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University of Nebraska - Lincoln, tasha.king@huskers.unl.edu

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INFLUENCE OF STRATEGIC SUPPLEMENTATION AND GENETIC POTENTIAL
FOR MILK YIELD ON FORAGE DIGESTIBILITY, AMINO ACID UTILIZATION,
AND LIVESTOCK PRODUCTION

by

Tasha M. King

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Under the Supervision of Professors

J. Travis Mulliniks

James C. MacDonald

Lincoln, Nebraska

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INFLUENCE OF STRATEGIC SUPPLEMENTATION AND GENETIC POTENTIAL
FOR MILK YIELD ON FORAGE DIGESTIBILITY, AMINO ACID UTILIZATION,
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Tasha M. King, Ph.D.

University of Nebraska, 2020

Advisors: J. Travis Mulliniks and James C. MacDonald

When consuming low-quality forages or at times when nutrient demands are high, ruminants may be unable to meet these requirements resulting in a state of negative energy balance. With feed costs contributing a large portion of production costs, providing dietary nutrients to maintain energy balance must be done so strategically. Additionally, producers must ensure consistent reproductive performance and offspring weaned to maintain a successful livestock enterprise. The objective of these studies was to evaluate the effect of glucogenic precursor supplementation and milk yield on forage digestibility, amino acid utilization, and livestock production. In experiment 1, a metabolism study was conducted on wethers receiving supplementation treatments with increasing levels of glucogenic precursors. Supplementation treatments providing 40 and 70 g of glucogenic potential (GP) resulted in greater OM digestibility ($P \leq 0.01$). Serum urea nitrogen (SUN) and circulating amino acid concentrations were also increased ($P < 0.01$) in the supplementation treatment providing 40 and 70 g of GP. Experiment 2 was conducted at the Gudmundsen Sandhills Laboratory (GSL) providing postpartum protein with and without calcium propionate to 2- and 3-yo March calving cows. Pregnancy rate and calf pre-weaning performance were not influenced ($P \geq 0.35$) by supplementation treatments of dam. Inclusion of calcium propionate tended ($P = 0.07$) to decrease circulating serum β -hydroxybutyrate concentrations. In Experiment 3, the impact of genetic potential for milk yield on cow reproductive and calf performance was evaluated

utilizing data collected from March calving cows at GSL from 2000 – 2018. Milk yield was estimated 4 times throughout the lactation period utilizing the weigh-suckle-weigh technique. Pregnancy rate and subsequent calf birth date were not influenced ($P \geq 0.43$) by level of milk production. Increasing dam milk production resulted in greater ($P < 0.01$) calf pre-weaning growth. Heavier calves at weaning maintained the weight advantage through slaughter ($P < 0.01$). In conclusion, providing increasing levels of protein can increase digestibility and circulating amino acid concentrations.

Additionally, GP supplementation tended to decrease ketone production suggesting improved energy metabolism. Increased milk production in the current study did not negatively impact reproductive performance and produced heavier calves at weaning.

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CHAPTER I. REVIEW OF LITERATURE

Introduction

The unique nature of the pre-gastric fermentation digestive system of the ruminant allows for the consumption of forages to meet dietary requirements. However, during varying physiological states, the nutrient uptake from low-quality forages may fail to meet the demand of the ruminant. Feed costs are associated with roughly 63% of variability in total cow-costs, which greatly effects profitability of an operation (Miller et al., 2001). Therefore, it is important to meet nutrient demands with minimal inputs, which is also dependent on genetic potential of the cowherd. To meet these fluctuating times of demand, supplementation may be necessary.

When evaluating a cow-calf system, the greatest time of nutrient demands is found at late gestation and early lactation. Depending on the calving season and environment, many cows may enter this time of high demand while grazing dormant range (Mulliniks and Adams, 2019). The decreased forage quality of dormant range combined with the increased energy demands of the cow can result in negative energy balance which has been shown to result in decreased reproductive success (Wathes et al., 2007; de Vries and Veerkamp, 2000; Hawkins et al., 2000). This can be detrimental to the efficiency of an operation as maintaining a yearly calving interval is vital for success. To maintain a yearly calving interval a cow must become bred within 82 d postpartum (NASEM, 2016). However, a greater interval in return to estrus was reported in young

cows (Wiltbank, 1970). Replacement heifers and young cows play a vital role in maintaining the health and productivity of the cow herd yet can create a challenge due to their greater nutritional requirements needed for meeting the demands of maintenance, growth, and production (i.e. lactation, gestation). Providing supplementation when nutrient availability of the forage is unable to meet the demands of the cow, can result in improvements in reproductive performance (Mulliniks and Adams, 2019; Hawkins et al., 2000). Therefore, by providing strategic supplementation to young cows to optimize reproductive efficiency and identify the ideal milk production the environment can support, opportunities to increase cow-calf operation output are available.

Utilization of Energy by Ruminants

Nutrients are supplied in a process that is considered a cycle. Feed is consumed which is digested breaking down into nutrients that are absorbed by animals (Bauman and Currie, 1980). These nutrients are then utilized by body tissues for maintenance, growth, and reserves. Nutrient availability and physiological state determine how nutrients are partitioned for use. Physiological states of lactation and gestation result in increased demand for nutrients from the fetus and mammary tissues. This results in homeorhetic changes or coordinated changes in metabolism to match the demands of the physiological state (Bauman and Currie, 1980).

To match the energy demands of maintenance and physiological state, ruminants utilize volatile fatty acids (VFA), glucose, and protein. Volatile fatty acids are produced when microbes in the rumen ferment dietary carbohydrates in the form of cellulose, hemicellulose, and starch consumed by the host (Church, 1988). The primary VFA

produced by the microbes are acetate, propionate, and butyrate. Upon absorption through the rumen wall, the utilization of the VFA differs (Preston and Leng, 1978). Butyrate is converted to ketone bodies for energy use in the epithelial tissue of the rumen wall. Acetate and propionate are absorbed into the portal bloodstream for transport to the liver. Propionate contributes carbon atoms directly to gluconeogenesis making it a vital gluconeogenic substrate (Engleking, 2015). Upon entering the liver, propionate enters hepatic gluconeogenesis (Brockman, 1990) and can be utilized as a precursor for 27 - 66% of glucose production (Engleking, 2015). However, acetate enters the liver through the portal vein and 80% of acetate remains unchanged, passing through to the peripheral tissues (Church, 1988). Acetate is a main precursor for lipogenesis and metabolism of acetate is often greater in adipose tissue due to the high level of activity of acetyl-CoA synthetase found in these tissues.

At the peripheral tissues, glucose enters cells through GLUT4 transporters and enters glycolysis to be converted to oxaloacetate. Acetate enters the cell via diffusion across a concentration gradient to enter the TCA cycle as acetyl-CoA. The first step of the TCA cycle is formation of citrate through the combination of acetyl-CoA and oxaloacetate. Acetate utilization is dependent upon glucose supply due to this requirement for oxaloacetate (Preston and Leng, 1978). Therefore, a diet of low-quality forage, which has an increased production of acetate and decreased production of propionate (Church, 1988) results in a decreased efficiency of acetate utilization. Increasing the glucogenic potential of the diet through improving forage quality or

supplement (i.e. glucogenic precursors, propionic salts) results in faster rate of acetate metabolism signaling improved energy metabolism (Cronjé et al., 1991).

Physiological State on Nutrient Demand

To achieve maximum efficiency of cow-calf systems, producers aim to have cows that maintain a yearly calving interval. However, to meet these demands, one must consider the variability of nutrient requirements throughout the year. Energy demand is lowest after weaning and increases into late gestation peaking at early lactation before decreasing again (NASEM, 2016). With the varying nutrient availability depending on environment and diet availability, cows will commonly go through a cyclic pattern of increases and decreases in cow BW and BCS.

Nutrient demand is even greater in young cows that still have not reached maturity. During the postpartum period, young cows are partitioning energy towards tissue, lactation, maintenance, and growth. Difficulty for dietary intake to match the energy demand can result in negative energy balance. In negative energy balance, cows mobilize body fat reserves producing glycerol for energy (de Vries and Veerkamp, 2000). This results in an increase in serum non-esterified fatty acid (NEFA) concentration which provides an additional energy source through oxidation. Maintaining energy balance is important as negative energy balance in cows can result in decreased fertility (Wathes et al., 2007) and loss of BCS and BW.

Late gestation results in an increase of maternal requirements between 30 and 50% (Bell, 1995). This increase in requirements is met between increases in voluntary

intake and the homeorhetic changes to metabolism. Bell (1995) reported fetal growth to utilize 46% of the maternal glucose supply. This agrees with results by Bauman and Currie (1980) that reported glucose to supply 50 to 70% of total energy substrates for fetal growth. To meet this increased demand for glucose, maternal gluconeogenesis increases made possible by the potential increased uptake of protein and glucose. Additionally, increased maternal metabolism of lipid substrates allows maternal glucose supply to be spared for fetal growth. This is supported by observations of increased serum NEFA concentrations during late gestation (Bell, 1995).

