Genetic variation in residual feed intake is associated with body composition, behavior, rumen, heat production, hematology, and immune competence traits in Angus cattle[1]

Robert M. Herd,[†,‡] Jose I. Velazco,[‡,‖] Helen Smith,[§] Paul F. Arthur,[¶,2] Brad Hine,[**] Hutton Oddy,[†] Robin C. Dobos,[†] and Roger S. Hegarty[‡]

[†]NSW Department of Primary Industries, Livestock Industries Centre, Armidale, NSW 2351, Australia; [‡]Environmental and Rural Science, University of New England, Armidale, NSW 2351, Australia; [‖]National Institute of Agricultural Research, Treinta y Tres 33000, Uruguay; [§]Local Land Services Agency, Braidwood 2622, NSW, Australia; [¶]NSW Department of Primary Industries, Elizabeth Macarthur Agricultural Institute, Menangle, NSW 2568, Australia; and **CSIRO Agriculture and Food, F.D McMaster Laboratory, Armidale, NSW 2350, Australia

ABSTRACT: This experiment was to evaluate a suite of biological traits likely to be associated with genetic variation in residual feed intake (**RFI**) in Angus cattle. Twenty nine steers and 30 heifers bred to be divergent in postweaning RFI (**RFIp**) and that differed in midparent RFI_p-EBV (**RFIp-EBVmp**) by more than 2 kg DMI/d were used in this study. A 1-unit (1 kg DM/d) decrease in RFI_p-EBV_{mp} was accompanied by a 0.08 kg (SE = 0.03; $P < 0.05$) increase in ADG, a 0.58 kg/d (0.17; $P < 0.01$) decrease in DMI, a 0.89 kg/kg (0.22; $P < 0.001$) decrease in FCR, and a 0.62 kg/d (0.12; $P < 0.001$) decrease in feedlot RFI (**RFIf**). Ultrasonically scanned depths of subcutaneous fat at the rib and rump sites, measured at the start and end of the RFI test, all had strong positive correlations with RFI_p-EBV_{mp}, DMI, and RFI_f (all r values ≥0.5 and $P < 0.001$). Variation in RFI_p-EBV_{mp} was significantly correlated ($P < 0.05$) with flight speed ($r = -0.32$), number of visits to feed bins ($r = 0.45$), and visits to exhaled-emission monitors ($r = -0.27$), as well as the concentrations of propionate ($r = -0.32$) and valerate ($r = -0.31$) in rumen fluid, white blood cell ($r = -0.51$), lymphocyte ($r = -0.43$), and neutrophil ($r = -0.31$) counts in blood. RFI_p-EBV_{mp} was also correlated with the cellular immune response to vaccination ($r = 0.25$; $P < 0.1$) and heat production in fasted cattle ($r = -0.46$; $P < 0.001$). Traits that explained significant variation ($P < 0.05$) in DMI over the RFI test were midtest metabolic-BW (44.7%), rib fat depth at the end of test (an additional 18%), number of feeder visits (additional 5.7%), apparent digestibility of the ration by animals (additional 2.4%) and white blood-cell count (2.1%), and the cellular immune response to vaccine injection (additional 1.1%; $P < 0.1$), leaving ~23% of the variation in DMI unexplained. The same traits (BW excluded) explained 33%, 12%, 3.6%, 3.7%, and 3.1%, and together explained 57% of the variation in RFI_f. This experiment showed that genetic variation in RFI was accompanied by variation in estimated body composition, behavior, rumen, fasted heat production, hematology, and immune competence traits, and that variation in feedlot DMI and RFI_f was due to differences in BW, scanned fatness, and many other factors in these cattle fed ad libitum and able to display any innate differences in appetite, temperament, feeding behavior, and activity.

[1]This work was funded by NSW Department of Primary Industries, University of New England, Meat and Livestock Australia and the Australian Government Department of Agriculture and Water Resources as part of the National Livestock Methane Program. The skilled technical assistance provided by Simon Bird, Steve Harden, David Mula, Karen Dibley, Peter Newman, Peter Kamphorst, Ian Higgins, Reg Woodgate, Chris Weber, Dan Ebert, Bill Johns, Graeme Bremner, Colin Crampton, Dominic Niemeyer, and Amy Bell is gratefully acknowledged. Brad Walmsley generously provided the equations used for calculating body composition. One of us (J.I.V.) was supported by an Australian Government scholarship funded by the Australian Agency for International Development and by the National Institute for Agricultural Research (INIA Uruguay).

[2]Corresponding author: paul.arthur@dpi.nsw.gov.au

Received November 5, 2018.
Accepted February 20, 2019.

Key words: body composition, cattle, digestibility, feed efficiency, immune competence

J. Anim. Sci. 2019.97:2202–2219
doi: 10.1093/jas/skz077

INTRODUCTION

Residual feed intake (**RFI**) is a measure of feed efficiency in beef cattle calculated as the difference between actual feed intake by an animal and its expected feed intake for maintenance and BW gain over a test period (Koch et al., 1963). It is a measure of feed efficiency calculated to be independent of BW and gain and potentially identifies variation in underlying physiological processes such as those that determine maintenance feed requirements (Archer et al., 1999). Herd and Arthur (2009) concluded that five major physiological processes are likely to contribute to variation in RFI, these being processes associated with the intake of feed, digestion of feed, metabolism (anabolism and catabolism associated with and including variation in body composition), physical activity, and thermoregulation. In their recent reviews, Cantalapiedra-Hijar et al. (2018) concluded that "the number of potential mechanisms involved in animal-to-animal variation in feed efficiency is huge", and Kenny et al. (2018) that "feed efficiency is multifactorial and complex trait" and more experimental information is needed "to unravel the biological regulation of the trait." The aim of this current experiment was to use Angus cattle bred to vary in genetic merit for RFI_p, to examine associations of genetic variation in RFI_p and the resulting phenotypic variation in feedlot DMI and RFI (**RFI_f**) with variation in measures of body composition, temperament, behavior, digestive function, hematology, immune competence, body temperature, heat production, maintenance energy requirements (**MER**), and energy budget. The traits studied included novel traits not previously examined but identified in the above reviews of RFI, and were all measured on the same animals during or following testing for RFI_f.

MATERIALS AND METHODS

Animals

This research was approved under NSW Department of Primary Industries Animal Research Authority ORA 13/16/004, and the University of New England Animal Research Authorities AEC14-002 and AEC14-036.

The Angus cattle were bred at the NSW DPI Agricultural Research Centre, Trangie, NSW, Australia. They were bred by AI using cows from the RFI postweaning (**RFI_p**) divergent selection lines described by Arthur et al. (2001) and stored semen from two sires from each line. The EBV for RFI_p (**RFI_p-EBV**) of the two low-RFI sires were −0.61 and −0.97 kg DM/d and for the two high-RFI sires +0.61 and +1.42 kg DM/d. By chance the sires differed slightly in their EBV for BW at 400 d of age (**400dBW-EBV**); being +40 and +68 kg for the two low-RFI sires, and +33 and +53 kg for the two high-RFI sires. The RFI_p-EBV of the dams joined to the two low-RFI sires ranged from 0.61 to −0.97 kg DM/d and for those joined to the two high-RFI sires from +0.61 to +1.42 kg DM/d. Values for the RFI_p and 400dBW-EBV were extracted from the Australian Angus Society EBV database on 7 February 2017 (https://www.angusaustralia.com.au/), and calculated without the RFI_f test data for the animals used in this experiment. At weaning on 26 February 2013, a total of 64 calves (30 steers: 34 heifers) were available for this experiment. They were grown on pastures until they reached feedlot entry weight of approximately 400 kg BW and were on average 579 (16; SD) d of age at the start of the RFI_f test.

RFI_f Test

Following a period of 5 wk for induction, adaption to the feedlot grain ration, and acclimation to feeding from the feed bins of a GrowSafe feed-intake recording system (GrowSafe Systems Ltd., Airdrie, Alberta, Canada), the cattle underwent a 10-wk RFI test. Heifers were kept together in a single feedlot pen and steers together in an adjacent pen, and the sexes were swapped between pens midway through the test period. Each feedlot pen was ~12.5 m wide × 40 m long, with the shorter side facing the feedlot laneway. Each pen contained 4 GrowSafe feed bins, side-by-side each other, and

positioned midway along the side of the pen facing the feedlot laneway. Each pen contained a single water trough located midway along the longer side of the pen. Mounted over the water trough were two GrowSafe Beef cattle-weighing platforms used to record the time of visits to the water trough by individual animals. Each pen also contained a single Greenfeed Emission Monitor (**GEM**; C-Lock Inc., Rapid City, South Dakota), positioned in one corner of the pen by the feedlot laneway, and used to measure methane production rate (**MPR**) from multiple short-term samplings. Details on the operation of the GEM and processing of data from both the GrowSafe feed bins and the GEM are given in Herd et al. (2016). Four of the 64 animals were removed before the RFI test by the feedlot manager for failing to adapt to eating from the feed bins. The RFI test commenced 3 February 2014, with the cattle ~19 months of age and 450 kg BW. One animal was removed from the RFI test because it stopped eating regularly from the feed bins and 59 animals (29 steers: 30 heifers) completed the RFI test. Forty-one animals (22 steers: 19 heifers) were judged to have sufficient number of valid records from the GEM to calculate MPR, being animals with a minimum of 30 records each of at least 3 min duration recorded over the RFI test, as recommended by Arthur et al.(2017).

The test ration was a high grain-content finishing ration that consisted of ~80% barley grain, 10% sorghum hay, 5% protein pellets, plus a proprietary mixture of molasses, water, and vitamin and mineral additives (fresh weight basis). Samples of this ration from the start, midway through, and end of the RFI test were sent to a commercial feed evaluation service (NSW Department of Primary Industries Feed Testing Service, Wagga Wagga NSW: http://www.dpi.nsw.gov.au/aboutus/services/das/feed-quality-service). The averaged reported content of the ration was 88% DM, 12% CP (DM basis), ether extract 4% of DM, 86% DM digestibility (**DMD**; two-stage in vitro digestion) and ME content of 13.2 MJ/kg DM (all determined following methods described in AFIA (2014) and ME calculated using equation 1 on page 91 of that publication). The mean value for DMD by cattle measured in the RFI_f test was 69.6% (method described below) and lower than the averaged reported value. A revised, lower value of 10.3 MJ/kg DM for the apparent ME content of the test ration was calculated using the same equation as for the reported value and is used in calculations of ME intake over the RFI test.

