Genetic parameters of methane emissions determined using portable accumulation chambers in lambs and ewes grazing pasture and genetic correlations with emissions determined in respiration chambers

Arjan Jonker,† Sharon M. Hickey,‡ Suzanne J. Rowe,§ Peter H. Janssen,¶ Grant H. Shackell,‖ Sarah Elmes,* Wendy E. Bain,* Janine Wing,* Gordon J. Greer,* Brooke Bryson,* Sarah MacLean,* Ken G. Dodds,* Cesar S. Pinares-Patiño,* Emilly A. Young,* Kevin Knowler,* Natalie K. Pickering,* and John C. McEwan‡

ABSTRACT: Methane (CH$_4$) emission traits were previously found to be heritable and repeatable in sheep fed alfalfa pellets in respiration chambers (RC). More rapid screening methods are, however, required to increase genetic progress and to provide a cost-effective method to the farming industry for maintaining the generation of breeding values in the future. The objective of the current study was to determine CH$_4$ and carbon dioxide (CO$_2$) emissions using several 1-h portable accumulation chamber (PAC) measurements from lambs and again as ewes while grazing ryegrass-based pasture. Many animals with PAC measurements were also measured in RC while fed alfalfa pellets at 2.0 × maintenance metabolizable energy requirements (MEM). Heritability estimates from mixed models for CH$_4$ and CO$_2$ production (g/d) were 0.19 and 0.16, respectively, when measured using PAC with lambs; 0.20 and 0.27, respectively, when measured using PAC with ewes; and 0.23 and 0.34, respectively, when measured using RC with lambs. For measured gas traits, repeatabilities of measurements collected 14 d apart ranged from 0.33 to 0.55 for PAC (combined lambs and ewes) and were greater at 0.65 to 0.76 for the same traits measured using RC. Genetic correlations ($r_g$) between PAC in lambs and ewes were 0.99 for CH$_4$, 0.93 for CH$_4$ + CO$_2$, and 0.85 for CH$_4$/(CH$_4$ + CO$_2$), suggesting that CH$_4$ emissions in lambs and ewes are the same trait. Genetic correlations between PAC and RC measurements were lower, at 0.62 to 0.67 for CH$_4$ and 0.41 to 0.42 for CH$_4$ + CO$_2$, likely reflecting different environmental conditions associated with the protocols used with the 2 measurement methods. The CH$_4$/(CH$_4$ + CO$_2$) ratio was the most similar genetic trait measured using PAC (both lambs and ewes, 63% and 66% selection efficiency, respectively) compared with CH$_4$ yield (g/kg DMI) measured using RC. These results suggest that PAC measurements have considerable value as a rapid low-cost method to estimate breeding values for CH$_4$ emissions in sheep.

Key words: animal variation, breeding, greenhouse gas, proxy, spot-sampling, young and mature

†This work was financially supported by the Pastoral Greenhouse Gas Research Consortium (P GGRC; www.pggrc.co.nz) and the New Zealand Agricultural Greenhouse Gas Research Centre (NZAGRC; www.nzagrc.co.nz).
‡Corresponding author: arjan.jonker@agresearch.co.nz
§Present address: International coordinator ‘The New Zealand Peru Dairy Support Project’, MINAGRI, Jr. Yauyos 258, Lima, Peru
¶Received December 10, 2017. Accepted May 5, 2018.
INTRODUCTION

Ruminants are globally a significant source of anthropogenic CH₄ emissions (Schaefer et al., 2016) and one of the main contributors to New Zealand’s greenhouse gas inventory (MFE, 2017). Methane emissions per unit of DMI (i.e., CH₄ yield) were found to be heritable and repeatable in sheep fed alfalfa pellets in respiration chambers (RC) (Pinares-Patiño et al., 2013), proving that animal breeding is an option to mitigate CH₄ emissions. However, progress in generating these breeding values for CH₄ emissions is slow and expensive when animals have to be measured in RC. There have also been concerns that measuring CH₄ under controlled conditions may not reflect CH₄ emitted under the grazing conditions that predominate on NZ farms. More rapid spot-sampling methods that can be used on-farm would speed up this selection progress and provide a cost-effective and relevant method to the farming industry for maintenance of breeding values in the future. These methods could also extend the use of similar breeding programs for sheep of different genetic background and in different countries, and provide greater power to estimate correlated animal traits. One such spot-sampling method, that can measure both gas concentrations and flux, is the portable accumulation chamber (PAC) (Goopy et al., 2011). The method involves placing sheep in a simple and transportable sealed container for 40 min to 1 h and measuring the accumulated gas concentrations at regular intervals using a monitoring device attached to a one-way valve. Results of CH₄ output from PAC have been shown to be heritable and repeatable, but are influenced by environmental factors (e.g., trial site, animal age, and diet) and to date have not been compared with strictly controlled conditions, such as RC (Goopy et al., 2016). Significantly, DMI cannot be measured directly in relation to PAC measurements, and therefore, CH₄ yield cannot be determined. The ratio of CH₄ to CO₂ or total gas (CH₄ + CO₂) has been suggested as a proxy for CH₄ yield where DMI is not known (Madsen et al., 2010), but its relationship to CH₄ yield in sheep has not been validated.

The objectives of the current study were to estimate the heritability and repeatability of CH₄- and CO₂-emission parameters determined using repeated PAC measurements as lambs and as ewes grazing ryegrass-based pasture. Furthermore, genetic and phenotypic correlations were determined for different gas emission traits within methods (PAC and RC) and animal category (lamb and ewe), and between PAC measures and the same gas emission parameters determined in RC from lambs fed alfalfa pellets.

MATERIALS AND METHODS

The animal experiments conducted adhered to the guidelines of the 1999 New Zealand Animal Welfare Act and AgResearch Code of Ethical Conduct. The PAC and RC trials of the current study were, respectively, approved by the AgResearch Invermay (Mosgiel, NZ) and AgResearch Grasslands (Palmerston North, NZ) Animal Ethics committees.

Animals

Animals in the dataset were from 7 New Zealand Sheep Improvement Limited (SIL) flocks including 1 AgResearch flock (SIL flock number 2638), 3 Central Progeny Test (CPT) flocks (SIL flock numbers 4640, 4757, 9153), 2 industry flocks (SIL flock numbers 2629, 4474), and the “methane yield selection” flock (SIL flock number 3633). In total, 3601 lambs were measured at least once using PAC or RC of which 2,255 lambs (born between 2012 and 2015; at 7 ± 3 [±StDev; range 4 to 13] mo of age) had 4,733 separate 1-h measurement records using PAC and 2,110 lambs (born in 2007 and between 2009 and 2015; at 9 ± 2 [range 6 to 12] mo of age) had 8,655 daily records using RC. Of these, 844 animals had both PAC and RC measurements as lambs. In addition, 1,251 (2,387 records) animals were measured in PAC as yearling/ewes (called ewe in this paper from this point onwards; 42 ± 19 [range 16 to 88] mo of age) of which 874 animals also had RC measurements as lambs and 698 had PAC measurements as lambs.

Flock, birth...
year, sex, and measurement year and season are detailed in Supplementary Table S1.

“Methane yield selection” flock animals (Pinares-Patiño et al., 2013) were progeny of maternal dual-purpose sires generated by the New Zealand industry CPT program (McLean et al., 2006), comprising Coopworth, Romney, Perendale, Texel, and composite breeds, where the latter breed consisted primarily of combinations of the former breeds with additional infusions of Finn and East Friesian. All rams were mated to composite ewes. Ewe progeny born in 2007 and between 2009 and 2011 (and also ram lambs born in 2009) was measured for CH₄ emissions per unit of feed DMI (i.e., CH₄ yield) (Pinares-Patiño et al., 2013), with the most extreme low and high 10% retained for further breeding. The lines were closed in 2012 and all sires used from 2012 onwards were born in the high and low CH₄-yield selection flocks, which are currently maintained at 100 ewes per line. Progeny from the CH₄-yield selection flock included in the dataset was born between 2010 and 2015.

