THE EFFECT OF DIET QUALITY ON DRY MATTER INTAKE, GENE EXPRESSION, GREENHOUSE GAS EMISSIONS, AND PRODUCTION MEASURES IN MATURE BEEF COWS

By

AMANDA LEIGH HOLDER

Bachelor of Science in Agronomy and Agriculture Business
College of the Ozarks
Point Lookout, Missouri
2015

Master of Science in Crop Science
University of Arkansas
Fayetteville, Arkansas
2018

Submitted to the Faculty of the
Graduate College of the
Oklahoma State University
in partial fulfillment of
the requirements for
the Degree of
DOCTOR OF PHILOSOPHY
July, 2021

THE EFFECT OF DIET QUALITY ON DRY MATTER INTAKE, GENE EXPRESSION, GREENHOUSE GAS EMISSIONS, AND PRODUCTION MEASURES IN MATURE BEEF COWS

Dissertation Approved:

Dr. David Lalman

Dissertation Adviser

Dr. James Rogers

Dr. Ryon Walker

Dr. Megan Rolf

Dr. Carla Goad

Dedicated to my
Mom and Dad,
Tor and Ad

I couldn't have done it without your love and support.

And to my trial cows:
Sweetpea, Pumpkin, Roxanne, Babs,
Anna, Stella, Queen Latifah, Rene, Ginny, Susie,
Eisa, Show Cow, Henrietta, Eileen,
Baby and Louie

For the same reason.

Name: AMANDA HOLDER

Date of Degree: JULY, 2021

Title of Study: THE EFFECT OF DIET QUALITY ON DRY MATTER INTAKE, GENE EXPRESSION, GREENHOUSE GAS EMISSIONS, AND PRODUCTION MEASURES IN MATURE BEEF COWS

Major Field: ANIMAL SCIENCE

Abstract: The purpose of this research was to examine the effect that diet quality had on dry matter intake, gene expression, greenhouse gas emissions, and production measures in gestating, mature Angus beef cows. Two experiments were conducted in consecutive years using a total of 90 gestating, non-lactating cows. In each trial, cows were assigned to one of two diet sequences, concentrate-forage (CF) or forage-concentrate (FC), so that age, body weight, body condition score, and days pregnant were equal across sequence. The diet sequence refers to the order in which two diets, a forage-only diet (HAY) and a concentrate-based diet (MIX) were consumed during two intake periods. In the first study, skeletal muscle from the semitendinosus muscle was collected at initiation, midpoint, and termination of the study and examination of the transcriptome through RNA-sequencing used to identify differentially expressed genes associated with diet quality and with intake classification (high or low intake). In total, 259 differentially expressed genes were associated with diet quality. In the second study, greenhouse gas emissions data was collected using a GreenFeed Emissions Monitoring (GEM) system to evaluate the effect of diet quality and diet sequence on gas emissions. During the first intake period, gas emissions were significantly associated with dry matter intake regardless of diet or sequence, highlighting the viability of using gas emissions data as a proxy for collection of intake data on both a forage diet and a concentrate-based diet.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER I

REVIEW OF THE LITERATURE

Introduction

The beef industry produces a high quality protein product using mainly waste products and non-utilizable plant components. The beef industry consists of three sectors: cow-calf, stocker or backgrounding, and feedlot. In the first two sectors, cow-calf and stocker, the primary goal is to sell pounds of calf, while the feedlot sector places value on carcass quality in addition to carcass size and weight. With the goal of maximizing pounds of calf sold at the cow-calf and stocker stage, an emphasis within the beef industry has been placed on increasing growth and other output traits and cattle prices have increased accordingly in the past decades (Brooks, 2015). However, the emphasis on production traits often results in unintentional increases in inputs to support the increased production. Decreasing input traits is rarely a focus of operations in the cow-calf sector even though feed costs make up 63% of the annual cost of owning a cow (Miller et al., 2001). The cow-calf and stocker sectors rely primarily on forage consumption through grazing or harvested forage. Feed costs associated with the cow-calf sector represents 70-74% of the total feed energy cost of beef production (Gregory, 1972; Rotz et al., 2019). Selection for increased feed efficiency can dramatically reduce production costs while simultaneously reducing energy lost to greenhouse gas emission and reducing the operation’s carbon footprint (Basarab et al., 2013).

Factors Affecting Feed Intake

Feed intake (FI) is measured in dry matter intake (DMI) or as residual feed intake (RFI) and is an important component of improved efficiency and sustainability in the beef industry. Feed intake is a complex topic with many interconnected factors affecting intake in individual animals. Among the factors affecting FI are diet parameters, physical restrictions, neural and hormonal feedback, genetic potential, and abiotic stressors (NASEM, 2016). Herd and Arthur (2009) suggested that variation in RFI could be attributed to metabolism, protein turnover, and stress (37%), between-animal differences in digestibility (10%), fermentation and heat production (9%), physical activity level (9%), body condition and composition (5%), and feeding activity and patterns (2%).

Diet parameters are perhaps one of the easiest factors affecting FI to manipulate. Diet processing level, moisture content, quality or energy content, palatability and digestibility can all impact the amount of given feed or forage an animal can, or will, consume (Ingvartsen, 1994). A meta-analysis evaluating the effect of diet energy content on DMI by comparing 48 study means found that as energy content of a feed decreases, DMI increases as animals consume more feed to meet energy requirements (Arelovick et al., 2008). Arelovick et al. (2008) found that as total dietary NDF (%) increased from 7.5 to 35.5%, DMI (%BW) increased linearly. Feeds with lower energy content are typically roughages or high-fiber feeds that are relatively bulky and filling compared to grains and byproducts, which have smaller particle size and higher energy content. Feed intake on high roughage, low energy diets then are restricted by rumen capacity and passage rate of digesta (Baile, 1981; Ingvartsen, 1994). Stretch receptors in the rumen provide satiety signals to the brain to stop feeding when the rumen is full though the animal's energy requirements may not have been met (Forbes and Barrio, 1992). Some practices may be employed to reduce the filling effect that roughage has in a diet. Decreasing particle size and adding moisture to the diet have both been shown to increase palatability and passage rate of a diet (Mertens, 1994). Decreasing the particle size of hay in a mixed diet from 28.7 mm to 2.9 mm increased passage rate in the hindgut but not in the reticulorumen (Tafaj et al., 2001). A negative relationship between water content of the diet and dry matter intake has been reported (Felton and Devries, 2010). Passage rate of digesta is under the control of physical constraints as well as chemostatic regulation. High energy, concentrate-based diets are thought to be regulated by chemostatic and hormonal responses to the animal's energy requirements (Roche et al., 2008). Thus, FI should eventually plateau when diet energy content reaches a level capable of meeting the animal's energy requirements (NASEM, 2016). This phenomenon is well documented in beef cattle consuming concentrate-based diets containing little fibrous roughage (Weiss et al., 2017; Jeon et al., 2019). An increase in quality of diet ingredients is not always economically possible, but can influence diet digestibility. Practices such as protein supplementation in low-quality forage diets can improve forage utilization and digestibility (Köster et al., 1996). This increase in digestibility can, in turn, increase passage rate and increase voluntary FI.

When animals consume a high quality, concentrate-based diet, there is less need for rumination and passage rate increases as the digestibility of the diet increases (Weiss et al., 2017). On these diets, FI is regulated by the central nervous system rather than through rumen fill (Baile, 1981). Metabolically active tissues such as visceral organs and adipose tissue provide chemical and hormonal feedback that induces satiety (Roche et al., 2008). Ghrelin, a hormone known to stimulate hunger, has been reported to be released in response to energy intake but not necessarily DMI (Wertz-Lutz et al., 2010). Satiety hormones include cholecystokinin (CCK), glucagon-like peptide-1 (GLP-1) and peptide YY (PYY) which are all produced in the intestine and secreted in response to diet nutrient content. The satiety hormones decrease gastric emptying and passage rate. Illius and Jessop (1996) report that an individual animal's intake is dependent on the animal's age genetic potential, growth stage, and immune factors. There is variation in prioritization of excess nutrients and an animal's ability to store or dispose of the excess (Illius and Jessop, 1996).

Abiotic stressors also play a role in FI. Brobeek et al. (1948) proposed that animals eat to maintain body temperature and stop eating to avoid hyperthermia. Since then, a well-defined relationship between body temperature and FI has been reported (Kadzere et al., 2002). Rumen receptors have been shown to be sensitive to temperature as well as the presence of feed. Feed intake is increased in cold-stressed animals and decreased in animals experiencing heat stress (Young, 1981; Kadzere et al., 2002). Diet ingredients may even be manipulated to alleviate heat stress in ruminants in order to reduce negative effects of heat on FI (Baldwin et al., 1980). Baldwin et al. (1980) found that increased fat content of the diet allows cattle to maintain FI with less heat production than a typical diet.

The conclusion can be drawn that while FI may be correlated in diets varying in concentrate content, FI data on one diet or another may not be interchangeable due to a difference in regulation mechanisms. This is an important assumption due to the difference in diet types common in the cow-calf sector of the beef industry compared to the feedlot sector.

Selecting for high performance animals in one sector may be inadvertently selecting for neutral or even poorly performing animals in another sector of the beef industry.

Measuring Feed Intake

Direct measurement of FI in grazing animals is not possible, however, techniques to estimate intake in grazing animals have been developed. These techniques focus on determination of fecal output and diet digestibility and measure concentrations of internal or external markers in fecal matter (Lippke, 2002). In order to provide accurate estimations of intake, a marker, whether internal or external, must adhere to the following characteristics originally described by Faichney (1975):

1. strictly non-absorbable,
2. does not alter and is not altered by the GI tract or the microbial population,
3. is physically similar to the material it should mark,
4. and does not interfere with other analyses.

The most common external marker fitting these characteristics is titanium dioxide (TiO_2). Titanium dioxide has some key advantages over the previously popular chromium oxide (Cr_2O_3). Chromium oxide has been reported to have carcinogenic effects which exclude it from continued use as an external marker (Sedman et al., 2006). Some possible difficulties with the use of external marker collection in a grazing setting include dosing of the marker, effects of diurnal marker recovery, and accurate collection of fecal samples (Lippke, 2002). While grazing, total fecal collection is difficult and requires extensive time and labor resources and the fact that titanium dioxide moves independently through the GI tract means that it can be used to measure total intake; however, shows a diurnal pattern of recovery (Lippke, 2002). In comparison, internal markers, those indigestible portions of the diet, do not follow a diurnal pattern, since they are not dosed individually. Two commonly used internal markers include indigestible neutral detergent fiber (iNDF) and acid detergent insoluble ash (ADIA) (Cochran et al., 1986). Determination of estimated intake using internal markers relies on comparison of internal markers in forage samples and fecal samples. In a monoculture, forage samples may prove a reliable source for comparison, however, many pasture settings are not a monoculture and individual selectivity plays a significant role in animal intake meaning that simple forage samples may not be an accurate estimate of the animal's diet (Lippke, 2002). To evaluate individual preference in a pasture setting, the use of plant-wax hydrocarbons (alkanes) has grown in popularity as grazing markers (Mayes and Dove, 2000). Alkanes are naturally occurring indigestible long-chained fatty acid compounds found in the waxy cuticle of plants. Dove and Mayes (1996) reported individual patterns of alkane concentrations for plant species meaning that diet composition could be determined by evaluating alkane composition in fecal matter.

With the factors listed above in mind, feed intake may be more easily observed in a confined environment where the test diet can be uniform and controlled. In confinement settings, automated intake systems may be used to collect individual intake data on animals housed as a group or animals may be penned separately for individual FI data collection. Some drawbacks of confinement feeding, whether group housed or individually penned, include alteration of physical and social behavior related to feeding (Custodio et al., 2016; Overvest et al., 2018). In an individual housing situation, animals are offered access to the diet ad libitum and feed refusals weighed back at 12 or 24-hour intervals. In this setting, an accurate estimation of intake can be gathered in a shorter testing period as the animal has no competition for access to feed. However, individual housing and feeding reduces the social component of feeding that would take place in a grazing or group housed environment. Animals housed as a group experience some of the social and behavioral aspects of feeding; however, feeding behavior will still be altered as access to the feed intake units is undoubtedly limited by how many animals each intake unit accommodates. The most common automated feeding systems are GrowSafe® System (Vytelle, Edmonton, Alberta), Insentec System (Onsentec, B.V., Marknesse, Netherlands), and SmartFeed (C-Lock Inc., Rapid City, South Dakota). Each of these systems are capable of measuring individual intake and feeding data in group-housed cattle by reading an electronic identification tag in the animal's ear or neck collar and assigning disappearance of feed to individual animals at each feeding event.

Feed Efficiency

Feed efficiency is typically evaluated using a feed conversion ratio (FCR) or a calculated RFI value comparing units of gain to units of feed consumed. Using these two measures of feed efficiency, more efficient animals have a lower FCR or RFI value, meaning they consume less feed per unit of gain, and less feed efficient animals have a higher FCR or RFI and consume more feed per unit of gain. Feed efficiency is typically determined in a post-weaning performance test. The Beef Improvement Federation (BIF) set guidelines for diet energy content, animal age, and length of study to ensure uniformity in post-weaning intake studies. According to the guidelines put forth, animals should be between 240 and 390 days of age and not more than a 60-day difference in age of animals within the same cohort. The minimum allowable energy concentration is 2.4 Mcal ME/kg (roughly 67% TDN) on a dry matter basis. While this is the minimum allowance, many intake studies utilize diets with greater energy content (up to 70-74% TDN). These test diets may be typical in a feedlot setting, but are equivalent to, if not greater than, the peak energy content of lush, vegetative forage in a pasture setting (Lahart et al., 2020). The average beef cow in the U.S. spends 8-10 months of the year consuming hay and grazing moderate to low quality forage ranging in TDN from 48-60%. This discrepancy between test diet and actual pasture diet can confound results related to efficiency of feed utilization. Results from recent studies report a moderate correlation between concentrate-based diet intake and forage-based diet intake (Cassady, 2016b; Martin et al., 2019; Foote et al., 2017). The strength of the correlation between "forage" and "concentrate" or higher-quality diets seems to be related to the difference or similarity of processing, moisture and forage quality between the diets in question. These correlations suggest that selection for reduced intake in animals consuming a concentrate-based diet may result in a modest reduction in low-quality forage intake. However, there is a negative correlation reported in the performance of animals consuming a concentrate-based mixed diet and the performance of animals consuming a forage-based diet (Cassady, 2016; Foote, 2017). This is important because the cattle industry has focused on selection for "greater performance" for over 60 years based on ranking of animals for growth traits while consuming a concentrate-based diet. These data suggest that little if any improvement has been made in efficiency of lower-quality forage utilization. The ability to identify cattle that perform well (gain well, produce adequate milk, and maintain body condition) while consuming less forage should lead to greater production efficiency.

