

RUNNING HEAD: Genetics of Feed Efficiency in Dairy Cattle

INVITED REVIEW: Genetic mechanisms underlying feed utilization and implementation of genomic selection for improved feed efficiency in dairy cattle

Luiz F. Brito^{1,2}, Hinayah R. Oliveira^{1,2}, Kerry Houlahan², Pablo A. S. Fonseca², Stephanie Lam², Adrien M. Butty², Dave J. Seymour^{2,3}, Giovana Vargas², Tatiane C. S. Chud², Fabyano F. Silva⁴, Christine F. Baes^{2,5,6}, Ángela Cánovas^{2,6}, Filippo Miglior^{2,6}, Flavio S. Schenkel^{2,6,*}

¹Department of Animal Sciences, Purdue University, West Lafayette, Indiana, 47907, USA

²Centre for Genetic Improvement of Livestock, Department of Animal Biosciences, University of Guelph, Guelph, Ontario, N1G 2W1, Canada

³Centre for Nutrition Modelling, Department of Animal Biosciences, University of Guelph, Guelph, Ontario, N1G 2W1, Canada

⁴Department of Animal Sciences, Federal University of Viçosa, Viçosa, Minas Gerais, 36570-000, Brazil

⁵Institute of Genetics, Vetsuisse Faculty, University of Bern, Bern, 3001, Switzerland

⁶These authors share last co-authorship

***Corresponding author:** Flavio S. Schenkel, Centre for Genetic Improvement of Livestock, Department of Animal Biosciences, University of Guelph, Guelph, Ontario, N1G 2W1, Canada.

Phone number: (+1) 519-824-4120, **E-mail:** schenkel@uoguelph.ca

Abstract: The economic importance of genetically improving feed efficiency has been recognized by cattle producers worldwide. It has the potential to considerably reduce costs, minimize environmental impact, optimize land and resource use efficiency, and improve the overall cattle industry's profitability. Feed efficiency is a genetically complex trait that can be described as units of product output (e.g. milk yield) per unit of feed input. The main objective of this review paper is to present an overview of the main genetic and physiological mechanisms underlying feed utilization in ruminants and the process towards implementation of genomic selection for feed efficiency in dairy cattle. In summary, feed efficiency can be improved via numerous metabolic pathways and biological mechanisms through genetic selection. Various studies have indicated that feed efficiency is heritable and genomic selection can be successfully implemented in dairy cattle with a large enough training population. In this context, some organizations have worked collaboratively to do research and develop training populations for successful implementation of joint international genomic evaluations. The integration of “-omics” technologies, further investments in high-throughput phenotyping, and identification of novel indicator traits will also be paramount in maximizing the rates of genetic progress for feed efficiency in dairy cattle worldwide.

Key Words: environmental footprint, feed efficiency, genomic selection, residual feed intake, rumen microbiome

Introduction

The global human population is expected to reach 9.8 billion by 2050 (FAOSTAT, 2019), and consequently there will be a substantial increase in food demand. In addition, the projected reduction in poverty and expansion of the middle class will reflect in a greater demand for larger amounts of high-quality meat and dairy products, produced under exemplary welfare conditions and leaving a minimal environmental footprint. As there are limited land and natural resources for production expansion, there is an urgent need to develop strategies to optimize the efficiency of food production.

The current worldwide cattle population has more than 1.5 billion animals; over 105 million cattle are raised in Canada and the United States alone (FAOSTAT, 2019). With feed currently being the largest expense in cattle production (Ho et al., 2013; Connor, 2015), a small improvement in nutrient utilization (i.e. better digestibility and/or greater nutrient absorption) can have major economic and environmental impacts. The reduction in feeding costs will positively impact not only the cattle producers' profitability, but also the final prices of meat and dairy products available to consumers.

The demand to optimize animal nutrition practices has led to important investments in research over the past decades. Consequently, the science of animal nutrition has evolved rapidly and resulted in major contributions to a better understanding of the nutritional physiology of cattle and its nutrient requirements. This had led to major advancements in diet formulation, supplementation, and techniques for food processing and storage (Eastridge, 2010; Coffey et al., 2016; Ondarza and Tricarico, 2017; Tedeschi et al., 2017). Despite the clear effectiveness of these developments, the need for a more permanent and cumulative solution has been envisioned

through genetic selection for a long time in various livestock species, including cattle (e.g. Freeman, 1967; Herd et al., 2003; Koch et al., 1963; Stone et al., 1960).

The economic importance of selecting for improved feed efficiency has been clearly recognized by cattle producers, due to its potential to reduce costs considerably, minimize environmental impacts (e.g. reduce nutrient loss in manure and methane intensity), optimize land and resource use efficiency, and improve the overall cattle industry profitability (Richardson and Herd, 2004; Basarab et al., 2013; Berry and Crowley, 2013). However, the inclusion of feed efficiency in cattle selection indexes used in commercial breeding programs has been delayed for various reasons, among them: 1) the limited amount of phenotypic records for feed efficiency and related variables, in commercial herds; 2) the differences in feed intake measurement protocols and data sources (e.g. different breeds, lactation stages, parities, diets); 3) unclear definition of the breeding goal (Berry and Crowley, 2013; Pryce et al., 2014; Connor, 2015; Hurley et al., 2016); and, 4) the lack of research on novel traits evaluated based on a systems biology level that could contribute to improve the accuracy genomic prediction of breeding values. In the case of beef cattle, there are even some additional challenges, including limited vertical integration of production, large diversity of genetic resources (breeds) within country and internationally, greater use of crossbreeding systems, and reduced use of artificial insemination compared to dairy cattle, which leads to weaker genetic linkage among populations, and consequently, less accurate genomic breeding values.

With the more recent advancements in genomic methods and technologies, selection for feed efficiency has become more feasible, as genomics can be used as a tool to transfer the knowledge generated on research farms to genetically-connected commercial populations (Connor, 2015). However, selection based on genomic information still requires genotyping of the

selection candidates, as well as continued collection of phenotypic and genotype records from genetically-representative individual animals (i.e. a training population). The main objective of this review is to present an overview of worldwide research efforts to unravel genetic, molecular and physiological mechanisms underlying the efficiency of feed utilization in ruminants, current knowledge on host-microbiota interactions, and the implementation process of genomic selection for improved feed efficiency in dairy cattle.

Definitions of feed efficiency and indicator traits

Dairy cattle breeding programs have been very successful in improving the main traits of interest for the industry (e.g. Miglior et al., 2017). The first step in moving genetic progress in a desired direction for any breeding program is the clear definition of the breeding goal. In this context, a feed efficient animal has been broadly defined as an animal that eats less without compromising performance, or an animal that produces more while consuming the same amount of feed. In other words, feed efficiency is related to the units of product output (e.g. milk production) per unit of feed input. These units are generally mass, energy, protein or an economic value (Vandehaar et al., 2016). It is also of interest to dairy cattle breeders to select animals that do not compromise other vital functions, such as reproduction and health, while breeding to achieve greater feed efficiency (Connor, 2015).

Feed efficiency is a complex trait, as feed intake and nutrient utilization are associated with many biological and physical mechanisms. For example, variability in feed efficiency can be due to variation in feed intake levels, digestion of feed and the associated energy costs, absorption of nutrients, metabolism, physiological stage, health status, rumen microbial metabolism, activity and thermoregulation (Herd et al., 2004; Herd and Arthur, 2007; Patience et al., 2015; Li et al., 2016).

Due to the challenging nature of measuring feed efficiency, many indicator traits have been proposed and utilized to assess feed efficiency, such as residual feed intake (**RFI**), residual solids production (**RSP**), and the use of milk mid-infrared spectroscopy (**MIR**) (Berry and Crowley, 2013; Koch et al., 1963; Coleman et al., 2010; Pryce et al., 2014; Connor, 2015; Hurley et al., 2016; Ondarza and Tricarico, 2017).

In 1963, Koch et al. suggested the use of RFI as an indicator of feed efficiency. The RFI variable, estimated through a regression model, corresponds to the difference (residual) between the observed and expected feed intake, where the expected feed intake is based on feeding requirements assessed according to metabolic body weight and level or quantity of product outcome. Other physiological activities that are energy demanding, such as maintenance and reproduction, can also be included in the calculations (Berry and Crowley, 2013; Pryce et al., 2014). Most commonly, RFI has been used in beef cattle research (Berry and Crowley, 2013). More recently, studies in dairy cattle have also been reported (e.g. Connor et al., 2019; Flay et al., 2019; Waghorn et al., 2012).

In dairy cattle, RFI is estimated by regressing dry matter intake (**DMI**) on a variety of physiological activities, which commonly include production (milk yield or energy corrected milk), metabolic body weight, changes in body weight or body condition score (**BCS**) and stage of lactation (Connor, 2015; Byskov et al., 2017; Seymour et al., 2019). Other residual traits have also been analyzed in place of RFI to obtain an estimate closer to the biology behind feed efficiency, such as RSP (Coleman et al., 2010). Similar to RFI, RSP represents the difference between observed milk solids production and that estimated via regression based on various activities (Coleman et al., 2010).

Another group of feed efficiency indicators are based on nutrient usage, such as energy and nitrogen efficiency, which considers nutrient partitioning between milk production and other physiological functions (Ondarza and Tricarico, 2017). For instance, energy conversion efficiency is the milk energy output divided by metabolizable energy intake. It has the advantage to consider diverse nutrient efficiency, however it does not account for mobilization of body reserves. Therefore, in order to account for body reserve changes, residual energy intake (**REI**) has been proposed (Mantysaari et al., 2012; Liinamo et al., 2015; Fischer et al., 2018). REI is the actual metabolizable energy intake minus the predicted energy requirement of the cow based on production, bodyweight, changes in bodyweight and/or BCS, and gestational energy needs (Mantysaari et al., 2012; Fischer et al., 2018).

As the costs to measure feed intake on individual animals are still high, alternative approaches to measure feed efficiency have been investigated. For instance, predictor traits that can be measured on a large number of animals for a relatively low cost through milk samples, blood, biosensors and automated recording systems are of great interest. Some examples of these include: infrared thermography (Montanholi et al., 2010), plasma concentrations of IGF-1 (Moore et al., 2005), milk MIR spectrometry (O'Donovan et al., 2014; Wallén et al., 2018), and milk fatty acid composition (Kelly et al., 2010). The use of MIR spectrometry to measure energy balance in dairy cattle began in 2011 (McParland et al., 2011; 2012). In addition MIR is widely used to determine major milk components, such as fat and protein. Shetty et al. (2017) used a partial least squares approach to estimate DMI based on MIR spectral data. While further studies are necessary, such models are a promising way to estimate individual energy intake (Dórea et al. 2018; Seymour et al., 2019). In summary, multiple alternatives to quantify individual variability of feed efficiency in dairy cattle have been proposed. As reviewed by Ondarza and Tricarico (2017), each one of

them has advantages and disadvantages. In order to make a better decision on the indicator trait to be used in a breeding program, it is of utmost value to understand the physiological mechanisms of feed utilization and the genetic architecture of the traits utilized. The next sections of this review will cover these aspects.

An overview of the main physiological mechanisms underlying feed efficiency

The expected physiological changes arising from genetic selection for improved feed efficiency are dependent on the feed efficiency metric (i.e. indicator trait) used. For example, gross feed efficiency is typically calculated as the ratio of milk output to feed intake. As a result, this trait can be improved by increasing milk yield, decreasing feed intake, or a combination of both strategies. More complex measures of feed efficiency, such as RFI (Koch et al., 1963), REI (Fischer et al., 2018), or net energy efficiency (Seymour et al., 2020), share some commonalities. This includes the concept of categorizing energy expenditures into maintenance, growth or production activities, and will be the focus of this section. However, regardless of the measure (i.e. trait) used, the physiology of feed efficiency can be partitioned into two main areas: those regulating voluntary feed intake, and those regulating the conversion of nutrients into milk. On another layer, the major components affecting feed efficiency can be divided into: those that alter maintenance or the portion of net energy that is captured in milk or body tissues (instead of being used for maintenance), and those that alter the conversion of gross energy to net energy (VandeHaar et al., 2016).

