

## Genetic and genomic solutions to improve feed efficiency and reduce environmental impact of dairy cattle

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**ABSTRACT:** Traits related to resource use efficiency are dry matter intake (DMI), residual feed intake (RFI) and methane (CH<sub>4</sub>) emission. In an experimental dataset of 588 heifers, we showed that it is possible to decrease CH<sub>4</sub> emission (predicted from DMI and ration composition) by selecting more efficient cows. Resource use efficiency phenotypes are difficult and expensive to measure, but genomic selection is a promising tool to enable selection for resource efficient cows. Using genomic selection, a reduction in predicted CH<sub>4</sub> (g/d) of 15% in 10 years is theoretically possible. For DMI, an international collaboration between 9 countries in Europe, US and Australasia has been established to assemble DMI data on >6,000 cows with phenotypes and genotypes. With all these developments, genetic selection is likely to make a major contribution to improving resource use efficiency, as long as feeding and management are adapted accordingly.

**Keywords:** Dairy cattle; Feed efficiency; Environmental impact

### Introduction

Improving resource use efficiency is of growing international interest. For example, it is well established that the release of greenhouse gases (GHG) is a contributing factor to climate change. The global livestock sector, particularly ruminants, contributes approximately 18% of total anthropogenic GHG emissions (Steinfeld et al. (2006)). Additionally, because feed costs represent >50% of total costs of dairy production, lowering feed intake, while maintaining the same production level and health and fertility status, is a viable approach to increase herd profitability.

This paper reviews our recent work to facilitate selective breeding for improved resource use efficiency. This entails firstly a strategy to enable selection for feed intake and efficiency, and secondly a strategy to reduce environmental impact of dairy cattle.

### Dry Matter Intake

**National efforts.** The major handicap in selecting for improved feed efficiency in dairy cattle is the lack of individual cow dry matter intake (DMI) records. Several countries, in recent decades, have recorded individual cow DMI (Svendsen et al. (1993); Veerkamp et al. (1994); Buckley et al. (2000); Veerkamp et al. (2000); Buttchereit et al. (2011);

Williams et al. (2011)). All these studies documented considerable variation in feed intake and feed efficiency; however, inconsistencies exist between studies in estimates of genetic (co)variances, because of large associated standard errors due to small datasets. Thus, it is difficult to draw firm conclusions. Moreover, a major limitation is that it is impossible to generate accurate breeding values for DMI from these small datasets which could be used in national breeding programs, since recording of DMI in a progeny testing scheme is generally not possible. Some countries initiated nucleus schemes where DMI was recorded on breeding cows, but the impact has, to date, been limited.

A novel approach to obtain breeding values for DMI in a population is to use genomic selection, where phenotypes (e.g., DMI) are measured in a subset of the population and genomic predictions are calculated for other animals that have genotypes, but no phenotypes. Since this approach is very appealing for facilitating selection for improved efficiency, we embarked on the necessary research early on. The first approach was based on one experimental dataset with ~600 first parity heifers in the Netherlands (Verbyla et al. (2010)). However, the size of the reference population from which the genomic prediction equations were derived was too small to achieve satisfactory levels of accuracy of genomic breeding values. The dataset was therefore expanded with data from nutritional experiments, and although the dataset grew to 3,179 lactations with at least one weekly record for DMI, this was still too limited for accurate genomic predictions (Veerkamp et al. (2014)). The same is true for other national datasets; they are simply too small to develop effective selection tools for dairy cattle within country.

**International collaboration.** One approach to possibly improve the accuracy of genomic prediction is to combine datasets from multiple populations. Several challenges, however, exist when combining phenotypes from several countries such as the possible existence of genotype by environment (GxE) interactions, as well as possible differences in trait definitions and recording schemes.

**Europe-Australia pilot study.** The aim of this pilot study was to estimate the accuracy of genomic prediction for DMI, when analysed together in a single-trait evaluation, or in a multi-trait evaluation, using both Australian data on growing heifers and European data on lactating first parity cows (De Haas et al. (2012)).

In total, DMI records were available on 1801 animals; 843 Australian (AU) growing heifers with records on DMI measured over approximately a 70 day test period at approximately 200 days of age (Williams et al. (2011); Pryce et al. (2012)), 359 Scottish (UK) and 588 Dutch (NL) lactating heifers with records on DMI during the first 100 days in milk (Banos et al. (2012); Veerkamp et al. (2012)). The genotypes used in this study were from the Illumina Bovine50 Beadchip with 54,001 single nucleotide polymorphisms (SNPs) (UK and NL), or the Illumina High Density Bovine SNP chip, which comprises 777,963 SNP markers (AU). The AU, UK and NL genomic data were matched using the SNP name. Quality controls were applied by carefully comparing the genotypes of 40 bulls that were available in each dataset. This resulted in a total of 30,949 SNPs being used in the analyses. Genomic predictions were estimated with genomic REML (G-REML), using ASReml (Gilmour et al. (2009)). The accuracy of genomic prediction was evaluated in 11 validation sets. The reference set (where animals had both DMI phenotypes and genotypes) was either 1) within AU, 2) within Europe (UK and NL combined), or, 3) with a multi-country reference set consisting of all data except the validation set.