Evaluating feed intake in a mature cow is challenging. Differences in environment, physiological differences, stage of production, and selection criteria of individual producers make uniformity across herds impossible. Annual changes in energy partitioning throughout the calving cycle affect intake in mature cows. Typically, a beef cow will partition energy first to basal metabolism (maintenance energy), followed by energy needed for activity, then necessary growth (bone, muscle, tissues necessary for life), and finally, production (Short et al., 1990). Within production requirements, energy will be partitioned first to pregnancy, followed by lactation, then energy reserves, and finally the estrous cycle (Short et al., 1990). The increase in cow size in some breeds may be due in part to heavier selection pressure for cows that would produce more milk and thus more pounds of calf at weaning. This may have increased the priority of nutrient partitioning to milk, though an increase in mature size would also increase the resources needed for basal metabolism and necessary growth of the animal indicating that those two goals be met first as listed in the hierarchy above (Short et al., 1990; Jenkins and Ferrell, 1992).

The beef cow's highest nutrient requirements are after parturition and just before breeding. During this time, the cow is approaching peak lactation, completing uterine involution, and is beginning to cycle. The energy requirements may be increased in fall-calving cows (compared to spring-calving) as this period typically coincides with a decrease in temperature. The lowest energy requirement for a beef cow is during the end of the second trimester and beginning of the third trimester as the only energy requirements during this time are maintenance and pregnancy. A fall-calving cow will typically be gestating and non-lactating during late spring and summer when forage quantity and quality are more than adequate to meet maintenance requirements. The increased energy available to the cow during this time leads to the cow retaining that energy as fat in preparation for calving and lactation in the fall. In comparison, the spring-calving cow will enter this gestation period during late fall when forage quality and quantity typically declines. Though this is the typical flow, in some animals or breeds, the priority of a few functions may be switched. For example, when the plane of nutrition increases during lactation, milk production also increases for many breeds. However, with Hereford cows the milk production may increase slightly, but this breed will start retaining the extra energy as body reserves while giving no additional milk (Jenkins and Ferrell, 1992).

Production traits in beef cattle are reported to be moderately heritable. Torres-Vazquez and Spangler (2016) reported heritabilities of 0.36 for yearling weight and 0.27 for intramuscular fat deposition. The current heritability reported by the American Angus Association for the milk EPD is 0.12, which encompasses all calf growth attributed to the dam. Dillard et al. (1978) reported a heritability of 0.44 for total milk yield in beef cattle. Therefore, selection for these traits will likely increase these performance and production traits. Maintenance energy requirements account for up to 75% of the total feed energy required by a mature cow and are defined as the amount of feed energy needed to result in zero net gain or loss of tissue in the animal’s body (Ferrell and Jenkins, 1985; NASEM, 2016). Variation in maintenance energy requirements exists between animals due to differences in mature weight, production potential, and visceral organ mass (Ferrell and Jenkins, 1985). Cattle with increased production (growth and milk) potential have increased maintenance energy requirements (Ferrell and Jenkins, 1985; Jenkins and Ferrell, 2007). In addition to variation in maintenance requirements due to production potential, variation in visceral organ mass may affect these requirements as well. Visceral organs make up 6-9% of an animal’s body weight but are responsible for 50% of the protein synthesis and for consuming up to 50% of oxygen intake (Chilliard et al., 1998). Increased visceral organ mass and production tissue mass (such as mammary tissue) leads to higher maintenance requirements in animals with high production potential. Increased production potential, and thus increased maintenance requirements, leads to a reduction in feed energy available for milk yield when the environment limits energy intake (Ferrell and Jenkins, 1984; Jenkins and Ferrell, 2007).

Ferrell and Jenkins (1984) evaluated energy requirements for four classifications of non-lactating crossbred cattle selected for growth and milk yield. The crossbred cows used were Angus x Hereford (moderate growth and moderate milk), Charolais x Hereford or Angus (high growth and moderate milk), Jersey x Hereford or Angus (moderate growth and high milk), or Simmental x Hereford or Angus (high growth and high milk). The cows were assigned to one of three energy intake levels (low, medium, or high) to represent different nutritional planes. Maintenance energy requirements were higher for animals with higher milk production potential than animals with low milk production potential, but did not differ due to growth potential alone. In addition, maintenance energy required in low milk potential cows during lactation was found to be 12% lower than in lactating cows with high milk potential (Montaño-Bermudez et al., 1990). Together, these two studies indicate than cows with higher milk potential require more energy during gestation and lactation than cows with lower milk potential.

In addition to the variation in FI attributed to increased production potential and energy content of the diet, stage of production and animal age also affect daily FI. Several recent studies reported moderate to strong phenotypic correlations for FI in beef heifers evaluated post-weaning and again as 3- or 5-year-old cows ranging from 0.57 to 0.78 when cattle were fed similar high-quality diets at each age (Cassady, 2016; Hardie, 2017). Across stages of production, correlations between post-weaning FI and lactating FI of mature cows ranged from 0.69 to 0.74 (Archer, 2002; Freetly, 2016). These correlations indicate that FI is repeatable across age and stage of production when similar are fed in each intake study. Repeatability of FI may be high on high-quality diets, however, studies are limited evaluating beef cattle consuming a moderate or low-quality diet. Black et al. (2013) evaluated 74 heifers in a post-weaning intake study and again as 3-year-old cows during their second lactation. During the study, heifers consumed a diet post-weaning composed of chopped bermudagrass hay (37.0%), corn gluten feed (35.0%), whole corn (16.0%), and cottonseed hulls (10.0%) and a mineral supplement and had a NE_g content of 0.9 Mcal/kg DM. The intake study in the same set of females as 3-year-olds was made up of bermudagrass silage (86.7%), dried distillers grains plus solubles (12.4%), and the same mineral supplement (0.9%) and had slightly lower energy content than the post-weaning study at 0.8 Mcal/kg NE_l, DM. In this study, a correlation of 0.63 was reported between the post-weaning stage and the lactating stage though the energy content of the diet during lactation was higher than a typical cow diet. In a similar study, Cassaday (2016) reported a correlation of 0.57 in a study evaluating 404 heifers over the course of 4 years in a post-weaning intake study and again as 5-year-old lactating cows. The diet in this study was closer to a typical cow diet consisting of grass or alfalfa hay (90.0%) and corn condensed distillers solubles (10.0%).

The ability to select for cows with reduced intake capable of maintaining an adequate level of production on a typical moderate to low-quality diet is a critical step toward improving profitability and sustainability in the beef industry. This ability would encourage the switch from evaluating productivity in terms of output traits to evaluating productivity in terms of input traits focusing primarily on FI.

Literature Cited

AAA. 2021. American Angus Association, Saint Joseph, MO.

Archer, J. A. 2002. Genetic variation in feed intake and efficiency of mature beef cows and relationships with postweaning measurments. 7th World Congress on Genetics Applied to Livestock Productino, Montpellier, France.

Arelovick, H. M., C. S. Abney, J. A. Vizcarra, and M. L. Galyean. 2008. Effects of dietary neutral detergent fiber on intakes of dry matter and net energy by dairy and beef cattle. The Professional Animal Scientist 24(5):375-383. doi: 10.15232/S1080-7446(15)30882-2

Baile, C. A. a. D.-F. M. A. 1981. Nature of Hunger and Satiety Control Systems in Ruminants. Journal of Dairy Science 64:1140-1152. doi: 10.3168/jds.S0022-0302(81)82693-8

Baldwin, R. L., N. E. Smith, J. Taylor, and M. Sharp. 1980. Manipulating Metabolic Parameters to Improve Growth Rate and Milk Secretion. Journal of Animal Science 51(6):1416-1428. doi: 10.2527/jas1981.5161416x

Basarab, J. A., K. A. Beauchemin, V. S. Baron, K. H. Ominski, L. L. Guan, S. P. Miller, and J. J. Crowley. 2013. Reducing GHG emissions through genetic improvement for feed efficiency: effects on economically important traits and enteric methane production. Animal 7(s2):303-315. doi: 10.1017/s1751731113000888

Black, T. E., K. M. Bischoff, V. R. G. Mercadante, G. H. L. Marquezini, N. Dilorenzo, C. C. Chase, 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. Journal of Animal Science 91(5):2254-2263. doi: 10.2527/jas.2012-5242

Brobeck, J. R. 1948. Food intake as a mechanism of temperature regulation. Yale J Biol Med 20(6):545-552.

Brooks, K. 2015. Annual and seasonal price patterns for cattle, University of Nebraska-Lincoln.

Cassady, C. J. 2016a. Effects of timing and duration of test period and diet type on intake and feed efficiency of Charolais-sired cattle. Journal of Animal Science 94(11):4748-4758. doi: doi:10.2527/jas.2016-0633.

Cassady, C. J. 2016b. Evaluation of intake and feed efficiency measures in beef cattle, University of Illinois, Urbana-ChampaignChilliard, Y., F. Bocquier, and M. Doreau. 1998. Digestive and metabolic adaptations of ruminants to undernutrition, and consequences on reproduction. Reproduction Nutrition Development 38(2):131-152. doi: 10.1051/rnd:19980201

Cochran, R. C., D. C. Adams, J. D. Wallace, and M. L. Galyean. 1986. Predicting Digestibility of Different Diets with Internal Markers: Evaluation of Four Potential Markers. Journal of Animal Science 63(5):1476-1483. doi: 10.2527/jas1986.6351476x

Custodio, S. A. s., M. P. P. Tomaz, D. A. L. Silva, R. Goulart, K. M. Dias, and E. R. Carvalho. 2016. Feeding Behavior of Beef Cattle Fed Different Forages and Housed in Individual or Collective Pens. Journal of Animal Behaviour and Biometeorology 5:20-28. doi: 10.14269/2318-1265/jabb.v5n1p20-28

Dillard, E. U., M. K. M. Yusuff, and O. W. Robison. 1978. Milk Production in Hereford Cows II. Heritabilities and Repeatabilities. Journal of Animal Science 47(1):137-141. doi: 10.2527/jas1978.471137x

Dove, H., and R. W. Mayes. 1996. Plant Wax Components: A New Approach to Estimating Intake and Diet Composition in Herbivores. The Journal of Nutrition 126(1):13-26. doi: 10.1093/jn/126.1.13

Faichney, G. J. 1975. The use of markers to partition digestion within the gastro-intestinal tract of ruminants. In: The 4th International Symposium on Ruminant Physiology, Sydney, NSW. p 277-291.

Felton, C. A., and T. J. Devries. 2010. Effect of water addition to a total mixed ration on feed temperature, feed intake, sorting behavior, and milk production of dairy cows. Journal of Dairy Science 93(6):2651-2660. doi: 10.3168/jds.2009-3009

Ferrell, C. L., and T. G. Jenkins. 1984. Energy Utilization by Mature, Nonpregnant, Nonlactating Cows of Different Types. Journal of Animal Science 58(1):234-243. doi: 10.2527/jas1984.581234x

Ferrell, C. L., and T. G. Jenkins. 1985. Cow Type and the Nutritional Environment: Nutritional Aspects. Journal of Animal Science 61(3):725-741. doi: 10.2527/jas1985.613725x

Foote, A. P., Richard, T., and Freetly, H. 2017. Changes in feed intake, growth, feed efficiency, and body composition of beef cattle fed forage then concentrate diets. Journal of Animal Science 95:64.

Forbes, J., and J. Barrio. 1992. Abdominal chemo- and mechanosensitivity in ruminants and its role in the control of food intake. Experimental Physiology 77(1):27-50. doi: 10.1113/expphysiol.1992.sp003581

Freetly, H. C., Huehn, L. A., Thallman, R. M., Snelling, W. M. 2016. Feed intake and production efficiency of beef cows Journal of Animal Science 94:114. (Abstract)

Gregory, K. E. 1972. Beef Cattle Type for Maximum Efficiency "Putting it all Together". Journal of Animal Science 34(5):881-884.

Hardie, L. 2017. The genetic basis and improvement of feed efficiency in lactating Holstein dairy cattle. Journal of Dairy Science 100(11):9061-9075. doi: doi:10.31274/etd-180810-5553

Herd, R. M., and P. F. Arthur. 2009. Physiological basis for residual feed intake1. Journal of Animal Science 87(suppl_14):E64-E71. doi: 10.2527/jas.2008-1345

Illius, A. W., and N. S. Jessop. 1996. Metabolic constraints on voluntary intake in ruminants. Journal of Animal Science 74(12):3052. doi: 10.2527/1996.74123052x

Ingvartsen, K. L. 1994. Models of voluntary food intake in cattle. Livestock Production Science 39(1):19-38. doi: https://doi.org/10.1016/0301-6226(94)90149-X

Jenkins, T. G., and C. L. Ferrell. 1992. Lactation characteristics of nine breeds of cattle fed various quantities of dietary energy. Journal of Animal Science 70(6):1652-1660. doi: 10.2527/1992.7061652x

Jenkins, T. G., and C. L. Ferrell. 2007. Daily dry matter intake to sustain body weight of mature, nonlactating, nonpregnant cows1. Journal of Animal Science 85(7):1787-1792. doi: 10.2527/jas.2006-678

Jeon, S., S. Jeong, M. Lee, J. Seo, D. K. Kam, J. H. Kim, J. Park, and S. Seo. 2019. Effects of reducing inclusion rate of roughages by changing roughage sources and concentrate types on intake, growth, rumen fermentation characteristics, and blood parameters of Hanwoo growing cattle (Bos Taurus coreanae). Asian-Australasian Journal of Animal Sciences 32(11):1705-1714. doi: 10.5713/ajas.19.0269

Kadzere, C. T., M. R. Murphy, N. Silanikove, and E. Maltz. 2002. Heat stress in lactating dairy cows: a review. Livestock Production Science 77(1):59-91. doi: 10.1016/s0301-6226(01)00330-x

Köster, H. H., R. C. Cochran, E. C. Titgemeyer, E. S. Vanzant, I. Abdelgadir, and G. St-Jean. 1996. Effect of increasing degradable intake protein on intake and digestion of low-quality, tallgrass-prairie forage by beef cows. Journal of Animal Science 74(10):2473. doi: 10.2527/1996.74102473x

Lahart, B., R. Prendiville, F. Buckley, E. Kennedy, S. B. Conroy, T. M. Boland, and M. McGee. 2020. The repeatability of feed intake and feed efficiency in beef cattle offered high-concentrate, grass silage and pasture-based diets. Animal 14(11):2288-2297. doi: 10.1017/s1751731120000853

Lippke, H. 2002. Estimation of Forage Intake by Ruminants on Pasture. Crop Science 42(3):869-872. doi: 10.2135/cropsci2002.8690

Mayes, R. W., and H. Dove. 2000. Measurement of dietary nutrient intake in free-ranging mammalian herbivores. Nutrition Research Reviews 13(1):107-138. doi: 10.1079/095442200108729025

Mertens, D. R. 1994. Regulation of Forage Intake, Forage Quality, Evaluation, and Utilization. p. 450-493.