**Portable Accumulation Chamber Measurements**

Sheep were measured in PACs made of polycarbonate sheet with an internal volume of 827 L (1.17-m length × 1.15-m height × 0.615-m width), similar to those described by Goopy et al. (2011). A standard protocol was used where the animals were set stocked for 3 d prior to measurement on ad libitum pasture allowance with average pre- and post-grazing pasture masses of 2500 ± 550 kg DM/ha and 2000 ± 375 kg DM/ha (from ground level), respectively, determined using a rising plate meter (Farmworks Systems Ltd., Feilding, NZ). The day before PAC measurements pasture pluck samples were collected from the paddock grazed, dried at 65 °C, ground through a 1-mm screen, and scanned by near-infrared spectroscopy for nutritional composition analysis as described by Corson et al. (1999). The average pasture DM content was 18 ± 3.1% and contained in % of DM (± StDev): 9 ± 0.8 ash, 4 ± 0.6 lipids, 22 ± 4.2 crude protein, 50 ± 7.8 neutral detergent fiber, 23 ± 2.9 acid detergent fiber, 12 ± 3.4 soluble sugars and starch, and 81 ± 4.5 organic matter digestibility (Supplementary Table S2).

Before PAC measurements, animals were taken off-pasture at 9:00 a.m., weighed to determine BW, and PAC measurements conducted in lots of 12 animals for 1 h each. The first PAC measurements did not start until the animals were off-pasture for at least 30 min. Animals remained off-pasture until they were recorded, and time after removal from pasture was also recorded. Gas measurements were recorded at the start (0 min), middle (~30 min), and end (~60 min) of the PAC measurement period along with exact time of measurement. Ambient temperature and atmospheric pressure were also recorded. Concentrations of CH₄, CO₂, and O₂ in the PACs were determined using an ENVICO Eagle 2 handheld gas meter (The Environmental Collective Ltd., Auckland, NZ) following the manufacturer’s instructions, including calibrations and filter checks. The analyzer was calibrated daily, before and after all sheep were measured in PAC, using standard gases containing 100, 1,000, and 5,000 ppm CH₄ (BOC Ltd., Auckland, NZ). Little analyzer drift was detected and the quoted accuracy of the data in the current study is ±5% of the recorded value. The difference between individual measurements at the start and end of the measurement period was transformed using the standard gas (at standard temperature and pressure) equations to calculate g CH₄, g CO₂, and g O₂ (Brouwer, 1965), with inclusion of a correction for the volume of the animals in the chamber. The volume of the animal was calculated as 1 kg BW = 1.01 liter. The sum of CO₂ and CH₄ production in moles (total gas) and molar CH₄/(CO₂ + CH₄) ratio was calculated, as was the respiration quotient (RQ; i.e., CO₂/O₂). Measurements discarded where the water seal was broken by animal activity or if other abnormalities were detected. After animal removal from the PAC, a household leaf blower was used to reduce CH₄ and CO₂ concentrations to background levels and the floor was cleaned before the next animals were measured.

Measurements on the same animals using PAC were repeated after approximately 14 d. A subset of 289 lambs had additional repeat PAC measurements in a different season (Supplementary Table S1). A subset of 32 ewes had repeat PAC measurements in a different season (summer then autumn in 2013), and 19 ewes (born 2009) had PAC measurements in different years (2013 and 2015). The full schedule of measurements using PAC is provided in Supplementary Table S1.

**Respiration Chamber Measurements**

RC measurements were performed with the standard protocol described by Pinares-Patiño et al. (2013) using a system with 24 open-circuit RC described in detail by Pinares-Patiño et al. (2012). Approximately 96 sheep between 6 and 12 mo of age were fed alfalfa pellets (Dunstan Nutrition Ltd., Hamilton, NZ) at 2.0 × maintenance metabolizable energy requirements (MEm) according to Australian Feed Standards (CSIRO, 2007), and measurements were performed for 2 × 48-h periods approximately 14 d apart (round). Animals were weighed before
and after RC measurements to determine their average BW. The management of the 4 groups of 24 animals was staggered consecutively in time. The RC doors were opened twice daily around 08:30 a.m. and 03:30 p.m. for approximately 15 min for excreta removal and feeding. Emissions for these periods were extrapolated by taking the average of the last 12 values before opening the door. There were 2,110 lambs (born 2007 and 2009–2015) with 8655-d measurements in RC between 2008 and 2016.

**Animal Allocation to PAC and RC**

Animals were allocated to chambers (PAC and RC) using a randomized incomplete block design where allocation to a measurement “lot” of 12 animals in PAC or to a measurement “group” of 24 animals in RC was random within sire but balanced across lots/groups. Subsequent allocation to individual chambers was random across the animals allocated to a lot/group. Analysis revealed no consistent chamber effects, but lot/group effects were significant. This approach reduced confounding between lots/groups and genetic relatedness of individuals.

**Statistical Analysis**

Initially, a variety of fixed effects and covariate models were tested for traits measured using PAC with lambs, using PAC with ewes and using RC with lambs. For PAC traits, fixed effects fitted included birth/rearing rank (brr; born single, twin or triplet, reared as single, twin, or triplet) and contemporary groups PACcg4 (birth year—birth flock—sex combination) and PACcg5 (measurement date—PAClot combination, where PAClot is a group of 12 animals measured together). Covariates fitted were BW at PAC measurement, birth day deviation from birth year within birth flock mean (bdev) and age of dam as both a linear (aod) and a quadratic (aod^2) function. For RC traits, fixed effects fitted included brr and contemporary groups cg4 (birth year—birth flock—sex combination) and cg5 (birth year—lot—group—round combination, where lot defines a mob of 96 animals, group is a submob of up to 24 animals within a lot measured contemporaneously, and round indicates the measurement time 14 d apart). Covariates fitted for RC traits were bdev, aod, and aod^2. PAC and RC trait final models were obtained by backwards elimination (Supplementary Table S3). A choice was made not to fit BW as a covariate, but to estimate the genetic parameters separately, so that both the genetic and phenotypic covariances of the trait with BW could be estimated. This allows greater flexibility for subsequent modeling.

An investigation into whether scaling was required to adjust for the variance within contemporary PAC groups with varying waiting times after removal off-pasture till PAC measurements, and RC groups measured at consecutive times, clearly indicated that it was necessary. This was undertaken by calculating the ratio of the actual value/group mean and multiplying by the overall trait mean. PAC traits, except for BW, were deviated from the contemporary group (PACcg5) mean; RC traits except for BW, CH_4 + CO_2, and CH_4(CH_4 + CO_2) were deviated from the contemporary group (cg5) mean. For nonscaled traits, PACcg5 and cg5 were fitted in the appropriate analysis model for PAC and RC traits, respectively.

Pedigree records were obtained from all 7 birth flocks of animals, born from 1990 to 2015. The data were analyzed in a mixed model using ASREML 3.0 (Gilmour et al., 2009), fitting animal as a random effect with covariances proportional to the numerator relationship matrix (Supplementary Table S3). Maternal random effects were fitted for PAC measures on lambs and for RC traits. Maternal random effect for PAC gas measures in ewes approached zero and was, therefore, excluded from the model. For PAC measures on lambs, permanent environmental effects between rounds 14 d apart, and between seasons, were included in the model. For PAC measures on ewes, an additional permanent environment effect was added to account for measurements in different years. For RC, permanent environment effects were added to account for measurements on separate days within a round and between rounds 14 d apart. Single trait analyses provided heritability (direct and maternal) and repeatability estimates (Table 1 and Supplementary Tables S4–S6), whereas bivariate analyses provided genetic and phenotypic correlations between traits (Tables 2 and 3 and Supplementary Table S7). The heritability (h^2) was classified as low (<0.2), moderate (0.2 to 0.4), or high (>0.4); r_g as low (<0.2), moderate (0.2 to 0.6), and high (>0.6); and repeatability and r_p as low (<0.35), moderate (0.35 to 0.75), and high (>0.75). The efficiency (Q) of indirect selection for CH_4 yield determined in RC using a single measure of emission traits in PAC was calculated as follows: Q = r_g (h_pac/h_rc), where h is the square root of the heritability (Table 4).