When DMI for each country was treated as the same trait (i.e., univariate analysis), using a multi-country reference set (uni-multi) the accuracy of genomic prediction increased for DMI for UK, compared to the accuracy achieved with a univariate analysis with just the national reference set. The accuracy did, however, not increase for AU and NL (Table 1).

**Table 1. The average of the approximated accuracy of genomic prediction of dry matter intake (DMI), calculated as the correlation between genomic breeding value (GEBV) and the true breeding value (TBV), estimated in a univariate, bivariate or trivariate run between Australia (AU), Europe (EU), United Kingdom (UK) and the Netherlands (NL), where “uni within” refers to the current situation with a national reference set. In all other analyses, a multi-country reference set was taken consisting of all data except the validation set (reproduced from De Haas et al. (2012))**

Country	uni within	uni multi	AU-EU	AU-UK-NL
AU	0.38	0.34	0.39	0.39
UK	0.30	0.33	0.32	0.33
NL	0.33	0.31	0.33	0.33

Extending the model to a bivariate (AU-EU) or trivariate (AU-UK-NL) model increased the accuracy of genomic prediction for DMI in all countries (De Haas et al. (2012)). The greatest accuracies were achieved in all countries when data were analysed with a trivariate model. Hence an important conclusion of this study was that a multi-trait model needs to be used to assume traits measured in

different environments are separate traits, and therefore treat both the GxE interaction and differences in trait definitions properly. Unfortunately, the combined dataset was still not large enough to provide accurate genomic predictions.

**global Dry Matter Initiative.** In a global initiative, DMI data from ten research herds in nine countries in Europe, US and Australasia were combined to address the major question: “Can accurate direct genomic breeding values (DGVs) be predicted for DMI for each country?”.

A total of 224,174 test-day records from 10,068 parity one to five records from 6,957 cows, as well as records from 1,784 growing heifers were available (Berry et al. (2014)). Genotype data of most of these animals were also available. The heifers from the Australian and New Zealand research herds were already genotyped at high density. The remaining genotypes were imputed from the Illumina Bovine50 beadchip to the Illumina high density beadchip (Pryce et al. (2014)). After editing, 591,213 genotypes on 5,999 animals remained.

**gDMI genetic parameters.** Random regression models were fit to the lactating cow test-day records and predicted feed intake at 70 days post calving was extracted from these fitted profiles (Berry et al. (2014)). The random regression model included a fixed polynomial regression for each parity separately as well as herd-year-season of calving and experimental treatment as fixed effects; random effects in the model included individual animal deviation from the fixed regression for each parity as well as mean herd-specific deviations from the fixed regression. Predicted DMI at 70-d post-calving was used as the phenotype for the genetic analyses undertaken using an animal repeatability model.

Heritability estimates of predicted cow feed intake 70-d post-calving was 0.34 across the entire dataset and varied, within population, from 0.08 to 0.52 (Berry et al. (2014)). Repeatability of feed intake across lactations was 0.66. Heritability of feed intake in the growing heifers was 0.20 to 0.34. The genetic correlation between feed intake in lactating cows and growing heifers was 0.67.

A combined pedigree and genomic relationship matrix ( $H^{-1}$  matrix) was used to improve linkages between populations for the estimation of genetic correlations of DMI in lactating cows; genotype information was available on 5,429 of the animals. Populations were categorized as North America, Grazing, Low input EU, and High input EU. Genetic correlation estimates for DMI between populations varied from 0.14 to 0.84 but were stronger (0.76 to 0.84) between the populations representative of high input production systems (Table 2). Genetic correlations with the grazing populations were weak to moderate varying from 0.14 to 0.57. These results suggest that genetic evaluations for DMI can be undertaken using data collated from interna-

tional populations; GxE interactions, however, with grazing production systems, in particular, need to be considered.