Miller, A. J., D. B. Faulkner, R. K. Knipe, D. R. Strohbehn, D. F. Parrett, and L. L. Berger. 2001. Critical Control Points for Profitability in the Cow-Calf Enterprise. The Professional Animal Scientist 17(4):295-302. doi: 10.15232/s1080-7446(15)31643-0

Montaño-Bermudez, M., M. K. Nielsen, and G. H. Deutscher. 1990. Energy requirements for maintenance of crossbred beef cattle with different genetic potential for milk. Journal of Animal Science 68(8):2279-2288. doi: 10.2527/1990.6882279x

NASEM. 2016. Nutrient Requirements of Beef Cattle 8th ed. The National Academies Press, Washington, DC.

Overvest, M. A., R. E. Crossley, E. K. Miller-Cushon, and T. J. Devries. 2018. Social housing influences the behavior and feed intake of dairy calves during weaning. Journal of Dairy Science 101(9):8123-8134. doi: 10.3168/jds.2018-14465

Roche, J. R., D. Blache, J. K. Kay, D. R. Miller, A. J. Sheahan, and D. W. Miller. 2008. Neuroendocrine and physiological regulation of intake with particular reference to domesticated ruminant animals. Nutrition Research Reviews 21(2):207-234. doi: 10.1017/s0954422408138744

Rotz, C. A., S. Asem-Hiablie, S. Place, and G. Thoma. 2019. Environmental footprints of beef cattle production in the United States. Agricultural Systems 169:1-13. doi: 10.1016/j.agsy.2018.11.005

Sedman, R. M., J. Beaumont, T. A. McDonald, S. Reynolds, G. Krowech, and R. Howd. 2006. Review of the Evidence Regarding the Carcinogenicity of Hexavalent Chromium in Drinking Water. Journal of Environmental Science and Health, Part C 24(1):155-182. doi: 10.1080/10590500600614337

Short, R. E., R. A. Bellows, R. B. Staigmiller, J. G. Berardinelli, and E. E. Custer. 1990. Physiological mechanisms controlling anestrus and infertility in postpartum beef cattle2. Journal of Animal Science 68(3):799-816. doi: 10.2527/1990.683799x

Tafaj, M., H. Steingass, and W. Drochner. 2001. Influence of hay particle size at different concentrate and feeding levels on digestive processes and feed intake in ruminants. 2. passage, digestibility and feed intake. Archiv für Tierernaehrung 54(3):243-259. doi: 10.1080/17450390109381981

Torres-Vázquez, J. A., and M. L. Spangler. 2016. Genetic parameters for docility, weaning weight, yearling weight, and intramuscular fat percentage in Hereford cattle1. Journal of Animal Science 94(1):21-27. doi: 10.2527/jas.2015-9566

Weiss, C. P., W. W. Gentry, C. M. Meredith, B. E. Meyer, N. A. Cole, L. O. Tedeschi, F. T. McCollum, and J. S. Jennings. 2017. Effects of roughage inclusion and particle size on digestion and ruminal fermentation characteristics of beef steers1. Journal of Animal Science 95(4):1707-1714. doi: 10.2527/jas.2016.1330

Wertz-Lutz, A. E., J. S. Jennings, and J. A. Clapper. 2010. Plasma ghrelin concentrations of beef cattle consuming a similar amount of dietary energy supplied by different ingredients1. Journal of Animal Science 88(7):2289-2299. doi: 10.2527/jas.2009-2447

Young, B. A. 1981. Cold Stress as it Affects Animal Production. Journal of Animal Science 52(1):154-163. doi: 10.2527/jas1981.521154x

CHAPTER II

DIFFERENTIAL GENE EXPRESSION IN RESPONSE TO VARIATION IN DIET QUALITY CONSUMED BY MATURE ANGUS COWS

Abstract: The objective of this study was to determine the effect of diet type on feed intake and gene expression in mature Angus cows. Forty-eight gestating commercial Angus cows (708 ± 52 kg; 7 ± 0.75 years old) were assigned to one of two diet sequences, concentrate-forage (CF) or forage-concentrate (FC), representing the order in which the two diets were consumed. In the first period, two of the four pens were assigned to the CF sequence and two to the FC sequence. Each pen contained an automatic waterer as well as four GrowSafe® feed intake units (GrowSafe System Ltd., Airdire, Alberta). During the first period, two of the four pens were assigned to a processed hay diet (10.0% CP, 1.98 Mcal ME/kg DM) while the other two pens consumed a mixed ration consisting of 43.0% hay, 22.0% corn, 24.0% soybean hulls, and 11.0% supplement, DM basis (11.7% CP, DM basis and 2.43 Mcal ME/kg DM). Following a 14-day adaptation period, feed intake was recorded for 50 days. Subsequently, diet type was switched and followed by 14 days of adaptation to the new diet and 50 days of feed intake measurement. Intake and performance data were analyzed as a crossover study using the PROC MIXED procedure in SAS v9.4. Pearson correlation coefficients were determined between phenotypic traits. Dry matter intake (DMI) on the two diets were correlated at r = 0.84 and r = 0.69 ($P < 0.001$) in the CF and FC sequence, respectively. In total, RNA sequencing of semitendinosus muscle tissue from the 12 highest intake cows and 12 lowest intake cows identified 259 differentially expressed genes (DEGs) associated with diet quality. Using the DEGs, enriched biological processes associated with energy metabolism and lipid biosynthesis were identified using g:Profiler. The results of this study support claims that genes controlling intake of low quality hay diets differ from those moderating intake on high quality, energy-rich diets.

Key words: beef cows, gene expression, dry matter intake, diet quality

Introduction

Feed efficiency is an important part of improving profitability and sustainability of the beef industry especially considering that selection for improved beef production centered on heavier weaning weights and rapid post-weaning gain (Lalman et al., 2019). Recently, measurement of post-weaning feed intake has been used to characterize feed efficiency in growing animals consuming diets varying in energy content (Cassady, 2016a; Foote, 2017). The goal of these post-weaning performance tests is to identify animals with an improved gain-to-feed ratio. In addition to phenotyping, genome-wide association studies (GWAS) have identified moderate to large effect QTL associated with measures of feed efficiency including dry matter intake (DMI), average daily gain (ADG), residual feed intake (RFI), and metabolic body weight (MBW) (Rolf et al., 2012; Seabury et al., 2017). The various QTL associated with each trait are thought to be a part of a larger pathway leading to variation in feed efficiency between individual animals (Cantalapiedra-Hijar, 2018).

Feed intake mechanisms controlling intake are different for concentrate-based or forage-based diets (Mertens, 1994). Intake of low-quality forage is driven primarily by rumen capacity, whereas intake of concentrate-based diets is typically driven by hormonal or neural mechanisms (Allen, 1996; Roche et al., 2008). These fundamental differences suggest that the genes controlling intake in forage diets may differ from genes controlling intake in concentrate-based diets. RNA sequencing has identified differentially expressed genes associated with RFI (Weber et al., 2016; Salleh et al., 2017). The primary objective of this study was to identify potential feed intake regulatory genes and pathways through evaluation of the transcriptome of skeletal muscle using

RNA sequencing. Skeletal muscle from the semitendinosus muscle was chosen as the tissue for RNA sequencing as skeletal muscle is responsible for 25% of maintenance energy requirements (Kelly et al., 2011). The role of skeletal muscle, and specifically mitochondrial energy production, in feed efficiency has been the subject of previous RNA-sequencing studies (Tizioto et al., 2016; Weber et al., 2016). A secondary objective of this study was to evaluate effects of diet quality on feed intake and gene expression in animals identified as high or low intake.

Materials and Methods

Phenotyping and Animal Management

The procedures and protocols conduced in this study were approved by Oklahoma State University (OSU) Animal Care and Use Committee (#AG-19-1). The trial was conducted in the GrowSafe® drylot pens at the Noble Research Institute Oswalt Ranch near Ardmore, OK. Using genomic panel scores produced by the Neogen Igenity Beef platform, 48 non-lactating, spring-calving, commercial Angus cows (7.0 ± 0.75 yrs old) were chosen from a larger herd to participate in the current study. The study was set up as a crossover design with the population assigned to one of two diet sequences: concentrate-forage (CF) or forage-concentrate (FC). Prior to trial initiation, spring calves were weaned and the cows were managed as a group for 14-d while transitioning from lactating to non-lactating. Following the post-weaning adjustment period, cows were randomly assigned to one of four pens, 12 cows per pen, during the first period with pens balanced for RFI genomic score, age, initial body weight, and days pregnant at trial initiation. The pens were each equipped with four GrowSafe® intake units (Vytelle, Edmonton, Alberta), GrowSafe® Beef water unitys (Vytelle, Edmonton, Alberta), with a metal roof over the feed intake units. Two pens were initially assigned to the hay (HAY) diet consisting of 100% chopped grass hay (10% protein, 53% total digestible nutrients (TDN)) and the other two pens were initially assigned to a mixed ration (MIX) with 43% chopped grass hay from the same harvest lot and 57% concentrate (11.7% protein, 67% TDN; Table 1). Hay fed in both diets was ground with a Haybuster Balebuster (DuraTech Industries International, Inc. Jamestown, ND). The HAY diet had 10% water added to reduce dust and improve ease of delivery.

Feed was delivered three times daily at 0700h, 1200h, and 1600h to provide adequate access to the diet for all cows and to avoid the need to overfill feed bunks in the pens fed the HAY diet for both feeding periods. GrowSafe® feeders were equipped with nets to reduce feed spillage due to head tossing and wind. At trial initiation, cows were allowed a 14-d adaptation period to adapt to the diet, feeders, and pen cohorts followed by a minimum of 50-d where intake data was collected (period 1). On d 64, dietary treatments were switched, and the second adaption period was initiated. Period 2 was initiated on d 78 and continued for 50 d. Regardless of period, cows assigned to MIX were fed 50% HAY and 50% MIX during adaptation for 7 d followed by 100% mix for 7 d. During adaptation for period 2, cows assigned to HAY (previously assigned to MIX) were fed 50% HAY and 50% MIX for 7 d followed by 100% HAY for 7 d. Body weight (BW) and body condition score (BCS; 3-person panel score; 1-9 scale) data were collected on two consecutive days at 0700h at initiation (d 14 and d 15 for period 1 and d 64 and d 65 for period 2) and termination (d 64 and d 65 for period 1 and d 114 and 115 for period 2) of each period. Single-day BW data was collected once at the mid-point of the study during each period. Average daily gain (ADG) was calculated for cows in each period by averaging the 2-d body weights at initiation and termination of each period and dividing by the days on feed in each period. Back fat, rump fat, and longissimus dorsi muscle area were determined by ultrasound (Aloka 500-V, Corometrics Medical Systems, Wallingford, CT) at the initiation and termination of each period by trained personnel. Feed intake data was collected and filtered following standard GrowSafe protocols. Intake data was excluded from final calculations for days when cows were removed from the pen for biopsy or ultrasound data collection for a significant period of time.

Sample Collection, RNA Extraction, Library Construction and Sequencing

Muscle tissue was collected on days 0, 64, and 115 from the semitendinosus muscle of each cow. The sample (100-200mg) was divided into two subsamples and stored in RNA-Later for at least four hours before freezing at -80°C to preserve RNA integrity until extraction. Extraction of RNA was performed using the standard protocol for the Qiazol®, RNeasy Mini Kit and extracted RNA was resuspended in 20 µl of RNase-free water. The quality and concentration of total RNA was evaluated using a NanoDrop® ND-1000 spectrophotometer (Thermo Scientific, Wilmington, DE). Integrity of RNA samples was evaluated using an Agilent 2100 Bioanalyzer (Santa Clara, CA). The 260/280 ratio for samples ranged from 1.72 to 2.91 (0.22 SD) and mean RNA integrity number from 6.6 to 8.6 (0.46 SD). Seventy-two samples (three time-point samples from twenty-four cows) were analyzed with RNA-seq. Sequencing of total RNA and library construction was performed by Novogene Co., Ltd. in Beijing, China. The RNA samples were sequenced using an Illumina HiSeq 2500 machine (Gene Denovo

Biotechnology Co., Guangzhou, China) and on average, 21,331,080 paired reads per sample were generated.

Only samples with more than 10 million reads and a Phred score >30 were retained, this excluded four samples. Sequence reads were mapped to the bovine reference genome (ARS-UCD1.2). Reads were mapped with 80% similarity for 90% of their length. Differentially expressed genes (DEGs) were identified by the DESeq2 package version 1.32.0 (Love et al., 2014). Differentially expressed genes were considered at a False Discovery Rae (FDR) <5%. Functional enrichment analysis of the DEGs was performed using g:Profiler (Raudvere et al., 2019). Gene Ontology (GO) terms and the Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway enrichment were used to find significant enrichment in DEGs identified against the background of all expressed genes.

Statistical Analysis

All data were analyzed as a crossover design using PROC MIXED in SASv.9.4 (SAS Institute Inc., Cary, NC). Individual animal was the experimental unit for all phenotypic traits. Sequence, period, and diet were included in the model as fixed effects. Pen and cow within sequence were included as random effects. Pearson correlation coefficients were calculated using the PROC CORR procedure (SAS Institute Inc., Cary, NC) was used to determine relationships between performance measures and DMI. Relationships were considered significant when $P \leq 0.05$. Correlations were considered "strong" where coefficients were equal to or greater than 0.70, "moderate" where coefficients were between 0.30 and 0.69, and "weak" where values were equal to or less than 0.29 (Cassady, 2016b).

Results and Discussion

Least squares means for performance and feed efficiency within sequence and diet are found in Table 2. For all measured traits excluding DMI (kg/d and %BW), there was a significant diet by sequence interaction observed ($P < 0.05$), for this reason, results are presented by diet sequence. Daily DMI of MIX was 26% greater than HAY regardless of sequence ($P < 0.01$). In the current study, HAY DMI (kg/d) and MIX DMI (kg/d) were highly correlated at r = 0.84 ($P < 0.0001$) and r = 0.69 ($P = 0.0002$) for the CF and FC sequence, respectively (Table 3.). Cassady et al. (2016) reported a phenotypic correlation of 0.56 for DMI in Charolais-sired calves consuming first a forage-based ration followed by a finishing ration where the forage-based ration consisted of alfalfa haylage and corn silage and the finishing ration contained 25% corn silage and 75% corn concentrates. In a study evaluating mature Charolais cows, a phenotypic correlation of 0.36 and a genetic correlation 0.83 was observed between DMI on a hay diet and a silage diet (Martin, 2019). However, in the experiment by Martin (2019), cows consumed less silage dry matter than hay, suggesting that feed intake for the silage diet was limited by factors other than diet digestibility.

The diet x sequence interaction for ADG was caused by lower ADG during the second period, regardless of diet (interaction $P < 0.01$). The difference in ADG could be attributed to increased body fat composition at the beginning of the second period and therefore, greater energy required per kg of weight gain (Buskirk et al., 1992; Bruns et al., 2004; NASEM, 2016), advancing pregnancy requiring more nutrients per d for fetal growth and fetal tissue development (NASEM, 2016), increased energy required to cope with cold stress associated with winter months during intake period.