Lactation results in an increase of nutrient requirements. In response to increased requirements for lactation, studies have reported increased forage intake (Hatfield et al., 1989; Johnson et al., 2003). Cows with a high milk EPD consumed 8% more forage than those with a low milk EPD (Johnson et al., 2003). This increase in forage consumption was reported at a rate of 0.33 and 0.37 kg in forage DMI in early and late lactation for every additional kg of increase in milk yield. Even with increased forage intake, cows can still be challenged for nutrition depending on the quality of forage available. With glucose serving as the sole precursor for lactose, lactation is a huge demand for glucose (Hawkins et al., 2000). In the last 60 d of gestation, Bauman and Currie (1980) reported fetal glucose and AA demand to be equivalent to the demand from mammary tissues that are producing 3 to 6 kg of milk per day. Bell (1995) reported the glucose requirement of mammary tissues in high producing dairy cows to be 2.5 times greater than the demands by the late gestation uterus. Glucose demand in goats has been reported to have a large increase in glucose demand from the mammary tissues starting at 2 d prepartum (Davis et

al., 1979). Blood flow was also increased starting 2 d prepartum and then increased again at 1 d postpartum. This agrees with the cyclic nature of lactogenesis (Svennersten-Sjaunja and Olson, 2005).

Glucogenic Precursor Supplementation

Glucogenic precursors provide a source of NADPH, which is required for the oxidation of acetate. If sufficient NADPH is not available, acetate would not be converted to beneficial fatty acids but rather be lost as heat (Preston and Leng, 1978). Increased acetate clearance rates suggest that a sufficient balance of acetate and glucogenic precursors is present. During times of limited propionate, glucogenic amino acids (AA) can be used as a source of glucose synthesis (Preston and Leng, 1978; Overton et al., 1999). Cronjé et al. (1991) provided supplemental propionate and protein to wethers consuming a forage diet and measured acetate flux. Both propionate and protein supplementation; increased the rate of acetate clearance; however, they are not equally used in gluconeogenesis. Propionate makes a net contribution to gluconeogenesis, while amino acids have varying levels of glucogenicity and are metabolized for other uses like maintenance of tissues. A portion of propionate is also converted to lactate, another glucogenic precursor. Efficient utilization of acetate is important, as it serves as an energy source and substrate for fat synthesis which can be beneficial during times of low glucose concentration.

The glucogenic potential of a diet can be estimated using an acetate tolerance test (ATT). Acetate is infused into the bloodstream and serum concentration is measured at various time points to identify how quickly the acetate diffuses from the bloodstream into the cell. As the glucogenic potential of the diet increases, rate at which the infused acetate is cleared from the bloodstream increases. However, when the glucogenic potential of the diet is low, acetate diffusion is unfavorable resulting in decreased acetate clearance.

Low-quality forage diets produce primarily acetate and butyrate with only roughly 15% of total VFA being propionate (Caton et al., 1988), which provides a low glucogenic potential of the diet. This decrease in production of propionate through ruminal fermentation can result in a decrease of glucogenic precursors. To have sufficient utilization of dietary energy, it is important to supply adequate glucogenic precursors to ensure metabolic function and reproductive performance (Hawkins et al., 2000). Evaluating low-quality forage diets with and without protein supplementation, McCollum and Galyean (1985) observed a potential shift in the acetate:propionate ratio with reduced acetate and increased propionate when supplemented.

Protein supplementation increases serum insulin resulting in greater uptake of glucose by adipose tissue and inhibition of fatty acid mobilization. Decreasing body tissue mobilization and utilizing a greater amount of the maternal glucose supply for the dam reduces the amount of nutrients available for lactation, resulting in a drop in milk production (Hunter and Magner, 1988). This reduces the overall energy demand of the dam; however, it may limit the production potential of the dam. In agreement, additional

supplemental protein did not increase milk production during early lactation (Marston et al., 1995); but supplementation of energy resulted in a 0.5 kg/d increase in milk production. Infusion of glucogenic substrates post-ruminally resulted in increases in milk yield and milk lactose content compared to control (Vanhatalo et al., 2003). Increases in milk lactose content suggests that the additional glucose supply allowed greater production. Waterman et al. (2006) reported similar results with increasing milk yield when RUP was supplemented above requirements as a source of glucogenic precursors. In contrast, when increasing the level of glucogenic precursor supplementation did not result in greater 24-h milk production (Patton et al., 2004; Mulliniks et al., 2011). Increasing levels of glucogenic precursor supplementation were not reported to affect milk constituent concentrations (Mulliniks et al., 2011). This suggests that milk production is more dependent on the type of energy provided in the diet compared to increasing levels of protein. This concurs with data from Perry et al. (1991) who provided dietary energy at 70% of the recommended level to postpartum cows resulting in decreased total milk yield and lower percentages of milk fat, protein, and total solids.

Increasing energy intake postpartum has been shown to shorten postpartum interval (Dunn et al., 1969; Wiltbank et al., 1964). Perry et al. (1991) reported cows receiving 70% of recommended dietary energy level to have a longer interval from parturition to first ovulation. This suggests that supplementing during the high nutrient demand of the postpartum period can improve fertility. Postpartum interval is of important consideration when producers are trying to return young cows to estrus. Development of supplementation strategies can be economical and improve reproductive

efficiency of the herd. Feeding rumen undegradable protein (RUP) postpartum in young cows initiated weight gain and pregnancy faster (Hunter and Magner, 1988; Wiley et al., 1991). Additionally, Mulliniks et al. (2011) provided a RUP supplement after parturition with varying levels of glucogenic precursor resulting in a favorable improvement in days to estrus and tended to have a quadratic response on pregnancy rate.

In contrary, Marston et al. (1995) reported energy supplementation to improve pregnancy rate when supplemented prepartum compared to postpartum supplementation. In agreement, Hight (1968) reported an effect of prepartum plane of nutrition on subsequent fertility even with consumption of improved plane of nutrition postpartum. Pre-calving energy level had the greatest influence on estrus early in the post-partum period (< 60 d) while post-calving energy level had the greatest influence on estrus later in the post-partum period (> 80 d; Dunn et al., 1969). Supplementing with RUP and RUP plus glucogenic precursors in postpartum cows had no impact on pregnancy rates (Waterman et al., 2006). These varying results in impacts on length of anestrus and pregnancy rate suggests that there may be a threshold of BCS or nutrition that may need to be observed before postpartum supplementation is beneficial for reproductive efficiency. Richards et al. (1986) reported the effect of postpartum nutrition is dependent on the BCS of the cow at calving. Cows that calved at a BCS > 5.0 returned to estrus at the same rate independent of the postpartum nutrition. However, cows that calved at a BCS < 4.0 had reduced percentage returning to estrus as level of postpartum nutrition decreased. In agreement, Dunn et al. (1969) reported feeding a high level of energy postpartum can help improve conception rate. Yet, a small number of cows fed low

energy in late gestation failed to show estrus at 40, 60 and 80-d post-parturition suggesting that post-partum nutrition levels cannot completely overcome failure to provide adequate energy pre-calving.

When energy is not being met by dietary intake, adipose tissue is broken down into NEFA. Therefore, increased serum concentrations of NEFA represents negative energy balance. Cows receiving no supplementation or a low level of RUP had greater NEFA concentration compared to cows receiving higher levels of RUP in late gestation (Sletmoen-Olson et al., 2000). This reflects the quality of the diet and physiological state. All treatments observed a linear decrease in NEFA concentration in the first 75 d of lactation. However supplementing RUP resulted in no difference in plasma NEFA concentration between late gestation and early lactation. Propionate has been shown to decrease ketogenesis by reducing the rate of oxidation of palmitate to ketone bodies in sheep hepatic cells of both starved and fed sheep (Lomax et al., 1983). Affecting both distribution of acetyl-CoA and the rate of β -oxidation, propionate may serve as a regulator of ketogenesis in the ruminant liver. However, studies supplemented increasing levels of glucogenic precursors and reported no effect on serum NEFA concentrations (Waterman et al., 2006; Mulliniks et al., 2011; Endecott et al., 2012). Beta-hydroxybutyrate is another ketone body that can be measured in serum to determine energy balance. While no effect was observed on NEFA serum concentration with varying levels of glucogenic potential, Mulliniks et al. (2011) observed a decrease in β -hydroxybutyrate concentration. When supplementation ceased, Endecott et al. (2012) observed concentration levels of β -hydroxybutyrate to double compared to serum

concentrations measured during time of supplementation. These data suggest that by increasing the glucogenic potential of cows during grazing or consumption of low-quality forages through supplementation, can help prevent negative energy balance in cows. By providing glucogenic precursors, mobilization of lipid stores for energy can be spared.