All measures of feed efficiency are dependent on the amount of feed consumed, and thus the regulation of voluntary feed intake is a major determinant of efficiency. The physiological regulation of DMI is a complex and multi-factorial process, and was comprehensively described

by Allen (2000), Allen et al. (2009) and Forbes (2000, 2007). The predominant factors known to affect voluntary feed intake in ruminants are reticulorumen distension due to gut fill, hepatic propionate flux, and the amount of lipid in the diet (Allen, 2000). Gut fill is considered to be the major limiting factor of feed intake in early lactation when energy demands are highest, where mechanoreceptors are triggered by reticulorumen wall distension and send negative feedback to the brain via the vagus nerve to reduce feed intake (Allen, 2000; Forbes, 2000). The hepatic oxidation theory (Allen et al., 2009) postulates that the oxidation of fuels in the liver, such as propionate, acts as a major signal integrator that regulates feed intake in response to whole body energy status. There is a plethora of other signaling mechanisms involved in the regulation of feed intake, and a single stimulus may act through multiple pathways (polymodality), as well as at different sites (polytopicity; Forbes, 2007).

After consuming feed, animals partition the available energy to various processes which are normally categorized into maintenance, growth, reproduction or production. In general, animals who partition a greater proportion of energy towards productive purposes are considered to be more feed efficient. The biological processes governing anabolic and catabolic processes are generally considered to be highly regulated and subject to strict thermodynamic constraints (Baldwin et al., 1980; Seymour et al., 2020), making genetic selection for improved efficiency of these pathways somewhat challenging. However, Bottje et al. (2019) have recently provided support for the theory that defective proteins in the electron transport chain may lead to suboptimal mitochondrial function and reduce the overall energetic efficiency of the animal. If the genes associated with these protein defects could be identified, genomic variants (e.g. single nucleotide polymorphisms – **SNPs**) could be given greater weight in sophisticated genomic prediction

methods that are currently available. However, this would likely necessitate increased selection intensity on dam of dam lines, as mitochondrial DNA is strictly maternal in origin.

An important physiological change associated with improved feed efficiency (RFI) in dairy cattle is a reduction in body size appropriate for the specific production system, as proposed by Dickerson (1978) and Vandehaar et al. (2016). This would serve to reduce the total energy partitioned towards maintenance processes, allowing for a greater proportion of energy to be directed towards productive purposes. While this will reduce milk yield and feed intake, selection for improved lactation persistency and management for extended lactations would help maintain total lactation milk yield (Capuco et al., 2006; Santschi et al., 2011a;b). Additionally, it is generally accepted that smaller body size is associated with improved fertility. Thus, animals would remain in the herd longer, which would result in fewer animals needed to produce a given volume of milk in a specific time period. Overall, these outcomes would likely improve the efficiency of both the individual animal and the overall herd. The nutritional management system in each farm also needs to be considered. As described by Vandehaar et al. (2016), under limited feeding and management, significant gains in feed efficiency can be captured by further diluting maintenance (e.g. smaller cows). However, the effect of multiples of maintenance on efficiency might be similar on weather animals are selected to produce more milk at a specific body weight, or the same milk yield with smaller body weight (Vandehaar et al., 2016).

Genetic architecture of feed efficiency

Genetic parameters for feed efficiency traits

Before including a trait in a genetic selection index, it is important to evaluate its heritability (h^2) and genetic variance in the population of interest, as well as its genetic correlation

with other economically important traits. These genetic parameters give insights into the rate of genetic progress that can be achieved per generation and contribute to better designing the genetic evaluation systems. Studies have indicated that feed efficiency, assessed in different ways using indicator traits, is moderately heritable (Table 1). For example, Williams et al. (2011) reported that genetic variation in RFI exists in dairy heifers and this could be an alternative to indirectly selecting dairy cows for improved feed efficiency, as it is easier to record feed intake in heifers (similar production and data collection systems as in beef cattle). Spurlock et al. (2012) estimated genetic parameters and made recommendations regarding traits related to energy balance, including DMI, bodyweight, BCS, energy-corrected milk production, and gross feed efficiency.

The h^2 estimates presented in Table 1 indicate that feed efficiency can be improved through genetic selection. The wide range of h^2 estimates reported in the literature are related to the different populations used in each study, as genetic parameters (such as h^2 estimates) are population-specific. Thus, this suggests the importance of (re-)estimating genetic parameters for each population. It is worth noting that selection for improved feed efficiency might also impact other economically important traits, due to genetic correlations between them. Examples of genetic correlations reported in the literature for different indicator traits of feed efficiency and some important economic traits in dairy cattle are summarized in Table 2. A detailed description of genetic correlations between feed efficiency indicators and other relevant traits can be found in Berry and Crowley (2013) and Manafiazar et al. (2016).

Functional candidate genes associated with feed efficiency

The development and availability of high-throughput “-omics” technologies (e.g. genomics, transcriptomics, proteomics, metabolomics, metagenomics) has enabled the

identification of numerous candidate regions associated with economically relevant traits (Cánovas et al., 2017). In this context, genome-wide association studies (**GWAS**) and transcriptomics using RNA-Sequencing (**RNA-Seq**) technology have contributed to the identification of functional candidate genes and genetic variants (e.g. SNPs; copy number of variations – **CNVs**; and, insertions and deletions – **INDELs**; Cánovas et al., 2010; Wickramasinghe et al., 2014). In beef cattle, GWAS and transcriptomics studies using RNA-Seq have enabled the identification of key regulators of biological processes and pathways linked to feed efficiency variability, including lipid and protein metabolism, ion transport, protein and amino acid glycosylation, and valine, leucine and isoleucine degradation (Rolf et al. 2012; Abo-Ismael et al. 2014; Olivieri et al. 2016; Duarte et al. 2019).

The integration of multiple “-omics” technologies through a systems biology approach is a powerful strategy for precisely identifying functional variants mapped in key regulator genes involved in the metabolic pathways affecting feed efficiency (Cánovas et al., 2017). Despite the low number of GWAS and RNA-Seq studies evaluating feed efficiency in dairy cattle, the combination of these results can be integrated to better understand the genetic architecture of feed efficiency in dairy cattle.

In this section we summarize the main studies, published up to date, that have applied GWAS and RNA-Seq to investigate the genetic mechanisms underlying feed efficiency. Table 3 presents the descriptive details of GWAS studies for feed efficiency in dairy cattle: breed(s), sample size, indicator trait, number of genetic markers, statistical method used, significance threshold, and number of significantly associated markers. Similarly, the descriptive details of transcriptomics studies using RNA-Seq comparing divergently-selected feed-efficient dairy cattle: breed(s), sample size, tissue analysed, indicator trait, statistical analysis, p-value threshold, and

number of differentially expressed genes are described in Table 4. Seven GWAS for feed efficiency indicator traits (e.g. RFI, DMI, milk energy, metabolic body weight) and three RNA-Seq studies comparing two divergent groups of feed-efficient animals were found in the literature (Tables 3 and 4). The majority of these studies focused on the Holstein breed, which is the most commonly raised breed for milk production around the world. However, Salleh et al. (2018) studied Jersey breed animals, in addition to a subset of Holstein cows. As there are breed differences for feed utilization performance (Berry and Crowley, 2013), one can assume that there would be differences at the gene expression level as well. However, the number of transcriptomics studies available using RNA-seq is still too limited to draw such conclusions.

The integration and evaluation of multiple levels of -omics data can provide a better understanding of the physiological processes underlying feed efficiency. In addition to transcriptomics, the combination of proteomic and metabolomic analysis is important to determine causal effect and provide functional validation. When considering the application of -omics in livestock studies, there is a lack of information on feed efficiency and, more specifically, in dairy cattle. Few studies have evaluated feed efficiency in dairy cattle using metabolomics and proteomics. Due to the lack of studies integrating multiple -omics technologies to study feed efficiency in dairy cattle, it is difficult to assess the consistency across studies. For instance, proteomics and metabolomics have only recently been performed to study feed efficiency in dairy cattle (Wang and Kadarmideen, 2019; Zhang et al., 2019). Metabolic profiling of blood plasma has been performed in Holstein and Jersey cattle, revealing multiple fatty acids with significantly different profiles between divergent feed efficiency groups and were functionally enriched for biological pathways associated with energy use and production (Wang and Kadarmideen, 2019). The integration of hepatic metabolomic and proteomic data of Holstein heifers divergent for feed

efficiency has revealed 29 metabolites and 60 proteins that were significantly different between low and high feed efficient heifers (Zhang et al., 2019). These studies provide useful biomarkers as indicators for feed efficiency in dairy cattle, however, integration and evaluation of multiple -omics technologies to study feed efficiency can improve the understanding of the whole biological system underlying feed efficiency through functional validation.

Regarding the study of feed efficiency in dairy cattle at the whole genome level using GWAS, a large number of regions were identified to be associated with feed efficiency traits, but the effect of each genomic region was small, indicating that feed efficiency is a polygenic trait. For instance, Hardie et al. (2017) reported that the 10 genomic windows explaining the majority of the genetic variance for RFI, accounted for only 5.38% and 4.80% of the genetic variance for RFI in first-parity and multiparous cows, respectively.

The transcriptomics analyses performed using RNA-Seq technology (Table 4) compared gene expression measured in the whole transcriptome between two divergently-selected group of animals based on feed efficiency. It is worth highlighting that Salleh et al. (2017, 2018) evaluated liver biopsies from the same set of animals (high-RFI = five Holstein and five Jersey cows; low-RFI = four Holstein and five Jersey cows), but used distinct statistical approaches. In all studies, RNA-seq was performed using liver tissue samples due to the key role of this organ in energy conversion and metabolic efficiency. In addition, Khansefid et al. (2017) also evaluated gene expression in white blood cells of divergently-selected cattle for RFI. The number of differentially expressed genes (**DEG**) varied substantially across studies. These results reinforce the polygenic nature of feed efficiency (as described by Salleh et al., 2018; Seymour et al., 2018). The use of methodologies such as the weighted gene co-expression network analysis (**WGCNA**) is a good alternative to identify hidden patterns of interactions between genes and consequently, contribute

to further understanding the biological processes associated with feed efficiency. This methodology is useful due to the fact that the individual identification of DEG can underestimate the complexity of the genetic architecture of quantitative traits, especially when the expression of genes acting in the biological processes tend to be correlated (Langfelder and Horvath, 2008). To date, no studies have exploited the identification of functional variants associated with feed efficiency traits using RNA-Seq data.

The level of overlapping and cross-validation among studies can greatly vary depending of the methodology used to perform the analyses. One of the main causes of the non-validation across studies is the lack of homogeneity in the population structure, phenotypes, statistical models, quality control thresholds, among others. For example, Fonseca et al. (2018) described a strong stratification for the list of positional and functional candidate genes as a function of the purpose of the breed (dairy or beef) and the phenotype evaluated when GWAS for male fertility traits were functionally integrated. Even though all the studies summarized in this current review were focused on feed efficiency, different phenotypes were used to access the feed efficiency in each study: RFI (using different models), daily-RFI and DMI. As each one of these phenotypes might be representing a different portion of the total feed efficiency of the animals, the candidate regions and genes can be expected to be different for each trait phenotype. Additionally,

the population structure and the statistical models applied in each study can substantially impact the detection power. Therefore, a very precise and careful approach must be performed in order to discuss and to point possible similarities and differences across the studies. This may be achieved by a proper meta-analysis, as more studies become available.