The cattle were measured for growth rate, feed intake, and feed efficiency following the test guidelines described by Exton (2001). At the start of the test, and then each fortnight, the cattle were walked from their pens to the nearby cattle processing yards where they were weighed. After weighing the cattle moved forward into a short section of cattle chute where they stood and a sample of feces (about 50 g) for subsequent determination of apparent DMD was collected by gloved hand from the rectum of each animal. Corresponding feed samples were taken 3 d before each fortnightly fecal sampling. Each animal was then moved forward onto a weighing platform, and then forward into a crush and two measures of temperament: crush score (**CS**) and flight speed (**FS**) were recorded. The CS was based on the amount of movement each animal made while standing in a crush, assessed visually on a five-point scale of agitation, where 1 = calm and 5 = highly agitated, as used in Cafe et al. (2011). The same person gave the CS throughout the test period and had been trained by the lead author of Cafe et al. (2011). FS (m/s) was calculated from the electronically recorded time for each animal to cover a known distance (~1.7 m) on release from the cattle crush, using the same equipment used in Cafe et al. (2011). The six fortnightly measurements of CS and FS were averaged to give a mean value over the RFI test for each animal.

Movement of individual animals within their feedlot pen during the RFI test was tracked using visitation data provided by the GrowSafe and GEM systems. The number of times per day each animal visited a GrowSafe feed bin (feed visits), visited the water trough via the GrowSafe Beef chute (water visits), and visited the GEM (**GEM visits**) in their pen was calculated. All visits, regardless of whether feed or water was consumed, or exhaled emissions were recorded, were tallied. Software was written to extract the temporal pattern of movement by each animal between machines over a day and used to calculate the minimum distance travelled for feeding and drinking activity per day (**DIST**) for each animal, recognizing that additional animal movements could occur that was not measured by this method. Data for the first and last days of the RFI test, day 37 when the animals were swapped between pens, and day 13 when an electrical-power failure interrupted data capture, were not used leaving daily records for each animal for 66 d. These records were averaged to produce a mean record for each animal for daily feed visits, water visits, GEM visits, and DIST over the RFI-test period.

On day 0 of the RFI test, and at the end of the test, an accredited technician used real-time ultrasound scanning to measure subcutaneous fat depth at the 12/13th rib (**RIBFAT**) and over the rump (Australian P8 site; **RUMPFAT**), cross-sectional area of the eye-muscle (M. longissimus dorsi; **EMA**) between the 12th and 13th ribs, and intramuscular or marbling fat as a percentage of fresh weight of muscle (**IMF**). Change (gain) in these traits over the RFI test was calculated as the difference between the end-of-test values and the start-of-test values. Fat and muscle content relative to the size of the animals was calculated by dividing fat depth and muscle cross-sectional area by animal BW on the day of scanning.

Digestive Function Traits

Whole-of-tract DMD, rumen concentrations of VFA and two methane emission traits were measured. Apparent digestibility of DM in the feed consumed by each animal was calculated indirectly using Silica (**Si**) as a naturally occurring indigestible internal marker. The use of Si concentration in feed and feces measured using portable X-ray fluorescence (**PXRF**) spectroscopy for digestibility studies in sheep and cattle has been validated by Barnett et al. (2016). Fortnightly fecal samples were collected into 70 mL plastic sample tubes, and feed samples in sealed polyethylene bags, and stored frozen at −20 °C for subsequent determination of Si concentration by the methods and PXRF machine used by Barnett et al. (2016). The frequency distribution of Si counts for the 300 fecal samples analyzed from animals in the feedlot test was checked for normality because ingestion of dirt or soil by an animal could result in elevated Si counts. The fecal samples were found to be skewed towards higher Si counts (skewness factors: 0.50) and results for samples with a Si count greater than three times the SD above the mean for all samples were discarded as being possibly contaminated: being two samples or 0.7% of all fecal samples. Results presented are mean values for 59 animals, with 56 animals having results for five fortnightly fecal samples, two animals with four results, and one animal with three results. The "count" for Si, with no conversion to Si concentration, was used to calculate DMD using the formula: DMD = 1 − (feed Si count/feces Si count).

On the final day of the RFI test, when the cattle were moved from their pens to be weighed, a rumen fluid sample was collected from each animal by aspiration through a flexible stomach tube, this being 1 to 2 h since access to feed. Rumen fluid was preserved by acidification and stored at 4 °C until concentrations of VFA were determined by GLC (Nolan et al., 2010). Three samples appeared to be poorly preserved when prepared for chromatography so only results for 56 of the 59 animals with RFI results are reported.

Methane produced by each animal over the RFI test was measured by GEM, as described above. To further quantify the loss of energy as methane from feed, methane yield (**MY**; g/d) was calculated using the formula: MY = MPR/DMI, with higher values representing more methane produced per unit of feed eaten. Following the RFI test and other posttest measurements at the feedlot the animals were measured for MPR and MY on a restricted allowance of roughage ration expected to result in higher levels of MY (Blaxter and Clapperton, 1965). These measurements were made in individual respiration chambers at the University of New England, Armidale, NSW, Australia. The animals were moved in groups of 10 to the animal house and were offered oaten hay chaff ad libitum for a minimum of 30 d. Then each group was offered an amount calculated to be the total weight of ration that the animals were to be offered subsequently in individual pens inside the animal house for a minimum of 4 d. The amount offered was calculated to be equivalent to 1.2 times the expected maintenance requirement for each animal. In the animal house, the cattle were accommodated and fed in individual pens for 2 d, then weighed, and put into individual respiration chambers for 2 d of measurement. This weight was used as the test weight (**TWT**) for the animal. This protocol meant that MPR on roughage was measured between 56 and 91 d after the end of the RFI test, provided time for the animal to adapt to the roughage ration and with all animals on a similar level of feed energy intake relative to BW when measured. The roughage ration, calculation of the feeding allowance, and measurement protocols are described in Herd et al. (2016).

Hematology, Stress Responsiveness, and Immune Competence Measurements

Following the RFI test, blood samples were taken on four occasions: the final day of the RFI test (baseline) and 4, 11, and 32 d after the RFI test (days 4, 11, and 32), via jugular venepuncture into evacuated tubes coated with EDTA anticoagulant

for hematological studies, or with no anticoagulant for stress responsiveness and immune competence studies. Blood samples for hematology were stored at 4 °C until reaching the laboratory where they were warmed to room temperature and analyzed on the same day. Blood samples for stress responsiveness and immune competence testing were stored at room temperature until they could be centrifuged (700 × *g*, 20 min), ~4 h later, and serum stored in multiple aliquots at −20 °C for subsequent analysis.

Hematology

Blood samples were analyzed on an automated hematology analyzer (Cell-Dyn 3500R, Abbott Diagnostics, North Ryde, NSW, Australia) with a specialized veterinary package installed. Parameters measured included red blood cell count (**RBC**), mean corpuscular volume (**MCV**), total white blood cell count (**WBC**), hemoglobin concentration (**HGB**), and platelet count (**PLT**). Differential white blood cell counts were also conducted to determine numbers of lymphocytes (**LYM**), neutrophils (**NEU**), monocytes (**MONO**), eosinophils (**EOS**), and basophils (**BAS**). Parameters calculated included %Lymphocytes (%**LYM**; =LYM/WBC as a %), %Neutrophils (%**NEU**; =NEU/WBC as a %), mean corpuscular hemoglobin (**MCH**; = (HGB/RBC) × 10, in picograms), hematocrit (**HCT**; = (RBC × MCV)/10, %), and mean corpuscular hemoglobin concentration (**MCHC**; = (HGB/HCT) × 100, in g/dL).

Stress Responsiveness

The acute phase protein haptoglobin (**HAPT**) has been shown to be a good candidate for measuring stress in cattle due to its half-life of 2 to 4 d, its latency to peak and has been shown to increase in response to production stressors such as weaning, transportation, social regrouping, and intensive management (Slocombe and Colditz, 2005). On the final day of the RFI test, a blood sample was taken to assess baseline levels of HAPT, and 4 d later, a second blood sample was taken to measure the change in serum HAPT levels following the additional handling, prolonged restraint, blood sampling, ultrasound scanning, and rumen fluid sampling of the cattle at the end of the RFI test. Serum HAPT was analyzed using the method described by Slocombe and Colditz (2012). Change in HAPT level (**ΔHAPT)** from baseline to day 4 is reported as the stress response.

Immune Competence Tests

The general immune competence of individual animals was assessed by measuring both the antibody and cellular immune responses (**AIR, CIR**) induced by vaccination with a clostridial vaccine (Aleri et al., 2015). Antibody and cellular responses were induced by vaccination with a commercial vaccine, ULTRAVAC 7in1 vaccine (Zoetis Australia, Sydney, Australia). A blood sample was taken for baseline antibody levels and then the vaccine was administered subcutaneously high on the neck as per manufacturer's instructions and at the manufacturer's recommended dose. Eleven days and 32 d later further blood samples were collected. Antibody production, specifically anti-tetanus toxoid serum IgG1, in response to the tetanus toxoid component of the multivalent vaccine was determined to assess AIR. Serum anti-tetanus toxoid IgG1was assayed using an indirect ELISA method described by (Aleri et al., 2015) and is reported in optical density (OD) units. Serum samples were assayed in quadruplicate and values corrected for control samples run on all plates. Baseline levels of antibody (due to routine previous vaccinations) were subtracted from days 11 to 32 levels and AIR is reported as the change in antibody level values at days 11 and 32 relative to baseline (**ΔAIR11** and **ΔAIR32**). The CIR was assessed by the magnitude of delayed-type hypersensitivity response to intradermal injection of the 7in1 vaccine. On day 32, 0.1 mL of 7in1 vaccine (test) or saline (control) was injected into the caudal tail fold using an insulin syringe with 30G needle. Before injection, injection sites were identified and the skin-fold thickness measured three times with Harpenden spring-loaded callipers (Baty International Ltd, West Sussex, UK) to provide a baseline skin-fold thickness. Forty-eight hours later, skin-fold thickness was again measured three times and the mean of the increase in skin-fold thickness (in mm) calculated to determine the cellular response due the saline control injection (**CIR ctrl**), and the 7in1 injection (**CIR test**).

Heat Production (HP) Traits

HP was calculated over the 2-d period that each animal was in the respiration chambers for MPR measurement. At the end of the 2-d period, being after a minimum of 8 d of being fed just above expected maintenance requirement, the animals were not fed and remained in the chambers for determination of their unfed HP over the next 24 h. This protocol follows that of Blaxter and Wainman

(1966) and HP is reported as unfed HP rather than fasting HP since it was measured over 1 d of fasting rather than during 4 to 5 d of fasting as employed by those authors. Fed and unfed HP were calculated using the equation of Brouwer (1965) and ignored energy loss in methane (unfed animals) and urinary N (fed and unfed animals). Without a direct measurement of O_2 consumed, it was calculated from the measured CPR and an assumed respiratory quotient (**RQ**). Values for RQ in sheep fed a range of conventional diets range from 0.9 to 1.1, while values below 0.8 appear to be found only in animals fasted for more than 48 h (Whitelaw, 1974). An RQ value of 1 was used for the animals fed a restricted roughage allowance. Heat production by each animal when unfed was calculated using a RQ value of 0.9, being intermediate between the values of 0.96 and 0.82 measured after 1 and 2 d of fasting in British-breed cattle of comparable weight to those in this experiment and that had being fed a restricted allowance of roughage ration prior to fasting (Blaxter and Wainman, 1966). Weight-specific HP (**HP-WT**) was calculated by dividing HP by TWT. Residual HP (**RHP**), representing more or less HP per unit of feed eaten was calculated by regressing individual animal HP against DMI, with the residuals being RHP.