Impact of Milk Production on Performance

A positive relationship between weaning weight and milk production has been observed with greater milk production resulting in heavier calves at weaning (Clutter and Nielsen, 1987; Abdelsamei et al., 2005). This has resulted in producers selecting breeding stock with potential for greater milk production. However, even with selection for greater milk production and calf growth, some regions of the United States have observed no increase in weaning calf BW (Lalman et al., 2019). Therefore, it is important to consider if the environmental and nutritional conditions allow the maximum potential to be met. With increased milk production, nutrient requirements for cows become increased (Ferrell and Jenkins, 1984; Montaña-Bermudez et al., 1990), which may result in negative energy balance or stunted production if nutrient availability is already limited.

Along with genetic potential, cow BW also has an impact on milk production. McMorris and Wilton (1986) reported a 0.3 kg increase of daily milk production per 100 kg of cow BW. Other studies have reported similar findings with heavier cows producing greater milk yields throughout the lactation period (Roche et al., 2007; Vaz et

al., 2016). While greater milk production is beneficial, the increase in cow BW and increase in milk production results in greater demand for nutrients. This supports findings from a study looking at differing levels of milk production in a high feed environment, which reported cow BW to have no influence on milk yield (Edwards et al., 2017). Nutrients were likely not limited in the high feed environment resulting in no impact on milk yield. A study evaluating metabolic body sizes in different breeds, reported maintenance requirements to be greatest for those cows that had the potential for greater milk production (Ferrell and Jenkins, 1984). This study reported that cows with the same potential for milk production, though at different metabolic sizes, had similar maintenance requirements suggesting that the muscular size of the cow has less impact on maintenance requirements compared to internal organ mass. In contrast, Montaña-Bermudez and Nielsen (1990) reported energy requirements to be higher by 10 and 12% for cows with medium and high milk production, respectively, over a low milk producing group.

Age of cow is generally associated with parity and lactation which can impact milk yield. Clutter and Nielsen (1987) reported milk production to increase in the first three lactations and plateau after that. These three lactations often fall in the cow age range that Lubritz et al. (1989) saw increasing milk yield from 2 to 5 years. However, the plateau that often occurs at maturity does not remain throughout the remaining productive lifetime of the cow. A decrease in milk production has been observed after 6 to 8 years of age (Lush and Shrode, 1950; Boggs et al., 1980). With these increases in cow milk

yield as cows reach maturity, days in milk has also been observed to increase (Roche et al., 2007).

One of the greatest impacts on production, efficiency, and overall profit for cow-calf producers is kg of calf weaned. Historically, positive associations between milk yield and weaning weight have been reported (Clutter and Nielsen, 1987; Abdelsamei et al., 2005). Milk from the dam makes the largest component of dietary intake for calves within the first 60 d postpartum. Providing ample supply and quality fat and protein impacts calf average daily gain (ADG). Milk yield resulted in a 71.3% variance in calf ADG (Gleddie and Berg, 1968), with biggest impact on variance observed in the second month of lactation. In agreement, cow-calf pairs maintained in a drylot setting had a high correlation between milk production and 70 d calf BW (Perry et al., 1991). While these improvements in pre-weaning ADG have been reported due to milk production (Gleddie and Berg, 1968; Beal et al., 1990; and Perry et al., 1991), others have not observed the improvements in pre-weaning ADG to impact final weaning weights (Ansotegui et al., 1991 and Edwards et al., 2017). Forage consumption by the calf increases after 60 d which can compensate for lack of milk intake by the suckling calf. This is likely due to the decrease in milk yield as the lactation period progresses, the increase in forage consumption by the calf, and the reduced reliance on milk for dietary nutrients.

With differences in pre-weaning ADG being observed when reliance on milk for dietary nutrients is greatest, the quality of forage consumed may impact whether the advantage of greater milk yield is observed in adjusted 205-d weaning weight. In a high forage quality environment, Edwards et al. (2017) observed no difference in weaning

weights between calves raised by dams with varying milk yields. In contrast, others have shown the advantage of greater milk production on calf ADG to result in greater weaning weights (Rutledge et al., 1971; Clutter and Nielsen, 1987; Minick et al., 2001; and Vaz et al., 2016). Calves from dams with lower milk yields averaged 9.96% less kg calf BW per 100 kg of cow BW at birth and 16.1% less kg of calf BW per 100 kg of cow BW at weaning (Vaz et al., 2016). Milk production producing greater calf weaning BW, results in calves entering the feedlot at a heavier BW. While milk production of the dam does not influence postweaning ADG, the heavier BW at entrance of the finishing period is maintained. Feedlot ADG was reported to be similar among calves from dams with varying levels of milk production (Abdelsamei et al., 2005). However, calves that entered the feedlot had decreased days on feed. In a study evaluating both Hereford and Herford \times dairy cattle, no correlation between milk production and postweaning performance was observed (Davis et al., 1985). However, it was noted that the maximum biological efficiency of milk production was not met potentially inhibiting detection of the true impact of milk production on progeny performance.

Increasing milk production results in greater energy demand which if not met by dietary intake can result in negative energy balance. This negative energy balance can have an unfavorable impact on reproductive performance. An issue associated with this is a delay in return to estrus. Evaluating the impact of milk production on return to estrus, Berry et al. (2003) reported no correlation between milk yield and time interval to first service. In contrast, others have reported a decrease in reproductive efficiency in young cows due to the metabolic demand caused by lactation (Mulliniks et al., 2013;

Hobbs et al., 2017). Increases in postpartum interval have been reported ranging from 1.4 to 5.5 d / kg milk produced (Boggs et al., 1980; Bartle et al., 1984; Mulliniks et al., 2013). These data suggest that nutrient availability may be too limited to meet the demands of lactation and reproductive performance. When grouping cows by milk yield, Edwards et al. (2017) reported cows that had a high milk yield producing roughly 12 kg at d 58 postpartum had reduced pregnancy rates. Cows that were grouped into moderate (~ 9 kg) and low (~ 6 kg) milk yields did not have affected pregnancy rates. Similar results were observed in dairy cows with those that had the greatest milk yield by 100 d postpartum having lower pregnancy rates after the first service (Buckley et al., 2007). Consideration of nutrient availability must be considered when comparing the benefits of greater milk production on calf performance and the impact it may have on reproductive efficiency.

Conclusion

In cow-calf production improving reproductive efficiency and increasing kilograms of calf weaned are critical for success. The demands of growth and lactation in young cows may result in a deficiency of energy increasing return to estrus. To maintain yearly calving interval, cows must return to estrus and conceive in a timely manner. Supplementing glucogenic precursors have the potential to decrease fatty acid mobilization and improve energy balance allowing nutrients to meet lactation demands and excess partitioned towards reproductive performance. However, further research needs to be conducted to determine if glucogenic precursors can improve nutrient utilization to provide a potential substitution to expensive RUP sources. Selecting for

greater milk production in cow-calf pairs grazing range has the potential for improved performance to produce heavier calves at weaning. However, it is necessary to remember that should nutrient availability become too limited, negative impacts on reproductive performance may occur.

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CHAPTER II. Effect of propionate salt supplementation on digestibility, energy metabolism, and rumen parameters in sheep

T. M. King*, J. K. Beard*, M. M. Norman*, H. C. Wilson*, J. C. MacDonald*, and J. T. Mulliniks[§]

*Department of Animal Science, University of Nebraska-Lincoln, Lincoln, NE 68583

[§]West Central Research and Extension Center, University of Nebraska, North Platte, NE 69101

Abstract:

Supplementation of glucogenic precursors in a roughage diet may increase production responses due to improved efficiencies of nutrient utilization. Therefore, the objective of this study was to determine the effect of supplemental glucogenic potential (GP) on forage digestibility, serum metabolites, energy utilization, and rumen parameters of growing wethers consuming a roughage diet (8.8% crude protein, 71.4% ash-free neutral detergent fiber). Crossbred wethers (49.1 ± 4.7 kg initial BW; $n = 16$) were utilized in a 4×4 replicated Latin Square design with four periods of 21 d. Supplements were designed to supplement increasing amount of GP: (1) no supplementation (CON; 0 g), (2) 40 g of calcium propionate (CAP; 30 g of GP), (3) 70 g of blood meal + 100 g of feather meal (BF; 40 g of GP), or (4) combination of CAP and BF (COMBO; 70 g of GP). Total fecal and urine collection was conducted from d 13 – 17 to calculate digestibility estimates and urinary losses. An acetate tolerance test (ATT) was administered on d 17 to determine the effect of GP on acetate clearance. Blood samples were taken on d 19

pre-prandial and 4 h post-prandial and were analyzed for serum concentrations of glucose and urea N (SUN). Rumen fluid was collected 4 h post-prandial on d 21 to determine supplementation effects on ruminal volatile fatty acid (VFA) and ammonia concentrations. Wethers receiving BF and COMBO supplementation had greatest ($P \leq 0.01$) DM and OM total tract digestibility. Supplementation did not affect ($P \geq 0.37$) NDF digestibility or digestible energy. Urinary nitrogen excretion was greatest ($P = 0.02$) for BF and COMBO. Circulating serum essential amino acid concentration was increased ($P < 0.01$) in BF and COMBO compared to CAP and CON. In addition, BF and COMBO had increased ($P < 0.01$) SUN concentrations compared to CAP and CON. Acetate half-life was not affected ($P = 0.39$) by supplementation strategy. However, area under the curve (AUC) for acetate was decreased ($P = 0.04$) with supplementation with no difference ($P \geq 0.80$) in glucose and insulin AUC. Ruminal propionate concentration was increased ($P \leq 0.01$) for CAP and COMBO supplementation resulting in decreased ($P \leq 0.01$) A:P ratio. These results suggest that supplementing above protein requirements may improve energy efficiency.