**Table 2. Genetic correlations (standard errors in parenthesis) between dry matter intake measured in groups of countries: North-America (NA), EU high-input (EUh), EU low-input (EUI) and Grazing (Gr) (reproduced from Berry et al. (2014))**

Region	NA	EUh	EUI
EUh	0.76 (0.21)		
EUI	0.79 (0.38)	0.84 (0.14)	
Gr	0.14 (0.43)	0.33 (0.20)	0.57 (0.43)

To estimate the pairwise genetic correlations between each separate country in gDMI, 55 bivariate analyses will be run to estimate all genetic and residual (co)variances for average predicted DMI at 70 DIM with an animal linear mixed models in ASReml (Gilmour et al. (2009)), using the  $H^{-1}$  matrix. The model to be used is:

$$Y_i Y_j = \mu + \text{fixed effects} + \text{animal} + e$$

where  $\mu$  is the overall mean.  $Y$  is the average predicted DMI at 70-d post-calving for cows, and the average DMI during the recording period for the growing heifers, and  $i$  and  $j$  are the ten research herds in gDMI, plus a set of 101 Australian lactating heifers that were recorded for DMI in their first parity as well. These records are included as a separate group. The fixed effects included in the model are parity and herd-year-season of calving. Finally, *animal* is fitted as a random additive genetic effect, distributed following  $N(0, A\sigma_g^2)$ , where  $A$  is the numerator relationship matrix based on the combined pedigree and genomic relationship matrix, and  $e$  is the residual term.

**gDMI genomic predictions.** The accuracies of genomic predictions of DMI in ten validation populations will be estimated by excluding each of those populations one at a time from the reference population (De Haas et al. (2014)). Validation populations are subsets of the dataset based on progeny groups of sires in the different countries, and each validation population represent all sources. With this approach it can be determined if the accuracy of a bull's DGV can be increased by using multi-country reference population.

Correlations will be calculated for each of the different validation populations between the DGVs estimated for all individuals in that validation population with an 11-trait (ten research herds plus the Australian lactating heifers) analysis in MiXBLUP (Mulder et al. (2010)) and their phenotype. The accuracy of true breeding values will then be approximated by dividing this correlation by the square root of the estimated heritability of DMI (0.27, see Berry et al. (2014)). These accuracies will finally be averaged across all 10 vali-

dation populations within each country to answer the question whether accurate DGVs can be predicted for DMI for each country.

### Predicted Methane Emission

Opportunities for nutritional and microbial manipulation to reduce enteric methane ( $CH_4$ ) emissions from livestock have been extensively researched and reviewed by several groups (e.g. (Beauchemin et al. (2008); McAllister and Newbold (2008))). An additional mitigation measure which is inexpensive and provides a long-term effect would be the use of natural variation to breed for animals with lower  $CH_4$  yield per unit intake [g  $CH_4$ /kg dry matter intake (DMI)] (Cavanagh et al. (2008); Vlaming et al. (2008)). Recent forums have begun to address the potential impact of animal genetics on emission intensity at individual animal and whole-farm levels (Chagunda et al. (2009); Wall et al. (2010)). Genetic improvement of livestock is a particularly cost-effective technology, producing permanent and cumulative changes in performance.

Measuring  $CH_4$  emission rates directly in animals is difficult and hinders direct selection on reduced  $CH_4$  emission. Therefore, in our first study we aimed to quantify phenotypic and genetic variation in predicted methane ( $CH_4$ ) emission (PME), and to examine the potential use of genomic selection to facilitate selection programs (De Haas et al. (2011)).

Dutch data from previous nutritional experiments were used, and records on daily DMI, weekly live weight and weekly milk production were available from 588 heifers (Veerkamp et al. (2000)). Predicted methane emission (g/d) is 6% of gross energy intake (method of International Panel on Climate Change (IPCC)) corrected for energy content of methane (55.65 kJ/g). All heifers were genotyped using the Illumina 50K SNP panel. Effects of SNPs were estimated using Bayesian stochastic search variable selection (SSVS; (George and McCulloch (1993))). A 10 fold cross-validation approach was employed to assess the accuracies of the two sets of predicted breeding values by correlating them with either PME or RFI.

The estimated heritability for PME was 0.35 (Table 3). Using the genetic standard deviations in PME, theoretical predictions can be made about the expected impact of genetic selection. When assuming a genetic progress of 0.22 genetic SD per year (e.g., as in a classical dairy cattle breeding program (Rendel and Robertson (1950))), it follows that the average PME can be reduced in ten years by about 13 kg per cow per lactation, i.e. from 120 kg to 107 kg per average cow per lactation, or from 13 grams per kg milk to 9 grams per kg milk. Thus there is clear potential for genetic improvement for these two traits, with a potential reduction of 11 and 26% in ten years, for PME (De Haas et al. (2011)).

**Table 3. The estimated heritability (on diagonal), phenotypic (above diagonal) and genetic correlation (below diagonal) for residual feed intake (RFI) and predicted methane emission (PME). The corresponding standard errors are shown in parentheses (reproduced from De Haas et al. (2011))**

	RFI	PME
RFI	0.40 (0.11)	0.72 (0.08)
PME	0.32 (0.06)	0.35 (0.12)

At first sight this might seem an excessively large improvement. However, when ranking the 588 cows in the current database on PME per kg milk, the 50 best cows produced 11.31 gram PME per kg milk, and the 50 worst cows produced 16.20 gram PME per kg milk (De Haas et al. (2011)). The worst cows produced therefore 42% more PME per kg milk than the best cows. An improvement up to 25% can thus be assumed to be realistic.

