected to be measured for CH$_4$ emission. Only complete records were used and if a chamber seal was broken or less than 95% of the offered feed was eaten on the day of measurement then the record was discarded.

For subsequent analysis CH$_4$ emissions were expressed as 24-h total gross emissions (g CH$_4$/day) and as CH$_4$ yield (g CH$_4$/kg DMI) per unit of ingested feed. LW (kg) was also measured on each measurement round. Additional performance information was downloaded from Sheep Improvement Limited (SIL), the New Zealand sheep genetic evaluation database, for the animals measured and their contemporaries. This was an attempt to make performance trait estimates more accurate and also allow unbiased estimates of maternal

Figure 1 A photograph of a sheep in a metabolic crate, within the respiration chamber. The measurement protocol used for the initial evaluation is presented below. An adaptation period of typically 21 days was followed by measurements within the respiration chamber conducted over 2 days for round 1 (M1 and M2). This was in turn followed by ~14 days of rest and then another 2-day round of measurements (M3 and M4).
effects where these were relevant. Additional information included pedigree records from the five birth flocks of animals, born 1990 to 2011, and the following animal information: sex (male, female), birth rearing rank (Brr), age of dam (aod), birth day deviation (bdev) from the contemporary group (flock, birth year, sex), weaning weight (WWT) grazing mob where "." indicates an interaction. Production trait measurements included: weaning weight at 3 months (kg; WWT), live weight at 6 or 8 months (kg; LW8), fleece weight at 12 months (kg; FW12), ultra sound eye muscle depth (mm; EMD), and dag score (faecal accumulation in the perineum region) at 3 or 8 months (DAG3, DAG8). Of these WWT, LW8, FW12 and EMD are standard production traits, whereas DAG3 and DAG8 are visually assessed traits commonly scored on a 6-point scale (0 no variations and flock and group treatment effects.

The data collected were analysed in a mixed model using ASREML 3.0 (Gilmour et al., 2009). Both single trait and bivariate analyses were undertaken. The former were for traits measured as part of the respiration chamber measurements g CH₄/day, g CH₄/kg DMI and LW while the latter were for estimating correlations of production and visual traits with g CH₄/day and g CH₄/kg DMI. Initial exploratory analyses were undertaken using a general linear model procedure, fitting a repeatability model (SAS, 2005) for g CH₄/day, g CH₄/kg DMI and LW. Fixed effects fitted included flock (flk), birth year (byr), sex, birth rearing rank (brr), grazing mob (trait mob), recording year (yr), round (1 to 3), lot (of 96 animals, 1 to 5), group (of up to 24 within year, 1 to 12), system (8 chambers to a system, 1 to 3), and chamber (1 to 24). Birthday deviation (bdev) and aod as linear and quadratic (aod²) were fitted as covariates. Interactions between these effects were tested and by a process of backwards elimination parsimonious models were selected (Table 1). For industry production traits standard fixed effects used by Sheep Improvement limited (www.sil.co.nz) were utilised (Pickering et al., 2011). For genetic estimates animal was fitted as a random effect with covariances proportional to the numerator relationship matrix along with permanent environmental effects (within round, between rounds and between years) where this was appropriate.

### Results and discussion

The final parsimonious models selected are described in Table 1. Chamber effects for CH₄ emissions were investigated, found to be non-significant, and therefore removed from final models. For the trait g CH₄/day gross emissions were influenced by LW (and associated feed offered) so factors that influenced these traits such as brr were significant. For the trait g CH₄/kg DMI only fixed effects affecting contemporary group formation were significant, presumably reflecting intrinsic factors including age, diet variations and flock and group treatment effects.

In total, 5236 daily complete CH₄ measurements were recorded (~4.3 records/animal; Table 2). The heritability of gross CH₄ emission was moderate with an estimate 0.29 ± 0.05 and the coefficient of variation was 12.9%; similar to that of LW. Given that CH₄ emission is influenced by intake, which in this situation is dependent on LW, of more interest is the heritability of CH₄ yield (g CH₄/kg DMI) which had a heritability estimate of 0.13 ± 0.03 and a coefficient of variation of 10.3%. These estimates are for a single 24-h measurement. Of considerable interest in any breeding programme is the repeatability of the measurement over both short (adjacent days), medium (several weeks) and long term (years) as these allow investigation of the consequences of repeated measurements and the utility of measurements taken at one age or time at subsequent measurements. The results for gCH₄/day show very high correlation between consecutive days, but the repeatability across rounds or across years is considerably lower at 0.53–0.55 compared to 0.80–0.88 for LW. This is perhaps not surprising, given that LW is a cumulative trait, whereas daily emissions are not. However, the results do suggest that a single g CH₄/day measurement contains a considerable amount of error and that recording consecutive days does little to mitigate this. It also suggests that repeatability
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### Table 2 Heritability (h²), repeatability estimates (±s.e.) for methane traits and LW at measurement

| Trait                  | n records | Mean | σp         | h² ± s.e. | Consecutive days | Across rounds | Across years |
|------------------------|-----------|------|------------|----------|-----------------|---------------|--------------|
| g CH₄/day              | 5236      | 24.6 | 3.18       | 0.29 ± 0.05 | 0.94 ± 0.003 | 0.55 ± 0.02  | 0.53 ± 0.02  |
| g CH₄/kg DMI           | 5235      | 15.7 | 1.62       | 0.13 ± 0.03 | 0.89 ± 0.005 | 0.26 ± 0.02  | 0.24 ± 0.02  |
| LW (kg)                | 4869      | 48.5 | 5.12       | 0.46 ± 0.07 | 0.93 ± 0.004 | 0.88 ± 0.01  | 0.80 ± 0.01  |

LW = live weight; DMI = dry matter intake.