**RESULTS**

**Heritability and Repeatability**

All PAC traits for lambs and ewes had moderate heritabilities between 0.16 and 0.35 (Table 1), except RQ, which was not heritable (Supplementary
Table 1. Heritability ($h^2$) and repeatability estimates (±SE) for gas emission parameters of lambs measured using respiration chambers and portable accumulation chambers (PAC) and ewes using PAC

| Trait                                      | Mean | StDev | $\sigma_r$ | $h^2$ (SE) | Repeatability (SE) |
|--------------------------------------------|------|-------|------------|------------|-------------------|
|                                            |      |       |            | Direct     | Maternal          |
|                                            |      |       |            | 14 day     | Season            |
|                                            |      |       |            |            | Year              |
| Respiration chamber, lambs                 |      |       |            |            |                   |
| BW, kg                                     | 45.9 | 8.00  | 4.80       | 0.35 (0.05)| 0.07 (0.03)       |
| CH$_4$ g/d                                 | 24.0 | 2.94  | 2.81       | 0.23 (0.04)| 0.05 (0.02)       |
| CO$_2$ g/d                                 | 1066 | 99.5  | 94.5       | 0.34 (0.05)| 0.03 (0.02)       |
| CH$_4$ + CO$_2$, mol/d                     | 25.5 | 3.71  | 2.34       | 0.33 (0.05)| 0.03 (0.02)       |
| CH$_4$/CH$_4$ + CO$_2$, mol/mol            | 0.059| 0.006 | 0.005      | 0.17 (0.03)| 0.03 (0.02)       |
| CH$_4$ yield, g/kg DMI                     | 16.0 | 1.42  | 1.39       | 0.13 (0.02)| 0.02 (0.02)       |
| Portable accumulation chamber, lambs       |      |       |            |            |                   |
| BW, kg                                     | 39.3 | 7.79  | 4.45       | 0.35 (0.07)| 0.08 (0.03)       |
| CH$_4$ g/d                                 | 7.5  | 2.12  | 2.13       | 0.19 (0.04)| 0.04 (0.02)       |
| CO$_2$ g/d                                 | 623  | 117.9 | 117.9      | 0.16 (0.04)| 0.07 (0.02)       |
| CH$_4$ + CO$_2$, mol/d                     | 14.6 | 2.73  | 2.73       | 0.16 (0.04)| 0.07 (0.02)       |
| CH$_4$/CH$_4$ + CO$_2$, mol/mol            | 0.032| 0.008 | 0.008      | 0.19 (0.04)| 0.02 (0.02)       |
| Portable accumulation chamber, ewes         |      |       |            |            |                   |
| BW, kg                                     | 69.9 | 8.96  | 7.60       | 0.33 (0.07)| 0.33 (0.07)       |
| CH$_4$ g/d                                 | 13.0 | 3.09  | 3.08       | 0.20 (0.05)| 0.39 (0.03)       |
| CO$_2$ g/d                                 | 987  | 232.9 | 229.7      | 0.27 (0.05)| 0.54 (0.02)       |
| CH$_4$ + CO$_2$, mol/d                     | 23.3 | 5.39  | 5.31       | 0.27 (0.05)| 0.54 (0.02)       |
| CH$_4$/CH$_4$ + CO$_2$, mol/mol            | 0.036| 0.009 | 0.009      | 0.25 (0.05)| 0.38 (0.03)       |

1Not determined.
2Maternal random effect for PAC gas measures in ewes approached zero and is, therefore, excluded from the model.

Table 2. Genetic correlation (±SE; below diagonal) and phenotypic correlation (±SE; above diagonal) for gas emission parameters within lambs in respiration chambers or portable accumulation chambers or within ewes in portable accumulation chambers

| Trait                                      | BW (Kg) | CH$_4$ (g/d) | CO$_2$ (g/d) | CH$_4$ + CO$_2$, mol/d | CH$_4$/CH$_4$ + CO$_2$, mol/mol | CH$_4$ g/kg DMI |
|--------------------------------------------|---------|--------------|--------------|------------------------|----------------------------------|-----------------|
| Respiration chamber, lambs                 |         |              |              |                        |                                  |                 |
| BW, kg                                     | –       | 0.61 (0.01)  | 0.78 (0.01)  | 0.78 (0.01)            | 0.04 (0.02)                      | 0.003 (0.02)    |
| CH$_4$ g/d                                 | 0.83 (0.04) | –           | 0.74 (0.01)  | 0.77 (0.01)            | –                                | 0.65 (0.01)     |
| CO$_2$ g/d                                 | 0.96 (0.01) | 0.84 (0.04) | –            | –                      | –                                | 0.67 (0.01)     |
| CH$_4$ + CO$_2$, mol/d                     | 0.93 (0.02) | 0.85 (0.04) | –            | –                      | –                                | 0.67 (0.01)     |
| CH$_4$/CH$_4$ + CO$_2$, mol/mol            | 0.09 (0.12) | 0.50 (0.09) | –0.01 (0.13) | 0.01 (0.11)            | –                                | 0.84 (0.01)     |
| CH$_4$, g/kg DMI                           | 0.02 (0.13) | 0.50 (0.10) | 0.06 (0.13)  | 0.07 (0.13)            | 0.93 (0.02)                      |                 |
| Portable accumulation chamber, lambs       |         |              |              |                        |                                  |                 |
| BW, kg                                     | –       | 0.43 (0.02)  | 0.36 (0.02)  | 0.38 (0.02)            | 0.22 (0.02)                      |                 |
| CH$_4$ g/d                                 | 0.52 (0.11) | –           | 0.41 (0.01)  | 0.44 (0.01)            | 0.79 (0.01)                      |                 |
| CO$_2$ g/d                                 | 0.55 (0.11) | 0.37 (0.14) | –            | –                      | –                                | 0.67 (0.01)     |
| CH$_4$ + CO$_2$, mol/d                     | 0.56 (0.11) | 0.41 (0.13) | –            | –                      | –                                | 0.67 (0.01)     |
| CH$_4$/CH$_4$ + CO$_2$, mol/mol            | 0.21 (0.14) | 0.82 (0.05) | –0.21 (0.15) | –0.17 (0.16)           | –                                |                 |
| Portable accumulation chamber, ewes         |         |              |              |                        |                                  |                 |
| BW, kg                                     | –       | 0.31 (0.02)  | 0.40 (0.02)  | 0.41 (0.02)            | –0.07 (0.03)                     |                 |
| CH$_4$ g/d                                 | 0.59 (0.13) | –           | 0.26 (0.03)  | 0.35 (0.02)            | 0.67 (0.02)                      |                 |
| CO$_2$ g/d                                 | 0.65 (0.10) | 0.32 (0.02) | –            | –                      | –                                | 0.67 (0.02)     |
| CH$_4$ + CO$_2$, mol/d                     | 0.65 (0.10) | 0.33 (0.15) | –            | –                      | –                                | 0.67 (0.02)     |
| CH$_4$/CH$_4$ + CO$_2$, mol/mol            | –0.13 (0.15) | 0.60 (0.02) | –0.60 (0.11) | –0.58 (0.11)           | –                                |                 |

Tables S4 and S5). Maternal heritability was significant for total gas and CO$_2$ production. These heritabilities were in a similar range to the heritability determined in RC (Table 1). Repeatabilities of gas measurement with PAC (for lambs and ewes) within season (14 d apart in PAC chambers) and between seasons were moderate, ranging from 0.21 to 0.54, which were, in general, lower than the comparable repeatabilities 14 d apart across rounds in RC. Repeatability of PAC gas
Table 3. Genetic and phenotypic correlations (±SE) for gas emission parameters determined using respiration chambers for lambs or portable accumulation chambers (PAC) for lambs and ewes