Despite the reduced number of GWAS and transcriptomics studies using RNA-Seq evaluating feed efficiency in dairy cattle, the findings currently reported are similar to those observed in beef cattle (Table 3). The similarities in results from beef and dairy cattle-based studies creates an opportunity to perform integrative analyses (e.g. meta-analyses, functional analyses) in order to reduce the number of false-positive associations, and consequently, fine map those variants with the strongest effects. Thereafter, the identification of functional candidate genes can be performed in a more efficient way. Once candidate genes are identified, the prospection of causal variants mapped within these genes can contribute to increasing the predictive ability of feed efficiency through the use of specific markers used in sophisticated genomic selection approaches (Hayes et al., 2013; Goddard et al., 2016; VanRaden et al., 2017).

The additional value of whole-genome sequence data

The use of genomic information derived from SNP chip arrays in genetic evaluation schemes is very efficient for multiple purposes (e.g. Georges et al., 2019). However, the inclusion of information from denser SNP arrays or whole-genome sequence data (**WGS**) is yet to be shown as advantageous. In this context, more recently, there has been an interest in selecting variants

based on WGS-based GWAS analyses and the incorporation of structural variants, especially CNVs, in GWAS and genomic predictions.

The identification of SNPs related to feed efficiency through WGS-based GWAS followed by functional analyses will enable the identification of variants with direct impact on feed efficiency. Therefore, the causal mutations can be included in the genomic predictions without the need to rely on linkage disequilibrium (**LD**; MacLeod et al., 2014). For instance, VanRaden et al. (2017a) reported an increase of 2.7% in the accuracy of genomic estimated breeding values (**GEBVs**) when performing WGS variant selection based on their estimated effect on a given trait.

Mielczarek et al. (2018) reported CNV variations within and between multiple European dairy cattle breeds. This variability in CNV might enable more accurate selection of animals with greater genetic merit for feed efficiency. Although few CNV studies have been performed in dairy cattle, those conducted reported identified multiple genomic regions associated with feed efficiency and other traits of interest. Based on 147 high-density (**HD**) Holstein genotypes, Hou et al. (2012) identified and partially validated CNVs that were only observed in high or low feed efficient animals. The authors also linked those CNVs with important metabolic pathways involved in feed utilization. However, the power of the study was small due to limited sample size. In addition, Zhou et al. (2018) identified 10 CNVs (based on the UMD3.1 reference assembly; Zimin et al., 2009) in Holstein cattle associated with RFI. One of these CNVs (BTA4:108,225,979-108,252,635 bp) was also associated with DMI. In addition, multiple regions were harboring olfactory receptor genes (e.g. *RXFP4*), which are likely indirectly related to feed efficiency through changes in feeding behavior (Soria-Gomez et al., 2014). For instance, the *RXFP4* gene is known to be related to appetite regulation and metabolism, providing a direct link to efficiency (Ang et al., 2017). Lastly, a region overlapping with a quantitative trait loci (**QTL**) associated with

average daily gain (**ADG**) on BTA7 (42,745,346-42,788,788) was also associated with RFI. The release of a better-quality reference genome assembly, i.e. ARS-UCD1.2 (Rosen et al., 2018), will enable the discovery of additional CNVs associated with feed efficiency. Furthermore, there are limitations on the number of individuals with phenotypic and WGS information. As more animals have phenotypes and WGS data become available on a larger number of individuals, more accurate results are expected to be obtained.

In addition to the individual genetic merit of dairy cattle, there are other factors that contribute to variability in feed efficiency. The next section will describe the role of the rumen microbiome on the efficiency of feed utilization in dairy cattle as well as its interaction with the genetic makeup of the individual host.

The role of rumen microbiome on feed efficiency

The rumen microbial community is a complex ecosystem composed mainly of bacteria, ciliate protozoa, fungi, and archaea, which interact with each other to digest fibrous feed (Williams and Coleman, 1997). Ruminants are dependent on the rumen microbial community to produce and serve as metabolic energy products to survive, and in return, the microbial community depends on the ruminant for a habitat to survive, resulting in a symbiotic host-microbial relationship. The function of the rumen to extract nutrients from feed and deliver metabolites to productive tissues, represents its large role in nutrient economy and whole-body metabolism (Baldwin and Connor, 2017). As feed efficiency is largely dependent on better partitioning of metabolic energy, the metabolic efficiency of the rumen microbiota is known to influence feed utilization, due to its large role in energy production and delivery to the host (Myer et al., 2015; Shabat et al., 2016).

The near-total exchange of rumen contents between two cows has revealed that ruminal pH and volatile fatty acid (VFA) concentration rapidly stabilizes within 24 hours after rumen content exchange. This implies that the rumen microbial community has the ability to adapt rapidly (Weimer et al., 2010), and the assembly of the microbial community could be partially determined by the host (Benson et al., 2010; Sasson et al., 2017; Difford et al., 2018; Wallace et al., 2019, Li et al., 2019). This suggests that ruminants may exhibit individual rumen microbial profiles, and that there could be a potential for genetic selection for a desirable rumen microbiome profile in combination with management of other environmental factors (e.g. diet). However, the understanding of the genetic basis underlying the interactions between the host's genetics and the rumen microbiome, along with its overall influence on feed efficiency is limited. This has led to recent studies using transcriptomics, meta-transcriptomic and metagenomics to investigate the role of the rumen microbiome, which is considered as "all the microbial genomes within the rumen microbial community".

Studying the rumen microbiome using "-omics" technologies

Previous studies have used metagenomic and meta-transcriptomic approaches to quantify microbial content/abundance and microbial gene expression, respectively, and its potential link to feed efficiency in cattle (Shabat et al., 2016; Li and Guan, 2017; Paz et al., 2018). Research investigating the rumen metagenome and its association with feed efficiency has revealed differential bacteria abundances across divergent rumen metabolic efficiencies by classifying specific bacteria using Operational Taxonomic Units (OTU; Paz et al., 2018), which measures various microbial species and their abundance. Specific OTU have been characterized in beef cattle across divergent feed efficiency groups and revealed that specific OTU abundance from

bacterial families, including *Prevotellaceae* and *Lachnospiraceae*, were associated with feed efficiency in beef steers (Paz et al., 2018). Feed efficiency and rumen microbiome have been previously associated (Hayes et al., 2013; Sasson et al., 2017; Paz et al., 2018), revealing that models used to explain feed efficiency traits (e.g. DMI; ADG; gain to feed ratio) explained up to 20% of the total variation in feed utilization when including OTU abundance parameters (Paz et al., 2018). This evidence suggests that microbial OTU abundance may serve as a predictor of feed efficiency (Paz et al., 2018).

The study of rumen meta-transcriptome has indicated that less efficient cattle exhibit more diverse microbial activities (Li and Guan, 2017). This supports the findings by Shabat et al. (2016), in which less feed efficient beef cattle exhibited higher richness of microbial gene content compared to more feed efficient beef cattle. These studies suggest that rumen microbiome content and function/activity may serve as a microbiome feature to genetically improve feed efficiency (Shabat et al., 2016; Li and Guan, 2017). To further improve the understanding of the associations between the rumen microbiome and host phenotypes, other “-omics” platforms should be considered, including meta-proteomics and metabolomics (Almeida et al., 2018; Hart et al., 2018).

Heritability estimates of rumen microbial features

The nature of the diverse community of the rumen microbiome has led to variation in characterizing and defining consistent rumen microbiome traits to estimate heritability and investigate correlations with production traits. The heritability of the rumen microbiome has primarily been estimated using taxonomic profiles, on an OTU abundance basis (Sasson et al., 2017; Difford et al., 2018; Wallace et al., 2019; Li et al., 2019). Additionally, more recent traits

used to estimate heritability of the rumen microbiome include microbial diversity indices and ratios between microbial groups (Li et al., 2019).

Using specific bacteria OTU abundance, a study on 78 Holstein-Friesian dairy cows estimated heritability of that trait at approximately 0.70 (Sasson et al., 2017). Furthermore, bacteria and archaea OTU abundance had heritability estimates ranged between 0.17 and 0.25, when the association between methane emissions (a trait correlated with efficiency of nutrient utilization) and rumen microbiome in lactating Holstein cows was analyzed (Difford et al., 2018). In Holstein and Nordic Red lactating dairy cows, Wallace et al. (2019) identified 39 heritable core (few specific targeted microorganisms) microbial OTUs, with heritability estimates ranging from 0.20 to 0.60.

Heritability estimates on rumen microbial traits have also been studied in beef cattle. For instance, the heritability of rumen bacterial diversity indices were estimated in Angus, Charolais, and Crossbreed, revealing heritability estimates of 0.23 (Shannon index) and 0.19 (Simpson index) (Li et al., 2019). In the same study, the heritability of bacterial or archaeal community component differences ranged from 0.15 to 0.25. Similarly, moderate heritability estimates were observed for total bacterial abundance (0.16), while heritability estimates for total archaeal abundance was lower (0.05). A wide range of heritability estimates have been reported for various microbial features, mainly due to differences across breeds, populations, analytical methods and diets (Sasson et al., 2017; Wallace et al., 2019; Li et al., 2019).

Host-microbiome genetic interactions and its influence on production traits in dairy cattle

Advances in transcriptomic, meta-transcriptomic (the measurement of host and microbial gene expression using RNA-Seq technology; Li and Guan, 2017) and metagenomic (amplicon-

sequencing to measure microbial content/abundance; Sasson, 2017; Wallace et al., 2019) sequencing approaches have led to opportunities to better understand rumen microbiome parameters and their relationship with host phenotypic expression. Li et al. (2019) reported 19 SNPs in the host genome associated with 14 rumen microbial taxa. Out of those 19 SNPs, five are located in known QTLs for cattle feed efficiency. Host-microbiome interactions has been widely studied in mice, flies and humans (Benson et al., 2010; Turpin et al., 2016; Fromont et al., 2019). However, to our best knowledge, Li et al. (2019) is the first report on the characterization of the link between the cattle genetic makeup and heritable microbial features. This is a research area in great expansion at the moment and therefore, major breakthroughs in this field are expected over the next few years.

Data collection and implementation of genomic evaluations

Genomic selection for improved feed efficiency

As previously outlined, the costs and feasibility of measuring individual feed intake (and related traits, such as bodyweight) in a large number of animals with pedigree information has limited the implementation of genetic selection for feed efficiency. Genomic selection has become widely available in the dairy cattle industry and enabled selection of breeding candidates based on their predicted genetic merit for feed efficiency. This is because animals from research herds can be used as a training population to estimate SNP effects, which are then used to predict GEBVs for selection candidates based on their own genotype (Veerkamp et al., 2015). In brief, genomic selection refers to the use of genome-wide genetic markers to predict breeding values of selection candidates (Meuwissen et al., 2001).

The accurate calculation of GEBVs depends on the estimation of SNP effects based on genomic and phenotypic datasets (i.e. training population). The size of the training population directly affects the GEBV accuracies (Goddard, 2009; Hayes and Goddard, 2008). However, the size of training population for feed efficiency in dairy cattle is still limited. Other factors that impact GEBV accuracy are: SNP panel density, trait heritability (Daetwyler et al., 2008; Goddard, 2009), the extent of the LD between SNP and QTL (Hayes et al., 2009; VanRaden et al., 2009), and the relationship between the training and validation or target populations (Habier et al., 2010; Pszczola et al., 2012).