Differences in body temperature (**TEMP**) have been shown to accompany differences in energy metabolism (Nielsen, 1966; Finch, 1986). Before being put into the respiration chambers each animal had rectal temperature recorded over 2 d. Rectal temperature was logged every 3 min over 2 d using the probe described by Lea et al. (2008) and averaged to calculate TEMP. Residual body temperature (**RTEMP**) was calculated by regressing TEMP against DMI, and the residuals (RTEMP) represent more or less energy metabolism per unit of feed eaten.

Energy Budgets

Energy budgets, being the sum of MER, energy loss due to activity (**Activity**), energy retained (**ER**) in body tissues deposited, and HP for energy lost in the process of tissue gain (**HPgain**), were calculated for each animal over the RFI_f test using data from this experiment and literature values.

Maintenance energy requirement (MJ/d) was calculated in two ways: first using literature equations (**MER lit**) and second from HP measured when unfed (**MER test**). MER lit was calculated as fasting metabolism (**FM**) using the equation: FM (MJ/d) = 0.53 × (fasted BW)$^{0.67}$ (ARC, 1980), with fasted or "shrunk" BW calculated as feedlot TWT × 0.96 (NRC, 1996); plus an activity allowance for nonfasting eating of 0.0043 MJ/d of BW as suggested by ARC (1980); plus an extra allowance of 0.1 times the predicted ME required for weight gain as recommended by SCA (2007). The calculation of MER test was as for MER lit except that FM used each animal's unfed HP.

Activity (MJ/d) was calculated as the sum of energy used for walking, standing while eating and standing in the feedlot yard, using either literature equations or test data (**Activity lit, Activity test**). For Activity test, energy used for walking was calculated for each animal by multiplying the estimated minimum distance walked by each animal (DIST) by 2 (as a conservative estimate of additional movement not measured) and multiplying this value by 2.6 kJ/km of horizontal walking for each kg of TWT (SCA, 2007). For Activity lit and Activity test, the energy used for standing while eating was calculated by multiplying the average time of 2 h spent standing while eating by British-breed cattle in this research feedlot (Fell and Clarke, 1993) by 2.5 kJ/h for each kg of TWT (SCA, 2007), and the energy cost for standing in the feedlot yard was calculated as 10 kJ/d for each kg of TWT as suggested by SCA (2007).

ER in tissue gain was calculated for each animal as the difference between the energy content of the empty body at the start and end of the RFI test. This required calculation of the energy as fat and protein in the empty body weight, and multiplying each by their respective energy densities. Empty body fat (**EBF**; kg) and protein (**EBP**; kg) were calculated using the model described by Walmsley et al. (2014), and used BW and RIBFAT as inputs, then multiplying EBF and EBP by their respective energy densities (39.3 and 23.6 MJ/kg). Heat production for tissue gain (HPgain) was calculated as ER as fat divided by 0.75 plus ER as protein divided by 0.20.

The predicted ME intake (**MEI**; MJ/d) of each animal over the RFI_f test was then calculated using the literature equations above (**MEI lit**) or from test data (**MEI test**) as MER (MER lit or MER test) plus Activity (Activity lit or Activity test) plus ER plus HPgain. The actual measured MEI for each animal was calculated as the sum of the ME in the feedlot ration consumed plus the ME in GEM-pellets consumed.

Traits and Statistical Analysis

Data for the 59 animals that completed the RFI test were used to calculate RFI_f. Daily intake

of feedlot ration and GEM pellets by each animal were summed and then averaged over the test period to calculate average DMI. Start BW, midtest BW, and ADG over the test were calculated for each animal from the linear regression of its fortnightly BW against day of test, with midtest BW being used as the TWT for the RFI test. To calculate RFI_f, in a linear model individual animal data for feedlot DMI were regressed against sex (steer or heifer), $TWT^{0.75}$ and ADG, with the residuals being RFI_f. Sex was not significant ($P > 0.1$) and was not included in the final linear model. The two traits: RFI_p and RFI_f are genetically correlated but they are technically classified as two different traits for the purpose of animal breeding because their genetic correlation does to meet the accepted 0.80 threshold (Jeyaruban et al., 2009). To test the consequences of breeding the next generation based on the RFI_p-EBV of the parents, the midparent RFI_p-EBV (**RFI_p-EBV_{mp}**) for each animal in the experiment was calculated as the mean of the sire and dam RFI_p-EBV and is the expected genetic merit of each animal for RFI_p, without using the animal's own RFI_f-test record in the calculation.

All analyses were performed using the SAS suite of software (SAS, 2012). The first analysis was of phenotypic variation in each of the production and physiological traits associated with variation in RFI_p-EBV_{mp}, that is, with the expected inherited genetic variation in RFI_p. To reduce the confounding effect that inherited differences in genetic merit for 400d-BW may have on the calculated magnitude of associations of traits with RFI_p-EBV_{mp}, midparent 400dBW-EBV (**400dBW-EBV_{mp}**) was fitted before RFI_p-EBV_{mp} in the model and the partial variation accounted for by RFI_p-EBV_{mp} calculated within PROC GLM as the partial eta-square option available within the procedure, with the square root of this value being equivalent to an *r* value. The magnitude of change in a trait per unit change in RFI_p-EBV_{mp} calculated as the regression coefficient provided by the solution option within Proc GLM.

The second analysis examined variation in feedlot DMI and RFI_f associated with variation in individual body composition and physiological traits. The strength of these associations was measured by calculating their correlation coefficients (*r* values) and *P*-value, and amount of variation in one trait explained by the other as r^2. Where required, the magnitude of change in a trait per unit change in another trait was calculated as the regression coefficient (*b*-value).

The third analysis was to check if associations RFI_p-EBV_{mp}, DMI, or RFI_f with hematological traits were maintained or changed over the days following the RFI test and in response to vaccination. Variation in each hematological trait was examined using GLM with day fitted as a class factor, RFI_p-EBV_{mp}, DMI, or RFI_f as continuous variables, and their interaction, with a statistically significant interaction being taken as evidence for change in the relationship between the hematological trait with RFI_p-EBV_{mp}, DMI, or RFI_f over time.

The fourth analysis was to explain as much of the phenotypic variation in feedlot DMI, RFI_f, and MEI as possible by variation in the body composition and physiological traits measured during or following the RFI_f test. This was achieved by using step-wise regression: DMI or RFI_f was regressed against a subset of the traits measured to calculate the magnitude of the incremental variation in DMI or RFI_f explained by inclusion of each trait in the model. The traits selected were from the different physiological categories and within a category were those with the highest correlation with DMI determined in the first analysis. For the analysis of MEI, traits that could be expressed in energy units or expected to be associated with ME requirements were used.

RESULTS

RFI_f Test

In the current experiment, $TWT^{0.75}$ explained 44.7% ($P < 0.001$) of the variation in DMI over the RFI test, ADG explained an additional 1.4% ($P > 0.1$), and together they explained 46.1% of the variation in DMI. For British-breed steers in large experiments in the research feedlot used in this experiment, the variance in DMI explained by $TWT^{0.75}$ had a mean of 29% recorded using the GrowSafe system (Torres-Vázquez et al., 2018) and 49% using an older feed-intake recording system (Robinson and Oddy, 2004), with the latter authors reporting that the amount of variation in DMI explained by BW and ADG declined in test groups of older, fatter animals. Descriptive statistics for animal production and scanned body composition traits measured over the RFI_f test are presented in Table 1. Genetic merit for RFI_p of the steers and heifers, as predicted by their RFI_p-EBV_{mp}, differed by over 2 kg/d of DMI. Lower RFI_p-EBV_{mp} was associated with higher ADG, lower DMI, lower FCR, and lower RFI_f over the RFI test. A 1-unit (1 kg DM/d) decrease in RFI_p-EBV_{mp} was accompanied

Table 1. Descriptive statistics for age, weight, feed intake, feed efficiency and initial, final, and gain in body composition traits over a 10-wk feedlot RFI test and their correlations with midparent RFI_p-EBV, DMI, and RFI for 59 Angus steers and heifers[1] bred to vary in genetic merit for postweaning RFI

						Correlations		
Trait	Units	Mean	SD	Min.	Max.	RFI_p-EBV_{mp}	DMI	RFI_f
RFI_p-EBV_{mp}	kg/d	0.03	0.83	−1.00	1.13	1	0.41**	0.57***
Start BW	kg	410	36	342	499	−0.04	0.68***	0.00
ADG	kg/d	1.40	0.20	0.97	1.95	−0.29*	0.17	0.00
TWT	kg	454	38	383	548	−0.09	0.67***	0.00
End BW	kg	504	42	417	600	−0.14	0.64***	0.00
DMI	kg/d	12.0	1.1	9.5	14.5	0.41**	1	0.73***
FCR	kg/kg	8.9	1.4	5.8	12.0	0.48***	0.39**	0.42**
RFI_f	kg/d	0.00	0.82	−1.80	1.83	0.57***	0.73***	1
Start RIBFAT	mm	5.2	1.6	2	10	0.55***	0.44***	0.48***
Start RUMPFAT	mm	6.6	2.2	3	13	0.55***	0.36**	0.49***
Start IMF	%	4.8	0.7	2.8	6.2	0.34**	0.24†	0.28*
Start EMA	cm^2	59.1	4.5	50	70	0.03	0.44***	−0.04
End RIBFAT	mm	10.6	2.0	7	16	0.57***	0.55***	0.57***
End RUMPFAT	mm	14.1	2.8	9	22	0.59***	0.42**	0.59***
End IMF	%	6.5	0.6	5.1	7.7	0.21	0.19	0.28*
End EMA	cm^2	77.5	5.0	67	88	−0.09	0.37**	−0.11
End RIBFAT-WT	mm/kg	0.021	0.004	0.013	0.031	0.64***	0.28*	0.56***
End RUMPFAT-WT	mm/kg	0.028	0.006	0.015	0.043	0.63***	0.14	0.55***
End IMF-WT	%/kg	0.013	0.002	0.008	0.016	0.25*	−0.28*	0.20
End EMAWT	cm^2/kg	0.154	0.010	0.138	0.182	0.09	−0.46***	−0.11
Gain in RIBFAT	mm	5.4	1.3	3	9	0.22†	0.31*	0.29*
Gain in RUMPFAT	mm	7.5	1.6	4	11	0.29*	0.24†	0.38**

[1]Comprising 29 steers and 30 heifers.