| Trait                      | Genotypic correlation          | Phenotypic correlation          |
|---------------------------|--------------------------------|--------------------------------|
|                           | BW    | CH₄    | CO₂   | CH₄ + CO₂ | CH₄/(CH₄ + CO₂) | BW    | CH₄    | CO₂   | CH₄ + CO₂ | CH₄/(CH₄ + CO₂) |
| PAC, lambs vs respiration chamber, lambs |        |        |       |          |                 |        |        |       |          |                 |
| BW, kg                    | 0.90(0.03) | 0.71(0.10) | 0.49(0.13) | 0.51(0.12) | 0.42(0.14) | 0.80(0.01) | 0.34(0.02) | 0.30(0.02) | 0.31(0.02) | 0.12(0.03) |
| CH₄, g/d                  | 0.63(0.09) | 0.67(0.11) | 0.04(0.17) | 0.08(0.17) | 0.73(0.13) | 0.54(0.02) | 0.27(0.02) | 0.19(0.02) | 0.20(0.02) | 0.14(0.02) |
| CO₂, g/d                  | 0.79(0.06) | 0.65(0.11) | 0.41(0.13) | 0.43(0.13) | 0.37(0.15) | 0.66(0.01) | 0.29(0.02) | 0.25(0.02) | 0.26(0.02) | 0.11(0.03) |
| CH₄ + CO₂, mol/d           | 0.82(0.05) | 0.68(0.11) | 0.40(0.13) | 0.42(0.13) | 0.44(0.15) | 0.69(0.01) | 0.32(0.02) | 0.26(0.02) | 0.27(0.02) | 0.12(0.03) |
| CH₄/(CH₄ + CO₂), mol/mol   | 0.08(0.15) | 0.38(0.15) | −0.36(0.18) | −0.33(0.18) | 0.57(0.13) | 0.05(0.03) | 0.08(0.02) | 0.01(0.02) | 0.01(0.02) | 0.08(0.02) |
| CH₄, g/kg DMI              | 0.05(0.16) | 0.34(0.15) | −0.38(0.18) | −0.35(0.18) | 0.54(0.14) | 0.03(0.03) | 0.08(0.02) | 0.01(0.02) | 0.01(0.02) | 0.08(0.02) |
| PAC, ewes vs. respiration chamber, lambs |        |        |       |          |                 |        |        |       |          |                 |
| BW, kg                    | 0.81(0.08) | 0.56(0.14) | 0.34(0.13) | 0.35(0.13) | 0.03(0.13) | 0.55(0.02) | 0.20(0.03) | 0.22(0.03) | 0.22(0.03) | −0.02(0.03) |
| CH₄, g/d                  | 0.57(0.11) | 0.62(0.14) | 0.15(0.14) | 0.16(0.14) | 0.32(0.15) | 0.36(0.03) | 0.16(0.03) | 0.13(0.03) | 0.13(0.03) | 0.03(0.03) |
| CO₂, g/d                  | 0.76(0.09) | 0.58(0.14) | 0.43(0.12) | 0.44(0.12) | −0.03(0.14) | 0.46(0.02) | 0.18(0.03) | 0.21(0.03) | 0.21(0.03) | −0.03(0.03) |
| CH₄ + CO₂, mol/d           | 0.75(0.09) | 0.59(0.13) | 0.41(0.12) | 0.41(0.12) | 0.01(0.14) | 0.48(0.02) | 0.19(0.03) | 0.22(0.03) | 0.23(0.03) | −0.02(0.03) |
| CH₄/(CH₄ + CO₂), mol/mol   | 0.14(0.14) | 0.33(0.15) | −0.18(0.14) | −0.17(0.14) | 0.50(0.14) | 0.06(0.03) | 0.05(0.02) | −0.01(0.03) | −0.01(0.03) | 0.06(0.02) |
| CH₄, g/kg DMI              | 0.15(0.14) | 0.29(0.15) | −0.22(0.15) | −0.23(0.15) | 0.47(0.14) | 0.03(0.03) | 0.06(0.02) | 0.01(0.02) | 0.01(0.02) | 0.05(0.02) |
| PAC, lambs vs PAC, ewes    |        |        |       |          |                 |        |        |       |          |                 |
| BW, kg                    | 0.89(0.07) | 0.65(0.11) | 0.58(0.13) | 0.60(0.13) | 0.36(0.15) | 0.67(0.02) | 0.30(0.03) | 0.31(0.03) | 0.32(0.03) | 0.10(0.03) |
| CH₄, g/d                  | 0.71(0.12) | 0.99(0.10) | 0.34(0.16) | 0.38(0.16) | 0.85(0.12) | 0.25(0.03) | 0.21(0.02) | 0.10(0.03) | 0.11(0.03) | 0.14(0.02) |
| CO₂, g/d                  | 0.61(0.12) | 0.44(0.13) | 0.93(0.08) | 0.93(0.08) | −0.25(0.15) | 0.24(0.03) | 0.12(0.03) | 0.31(0.02) | 0.31(0.02) | −0.07(0.03) |
| CH₄ + CO₂, mol/d           | 0.62(0.12) | 0.46(0.13) | 0.93(0.08) | 0.93(0.08) | −0.23(0.15) | 0.24(0.03) | 0.13(0.03) | 0.31(0.02) | 0.31(0.02) | −0.07(0.03) |
| CH₄/(CH₄ + CO₂), mol/mol   | 0.10(0.14) | 0.38(0.14) | −0.65(0.12) | −0.62(0.12) | 0.85(0.09) | 0.02(0.03) | 0.08(0.03) | −0.17(0.03) | −0.17(0.03) | 0.18(0.02) |
measurements of ewes between years was lower than within and between seasons of the same year. Repeatability was in general lower for the ratio traits [CH\textsubscript{4}/(CH\textsubscript{4} + CO\textsubscript{2}) and CH\textsubscript{4} yield] than for gas production traits (CH\textsubscript{4}, CO\textsubscript{2}, and CH\textsubscript{4} + CO\textsubscript{2}) for all 3 measurement datasets.

**Genetic and Phenotypic Correlations**

Genetic and phenotypic correlations within and between measurement datasets are given in Tables 2 and 3 and Supplementary Table S7. Within all 3 measurement datasets, \( r_g \) were moderate to high (0.52 to 0.96) for BW with gross emissions (CH\textsubscript{4}, CO\textsubscript{2}, and CH\textsubscript{4} + CO\textsubscript{2}) and for CH\textsubscript{4} (g/d) with CH\textsubscript{4}/(CH\textsubscript{4} + CO\textsubscript{2}) (0.50 to 0.82) (Table 2). Corresponding \( r_g \) were also moderate to high (0.31 to 0.78) for BW with gross emissions (CH\textsubscript{4}, CO\textsubscript{2}, and CH\textsubscript{4} + CO\textsubscript{2}) and for CH\textsubscript{4} with CH\textsubscript{4}/(CH\textsubscript{4} + CO\textsubscript{2}) (0.65 to 0.79). Genetic correlations of CH\textsubscript{4} production with CO\textsubscript{2} or CH\textsubscript{4} + CO\textsubscript{2} were moderate (0.32 to 0.41) when measured using PAC (both lambs and ewes), whereas these correlations were strong in RC (0.84 and 0.85). Phenotypic correlations of CH\textsubscript{4} production with CO\textsubscript{2} or CH\textsubscript{4} + CO\textsubscript{2} also were moderate (0.26 to 0.44) when measured using PAC (both lambs and ewes), whereas these correlations were moderate to strong in RC (0.74 to 0.77). Within RC data, CH\textsubscript{4} yield (g/kg DMI) had a strong \( r_p \) and \( r_g \) with CH\textsubscript{4}/(CH\textsubscript{4} + CO\textsubscript{2}) ratio (0.93 and 0.84, respectively) and moderate \( r_g \) and \( r_p \) with CH\textsubscript{4} production (0.50 and 0.67, respectively). Methane yield did not correlate significantly with other parameters. Genetic correlations for all gas emission parameters (e.g., CH\textsubscript{4} and CO\textsubscript{2}) of PAC measurements as lambs and ewes were very strong (\( r_g = 0.85 \) to 0.99) and much higher than the corresponding \( r_g \) (0.18 to 0.31) (Table 3).

Genetic correlations of PAC measurements as lambs and ewes with the same parameters measured in RC were moderate (\( r_g = 0.41 \) to 0.67), but higher than corresponding \( r_g \) (0.06 to 0.27). The efficiency (Q) of indirect selection for RC CH\textsubscript{4} yield using PAC traits (both as lambs and ewes) was greater using CH\textsubscript{4}/(CH\textsubscript{4} + CO\textsubscript{2}) ratio and CH\textsubscript{4} production with CH\textsubscript{4} + CO\textsubscript{2} as a covariate than for CH\textsubscript{4} production without a covariate or with BW as a covariate (Table 4).