As previously discussed, RFI is one of the most common indicator traits of feed efficiency in research settings. Genomic selection for RFI has been shown to be feasible, but the accuracies are still lower compared to other traits (Table 5; Calus et al., 2013; Gonzalez-Recio et al., 2014). Some studies have reported that a training population containing more than 30,000 individuals would be required to achieve satisfactory reliabilities for traits with heritability estimates of 0.2 such as RFI (Calus et al., 2013; Connor, 2015; Gonzalez-Recio et al., 2014). Despite the fact that GEBV accuracies for RFI in dairy cows are usually lower than the accuracies obtained for production traits, they are expected to increase as the training populations keep growing. For instance, Pryce et al. (2012) reported GEBV accuracies for RFI of 0.31 to 0.37, when using a HD SNP panel and independent cross-validation datasets from Australia and New Zealand, respectively.

Gonzalez-Recio et al. (2014) described the implementation of heifer feed efficiency in the Australian selection index, using genomic selection and its impact in the industry. In 2015, the same research group (Pryce et al., 2015) defined and described the implementation of genetic evaluation for “feed saved” as a new indicator of feed efficiency in dairy cows. Feed saved

combines RFI with mature bodyweight estimated using estimated breeding values for predicting maintenance costs, so that feed requirements are quantified in a single breeding value. Since April 2015, feed saved has been included as part of the Australian national selection index.

Negussie et al. (2019) used simulated data to estimate accuracies of genomic prediction for different DMI recording scenarios (once weekly, once monthly, every two, three and four months) using different sizes of training populations in dairy cattle in order to develop future innovative phenotyping strategies of recording DMI. The authors reported that the accuracy of genomic predictions associated with the five recording scenarios indicated that the use of a large training population and the adoption of a less-frequent DMI sampling scenario is an advantageous strategy when considering accuracy, logistic, and cost implications. GEBV accuracies for DMI and RFI that have been reported in the literature are summarized in Table 5. These results indicate that there is still room for improving the prediction of GEBVs.

Some alternatives have been investigated to increase the training population for feed efficiency, including the use of data from nutrition studies (Tempelman et al., 2015; Veerkamp et al., 2014) and combining data from different countries (Banos et al., 2012; Berry et al., 2014; de Haas et al., 2012; Pryce et al., 2012; Tempelman et al., 2015) or breeds (Khansefid et al., 2014). It is worth noting that in the last few years, a collaboration group named The global Dry Matter Initiative (**gDMI**) has been created to combine feed intake records, which included 10 research herds from nine countries (de Haas et al., 2015; Berry et al., 2014).

Data collection and international collaborations for data sharing

To genetically select animals for improved feed efficiency, at least, pedigree information and individual phenotypic records associated with feed intake and production traits are required.

The simplest way to record DMI is based on the amount of feed offered and refused by each cow per day, with the associated DM percentage (Seymour et al., 2019). Other important variables to be recorded for assessment of feed efficiency are milk production and composition, lactation stage, water intake, diet composition, bodyweight and BCS over the course of lactation, health/disease events, and reproductive performance traits. It is important to notice that even if not all these variables are used in the genetic/genomic evaluations, they might be useful in the future for research and also selection purposes. Furthermore, the costs to record these additional traits are low compared to the cost of individual feed intake recording (Veerkamp et al., 2015).

There are various automated systems available for feed intake recording, including Calan Broadbent (American Calan Inc. Northwood, NH), Gallagher Animal Management Systems (Hamilton, New Zealand), GrowSafe® Feed Intake System (GrowSafe Systems, Ltd., Airdrie, AB, Canada), and the RIC system (i.e. Insentec; Hokofarm Group B.V., Marknesse, the Netherlands). These systems are mostly based on radio-frequency identification to track and record individual feed intake as well as feeding behavior (e.g. number of visits per day, intake duration, time of intake). As discussed by Connor (2015), the use of these systems in dairy cattle has been limited to research herds or growing heifers. The use of automated feed monitoring systems in larger groups of lactating cows is hindered by the limited feeding capacity of the automated feed bunks, meaning that significantly fewer cows can be fed from a single bunk relative to growing cattle to accommodate substantially greater intakes of lactating cows (Connor, 2015).

It is well established that the success and long-term sustainability of any livestock breeding program is largely dependent on the amount and quality of pedigree, phenotypic and genotypic data available for genetic and genomic evaluations. As feed efficiency is difficult and expensive to measure, a global effort to enlarge training population for genomic evaluations is crucial and

has the potential to greatly benefit all groups involved in the project. In addition to gDMI, the Efficient Dairy Genome Project (**EDGP**, <http://genomedairy.ualberta.ca/>) is a large international research project led by Canadian institutions aiming to develop strategic research, tools, and the whole infrastructure to implement genetic and genomic evaluations for improved feed efficiency and reduced methane emissions in dairy cattle. In this regard, the EDGP database was developed in 2017 to allow data sharing among international collaborators from six countries (Australia, Canada, Denmark, Switzerland, United Kingdom and United States) to facilitate development of an international genetic evaluation for feed efficiency. This goal is likely possible due to the high level of relatedness of the Holstein population, the most common dairy breed with records for feed efficiency. Moreover, all collaborators are members of the International Committee for Animal Recording (**ICAR**, www.icar.org) providing standardized information on production records.

Incorporating feed efficiency into breeding programs

National organizations and private companies began implementing the selection of feed efficiency into their breeding programs in 2014 (Pryce et al., 2014). Each organization incorporated indirect measures of feed efficiency such as production levels, body weight (or predicted body weight), and conformation traits into their selection indexes (VanRaden et al., 2007; Veerkamp et al., 2013; Pryce et al., 2014, 2015). In Australia, animals that are one standard deviation above the mean for the Feed Saved trait, consume 65 kg less feed per year, while maintaining the same levels of production (Pryce et al., 2018). The USA developed a composite index into their national evaluation, which uses milk, fat, protein, and predicted body weight to predict feed efficiency (Holstein Association USA, 2017). New Zealand indirectly selects for feed efficiency by selecting on milk production, live weight and BCS simultaneously (Pryce et al.,

2014; DairyNZ, 2016). In the Netherlands, GEBVs for saved feed costs have been available since December 2017 (CRV, 2018). This value is expressed in euros per cow per lactation, where the feed for both production and maintenance are considered (de Jong et al., 2019). Furthermore, there are many other countries worldwide, including Canada, which are working towards including feed efficiency into their national breeding programs.

The inclusion of feed efficiency into breeding objectives is not exclusive to national organizations. Private breeding companies (e.g. CRV, Select Sires, GENEX, STGenetics and Viking Genetics) are also promoting GEBVs for more efficient cows through their own selection strategies. For instance, CRV generates the NVI, which is the total merit index used in the Netherlands and Flanders, which includes a saved-feed-cost trait. In other countries, such as the USA, CRV offers the Better Life Efficiency program, which identifies bulls that have a high lifetime production to lifetime feed intake ratio (CRV, 2019). Recently, Viking Genetics also released a saved feed index. They are working towards implementing an index based on two indicator traits: maintenance efficiency, which captures the energy requirements for maintenance, and metabolic efficiency, which reflects how efficiently the eaten feed is utilized (Viking Genetics, 2019). Bulls with a score of 5% for Better Life Efficiency have been reported to have daughters that can produce an additional 680 kg of milk with the same amount of feed, which would translate into an additional \$295 per cow per lifetime. Select Sires also developed a selection index, FeedPRO® that focuses on producing moderately sized cows while maintaining production levels (Select Sires Inc., 2019). Daughters of FeedPRO® bulls have been reported to produce on average 13 to 18 cents more per day (Select Sires Inc., 2019). The FeedPRO® index is also correlated at 0.90 to TPI, a total selection index in the USA. Production Efficiency (PREF\$), an index from GENEX, has also been reported to result in higher yielding cows with lower feed costs. This sub-

index makes up 47% of the ICC\$TM index, with emphasis on marginal feed costs, fat, protein and milk yield (Genex Cooperative, 2018). EcoFeedTM, a feed efficiency index developed by STgenetics, is designed to encompass environmental, metabolic and genomic factors affecting dairy cattle profitability from birth to culling (STgenetics, 2018). Daughters of bulls that are 5 points above the average are reported to consume 0.45 kg less feed per day, while maintaining similar production levels (STgenetics, 2018). While many companies and national breeding organizations are moving towards the inclusion of feed efficiency into their breeding programs, there is no consensus on the optimal way to include these traits. It is worth mentioning that the descriptions of the selection indexes mentioned above were provided by the companies and some details might have been omitted by them. Further research is required to compare different approaches and define an optimal strategy.

Conclusions

Feed efficiency, assessed based on different indicators, is a heritable trait and can be improved through genetic and genomic selection. The breeding goal needs to be refined and indicator traits that can be easily measured at low cost should be identified. Feed efficiency is a polygenic trait influenced by many genetic variants, regulator genes and structural variations. With the important role of the rumen microbiota on feed efficiency and evidence of host genetic influence on the rumen microbiome profile, further evaluation of rumen microbial features may lead to its prospective use as an indicator trait, or use in future genomic selection models. The accuracy of genomic predictions for feed efficiency are still low, but are expected to increase as training populations are enlarged and additional functional information could be included from transcriptomics and other –“omics” technologies. In this regard, various groups around the world

are collaboratively working to refine the methods used in the evaluations as well as enlarging the datasets used for genomic evaluations.

Acknowledgements

We gratefully acknowledge funding by the Efficient Dairy Genome Project, funded by Genome Canada (Ottawa, Canada), Genome Alberta (Calgary, Canada), Ontario Genomics (Toronto, Canada), Alberta Agriculture and Forestry (Edmonton, Canada), Ontario Ministry of Research and Innovation (Toronto, Canada), Ontario Ministry of Agriculture, Food and Rural Affairs (Guelph, Canada), Canadian Dairy Network (Guelph, Canada), GrowSafe Systems (Airdrie, Canada), Alberta Milk (Edmonton, Canada), Victoria Agriculture (Melbourne, Australia), Scotland's Rural College (Edinburgh, UK), USDA Agricultural Research Service (Beltsville, United States), Qualitas AG (Zug, Switzerland), Aarhus University (Aarhus, Denmark). C. Baes and F. Schenkel acknowledge financial contributions from the Dairy Research Cluster 3 (Dairy Farmers of Canada and Agriculture and Agri-Food Canada) under the Canadian Agricultural Partnership AgriScience Program.

Literature cited

- Abo-Ismael, M.K., Vander Voort, G., Squires, J.J., Swanson, K.C., Mandell, I.B., Liao, X., Stothard, P., Moore, S., Plastow, G., and Miller, S.P. 2014. Single nucleotide polymorphisms for feed efficiency and performance in crossbred beef cattle. *BMC Genet.* **15**:14.
- Allen, M. S. 2000. Effects of diet on short-term regulation of feed intake by lactating dairy cattle. *J. Dairy Sci.* **83**(7):1598-1624.