### Enteric Methane Emission

Predicted methane emission as described above was a function of DMI, and little information is known on opportunities to mitigate direct enteric CH<sub>4</sub> via animal genetics. However, genetic diversity in a range of digestive parameters likely to be associated with enteric CH<sub>4</sub> production was apparent when reviewed by Hegarty (2004). The prospect for selection for a CH<sub>4</sub> trait was investigated by multiple groups; some identified variation in CH<sub>4</sub> traits amenable to animal selection (Robinson et al. (2010)) but some did not (Munger and Kreuzer (2008)). More recent research in beef (Donoghue et al. (2013)) and sheep (Pinares-Patino et al. (2013)) is increasingly supportive of CH<sub>4</sub> traits being heritable with improvement by direct selection achievable. In dairy cattle, a heritability for CH<sub>4</sub> (measured in ppm; i.e., as a concentration) of 0.21 has been documented (Lassen and Lovendahl (2013)).

Enteric CH<sub>4</sub> emissions (as g CH<sub>4</sub>/day, or as g CH<sub>4</sub>/kg DMI) can certainly be described as a “difficult to measure trait”. Methods to measure enteric CH<sub>4</sub> emissions currently available are expensive and time consuming and subject animals to artificial environments. The methods to measure CH<sub>4</sub> emissions under natural production environments (pasture, feedlot or dairy feeding station) sample CH<sub>4</sub> for only a part of a day and require repeat measurements; moreover their precision is questioned. Genomic selection opens the possibility to efficiently select for these difficult to measure traits.

To date no genomic studies have been performed on direct enteric CH<sub>4</sub>, but improvements in reducing CH<sub>4</sub> can possibly be made through selection on associated traits (e.g. feed efficiency (Verbyla et al. (2010))), or through selection on CH<sub>4</sub> predicted from feed intake and diet composition (De

Haas et al. (2011)). In dairy cattle, a positive genetic correlation between residual feed intake (RFI) and PME of 0.32 indicates that cows with lower RFI have lower PME as well (Table 3). Genomic selection of RFI and PME showed that the genomic model produced breeding values with reliability double (for RFI) or triple (for PME) that of the breeding values produced by the polygenic model (Table 4). This strengthens the hypothesis that genomic selection opens the possibility to efficiently select for these difficult to measure traits.

**Table 4. Reliabilities of estimated breeding values (EBV) based on pedigree information only, and direct genomic values (DGV) based on both pedigree and marker (SNP) information for residual feed intake (RFI) and predicted enteric methane emission (PME) (reproduced from Verbyla et al. (2010) and De Haas et al. (2011))**

	RFI	PME
EBV (Pedigree)	0.14	0.04
DGV (Pedigree + SNP)	0.27	0.14

It has been shown in beef that selection for RFI (less feed / weight and weight gain) results in lower total emissions of CH<sub>4</sub>/head and a small non-significant increase in methane yield (g CH<sub>4</sub>/kg DMI) (Hegarty et al. (2007)). Selection for low methane yield has been demonstrated in sheep (Pinares-Patino et al. (2013)). The mechanisms that contribute to genetic variation in methane yield (gCH<sub>4</sub>/kg DMI) of individual animals may include: reduced fermentation of organic matter in the rumen, due to shorter retention time of digesta and smaller rumen volume, different microbial population in the rumen and potentially reductive acetogenesis. The extent to which these combine to produce natural variation in CH<sub>4</sub> yield is unknown, but data from measurements of CH<sub>4</sub> yield by sheep in respiration chambers suggest that the coefficient of variation is 10.3% (Pinares-Patino et al. (2013)) and for cattle is 14%. This shows that not only for PME, but also for direct enteric CH<sub>4</sub>, it would not be unreasonable to anticipate a response to long term selection to exceed 2 SDs from the mean, suggesting that a reduction of up to 25% in CH<sub>4</sub> yield may be feasible through selection of livestock for low CH<sub>4</sub> yield (Pickering et al. (2014)). \

### Overall conclusions

The genetic and genomic solutions to improve feed efficiency and reduce environmental impact of dairy cattle were examined. The major handicap in selecting for improved feed efficiency is the lack of individual DMI records on large numbers of animals. Our national and international efforts provide possible solutions on how to overcome this, and have shown that genetic solutions to improve resource efficiency traits are possible. International collaboration to assemble data on more cows improves the accuracy and

genetic gain. Higher accuracies and larger genetic gains can be achieved by using genomic selection for these minimally recorded traits. With all the developments, genetic selection is likely to make a major contribution to the improvement of resource use efficiency, as long feeding and management are adapted accordingly.

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