Key words: forage digestibility, lambs, propionate salt, protein supplementation

Introduction

Supplementation of glucogenic precursors and rumen undegradable (RUP) may increase production responses due to improved efficiencies of nutrient utilization. In forage-based production systems, ruminal production of acetate compared to propionate can result in imbalanced acetate:propionate ratio (McCollum and Galyean, 1985; Cronjé et al., 1991), resulting in negative modifications in energy metabolism. Sanchez et al.

(2014) reported supplementation of propionate source to decrease the acetate:propionate ratio, while others (DelCurto et al., 1990; Salisbury et al., 2004) have observed a similar response when supplementing protein. Ferrell et al. (1999) observed greater digestible energy and available amino acids when a combination of energy and RUP were supplemented to a diet of low-quality hay. Providing growing lambs with supplemental RUP consuming low-protein forage diets resulted in increased feed intake, rate of growth and improved feed efficiency with an additional growth response observed due to an increase of post-ruminal glucose (Kempton et al., 1978). However, the additional growth response due to post-ruminal increase of glucose was only observed once RUP requirements were met. In addition, continuous duodenal infusion of glucose resulted in increased growth rate and improved feed conversion for lambs consuming a low-protein diet regardless of supplemental bypass protein (Leng et al., 1978). Increasing post-ruminal supply of propionate increases fatty acid and acetate hindlimb uptake of growing lambs (Majdoub et al., 2003). Similarly, Mulliniks et al. (2011) reported that increasing glucogenic precursors with RUP and 40 g/d calcium propionate enhanced energy metabolism by increasing the rate that acetate is metabolized in young, lactating range beef cows grazing dormant forage. Our hypothesis was that providing increased levels of glucogenic precursors would increase acetate utilization and efficiency in growing lambs on a forage-based diet. Therefore, the objective of this study was to determine the effect of supplemental glucogenic potential (GP) on forage digestibility, serum metabolites, rumen parameters, and energy utilization of a forage diet.

Materials and Methods

All animal care and management procedures used were reviewed and approved by the University of Nebraska Institutional Care and Animal Use Committee (IACUC #1678).

Sixteen crossbred wethers (49.1 ± 4.7 kg initial BW) were utilized to determine forage digestibility, blood and rumen parameters, and acetate utilization. Wethers were sorted into 4 blocks based on initial BW in a 4×4 replicated Latin Square design. Wethers were assigned randomly within each period to 1 of 4 treatments to provide 0, 30, 40, and 70 g of GP: (1) control (**CON**; 0 g of GP), (2) 40 g of NutroCal (**CAP**; Ca-propionate, 30 g of GP; Kemin Industries Inc., Des Moines, IA), (3) 70 g of blood meal and 100 g of feather meal (**BF**; 40 g of GP), or (4) combination of GP and BF (**COMBO**; 70 g of GP). Crude protein (CP) percentage provided from blood meal and feather meal was calculated by adding grams of CP from each and dividing by the total 170 g of supplement. Values for CP and RUP % were taken from NASEM (2016). The combination of supplements for the BF treatment was 92.6% CP and 57.9% digestible rumen undegradable protein (RUP; % CP). Grams of GP from RUP was calculated assuming 40% of digestible RUP is glucogenic (Preston and Leng, 1987). Nutrocal contains 80% propionate which is 95% glucogenic (Steinhour and Bauman, 1988), allowing for calculation of the GP it provides. Brome grass hay (8.8% CP, 90.9% organic matter (OM), 71.4% ash-free neutral detergent fiber (NDF_{om}), 44.8% acid detergent fiber (ADF)) was ground with a tub grinder through a 2.5-cm screen and fed at a constant 2% BW. An ounce of commercial mineral + vitamin premix was offered daily to all wethers.

Periods were 21-d in length allowing for 12-d of diet adaptation, 5-d of total fecal and urine collection, and 4-d for metabolism collections. Wethers were fed brome grass hay twice daily at 0800 and 1700 h, with 50% of daily DM at each feeding. Supplementation occurred at 0730 h each day. Wethers receiving BF supplementation were adapted at levels of 40, 60, and 80% total supplementation on d 1-3 of each period, respectively. Feed refusals were taken daily prior to supplementation. On d 12, wethers were placed in metabolism crates at 1700 h for total fecal collection. Fecal bags were emptied and recorded at 0800 and 1700 h daily, 10% of each fecal collection was retained for data analysis and stored at 2.8°C until the end of the period. Five percent of each fecal collection was composited by period and lyophilized (VirTis Freezemobile 25ES, SP Scientific, Gardiner, NY). Urine was collected daily via gravity flow into covered tubs below metabolism crates. Tubs contained 100 mL of 1 M hydrochloric acid to prevent volatilization of N and was replaced daily. At 1700 h, tubs were removed, weight was recorded, and 10% of total urine collection was retained and stored at 2.8°C until further analysis. Urine was thawed and boiled to reduce water content prior to further analysis (Judy et al., 2019). Beakers filled with urine were placed into a boiling water bath (Ankom Technology, Macedon, NY) underneath a hood. Urine paste was then lyophilized (VirTis Freezemobile 25ES, SP Scientific, Gardiner, NY) and analyzed. Percentage nitrogen of urine was analyzed utilizing a LECO nitrogen analyzer (LECO, St. Joseph, MI). Energy lost from urine was analyzed with a Parr 6400 calorimeter (Parr Instrument Company, Moline, IL). Feed refusals were taken d 10 to 15 and feed samples taken d 12 and 19 were dried at 60°C for 72 h to correct daily dry matter intake. Fecal, feed, and feed refusal samples were ground through a 1-mm screen of a Wiley mill and

analyzed for OM, NDF_{om}, and ADF. Analysis for NDF_{om} and ADF was conducted using the beaker method (Van Soest et al., 1991). Gross energy was analyzed using a Parr 6400 calorimeter (Parr Instrument Company, Moline, IL) for individual fecal samples, composite feed samples, and composite feed refusal samples for each period. Caps containing 2.0 g of sample and 0.4 g of mineral oil sat for a minimum of 12 h prior to being bombed for determination of gross heat. Digestible energy was then calculated by subtracting the energy lost in feces from GE of feed intake (NASEM, 2016).

An acetate tolerance test (ATT) was conducted on d 17 to analyze acetate clearance as affected by GP of treatments. Jugular catheters were inserted the morning of the ATT, through which a 20% acetic acid solution was infused at 1.25 mL/kg of BW. Blood samples were then collected (~7 mL) -1, 0, 1, 3, 5, 7, 10, 15, 30, 60, and 90 min relative to infusion. Samples were placed in Corvac serum separator tubes, cooled, and centrifuged at 2,000 x g at 4°C for 20 min. Serum was collected and stored at -20°C for later analysis of acetate, insulin, and glucose concentrations. Serum was filtered with a centrifugal filter device for 100 min at 4°C at 5,000 x g for deproteinization (Amicon Ultra-2 centrifugal device, Millipore Corp., Burlington, MA). Filtered serum was mixed at a 5:1 ratio with 25% metaphosphoric acid containing 2 g/L of 2-ethyl butyric acid as an internal standard. Acetate concentration was analyzed via gas chromatography adapted from the method of Goetsch and Galyean (1983). The half-life of acetate was calculated as the time required for a 50% decrease from peak serum concentration (Kaneko, 1989). Serum were analyzed for glucose concentration by lab in the Biomedical and Obesity Research Core (BORC) of the Nebraska Center for Prevention of Obesity Diseases

(NPOD). Serum acetate and glucose area under the curves (AUC) were calculated using the trapezoidal method.

On d 19, a blood sample was taken pre-prandial at 0730 h and 4 h post-prandial at 1230 h via venipuncture from the jugular vein and the saphenous vein found in the hindlimb into serum separator vacuum tubes (Corvac, Kendall Healthcare, St. Louis, MO). Serum samples were analyzed for glucose, urea N (SUN), non-esterified fatty acids (NEFA), and circulating amino acid (AA) concentrations. Glucose and SUN were also analyzed by the BORC lab of NPOD. Amino acid concentrations were analyzed using the EZ:faast™ For Free (Physiological) Amino Acid Analysis kit (Phenomenex, Torrance, CA) for gas chromatography (GC). Serum samples were analyzed for NEFA concentration utilizing the WAKO HR Series NEFA-HR(2) (FUJIFILM Wako Diagnostics U.S.A., Mountain View, CA).