**DISCUSSION**

**Genetic Parameters of Methane Production**

The heritability estimates for CH\textsubscript{4} production (g/d) and CH\textsubscript{4} yield (g/kg DMI) (0.23 and 0.13, respectively) with RC in the current study, which included an additional 3,400 records in addition to the record reported in Pinares-Patiño et al. (2013), remained similar to those previously reported (0.29 and 0.13, respectively)

---

**Table 4. Heritability \((h^2)\) (±SE) of emission parameters (determined using respiration chambers [RC] for lambs or portable accumulation chambers for lambs and ewes) without or with covariates, genetic correlations \((r_g)\) (±SE), and efficiency \((Q)\) for a single emission parameter measure to select indirectly for RC CH\textsubscript{4} yield (g/kg DMI)**

| Trait | Covariate | \(h^2\) (±SE) | \(r_g\) with RC CH\textsubscript{4} yield | Q value (%) |
|-------|-----------|---------------|---------------------------------|-------------|
| Respiration chamber, lambs | | | | |
| CH\textsubscript{4}, g/d | – | 0.27 (0.04) | 0.50 (0.10) | 73 |
| | BW, kg | 0.27 (0.04) | 0.50 (0.10) | 73 |
| | CO\textsubscript{2}, g/d | 0.20 (0.03) | 0.76 (0.06) | 95 |
| | CH\textsubscript{4} + CO\textsubscript{2}, mol/d | 0.20 (0.03) | 0.78 (0.06) | 97 |
| Portable accumulation chamber, lambs | | | | |
| CH\textsubscript{4}, g/d | – | 0.18 (0.03) | 0.93 (0.02) | 105 |
| | BW, kg | 0.18 (0.04) | 0.34 (0.15) | 40 |
| | CO\textsubscript{2}, g/d | 0.18 (0.04) | 0.51 (0.14) | 60 |
| | CH\textsubscript{4} + CO\textsubscript{2}, mol/d | 0.18 (0.04) | 0.53 (0.14) | 61 |
| Portable accumulation chamber, lambs | | | | |
| CH\textsubscript{4}, g/d | – | 0.16 (0.03) | 0.54 (0.14) | 61 |
| | BW, kg | 0.19 (0.05) | 0.29 (0.15) | 35 |
| | CO\textsubscript{2}, g/d | 0.19 (0.05) | 0.42 (0.15) | 49 |
| | CH\textsubscript{4} + CO\textsubscript{2}, mol/d | 0.19 (0.05) | 0.43 (0.15) | 51 |
| Portable accumulation chamber, lambs | | | | |
| CH\textsubscript{4}, g/d | – | 0.24 (0.05) | 0.48 (0.15) | 65 |

\(Q = r_g(h_{PAC\text{emission\ trait}}/h_{RC\text{CH}_4\text{yield}})^2\), where \(h\) is the square root of the heritability.
(Pinares-Patiño et al., 2013). Similar heritability estimates for CH$_4$ production and yield (0.27 and 0.22, respectively) were found for Angus beef cattle when measured in RC (Hayes et al., 2016). The heritability estimates for CH$_4$ production in PAC as lambs and ewes were similar (0.19 and 0.20, respectively) to those in RC in the current study and similar heritability estimates were reported for sheep at different sites in Australia measured 2 or 3 times in PAC for 1 h (0.19 and 0.23, respectively) (Robinson et al., 2014). There was a strong $r_g$ for CH$_4$ production measured in PAC as lambs and ewes with CH$_4$ production in RC (0.67 and 0.62, respectively) in the current study, suggesting that they are similar genetic traits. Repeatability of CH$_4$ production approximately 14 d apart in PAC (both as lambs and ewes) was, however, only about half the repeatability in RC (0.33 to 0.39 vs. 0.65), which is consistent with previous findings in sheep (0.24 vs. 0.49, respectively), reported by Bickell et al. (2011). During the course of PAC measurements, the quality of the pasture varied over time, DMI varied due to ad libitum feed being offered, and the time of removal from pasture until the time of PAC measurement differed for the same individual sheep on which for PAC measurements were made 14 d apart. In contrast, the batch of alfalfa pellets, the feeding level, and measurement time were the same for RC measurements made on the same individual sheep 14 d apart. These likely explain the lower repeatability with PAC compared with the controlled experimental conditions during RC measurements. Interestingly, repeatability of CH$_4$ production in PAC 14 d apart and in different seasons (within 1 yr) was similar, suggesting that the accuracy of the measurement remained consistent over a long time period. Donoghue et al. (2016) also found a similar repeatability of CH$_4$ production and CH$_4$ yield in beef cattle when comparing a RC measurement and repeat measurements after between 61 and 120 d and after between 121 and 450 d.

**Genetic Parameters of Methane Yield**

Methane yield (g/kg DMI) could not be determined with grazing sheep measured using PAC since DMI is not known. There was, however, a moderately positive $r_g$ for CH$_4$ production measured in PAC both as lambs and ewes (0.34 and 0.29) with CH$_4$ yield measured in RC, and an $r_g$ of 0.50 for CH$_4$ production measured in RC with CH$_4$ yield measured in RC. These suggest that genetic selection for reduced CH$_4$ production in PAC will also reduce CH$_4$ yield, with a moderate efficiency (Q-value) of 40% and 35% for lambs and ewes, respectively.

The main driver of CH$_4$ production is DMI, explaining 76% and 90% of the variation in sheep and cattle fed cut pasture (Swainson et al., 2018; Jonker et al., 2017a). Selection for reduced CH$_4$ production might therefore result in selection of animal with lower DMI. This is not a problem if these animals have similar performance (e.g., low residual feed intake trait and same number of lambs born), whereas animal numbers on the farm remain the same. However, if growth performance is proportionally reduced, or lambs born per ewe are decreased, then total gross emissions of the farm system might remain similar or increase. However, selection for reduced CH$_4$ production can also result in selection of animals with reduced CH$_4$ yield either due to increased DMI effects (Swainson et al., 2018) or due to sheep genetics (Pinares-Patiño et al., 2013). Therefore, if selecting animals with similar performance traits (McLean et al., 2006; Byrne et al., 2012; Paganoni et al., 2017), selection for PAC CH$_4$ production should result in animals with either lower DMI (but similar performance) or lower CH$_4$ yield (or both) and therefore reduce gross farm system emissions. This selection might, however, result in a wide range of phenotypes with low CH$_4$ emissions, and this might result in animals with different CH$_4$ genotypes when individuals with different underlying phenotypes are used for breeding.

**Relationship of CO$_2$ and CH$_4$ Emissions**

Voluntary DMI with ad libitum feed offered is in general related to the BW of the animal (CSIRO, 2007) and to gaseous carbon emissions (Aubry and Yan, 2015; Jonker et al., 2016). The use of animal BW or CH$_4$ + CO$_2$ emissions as a covariate for CH$_4$ production (Herd et al., 2016) or using the ratio of CH$_4$ to CH$_4$ + CO$_2$ emissions (Madsen et al., 2010; Lassen et al., 2012) has therefore been suggested as indicators of CH$_4$ yield when DMI is unknown. The genetic correlation with CH$_4$ yield in RC was greatest for the CH$_4$/CH$_4$ + CO$_2$ ratio measured with PAC (both as lambs and ewes) ($r_g = 0.54$ and 0.47), slightly lower for CH$_4$ production with CH$_4$ + CO$_2$ emissions as a covariate ($r_g = 0.53$ and 0.43), and much lower for CH$_4$ production with BW as a covariate ($r_g = 0.40$ and 0.29). Within RC, $r_g$ with CH$_4$ yield was stronger for the CH$_4$/CH$_4$ + CO$_2$ ratio ($r_g = 0.93$) than for CH$_4$ production with either CH$_4$ + CO$_2$ emissions ($r_g = 0.78$) or BW ($r_g = 0.53$) as a covariate. Therefore, CH$_4$/CH$_4$ + CO$_2$ ratio appears to most genetically similar to CH$_4$ yield, following
by $\text{CH}_4$ production with $\text{CH}_4 + \text{CO}_2$ emissions as a covariate, whereas $\text{CH}_4$ production with or without BW as a covariate appeared genetically less similar to $\text{CH}_4$ yield in the current study. In growing beef cattle, however, there was a strong $r_g$ and $r_p$ between $\text{CH}_4$/per unit of BW and $\text{CH}_4$ yield (Donoghue et al., 2013; Herd et al., 2014). This metric has been suggested as a possible measure of $\text{CH}_4$ intensity (per unit of animal product) in growing sheep and beef cattle (Donoghue et al., 2013; Herd et al., 2014; Robinson et al., 2014), which is probably suitable for growing animals of similar age but might result in the selection of heavier animals when determined in mature ewes.