[2]RFI_p-EBV_{mp} = midparent EBV for postweaning RFI; TWT = midtest BW; FCR = feed conversion ratio; RIBFAT = subcutaneous fat depth at the 12/13th rib; RUMPFAT = subcutaneous fat depth at the Australian P8 rump site; IMF = intramuscular fat content of the eye muscle; EMA = cross-sectional area of the eye muscle between the 12th and 13th rib at the start of test; RIBFAT-WT = RIBFAT divided by BW; RUMPFAT-WT = RUMPFAT divided by BW; IMF-WT = IMF divided by BW; EMA-WT = EMA divided by BW; RFI_f = RFI measured in the feedlot; RFI = residual feed intake; RFI_p = RFI measured postweaning.

†$P \leq 0.1$; *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

by a 0.076 kg/d (SE = 0.033; $P < 0.05$) increase in ADG, a 0.58 kg DM/d (0.17; $P < 0.01$) decrease in DMI, a 0.89 kg/kg (0.22; $P < 0.001$) decrease in FCR, and a 0.62 (0.12; $P < 0.001$) kg/d decrease in RFI_f. Phenotypic variation in RFI_f was independent of BW and ADG (as expected from the calculation of RFI_f), and lower RFI_f was associated with lower DMI and lower FCR. Of the scanned body composition traits, RIBFAT and RUMPFAT measured at the start and end of the test had strong positive correlations with RFI_p-EBV_{mp}, DMI, and RFI_f. RIBFAT and RUMPFAT relative to BW at the end of the test, and gain in these two fat traits over the test, were also positively correlated with RFI_p-EBV_{mp} and RFI_f. Gain in IMF and EMA over the RFI test was not correlated with either RFI_p-EBV_{mp} or RFI_f, but at the end of the RFI-test IMF was positively correlated with RFI_f, and IMF-WT was positively correlated with RFI_p-EBV_{mp}. The strongest association between genetic variation in RFI_p and change in one of these body composition traits was with RIBFAT-WT measured at the end of the RFI test ($r = 0.64$), with RFI_p-EBV_{mp} explaining 41% (0.64^2) of the variation in RIBFAT-WT at the end of the RFI test. Variation in RIBFAT-WT at the end of the RFI test explained 7.8% (0.28^2) of the variation in DMI and 32% (0.56^2) of the variation in RFI_f.

Behavioral and Physiological Traits

Descriptive statistics for temperament, movement, and digestive function traits measured over or at the end of the RFI_f test are presented in Table 2 and for traits measured in the animal house are presented in Table 3. In the RFI_f-test variation in RFI_p-EBV_{mp}, DMI and RFI_f were negatively correlated with FS meaning that animals with lower RFI (both genetic-RFI_p and phenotypic-RFI_f) and lower DMI exited the cattle crush faster than animals with higher RFI and DMI. Variation in RFI_p-EBV_{mp} explained 10% (-0.32^2) of the variation in

Table 2. Descriptive statistics for temperament, movement, and rumen function traits for a 10-wk feedlot RFI test and their correlations with midparent RFI_p-EBV, DMI and RFI for 59 Angus steers and heifers[1] bred to vary in genetic merit for postweaning RFI

						Correlations		
Trait	Units	Mean	SD	Min.	Max.	RFI_p-EBV_{mp}[2]	DMI	RFI_f
Temperament								
Crush score	1 to 5	2.6	0.5	*2.0*	3.8	−0.12	−0.18	−0.05
Flight speed	m/s	2.0	0.8	0.9	4.0	−0.32*	−0.29*	−0.22†
Movement								
Feeder visits	Counts/d	42.4	8.2	24.7	61.6	0.45***	0.23†	0.53***
Water visits	Counts/d	12.8	3.7	7.2	23.4	0.16	−0.19	−0.02
GEM visits	Counts/d	9.2	3.3	0.0	16.2	−0.27*	−0.09	−0.03
Distance	m/d	325	53	236	458	0.20	−0.10	0.12
Digestive function								
DMD	%	69.6	2.0	65.9	75.0	−0.13	−0.27*	−0.29*
Total VFA[3]	mM	77	24	29	132	−0.14	−0.17	−0.16
Acetate	mM	40	12	17	73	−0.08	−0.13	−0.13
Propionate	mM	26	11	5.4	52	−0.24†	−0.21	−0.17
Butyrate	mM	7.7	4.0	2.6	25	0.02	−0.04	−0.06
Isobutyrate	mM	0.46	0.21	0.13	1.05	−0.07	−0.08	−0.10
Valerate	mM	0.87	0.35	0.30	1.8	−0.31*	−0.38**	−0.35**
Isovalerate	mM	2.0	1.3	0.7	8.8	0.11	0.10	0.00
Acetate %	%	53	4.4	44	64	0.23†	0.22	0.13
Propionate %	%	33	7.4	14	44	−0.24†	−0.21	−0.09
Butyrate %	%	10	3.6	5.7	23	0.13	0.10	0.01
MPR[4]	g/d	144	16	118	190	0.06	0.28†	−0.05
MY[4]	g/kg	12.0	1.5	9.8	16.3	−0.23	−0.48**	−0.54***

[1]Comprising 29 steers and 30 heifers.

[2]Midparent EBV for RFI_p.

[3]N = 56 animals.

[4]N = 41 animals (comprising 22 steers and 19 heifers) with sufficient valid records obtained from the GEM.

RFI = residual feed intake; RFI_p = RFI measured postweaning; RFI_f = RFI measured in the feedlot; GEM = Greenfeed Emission Monitor; DMD = DM digestibility; MPR = methane production rate; MY = methane yield.

†$P \leq 0.1$; *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

FS and variation in FS explained 8.4% (-0.29^2) of the variation in DMI and 4.9% (-0.22^2) of the variation in RFI_f. There was no significant correlation for CS with RFI_p-EBV_{mp}, DMI, or RFI_f.

Of the four movement traits recorded over the RFI test, number of feeder visits had a strong positive correlation with both RFI_p-EBV_{mp} and RFI (Table 2). Variation in RFI_p-EBV_{mp} explained 20% (0.45^2) of variation in feeder visits and variation in feeder visits per day explained 5.2% (0.23^2) of the variation in DMI and 28% (0.53^2) of the variation in RFI_f. Feedlot DMI explained 5.2% ($P < 0.05$) of the variation in feeder visits, but even after accounting for this, RFI_p-EBV_{mp} still explained an additional 18% ($P < 0.001$) of the remaining variation in feeder visits, and feeder-visits 29% ($P < 0.001$) of the remaining variation in RFI_f. Neither the number of water visits or the estimate of minimum distance walked was associated with variation in RFI_p-EBV_{mp}, DMI, or RFI_f. There was a weak negative correlation between number of GEM visits and RFI_p-EBV_{mp} but no significant correlation with DMI or RFI_f.

Lower DMI and lower RFI_f over the feedlot test, but not lower RFI_p-EBV_{mp}, were associated with higher DMD and higher MY (Table 2). Lower RFI_p-EBV_{mp} and lower RFI_f were associated with higher MY on the roughage diet (Table 3). The value of $r = -0.29$ indicated that 8.2% of the variation in RFI_f could be explained by variation in DMD. On the feedlot-test ration higher DMD was associated with a higher MY ($r = 0.38$; $P < 0.05$). For the other rumen function trait measured: VFA, RFI_p-EBV_{mp} was negatively correlated with propionate in rumen fluid sampled at the end of the RFI test, both as a concentration and as a percentage of total VFA (Table 2), negatively correlated with valerate, present at a low concentration compared with

Table 3. Descriptive statistics for age, BW, feed intake, body composition, body temperature, gut function, and metabolic rate traits measured on 57 Angus steers and heifers[1] bred to vary in genetic merit for post-weaning RFI fed a restricted allowance of roughage ration in individual pens followed by fasting, and their correlations with midparent RFI_p-EBV and with feedlot-test DMI and RFI

						Correlations		
Trait	Units	Mean	SD	Min.	Max.	RFI_p-EBV_{mp}[1]	DMI	RFI_f[2]
TWT	kg	544	43	460	640	−0.12	0.54***	−0.07
DMI	kg/d	7.59	0.67	5.36	8.66	0.15	0.40**	0.04
Body composition								
RIBFAT	mm	11.3	2.0	7	18	0.51***	0.49***	0.53***
RUMPFAT	mm	16.4	3.2	10	24	0.57***	0.36**	0.58***
IMF	%	6.6	0.7	5.1	7.9	0.23†	0.07	0.14
EMA	cm^2	79.6	3.9	70	86	−0.19	0.30*	−0.15
RIBFAT-WT	mm/kg	0.021	0.004	0.012	0.032	0.56***	0.24†	0.54***
RUMPFAT-WT	mm/kg	0.030	0.006	0.016	0.042	0.60***	0.12	0.55***
Body protein	kg	78.5	6.8	64.9	96.7	−0.34*	0.35**	−0.26*
Body fat	kg	121	14.8	94.1	161	0.36**	0.66***	0.38**
Body temperature								
TEMP	°C	38.7	0.38	38.0	39.8	−0.17	−0.13	−0.19
RTEMP	°C	0.00	0.35	−0.63	0.94	−0.25†	−0.18	−0.31*
Digestive function								
MPR	g/d	144	18	99	195	−0.16	0.11	−0.37**
MY	g/kg	19.0	1.9	15.7	25.6	−0.31*	−0.20	−0.48***
Fed heat production								
CPR	g/d	4399	291	3566	5135	−0.20	0.40**	−0.10
HP	MJ/d	47.0	3.1	38.2	54.9	−0.20	0.41**	−0.09
HP-WT	MJ/kg	0.387	0.004	0.078	0.100	−0.06	−0.33*	−0.01
RHP	MJ/d	0	2.3	−4.8	5.4	−0.36**	0.19	−0.15
HP-EBP	MJ/kg	0.627	0.030	0.581	0.703	0.29*	−0.08	0.34*
Unfed heat production								
CPR	g/d	3027	255	2563	3644	−0.46***	0.17	−0.25†
HP	MJ/d	35.4	3.0	30.0	42.7	−0.46***	0.17	−0.25†
HP-WT	MJ/kg	0.065	0.005	0.054	0.08	−0.35**	−0.37**	−0.21
HP-EBP	MJ/kg	0.472	0.034	0.391	0.561	−0.16	−0.24†	−0.01

[1]Comprising 28 steers and 29 heifers.

[2]Midparent EBV for RFI_p.