### Table 3 Estimates of SIL production trait heritabilities (h²) (±s.e.) and genetic (rg) and phenotypic (rp) correlations with methane traits

| Trait               | n records | Mean   | σp         | Direct h² ± s.e. | dam h² ± s.e. | 2-trait with g CH₄/day | 2-trait with g CH₄/kg DMI |
|---------------------|-----------|--------|------------|------------------|----------------|-----------------------|--------------------------|
| WWT (kg)            | 48591     | 27     | 4.11       | 0.23 ± 0.01      | 0.23 ± 0.01    | 0.71 ± 0.07           | 0.43 ± 0.02              |
| LW8 (kg)            | 34742     | 40     | 4.95       | 0.56 ± 0.01      | –              | 0.80 ± 0.04           | 0.52 ± 0.02              |
| FW12 (kg)           | 15186     | 3.1    | 0.48       | 0.53 ± 0.02      | –              | 0.11 ± 0.07           | 0.12 ± 0.03              |
| EMD (mm)            | 22141     | 26.7   | 2.86       | 0.50 ± 0.02      | –              | 0.42 ± 0.10           | 0.32 ± 0.03              |
| DAG3 score          | 22809     | 1.03   | 1.12       | 0.43 ± 0.02      | –              | –0.18 ± 0.07          | –0.06 ± 0.03             |
| DAG8 score          | 8072      | 1.14   | 1.25       | 0.51 ± 0.03      | –              | –0.07 ± 0.12          | 0.01 ± 0.04              |

SIL = Sheep Improvement Limited; DMI = dry matter intake; WWT = weaning weight; LW = live weight; FW = fleece weight; EMD = eye muscle depth.

The results presented, comprehensively show that both gross CH₄ (g CH₄/day) and CH₄ yield (g CH₄/kg DMI) are heritable and repeatable traits. Although variation in intake accounts for a significant fraction of the phenotypic and genetic variation in CH₄ emissions, there remains a component that is independent of intake and offers hope for genetic selection as a potential option to reduce CH₄ yield emissions. On the basis of the limited results of Pinares-Patiño et al. (2011a and 2011c), whose animals formed a part of this work, it would appear that these differences remain across a variety of diets and ages. These diets span the likely range to be encountered in temperate climates when grazing improved pastures or feedlot conditions. As previously identified by Janssen (2010) in his review and also by Pinares-Patiño et al. (2003 and 2011a): reduced CH₄ emission is also associated with increased rumen flow rates.
associated with reduced productivity for any of the traits examined to date, albeit a wider range of traits and ages needs to be examined. This includes adult maternal traits which have major effects on farm scale CH₄ emissions as well as productivity (Cottle and Connington, 2012; Ludemann et al., 2012). Until reliable estimates for these are available (estimated to be late 2014 for this experiment) updating modelling studies will be premature.

It is acknowledged that the current work suffers a weakness in that feed intake was controlled during CH₄ emission measurement, which consisted of a brief period of around 4 days. However, productivity was measured almost exclusively at pasture where feeding levels varied with season from near maintenance to ad libitum. In addition, grazing pasture also allowed the animals to vary their individual intakes and chewing and rumination behaviour. Thus, a key research task is to validate that the CH₄ differences observed in chambers are also present in animals when grazing pasture. Work is currently underway to examine this across seasons and feeding levels using both portable accumulation chambers (Goopy et al., 2011) and using the SF₆ tracer technique (Johnson et al., 1994; Pinares-Patiño et al., 2011b).

Another aspect worthy of investigation is measurement of voluntary feed intake and feed efficiency in these animals. It is perhaps unfortunate that worldwide investigations into CH₄ emissions and feed efficiency have been undertaken largely independently of each other to date with only very limited contemporaneous measurement of both traits.

A further research priority is to confirm that similar relationships are present in beef and dairy cattle. The development and validation of the utility of brief measurements using ‘sniffers’ for cattle as reported by Garnsworthy et al. (2012) offers the potential that this can be done on a suitable scale to obtain useful genetic estimates. Finally, almost all of the work to date has been conducted in temperate climates using C3 grass, legume or grain diets; and there is an urgent need to assess whether these results have any relevance to tropically adapted ruminants grazing lower quality C4 pasture diets.

Divergent selection lines are being developed from the extremes of the animals identified in this research and the following research topics are underway or planned. Firstly, progeny from the divergent selection lines will continue to be measured using the current protocol and selection will be on the basis of breeding values for g CH₄/kg DMI. For both the original screened animals and the selection lines detailed measurements of production traits will be undertaken including maternal traits such as milk production and composition, reproductive status and adult LW, condition score and wool production. The intention is to monitor all traits that currently contribute to New Zealand dual-purpose sheep selection indexes (http://www.sil.co.nz) and to estimate their genetic correlations with the CH₄ emission traits. Secondly, studies are underway using the selection lines and existing data to determine the opportunities for indirect selection or rapid measurements of CH₄ emissions in order to rank animals. Finally, studies have commenced to examine what, if any, differences are present in the selection lines with regards to rumen anatomy, physiology and microbiology. This includes studies at different physiological ages, with different measurement techniques and for different diets.

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Supplementary materials

For supplementary materials referred to in this article, please visit http://dx.doi.org/10.1017/S1751731113000864

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