RNA-Seq was used to identify differentially expressed genes (DEGs) associated with intake when diet energy content varied. Least squares means for performance and feed efficiency within sequence and diet for cows for which RNA-seq libraries were generated are found in Table 5. To evaluate the effect of diet quality on gene expression, DMI on the MIX diet was compared to DMI on the HAY diet regardless of diet sequence or intake classification of cows. Of the 30,589 expressed genes evaluated, 259 were identified as significantly ($P < 0.05$) differentially expressed between the two diets. On the MIX diet, 119 genes were down-regulated and 140 up-regulated compared to the HAY diet. Of the significant DEGs, 10 genes showed a greater than 4 fold change in cows consuming the MIX diet compared to the HAY diet (Figure 1).

Biological processes affected by DEGs were investigated using an enrichment analysis of DEGs in response to diet quality. Using only the 10 DEGs with a fold change greater than 4, 8 significantly ($P < 0.05$) enriched biological processes were identified. The most significantly enriched processes identified are listed in Table 6 fell into two main categories: gas cycling or respiration and lipid synthesis. The genes associated with the gas cycling category were ‘hemoglobin, beta’ (HBB) and ‘hemoglobin alpha 2’ (HBA). The genes associated with lipid synthesis and adipogenesis were ‘leptin’ (LEP), ‘phosphoenolpyruvate carboxykinase 2’ (PCK2), ‘thyroid hormone responsive’ (THRSP), and ‘ELOVL fatty acid elongase 6’ (ELOVL6). The lipid biosynthetic process was significantly enriched in cows consuming the MIX diet across sequence. A 60-fold increase in expression of lipogenic transcription factor THRSP has been reported in Angus crossbred steers consuming high starch diets (Graugnard et al., 2009). An increase in THRSP expression is reported to be a result of increased intramuscular fat (IMF) content in beef cows (Schering et al., 2017). Thus, up-regulation of THRSP would be expected in cows consuming the MIX diet as body fat (both BF and IMF) increased in cows consuming MIX. In addition, the up-regulation of ELOVL6 has been previously reported as affecting lipid metabolism in bovine adipose cells (Junjvlieke et al., 2020). ELOVL6 is responsible for increasing concentrations of arachidonic acid and stearic acid and was up-regulated in beef cattle fed high-quality diets. The role of leptin (LEP) in controlling appetite is well documented (Giblin et al., 2010; Foote et al., 2015). A positive correlation exists between leptin serum concentration and both DMI and BF thickness indicating that animals consuming more feed have higher concentrations of circulating leptin (Kelly et al., 2010; Foote et al., 2015).

Four genes were identified as differentially expressed between high and low intake cows on the MIX diet and two genes were differentially expresses between intake classifications on the HAY diet. The four DEGs associated with the MIX diet were 'major histocompatibility complex class 1, A' (BoLA), 'ras-related GPR' (RRaD), 'MAS-related GPR' (MRGPRF), and 'serine peptidase inhibitor (SERPINE1). The three genes, BoLA, RRaD, and MRGPRF are reported to have functions in immune and inflammation response. In beef cattle, the enrichment of immune and inflammation pathways has been well documented in growing cattle consuming a diet high in concentrates (Paradis et al., 2015; Zarek et al., 2017). Previous studies evaluating gene expression in beef cattle found that cattle with improved feed efficiency expend less energy toward fighting the inflammation response to the diet and instead put that energy toward production (Alexandre et al., 2015; Paradis et al., 2015). The two DEGs associated with the HAY diet were 'integrin subunit alpha 2' (ITGA2) and 'meteorin-like, glial cell differentiation regulator' (METRNL). The gene, METRNL, was upregulated in high intake cows and is reported to promote energy expenditure within skeletal muscle and improve glucose tolerance (Rajesh et al., 2014).

Conclusively, this study highlights the need to evaluate feed efficiency in mature cows on a moderate to low-quality diet more typical of the nation's cowherd. There was a positive correlation observed between feed intake and a negative correlation observed for average daily gain on the two diets suggesting that selection for animals that perform well on the concentrate-based diet will result in animals with marginal performance in a grazing environment. Transcriptome profiling of high and low intake beef cows consuming diets with varying energy content was used to identify significant DEGs and functionally enriched biological processes. The DEGs identified for diet quality were primarily involved in lipid metabolism and synthesis while DEGs associated with feed intake were shown to be involved in immune function and inflammatory response.

Literature Cited

Alexandre, P. A., L. J. A. Kogelman, M. H. A. Santana, D. Passarelli, L. H. Pulz, P. Fantinato-Neto, P. L. Silva, P. R. Leme, R. F. Strefezzi, L. L. Coutinho, J. B. S. Ferraz, J. P. Eler, H. N. Kadarmideen, and H. Fukumasu. 2015. Liver transcriptomic networks reveal main biological processes associated with feed efficiency in beef cattle. BMC Genomics 16(1)doi: 10.1186/s12864-015-2292-8

Allen, M. S. 1996. Physical constraints on voluntary intake of forages by ruminants. Journal of Animal Science 74(12):3063. doi: 10.2527/1996.74123063x

Bruns, K. W., R. H. Pritchard, and D. L. Boggs. 2004. The relationships among body weight, body composition, and intramuscular fat content in steers. Journal of Animal Science 82(5):1315-1322. doi: 10.2527/2004.8251315x

Buskirk, D., R. Lemenager, and L. A. Horstman. 1992. Estimation of net energy requirements (NEm and NE delta) of lactating beef cows. Journal of animal science 70:3867-3876. doi: 10.2527/1992.70123867x

Cantalapiedra-Hijar, G. M. A.-I., G. Carstens, L. Guan, R. Hegarty, D. Kenny, and I. Ortigues-Marty. . 2018. Review: Biological determinants of between-animal variation in feed efficiency of growing beef cattle. Animal 12:321-335. doi: 10.1017/S1751731118001489

Cassady, C. J. 2016a. Effects of timing and duration of test period and diet type on intake and feed efficiency of Charolais-sired cattle. Journal of Animal Science 94(11):4748-4758. doi: doi:10.2527/jas.2016-0633.

Cassady, C. J. 2016b. Evaluation of intake and feed efficiency measures in beef cattle, University of Illinois, Urbana-Champaign.

Foote, A. P., K. E. Hales, L. A. Kuehn, D. H. Keisler, D. A. King, S. D. Shackelford, T. L. Wheeler, and H. C. Freetly. 2015. Relationship of leptin concentrations with feed intake, growth, and efficiency in finishing beef steers. Journal of Animal Science 93(9):4401-4407. doi: 10.2527/jas.2015-9339

Foote, A. P., Richard, T., and Freetly, H. 2017. Changes in feed intake, growth, feed efficiency, and body composition of beef cattle fed forage then concentrate diets. Journal of Animal Science 95:64.

Giblin, L., S. T. Butler, B. M. Kearney, S. M. Waters, M. J. Callanan, and D. P. Berry. 2010. Association of bovine leptin polymorphisms with energy output and energy storage traits in progeny tested Holstein-Friesian dairy cattle sires. BMC Genetics 11(1):73. doi: 10.1186/1471-2156-11-73

Graugnard, D. E., P. Piantoni, M. Bionaz, L. L. Berger, D. B. Faulkner, and J. J. Loor. 2009. Adipogenic and energy metabolism gene networks in longissimus lumborum during rapid post-weaning growth in Angus and Angus × Simmental cattle fed high-starch or low-starch diets. BMC Genomics 10(1):142. doi: 10.1186/1471-2164-10-142

Junjvlieke, Z., R. Khan, C. Mei, G. Cheng, S. Wang, S. H. A. Raza, J. Hong, X. Wang, W. Yang, and L. Zan. 2020. Effect of ELOVL6 on the lipid metabolism of bovine adipocytes. Genomics 112(3):2282-2290. doi: 10.1016/j.ygeno.2019.12.024

Kelly, A. K., M. McGee, D. H. Crews, A. G. Fahey, A. R. Wylie, and D. A. Kenny. 2010. Effect of divergence in residual feed intake on feeding behavior, blood metabolic variables, and body composition traits in growing beef heifers1. Journal of Animal Science 88(1):109-123. doi: 10.2527/jas.2009-2196

Kelly, A. K., S. M. Waters, M. McGee, R. G. Fonseca, C. Carberry, and D. A. Kenny. 2011. mRNA expression of genes regulating oxidative phosphorylation in the muscle of beef cattle divergently ranked on residual feed intake. Physiol Genomics 43(1):12-23. doi: 10.1152/physiolgenomics.00213.2009

Lalman, D. L., C. E. Andresen, C. L. Goad, L. Kriese-Anderson, M. E. King, and K. G. Odde. 2019. Weaning weight trends in the US beef cattle industry. Applied Animal Science 35(1):57-65. doi: 10.15232/aas.2018-01797

Love, M. I., W. Huber, and S. Anders. 2014. Moderated estimation of fold change and dispersion for RNA-seq data with DESeq2. Genome Biology 15(12)doi: 10.1186/s13059-014-0550-8

Martin, P., Taussat, S., Vinet, A., Krauss, D., Maupetit, D., and Renand, G. . 2019. Genetic parameters and genome-wide association study regarding feed efficiency and slaughter traits in Charolais cows. Journal of Animal Science 97:3684-3698. doi: 10.1093/jas/skz240

Mertens, D. R. 1994. Regulation of Forage Intake, Forage Quality, Evaluation, and Utilization. p. 450-493.

NASEM. 2016. Nutrient Requirements of Beef Cattle 8th ed. The National Academies Press, Washington, DC.

Paradis, F., S. Yue, J. R. Grant, P. Stothard, J. A. Basarab, and C. Fitzsimmons. 2015. Transcriptomic analysis by RNA sequencing reveals that hepatic interferon-induced genes may be associated with feed efficiency in beef heifers. Journal of Animal Science 93(7):3331-3341. doi: 10.2527/jas.2015-8975

Rajesh, Jonathan, James, Katrin, J. Lou, I. Lokurkar, Mark, Jorge, Christiane, James, Donny, J. Lachey, S. Gygi, J. Seehra, John, and Bruce. 2014. Meteorin-like Is a Hormone that Regulates Immune-Adipose Interactions to Increase Beige Fat Thermogenesis. Cell 157(6):1279-1291. doi: 10.1016/j.cell.2014.03.065

Raudvere, U., L. Kolberg, I. Kuzmin, T. Arak, P. Adler, H. Peterson, and J. Vilo. 2019. g:Profiler: a web server for functional enrichment analysis and conversions of gene lists (2019 update). Nucleic Acids Research 47(W1):W191-W198. doi: 10.1093/nar/gkz369

Roche, J. R., D. Blache, J. K. Kay, D. R. Miller, A. J. Sheahan, and D. W. Miller. 2008. Neuroendocrine and physiological regulation of intake with particular reference to domesticated ruminant animals. Nutrition Research Reviews 21(2):207-234. doi: 10.1017/s0954422408138744

Rolf, M. M., J. F. Taylor, R. D. Schnabel, S. D. McKay, M. C. McClure, S. L. Northcutt, M. S. Kerley, and R. L. Weaber. 2012. Genome-wide association analysis for feed efficiency in Angus cattle. Animal Genetics 43(4):367-374. doi: 10.1111/j.1365-2052.2011.02273.x

Salleh, M. S., G. Mazzoni, J. K. Höglund, D. W. Olijhoek, P. Lund, P. Løvendahl, and H. N. Kadarmideen. 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 Genomics 18(1)doi: 10.1186/s12864-017-3622-9

Schering, L., E. Albrecht, K. Komolka, C. Kühn, and S. Maak. 2017. Increased expression of thyroid hormone responsive protein (THRSP) is the result but not the cause of higher intramuscular fat content in cattle. International Journal of Biological Sciences 13(5):532-544. doi: 10.7150/ijbs.18775

Seabury, C. M., D. L. Oldeschulte, M. Saatchi, J. E. Beever, J. E. Decker, Y. A. Halley, E. K. Bhattarai, M. Molaei, H. C. Freetly, S. L. Hansen, H. Yampara-Iquise, K. A. Johnson, M. S. Kerley, J. Kim, D. D. Loy, E. Marques, H. L. Neibergs, R. D. Schnabel, D. W. Shike, M. L. Spangler, R. L. Weaber, D. J. Garrick, and J. F. Taylor. 2017. Genome-wide association study for feed efficiency and growth traits in U.S. beef cattle. BMC Genomics 18(1)doi: 10.1186/s12864-017-3754-y

Tizioto, P. C., L. L. Coutinho, P. S. N. Oliveira, A. S. M. Cesar, W. J. S. Diniz, A. O. Lima, M. I. Rocha, J. E. Decker, R. D. Schnabel, G. B. Mourão, R. R. Tullio, A. Zerlotini, J. F. Taylor, and L. C. A. Regitano. 2016. Gene expression differences in Longissimus muscle of Nelore steers genetically divergent for residual feed intake. Scientific Reports 6(1):39493. doi: 10.1038/srep39493

Weber, K. L., B. T. Welly, A. L. Van Eenennaam, A. E. Young, L. R. Porto-Neto, A. Reverter, and G. Rincon. 2016. Identification of Gene Networks for Residual Feed Intake in Angus Cattle Using Genomic Prediction and RNA-seq. PLOS ONE 11(3):e0152274. doi: 10.1371/journal.pone.0152274

Zarek, C. M., A. K. Lindholm-Perry, L. A. Kuehn, and H. C. Freetly. 2017. Differential expression of genes related to gain and intake in the liver of beef cattle. BMC Research Notes 10(1)doi: 10.1186/s13104-016-2345-3

Table 2.1. Composition of diets, %DM

Item	HAY	MIX
Bermudagrass hay	100.0	43.0
Rolled corn	-	22.0
Soybean hulls	-	24.0
Liquid feed	-	7.5
Mineral mix[1,2]	-	3.5
Analyzed Values		
Moisture (%)	9.0	19.0
TDN (%)	53.0	67.2
Crude protein (%)	10.0	11.7
NEm, Mcal/kg	1.1	1.6
ME, Mcal/kg	1.9	2.4

[1] Mineral mix consisted of cottonseed meal (45%), limestone (32.5%), salt (10%), sodium bicarbonate (7.5%), and molasses (5%)

[2] On the HAY diet, cows had free choice access to a commercial mineral

Table 2.2. Performance, intake, and efficiency response of gestating beef cows fed a forage only and forage-concentrate diet in a crossover feeding design[1]

Item	FC[2]	CF[2]	Diet[3]	Seq[3]	Diet*Seq[3]
No. of cows	24	22	-	-	-
BW, kg			<0.0001	0.0103	<0.0001
HAY	662.8 (9.4)^{a}	759.4 (10.1)^{b}			
MIX	734.4 (10.8)^{bc}	715.6 (10.5)^{c}			
BCS			<0.0001	0.0001	<0.0001
HAY	5.5 (0.10)^{a}	6.8 (0.12)^{b}			
MIX	6.4 (0.13)^{c}	6.5 (0.12)^{bc}			
ADG, kg/d			<0.0001	0.0462	<0.0001
HAY	0.80 (0.05)^{a}	0.09 (0.05)^{b}			
MIX	0.78 (0.05)^{a}	1.23 (0.09)^{c}			
ABF, cm			0.0018	0.3854	<0.0001
HAY	0.31 (0.03)^{a}	0.53 (0.04)^{bc}			
MIX	0.54 (0.04)^{b}	0.42 (0.04)^{abc}			
ARF, cm			0.0004	0.0045	<0.0001
HAY	0.39 (0.04)^{a}	0.69 (0.04)^{b}			
MIX	0.56 (0.04)^{c}	0.60 (0.04)^{bc}			
AIMF, %			0.3360	0.0462	0.0078
HAY	7.0 (0.23)^{a}	7.9 (0.28)^{b}			
MIX	7.1 (0.21)^{a}	7.6 (0.24)^{ab}			
DMI, kg/d			<0.0001	0.3400	0.0670
HAY	9.9 (0.50)^{a}	10.9 (0.46)^{a}			
MIX	14.6 (0.43)^{b}	14.8 (0.57)^{b}			
DMI, %BW			<0.0001	0.8857	0.0214
HAY	1.53 (0.0007)^{a}	1.45 (0.0007)^{a}			
MIX	2.01 (0.0005)^{b}	2.11 (0.0009)^{b}			
G:F			<0.0001	0.0003	<0.0001
HAY	0.07 (0.005)^{a}	0.01 (0.004)^{b}			
MIX	0.05 (0.003)^{c}	0.08 (0.005)^{a}			

[1] Except for number of cows, values in the table are mean ± (standard error).