- Allen, M.S., Bradford, B.J. and Oba, M. 2009. Board Invited Review: The hepatic oxidation theory of the control of feed intake and its application to ruminants. *J. Animal Sci.* **87(10)**:3317-3334.
- Almeida, R.T.R., Prado, R.M., Porto, C., Santos, G.T., Huws, S.A., and Pilau, E.J. 2018. Exploring the rumen fluid metabolome using liquid chromatography-high-resolution mass spectrometry and Molecular Networking. *Nature: Scient. Rep.* **8**:17971.
- Ang, S.Y., D.S. Hutchinson, B.A. Evans, M.A. Hossain, N. Patil, R.A.D. Bathgate, M. Kocan, and R.J. Summers. 2017. The actions of relaxin family peptides on signal transduction pathways activated by the relaxin family peptide receptor *RXFP4*. *Naunyn. Schmiedebergs. Arch. Pharmacol.* **390**:105–111.
- Baldwin, R.L., Smith, N.E., Taylor, J. and Sharp, M. 1980. Manipulating metabolic parameters to improve growth rate and milk secretion. *J. Animal Sci.* **51(6)**:1416-1428.
- Baldwin, R.L. and Connor, E.E. 2017. Rumen function and development. *Vet. Clin. North America: Food Anim. Pract.* **33**:427-439.
- Banos, G., and Coffey, M.P. 2012. Technical note: prediction of live weight from linear conformation traits in dairy cattle. *J. Dairy Sci.* **95**:2170–2175.
- Basarab, J.A., Beauchemin, K.A., Baron, V.S., Ominski, K.H., Guan, L.L., Miller, S.P., and Crowley, J.J. 2013. Reducing GHG emissions through genetic improvement for feed efficiency: effects on economically important traits and enteric methane production. *Animal.* **7(Suppl 2)**:303–315.
- Benson, A.K., Kelly, S.A., Legge, R., Ma, F., Low, S. J., Kim, J., Zhang, M., Oh, P. L., Nehrenberg, D., Hua, K., Kachman, S. D., Moriyama, E. N., Walter, J., Peterson, D. A., and Pomp, D. 2010. Individuality in gut microbiota composition is a complex polygenic

trait shaped by multiple environmental and host genetic factors. *Proc. Natl. Acad. Sci. USA*. **107**:18933-18938.

Berry, D.P., and Crowley, J.J. 2013. Cell biology symposium: Genetics of feed efficiency in dairy and beef cattle. *J. Anim. Sci.* **91(4)**:1594-613.

Berry, D.P., Coffey, M.P., Pryce, J.E., De Haas, Y., Lovendahl, P., Krattenmacher, N., Crowley, J.J., Wang, Z., Spurlock, V., Weigel, K., Macdonald, K., and Veerkamp, R.F. 2014. International genetic evaluations for feed intake in dairy cattle through the collation of data from multiple sources. *J. Dairy Sci.*, **97(6)**:3894-3905.

Bilal, G., Cue, R. I., and Hayes., J. F. 2016. Genetic and phenotypic associations of type traits and body condition score with dry matter intake, milk yield, and number of breedings in first lactation Canadian Holstein cows. *Can. J. Anim. Sci.* **96(3)**: 434-447.

Bottje, W. G., and Carstens, G.E. 2019. BOARD INVITED REVIEW: Oxidative stress and efficiency: the tightrope act of mitochondria in health and disease. *J. Animal Sci.* **97(8)**:3169-3179.

Byskov, M.V., Fogh, A., and Løvendahl, P. 2017. Genetic parameters of rumination time and feed efficiency traits in primiparous Holstein cows under research and commercial conditions. *J. Dairy Sci.* **100(12)**:9635-9642.

Calus, M.P., De Haas, Y., Pszczola, M., and Veerkamp, R. 2013. Predicted accuracy of and response to genomic selection for new traits in dairy cattle. *Animal*, **7(2)**:183-191.

Cánovas, A., Rincon, G., Islas-Trejo, A., Wickramasinghe, S., Medrano, J.F. 2010. SNP discovery in the bovine milk transcriptome using RNA-Seq technology. *Mamm. Gen.* **21(11-12)**:592-598.

- Canovas, A., Casellas, J., Thomas, M., Medrano, J.F. 2017. Applying new genomic technologies to accelerate genetic improvement in beef and dairy cattle. *Proc. Assoc. Advmt. Anim. Breed. Genet.* **22**:377-383
- Capuco, A.V., Annen, E., Fitzgerald, A.C., Ellis, S.E., and Collier, R.J. 2006. Mammary cell turnover: relevance to lactation persistency and dry period management. *Ruminant physiology: Digestion, metabolism and impact of nutrition on gene expression, immunology and stress*, Wageningen Academic Publishers, Wageningen, the Netherlands **(2006)**:363-388.
- Coleman, J., Berry, D.P., Pierce, K.M., Brennan, A., and Horan, B. 2010. Dry matter intake and feed efficiency profiles of 3 genotypes of Holstein-Friesian within pasture-based systems of milk production. *J. Dairy Sci.* **93**:4318–4331.
- Connor, E.E. 2015. Invited review: Improving feed efficiency in dairy production: Challenges and possibilities. *Animal*. **9(3)**:395-408.
- Connor, E. E., J. L. Hutchison, C. P. Van Tassell, and J. B. Cole. 2019. Defining the optimal period length and stage of growth or lactation to estimate residual feed intake in dairy cows. *J. Dairy Sci.* 102(7):6131-6143.
- CRV. 2018. Save on Feed not used for milk production. Access date: September 27, 2019. Available at: <https://www.crv4all-international.com/wp-content/uploads/2018/03/Art.-Saved-Feed-VT-ENG.pdf>.
- CRV. 2019. Better Life Efficiency. Access date: September 26, 2019. Available at: <https://www.crv4all.us/service/better-life-efficiency/>.
- Daetwyler, H.D., Villanueva, B., and Woolliams, J.A. 2008. Accuracy of predicting the genetic risk of disease using a genome-wide approach. *PLoS One*, **3(10)**:e3395.

- DairyNZ. 2016. Breeding values. Access date: September 25, 2019. Available at: <https://www.dairynz.co.nz/animal/animal-evaluation/interpreting-the-info/breeding-values/>
- De Haas, Y., Calus, M.P.L., Veerkamp, R.F., Wall, E., Coffey, M.P., Daetwyler, H.D., Hayes, B.J., and Pryce, J.E. 2012. Improved accuracy of genomic prediction for dry matter intake of dairy cattle from combined European and Australian data sets. *J. Dairy Sci.*, **95**(10): 6103-6112.
- de Jong, G., Schopen, G.C.B., Bouwmeester-Vosman, J.J., van der Linde, C., de Haas, Y., and Veerkamp, R.F. 2019. Feed intake genetic evaluation: progress and an index for saved feed costs. *Interbull Bulletin No. 55*, Cincinatti, OH, USA.
- Dickerson, G.E. 1978. Animal size and efficiency: Basic concepts. *Animal Prod.* **27**:367-379.
- Difford, G.F., Plichta, R.P., Løvendahl, P., Lassen, J., Noel, S.J., Højberg, O., Wright, Zhu, Z., Kristensen, L., Nielsen, H.B., Guldbrandtsen, B. and Sahana, G. 2018. Host genetics and the rumen microbiome jointly associate with methane emissions in dairy cows. *PLoS Genetics*. **14**:10.
- Difford, G.F., Løvendahl, P., Veerkamp, R.F., Bovenhuis, H., Visker, M.H.P.W., Lassen, J. and de Haas, Y., 2020. Can greenhouse gases in breath be used to genetically improve feed efficiency of dairy cows? *J. Dairy Sci.* **103**(3):2442-2459
- Dórea, J.R., Rosa, G.J.M., Weld, K.A., and Armentano, L.E. 2018. Mining data from milk infrared spectroscopy in residual energy intake among lactating Holstein cows. *J. Dairy Sci.* **101**:5878-5889.
- Duarte, D., Newbold, C., Detmann, E., Silva, F., Freitas, P., Veroneze, R., and Duarte, M. 2019. Genome-wide association studies pathway-based meta-analysis for residual feed intake in

- beef cattle. *Anim. Genet.* **50**: 150–153.
- Erbe, M., Hayes, B.J., Matukumalli, L.K., Goswami, S., Bowman, P.J., Reich, C.M., Mason, B.A., and Goddard, M.E. 2012. Improving accuracy of genomic predictions within and between dairy cattle breeds with imputed high-density single nucleotide polymorphism panels. *J. Dairy Sci.*, **95**(7):4114-4129.
- Fischer, A., Friggens, N.C., Berry, D.P., and Faverdin, P. 2018. Isolating the cow-specific part of residual energy intake in lactating dairy cows using random regressions. *Animal* **12**(7):1396-1404.
- Flay, H. E., B. Kuhn-Sherlock, K. A. Macdonald, M. Camara, N. Lopez-Villalobos, D. J. Donaghy, and J. R. Roche. 2019. Hot topic: Selecting cattle for low residual feed intake did not affect daily methane production but increased methane yield. *J. Dairy Sci.* 102(3):2708-2713.
- Fonseca, P.A.D.S., dos Santos, F.C., Lam, S., Suárez-Vega, A., Miglior, F., Schenkel, F.S., Diniz, L.D.A.F., Id-Lahoucine, S., Carvalho, M.R.S. and Cánovas, A., 2018. Genetic mechanisms underlying spermatogenic and testicular traits within and among cattle breeds: systematic review and prioritization of GWAS results. *J. Animal Sci.* 96(12), pp.4978-4999.
- Forbes, J. M., 2000. Physiological and metabolic aspects of feed intake control. In: *Farm Animal Metabolism and Nutrition*. 319-333.
- Forbes, J. M., 2007. A personal view of how ruminant animals control their intake and choice of food: minimal total discomfort. *Nutr. Res. Rev.* **20**(2):132-146.
- Fromont, C., Adair, K.L., and Douglas, A.E. 2019. Correlation and causation between the microbiome, *Wolbachia* and host functional traits in natural populations of drosophilid flies. *Mol. Ecol.* **28**:1826–1841.
- Genex Cooperative. 2018. ICCSTM Index for Holsteins. Accessed September 24, 2019. Available

at:<https://www.genex.coop/dairy-cattle-artificial-insemination-services/ideal-commercial-cow-index/icc-sub-indexes>

- Georges, M., Charlier, C., and Hayes, B. 2019. Harnessing genomic information for livestock improvement. *Nat. Rev. Genet.* **20**:135–156.
- Goddard, M. 2009. Genomic selection: Prediction of accuracy and maximization of long-term response. *Genetica*, **136**(2):245-257.
- Goddard, M.E., Kemper, K.E., MacLeod, I.M., Chamberlain, A.J., and Hayes, B.J. 2016. Genetics of complex traits: Prediction of phenotype, identification of causal polymorphisms and genetic architecture. *Proc. R. Soc. B Biol. Sci.* **283**:1835.
- Gonzalez-Recio, O., Coffey, M., and Pryce, J. 2014. On the value of the phenotypes in the genomic era. *J. Dairy Sci.*, **97**(12):7905-7915.
- Habier, D., Tetens, J., Seefried, F-R., Lichtner, P. and Thaller, G. 2010. The impact of genetic relationship information on genomic breeding values in German Holstein cattle. *Genet. Sel. Evol.*, **42**:5.
- Hardie, L.C., VandeHaar, M.J., Tempelman, R.J., Weigel, K.A., Armentano, L.E., Wiggans, G.R., Veerkamp, R.F., de Haas, Y., Coffey, M.P., Connor, E.E., Hanigan, M.D., Staples, C., Wang, Z., Dekkers, J.C.M., and Spurlock, D.M. 2017. The genetic and biological basis of feed efficiency in mid-lactation Holstein dairy cows. *J. Dairy Sci.* **100**(11):9061–9075
- Hart, E. H., Creevey, C. J., Hitch, T., and Kingston-Smith, A. H. 2018. Meta-proteomics of rumen microbiota indicates niche compartmentalisation and functional dominance in a limited number of metabolic pathways between abundant bacteria. *Nature Scient. Reports.* **8**:10504.