The heritability estimates for the $\text{CH}_4/(\text{CH}_4 + \text{CO}_2)$ ratio (0.17 to 0.25) and $\text{CH}_4$ production with either $\text{CH}_4 + \text{CO}_2$ emissions (0.19 to 0.20) or BW (0.15 to 0.27) as a covariate determined using RC or PAC with lambs and ewes were also, in general, slightly greater than for $\text{CH}_4$ yield in RC (0.13). A similar heritability estimate (0.16) for $\text{CH}_4$/CO$_2$ ratio was found in dairy cows when measured in multiple spot-samples during visits to milk robots (Lassen and Lovendahl, 2016). Heritability of $\text{CH}_4$ production with BW as a covariate (0.19) was previously found to be similar to $\text{CH}_4$ yield (0.21) in beef cattle measured in RC (Donoghue et al., 2013). Altogether these results suggest that $\text{CH}_4/(\text{CH}_4 + \text{CO}_2)$ ratio and $\text{CH}_4$ production with $\text{CH}_4 + \text{CO}_2$ emissions as a covariate in PAC are the most similar genetic traits to $\text{CH}_4$ yield in RC.

Methane yield in sheep decreases with increasing DMI, even with small increases in DMI (Swainson et al., 2018; Jonker et al., 2018), whereas the $\text{CH}_4$/CO$_2$ ratio increases with increasing DMI (Lassen et al., 2012; Jonker et al., 2016). Also, the $\text{CH}_4$/CO$_2$ ratio is not constant during the day, with the ratio being higher during the day than during the night in lactating dairy cows (Lassen et al., 2012; Brask et al., 2015). It decreases in sheep with increasing time off-pasture (Robinson et al., 2014). Robinson et al. (2014) found that there was quite a steep decline in $\text{CH}_4$ emissions in sheep with increasing time from pasture, whereas CO$_2$ emissions declined only slightly during the same time. Scaling of contemporary groups (12 animals measured at once in separate PAC units) was performed in the current study relative to overall mean value, which should eliminate this effect of time off-pasture. Within RC measurements from 684 sheep, it was found that there is a strong positive $r_g$ for $\text{CH}_4$ production (0.89) and yield (0.76) separated in $24 \times 1$-h intervals with daily average of the same parameters (McEwan et al., 2012), suggesting that sampling time would have a minor effect on the genetic selection. This correlation was, however, not reported for the $\text{CH}_4/(\text{CH}_4 + \text{CO}_2)$ ratio.

Hellwing et al. (2013) found that $\text{CH}_4$ production predicted with $\text{CH}_4$/CO$_2$ ratio explained 55% of the variation in actual $\text{CH}_4$ production in lactating dairy cows, with the majority of the rest of the variation explained by differences in $\text{CH}_4$, CO$_2$, milk production, and BW. There was a negligible to low $r_g$ and $r_p$ for BW with $\text{CH}_4/(\text{CH}_4 + \text{CO}_2)$ within any of 3 datasets (PAC with lambs and ewes and lambs in RC) in the current study, suggesting that selection for $\text{CH}_4/(\text{CH}_4 + \text{CO}_2)$ would not lead to selection for animals with a greater BW. There was a positive $r_g$ and $r_p$ for $\text{CH}_4$ production with $\text{CH}_4/(\text{CH}_4 + \text{CO}_2)$ in all 3 datasets in the current study, which was also observed, in terms of $r_p$, in sheep (Robinson et al., 2016) and in 3 datasets generated from beef cattle fed 3 different diets (Herd et al., 2016). A similar trend was found within RC, with $\text{CH}_4$ production having a positive $r_g$ and $r_p$ with $\text{CH}_4$ yield in the current study, as also found in growing beef cattle (Donoghue et al., 2013; Herd et al., 2014). These positive relationships are favorable traits indicating that selection for reduced $\text{CH}_4/(\text{CH}_4 + \text{CO}_2)$ or $\text{CH}_4$ yield will also reduce gross $\text{CH}_4$ emissions. In ewes (but not in lambs with either RC or PAC), however, the daily CO$_2$ and $\text{CH}_4 + \text{CO}_2$ production had moderate negative $r_g$ and $r_p$ with $\text{CH}_4$ yield in RC (0.19 to 0.20) within any of 3 datasets (PAC with lambs and ewes and lambs in RC). The selection for reduced $\text{CH}_4/(\text{CH}_4 + \text{CO}_2)$ using PAC in ewes might, therefore, concurrently result in selection for increased CO$_2$ and $\text{CH}_4 + \text{CO}_2$ production, which had positive correlations with gross $\text{CH}_4$ emissions. To avoid unintended selection for increased CO$_2$ and $\text{CH}_4 + \text{CO}_2$ production (strong positive relationship with DMI), which might happen as one selects for reduced $\text{CH}_4/(\text{CH}_4 + \text{CO}_2)$ at least with PAC in ewes, it has been suggested that $\text{CH}_4$ production with $\text{CH}_4 + \text{CO}_2$ as a covariate or regressed against $\text{CH}_4 + \text{CO}_2$ production with a residual $\text{CH}_4$ production trait (Herd et al., 2016) could be a suitable metric. However, in practice, the best approach is to use the heritability and $r_g$ of the individual objective traits weighted by their economic value, either positive for production traits or negative for inputs such as DMI (related to CO$_2$ or $\text{CH}_4 + \text{CO}_2$ production) and $\text{CH}_4$ production. This approach does, however, require accurate estimates of the $r_p$ PAC and RC emissions and with DMI, which is currently in progress.

**Emissions From Lambs and Ewes**

Genetic selection for reduced $\text{CH}_4$ emissions in NZ has been performed in growing lambs to make the fastest genetic progress possible (Pinares-Patiño et al., 2013). Approximately 20 million ewe population that produces approximately 25 million lambs (tailed or marketed) in NZ (StatsNZ, 2015),
however, contributes to the majority of all CH₄ emissions (approximately 75% to 80%) from sheep in NZ (Muetzel and Clark, 2015; Swainson et al., 2018). It is therefore very important to prove that the low CH₄ trait selected in lambs is also expressed in ewes. Previous results from sheep fed alfalfa pellets measured in RC indicated that CH₄ production and CH₄ yield traits were similarly repeatable for measurements 14 d apart and across different years (Pinares-Patiño et al., 2013), and suggest that these traits were also expressed at adult age. There were strong $r_g$ (0.85 to 0.99) for all traits measured using PAC in lambs compared with those measured using PAC in ewes in the current study. The $r_g$ for emission traits measured in RC (lambs) were similar to emission traits measured using PAC with either lambs (0.41 to 0.67) or ewes (0.41 to 0.62) in the current study. The heritability and repeatability of all emission parameters determined in PAC were also similar for lambs and ewes. These suggest that emission traits determined as lambs were genetically similar to those measured as ewes. Robinson et al. (2016) also found a strong $r_p$ between CH₄ and CO₂ production (0.64 and 0.75, respectively) measured using PAC as lambs and as ewes; Paganoni et al. (2017) found a strong $r_g$ for PAC measured CH₄ production (>0.60) at post-weaning, yearlings, and as adult sheep; and Oddy et al. (2018) found no interaction between progeny sire and measurement age (12, 21, and 28 mo of age). Opposite, Dominik and Oddy (2015) suggested, based on repeatability estimates, that CH₄ traits measured using PAC in Merino ewes (15 mo) on pasture were not a reliable indicator of the adult (27 mo) CH₄ trait. However, the range in repeatabilities they reported (0.17 to 0.40) for CH₄ with BW as a covariate was in a similar range to those in the current study for the same trait (0.10 to 0.40). Recent meta-analysis of RC data from sheep fed cut pasture suggested that also the regression for CH₄ emissions and DMI was similar for lambs (<1 yr) and older sheep (>1 yr) (Muetzel and Clark, 2015; Swainson et al., 2018). Altogether, these suggest that the CH₄ emission trait selected in lambs is a genetic trait that is also expressed when the animal is an ewe as also suggested by Oddy et al. (2018). However, they recommended to measure CH₄ traits in dry ewes, not when pregnant or lactating, although without clear explanation other than that CH₄ yield was lower in pregnant and lactating sheep than in dry ewes and lambs.