[2] FC = forage-concentrate diet sequence; CF = concentrate-forage diet sequence

[3] *P*-values for effects of diet, sequence, and the interaction of diet and sequence.

[abc] Means within item followed by the same letter are not significantly different ($P < 0.05$)

Table 2.3. Correlations between performance, intake, and efficiency response for each diet during the first intake period. Above the diagonal are correlations for the CF sequence and below the diagonal are correlations for the FC sequence during period 1.

	BW	BCS	ADG	DMI	%BW	G:F	ABF	AIMF	ARF
BW		0.73***	-0.29	-0.09	-0.47*	-0.31	0.63**	0.07	0.61**
BCS	0.60**		-0.10	0.05	-0.26	-0.17	0.64**	0.33	0.72***
ADG	0.03	-0.03		0.61**	0.64**	0.85***	-0.40	0.12	-0.21
DMI	0.43	0.28	0.31		0.91***	0.13	-0.43*	0.13	0.00
%BW	0.15	0.11	0.34	0.95***		0.22	-0.64**	0.03	-0.23
G:F	-0.35	-0.32	0.68***	-0.44*	-0.36		-0.27	0.10	-0.31
ABF	0.38	0.73***	-0.20	-0.14	-0.27	-0.17		0.07	0.70***
AIMF	0.12	0.49*	-0.03	-0.05	-0.10	-0.05	0.53**		0.15
ARF	0.53**	0.73**	-0.06	0.00	-0.16	-0.12	0.82***	0.29	

$^{***}P < 0.001$, $^{**} P < 0.01$, $^{*} P < 0.05$

Table 2.4. Correlations between performance, intake, and efficiency response for each diet during the first intake period. Above the diagonal are correlations for the CF sequence and below the diagonal are correlations for the FC sequence during period 2.

	BW	BCS	ADG	DMI	%BW	G:F	ABF	AIMF	ARF
BW	.	0.64**	0.14	0.05	-0.33	0.13	0.53*	0.05	0.57**
BCS	0.62**	.	-0.04	0.10	-0.14	-0.05	0.65**	0.26	0.67***
ADG	-0.21	-0.06	.	0.13	0.05	0.99***	0.16	-0.10	-0.04
DMI	0.39	0.46*	0.03	.	0.92***	0.04	-0.10	-0.03	-0.04
%BW	-0.13	0.14	0.15	0.85***	.	-0.04	-0.32	-0.08	-0.25
G:F	-0.35	-0.26	0.86***	-0.47*	-0.31	.	0.18	-0.11	-0.05
ABF	0.35	0.51*	-0.05	0.17	-0.05	-0.14	.	0.15	0.15
AIMF	0.06	0.49*	0.20	0.17	0.09	0.08	0.31	.	-0.02
ARF	0.45*	0.58**	-0.26	0.17	-0.10	-0.31	0.66***	0.32	.

$^{***}P < 0.001$, $^{**}P < 0.01$, $^{*}P < 0.05$

Table 2.5. Performance, intake, and efficiency response for cows for which RNA-seq libraries were generated.[1]

Item	CF High[2]	CF Low[2]	FC High[2]	FC Low[2]
No. of cows	6	6	6	6
BW, kg				
HAY	758.0 (20.1)[a]	770.0 (17.8)[a]	671.0 (14.5)[b]	645.7 (24.6)[b]
MIX	696.8 (19.7)[a]	738.4 (17.7)[a]	750.2 (16.2)[a]	718.2 (27.8)[a]
BCS[4]				
HAY	6.8 (0.3)[a]	7.0 (0.3)[a]	5.6 (0.1)[b]	5.3 (0.3)[b]
MIX	6.3 (0.3)[a]	6.7 (0.2)[a]	6.8 (0.1)[a]	6.0 (0.3)[a]
ADG, kg/d				
HAY	0.3 (0.1)[a]	0.2 (0.1)[a]	1.1 (0.1)[b]	0.9 (0.1)[b]
MIX	1.7 (0.2)[a]	1.0 (0.2)[b]	1.0 (0.1)[b]	1.0 (0.1)[b]
ABF, cm				
HAY	0.5 (0.1)[ac]	0.6 (0.1)[a]	0.2 (0.0)[b]	0.3 (0.1)[bc]
MIX	0.3 (0.1)[a]	0.6 (0.1)[b]	0.4 (0.0)[ab]	0.5 (0.1)[b]
DMI, kg/d				
HAY	15.1 (1.0)[a]	10.9 (0.8)[b]	14.5 (0.6)[a]	8.7 (0.5)[c]
MIX	20.0 (0.8)[a]	13.6 (0.6)[a]	18.6 (0.5)[b]	14.6 (0.8)[b]
DMI, %BW				
HAY	2.0 (0.1)[a]	1.4 (0.1)[b]	2.2 (0.1)[a]	1.3 (0.1)[b]
MIX	2.9 (0.1)[a]	1.9 (0.1)[b]	2.5 (0.1)[c]	2.0 (0.1)[b]
G:F				
HAY	0.015 (0.01)[a]	0.016 (0.01)[a]	0.076 (0.01)[b]	0.098 (0.01)[b]
MIX	0.079 (0.01)[a]	0.072 (0.01)[a]	0.051 (0.01)[a]	0.065 (0.01)[a]

[1] Except for number of cows, values in the table are mean ± (standard error).

[2] CF high = concentrate-forage sequence, high intake; CF low = concentrate-forage sequence, low intake; FC high = forage-concentrate sequence, high intake; FC low = forage concentrate sequence, low intake

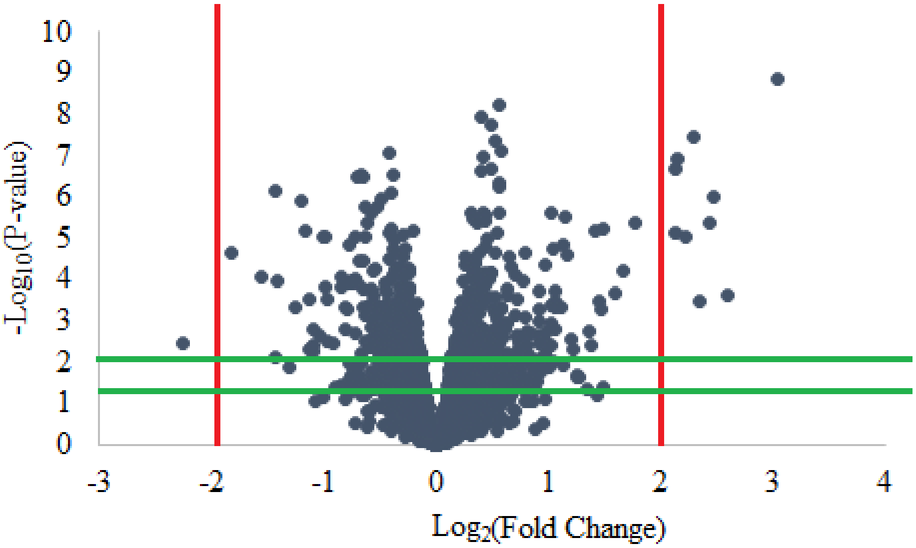
[abc] Means followed by the same letter within row are not significantly different ($P < 0.05$)

Table 2.6. Significantly enriched biological processes identified using the 10 most significantly up-regulated DEGs (10DEGs) and 259 DEGs (Total DEGs).[1]

Profiling Group	Biological Process	DEGs	*P* – Value
10DEGs	Hemoglobin complex	HBA, HBB	0.0014
	Oxygen binding and transport	HBA, HBB	0.0058
	Adipogenesis	PCK2, LEP	0.0301
	Lipid biosynthetic process	PCK2, LEP, ELOVL6, THRSP	0.0460
Total DEGs	Pentose phosphate pathway	TALDO1, TKT, G6PD	0.0027
	Lipid biosynthetic process	20+ genes	0.0030
	Glycerophospholipid biosynthetic pathway	15 + genes	0.0039

[1] 10 most significantly up-regulated DEGs are a subset of the 259 significant ($P < 0.05$) DEGs identified consisting of only genes with a > 4 fold change.

Figure 2.1. Volcano plot depicting a total of 30,589 genes expressed in muscle tissue from cows consuming diets varying in energy content. The vertical red lines at Log_2(fold change) of -2 and 2 represent gene expressions of 25% (Log_2(fold change) = -2) and 400% (Log_2(fold change) = 2) in concentrate-based diet (MIX) compared to the forage baseline diet (HAY). A positive fold change refers to genes up-regulated in cows consuming the MIX diet compared to the HAY diet, while a negative fold change refers to genes down-regulated in cows consuming the MIX diet. The green lines at $-\text{Log}_{10}(P\text{-Value}) = 1.3$ and 2 represent significance thresholds of $P < 0.05$ and $P < 0.01$, respectively.



CHAPTER III

EFFECTS OF DIET ON FEED INTAKE, WEIGHT CHANGE, AND GAS EMISSIONS IN MATURE ANGUS COWS

Abstract: The objective of this study was to examine the effects of diet energy density on ranking for dry matter intake (DMI), residual feed intake (RFI) and greenhouse gas emissions. Forty-two mature, gestating Angus cows (600 ± 69 kg BW; and BSC 5.3 ± 1.1) with a wide range in DMI EPD (-1.38 to 2.91) were randomly assigned to 2 diet sequences, forage-concentrate (FC) or concentrate-forage (CF), determined by the diet they consumed in each period (forage or concentrate). The forage diet consisted of long-stem native grass hay plus protein supplement (HAY). The concentrate diet consisted of 35% chopped grass hay and 65% concentrate feeds on a dry matter basis (MIX). The cows were adapted to the diet and the SmartFeed individual intake units for 14 days followed by a minimum of 52 days of intake data collection for each period. The GreenFeed Emission Monitoring system was used to determine CO_2, O_2, and CH_4 flux. Data were analyzed in a crossover design using a mixed model including diet, period, and sequence as fixed effects and pen and cow within sequence as random effects. For all measured traits excluding DMI, there was a diet by sequence interaction ($P < 0.05$). The correlation between MIX and HAY DMI was 0.41 ($P = 0.067$) and 0.47 ($P = 0.03$) for FC and CF sequences, respectively. There was no relationship ($P > 0.66$) between HAY and MIX average daily gain, regardless of sequence. Fifty seven percent of the variation in DMI was explained by metabolic BW, average daily gain (ADG), and body condition score for both diets during the first period. During the second period, the same three explanatory variables accounted for 38 and 37 percent of the variation in DMI for MIX and HAY diets, respectively. The negative relationship between body condition score and DMI was more pronounced when cows consumed the MIX diet. The correlation between MIX and HAY RFI was -0.20 ($P = 0.41$) and 0.31 ($P = 0.18$) for FC and CF sequences, respectively. During the first period, correlations for CO_2, CH_4, and O_2 with MIX DMI were 0.69, 0.81 and 0.56 ($P \leq 0.015$), respectively and 0.76, 0.74 and 0.64 ($P < 0.01$) with HAY DMI. During the second period, correlations for CO_2, CH_4, and O_2 with MIX DMI were 0.62, 0.47 and 0.56 ($P \leq 0.11$), respectively. However, HAY DMI during the second period was not related to gas flux ($P > 0.47$). Results from this experiment indicate that feed intake for energy-diverse diets are moderately correlated while ADG is not related. While further experimentation is necessary, gas flux data has potential as a proxy to determine relative feed intake in beef cows.

Key words: beef cows, greenhouse gas emissions, dry matter intake

Introduction

Identifying beef cattle that reach desired market composition and weight rapidly while efficiently converting nutrients to red meat yield is a research focus in the beef industry (Strydom, 2016). The growing interest in environmental and economic sustainability have resulted in increased selection for growth rate and feed efficiency and adoption of technologies for measuring feed intake and efficiency, and tools such as feed additives, and growth promoting implants (Capper, 2011; Cantalapiedra-Hijar, 2018). In the U.S., most cow/calf enterprises rely on grazed forage with substantial seasonal and year-to-year variation in availability and nutritive value (Drouillard, 2018). Harvested forage feeding and (or) concentrate supplementation is used to meet nutrient requirements when grazed forage availability or nutritive value is limited (Drouillard, 2018; Tedeschi et al., 2019). However, the negative relationship between total feed costs and particularly, purchased and harvested feed costs, to enterprise profitability is well documented (Ramsey et al., 2005; Mulliniks et al., 2015; Tonsor and Schulz, 2015). Considering that the cow-calf sector of the beef industry accounts for 74% of total feed energy utilized in the production of beef, efficiency of forage utilization by the cow/calf sector is of critical importance to overall efficiency of food production, security, and sustainability (Kenny et al., 2018).

Dry matter intake and feed efficiency traits are known to have moderate heritability in growing animals ranging from 0.39 to 0.84 (Rolfe et al., 2011; Mao et al., 2013; Retallick et al., 2017; Freetly et al., 2020). Average daily gain is also considered moderately heritable in growing animals with a heritability ranging from 0.26 to 0.53 (Rolfe et al., 2011; Freetly et al., 2020). While many feed intake studies revolve around growing animals in the feedlot sector, recent studies have documented the relationship between DMI and efficiency during post-weaning, an animal's growing phase and later as mature cows (Archer, 2002; Black et al., 2013; Cassady, 2016; Freetly, 2016; Freetly et al., 2020). However, the bulky and difficult to manage characteristics of a mature cow diet, namely long-stemmed grass or hay, poses an obstacle to collecting reliable information on intake and efficiency of mature cows in a typical forage grazing system.