- Hayes, B.J., and Goddard, M.E. 2008. Technical note: Prediction of breeding values using marker derived relationship matrices. *J. Anim. Sci.*, **86(9)**:2089-2092.
- Hayes, B.J., Bowman, P.J., Chamberlain, A.J., Verbyla, K., and Goddard, M.E. 2009. Accuracy of genomic breeding values in multi-breed dairy cattle populations. *Genet. Sel. Evol.*, **41**:51.
- Hayes, B.J., Lewin, H.A., and Goddard, M.E. 2013. The future of livestock breeding: Genomic selection for efficiency, reduced emissions intensity, and adaptation. *Trends Genet.*, **29(2)**:206-214
- Herd, R.M., and Arthur, P.F. 2008. Physiological basis for residual feed intake. *J Anim Sci.* **87(14 Suppl)**:E64-71.
- Herd, R. M., Oddy, V.H., and Richardson, E.C. 2004. Biological basis for variation in residual feed intake in beef cattle: Review of potential mechanisms. *Aust. J. Exp. Agric.* doi:10.1071/ea02220.
- Holstein Association USA. 2017. TPI formula. Accessed September 26, 2019. Available at: www.holsteinusa.com/genetic_evaluations/ss_tpi_formula.html
- Hou, Y., Bickhart, D.M., Chung, H., Hutchison, J.L., Norman, H.D., Connor, E.E., and Liu, G.E. 2012. Analysis of copy number variations in Holstein cows identify potential mechanisms contributing to differences in residual feed intake. *Funct. Integr. Genom.* **12**:717–723.
- Hurley, A.M.M., López-Villalobos, N., McParland, S., Kennedy, E., Lewis, E., O'Donovan, M., Burke, J.L.L., and Berry, D.P.P. 2016. Inter-relationships among alternative definitions of feed efficiency in grazing lactating dairy cows. *J. Dairy Sci.* **99(1)**:468-79.
- Jensen, J., Hohenboken, W. D., Madsen, P., and Andersen, B. B. 1995. Sire× nutrition interactions and genetic parameters for energy intake, production and efficiency of nutrient utilization

- in young bulls, heifers and lactating cows. *Acta Agriculturae Scandinavica A-Animal Sciences* **45**(2):81-91.
- Kelly, A.K., McGee, M., Crews, D.H., Fahey, A.G., Wylie, A.R., and Kenny, D.A. 2010. Effect of divergence in residual feed intake on feeding behavior, blood metabolic variables, and body composition traits in growing beef heifers. *J. Anim. Sci.* **88**(1):109-23.
- Khansefid, M., Millen, C.A., Chen, Y., Pryce, J.E., Chamberlain, A.J., Vander Jagt, C.J., Gondro, C., and Goddard, M.E. 2017. Gene expression analysis of blood, liver, and muscle in cattle divergently selected for high and low residual feed intake. *J. Anim. Sci.* doi:10.2527/jas2016.1320.
- Koch, R.M., Gregory, K.E., Chambers, D., and Swiger, L.A. 1963. Efficiency of feed use in beef cattle. *J. Anim. Sci.* **20**:1-13.
- Köck, A., Ledinek, M., Gruber, L., Steininger, F., Fuerst-Waltl, B., and Egger-Danner, C. 2018. Genetic analysis of efficiency traits in Austrian dairy cattle and their relationships with body condition score and lameness. *J. Dairy Sci.* **101**(1): 445-455.
- Korver, S., Van Eekelen, E. A. M., Vos, H., Nieuwhof, G. J., and Van Arendonk, J. A. M. 1991. Genetic parameters for feed intake and feed efficiency in growing dairy heifers. *Livest. Prod. Sci.* **29**(1):49-59.
- Langfelder, P., and Horvath, S. 2008. WGCNA: An R package for weighted correlation network analysis. *BMC Bioinformatics*. doi:10.1186/1471-2105-9-559.
- Li, F., Zhou, M., Ominski, K., and Guan, L.L. 2016. Does the rumen microbiome play a role in feed efficiency of beef cattle? *J. Anim. Sci.* **94**:44–48.
- Li, B., Fang, L., Null, D.A., Hutchison, J., Connor, E., VanRaden, P., VandeHaar, M., Tempelman, R., Weigel, K., and Cole, J. 2019. High-density genome-wide association study for residual

- feed intake in Holstein dairy cattle. *J. Dairy Sci.* doi.org/10.3168/jds.2019-16645
- Li, F., and Guan, L.L. 2017. Metatranscriptomic profiling reveals linkages between the active rumen microbiome and feed efficiency in beef cattle. *Appl. Envir. Microb.* **83**:e00061-17.
- Li, F., Li, C., Chen, Y., Liu, J., Zhang, C., Irving, B., Fitzsimmons, C., PLastow, G., and Guan, L.L. 2019. Host genetics influence the rumen microbiota and heritable rumen microbial features associate with feed efficiency in cattle. *Microbiome.* **7**(1).
- Lin, Z., Macleod, I. and Pryce, J.E., 2013. Estimation of genetic parameters for residual feed intake and feeding behavior traits in dairy heifers. *J. Dairy Sci.* **96**(4):2654-2656.
- Liinamo, A.E., Mäntysaari P., Lidauer M.H., and Mäntysaari E.A. 2015. Genetic parameters for residual energy intake and energy conversion efficiency in Nordic Red dairy cattle. *Acta. Agriculturae Scandinavica, Section A.* **65**(2): 63-72.
- Lu, Y., Vandehaar, M.J., Spurlock, D.M., Weigel, K.A., Armentano, L.E., Connor, E.E., Coffey, M., Veerkamp, R.F., de Haas, Y., Staples, C.R., Wang, Z., Hanigan, M.D., and Tempelman, R.J. 2018. Genome-wide association analyses based on a multiple-trait approach for modeling feed efficiency. *J. Dairy Sci.* **101**(4):3140-3154
- Lu, Y., Vandehaar, M.J., Spurlock, D.M., Weigel, K.A., Armentano, L.E., Staples, C.R., Connor, E.E., Wang, Z., Bello, N.M., and Tempelman, R.J. 2015. An alternative approach to modeling genetic merit of feed efficiency in dairy cattle. *J. Dairy Sci.* **98**(9):6535-51.
- MacLeod, I.M., Hayes, B.J., and Goddard, M.E. 2014. The effects of demography and long-term selection on the accuracy of genomic prediction with sequence data. *Genetics.* **198**:1671–1684.
- Manafiazar, G., Goonewardene, L., Miglior, F., Crews, D.H., Basarab, J.A., Okine, E. and Wang, Z., 2016. Genetic and phenotypic correlations among feed efficiency, production and

- selected conformation traits in dairy cows. *Animal*, **10**(3):381-389.
- Mantysaari, P., Liinamo, A.-E., Mantysaari, E.A. 2012. Energy efficiency and its relationship with milk, body, and intake traits and energy status among primiparous Nordic Red dairy cattle. *J. Dairy Sci.*, **95**(6):3200-3211.
- McParland, S., Banos, G., McCarthy, B., Lewis, E., Coffey, M.P., O'Neil, C., O'Donovan, M., Wall, E., and Berry, D.P. 2012. Validation of mid-infrared spectrometry in milk for predicting body energy status in Hostein-Fresian cows. *J. Dairy Sci.* **95**:7225-7235.
- Meuwissen, T.H.E., Hayes, B.J., and Goddard, M.E. 2001. Prediction of total genetic value using genome-wide dense marker maps. *Genetics*, **157**(4):1819-1829.
- Mielczarek, M., Fraszczak, M., Nicolazzi, E., Williams, J.L., and Szyda, J. 2018. Landscape of copy number variations in *Bos taurus*: Individual - and inter-breed variability. *BMC Genom.* **19**:410.
- Miglior, F., Fleming, A., Malchiodi, F., Brito, L.F., Martin, P., and Baes, C.F. 2017. A 100-Year Review: Identification and genetic selection of economically important traits in dairy cattle. *J. Dairy Sci.* 100.
- Montanholi, Y.R., Swanson, K.C., Palme, R., Schenkel, F.S., McBride, B.W., Lu, D., and Miller, S.P. 2010. Assessing feed efficiency in beef steers through feeding behavior, infrared thermography and glucocorticoids. *Animal*. **4**(5):692-701
- Moore, K.L., Johnston, D.J., Graser, H.U., and Herd, R. 2005. Genetic and phenotypic relationships between insulin-like growth factor-I (IGF-I) and net feed intake, fat, and growth traits in Angus beef cattle. *Aust. J. Agric. Res.* **56**(3):211-218.
- Myer, P.R. 2019. Bovine genome-microbiome interactions: Metagenomic frontier for the selection of efficient productivity in cattle systems. *Systems* **4**:e00103-19.

- Myer, P.R., Smith, T.P., Wells, J.E., Kuehn, L.A. and Freetly H.C.I. 2015. Rumen microbiome from steers differing in feed efficiency. *PLoS ONE* **10**:e0129174.
- Negussie, E., Mehtiö, T., Mäntysaari, P., Løvendahl, P., Mäntysaari, E.A., and Lidauer, M.H. 2019. Reliability of breeding values for feed intake and feed efficiency traits in dairy cattle: When dry matter intake recordings are sparse under different scenarios. *J. Dairy Sci.*, **102(8)**:7248-7262.
- O'Donovan, M., Berry, D.P., Kennedy, E., Moore, S.G., Lewis, E., Pryce, J.E., McCarthy, B., McParland, S., and Butler, S.T. 2014. Mid-infrared spectrometry of milk as a predictor of energy intake and efficiency in lactating dairy cows. *J. Dairy Sci.* **97(9)**:5863-71.
- Olivieri, B.F., Mercadante, M.E.Z., Cyrillo, J.N.D.S.G., Branco, R.H., Bonilha, S.F.M., De Albuquerque, L.G., De Oliveira Silva, R.M., and Baldi, F. 2016. Genomic regions associated with feed efficiency indicator traits in an experimental nellore cattle population. *PLoS One*. **11(10)**:e0164390
- Ondarza, M.B., and Tricarico, J.M. 2017. Review: Advantages and limitations of dairy efficiency measures and the effects of nutrition and feeding management interventions. *Prof. Anim. Sci.* **33**:393–400.
- Patience, J.F., Rossoni-Serão, M.C., and Gutiérrez, N.A. 2015. A review of feed efficiency in swine : biology and application. *J. Anim. Sci. Biotechnol.* **1**:9.
- Paz, H.A., Hales, K.E., Wells, J.E., Kuehn, L.A., Freetly, H.C., Berry, E.D., Flythe, M.D., Spangler, M.L. and Fernando, S.C. 2018. Rumen bacterial community structure impacts feed efficiency in beef cattle. *J. Animal Sci.* **96**:1045-1058.
- Pryce, J.E., Arias, J., Bowman, P.J., Davis, S.R., Macdonald, K.A., Waghorn, G.C., Wales, W.J., Williams, Y.J., Spelman, R.J., and Hayes, B.J. 2012. Accuracy of genomic predictions of

- residual feed intake and 250-day bodyweight in growing heifers using 625,000 SNP markers. *J. Dairy Sci.*, **95**(4):2108-2119.
- Pryce, J.E., Wales, W.J., De Haas, Y., Veerkamp, R.F., and Hayes, B.J. 2014. Genomic selection for feed efficiency in dairy cattle. *Animal*. **8**(1):1-10.
- Pryce, J.E., Gonzalez-Recio, O., Nieuwhof, G., Wales, W.J., Coffey, M.P., Hayes, B.J., and Goddard, M.E. 2015. Hot topic: Definition and implementation of a breeding value for feed efficiency in dairy cows. *J. Dairy Sci.* **98**:7340–7350.
- Pryce, J.E., Nguyen, T.T.T., Axford, M., Nieuwhof, G., and Shaffer, M. 2018. Symposium review: Building a better cow - The Australian experience and future perspectives. *J. Dairy Sci.* **101**:3702-3713.
- Pszczola, M., Strabel, T., van Arendonk, J.A.M., and Calus, M.P.L. 2012. The impact of genotyping different groups of animals on accuracy when moving from traditional to genomic selection. *J. Dairy Sci.*, **95**(9):5412-5421.
- Rolf, M.M., Taylor, J.F., Schnabel, R.D., McKay, S.D., McClure, M.C., Northcutt, S.L., Kerley, M.S., and Weaber, R.L. 2012. Genome-wide association analysis for feed efficiency in Angus cattle. *Anim. Genet.* 43(4): 367–374.
- Rosen, B., Bickhart, D., Schnabel, R., Koren, S., Elsik, C., Zimin, A., Dreischer, C., Schultheiss, S., Hall, R., Schroeder, S., Van Tassell, C., Smith, T., and Medrano, J. 2018. Modernizing the Bovine Reference Genome Assembly. In: *Proc. World Congr. Genet. Appl. to Livest. Prod.* 802, Auckland, New Zealand.
- Ross, E.M., Moate, P.J., Bath, C.R., Davidson, S.E., Sawbridge, T.I., Guthridge, K.M., Cocks, B.G., and Hayes, B.J. 2012. High throughput whole rumen metagenome profiling using untargeted massively parallel sequencing. *BMC Genet.* **13**:53.