RFI = residual feed intake; RFI_p = RFI measured postweaning; TWT = midtest BW; RIBFAT = subcutaneous fat depth at the 12th/13th rib; RUMPFAT = subcutaneous fat depth at the Australian P8 rump site; IMF = intramuscular fat content of the eye muscle; EMA = cross-sectional area of the eye muscle between the 12th and 13th rib at the start of test; RIBFAT-WT = RIBFAT divided by BW; RUMPFAT-WT = RUMPFAT divided by BW; TEMP = deep rectal temperature; RTEMP = residual TEMP calculated by DMI; MPR = methane production rate; MY = methane yield; CPR = carbon dioxide production rate; HP = heat production; HP-WT = HP divided by TWT; HP-EBP = HP divided by empty body protein weight; RHP = residual heat production calculated from DMI.; RFI_f = RFI measured in the feedlot.

†$P \leq 0.1$; *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

other VFA, and positively correlated with acetate as a percentage of total VFA. Only valerate was correlated with DMI or RFI_f.

Body temperature (TEMP) measured on restricted roughage feeding was not correlated with RFI_p-EBV_{mp} or RFI_f, but RTEMP was negatively correlated with both RFI_p-EBV_{mp} and RFI_f (Table 3). This meant cattle that were genetically lower for RFI_p or that had demonstrated lower RFI_f in the feedlot test had a higher energy production than expected for their feed intake. Variation in RTEMP explained 9.6% (-0.31^2) of the phenotypic variation in RFI_f.

Hematological, Stress Responsiveness, and Immune Competence

Descriptive statistics for hematological, stress responsiveness, and immune competence traits measured at the end of the RFI_f test are presented in Table 4. Of the hematological traits, WBC and LYM measured in blood at the end of the RFI test had strong negative correlations with RFI_p-EBV_{mp}, DMI, and RFI_f meaning that lower values for the three traits were associated with higher levels of WBC and LYM. The directions of the correlations for the other WBC types: NEU, MONO, EOS,

Table 4. Descriptive statistics for hematological, stress responsiveness, and immune competence traits measured on 59 Angus steers and heifers[1] bred to vary in genetic merit for postweaning RFI at the end of their 10-wk feedlot RFI test while still on the feedlot test ration ad libitum, and their correlations with midparent RFI_P-EBV and with feedlot-test DMI and RFI

						Correlations		
Trait	Units	Mean	SD	Min.	Max.	RFI_p-EBV_{mp}[2]	DMI	RFI_f
Hematology								
White blood cells	10^6/mL	9.67	1.78	5.78	14.59	−0.51***	−0.29*	−0.42***
Lymphocytes	10^6/mL	4.84	0.98	3.25	7.67	−0.43***	−0.34**	−0.54***
Neutrophils	10^6/mL	3.73	1.29	1.74	8.47	−0.31*	−0.12	−0.11
Monocytes	10^6/mL	0.76	0.19	0.35	1.24	−0.20	−0.14	−0.19
Eosinophils	10^6/mL	0.21	0.21	0.01	1.30	−0.21	0.02	−0.19
Basophils	10^6/mL	0.14	0.05	0.06	0.30	−0.14	−0.18	−0.15
Red blood cells	10^9/mL	7.37	0.63	6.12	8.56	−0.06	0.00	0.03
Hemoglobin	g/dL	13.6	1.1	11.4	15.9	−0.06	0.07	0.12
Hematocrit	%	42.0	3.0	35.0	48.0	−0.03	0.11	0.16
MCV	fL	57.2	4.2	46.2	66.3	−0.06	0.12	0.14
MCH	pg	18.5	1.3	15.3	21.1	0.02	0.09	0.11
MCHC	g/dL	32.3	0.9	29.6	34.1	−0.09	−0.09	−0.08
Platelets	10^6/mL	549	146	212	851	−0.09	−0.41**	−0.38**
%Lymphocyte	%	50.5	7.9	33.3	69.5	0.07	−0.12	−0.18
%Neutrophil	%	38.0	7.9	22.1	58.1	−0.10	0.09	0.18
Stress responsiveness								
ΔHAPT	mg/mL	0.08	0.39	−0.93	2.11	0.08	0.10	0.14
Immune competence								
AIR11	OD units	1.11	0.30	0.45	1.91	−0.04	−0.07	−0.15
AIR32	OD units	0.71	0.25	0.10	1.28	0.12	−0.02	−0.02
CIR-ctrl	mm	0.63	0.72	−0.93	2.27	−0.01	−0.20	−0.04
CIR test	mm	8.04	3.06	4.30	23.1	0.25†	0.32*	0.28*

[1]Comprising 29 steers and 30 heifers.

[2]Midparent EBV for postweaning RFI.

RFI = residual feed intake; RFI_p = %RFI measured postweaning; MCV = mean corpuscular volume; MCH = mean corpuscular hemoglobin; MCHC = mean corpuscular hemoglobin concentration; ΔHAPT = change in serum haptoglobin levels from day 0 to day 4 after the RFI test; AIR11 = antibody immune response reported as change in antibody level values at day 11 relative to day 0 after the RFI test; AIR32 = AIR reported as change in antibody level values at day 32 relative to day 0 after the RFI test; CIR ctrl = cellular immune response to saline control injection; CIR test = CIR relative to control in response to vaccine injection; RFI_f = RFI measured in the feedlot.

*$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

and BASO with the three traits were all negative but only the correlation for NEU with RFI_p-EBV_{mp} was statistically significant. Variation in RFI_p-EBV_{mp} explained 27% of the variation in WBC (-0.51^2) and 19% of the variation LYM (-0.43^2). The counts for LYM and NEU as percentages of total WBC were not correlated with either RFI trait implying that neither WBC type was differentially decreased as RFI-trait values increased. Variation in DMI and RFI_f was negatively correlated with WBC, LYM, and PLT and the latter three traits explained 8.4% (-0.29^2), 12% (-0.34^2), and 17% (-0.41^2), respectively, of the variation in DMI and 18% (-0.42^2), 29% (-0.54^2), and 14% (-0.38^2) of the variation in RFI_f. For the 15 hematological traits in Table 4, day after the end of the RFI test had a significant ($P < 0.05$) effect on: WBC, NEU, MONO, BASO, RBC, HGB, HCT, MCHC, PLT, and NEU% with values for each trait being highest on days: 11, 11, 4, 0, 32, 32, 32, 11, 11, and 11, respectively, after the end of the RFI test. The statistically significant negative associations between the three traits: WBC, LYM, and NEU with RFI_p-EBV_{mp} at the end of the RFI test shown in Table 4 continued to be significant ($P < 0.05$), and in addition the negative relationships for MONO and EOS became significant ($P < 0.05$). The statistically significant negative associations between the three traits: WBC, LYM, and PLT with DMI at day 0 shown in Table 4 continued to be significant ($P < 0.05$), and in addition the negative relationships for MONO and BASO became significant ($P < 0.05$). The statistically significant negative associations between the three traits: WBC, LYM, and PLT with RFI_f at day 0 shown in Table 4 continued to be significant ($P < 0.05$), and in addition,

the negative relationships for BASO and LYM%, and positive relationship for NEU%, with RFI_f became significant ($P < 0.05$). For all 15 traits, there was no significant ($P < 0.05$) interaction for their relationship with RFI_p-EBV_{mp}, DMI, or RFI_f and day of sampling confirming that the relationships shown in Table 4 were maintained for at least 1 mo following the RFI test and were not influenced by vaccination.

Stress responsiveness was measured as the change in HAPT following the stress imposed by animal handling at the end of the RFI test. Serum HAPT levels changed little, from a mean of −0.05 (SD 0.24) mg/mL on the day of handling, to 0.02 (SD 0.37) mg/mL measured 4 d later. The ΔHAPT (Table 4) was not associated ($P > 0.1$) with variation in RFI_p-EBV_{mp}, DMI, or RFI_f.

Immune competence was assessed using measures of AIR and CIR. Serum antibody levels increased from background levels of 0.41 (SD 0.21) OD units on the day of vaccination administration to 1.52 (0.35) OD units 11 d later and had begun to decline 32 d after vaccine administration (1.12 ± 0.30 OD units). There was no association ($P > 0.1$) between background levels with RFI_p-EBV_{mp}, DMI, or RFI_f ($r = -0.01$, −0.18, and −0.11, respectively). The increases above background levels in antibody levels at 11 and 32 d after vaccine administration (AIR11 and AIR32; Table 4) were also not associated ($P > 0.1$) with variation in RFI_p-EBV_{mp}, DMI, or RFI_f. When assessing CIR, a minor increase in skin-fold thickness was observed at the site of saline injection (CIR ctrl), from 5.6 (0.8) mm on the day of injection to 6.2 (1.0) mm measured 2 d later. Skin-fold thickness increased considerably after the 7in1 injection (CIR test), from 0.6 mm to 8.0 mm, and the magnitude of the responses were significantly positively correlated with RFI_p-EBV_{mp}, DMI, and RFI_f, meaning that vaccination induced smaller CIR in cattle with lower genetic RFI_p, lower DMI, or lower RFI_f. Variation in RFI_p-EBV_{mp} explained 6.4% ($r = 0.25^2$) of the variation in CIR test, and CIR test explained 10% (0.32^2) and 7.8% (0.28^2) of the variation in DMI and RFI_f, respectively.

Heat Production Traits

The traits: HP and HP-WT measured on the cattle when fed a restricted allowance of roughage diet were not correlated with RFI_p-EBV_{mp} or with RFI_f recorded in the feedlot test, but RHP was negatively correlated with RFI_p-EBV_{mp} (Table 3). These meant that cattle genetically lower for RFI_p were producing more heat relative to their feed intake than genetically higher RFI_p cattle. As reported above, genetically lower RFI_p was also associated with higher MY on the roughage ration, and the higher RHP is consistent with greater DMD and extraction of ME from the roughage ration by genetically lower RFI_p cattle. The strong positive associations for scanned fatness traits at the end of the feedlot with RFI_p-EBV_{mp} and RFI_f persisted at the roughage test. This variation in body composition may have also been associated with the observed variation in metabolic rate traits, so HP per kilogram of EBP (**HP-EBP**) was calculated to adjust for this variation in body composition and in recognition that body protein is generally considered to be more metabolically active than body fat. To test the association for HP with variation in body composition, HP was modeled against EBP and EBF measured at the roughage test. If fitted first, EBP explained 71% ($P < 0.001$) of the variation in HP, and EBF explained <1% ($P > 0.1$) of the remaining variation in HP. If EBF was fitted first it explained 8.2% ($P < 0.001$) of the variation in HP and EBP explained 63% ($P < 0.001$) of the remaining variation in HP. Both RFI_p-EBV_{mp} and RFI_f were positively correlated ($P < 0.05$) with HP-EBP meaning that cattle with lower values for either RFI trait were producing less heat per kilogram of EBP. During fasting, both HP and HP-WT were negatively correlated ($P < 0.01$) with RFI_p-EBV_{mp} meaning that cattle genetically lower for RFI_p were producing more heat than cattle genetically higher for RFI_p. Unfed HP was less strongly associated with variation in body composition than was fed HP, with EBP alone explaining 41% ($P < 0.001$) and EBF alone explaining <1% ($P > 0.1$) of the variation in unfed HP. In these unfed cattle there was no association of HP-EBP with RFI_p-EBV_{mp} or RFI_f.