Since direct measurement of DMI in grazing animals on pasture is not possible, indirect measurements of intake have become more popular and reliable. One method of indirect measurement is evaluation of greenhouse gas emissions data. The GreenFeed Emission Monitoring System (GEM; C-Lock Inc., Rapid City, SD) collects multiple short-term breath measures and estimates emissions of CO2 and CH4 and consumption of oxygen (O2). Using a GEM system, Arthur et al. (2018) reported a strong, significant correlation of 0.70 between DMI and CO2 production in beef heifers consuming a silage diet. Therefore, the objective of the current study was to examine the effects of diet quality on greenhouse gas emissions, DMI, and performance in mature cows consuming a high-quality concentrate-based diet or a low-quality hay. With moderate heritability for various feed intake and efficiency measures, considerable genetic improvement could be made by selecting for more efficient animals based on these measures.

Materials and Methods

The procedures and protocols conduced in this study were approved by Oklahoma State University (OSU) Animal Care and Use Committee (#AG-19-1). The study was conducted using the Kenneth and Caroline Eng Pens located at the Range Cow Research Center, North Range Unit near Stillwater, OK. Forty-two non-lactating, fall-calving, registered Angus cows (5.8 ± 2.2 yr old; 599.8 ± 68.8 kg BW) were included. All cows had previously had tissue samples submitted to the American Angus Association (Saint Joseph, MO) for parental verification and genomic testing on the AngusGS platform.

On d 0, cows (n = 21) were assigned to one of two diets (Table 1) at initiation of the first experimental period to create two blocks of equal average age, initial BW, and days pregnant. Five experimental pens were equipped with two SmartFeed individual intake units (SmartFeed, C-Lock Inc., Rapid City, South Dakota), one SmartFeed Pro individual intake unit (SmartFeed Pro, C-Lock Inc., Rapid City, South Dakota), automatic livestock watering tanks (MiraFount A3465, Miraco Automatic Livestock Waterers, Grinnell, Iowa), shade cloths, and windbreaks on the north and south perimeters. Three pens were designated as HAY treatment pens with hay baskets mounted on the SmartFeed units and the SmartFeed Pro unit used to deliver the protein supplement. Seven cows were assigned to each HAY pen based on initial BW, age, and dry matter intake expected progeny difference (**DMI EPD**). Two pens were designated as MIX pens with the MIX fed in the two SmartFeed units as well as the one SmartFeed Pro unit in each pen. Ten and eleven cows were assigned to the two MIX pens based on age, initial BW, and DMI EPD. The hay diet (**HAY**) consisted of unprocessed native grass hay supplemented with cottonseed meal. Supplement was dispensed through the SmartFeed Pro units set to limit supplement intake to 0.25% of BCS-5-adjusted body weight (**BW**; NASEM 2016). The mixed diet (**MIX**) contained processed hay from the same harvest lot as the HAY treatment, concentrate feeds, and a molasses-based liquid supplement (Table 1).

Cows were provided ad libitum access to their respective diets throughout the experiment. HAY feeders were filled daily at 0700 h, 1400 h, and 1900 h. Supplement was delivered once daily at 0700 h. Feeders designated for the MIX diet were filled daily at 0700 h and 1400 h and were equipped with a slotted wooden lid to minimize feed waste around the perimeter of the feed bunk. Each period began with a 14-d adaptation where cows acclimated to the individual intake units, pen cohorts, and the study diet. Period 1 began on d-1 and continued to d-67 with the first 14-d being adaptation. At the beginning of each study period, cows assigned to the MIX diet were stepped up from the previous diet to the MIX through three transition periods (Table 2). On d-68, the second 14-d adaptation period was initiated to transition cows to the opposite dietary treatment. Cows stepping up from the HAY diet to the MIX diet followed the transition diets outlined in Table 2, while cows stepping down from MIX to HAY had ad libitum access to HAY and were offered 50% of their previous week's MIX intake for the first 3-d followed by 25% of the MIX intake for 4-d. Period 2 was initiated on d-80 and continued through d-131.

Feed intake data were excluded from the final intake calculation for d with precipitation events and technical or mechanical difficulties resulting in insufficient feeding time for all animals. All feeders were cleaned out weekly or after a rain event and recalibrated according to manufacturer instructions. Round-baled native grass hay was weighed as a whole bale before feeding. HAY intake data were considered valid if 90% of the original bale weight was recovered through cow intake or in weighing back orts. In the same manner, the amount of MIX fed into each feeder was recorded from the mix wagon scale and compared to animal intake and orts. MIX intake data were considered valid if 95% of the original amount fed was recovered.

Body weight data were collected on consecutive days 0700 h at initiation and termination of each period. Body weight data was also collected at 14-d intervals for the duration of the study during each period. Body condition scores (scale = 1 to 9; (Wagner et al., 1988) were assigned at the initiation, midpoint, and termination of each period by two trained personnel. Back fat, rump fat, and longissimus dorsi muscle area were determined by ultrasound (Aloka 500-V, Corometrics Medical Systems, Wallingford, CT) at the initiation and termination of each period by trained personnel. Initial BW, final BW and average daily gain (ADG) was computed for each cow within period by regressing body weight on day of study (Ferrell and Jenkins, 1984).

The GreenFeed Emission Monitoring system (GEM) (GreenFeed, C-Lock Inc., Rapid City, South Dakota) was used to determine daily carbon dioxide (CO_2), methane (CH4) output, and daily oxygen (O_2) consumption. The GEM dispenses a small amount of pelleted feed and collects breath samples as the animal consumes the bait feed. The GEM was rotated through the pens after adaptation to the diet in each period spending no less than nine days in each pen. In order to achieve a diurnal usage pattern, animals were limited to four visits to the GEM in a 24-h period and could visit the GEM no sooner than four hours after the previous visit. As recommended by C-Lock Inc., only animals having at least 20 gas emissions records lasting a minimum of three minutes per record were included in the final dataset (Manafiazar et al., 2016; Gunter and Beck, 2018; Zimmerman, 2020). Gas emissions data was used to calculate heat production (HP) using the following equation (Pereira, 2015):

$$HP = [(4.96 + 16.07 \div RQ) \times CO2] \div 1{,}000$$

Where HP is heat production in MJ/d, RQ is the respiratory quotient calculated by dividing CO_2 production (l/d) by O_2 consumption (l/d), and CO_2 is carbon dioxide production in liters per day.

Statistical Analyses

Residual feed intake (RFI) was computed independently for each diet within sequence as the residual from mixed model regression (PROC MIXED; SAS Inst. Inc., Cary, NC) of DMI on ADG, period-average $BW^{0.75}$, and period-average BCS. Cow age and period-average days pregnant were included as random variables. Phenotypic data were analyzed as a crossover design using PROC MIXED in SASv.9.4 (SAS Institute Inc., Cary, NC). Individual animal was the experimental unit. Sequence, diet, and period were included in the model as fixed effects. Pen and cow within sequence were included as random effects. Pearson correlation coefficients were calculated using the PROC CORR procedure (SAS Institute Inc., Cary, NC) was used to determine relationships between performance measures, DMI, and gas emissions. Relationships were considered significant when $P \leq 0.05$. Correlations were considered “strong” where coefficients were equal to or greater than 0.70, “moderate” where coefficients were between 0.30 and 0.69, and “weak” where values were equal to or less than 0.29 (Cassady, 2016).

Results and Discussion

For all measured traits excluding DMI, there was a significant diet by sequence interaction observed ($P < 0.05$), and therefore, least squares means for performance and efficiency measures are presented by diet sequence (Table 3). The diet x sequence interaction for BW, BCS, and BF is due to lesser values for each trait during the first period and greater values for each trait during the second period regardless of diet. In contrast, the diet x sequence interaction for ADG and G:F is due to greater values for each trait during the first period and lesser values for each trait during the second period regardless of diet. These results suggest that cows experienced compensatory tissue gain during the first period regardless of diet. This may be a result of previous negative energy balance during lactation in combination with reduced energy requirement after calves were weaned and milk production ceased (NASEM, 2016).

In contrast to other performance traits shown in Table 3, mean daily DMI within diet did not differ by sequence ($P = 0.95$) and was 63% greater for MIX compared to HAY ($P < 0.01$). This agrees with the positive relationship between diet energy density and feed intake documented in several extensive reviews (NRC, 1996; Coleman et al., 2014). In the current experiment, hay used in the HAY diet was not processed although the hay used in the MIX diet was ground as previously described. Reducing forage particle size decreases the filling effect and results in increased forage DMI due to increased particle passage rate through the rumen (Galyean, 1993; Nasrollahi et al., 2015). The influence of passage rate and DMI are more pronounced for low-quality forages (Merchen, 1994; Minson, 1994). For this reason, the increased intake in the MIX treatment is likely due, in part, to the combined effects of diet digestibility and forage particle size.

Figure 3.1 shows the relationship of observed feed intake to predicted feed intake using Eq. 10-5 from NASEM (2016) for both sequences and both diets. The NASEM equation provided a reasonably accurate estimate of low-quality forage intake (HAY RMSD = 1.7 kg/d) although grossly underestimated MIX diet intake (MIX RMSD = 7.0

kg/d; intercept and slope differ from zero $P < 0.01$). Heat production calculated for CF cows consuming the MIX diet (278.8 kcal/kg MBW) was similar to HP reported by Pereira et al. (2015; 267.6 kcal/kg MBW) in mature Holstein cows consuming a corn silage and concentrate diet. Oxygen consumption is not reported often in the literature, however, in two recent papers reporting HP (MJ/d), O2 consumption ranged from 4.0 to 9.3 kg/d (Pereira, 2015; Caetano et al., 2018).

Several previous studies have evaluated the relationship between greenhouse gas emissions and DMI in growing beef steers (Velazco et al., 2017; Arthur, 2018; Caetano et al., 2018), yearling beef heifers (Fitzsimons et al., 2013; Manafiazar et al., 2016; Renand and Maupetit, 2016; Arthur et al., 2017; Gunter and Beck, 2018), and dairy cattle (Pereira, 2015). Gas emissions data for the current study is presented in Table 4. Pereira (2015) reported emissions of 13.6 kg/d and 0.5 kg/d for CO_2 and CH_4, respectively, and O_2 consumption of 9.3 kg/d in lactating dairy cows. Gas emissions for CF cows during the first period were comparable to these values at 14.3 kg/d, 0.5 kg/d, and 10.9 kg/d for CO_2, CH_4, and O_2, respectively. When presented on a g/kg MBW basis, each of the gas emissions parameters fell within the literature- established range. Carbon dioxide emissions of 74.7 and 105.6 g/kg MBW for FC and CF cows, respectively, were observed in the first study period. These values fell within observed CO_2 emissions ranging from 36.0 – 101.4 g/kg MBW (Pereira, 2015; Donoghue et al., 2020). In the current study, FC CH_4 production (2.3 g/kg MBW) during the first period was similar to previous studies with animals consuming grass hay (2.6 g/kg MBW, (Arthur et al., 2017)), pasture (2.3 g/kg MBW, (Velazco et al., 2017)), or grass silage (2.5 g/kg MBW, (Manafiazar et al., 2016)).

Heat production was calculated according to Pereira et al. (2015) with the amendment that rather than using a standard 0.95 for RQ, RQ was determined by dividing the amount of CO_2 (L/d) produced by the amount of O_2 consumed (L/d) for each cow (Manafiazar et al., 2016). A RQ of ~0.7 indicates an animal obtaining calories from fat metabolism, whether from the diet or from fat stores metabolized during negative energy balance (Kim, 2013). Since the animals in this study were neither fasted nor fed a concentrated fat diet, RQ approximating 1.0 was expected. Nevertheless, RQ values were lower for cows fed HAY in the FC compared to the CF sequence, whereas RQ for cows fed MIX was similar regardless of sequence (interaction $P < 0.01$). An explanation for greater RQ during CF is unclear although could be attributed to a reduction in cows voluntarily utilizing the GEM. Since CF cows during the second period are gaining only 0.38 kg/d, it is reasonable to assume that much of their weight gain is attributed to fetal and maternal tissue rather than an increase in body fat as they are within the last trimester of pregnancy during the second period (NASEM, 2016). This possible slight negative energy balance would mean that CF cows were utilizing fat stores in the second period, which would result in a lower RQ.

Equations used to predict DMI for the RFI calculation are shown in Table 5. Average daily gain, MBW, and BCS explained 57% of the variation in DMI for both the HAY and the MIX diet during the first period. In contrast, these three variables accounted for 37% and 38% of the variation in HAY DMI and MIX DMI, respectively, during the second period. Replacing BCS with period change in back fat did not improve the coefficient of determination in any of the four treatment combinations. Replacing BCS with period-average back fat resulted in increased coefficient of determination in only one of the four treatment combinations: MIX diet in the FC sequence (R^2 = 0.48; DMI, kg/d = 8.01 + 0.0942 x $BW^{0.75}$ + 2.853 x ADG – 5.167 x AvgBF).

Little has been published to quantify the effects of body composition on feed intake in beef cows. A negative relationship between BCS and DMI in beef cows is suggested (NRC, 1996). Fox et al. (1988) estimated DMI declined by 2.7% for each one percent increase in body fat composition for growing cattle within the range of 21.3 to 31.5% body fat (Fox et al., 1988). Assuming each unit gain in BCS is equivalent to 3.8% increase in body fat composition (NASEM, 2016), HAY diet DMI decreased by 1.5% and MIX diet DMI decreased by 2.4% for each one percent increase in body fat composition.

Several intake and efficiency studies have been conducted in growing animals fed a growing diet followed by a finishing diet (Durunna et al., 2011; Cassady et al., 2016; Russell et al., 2016; Foote et al., 2017; Lahart et al., 2020). In these studies, feed intake during the forage period was positively correlated to feed intake during the finishing period, ranging from r= 0.41 to 0.58 (Cassady et al., 2016; Lahart et al., 2020). In the current study, correlations of concentrate to forage intake fell within this established range. First period MIX DMI (kg/d) was correlated to second period HAY DMI (kg/d) at r= 0.47 (P = 0.03) and there was a tendency in the FC sequence for HAY DMI (kg/d) to be moderately correlated to MIX DMI (kg/d; r = 0.41, P = 0.07). Martin et al. (2019) reported a phenotypic correlation of 0.36 in mature Charolais cows consuming first a grass hay diet followed by a corn silage-based diet (Martin, 2019). Even though energy density, forage processing and moisture content were purposely dissimilar in the HAY and MIX dietary treatments, phenotypic correlations for DMI among diets were moderate and similar to previously reported values.