- Salleh, M.S., Mazzoni, G., Höglund, J.K., Olijhoek, D.W., Lund, P., Løvendahl, P., and Kadarmideen, H.N. 2017. RNA-Seq transcriptomics and pathway analyses reveal potential regulatory genes and molecular mechanisms in high- and low-residual feed intake in Nordic dairy cattle. *BMC Genom.* **18**:258.
- Salleh, S.M., Mazzoni, G., Løvendahl, P., and Kadarmideen, H.N. 2018. Gene co-expression networks from RNA sequencing of dairy cattle identifies genes and pathways affecting feed efficiency. *BMC Bioinf.* **19**:513.
- Santschi, D.E., Lefebvre, D.M., Cue, R.I., Girard, C.L., and Pellerin, D. 2011a. Complete-lactation milk and component yields following a short (35-d) or a conventional (60-d) dry period management strategy in commercial Holstein herds. *J. Dairy Sci.* **94**(5):2302-2311.
- Santschi, D.E., Lefebvre, D.M., Cue, R.I., Girard, C.L., and Pellerin, D. 2011b. Economic effect of short (35-d) compared with conventional (60-d) dry period management in commercial Canadian Holstein herds. *J. Dairy Sci.* **94**(9):4734-4743.
- Sasson, G., Ben-Shabat, S.K., Seroussi, E., Doron-Faigenboim, A., Shterzer, N., Yaacoby, S., Miller, M. E. B., White, B. A., Halperin, E., and Mizrahi, I. 2017. Heritable bovine rumen bacteria are phylogenetically related and correlated with the cow's capacity to harvest energy from its feed. *ASM.* **8**:4(e00703-17).
- Select Sires Inc. 2019. FeedPRO® Sires. Accessed September 25, 2019. Available at: www.selectsires.com/designations/feedpro.html?version=20180803.
- Seymour, D.J., Cánovas, A., Baes, C.F., Chud, T.C.S., Osborne, V.R., Cant, J.P., Brito, L.F., Gredler-Grandl, B., Finocchiaro, R., Veerkamp, R.F., de Haas, Y., and Miglior, F. 2019. Invited review: Determination of large-scale individual dry matter intake phenotype in dairy cattle. *J. Dairy Sci.* **102**(9):7655–7663.

- Seymour, D., Miglior, F., Fonseca, P., Lam, S., Brito, L., Suárez-Vega, A., Cánovas, A. 2018. A comparison of weighted gene co-expression networks in high-and low-feed efficiency dairy cattle. *J. Animal Sci.* **96(suppl_3)**:146-147.
- Shabat, S.K., Sasson, G., Doron-Faigenboim, A., Durman, T., Yaacoby, S., Berg Miller, M. E., White, B. A., Shterzer, N. and Mizrahi, I. 2016. Specific microbiome-dependent mechanisms underlie the energy harvest efficiency of ruminants. *ISME J.* **10**:2958–2972.
- Shonka, B. N., Tao, S., Dahl, G. E., and Spurlock, D. M. 2015. Genetic regulation of prepartum dry matter intake in Holstein cows. *J. Dairy Sci.* **98**(11):8195-8200.
- Soria-Gomez, E., Bellocchio, L., and Marsicano, G. 2014. New insights on food intake control by olfactory processes: The emerging role of the endocannabinoid system. *Mol. Cell. Endocrinol.* **397**:59–66.
- Svendsen, M., Skipenes, P., and Mao, I. L. 1993. Genetic parameters in the feed conversion complex of primiparous cows in the first two trimesters. *J. Animal Sci.* **71**(7): 1721-1729.
- STgenetics. 2018. EcoFeed™ by STgenetics®. Accessed September 28, 2019. stgen.com/article/article.aspx?code=4247&language=English.
- Tempelman, R.J., Spurlock, D.M., Coffey, M., Veerkamp, R.F., Armentano, L.E., Weigel, K.A., De Haas, Y., Staples, C.R., Connor, E.E., Lu, Y. and VandeHaar, M.J., 2015. Heterogeneity in genetic and nongenetic variation and energy sink relationships for residual feed intake across research stations and countries. *J. Dairy Sci.*, **98**(3):2013-2026.
- Tetens, J., Thaller, G., and Krattenmacher, N. 2014. Genetic and genomic dissection of dry matter intake at different lactation stages in primiparous Holstein cows. *J. Dairy Sci.* **97**(1):520-31.

- Turpin, W., Espin-Garcia, O., Xu, W., Silverberg, M. S., Kevans, D., Smith, M. I., Guttman, D. S., Griffiths, A., Panaccione, R., Otley, A., Xu, L., Shestopaloff, K., Moreno-Hagelsieb, G. 2016. Association of host genome with intestinal microbial composition in a large healthy cohort. *Nature Genet.* **48**:1413–7.
- Vallimont, J. E., Dechow, C. D., Daubert, J. M., Dekleva, M. W., Blum, J. W., Barlieb, C. M., Liu, W., Varga, G. A., Heinrichs, A. J., and Baumrucker, C. R. 2010. Genetic parameters of feed intake, production, body weight, body condition score, and selected type traits of Holstein cows in commercial tie-stall barns. *J. Dairy Sci.* **93**(10): 4892-4901.
- Vallimont, J. E., Dechow, C. D., Daubert, J. M., Dekleva, M. W., Blum, J. W., Barlieb, C. M., Liu, W., Varga, G. A., Heinrichs, A. J., and Baumrucker, C. R. 2011. Heritability of gross feed efficiency and associations with yield, intake, residual intake, body weight, and body condition score in 11 commercial Pennsylvania tie stalls. *J. Dairy Sci.* **94**(4): 2108-2113.
- Vallimont, J.E., Dechow, C.D., Daubert, J.M., Dekleva, M.W., Blum, J.W., Liu, W., Varga, G.A., Heinrichs, A.J. and Baumrucker, C.R., 2013. Feed utilization and its associations with fertility and productive life in 11 commercial Pennsylvania tie-stall herds. *J. Dairy Sci.*, **96**(2):1251-1254.
- Van Arendonk, J. A. M., Nieuwhof, G. J., Vos, H., and Korver, S. 1991. Genetic aspects of feed intake and efficiency in lactating dairy heifers. *Livest. Prod. Sci.* **29**(4):263-275.
- VandeHaar, M. J., Armentano, L.E., Weigel, K., Spurlock, D.M., Tempelman, R.J., and Veerkamp, R. 2016. Harnessing the genetics of the modern dairy cow to continue improvements in feed efficiency. *J. Dairy Sci.* **99**(6):4941–4954.

- VanRaden, P.M., Van Tassell, C.P., Wiggans, G.R., Sonstegard, T.S., Schnabel, R.D., Taylor, J.F. and Schenkel, F.S. 2009. Invited review: Reliability of genomic predictions for North American Holstein bulls. *J. Dairy Sci.*, **92(1)**:16-24.
- VanRaden, P.M., Wiggans, G.R., Tooker, M.E., Megonigal, J.H., and Cole, J.B. 2007. Genetic Evaluations for Mixed-Breed Populations. *J. Dairy Sci.* **90(5)**:2434-41.
- VanRaden, P.M., Tooker, M.E., O'Connell, J.R., Cole, J.B., Bickhart, D.M., O'Connell, J.R., Cole, J.B., and Bickhart, D.M. 2017a. Selecting sequence variants to improve genomic predictions for dairy cattle. *Genet. Sel. Evol.* **49**:32.
- Van Raden, P.M., Wright, J.R., Connor, E.E., Vandehaar, M., Tempelman, R.J., Liesman, J., Weigel, K. 2017b. Preliminary genomic predictions of feed saved for 1.4 million Holsteins. *J. Dairy Sci.* **100(Suppl. 2)**:200–201(abstr. 209).
- Veerkamp, R.F., Pryce, J.E., Spurlock, D., Berry, D., and Coffey, M. 2013. Selection on feed intake or feed efficiency: A position paper from gDMI breeding goal discussions.
- VikingGenetics. 2019. Saved Feed Index Released in the Nordics. Accessed September 26, 2019. Available at: www.vikinggenetics.com/media/news?show=bedg.
- Waghorn G.C., Macdonald, K.A., Williams, Y., Davis, S.R., Spelman, R.J. 2012. Measuring residual feed intake in dairy heifers fed an alfalfa (*Medicago sativa*) cube diet. *J. Dairy Sci.* **95(3)**:1462-71.
- Wallace, R.J., Sasson, G., Garnsworthy, P.C., Tapio, I., Gregson, E., Bani, P., Huhtanen, P., Bayat, A.R., Strozzi, R., Biscarini, F., Snelling, T.J., Saunders, N., Potterton, S.L., Craigon, J., Minuti, A., Trevisi, E., Callegari, M.L., Cappelli, F.P., Cabezas-Garcia, E. H., Vilkki, J., Pinares-Patino, C., Fliegerová, K. O., Mrázek, J., Sechovcová, H., Kopečný, J., Bonin, A., Boyer, F., Taberlet, P., Kokou, F., Halperin, E., Williams, J.L., Shingfield, K.

- J. and Mizrahi, I. 2019. A heritable subset of the core rumen microbiome dictates dairy cow productivity and emissions. *Sci. Adv.* **5**:7.
- Wallén, S.E., Prestløkken, E., Meuwissen, T.H.E., McParland, S. and Berry, D.P., 2018. Milk mid-infrared spectral data as a tool to predict feed intake in lactating Norwegian Red dairy cows. *J. Dairy Sci.*, **101**(7):6232-6243.
- Weimer, P.J., Stevenson, D.M., Mantovani, H.C. and Man, S.L.C. 2010. Host specificity of the ruminal bacterial community in the dairy cow following near-total exchange of ruminal contents. *J. Dairy Sci.* **93**:5902–5912.
- Wickramasinghe, S., Cánovas, A., Rincón, G., Medrano, J.F. 2014. RNA-sequencing: a tool to explore new frontiers in animal genetics. *Livest. Sci.* **166**:206-216.
- Williams, Y.J., Pryce, J.E., Grainger, C., Wales, N. Linden, M., Porker, W.J., and Hayes, B.J. 2011. Variation in residual feed intake in Holstein Friesian dairy heifers in southern Australia. *J. Dairy Sci.* **94**:4715–4726.
- Williams, A.G. and Coleman, G.S. 1997. *The Rumen Microbial Ecosystem* (Chapman & Hall, 1997).
- Wolc, A., Arango, J., Settar, P., Fulton, J.E., O’Sullivan, N.P., Preisinger, R., Habier, D., Fernando, R., Garrick, D.J., Hill, W.G., and Dekkers, J.C.M. 2012. Genome-wide association analysis and genetic architecture of egg weight and egg uniformity in layer chickens. *Anim. Genet.* **43**(1):87-96.
- Yao, C., Spurlock, D.M., Armentano, L.E., Page, C.D., VandeHaar, M.J., Bickhart, D.M., and Weigel, K.A. 2013. Random Forests approach for identifying additive and epistatic single nucleotide polymorphisms associated with residual feed intake in dairy cattle. *J. Dairy Sci.* **96**(10):6716-29.