Energy Budget

Descriptive statistics for BW, body composition, MER, Activity, and ER and HPgain for fat and protein for the cattle over the RFI_f test are given in Table 5. The EBP of animals at the start and end of the RFI test was negatively correlated with genetic variation in RFI (RFI_p-EBV_{mp}) but not RFI_f. Fat in the empty body (EBF) at the start and end of the RFI test was positively correlated with both RFI_p-EBV_{mp} and RFI_f. The mean values for MER calculated using either literature values or using data from this test were very close, but MER test was negatively correlated with RFI_p-EBV_{mp} reflecting the use of individual-animal unfed-HP in

Table 5. Descriptive statistics for BW, predicted body composition, energy requirements, and energy budgets for the 59 Angus steers and heifers[1] in the feedlot RFI test, and their correlations with midparent RFI_p-EBV and with feedlot-test DMI and RFI

						Correlations		
Trait	Units	Mean	SD	Min.	Max.	RFI_p-EBV_{mp}[2]	DMI	RFI_f
Start-WT	kg	410	36	342	499	−0.04	0.68***	0.00
Start-EBP	kg	62.5	5.9	50.8	80.7	−0.28*	0.50***	−0.17
Start-EBF	kg	60.8	11.4	34.8	90.8	0.47***	0.62***	0.41**
End-WT	kg	504	42	417	600	−0.14	0.64***	0.00
End-EBP	kg	70.1	6.2	57.8	88.1	−0.40**	0.44***	−0.21
End-EBF	kg	106	13.6	77.8	137	0.40**	0.75***	0.44***
Maintenance: MER lit	MJ/d	40.5	2.6	34.6	46.0	−0.14	0.62***	0.04
Maintenance: MER test	MJ/d	40.4	3.7	33.1	49.8	−0.36**	0.33*	−0.11
MER test WT	MJ/kg	0.089	0.006	0.075	0.102	−0.33*	−0.37**	−0.15
MER test Protein	MJ/kg	0.611	0.038	0.511	0.686	−0.07	−0.21	0.10
Activity lit	MJ/d	6.8	0.6	5.7	8.2	−0.09	0.67***	0.00
Activity test	MJ/d	7.6	0.6	6.4	9.2	−0.05	0.67***	0.03
ME for gain	MJ/d	75.7	10.5	52.8	102	0.55***	0.93***	0.84***
ME for gain ADG	MJ/kg	55.1	11.2	30.7	79.3	0.56***	0.47***	0.55***
ME for gain Protein	MJ/kg	793	357	327	1948	0.35**	0.32*	0.27*
ME requirement: MEI lit	MJ/d	122	13	92	155	−0.16	0.42**	0.09
ME requirement: MEI test	MJ/d	123	14	91	156	−0.21	0.37**	0.05
Actual ME intake	MJ/d	124	11	98	150	0.41**	1	0.74***

[1]Comprising 29 steers and 30 heifers.

[2]Midparent EBV for postweaning RFI.

RFI = residual feed intake; RFI_p = RFI measured postweaning; EBP = empty body protein weight; EBF = empty body fat weight; MER = maintenance energy requirement; MER lit = MER calculated based on literature equations; MER test = MER calculated using data from this current test; MER test WT=MER test/midtest BW; MER test protein=MER test/midtest EBP; Activity = energy used for activity; Activity lit = Activity calculated based on literature equations; Activity test = Activity calculated using data from this current test; ME for gain = ME for energy retained and heat production for gain in body protein and fat; ME for gain ADG=ME for gain/ADG; ME for gain Protein=ME for gain/gain in body protein; MEI = ME intake; MEI lit = MEI calculated based on literature equations; MEI test = MEI calculated using data from this current test; RFI_f = RFI measured in the feedlot.

*$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

the calculation of MER test. Calculation of MER lit assumed no between-animal variation in FM, and MER lit was not correlated with RFI_p-EBV_{mp}. Neither estimate of MER was correlated with RFI_f. The mean value for Activity test was slightly higher than that for Activity lit reflecting the addition of energy used for DIST walked in the calculation of Activity test. Neither activity estimate was correlated with RFI_p-EBV_{mp} or RFI_f. Gain in protein, ER in protein gain, and HP for protein gain were negatively correlated with RFI_p-EBV_{mp} but not RFI_f. Gain in fat, ER in fat gain, and HP for fat gain were not correlated with RFI_p-EBV_{mp} or RFI_f. The results show that the animals with lower RFI_p-EBV_{mp} had less fat and more protein in their bodies, but were predicted not to have a lower total MEI than animals with higher RFI_p-EBV_{mp}. The mean values for the two predictions of MEI were very close to the mean value for the actual measured MEI by the animals, and although the correlations between actual MEI with MEI lit and MEI test were positive, with r values of 0.42 and 0.37 (both $P < 0.01$), each explained only 17% and 13%, respectively, of the variation in actual MEI.

Sources of Variation in DMI, RFI, and MEI

The amount of variation in feedlot DMI, RFI, and MEI explained by BW, ADG, and traits in seven physiological categories is presented in Table 6. The latter seven traits were fitted in descending order by amount of variation in DMI explained by each trait separately (see Tables 1 to 4), meaning that RIBFAT at the end of the RFI test was fitted first and unfed HP was fitted last. Together TWT, ADG, and the seven traits explained 77.3% of the variation in DMI (or 76.2% if the two nonsignificant traits were excluded), with the fatness trait explaining 18% of the total variation, and with four other traits explaining smaller, but still statistically significant ($P < 0.1$), amounts of variation in DMI. FS and unfed HP no longer explained

Table 6. Variance in feedlot DMI, RFI, and MEI and the incremental percentage of variation explained by selected traits[1] within physiological categories after attributing variation explained by the preceding traits, the cumulative percentage of variation explained, and the percentage of variance unexplained. Data is for 59 Angus steers and heifers[2] bred to vary in genetic merit for postweaning RFI

Category and trait	Incremental and (cumulative) variation in DMI	Incremental and (cumulative) variation in RFI	Incremental and (cumulative) variation in MEI
Variance, kg/d or MJ/d	1.24	0.67	129
BW and gain			
Midtest $BW^{0.75}$, kg	44.7%*** (44.7%)	—	—
ADG, kg/d	1.4%^{ns} (46.1%)	—	—
Body composition			
End RIBFAT, mm	18.3%*** (64.4%)	32.7%*** (32.7%)	—
Midtest Protein, MJ	—	—	22.6%*** (22.6%)
Midtest Fat, MJ	—	—	39.7%*** (62.3%)
Movement			
Feeder visits, visits/d	5.7%** (70.0%)	11.7%** (44.4%)	—
Activity test, MJ/d	—	—	0.0%^{ns} (62.3%)
Temperament			
Flight speed, m/s	0.0%^{ns} (70.1%)	0.0%^{ns} (44.4%)	0.6%^{ns} (62.9%)
Digestive function			
DMD, %	2.4%* (72.5%)	3.6%† (48.0%)	2.6%† (65.5%)
Hematology			
WBC, counts	2.1%* (74.6%)	3.7%* (51.7%)	2.8%* (68.3%)
Immune competence			
CIR test, mm	1.6%† (76.2%)	3.1%† (54.8%)	1.4%† (70.1%)
Heat production			
Unfed HP-WT, kg	1.1%^{ns} (77.3%)	2.0%^{ns} (56.8%)	0.4%^{ns} (71.5%)
Variance unexplained, %	22.7%	43.2%	28.5%

[1]Within categories, the trait with the strongest significant correlation with feedlot DMI was selected – see Tables 1 to 4.

[2]Comprising 29 steers and 30 heifers.

RFI = residual feed intake; MEI = ME intake; End RIBFAT = subcutaneous fat depth at the 12th/13th rib at the end of the RFI test; DMD = digestibility of DM; WBC = white blood cell; CIR test = cellular immune response relative to saline control in response to vaccine injection; HP-WT = heat production divided by BW.

$^{ns}P > 0.1$; $^{\dagger}P \leq 0.1$; $^{*}P < 0.05$; $^{**}P < 0.01$; $^{***}P < 0.001$.

significant variation in DMI. Approximately 23% of the phenotypic variation in DMI remained unexplained.

RFI_f was fitted against the seven traits listed in Table 6 in the same descending order as above for feedlot DMI. Together, the seven traits explained 57% of the variation in RFI_f (or 55% if the two nonsignificant traits were excluded), the fatness trait explaining 33%, and with four other traits explaining smaller, but still statistically significant ($P < 0.1$), amounts of the remaining variation in RFI_f. FS and unfed HP no longer explained significant variation in RFI_f. Approximately 43% of the phenotypic variation in RFI_f remained unexplained.

Feedlot MEI was fitted against traits that could be expressed in energy units or expected to be associated with ME requirements. The traits are listed in Table 6 and were fitted in the same descending order as above for DMI. Together the energy stored by midtest as fat and protein explained 62% of the variation in MEI, with three other traits explaining smaller, but still statistically significant ($P < 0.1$), amounts of the remaining variation in MEI. Together all the traits listed in Table 6 explained 72% of the variation in MEI (or 70% if the three nonsignificant traits were excluded). Activity, FS and unfed HP no longer explained significant variation in MEI. Approximately 28% of the phenotypic variation in feedlot MEI remained unexplained.

DISCUSSION

In the current experiment, the mean values for the two predictions of MEI for the cattle were very close to the mean value for the actual MEI by the animals. This confirmed that calculation of MEI based on known biological processes associated with maintenance, activity, and gain in body composition was able to predict the mean for actual MEI of the

cattle over the RFI test. Variation between animals in predicted MEI was present reflecting the differences between animals in BW and in body composition over the RFI test. However, prediction of MEI based on known biological processes was not able to explain most the observed variation in actual MEI and presumably because the prediction assumed no between-animal difference in the energetic efficiency of the processes of protein or fat gain, and for MEI lit also assumed no difference in individual maintenance requirement. In an earlier experiment on cattle from the same RFI_p divergent-selection lines, Lines et al. (2018) reported that, compared with low-RFI animals, the extra feed energy consumed by high-RFI animals could be attributed to ER as fat. That 100% of the variation in DMI in the current experiment could not be accounted for by BW and composition of gain is likely due to the difference in housing and feeding of cattle between the two experiments. The cattle in this current experiment were housed in feedlot pens, fed ad libitum and had a CV for DMI of 9.2%, being greater than the CV for their start BW of 8.8%. In the experiment of Lines et al. (2018), the cattle were housed in individual stalls and were individually fed a restricted ration formulated to provide ME equivalent to either 1.05 or 1.8 times each animals expected maintenance requirement. Even at the 1.8 times maintenance level of feeding the CV for DMI was ~6%, much less than the CV of 9% for start BW (both CV calculated from the SEM and means in Lines et al., 2018). Further, the cattle were housed and fed in individual small pens so energy expended on activity would have been minimal, with little opportunity to display any innate difference in feeding or locomotor behavior. It would therefore appear that the feedlot-fed cattle in the current experiment were able to express innate differences in appetite and activity that resulted in prediction of DMI based on expected requirements for maintenance and for gain in body tissue alone not explaining all the variation in their DMI.