Two recent studies reported phenotypic correlations of -0.30 and -0.09 for ADG of animals consuming a concentrate-based mixed diet and the ADG of animals consuming a forage-based diet (Cassady et al, 2016; Foote et al., 2017, respectively). In agreement, we found no significant correlation for ADG regardless of sequence ($P >$ 0.66). The diet by sequence interaction for ADG was due to slower rates of weight gain during the second period regardless of diet. Increasing BW and body fat deposition are known to be negatively correlated with ADG (Bruns et al., 2004). While cows consuming MIX gained at a faster rate during the CF sequence, on average, ADG was similar to that that reported by Freetly et al. (2020; 1.54 kg) where cows were fed a diet with similar energy density. As expected, G:F was greater when cows consumed the MIX diet compared to the HAY diet, regardless of sequences. Residual feed intake calculated for FC on HAY was positively correlated to HAY CH_4 emissions (r = 0.67, P = 0.0011), but no significant correlation between RFI on either diet was observed between gas emissions for CF cows.

Caetano et al. (2018) reported a strong correlation between metabolizable energy intake and daily CO_2 production (r = 0.73) when cows consumed a chaff pellet and oat hay diet. Similarly, Arthur et al. (2018) reported a correlation of 0.84 between roughage DMI and CO_2 production. On the current study, a strong correlation (r = 0.76) falling between the established range was observed between HAY DMI in FC cows and CO_2 production. Arthur et al. (2018) also reported a correlation of 0.83 between concentrate-based DMI and CO_2 production which was higher than the r = 0.69 correlation observed between MIX DMI and CO_2 production in CF cows on the current study. Bird-Garner et al. (2017) reported a strong correlation between CH_4 production and DMI for forage-based diets and grain-based diets of r = 0.74 and r = 0.66, respectively. On the current study, strong correlations of r = 0.74 and r = 0.81 were observed between CH_4 and DMI on HAY and MIX during the first period. Because correlations between CO_2, O_2, and CH_4 and DMI values were strong and positive for both HAY and MIX diets during period 1, least squares regression equations were developed using gas flux and DMI values from both treatments during period 1. Each additional kg of feed intake was associated with a linear increase ($P < 0.01$) in CO_2, O_2, and CH_4 flux (0.51, 0.37 and 0.02 kg/d, respectively). In addition to previously reported correlations, data from the current study emphasize the potential for gas flux data to be used as a proxy for measuring DMI directly. The ability to use indirect intake measures such as the GEM system for selecting animals with increased forage utilization efficiency would greatly enhance the profitability and environmental sustainability of beef production.

Literature Cited

Archer, J. A. 2002. Genetic variation in feed intake and efficiency of mature beef cows and relationships with postweaning measurments 7th World Congress on Genetics Applied to Livestock Productino, Montpellier, France.

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 measures1,2. Journal of Animal Science 95(2):645-656. doi: 10.2527/jas.2016.0700

Arthur, P. F., Bird-Gardiner, T., Barchia, I. M., Donoghue, K. A., and Herd, R. M. 2018. Relationships among carbon dioxide, feed intake, and feed efficiency traits in ad libitum fed beef cattle. Journal of Animal Science 96:4859-4867. doi: 10.1093/jas/sky308

Black, T. E., K. M. Bischoff, V. R. G. Mercadante, G. H. L. Marquezini, N. Dilorenzo, C. C. Chase, 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 cows1. Journal of Animal Science 91(5):2254-2263. doi: 10.2527/jas.2012-5242

Bruns, K. W., R. H. Pritchard, and D. L. Boggs. 2004. The relationships among body weight, body composition, and intramuscular fat content in steers. Journal of Animal Science 82(5):1315-1322. doi: 10.2527/2004.8251315x

Caetano, M., M. J. Wilkes, W. S. Pitchford, S. J. Lee, and P. I. Hynd. 2018. Energy relations in cattle can be quantified using open-circuit gas-quantification systems. Animal Production Science 58(10):1807. doi: 10.1071/an16745

Cantalapiedra-Hijar, G. M. A.-I., G. Carstens, L. Guan, R. Hegarty, D. Kenny, and I. Ortigues-Marty. . 2018. Review: Biological determinants of between-animal variation in feed efficiency of growing beef cattle. Animal 12:321-335. doi: 10.1017/S1751731118001489

Capper, J. L. 2011. The environmental impact of beef production in the United States: 1977 compared with 2007. Journal of Animal Science 89(12):4249-4261. doi: 10.2527/jas.2010-3784

Cassady, C. J. 2016. Evaluation of intake and feed efficiency measures in beef cattle, University of Illinois, Urbana-Champaign.

Cassady, C. J., T. L. Felix, J. E. Beever, and D. W. Shike. 2016. Effects of timing and duration of test period and diet type on intake and feed efficiency of Charolais-sired cattle1. Journal of Animal Science 94(11):4748-4758. doi: 10.2527/jas.2016-0633

Coleman, S. W., S. A. Gunter, J. E. Sprinkle, and J. P. S. Neel. 2014. BEEF SPECIES SYMPOSIUM: Difficulties associated with predicting forage intake by grazing beef cows1,2. Journal of Animal Science 92(7):2775-2784. doi: 10.2527/jas.2013-7090

Donoghue, K. A., T. Bird-Gardiner, R. M. Herd, R. S. Hegarty, and P. F. Arthur. 2020. Genetic variance and covariance components for carbon dioxide production and postweaning traits in Angus cattle. Journal of Animal Science 98(9)doi: 10.1093/jas/skaa253

Drouillard, J. S. 2018. Current situation and future trends for beef production in the United States of America — A review. Asian-Australasian Journal of Animal Sciences 31(7):1007-1016. doi: 10.5713/ajas.18.0428

Durunna, O. N., F. D. N. Mujibi, L. Goonewardene, E. K. Okine, J. A. Basarab, Z. Wang, and S. S. Moore. 2011. Feed efficiency differences and reranking in beef steers fed grower and finisher diets1. Journal of Animal Science 89(1):158-167. doi: 10.2527/jas.2009-2514

Ferrell, C. L., and T. G. Jenkins. 1984. Relationships among Various Body Components of Mature Cows. Journal of Animal Science 58(1):222-233. doi: 10.2527/jas1984.581222x

Fitzsimons, C., D. A. Kenny, M. H. Deighton, A. G. Fahey, and M. McGee. 2013. Methane emissions, body composition, and rumen fermentation traits of beef heifers differing in residual feed intake1. Journal of Animal Science 91(12):5789-5800. doi: 10.2527/jas.2013-6956

Foote, A. P., R. G. Tait, and H. C. Freetly. 2017. 130 Changes in feed intake, growth, feed efficiency, and body composition of beef cattle fed forage then concentrate diets. Journal of Animal Science 95(suppl_4):64-64. doi: 10.2527/asasann.2017.130

Fox, D. G., C. J. Sniffen, and J. D. O'Connor. 1988. Adjusting Nutrient Requirements of Beef Cattle for Animal and Environmental Variations. Journal of Animal Science 66(6):1475-1495. doi: 10.2527/jas1988.6661475x

Freetly, H. C., Huehn, L. A., Thallman, R. M., Snelling, W. M. 2016. Feed intake and production efficiency of beef cows Journal of Animal Science 94:114. (Abstract)

Freetly, H. C., L. A. Kuehn, R. M. Thallman, and W. M. Snelling. 2020. Heritability and genetic correlations of feed intake, body weight gain, residual gain, and residual feed intake of beef cattle as heifers and cows. Journal of Animal Science 98(1)doi: 10.1093/jas/skz394

Galyean, M. L. a. A. L. G. 1993. Forage Cell Wall Structure and Digestibility. American Society of Agronomy, Crop Science Society of America, and Soil Science Society of America, Madison, WI.

Gunter, S. A., and M. R. Beck. 2018. Measuring the respiratory gas exchange by grazing cattle using an automated, open-circuit gas quantification system1. Translational Animal Science 2(1):11-18. doi: 10.1093/tas/txx009

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(9):1815-1826. doi: 10.1017/s1751731118000976

Kim, D. H., McLeod, K. R., Koontz, A. F., Foote, A. P., Klotz, J. L., and Harmon, D. L. 2013. Effect of intake on fasting heat production, respiratory quotient and plasma metabolites measured using the washed rumen technique. Animal 9(1):58-66. doi: 10.1017/S1751731114002183

Lahart, B., R. Prendiville, F. Buckley, E. Kennedy, S. B. Conroy, T. M. Boland, and M. McGee. 2020. The repeatability of feed intake and feed efficiency in beef cattle offered high-concentrate, grass silage and pasture-based diets. Animal 14(11):2288-2297. doi: 10.1017/s1751731120000853

Manafiazar, G., S. Zimmerman, and J. Basarab. 2016. Repeatability and variability of short-term spot measurement of methane and carbon dioxide emissions from beef cattle using GreenFeed Emissions Monitoring System. Canadian Journal of Animal Science doi: 10.1139/cjas-2015-0190

Mao, F., L. Chen, M. Vinsky, E. Okine, Z. Wang, J. Basarab, D. H. Crews, and C. Li. 2013. Phenotypic and genetic relationships of feed efficiency with growth performance, ultrasound, and carcass merit traits in Angus and Charolais steers1. Journal of Animal Science 91(5):2067-2076. doi: 10.2527/jas.2012-5470

Martin, P., Taussat, S., Vinet, A., Krauss, D., Maupetit, D., and Renand, G. . 2019. Genetic parameters and genome-wide association study regarding feed efficiency and slaughter traits in Charolais cows. Journal of Animal Science 97:3684-3698. doi: 10.1093/jas/skz240

Merchen, N. R. a. L. D. B. 1994. Processes of digestion and factors influencing digestion of forage-based diets by ruminants. . American Society of Agronomy, Crop Science Society of America, and Soil Science Society of America, Madison, WI.

Minson, D. J. a. J. R. W. 1994. Prediction of intake as an element of forage quality. American Society of Agronomy, Crop Science Society of America, and Soil Science Society of America, Madison, WI.

Mulliniks, J. T., A. G. Rius, M. A. Edwards, S. R. Edwards, J. D. Hobbs, and R. L. G. Nave. 2015. FORAGES AND PASTURES SYMPOSIUM: Improving efficiency of production in pasture- and range-based beef and dairy systems1. Journal of Animal Science 93(6):2609-2615. doi: 10.2527/jas.2014-8595

NASEM. 2016. Nutrient Requirements of Beef Cattle 8th ed. The National Academies Press, Washington, DC.

Nasrollahi, S. M., M. Imani, and Q. Zebeli. 2015. A meta-analysis and meta-regression of the effect of forage particle size, level, source, and preservation method on feed intake, nutrient digestibility, and performance in dairy cows. Journal of Dairy Science 98(12):8926-8939. doi: 10.3168/jds.2015-9681

NRC. 1996. Nutrient REquirements of Beef Cattle. 7th revised edition ed. National Academy Press, Washington, DC.

Pereira, A. B. D., Utsumi, S. A., Dorich, C. D., and Brito, A. F. 2015. Integrating spot short-term measurments of carbon emissions and backward dietary energy partition calculations to estimate intake in lactating dairy cows fed ad libitum or restricted. Journal of Dairy Science 98:8913-8925. doi: 10.3168/jds.2015-9659

Ramsey, R., D. Doye, C. Ward, J. McGrann, L. Falconer, and S. Bevers. 2005. Factors affecting beef cow-herd costs, production, and profits. Journal of Agriculture and Applied Economics 31(1):91-99. doi: 10.22004/ag.econ.43687

Renand, G., and D. Maupetit. 2016. Assessing individual differences in enteric methane emission among beef heifers using the GreenFeed Emission Monitoring system: effect of the length of testing period on precision. Animal Production Science 56(3):218. doi: 10.1071/an15429

Retallick, K. J., J. M. Bormann, R. L. Weaber, M. D. Macneil, H. L. Bradford, H. C. Freetly, K. E. Hales, D. W. Moser, W. M. Snelling, R. M. Thallman, and L. A. Kuehn. 2017. Genetic variance and covariance and breed differences for feed intake and average daily gain to improve feed efficiency in growing cattle. Journal of Animal Science 95(4):1444-1450. doi: 10.2527/jas.2016.1260

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 selection123. Journal of Animal Science 89(11):3452-3459. doi: 10.2527/jas.2011-3961

Russell, J. R., N. O. Minton, W. J. Sexten, M. S. Kerley, and S. L. Hansen. 2016. Influence of feed efficiency classification on diet digestibility and growth performance of beef steers1. Journal of Animal Science 94(4):1610-1619. doi: 10.2527/jas.2015-9949

Strydom, P. E. 2016. Performance-enhancing technologies of beef production. Animal Frontiers 6(4):22-30. doi: doi:10.2527/af.2016-0040

Tedeschi, L. O., G. Molle, H. M. Menendez, A. Cannas, and M. A. Fonseca. 2019. The assessment of supplementation requirements of grazing ruminants using nutrition models. Translational Animal Science 3(2):811-828. doi: 10.1093/tas/txy140

Tonsor, G. T., and L. L. Schulz. 2015. BEEF SPECIES SYMPOSIUM: Economic considerations related to U.S. beef herd expansion1. Journal of Animal Science 93(9):4227-4234. doi: 10.2527/jas.2014-8473

Velazco, J. I., R. M. Herd, D. J. Cottle, and R. S. Hegarty. 2017. Daily methane emissions and emission intensity of grazing beef cattle genetically divergent for residual feed intake. Animal Production Science 57(4):627. doi: 10.1071/an15111

Wagner, J. J., K. S. Lusby, J. W. Oltjen, J. Rakestraw, R. P. Wettemann, and L. E. Walters. 1988. Carcass Composition in Mature Hereford Cows: Estimation and Effect on Daily Metabolizable Energy Requirement During Winter. Journal of Animal Science 66(3):603-612. doi: 10.2527/jas1988.663603x

Zimmerman, S. 2020. GreenFeed Data Processing Questions. In: A. L. Holder (ed.).

Table 3.1. Composition of diets, %DM

		Treatment[1]	
Item	Supplement[2]	HAY	MIX
Ingredient, % of DM			
Native grass hay	-	100.0	35.4
Rolled corn	-	-	26.0
Soybean hulls	-	-	25.5
Liquid feed[3]	-	-	7.1
Cottonseed meal	95.0	-	5.2
Mineral mix[4]	5.0	-	0.8
Chemical composition, DM basis			
Dry matter, %	88.1	92.8	78.5
Crude protein, %	36.2	7.4	12.5
TDN, %	63.0	49.0	69.0
ME, Mcal/kg	2.83	1.9	2.5

[1] Hay = native tallgrass prairie hay offered on an ad libitum basis plus supplement fed individually daily at the rate of 0.25% of BCS-5-adjusted BW. MIX = total mixed ration offered on an ad libitum basis.

[2] Supplement = protein and mineral supplement fed to cows receiving the hay diet

[3] Liquid feed = 16.8 % crude protein, 83.0% TDN, 3.0 Mcal ME/kg

[4] Classic Aureo® FC C6000; Ca, 9.0%; P, 9.5%; NaCl, 14.0%; Mg, 6.0%; Cu, 750 ppm; I, 35 ppm; Mn, 2,900 ppm; Se, 10 ppm; Zn, 3,000 ppm; Vit A, 100,000 IU/lb; Vit D-3, 10,000 IU/lb; Vit E, 400 IU/lb; Chlortetracycline 3.0 g/lb

[3] Mineral mix was included in the supplement in the HAY diet and blended in the MIX diet

Table 3.2. Transition period diets, feeding methods, and days fed.