**Repeatability of Emissions on Different Diets**

The initial studies screening sheep for low CH₄ emission traits in NZ were performed while sheep were fed alfalfa pellets at a fixed feeding level (Pinares-Patiño et al., 2013), which is very different from the typical grazing conditions on NZ farms. The average CH₄ yield on alfalfa pellets is very low, at approximately 16 g/kg DMI (Pinares-Patiño et al., 2013) compared with the CH₄ yield of approximately 21 to 23 g/kg DMI more typically found in sheep fed cut pasture in RC (Muetzel and Clark, 2015; Swainson et al., 2018). Genotype × environment interactions were found for CH₄ emissions in some studies, including effects of diet changes, although these effects mainly affected the magnitude of differences between CH₄ selection lines (Pinares-Patiño et al., 2003; Pinares-Patiño et al., 2011b; Robinson et al., 2015; Goopy et al., 2016). There is therefore some uncertainty if the CH₄ emission trait selected on a particular diet is also expressed when animals are fed a different diet. The similar heritability and repeatability and moderate $r_p$ for CH₄/(CH₄ + CO₂) in PAC off-pasture with CH₄/(CH₄ + CO₂) in RC on alfalfa pellets in the current study suggest that both are a similar genetic trait. Differences in CH₄ yield between the low and high CH₄ yield line sheep ($n = 96$) were 10% when fed alfalfa pellets and on average 9% when fed cut pasture in 3 periods measured using RC (Jonker et al., 2017b). Selection of low CH₄ yield sheep on relatively fibrous low energy mixed alfalfa:oaten chaff in Australia resulted in selection of animals with a smaller rumen and increased rumen digesta passage kinetics (Goopy et al., 2014), which is similar to phenotypic findings for sheep with low CH₄ yield selected on alfalfa pellets in NZ (Pinares-Patiño et al., 2011a; Bain et al., 2014; Elmes et al., 2014). These suggest that a CH₄ emission trait selected in sheep on a particular diet is also expressed when the same animals are fed another diet.

**Phenotypic Parameters of Methane Emissions**

In general, $r_p$ were lower (0.004 to 0.27) than $r_g$ (0.41 to 0.67) for PAC emission parameters (as lambs and ewes) with RC emission parameters. Environmental conditions imposed before and during PAC vs. RC measurements were different, which likely explains the low $r_p$ in the current study. During RC, alfalfa pellets were fed at a fixed feeding level and the same batch of pellets was fed during the repeat measurement after 14 d, whereas before PAC, animals had ad libitum access to pasture, likely resulting in more variable DMI among animals. Pasture quality also varied during repeat measurements. In addition, age and BW differed during PAC and RC measurements. Robinson et al. (2015) also found negligible $r_p$ (0.04 to 0.19) for sheep measured using PAC at 5 occasions using different measurement...
protocols with measurements of the same sheep in RC. The $r_p$ of CH$_4$ production measured using PAC and RC were, however, much greater (0.56 to 0.71) when animals entered PAC direct after removal from RC (Bickell et al., 2011; Goopy et al., 2011). The regression for PAC with RC measurements presented by Goopy et al. (2011) revealed some bias, with an underestimation of CH$_4$ emissions at low CH$_4$ concentrations in PAC. This might explain the low CH$_4$ production estimated using PAC in the current study relative to CH$_4$ production from sheep fed cut pasture in RC (Muetzel and Clark, 2015; Swainson et al., 2018). The CH$_4$ production estimates using PAC in ewes in the current study were, however, in a similar range as previously found using PAC (Robinson et al., 2014; Robinson et al., 2015; Goopy et al., 2016). Accuracy of PAC measurements was similar when performed for 1 and 2 h (Goopy et al., 2011) and also when performed for 40 min and 1 h (Robinson et al., 2014). Therefore, more frequent spot-sampling will likely be required to estimate absolute values more precisely. Even so, the current protocol with 2 PAC samples off-pasture is suitable to estimate relative differences in CH$_4$ emissions for genetic selection.

**CONCLUSIONS**

The ranking of CH$_4$ emission traits in sheep for the purpose of selection for low emissions using PAC was highly correlated ($r_p$) with selection using RC. Furthermore, methane emissions measured in lambs were highly genetically correlated (>0.9) with measures as ewes. There were some differences, mainly attributed to the ability of the animal to vary DMI, variable pasture quality on offer, and shorter sampling time during PAC measurements. PAC have several major advantages over RC as they enable measurement of methane emissions on farm at pasture, at lower cost, and more animals can be measured in a shorter time. Before wide-scale industry use, accurate estimates of the genetic correlations of PAC and RC gas measurements with DMI are required.

**SUPPLEMENTARY DATA**

Supplementary data are available at *Journal of Animal Science* online.

**LITERATURE CITED**

Aubry, A., and T. Yan. 2015. Meta-analysis of calorimeter data to establish relationships between methane and carbon dioxide emissions or oxygen consumption for dairy cattle. Anim. Nutr. 1:128–134. doi:10.1016/j.aninu.2015.08.015

Bain, W. E., L. Bezuidenhout, N. B. Jopson, C. S. Pinares-Patiño, and J. C. McEwan. 2014. Rumen differences between sheep identified as being low or high emitters of greenhouse gas. Proc. Assoc. Advmt. Anim. Breed. Genet. 20:376–378.

Bickell, S. L., D. L. Robinson, A. F. Toovey, J. P. Goopy, R. S. Hegarty, D. K. Revell, and P. E. Vercoe. 2011. Four week repeatability of daily and one hour methane production of mature merino wethers fed ad libitum. Proc. Assoc. Advmt. Anim. Breed. Genet. 19:415–418.

Brask, M., M. R. Weisbjerg, A. L. Hellwing, A. Bannink, and P. Lund. 2015. Methane production and diurnal variation measured in dairy cows and predicted from fermentation pattern and nutrient or carbon flow. Animal 9:1795–1806. doi:10.1017/S1751731115001184

Brouwer, E. 1965. Report of sub-committee on constants and factors. In: Blaxter, K.L., editor Energy metabolism of farm animals. EAAP Publ. No. 11. Academic Press, London, UK. p. 441–443.

Byrne, T. J., C. I. Ludemann, P. R. Amer, and M. J. Young. 2012. Broadening breeding objectives for maternal and terminal sheep. Livest. Sci. 144:20–36. doi:10.1016/j.livsci.2011.10.010

Corson, D. C., G. C. Waghorn, M. J. Ulyatt, and J. Lee. 1999. NIRS: Forage analysis and livestock feeding. Proc. N.Z. Grassl. Assoc. 61:127–132.

CSIRO. 2007. Nutrient requirements of domesticated ruminants. CSIRO Publishing, Melbourne, Victoria, Australia.

Dominik, S., and V. H. Oddy. 2015. Repeatabilities for methane emissions in Merino ewes on pasture across different ages. Proc. Assoc. Advmt. Anim. Breed. Genet. 21:110–113.

Donoghue, K. A., T. Bird-Gardiner, P. F. Arthur, R. M. Herd, and R. S. Hegarty. 2016. Repeatability of methane emission measurements in Australian beef cattle. Anim. Prod. Sci. 56:213–217. doi:10.1071/AN15573

Donoghue, K. A., R. M. Herd, S. H. Bird, P. F. Arthur, and R. F. Hegarty. 2013. Preliminary genetic parameters for methane production in Australian beef cattle. Proc. Assoc. Advmt. Anim. Breed. Genet. 20:290–293.

Elmes, S. N., W. E. Bain, G. J. Greer, S. M. Hickey, E. A. Young, N. K. Pickering, S. J. Rowe, K. J. Knowler, C. S. Pinares-Patiño, and J. C. McEwan. 2014. An exploratory investigation of the effects of selection for divergence in methane emissions on rumen digesta and carcass traits in eight-month-old sheep. Proc. N.Z. Soc. Anim. Prod. 74:142–144.

Gilmour, A. R., B. J. Gogel, R. Cullis, and R. Thompson. 2009. ASReml user guide release 3.0. VSN Int. Ltd., Hemel Hempstead, UK.

Goopy, J. P., A. Donaldson, R. Hegarty, P. E. Vercoe, F. Haynes, M. Barnett, and V. H. Oddy. 2014. Low-methane yield sheep have smaller rumens and shorter rumen retention time. Br. J Nutr. 111:578–585. doi:10.1017/S0007114513002936

Goopy, J. P., D. L. Robinson, R. T. Woodgate, A. J. Donaldson, V. H. Oddy, P. E. Vercoe, and R. S. Hegarty. 2016. Estimates of repeatability and heritability of methane production in sheep using portable accumulation chambers. Anim. Prod. Sci. 56:116–122. doi:10.1071/AN13370

Goopy, J. P., R. Woodgate, A. Donaldson, D. L. Robinson, and R. S. Hegarty. 2011. Validation of a short-term methane measurement using portable static chambers to estimate daily methane production in sheep. Anim. Feed Sci. Technol. 166–167:219–226. doi:10.1016/j.anifeedsci.2011.04.012
Hellwing, A. L. F., P. Lund, J. Madsen, and M. R. Weisbjerg. 2013. Comparison of enteric methane production predicted from the CH$_4$/CO$_2$ ratio and measured in respiration chambers. Adv. Anim. Biosci. 4:557.