Rumen samples were collected 4 h post-prandial at 1230 h on d 21. A sample of contents (40 mL) were collected through oral lavage, snap frozen in liquid nitrogen, and stored at -20°C until analysis. Samples were thawed and centrifuged at 5,000 x g for 20 minutes prior to analysis for VFA and ruminal ammonia concentration. For analysis of VFA concentration, 2.0 mL were pipetted into test tubes. To each test tube, 0.5 mL of ice cold 25% meta-phosphoric acid/crotonic acid solution was added and then vortexed. Test tubes were then refrigerated at 4°C for 30 min and then centrifuged at 10,000 x g for 15 minutes. Tuberculin syringes are filled with 3.0 mL of supernatant and filtered through a filter-tip syringe into a GC vial and analyzed for VFA concentration on the GC. For ruminal ammonia concentration, 40 µL of rumen fluid plus 40 µL of H₂O were dispensed

into plastic test tubes. Phenol reagent was added at 2.5 mL followed by 2.0 mL of alkaline hypochlorite reagent. Tubes were then vortexed and incubated in a 37°C water bath for 10 min. Then 300 µL was pipetted from each tube into the wells of a microtiter plate and absorbance was read on each plate at 550 nm. A standard curve was calculated using linear regression where: $x = \text{absorbance}$ and $y = \text{concentration}$. Sample absorbances were applied to standard curve calculation to determine concentration.

Statistical Analysis

Total tract digestibility and rumen parameters data were analyzed as a Latin Square design using the MIXED procedure (SAS Inst. Inc., Cary, NC, USA). Lambs were blocked by weight into light and heavy blocks. Data were analyzed with lamb serving as experimental unit, with supplementation type and period as fixed effects. Lamb within period served as a random effect. Acetate half-lives were estimated for each animal by regressing the logarithmically transformed acetate concentrations over time (Kaneko, 1989). Area under the curves were determined for acetate and glucose using the trapezoidal summation method. Serum data were analyzed as repeated measures with time of blood collection serving as repeated factor with an autoregressive covariate structure. Treatment by location and treatment by time interactions were not significant ($P > 0.05$) and were removed from the model. Significance level was set at $P \leq 0.05$.

Results

Wethers receiving BF and COMBO supplementation had greater ($P < 0.01$; Table 1) DM and OM total tract digestibilities compared to the CAP and CON treatments.

Supplemental treatments had no impact ($P = 0.93$) on NDF_{om} digestibility. Wethers receiving BF supplementation had greatest ($P = 0.03$) ADF digestibility while CAP had the lowest ADF digestibility. Wethers fed CON and COMBO treatments had intermediate ADF digestibilities with COMBO having similar ADF digestibility to both BF and CON ($P \geq 0.06$). Total intake of DM, OM, and ADF increased ($P < 0.01$) with increasing GP supplementation, which was expected due to total intake including basal and supplementation amounts. However, wethers consuming CON consumed a greater quantity of hay resulting in greater ($P = 0.02$; Table 2) forage DM intake compared to the supplemental treatments forage DM intake. Forage OM and ADF intake did not differ ($P > 0.05$) among treatments. However, forage NDF intake tended ($P = 0.08$) to be greater for CON compared to their counterpart supplemental treatments. Digestible and urinary energy did not differ ($P \geq 0.57$) among treatments. However, urinary N loss was affected ($P = 0.02$; Table 3) with BF and COMBO having greater losses compared to CAP and CON.

Circulating serum glucose concentration was not influenced ($P \geq 0.47$, Table 4) by supplementation in both jugular and saphenous veins samples. The addition of RUP supplementation in BF and COMBO resulted in increased ($P < 0.01$) circulating SUN compared to CON and CAP. In pooled samples across treatments, serum concentrations of SUN were observed ($P < 0.01$) to be lower pre-prandial compared to post-prandial concentrations. No differences ($P = 0.27$) were observed among treatments for jugular serum NEFA concentrations. However, a difference in timing of sample taken was observed ($P < 0.01$), with serum NEFA concentrations being greater at pre-prandial draw

compared to post-prandial. No treatment by time interactions were observed for serum concentrations of glucose, SUN, or NEFA ($P > 0.05$).

Total circulating serum AA concentrations were increased ($P < 0.01$; Table 5) in lambs consuming BF and COMBO supplemental treatment. Inclusion of RUP supplement in BF and COMBO resulted in an increased ($P < 0.01$) concentration of essential amino acids (EAA) compared to CAP and CON. However, non-EAA were not influenced ($P = 0.40$) by supplemental treatments. Glucogenic, ketogenic, and AA that are both glucogenic and ketogenic were increased ($P < 0.01$) in wethers fed BF and COMBO. Measuring free amino acids individually, all were greater ($P < 0.05$; Table 6) in wethers consuming BF and COMBO except for histidine, glutamine, and tyrosine. Pooling supplemental treatments, time impacted ($P < 0.01$; Table 7) total AA, non-essential AA, and ketogenic AA concentrations. Total AA and non-essential AA were greater at 4 h post-prandial. Ketogenic AA concentrations were decreased post-prandial. Essential AA concentration tended ($P = 0.09$) to decrease post-prandial. Location also impacted circulating serum AA concentrations. Total AA, essential AA, non-essential AA, glucogenic, ketogenic, and AA that are both glucogenic and ketogenic concentrations were decreased ($P < 0.01$) in samples from the saphenous vein compared to the jugular vein. Specifically, free AA concentrations for glycine, valine, isoleucine, threonine, serine, proline, asparagine, methionine, phenylalanine, glutamine, ornithine, lysine, tyrosine, and tryptophan were observed to be lower ($P < 0.05$) in samples taken from the saphenous compared to their jugular counterparts.

Acetate half-life was not influenced ($P = 0.39$; Table 8) by supplemental treatments. However, acetate AUC was influenced ($P = 0.04$) by supplemental treatments. Wethers on BF and COMBO supplements had decreased ($P \leq 0.04$) acetate AUC compared to CON wethers. Whereas wethers fed CAP had a tendency ($P = 0.08$) to have a decreased AUC compared to CON. However, glucose and insulin AUC were not different ($P = 0.80$ and 0.84 ; respectively) among supplemental treatments.

Rumen ammonia concentration was affected ($P < 0.01$; Table 9) by supplementation. Wethers fed BF had greater ($P < 0.01$) ruminal ammonia concentration compared to CAP and CON and tended ($P = 0.10$) to be greater than COMBO. Control and CAP supplemental treatments did not differ ($P = 0.84$) and were lower ($P < 0.01$) than COMBO. Total VFA concentration had a tendency ($P = 0.10$; Table 10) to be impacted by supplement. Acetate concentration was not influenced ($P = 0.61$) by supplemental treatments. However, supplementation had an effect ($P < 0.01$) on propionate concentration. Wethers receiving CAP and COMBO had greater ($P < 0.01$) ruminal propionate concentration than CON and BF. Control and BF did not differ ($P = 0.66$) in propionate concentration. In addition, ruminal butyrate concentration did not differ ($P = 0.76$) among supplementation treatments. Ruminal concentration of valerate differed ($P = 0.04$) among supplementation treatments. Wethers receiving BF and COMBO had greater ($P \leq 0.04$) concentration of valerate compared to CON. Wethers receiving CAP had similar ($P \geq 0.30$) valerate concentrations to CON and BF and tended ($P = 0.08$) to be lower than COMBO. Acetate to propionate (A:P) ratio was affected ($P < 0.01$) by supplement. Wethers fed CAP had a lower ($P < 0.01$) A:P ratio than those

receiving BF or CON but did not differ ($P = 0.58$) in A:P ratio from COMBO. Control and BF treatments did not differ ($P = 0.77$) in A:P ratio.

Discussion

Protein supplementation has been shown to increase intake and digestibility of low-quality forages (Owens et al., 1991) and increase rate of fermentation and microbial protein flow to the small intestine (Kunkle et al., 2000). While the current study offered forage at 2% BW, an increase in forage intake was observed in the control group compared to the treatments receiving supplementation. Supplementation of RUP in both BF and COMBO increased DM and OM total tract digestibility compared to treatments without additional RUP supplementation. The similar total tract OM digestibility between CAP and CON agrees with results by others that observed no differences in OM or NDF digestibility when supplementing with varying glucogenic precursor sources (Vanhatalo et al., 2003; Sanchez et al., 2014). However, increased DM and OM digestibility with RUP supplementation has been shown in sheep consuming low-quality forages (Ferrell et al., 1999). In the current study, NDF digestibility was not influenced by supplemental treatments; however, supplementation did influence ADF digestibility. Greatest ADF digestibility was observed in wethers consuming the BF supplementation but was not different from wethers fed COMBO. Wethers receiving no supplementation (CON) had an ADF digestibility intermediate to COMBO and CAP. This suggests that the RUP has a greater effect on ADF digestibility compared to propionic salt supplementation only. In contrast, Reed et al. (2007), supplemented steers on low-quality grass hay and reported no differences in total tract digestibility between no supplement

and RUP supplement groups. However, RUP supplemented steers did have a tendency for improved ruminal ADF digestibility compared to the no supplement control.