Diet	Diet Composition	Days Fed
Transition 1	75% chopped hay 25% MIX	Period 1: d 1 – 5 Period 2: d 68 - 72
Transition 2	50% chopped hay 50% MIX	Period 1: d 6 – 9 Period 2: d 73 - 76
Transition 3	25% chopped hay 75% MIX	Period 1: d 10 – 13 Period 2: d 77 - 79

Table 3.3. Performance, intake, and efficiency response of gestating beef cows fed a forage only and forage-concentrate diet in a crossover feeding design[1]

Item	FC[2]	CF[2]	Diet[3]	Seq[3]	Diet*Seq[3]
No. of cows	21	21	-	-	-
BW, kg			<0.0001	0.1627	<0.0001
HAY	648.0 (16.8)^{a}	738.2 (13.5)^{b}			
MIX	725.3 (15.7)^{bc}	695.0 (13.9)^{c}			
BCS			0.4450	0.0141	<0.0001
HAY	5.9 (0.21)^{a}	7.8 (0.10)^{b}			
MIX	7.3 (0.14)^{c}	6.5 (0.16)^{d}			
ADG, kg/d			<0.0001	0.7719	0.0001
HAY	0.51 (0.06)^{a}	0.09 (0.07)^{b}			
MIX	1.12 (0.13)^{c}	1.48 (0.10)^{d}			
ABF, cm			0.0544	0.0004	<0.0001
HAY	0.28 (0.03)^{a}	0.96 (0.05)^{b}			
MIX	0.73 (0.04)^{c}	0.40 (0.02)^{d}			
ARF, cm			<0.0001	0.0011	0.0264
HAY	0.46 (0.05)^{a}	0.61 (0.04)^{a}			
MIX	1.20 (0.06)^{b}	1.53 (0.07)^{c}			
DMI, kg/d			<0.0001	0.9469	0.9107
HAY	12.0 (0.41)^{a}	11.9 (0.34)^{a}			
MIX	20.0 (0.64)^{b}	20.0 (0.49)^{b}			
DMI, %BW			<0.0001	0.4539	0.0014
HAY	1.87 (0.06)^{a}	1.62 (0.05)^{b}			
MIX	2.77 (0.10)^{c}	2.89 (0.07)^{c}			
G:F			<0.0001	0.0636	<0.0001
HAY	0.06 (0.004)^{a}	0.03 (0.004)^{b}			
MIX	0.07 (0.004)^{c}	0.08 (0.004)^{d}			

[1] Except for number of cows, values in the table are mean ± (standard error).

[2] FC = forage-concentrate diet sequence; CF = concentrate-forage diet sequence.

[3] P-values for effects of diet, sequence, and the interaction of diet and sequence.

[abcd] Means followed by the same letter within item are not significantly different ($P < 0.05$)

Table 3.4. Gas emissions measures of gestating beef cows fed a forage only and forage-concentrate diet in a crossover feeding design[1]

Item	FC	CF	Diet	Seq	Diet*Seq
CO_2 kg/d			<0.0001	0.0004	<0.0001
HAY	9.6 (0.21)[a]	8.8 (0.24)[b]			
MIX	11.1 (0.35)[c]	14.3 (0.29)[d]			
CH_4 kg/d			<0.0001	0.3062	0.0002
HAY	0.3 (0.01)[a]	0.3 (0.01)[b]			
MIX	0.5 (0.02)[c]	0.5 (0.01)[d]			
O_2 kg/d			<0.0001	0.0029	<0.0001
HAY	7.4 (0.17)[a]	6.0 (0.12)[b]			
MIX	8.0 (0.27)[a]	10.9 (0.24)[c]			
RQ			0.1999	0.0194	<0.0001
HAY	0.9 (0.01)[a]	1.1 (0.02)[b]			
MIX	1.0 (0.01)[b]	1.0 (0.01)[b]			
HP MJ/d			<0.0001	0.0017	<0.0001
HAY	107.4 (2.45)[a]	90.0 (1.94)[b]			
MIX	117.9 (3.88)[c]	158.1 (3.34)[d]			

[1] Values in the table are mean ± (standard error).

[2] FC = forage-concentrate diet sequence; CF = concentrate-forage diet sequence.

[3] *P*-values for effects of diet, sequence, and the interaction of diet and sequence.

[abcd] Means followed by the same letter within item are not significantly different ($P < 0.05$).

Table 3.5. Regression equations predicting dry matter intake using metabolic body weight, average daily gain and body condition score

Sequence[1]	Diet[2]	Intercept	MBW[3]	ADG[3]	BCS[3]	AIC[4]	BIC[4]
FC	HAY	2.423	0.0922***	2.985***	-0.657*	67.1	63.1
FC	MIX	17.15**	0.0954	2.960*	-1.943*	80.8	78.8
CF	HAY	4.416	0.0941*	2.946**	-0.786	70.5	66.5
CF	MIX	7.018	0.1551**	2.283**	-1.756**	78.2	74.2

$^{*}P < 0.1$, $^{**}P < 0.05$, $^{***}P < 0.01$

[1] FC = diet sequence consuming HAY during period 1 and MIX during period 2; CF = diet sequence consuming MIX during period 1 and HAY during period 2

[2] HAY = long-stemmed, native grass hay individually supplemented with cottonseed meal (CSM) at 0.25% of BCS-5-adjusted BW; MIX = concentrate-based diet consisting of 35% chopped native grass hay and 65% concentrate (soybean hulls, rolled corn, liquid feed)

[3] MBW = metabolic body weight (kg); ADG = average daily gain (kg); BCS = body condition score (1-9 scale)

[4] AIC = Akaike’s Information Criteria; BIC = Bayesian Information Criteria

Table 3.6. Correlations between gas emissions, intake, and performance traits by diet sequence during the first intake period. Correlations for the CF sequence are found above the diagonal and for the FC cows, below the diagonal.

	BW	ADG	DMI	G:F	RFI	CO_2	CH_4	O_2	HP	RQ
BW		0.26	0.34	-0.16	0.00	0.73^{***}	0.62^{**}	0.88^{***}	0.85^{***}	-0.43
ADG	0.29		0.71^{***}	0.92^{***}	0.00	0.67^{**}	0.73^{***}	0.47^{*}	0.52^{*}	0.28
DMI	0.49^{*}	0.63^{**}		0.45^{*}	0.60^{**}	0.69^{**}	0.81^{***}	0.56^{*}	0.60^{**}	0.15
G:F	0.31	0.82^{***}	0.38		-0.32	0.50^{*}	0.51^{*}	0.32	-0.36	-0.29
RFI	0.00	0.00	0.61^{**}	-0.28		0.24	0.28	0.22	0.23	0.01
CO_2	0.67^{***}	0.39	0.76^{***}	0.16	0.40		0.92^{***}	0.89^{***}	0.93^{***}	0.04
CH_4	0.19	0.33	0.74^{***}	0.05	0.67^{**}	0.79^{***}		0.78^{***}	0.83^{***}	0.12
O_2	0.75^{***}	0.20	0.64^{**}	0.04	0.34	0.93^{***}	0.65^{**}		0.99^{***}	-0.41
HP	0.74^{***}	0.24	0.67^{***}	0.07	0.36	0.96^{***}	0.69^{***}	0.99^{***}		-0.33
RQ	-0.28	0.50^{*}	0.29	0.32	0.10	0.10	0.33	-0.26	-0.18	

$^{***}P < 0.001$, $^{**}P < 0.01$, $^{*}P < 0.05$

Table 3.7. Correlations between gas emissions, intake, and performance traits by diet sequence during the second intake period. Correlations for the CF sequence are found above the diagonal and for the FC cows, below the diagonal.

	BW	ADG	DMI	G:F	RFI	CO$_2$	CH$_4$	O$_2$	HP	RQ
BW		-0.21	0.25	-0.46	0.00	0.50^{*}	0.63^{**}	0.77^{***}	0.71^{***}	-0.16
ADG	-0.15		0.56^{**}	0.98^{***}	0.00	0.24	-0.22	-0.22	-0.24	0.11
DMI	0.11	0.22		0.34	0.74^{***}	0.12	0.18	0.15	0.13	0.04
G:F	-0.14	0.80^{***}	-0.04		-0.02	-0.39	-0.40	-0.43	-0.44	-0.07
RFI	0.00	0.00	0.72^{***}	-0.43		0.10	0.11	0.00	0.01	0.23
CO$_2$	0.08	0.54	0.62^{*}	0.15	0.40		0.80^{***}	0.84^{***}	0.92^{***}	0.64^{**}
CH$_4$	-0.35	0.47	0.46	0.18	0.23	0.74^{**}		0.74^{***}	0.78^{***}	0.43
O$_2$	0.35	0.47	0.56^{*}	0.11	0.36	0.93^{***}	0.47		0.98^{***}	0.13
HP	0.30	0.49	0.58^{*}	0.11	0.38	0.95^{***}	0.53	1.00^{***}		0.30
RQ	-0.74^{**}	0.04	0.04	0.01	0.07	0.05	0.62^{*}	-0.32	-0.25	

$^{***}P < 0.001$, $^{**}P < 0.01$, $^{*}P < 0.05$

Figure 3.1. Observed vs predicted dry matter intake (DMI, NASEM, 2016; DMI, kg/d = [BW$^{0.75}$ * (0.04997 * NEm^2 + 0.04631)] / Feed NEm, Mcal/kg. Solid line = Unity or Y = X and dashed line represents linear regression of observed intake on predicted intake. Intercept and slope differ from 0 (*P* < 0.01) and overall root mean squares deviation = 5.1 kg/

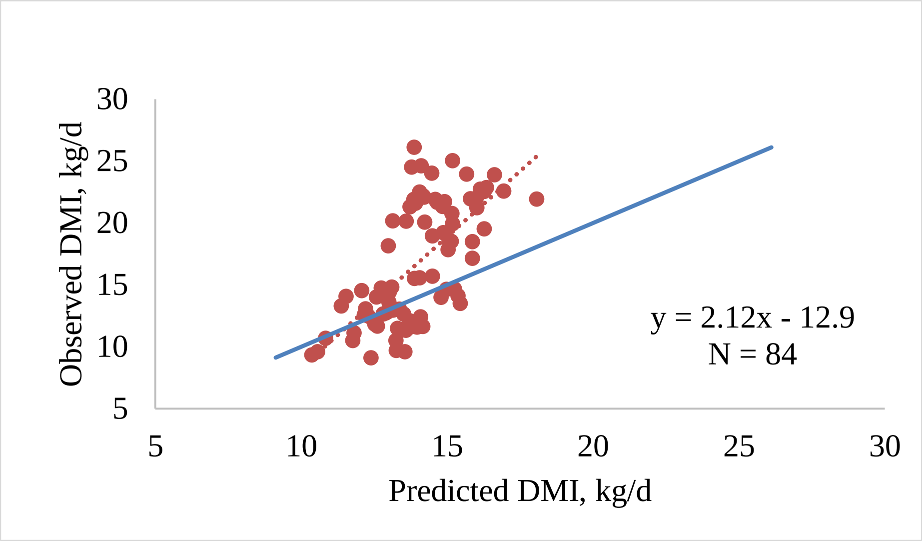
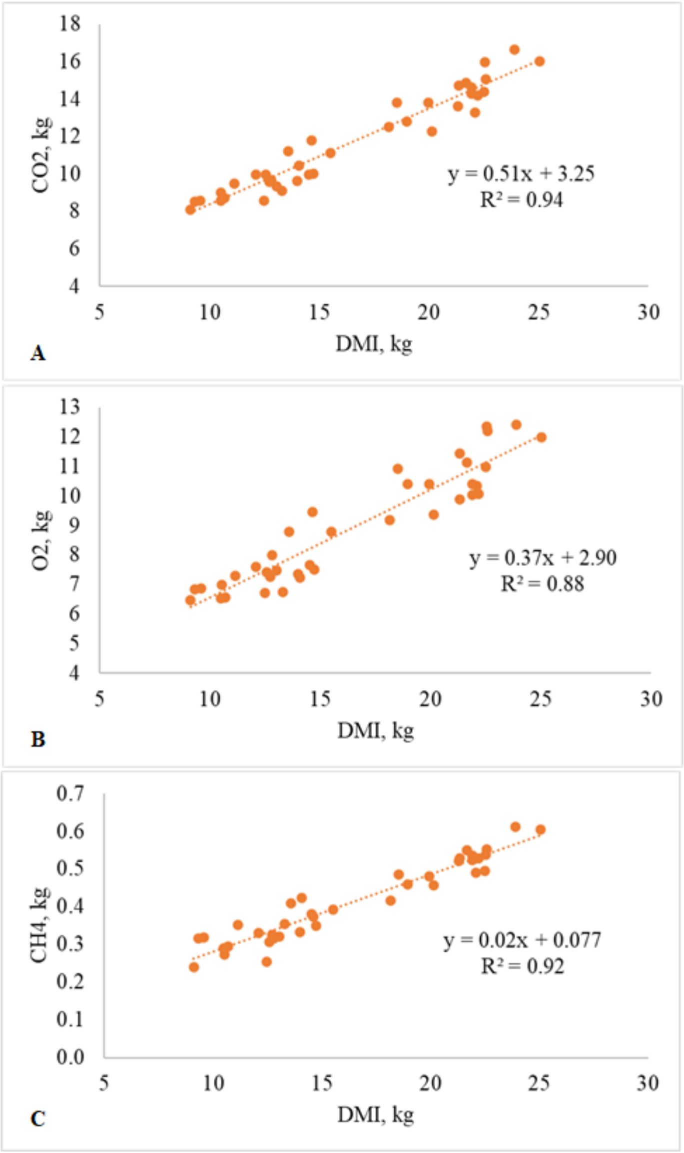


Figure 3.2.

(**A**)Relationship of daily dry matter intake (DMI) to daily CO_2 production for a forage diet (HAY) and a concentrate-based diet (MIX) during first period only; (**B**) relationship of daily DMI to daily O_2 consumption for HAY and MIX diets during first period only; (**C**) relationship of daily DMI to daily CH_4 excretion for HAY and MIX diets during first period only.



VITA

Amanda Leigh Holder

Candidate for the Degree of

Doctor of Philosophy

Dissertation: THE EFFECT OF DIET QUALITY ON DRY MATTER INTAKE, GENE EXPRESSION, GREENHOUSE GAS EMISSIONS, AND PRODUCTION MEASURES IN MATURE BEEF COWS

Major Field: Animal Science

Biographical:

Education:

Completed the requirements for the Doctor of Philosophy in Ruminant Nutrition at Oklahoma State University, Stillwater, Oklahoma in July, 2021.

Completed the requirements for the Master of Science in your major at University of Arkansas, Fayetteville, Arkansas in 2018.

Completed the requirements for the Bachelor of Science in Agronomy and Agriculture Business at College of the Ozarks, Point Lookout, Missouri in 2015.