Herd, R. M., P. F. Arthur, S. H. Bird, K. A. Donoghue, and R. S. Hegarty. 2014. Genetic variation for methane traits in beef cattle. In: 10th World Congr. Genet. Appl. Livest. Prod. Vancouver, BC, Canada.

Herd, R. M., J. I. Velazco, P. F. Arthur, and R. S. Hegarty. 2016. Proxies to adjust methane production rate of beef cattle when the quantity of feed consumed is unknown. Anim. Prod. Sci. 56:231–237. doi:10.1071/AN15477

Jonker, A., G. Molano, C. Antwi, and G. C. Waghorn. 2016. Enteric methane and carbon dioxide emissions measured using respiration chambers, the sulfur hexafluoride tracer technique, and a greenfeed head-chamber system from beef heifers fed alfalfa silage at three allowances and four feeding frequencies. J. Anim. Sci. 94:4326–4337. doi:10.2527/jas.2016-0646

Jonker, A., G. Molano, J. Koolaard, and S. Muetzel. 2017a. Methane emissions from lactating and non-lactating dairy cows and growing cattle fed fresh pasture. Anim. Prod. Sci. 57:643–648. doi:10.1071/AN15656

Jonker, A., E. Sandoval, P. Boma, S. Hickey, J. McEwan, P. H. Janssen, and S. Rowe. 2017b. Sheep selected for divergent methane yield on lucerne pellets also express the same trait when fed fresh pasture. Adv. Anim. Biosci. 8:206.

Jonker, A., G. Molano, E. Sandoval, P. S. Taylor, C. Antwi, S. Olinga, and G. P. Cosgrove. 2018. Methane emissions differ between sheep offered a conventional diploid, a high-sugar diploid or a tetraploid perennial ryegrass cultivar at two allowances at three times of the year. Anim. Prod. Sci. 58:1043–1048. doi:10.1071/AN15597

Lassen, J., and P. Lovendahl. 2016. Heritability estimates for enteric methane emissions from Holstein cattle measured using noninvasive methods. J. Dairy Sci. 99:1959–1967. doi:10.3168/dsj.2015-10012

Lassen, J., P. Lovendahl, and J. Madsen. 2012. Accuracy of noninvasive breath methane measurements using Fourier transform infrared methods on individual cows. J. Dairy Sci. 95:890–898. doi:10.3168/dsj.2011-4544

Madsen, J., B. S. Bjerg, T. Hvelplund, M. R. Weisbjerg, and P. Lund. 2010. Methane and carbon dioxide ratio in excreted air for quantification of the methane production from ruminants. Livest. Sci. 129:223–227. doi:10.1016/j.livsci.2010.01.001

McEwan, J. C., S. M. Hickey, S. M. Young, K. G. Dodds, S. McLean, G. Molano, E. Sandoval, H. Kjestrup, C. Hunt, and C. Pinares-Patiño. 2012. Heritability estimates for hourly measures of methane emissions. 33rd Conf. Int. Soc. Anim. Genet. No. P4021, Cairns, Australia.

McLean, N. J., N. B. Jopson, A. W. Campbell, K. Knowler, M. Behrent, G. Cruckshank, C. M. Logan, P. D. Muir, T. Wilson, and J. C. McEwan. 2006. An evaluation of sheep meat genetics in New Zealand: the central progeny test (CPT). Proc. N.Z. Soc. Anim. Prod. 66:368–372.

MfE. 2017. New Zealand’s greenhouse gas inventory 1990–2015. http://www.mfe.govt.nz/node/23304/ (Accessed 27 April 2018.)

Muetzel, S., and H. Clark. 2015. Methane emissions from sheep fed fresh pasture. N.Z. J. Agric. Res. 58:472–489. doi:10.1080/00288283.2015.1090460

Oddy, V. H., A. J. Donaldson, M. Cameron, J. Bond, S. Dominik, and D. L. Robinson. 2018. Variation in methane production over time and physiological state in sheep. Anim. Prod. Sci. doi:10.1071/AN17447

Paganoni, B., G. Rose, C. Macleay, C. Jones, D. J. Brown, G. Kearney, M. Ferguson, and A. N. Thompson. 2017. More efficient sheep produce less methane and carbon dioxide when eating high-quality pellets. J. Anim. Sci. 95:3839–3850. doi:10.2527/jas2017.1499

Pinares-Patiño, C. S., S. H. Ebrahimi, J. C. McEwan, H. Clark, and D. Luo. 2011a. Is rumen retention time implicated in sheep differences in methane emission? Proc. N.Z. Soc. Anim. Prod. 71:219–222.

Pinares-Patiño, C. S., S. M. Hickey, E. A. Young, K. G. Dodds, S. MacLean, and G. Molano. 2013. Heritability estimates of methane emissions from sheep. Animal 7:316–321. doi:10.1017/S1751731113000864

Pinares-Patiño, C. S., C. Hunt, R. Martin, J. West, P. Lovejoy, and G. C. Waghorn. 2012. Chapter 1: New Zealand Ruminant Methane Measurement Centre, AgResearch, Palmerston North. In: C. S. Pinares-Patiño and G. C. Waghorn, editors, Technical manual on respiration chamber design. Ministry of Agriculture and Forestry, Wellington, New Zealand.

Pinares-Patiño, C. S., J. C. McEwan, K. G. Dodds, E. A. Cárdenas, R. S. Hegarty, J. P. Koolaard, and H. Clark. 2011b. Repeatability of methane emissions from sheep. Anim. Feed Sci. Technol. 166–167:210–218. doi:10.1016/j.anifeedsci.2011.04.068

Pinares-Patiño, C. S., M. J. Ulyatt, K. R. Lassey, T. N. Barry, and C. W. Holmes. 2003. Persistence of differences between sheep in methane emission under generous grazing conditions. J. Agric. Sci. 140:227–233. doi:10.1017/S0021859603003071

Robinson, D. L., M. Cameron, A. J. Donaldson, S. Dominik, and V. H. Oddy. 2016. One-hour portable chamber methane measurements are repeatable and provide useful information on feed intake and efficiency. J. Anim. Sci. 94:4376–4387. doi:10.2527/jas.2016-0620

Robinson, D. L., J. P. Goopy, R. S. Hegarty, and V. H. Oddy. 2015. Comparison of repeated measurements of methane production in sheep over 5 years and a range of measurement protocols. J. Anim. Sci. 93:4637–4650. doi:10.2527/jas.2015-9092

Robinson, D. L., J. P. Goopy, R. S. Hegarty, V. H. Oddy, A. N. Thompson, A. F. Toovey, C. A. Macleay, J. R. Bregal, R. T. Woodgate, A. J. Donaldson, et al. 2014. Genetic and environmental variation in methane emissions of sheep at pasture. J. Anim. Sci. 92:4349–4363. doi:10.2527/jas.2014-8042

Schafer, H., S. E. Mikaloff Fletcher, C. Veidt, K. R. Lassey, G. W. Brailsford, T. M. Bromley, E. J. Dlugokencky, S. E. Oddy, V. H., A. J. Donaldson, M. Cameron, J. Bond, S. Dominik, and D. L. Robinson. 2016. A 21st-century shift from fossil-fuel to biogenic methane emissions indicated by δ¹³CH$_4$. Science 352:80–84. doi:10.1126/science.aad2705.

StatsNZ. 2015. Agricultural production statistics: June 2015 (final). http://archive.stats.govt.nz/browse_for_stats/industry_sectors/agriculture-horticulture-forestry/AgriculturalProduction_final_HOTPJun15final/Tables.aspx (Accessed 27 April 2018.)

Swainson, N., S. Muetzel, and H. Clark. 2018. Updated predictions of enteric methane emissions from sheep suitable for use in the New Zealand national greenhouse gas inventory. Anim. Prod. Sci. 58:973–979. doi:10.1071/AN15766