In this current experiment, genetic variation in RFI_p was associated with variation in a number of physiological processes, notably in traits related to feeding behavior, temperament, digestive function, hematology, immune competence, and HP, and in turn these traits, with the exception of HP, contributed to variation in DMI and RFI_f over the RFI test. From an energetic perspective, differences in feeding behavior become important if they influence energy expenditure. There was a positive association for RFI (both genetic and phenotypic) with number of visits to the feed bins by cattle in the feedlot, but the increase in feeder visits associated with higher RFI was greater than that just associated with higher intake alone, and feed visits explained an additional variation in DMI and RFI_f not explained by BW and fatness. More time spent eating in high-RFI cattle is a consistent observation over many experiments, but associations between RFI and other feeding and nonfeed related activities are less consistent (Kenny et al., 2018). Through not simply the case in this experiment, Cantalapiedra-Hijar et al. (2018) caution that many associations between feeding behavior and RFI reported in the literature may reflect differences in feed intake between low- and high-RFI cattle rather than being a source of variation in feed efficiency. The temperament trait, FS, while negatively correlated with RFI (both genetic and phenotypic), did not explain any additional variation in DMI or RFI_f beyond that already explained by BW, fatness, and feed visits. Significant negative genetic correlations between FS and RFI in beef cattle have been reported by Nkrumah et al. (2007) and Rolfe et al. (2011), but the same authors and Black et al. (2013) reported no significant phenotypic associations, leading Rolfe et al. (2011) to conclude that FS would not be recommended as an indicator trait for selection to change feed efficiency.

Cattle are less able than other ruminants in the ability to masticate whole grain (NRC, 1996) and grinding of samples of the feedlot ration before in vitro feed evaluation likely resulted in the reported values for DMD being potential maximum values. Further, incomplete milling of grain during feedlot ration preparation can result in some passing through the gut of cattle undigested, with barley (the grain in the feedlot ration used in this experiment) being more resistant to digestion than other grains such as wheat and oats (Toland, 1976; Campling, 1991). The research feedlot used a roller mill to crack grain and in virtually every sample of faces collected from animals some undigested grain was present. A consequence was that the DMD and ME content of the feedlot ration appeared to be overestimated by the commercial feed testing service used, and a lower value for the ME content was used in calculations of results from the RFI test, as described above in the Methods section. In this current experiment, with the cattle offered a high-energy feedlot ration ad libitum, higher DMD accompanied both lower DMI and lower RFI_f. Higher MY is indicative of more feed energy being released during digestion due to increased fermentation of feed in the rumen and the negative association for MY with RFI on the feedlot and roughage rations provide additional evidence that

greater digestion of feed accompanied lower RFI. Variation in DMD explained additional variation in DMI and RFI_f not explained by BW, fatness, and feed visits. The evidence for an association between DMD and RFI from other experiments is inconsistent, with Cantalapiedra-Hijar et al. (2018) observing that low RFI cattle often exhibit an increase in whole-tract DMD, but Kenny et al. (2018) not finding consistent evidence of such an association from the literature they reviewed. Both sets of authors caution there is evidence that the higher DMD in low RFI cattle might be mostly the consequence of a lower DMI and slower rate of passage of digesta leading to a higher DMD, and not likely the opposite. Of the VFA examined, only valerate, which was present in rumen fluid at a much lower concentration than other VFA, show an association with RFI and DMI (both genetic and phenotypic). In their review, Kenny et al. (2018) found no consistent evidence for association between RFI and production and composition of VFA.

Difference in stress physiology between high and low-RFI cattle, both as a marker trait for RFI and a mechanism for difference in metabolic response, was reviewed by Kenny et al. (2018) and Cantalapiedra-Hijar et al. (2018) and both sets of authors found no consistent compelling experimental evidence to support either hypothesis. In the current experiment, the responses 4 d after the end-of-test handling procedures in HAPT levels, and RBC and WBC counts (short term, hours to days, and responses to stress: Richardson et al., 2002), above baseline levels were not significant. The result for ΔHAPT indicated that these procedures were not as stressful as the production procedures examined by Slocombe and Colditz (2005) who reported changes in HAPT levels more than 10-fold higher than the average value in this experiment.

Lymphocytes and the CIR contribute to adaptive immune system function which is activated in response to exposure to specific pathogens. The current experiment found that lower RFI (both phenotypic and genetic) associated with maintenance of higher levels of LYM over time following the RFI test, whereas a stronger CIR was induced by vaccination in higher RFI (both phenotypic and genetic) relative to lower RFI animals. In this experiment, variation in WBC and CIR explained additional variation in DMI, RFI_f, and MEI to that explained by BW, gain, body composition, movement, and temperament. The energy cost of mounting an immune response is comparable in magnitude to energy costs for reproduction and growth, and even the cost of maintaining an immune system is much larger than might be expected based on the absolute mass of the immune system which is such a minor contributor to body mass (Lochmiller and Deerenberg, 2000). These authors provide examples across a range of mammalian species of 10% to 30% increases in metabolic rate to support upregulation of the immune system, and improvements in growth rate 5% to 23% following administration of antibiotics in diets given to young growing farm animals. Lymphocytes have a lifetime in blood of weeks so there will be an ongoing energy and protein cost associated with synthesizing and maintaining LYM levels. There is also expected to be a nontrivial energy cost in mounting enhanced immune responses to pathogen challenges, as was observed for CIR in higher vs. lower-RFI cattle in the current experiment. These results suggest maintaining a higher LYM count is potentially an energy saving and more immediate strategy to combat infection in lower versus higher-RFI cattle. That a less-strong CIR was associated with lower RFI does support the contention of Rauw (2012) that selection for productivity traits can be associated with compromise in immune system response and robustness. However, there is experimental evidence that the converse need not be true, with selection for immune response being shown not to be associated with compromise in production (Wagter et al., 2003; Stoop et al., 2016).

There is emerging evidence that higher feed efficiency (low RFI) is characterized by both lower maintenance energy requirement and a higher partial efficiency of use of ME for growth (Cantalapiedra-Hijar et al., 2018). Fasting HP is a major determinant of maintenance energy requirement, and in this current experiment, unfed HP and HP-WT were negatively correlated with genetic variation in RFI_p and phenotypic RFI_f. Higher RTEMP associated with lower RFI (both genetic and phenotypic) meant cattle that genetically lower for RFI_p or that had demonstrated lower RFI_f in the feedlot test had a higher energy production than expected for their feed intake. These associations for HP disappeared when expressed per kilogram of body protein providing evidence that the apparent increase in maintenance energy requirement in genetically lower RFI_p cattle was due to their increase in lean body content. Lines et al. (2018) calculated HP for animals fed at ~105% of their expected MER from CO_2 entry-rate data and reported no significant difference in HP, either as MJ/d or MJ/kg $BW^{0.75}$/d, between high and low RFI selection-line heifers, even without correction for differences in subcutaneous fat depth between the lines. Together, the results from both experiments provide

no evidence of an association between HP during fasting or at near-maintenance levels of feeding with genetic variation in RFI_p in animals compared at similar body composition. Difference in protein metabolism has been postulated as a contributor to variation in RFI, with lower rates of protein turnover associated with lower RFI (Herd and Arthur, 2009; Cantalapiedra-Hijar et al., 2018). The experiment by Lines et al. (2018), using an AA tracer technique, found no significant difference between RFI selection lines either in protein metabolism or in the HP for protein metabolism at either of two levels of feeding (105% and 180% expected MER). This led them to conclude that change in the energetic efficiency of protein gain did not accompany divergent selection for RFI_p in this population of cattle. Variation in protein metabolism has been shown to accompany divergent selection for growth and other traits in domestic animals (reviewed by Oddy, 1999). In their review, Cantalapiedra-Hijar et al. (2018) remarked that there is a scarcity of protein turnover data obtained in beef cattle through isotopic methods in vivo, but concluded that most reported studies support a role for variation in protein turnover to difference in feed efficiency. Whether reduction in maintenance requirement or improved partial efficiency of growth, or both, more generally accompanies low RFI needs to be confirmed in future studies (Cantalapiedra-Hijar et al., 2018).

This current experiment with Angus cattle descendent from unique divergent selection lines genetic variation in RFI_p was associated with variation in feed efficiency, measured as either RFI or FCR, and strongly associated with variation in body composition traits. Consistent with the findings from reviews of a large number of published experiments (Cantalapiedra-Hijar et al., 2018; Kenny et al., 2018), variation in estimated body composition alone did not fully explain the observed variation in DM intake and ME intake in these cattle being fed ad libitum and able to display any innate difference in appetite, temperament, feeding behavior and activity in the feedlot. There was evidence for an unfavorable negative association for genetic variation in RFI_p with higher HP by animals when fasted, as a consequence of the higher body-protein content in the genetically lower RFI animals. These results support the growing body of evidence presented by Cantalapiedra-Hijar et al. (2018) that low RFI may be accompanied by lower maintenance requirement and/or higher partial efficiency of ME use for growth. The corollary that needs to be tested is that RFI as a trait for selection is more likely to result in improved feed efficiency for growing animals in environments where feed is plentiful (e.g., feedlots and high-quality abundant pastures) than in mature animals feeding at levels just above maintenance (e.g., cows in many extensive pasture-based enterprises).

Conflict of interest statement. None declared.

LITERATURE CITED

AFIA. 2014. AFIA – laboratory methods manual (version 8). https://www.afia.org.au/files/AFIALabManua_v8_rm.pdf. – [accessed 9 January 2019].

Aleri, J. W., B. C. Hine, M. F. Pyman, P. D. Mansell, W. J. Wales, B. Mallard, and A. D. Fisher. 2015. Assessing adaptive immune response phenotypes in Australian Holstein-Friesian heifers in a pasture-based production system. J. Anim. Sci. 93:3713–3721. doi:10.2527/jas.2015-9078

ARC. 1980. The nutrient requirements of ruminant livestock. Technical review by an Agricultural Research Council Working Party. Slough: Commonwealth Agricultural Bureaux.