Urinary N excretion was increased in wethers receiving BF and COMBO compared to CON and CAP. In agreement, Salisbury et al. (2004) reported increased urinary N excretion in lambs receiving supplemental RUP compared to their counterparts receiving no supplement. While not statistically significant, the numerical decrease between BF and COMBO is similar to results from Ørskov et al. (1999) who observed reductions in urinary N excretion when glucose was infused intragastrically.

Serum glucose concentrations were similar among all treatments in this study. In agreement, Jenkins and Thonney (1988) reported no difference in plasma glucose concentration with increasing GP of diet. In contrast, Mulliniks et al. (2011) reported circulating serum glucose concentrations increased linearly with increased consumption of glucogenic precursors in young, lactating range cows grazing low-quality forage. Reed et al. (2007) reported no difference in blood glucose concentrations among steers being supplemented with increasing levels of protein. However, Reed et al. (2007) did observe an increase in SUN due to protein supplementation. Similarly, the current study had increased circulating SUN concentrations in wethers receiving RUP supplementation. Similarly, SUN concentrations were greater in wethers consuming a low-quality hay with RUP supplementation compared to no supplement (Ferrell et al., 1999).

Once N requirements of the ruminant have been met, additional AA can contribute to protein accretion or be oxidized (Lobley, 1992). Due to the lack of glucose absorbed through the small intestine of a ruminant, gluconeogenesis is a continual

process occurring in the liver of ruminants in constant need of substrates. Amino acids are estimated to contribute 5 – 7 % of glucose produced through gluconeogenesis by the ruminant (Engelking, 2015). In times of high demand or when dietary intake does not meet nutrient requirements, muscle can be mobilized and broken down into AA for utilization. These AA can then be utilized as glucogenic precursors for gluconeogenesis (McCabe and Boerman, 2020). Infusion of casein as a glucogenic precursor source resulted in increased essential AA and branch chained AA in lactating dairy cows (Vanhatalo et al. 2003). Infusing casein in pregnant ewes also increased circulating glucose concentrations compared to control (Barry and Manley, 1985). Similar results were reported in the current study with increased circulating serum essential and glucogenic AA concentrations in wethers receiving BF and COMBO supplementation. Increased circulating serum glucogenic AA concentrations reflected this observation of greater essential AA concentrations. Vanhatalo et al. (2013) observed greater milk lactose production when essential AA concentrations were increased supporting the contribution of glucogenic AA towards gluconeogenesis. However, this utilization of AA for energy can be detrimental to protein accretion. Barry and Manley (1985) reported decreases in fetal growth on a diet of kale that required mobilization of muscle to maintain pregnancy. Optimal metabolizable energy balance was reported when a combination of glucose + casein was infused (Barry and Manley, 1985). This suggests that a balance of energy and N must be met to maximize AA efficiency. Infusing low levels of glucose into fasted steers decreased urinary N excretion suggesting that inclusion of glucose or a glucogenic precursor would create a protein sparing effect (Ørskov and MacLeod, 1990). This protein sparing effect may act in two ways: by

allowing AA or N to go towards accretion instead of energy or prevent catabolism of muscle for energy. While both BF and COMBO, had greater AA concentrations than CAP, the addition of calcium propionate to COMBO did not further improve the AA utilization above BF.

Acetate clearance can be used as an indication of the GP of a diet and reveals efficiency of oxidative metabolism (Cronjé et al., 1991). Acetate half-life was not influenced by GP of diets in the current study. In contrast, Mulliniks et al. (2011) reported increased clearance rate of acetate when increasing levels of GP were supplemented. Acetate half-life in the current study were similar to those reported in previous studies where animals were consuming low-quality forage diets (Cronjé et al., 1991; Endecott et al., 2012). However, acetate half-life has been reported to be as quick as 10 min (Preston and Leng, 1987), approximately 2.5 to 3 times quicker than reported in this current study, suggesting that opportunities exist to increase oxidative metabolism. The decrease observed in acetate AUC for BF and COMBO compared to the CON suggests that meeting RUP requirements improved acetate utilization. A tendency for CAP to have a decreased acetate AUC compared to CON suggests that the increased GP of the diet will improve acetate uptake, but RUP requirements may need to be met to improve acetate utilization. The tendency for improved acetate utilization with increasing levels of GP and no change in circulating serum glucose concentration, suggests that the wether is capable of maintaining glucose concentration in circulating serum even with increased dietary GP (Kaneko, 1989). Yet, the increased uptake of

acetate leads us to conclude that peripheral tissue concentration of glucose is increased with increasing levels of dietary GP.

Total VFA concentration tended to be increased in CAP and COMBO fed wethers. Similarly, Sanchez et al. (2014) did not observe a difference in total VFA concentration when supplementing propionate as calcium propionate or *Propinobacterium* in heifers fed low-quality hay. Calcium propionate supplementation in the current study resulted in greater production of propionate, the VFA with the greatest contribution to gluconeogenesis (Aiello et al., 1989). This resulted in a lower A:P ratio for CAP and COMBO supplemental treatments. Other studies supplementing propionate have also shown an increase in propionate production and decrease of A:P ratio (van Houtert and Leng, 1993; Sanchez et al., 2014). While not observed in the current study with BF supplementation others have observed an increase in propionate concentrations and decrease A:P ratio in supplementation of RUP to ruminants consuming roughages (DelCurto et al., 1990; Salisbury et al., 2004).

Ruminal ammonia N concentration was greatest for BF and COMBO, which is to be expected due to the nature of the protein supplementation resulting in greater N intake. Increased rumen ammonia N due to protein supplementation has been previously observed in protein supplementation on low-quality forages (Salisbury et al., 2004; Reed et al., 2007). DelCurto et al. (1990) supplemented steers consuming roughage with varying levels of protein and energy and observed an increase in ruminal ammonia N concentration for those being supplemented with high levels of protein. Sanchez et al. (2014) observed a tendency for ammonia concentration to be decreased in diets

supplemented with a propionate source compared to the control. In contrast, the current study had similar ammonia concentrations between CON and CAP at 5.30 and 5.17 mg/dL, respectively.

Implications

Results from this study would suggest supplementing protein increases circulating serum concentrations of glucogenic AA, which can be utilized in gluconeogenesis increasing supply of glucose. Increasing glucogenic precursors with rumen undegradable protein resulted in improved efficiency of nutrient and acetate utilization in growing lambs fed a moderate-quality hay. Providing propionate salts as a supplement in lambs consuming moderate-quality hay resulted in an increased propionate concentration resulting in a decreased A:P ratio. However, the increased propionate supply by providing propionate salts did not result in a protein sparing impact.

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Table 2.1. Total tract digestibilities and digestible energy for wethers supplemented with glucogenic precursors fed a forage-based diet

	Supplementation Treatment				SEM	P-value
	CON ¹	CAP ²	BF ³	COMBO ⁴		
DM						
Total intake ⁵ , kg/d	1.03 ^d	1.05 ^c	1.16 ^b	1.21 ^a	0.02	< 0.01
Digestibility, %	37.4 ^b	36.6 ^b	43.0 ^a	42.9 ^a	0.98	< 0.01
OM						
Total intake, kg/d	0.95 ^d	0.97 ^c	1.10 ^b	1.13 ^a	0.02	< 0.01
Digestibility, %	42.6 ^b	43.6 ^b	49.8 ^a	49.8 ^a	1.11	< 0.01
NDFD_{om}⁶						
Total intake, kg/d	0.70	0.70	0.70	0.70	0.02	0.98
Digestibility, %	44.8	45.2	45.8	45.3	1.28	0.93
ADF						
Total intake, kg/d	0.46 ^b	0.46 ^b	0.49 ^a	0.50 ^a	0.01	< 0.01
Digestibility, %	35.6 ^{bc}	35.4 ^c	39.2 ^a	38.5 ^{ab}	1.31	0.03
Digestible Energy, Mcal/kg	1.69	1.74	1.63	1.65	0.05	0.37

^{a-d}Means within a row with different superscripts differ ($P < 0.05$).

¹CON: No supplementation.

²CAP: Supplementation of 40 g of NutroCal (Kemin Industries Inc., Des Moines, IA).

³BF: Supplementation of 70 g of blood meal + 100 g of feather meal.

⁴COMBO: Supplementation of 40 g of calcium propionate + 70 g of blood meal + 100 g of feather meal.

⁵Total intake = basal diet + supplementation + mineral.

⁶NDF_{om} = ash-free NDF.