- Wang, X., and Kadarmideen, H. N. 2019. Metabolomics Analyses in High-Low Feed Efficient Dairy Cows Reveal Novel Biochemical Mechanisms and Predictive Biomarkers. *Metabolites*, **9**(7). doi: 10.3390/metabo9070151.
- Zhang, J., Shi, H., Cao, Z., Yang, H., and Wang, Y. 2019. Integrative hepatic metabolomics and proteomics reveal insights into the mechanism of different feed efficiency with high or low dietary forage levels in Holstein heifers. **194:1-13**. doi:10.1016/j.jprot.2018.12.026.
- Zhou, Y., Connor, E.E., Wiggans, G.R., Lu, Y., Tempelman, R.J., Schroeder, S.G., Chen, H., and Liu, G.E. 2018. Genome-wide copy number variant analysis reveals variants associated with 10 diverse production traits in Holstein cattle. *BMC Genom.* **19**:314.
- Zimin, A.V, Delcher, A.L., Florea, L., Kelley, D.R., Schatz, M.C., Puiu, D., Hanrahan, F., Pertea, G., Van Tassell, C.P., Sonstegard, T.S., Marçais, G., Roberts, M., Subramanian, P., Yorke, J.A. and Salzberg, S.L. 2009. A whole-genome assembly of the domestic cow, *Bos taurus*. *Genome Biol.* **10**:R42.

Tables

Table 1. Heritability estimates for different indicator traits of feed efficiency in dairy cattle.

Trait	Reference	$h^2 \pm SE^a$	Breed
Dry matter intake	Vallimont et al. (2010)	0.18±0.06	Holstein
	Williams et al. (2011)	0.17±0.10	Holstein Friesian
	Liinamo et al. (2012)	0.23±0.12	Nordic Red dairy cattle
	Tetens et al. (2014)	0.37±0.04	Holstein
	Shonka et al. (2015)	0.52±0.13	Holstein
	Bilal et al. (2016)	0.12±0.01	Canadian Holstein
	Byskov et al. (2017)	0.37±0.06	Holstein
	Lu et al. (2018)	0.23±0.02	NA ^b
Energy intake	Köck et al. (2018)	0.07±0.03 to 0.13±0.02	Fleckvieh, Brown Swiss and Holstein
Energy-corrected milk	Köck et al. (2018)	0.08±0.03 to 0.12±0.02	Fleckvieh, Brown Swiss and Holstein
Residual Feed Intake	Hurley et al. (2017)	0.04±0.08 to 0.11±0.08	Holstein Friesian
	Van Arendonk et al. (1991)	0.19±0.12	Dutch and Holstein Friesian
	Krover et al. (1991)	0.22±0.11	Dairy cattle raised in The Netherlands (breed not specified)
	Jensen et al. (1995)	0.36±0.17	Red Danish, Danish Friesian, Danish Red and White, and Jersey
	Svendsen et al. (1993)	0.02±0.08	Dual-purpose Norwegian cattle
	Vallimont et al. (2011)	0.01±0.05	Holstein
	Williams et al. (2011)	0.27±0.12	Holstein Friesian
	Byskov et al. (2017)	0.23±0.05	Holstein
	Lu et al. (2018)	0.16±0.02	NA

^a $h^2 \pm SE$: heritability \pm standard error.

^b NA: Data from international dairy consortium, included several breeds (not specified).

Table 2. Examples of genetic correlations between different indicator traits of feed efficiency and production and production-related traits in dairy cattle.

Feed efficiency trait	Trait	Reference	$r_g \pm SE^a$
Dry matter intake	Milk yield	Gonzalez-Recio et al. (2014)	0.10±0.11
		Vallimont et al. (2010)	0.51±0.32
	Fat- and protein-corrected milk yield	Difford et al. (2020)	0.83±0.04
	Fat yield	Gonzalez-Recio et al. (2014)	-0.03±0.10
		Vallimont et al. (2010)	0.53±0.34
	Protein yield	Gonzalez-Recio et al. (2014)	-0.11±0.08
		Vallimont et al. (2010)	0.55±0.37
	Somatic cell score	Vallimont et al. (2010)	-0.15±0.28
	Bodyweight	Liinamo et al. (2012)	0.54 to 1.00
		Vallimont et al. (2010)	0.52±0.35
	Natural logarithm of methane	Difford et al. (2020)	0.60±0.13
	Natural logarithm of carbon dioxide	Difford et al. (2020)	0.42±0.13
	Productive life	Vallimont et al. (2013)	0.49±0.18
	Meal size	Lin et al. (2013)	0.18±0.15
	Eating rate	Lin et al. (2013)	0.11±0.14
	Feeding duration	Lin et al. (2013)	0.48±0.12
	Number of meals	Lin et al. (2013)	0.03±0.16
	Days open	Vallimont et al. (2013)	-0.14 ± 0.29
	Body condition score	Gonzalez-Recio et al. (2014)	0.37±0.32
		Liinamo et al. (2012)	0.11 to 0.45
		Vallimont et al. (2010)	0.37±0.46
Residual Feed Intake	Milk yield	Veerkamp et al. (1994)	-0.11 to 0.07
		Gonzalez-Recio et al. (2014)	0.07±0.08
	Fat yield	Gonzalez-Recio et al. (2014)	0.02±0.07
	Protein yield	Gonzalez-Recio et al. (2014)	0.03±0.07
		Veerkamp et al. (1994)	-0.11 to -0.02
	Lactose	Veerkamp et al. (1994)	-0.19 to -0.05
	Bodyweight	Korver et al. (1990)	0.03
		Van Arendonk et al. (1991)	0.01
	Natural logarithm of methane	Difford et al. (2020)	0.42±0.23
	Natural logarithm of carbon dioxide	Difford et al. (2020)	0.48±0.24
	Productive life	Vallimont et al. (2013)	-0.23±0.29
	Days open	Vallimont et al. (2013)	-0.50±0.40
	Meal size	Lin et al. (2013)	-0.06±0.16
	Eating rate	Lin et al. (2013)	0.06±0.16

Feeding duration	Lin et al. (2013)	0.27±0.15
Number of meals	Lin et al. (2013)	-0.07±0.17
Body condition score	Gonzalez-Recio et al. (2014)	0.71±0.32
	Veerkamp et al. (1994)	0.33 to 0.36

^a $r_g \pm SE$: additive genetic correlation \pm standard error.

Table 3. Summary of Genome-Wide Association Studies (GWAS) performed in dairy cows to identify genomic regions associated with feed efficiency traits.

Breed, country, reference	N	Trait	N of SNPs	Statistical analysis	Significance threshold	N of significant markers
Holstein, USA, (Li et al. 2019)	5,610	RFI	278,254 (after quality control)	Single-step GWAS	Top-20 SNPs and 5-SNP sliding windows	20 single-SNPs 20 5-SNPs sliding windows
Holstein, Scotland, Netherlands, Canada, and USA, (Lu et al. 2018)	4,916	RFI based on classical model and multiple-trait (MT) model (Lu et al. 2015) using dry matter intake (DMI), milk energy (MILKE), and metabolic body weight (MBW)	57,347 (after quality control)	Single-SNP marker association and windows-based association (1 Mb non-overlapping windows)	Multiples, based on Bonferroni adjustment at genome-wide type I error rate of 5%	MBW = 4 SNPs, 3 windows RFI = 2 windows MT = 2 windows DMI = 2 windows MILKE = 1 window MBW = 3 windows
Holstein, USA, (Zhou et al. 2018)	473	Residual feed intake and Dry matter intake	454 Copy Number Variants (CNVs)	Multiple linear regression	p-value<0.05 after FDR correction	RFI = 10 CNVs DMI = 1 CNV
Holstein, USA, Canada, Netherlands and United Kingdom, (Hardie et al. 2017)	4,916	RFI was calculated as the residual of the regression of dry matter intake (DMI) on milk energy (MILE), metabolic body weight (MBW), change in body	60,671 markers	BayesB and BayesC	Top-10 genomic windows	RFI = 5.38% (PM) and 4.80% (MP) of total genetic variance explained by the top 10 windows DMI = 9.18% (PM) and 5.54% (MP)

		weight, and systematic effects. RFI, DMI, MILKE, and MBW, bivariate analyses were performed for each trait as a separate trait within parity group.				MILKE = 7.12% (PM) and 5.08% (MP)
						MBW = 9.31% (PM) and 9.8% (MP)
Holstein, Germany, (Tetens et al. 2014)	681	DIM 11, 30, 80, 130, and 180	40,407 (after pruning for loci >10% of missing genotypes, MAF<0.05 and markers without position)	Linear mixed model approach implemented in the package GEMMA	p-value<0.05 after Bonferroni correction.	DMI11= 4 markers DMI30= 8 markers DMI80= 3 markers DMI130= 5 markers DMI180= 7 markers
Holstein, USA, (Yao et al. 2013)	395	Daily RFI from 50 to 150 d postpartum	42,275	Random Forest algorithm	Importance score (Δ MSE%)	188 markers
Holstein, Australia and New Zealand, (Pryce et al. 2012)	1,782	RFI	624,930	Bayes A and Bayes	The 1,000 largest SNP effects ranked on absolute value were selected.	1,000 markers

4
5
6
7

Table 4. Summary of RNA sequencing studies comparing divergent feed efficient groups (based on Residual feed intake, RFI) in dairy cattle.

Breed, country	Sample size	Tissue	Significance threshold	N of differentially expressed genes	Reference
Holstein (H) and Jersey (J), Denmark	High-RFI ^a = 5 (H) and 5 (J) Low-RFI ^a = 4 (H) and 5 (J)	Liver	p-value < 0.05	Holstein: 11 modules of co-expressed genes Jersey: 4 modules of co-expressed genes	(Salleh et al., 2018)
Holstein (H) and Jersey (J), Denmark	High-RFI ^a = 5 (H) and 5 (Jersey) Low-RFI ^a = 4 (H) and 5 (J)	Liver	FDR < 5%	70 (H for model 1) 19 (J for model 1) 2 (J for model 2)	(Salleh et al., 2017)
Holstein, Australia	High-RFI ^b = 9 animals Low-RFI ^b = 10 animals	Liver and white blood cells	FDR < 5%	Liver RFI = 473 Liver GEBV = 526 WBC RFI = 4,817 WBC GEBV = 137	(Khansefid et al., 2017)

^a RFI groups were defined based on the ranking of random effects solutions (from a fixed linear regression on metabolic body weight, daily live weight change, daily body condition score change (fitted with a Legendre polynomial), and energy corrected milk yield.) for 200 animals, where the top and bottom animals were selected.

^b RFI groups were defined using the top and bottom 10% animals from the RFI distribution in a population composed by 843 animals (average RFI = 0, and SD = 0.19; Williams et al., 2011).

Table 5. Accuracies of genomic predictions for indicator traits of feed efficiency.

Trait	Reference	Average accuracy
Dry matter intake	de Haas et al. (2012)	0.35
	de Haas et al. (2015)	0.37
	Mujibi et al. (2011)	0.20
	Boloorma et al. (2013)	0.32
	Negussie et al. (2019)	0.42 – 0.57 (simulation-based)
Residual feed intake	Pryce et al. (2012)	0.40
	Mujibi et al. (2011)	0.43
	Boloorma et al. (2013)	0.43
	VanRaden et al. (2017b)	0.44
	Negussie et al. (2019)	0.22 – 0.50 (simulation-based)