Archer, J. A., E. C. Richardson, R. M. Herd, and P. F. Arthur. 1999. Potential for selection to improve efficiency of feed use in beef cattle: a review. Aust. J. Agric. Res. 50:147–161. doi:10.1071/A98075

Arthur, P. F., J. A. Archer, D. J. Johnston, R. M. Herd, E. C. Richardson, and P. F. Parnell. 2001. Genetic and phenotypic variance and covariance components for feed intake, feed efficiency, and other postweaning traits in Angus cattle. J. Anim. Sci. 79:2805–2811.

Arthur, P. F., I. M. Barchia, C. Weber, T. Bird-Gardiner, K. A. Donoghue, R. M. Herd, and R. S. Hegarty. 2017. Optimizing test procedures for estimating daily methane and carbon dioxide emissions in cattle using short-term breath measures. J. Anim. Sci. 95:645–656. doi:10.2527/jas.2016.0700

Barnett, M. C., N. A. Forster, G. A. Ray, L. L., C. N. Guppy, and R. S. Hegarty. 2016. Using portable X-ray fluorescence (pXRF) to determine fecal concentrations of non-absorbable digesta kinetic and digestibility markers in sheep and cattle. Anim. Feed Sci. Technol. 212:35–41. doi:10.1016/j.anifeedsci.2015.12.015

Black, T. E., K. M. Bischoff, V. R. Mercadante, G. H. Marquezini, N. Dilorenzo, C. C. Chase, Jr, S. W. Coleman, T. D. Maddock, and G. C. Lamb. 2013. Relationships among performance, residual feed intake, and temperament assessed in growing beef heifers and subsequently as 3-year-old, lactating beef cows. J. Anim. Sci. 91:2254–2263. doi:10.2527/jas.2012-5242

Blaxter, K. L., and J. L. Clapperton. 1965. Prediction of the amount of methane produced by ruminants. Br. J. Nutr. 19:511–522. doi:10.1079/BJN19650046

Blaxter, K. L., and F. W. Wainman. 1966. The fasting metabolism of cattle. Br. J. Nutr. 20:103–111. doi: 10.1079/BJN19660012

Brouwer, E. 1965. Report of sub-committee on constants and factors. In: K. Blaxter (ed.) 3rd symposium on energy metabolism. London, Troon, Scotland: Academic Press; p. 441–443.

Cafe, L. M., D. L. Robinson, D. M. Ferguson, B. L. McIntyre, G. H. Geesink, and P. L. Greenwood. 2011. Cattle temperament: persistence of assessments and associations with productivity, efficiency, carcass and meat quality traits. J. Anim. Sci. 89:1452–1465. doi:10.2527/jas.2010-3304

Campling, R. C. 1991. Processing cereal grains for cattle –a review. Livest. Prod. Sci. 28:223–234. doi:10.1016/0301-6226(91)90144-F

Cantalapiedra-Hijar, G., M. Abo-Ismail, G. E. Carstens, L. L. Guan, R. Hegarty, D. A. Kenny, M. McGee, G. Plastow, A. Relling, and I. Ortigues-Marty. 2018. Review: biological determinants of between-animal variation in feed efficiency of growing beef cattle. Animal. 12(s2):s321–s335. doi:10.1017/S1751731118001489

Exton, S. 2001. Testing beef cattle for net feed efficiency – standards manual. 2nd ed. http://www.dpi.nsw.gov.au/agriculture/livestock/beef/breeding/general/feed-efficiency. – [accessed 30 March 2001).

Fell, L. R., and M. R. Clarke. 1993. Behaviour of lot-fed cattle. In: Recent advances in animal nutrition, Armidale, Australia; p 107–116. http://livestocklibrary.com.au/handle/1234/19680

Finch, V. A. 1986. Body temperature in beef cattle: its control and relevance to production in the Tropics. J. Anim. Sci. 62(2):531–542. doi: 10.2527/jas1986.622531x

Herd, R. M., and P. F. Arthur. 2009. Physiological basis for residual feed intake. J. Anim. Sci. 87(14 Suppl.):E64–E71. doi:10.2527/jas.2008-1345

Herd, R. M., J. I. Velazco, P. F. Arthur, and R. F. Hegarty. 2016. Associations among methane emission traits measured in the feedlot and in respiration chambers in Angus cattle bred to vary in feed efficiency. J. Anim. Sci. 94:4882–4891. doi:10.2527/jas.2016-0613

Jeyaruban, M. G., D. J. Johnston, and H.-U. Graser. 2009. Genetic association of net feed intake measured at two stages with insulin-like growth factor-I, growth and ultrasound scanned traits in Angus cattle. Proc. Assoc. Advmt. Anim. Breed. Genet. 18:584–587. http://www.aaabg.org/proceedings/2009/jeyaruban584.pdf

Kenny, D. A., C. Fitzsimons, S. M. Waters, and M. McGee. 2018. Invited review: improving feed efficiency of beef cattle - the current state of the art and future challenges. Animal 12:1815–1826. doi:10.1017/S1751731118000976

Koch, R. M., L. A. Swiger, D. Chambers, and K. E. Gregory. 1963. Efficiency of feed use in beef cattle. J. Anim. Sci. 22:486–494. doi:10.2527/jas1963.222486x

Lea, J. M., D. D. O. Niemeyer, M. T. Reed, A. D. Fisher, and D. M. Ferguson. 2008. Development and validation of a simple technique for logging body temperature in free-ranging cattle. Aust. J. Exp. Agric. 48(7):741–745. doi: 10.1071/EA07422

Lines, D. S., C. D. K. Bottema, R. M. Herd, V. H. Oddy, and W. S. Pitchford. 2018. Selection for residual feed intake affects appetite and body composition rather than energetic efficiency. Anim. Prod. Sci. 58(1):175–184. doi:10.1071/AN13321

Lochmiller, R. L., and C. Deerenberg. 2000. Trade-offs in evolutionary immunology: just what is the cost of immunity? Oikos. 88(1):87–98.doi:10.1034/j.1600-0706.2000.880110.x

Nielsen, B. 1966. Regulation of body temperature and heat dissipation at different levels of energy-and heat production in man. Acta Physiol. Scand. 68(2):215–227. doi: 10.1111/j.1748-1716.1966.tb03420.x

Nkrumah, J. D., J. D. H. Crews, J. A. Basarab, M. A. Price, E. K. Okine, Z. Wang, C. Li, and S. S. Moore. 2007. Genetic and phenotypic relationships of feeding behavior and temperament with performance, feed efficiency, ultrasound, and carcass merit of beef cattle1. J. Anim. Sci. 85(10):2382–2390. doi: 10.2527/jas.2006–657

Nolan, J. V., R. S. Hegarty, J. Hegarty, I. R. Godwin, and R. Woodgate. 2010. Effects of dietary nitrate on fermentation, methane production and digesta kinetics in sheep. Anim. Prod. Sci.. 50:801–806. doi:10.1071/AN09211

NRC. 1996. Nutrient Requirements of Beef Cattle. 7th ed. Washington, DC: National Academy Press.

Oddy, V. H. 1999. Protein metabolism and nutrition in farm animals: an overview. In: G. E. Lobley, A. White, and J. C. Mackae, editors, Protein metabolism and nutrition. Wageningen, Netherlands: European Association for Animal Production; p. 9–23.

Rauw, W. M. 2012. Immune response from a resource allocation perspective. Front. Genet. 3:267. doi:10.3389/fgene.2012.00267

Richardson, E. C., R. M. Herd, I. G. Colditz, V. H. Oddy, J. A. Archer, and P. F. Arthur. 2002. Blood cell profiles of steer progeny from parents selected for and against residual feed intake. Aust. J. Exp. Agric. 42:901–908. doi:10.1071/EA01098

Robinson, D. L., and V. H. Oddy. 2004. Genetic parameters for feed efficiency, fatness, muscle area and feeding behaviour of feedlot finished beef cattle. Livest. Prod. Sci. 90(2):255–270. doi:10.1016/j.livprodsci.2004.06.011

Rolfe, K. M., W. M. Snelling, M. K. Nielsen, H. C. Freetly, C. L. Ferrell, and T. G. Jenkins. 2011. Genetic and phenotypic parameter estimates for feed intake and other traits in growing beef cattle, and opportunities for selection. J. Anim. Sci. 89:3452–3459. doi:10.2527/jas.2011-3961

SAS. 2012. SAS STAT software, version 9.4 of the SAS system for windows. Copyright © 2002–2012. Cary, NC: SAS Institute Inc.

SCA. 2007. Nutrient requirements of domesticated ruminants. Collingwood, Australia: CSIRO Publications.

Slocombe, L. L., and I. G. Colditz. 2005. Evaluating the stress of production in cattle using haptoglobin 5th international colloquium on animal acute phase proteins. Dublin, Ireland: Scientific Committee and Enterprise Ireland, Biotechnology Directorate; p. 30.

Slocombe, L. L., and I. G. Colditz. 2012. A method for determining the concentration of haptoglobin in cattle blood following haemolysis caused at collection. Res. Vet. Sci. 93(1):190–194. doi: 10.1016/j.rvsc.2011.05.012

Stoop, C. L., K. A. Thompson-Crispi, S. L. Cartwright, and B. A. Mallard. 2016. Short communication: variation in production parameters among Canadian Holstein cows classified as high, average, and low immune responders. J. Dairy Sci. 99:4870–4874. doi:10.3168/jds.2015-10145

Toland, P. C. 1976. The digestibility of wheat, barley or oat grain fed either whole or rolled at restricted levels with hay to steers. Aust. J. Exp. Agric. Anim. Husb. 16:71–75. doi:10.1071/EA9760071

Torres-Vázquez, J. A., J. H. J. van der Werf, and S. A. Clark. 2018. Genetic and phenotypic associations of feed efficiency with growth and carcass traits in Australian Angus cattle. J. Anim. Sci. 96:4521–4531. doi:10.1093/jas/sky325

Wagter, L. C., B. A. Mallard, B. N. Wilkie, K. E. Leslie, P. J. Boettcher, and J. C. Dekkers. 2003. The relationship between milk production and antibody response to ovalbumin during the peripartum period. J. Dairy Sci. 86:169–173. doi:10.3168/jds.S0022-0302(03)73597-8

Walmsley, B. J., M. J. McPhee, and V. H. Oddy. 2014. Development of the BeefSpecs fat calculator to assist decision making to increase compliance rates with beef carcass specifications. Anim. Prod. Sci. 54(12):2003–2010. doi: 10.1071/AN14611

Whitelaw, F. G. 1974. Measurement of energy expenditure in the grazing ruminant. Proc. Nutr. Soc. 33:163–172. doi: 10.1079/PNS19740030