Table 2.2. Daily forage intakes for wethers supplemented with glucogenic precursors

	Supplementation Treatment				SEM	P-value
	CON ¹	CAP ²	BF ³	COMBO ⁴		
DM Intake, kg/d	1.01 ^a	0.99 ^b	0.98 ^b	0.99 ^b	0.02	0.02
OM Intake, kg/d	0.94	0.94	0.94	0.94	0.02	0.94
NDFD _{om} ⁵ Intake, kg/d	0.68	0.67	0.66	0.67	0.02	0.08
ADF Intake, kg/d	0.46	0.46	0.46	0.46	0.01	0.97

^{a-b}Means within a row with different superscripts differ ($P < 0.05$).

¹CON: No supplementation.

²CAP: Supplementation of 40 g of NutroCal (Kemin Industries Inc., Des Moines, IA).

³BF: Supplementation of 70 g of blood meal + 100 g of feather meal.

⁴COMBO: Supplementation of 40 g of calcium propionate + 70 g of blood meal + 100 g of feather meal.

⁵NDF_{om} = ash-free NDF.

Table 2.3. Effect of supplement on urinary energy and nitrogen losses for wethers consuming a forage- based diet supplemented with glucogenic precursors

Urinary loss	Supplementation Treatment				SEM	P-value
	CON ¹	CAP ²	BF ³	COMBO ⁴		
Energy, Mcal	0.80	0.98	1.08	1.04	0.19	0.71
Nitrogen, g	42.0 ^b	56.4 ^b	106 ^a	88.4 ^a	15.8	0.02

^{a,b}Means with differing superscripts are different ($P < 0.05$).

¹CON: No supplementation.

²CAP: Supplementation of 40 g of NutroCal (Kemin Industries Inc., Des Moines, IA).

³BF: Supplementation of 70 g of blood meal + 100 g of feather meal.

⁴COMBO: Supplementation of 40 g of calcium propionate + 70 g of blood meal + 100 g of feather meal.

Table 2.4. Impact of glucogenic precursor supplementation on serum metabolites of wethers fed a forage-based diet

Measurements	Supplementation Treatment				SEM	P-values		
	CON ¹	CAP ²	BF ³	COMBO ⁴		Trt	Time	Trt x Time
Jugular Glucose mg/dL	55.4	54.1	55.8	55.8	1.93	0.87	< 0.01	0.57
Saphenous Glucose mg/dL	56.7	54.8	55.5	58.0	1.84	0.47	< 0.01	0.16
Jugular SUN ⁵ , mg/dL	11.3 ^b	10.6 ^b	25.9 ^a	25.5 ^a	1.12	< 0.01	< 0.01	0.23
Saphenous SUN, mg, dL	11.6 ^b	11.2 ^b	25.7 ^a	25.2 ^a	1.09	< 0.01	< 0.01	0.13
Jugular NEFA ⁶ , mg/dL	3.06	2.86	2.79	2.73	0.12	0.27	< 0.01	0.45

^{a,b}Means with differing superscripts are different ($P < 0.05$).

¹CON: No supplementation.

²CAP: Supplementation of 40 g of NutroCal (Kemin Industries Inc., Des Moines, IA).

³BF: Supplementation of 70 g of blood meal + 100 g of feather meal.

⁴COMBO: Supplementation of 40 g of calcium propionate + 70 g of blood meal + 100 g of feather meal.

⁵SUN = serum urea N

⁶NEFA = non-esterified fatty acids

Table 2.5. Effect of supplemental treatment on circulating serum amino acid (AA) concentration

	Supplemental Treatment				SEM	<i>P</i> -value
	CON ¹	CAP ²	BF ³	COMBO ⁴		
Total AA, nMol/mL	131.5 ^b	125.8 ^b	150.9 ^a	148.1 ^a	9.43	< 0.01
EAA ⁵	19.45 ^b	18.96 ^b	34.78 ^a	33.66 ^a	2.34	< 0.01
Non-EAA ⁵	113.2	107.9	117.3	115.6	7.91	0.40
Glucogenic	36.69 ^b	36.01 ^b	52.48 ^a	51.66 ^a	3.72	< 0.01
Ketogenic	5.76 ^b	5.51 ^b	8.80 ^a	8.75 ^a	0.67	< 0.01
Gluco-Ketogenic	4.40 ^b	4.53 ^b	5.39 ^a	5.49 ^a	0.33	< 0.01

^{a-b}Means within a row with different superscripts differ ($P < 0.05$).

¹CON: No supplementation.

²CAP: Supplementation of 40 g of NutroCal (Kemin Industries Inc., Des Moines, IA).

³BF: Supplementation of 70 g of blood meal + 100 g of feather meal.

⁴COMBO: Supplementation of 40 g of calcium propionate + 70 g of blood meal + 100 g of feather meal.

⁵EAA = essential amino acids

Table 2.6. Effect of supplemental treatment on individual free amino acid (AA) concentration

Amino Acid, nMol/mL	Supplemental Treatment				SEM	P-value
	CON ¹	CAP ²	BF ³	COMBO ⁴		
Alanine	4.76 ^a	4.40 ^a	3.75 ^b	3.60 ^b	0.33	< 0.01
Glycine	9.10 ^b	9.58 ^b	11.31 ^a	11.72 ^a	0.84	< 0.01
Valine	6.42 ^b	6.10 ^b	16.86 ^a	15.85 ^a	1.13	< 0.01
Leucine	1.34 ^b	1.26 ^b	3.70 ^a	3.47 ^a	0.26	< 0.01
Isoleucine	1.44 ^b	1.38 ^b	1.90 ^a	1.84 ^a	0.17	< 0.01
Threonine	1.95 ^b	1.88 ^b	2.65 ^a	2.62 ^a	0.22	< 0.01
Serine	1.95 ^b	2.12 ^b	3.50 ^a	3.76 ^a	0.14	< 0.01
Proline	1.30 ^b	1.33 ^b	2.47 ^a	2.41 ^a	0.18	< 0.01
Asparagine	0.92 ^b	0.91 ^b	1.21 ^a	1.18 ^a	0.10	< 0.01
Methionine	0.20 ^a	0.19 ^a	0.16 ^b	0.17 ^{ab}	0.02	0.04
Phenylalanine	0.89 ^b	0.91 ^b	1.22 ^a	1.31 ^a	0.08	< 0.01
Glutamine	6.50	5.50	6.99	6.67	0.95	0.17
Ornithine	1.47 ^b	1.50 ^b	2.53 ^a	2.42 ^a	0.36	< 0.01
Lysine	4.44 ^b	4.20 ^b	5.12 ^a	5.28 ^a	0.45	< 0.01
Histidine	2.03	1.91	2.28	2.12	0.29	0.34
Tyrosine	0.91	0.96	1.00	1.05	0.08	0.13
Tryptophan	1.23 ^b	1.35 ^a	1.35 ^a	1.37 ^a	0.08	0.04

^{a-b}Means within a row with different superscripts differ ($P < 0.05$).

¹CON: No supplementation.

²CAP: Supplementation of 40 g of NutroCal (Kemin Industries Inc., Des Moines, IA).

³BF: Supplementation of 70 g of blood meal + 100 g of feather meal.

⁴COMBO: Supplementation of 40 g of calcium propionate + 70 g of blood meal + 100 g of feather meal.

Table 2.7. Effects of time (h) and location on amino acid (AA) concentrations

Conc., nMol/mL	Time ¹ , h		SEM	P-value	Location		SEM	P-value
	0800	1200			Jugular	Saphenous		
Total AA	125.5	152.6	8.76	< 0.01	147.6	130.6	8.72	< 0.01
EAA ²	27.74	25.69	2.18	0.09	29.48	23.95	2.17	< 0.01
Non-EAA ²	99.14	127.8	7.28	< 0.01	119.4	107.6	7.32	< 0.01
Glucogenic	43.27	45.15	3.43	0.33	47.55	40.87	3.44	< 0.01
Ketogenic	7.64	6.77	0.62	0.01	8.19	6.22	0.62	< 0.01
Gluco-Ketogenic	5.07	4.83	0.31	0.16	5.44	4.46	0.31	< 0.01
Free AA								
Alanine	3.64	4.61	0.31	< 0.01	4.21	4.04	0.31	0.34
Glycine	9.78	11.1	0.78	< 0.01	11.42	9.43	0.78	< 0.01
Valine	11.7	10.92	1.05	0.19	12.6	10.2	1.05	< 0.01
Leucine	2.48	2.40	0.24	0.49	2.79	2.09	0.24	< 0.01
Isoleucine	1.73	1.55	0.16	0.04	1.82	1.46	0.16	< 0.01
Threonine	2.18	2.36	0.21	0.12	2.49	2.06	0.21	< 0.01
Serine	2.71	2.95	0.10	0.09	3.13	2.53	0.10	< 0.01
Proline	1.95	1.80	0.17	0.08	2.08	1.67	0.17	< 0.01
Asparagine	1.23	0.86	0.09	< 0.01	1.13	0.96	0.09	< 0.01
Methionine	0.18	0.18	0.02	0.89	0.19	0.17	0.02	0.05
Phenylalanine	1.01	1.16	0.08	< 0.01	1.17	1.00	0.08	< 0.01
Glutamine	7.22	5.61	0.89	< 0.01	7.49	5.35	0.89	< 0.01
Ornithine	2.22	1.73	0.33	< 0.01	2.20	1.75	0.33	0.01
Lysine	5.13	4.39	0.42	< 0.01	5.41	4.11	0.42	< 0.01
Histidine	2.15	2.02	0.27	0.40	2.01	2.16	0.27	0.29
Tyrosine	0.94	1.02	0.07	0.03	1.09	0.87	0.07	< 0.01
Tryptophan	1.47	1.18	0.07	< 0.01	1.44	1.21	0.07	< 0.01

¹CON: No supplementation; ²CAP: Supplementation of 40 g of NutroCal (Kemin Industries Inc., Des Moines, IA).; ³BF: Supplementation of 70 g of blood meal + 100 g of feather meal.; ⁴COMBO: Supplementation of 40 g of calcium propionate + 70 g of blood meal + 100 g of feather meal.; ⁵EAA = essential amino acids

Table 2.8. Effect of supplement on acetate tolerance test for wethers consuming a forage-based diet supplemented with glucogenic precursors

Acetate tolerance test response	Supplementation Treatment				SEM	P-value
	CON ¹	CAP ²	BF ³	COMBO ⁴		
Acetate half-life, min	39	33	26	31	6	0.39
Acetate AUC ⁵	298 ^a	242 ^{ab}	205 ^b	228 ^b	24.3	0.04
Glucose AUC ⁵	310	310	326	316	15.7	0.80
Insulin AUC ⁵	31.5	32.8	36.8	32.1	5.06	0.84

^{a,b}Means with differing superscripts are different ($P < 0.05$).

¹CON: No supplementation.

²CAP: Supplementation of 40 g of NutroCal (Kemin Industries Inc., Des Moines, IA).

³BF: Supplementation of 70 g of blood meal + 100 g of feather meal.

⁴COMBO: Supplementation of 40 g of calcium propionate + 70 g of blood meal + 100 g of feather meal.

⁵AUC: area under curve.

Table 2.9. Impact of glucogenic precursor supplementation on rumen parameters of wethers consuming a forage-based diet

Parameter	Supplementation Treatment				SEM	P-value
	CON ¹	CAP ²	BF ³	COMBO ⁴		
Total VFA Conc., mMol	49.7	58.1	45.5	56.3	3.91	0.10
Acetate, %	70.6	54.7	69.2	55.4	2.34	0.61
Butyrate, %	6.72	5.52	6.62	5.33	0.26	0.76
Propionate, %	20.3 ^b	37.9 ^a	20.1 ^b	36.1 ^a	1.51	< 0.01
A:P Ratio	3.51 ^a	1.56 ^b	3.47 ^a	1.63 ^b	0.10	< 0.01
Ammonia, mg/dL	5.30 ^b	5.17 ^b	9.70 ^a	8.62 ^a	0.46	< 0.01

^{a,b}Means with differing superscripts are different ($P < 0.05$).

¹CON: No supplementation.

²CAP: Supplementation of 40 g of NutroCal (Kemin Industries Inc., Des Moines, IA).

³BF: Supplementation of 70 g of blood meal + 100 g of feather meal.

⁴COMBO: Supplementation of 40 g of calcium propionate + 70 g of blood meal + 100 g of feather meal.

CHAPTER III. Effect of glucogenic precursor supplementation on postpartum cow
performance

T. M. King[§], J. K. Beard[§], J. C. MacDonald^{*}, J. A. Musgrave[§], and J. T. Mulliniks[§]

^{*}Department of Animal Science, University of Nebraska-Lincoln, Lincoln, NE 68583

[§]West Central Research and Extension Center, University of Nebraska, North Platte, NE
69101

Abstract:

Dietary consumption of low-quality forages by range beef cows results in an unfavorable acetate:propionate ratio, which negatively affects energy metabolism. The imbalance of acetate and propionate in lactating range cows can cause metabolic dysfunctions by incomplete oxidation and decreased reproductive performance. In a 2-yr study at the Gudmundsen Sandhills Laboratory near Whitman, NE, March-calving young range cows were individually supplemented with one of two supplements: 1) distillers grain-based range cube (CON) protein supplement or 2) CON with calcium propionate (CAP) incorporated to be provided at 40 g/hd/0.908 kg. Supplements were provided at a rate of 0.908 kg/d. Supplementation started approximately 10 d after parturition and continued through the first of June for an average of 70 d postpartum. Cow body weight (BW) and body condition score (BCS) were collected weekly. Weekly blood samples were taken beginning 45 d postpartum. An acetate tolerance test was conducted to determine acetate utilization. Cow BW, BCS, and pregnancy rate were not different ($P > 0.05$) between supplementation treatment. Calf pre-breeding BW, pre-weaning ADG, and 205-d calf BW were not influenced ($P \geq 0.40$) by dam supplementation. Cows receiving CAP

tended to have decreased ($P = 0.07$) circulating serum β -hydroxybutyrate concentrations compared to CON. Postpartum supplements did not influence ($P > 0.05$) non-esterified fatty acid concentration. Acetate area under the curve (AUC) from the acetate tolerance challenge was not impacted ($P = 0.18$) by supplementation treatments. Acetate half-life had a tendency ($P = 0.09$) to be greater for CAP than CON. Milk production was not influenced ($P = 0.39$) by postpartum supplements. The results from this study suggest that supplying a protein supplement with additional calcium propionate did not improve cow BW or BCS. However, addition of calcium propionate tended to decrease β -hydroxybutyrate concentration indicating lower ketone production in young postpartum cows suggesting improved dietary energy utilization.

Key words: cow-calf performance, glucogenic potential, postpartum supplementation

Introduction

Lactation of first- and second-calf cows is a time of high nutrient demand due to the increased requirements of lactation as well as growth. These high demands may not be met when cows are grazing dormant forage resulting in a negative energy balance postpartum. Consuming dormant range favors acetate production resulting in a shift in the acetate:propionate ratio which can negatively affect energy metabolism (McCollum and Galyean, 1985; Cronjé et al., 1991). Cows in negative energy balance result in decreased fertility (Wathes et al., 2007) and loss of body condition score (BCS) and body weight (BW). Supplementation to increase glucogenic activity through glucogenic precursors increase the glucogenic potential (GP) of the diet altering gluconeogenesis and energy metabolism ensuring metabolic function and reproductive performance (Hawkins

et al., 2000). Providing an additional or increasing source of energy can allow cows to repartition energy during this critical period meeting lactation demands and allowing increased availability of nutrients to go towards repair of reproductive tissue. Previous studies (Hunter and Magner, 1988; Wiley et al., 1991; Mulliniks et al., 2011) have observed improvement in return to estrus when supplementing with RUP. Our hypothesis was that increasing the GP of the diet would result in maintenance or improvement of BCS and BW and improve reproductive performance of young cows. The objectives of this study were to determine the impact of increasing supplemental GP on young cow BW change, BCS, energy metabolism, pregnancy rate, and calf weaning BW.

Materials and Methods

All animal care and management procedures were reviewed and approved by the University of Nebraska Institutional Care and Animal Use Committee (IACUC approval number 1474).

This study was conducted over a two-year period (2019 – 2020) utilizing cows from the March calving herd at the University of Nebraska Gudmundsen Sandhills Laboratory (GSL) located near Whitman, NE. Supplementation was offered for an average 70 d postpartum. Cows (n = 125) were Husker Reds (5/8 Red Angus, 3/8 Simmental) in their first or second parity. Cows were stratified by pre-calving BW (late December; 470.2 ± 46.9 kg) and assigned randomly to a supplementation treatment upon calving. Supplementation (Table 1) was provided at a rate of 0.908 kg/d with treatments being: a) dried distillers grain-based range cube (**CON**; Farmers Ranchers Co-op,

Ainsworth, NE) or b) CON with calcium propionate (**CAP**; Kemin Industries Inc., Des Moines, IA) incorporated into the cube to provide 40 g per hd per 0.908 kg. In year one, 50 3-yr-old cows were placed in a dry lot and fed meadow hay (Table 2) with a bale processor daily at a rate of 13 kg/hd. In yr 1, cows were individually supplemented in stanchions twice a week (Monday and Friday) at 0800 h. In year two, 75 2- and 3-yr-old cows were placed into an adjacent pasture (19.4 ha) upon calving. Cows were allowed to consume range and meadow hay was provided with a bale processor at a rate of 13 kg/hd. Supplement was offered daily by Super SmartFeed (SSF; C-Lock Inc., Rapid City, SD). The SSF is an electronic individual feeding system that drops an allotted amount of feed upon reading the cow's electronic identification (EID) tag. Once allotted daily supplement was consumed, cows were not dispensed any more supplement. Average daily intake of the supplement across throughout the feeding period was 0.858 kg. The average frequency of visits was 92% across the trial. Cows that failed to visit the feeder 85% of their days on trial, were removed from the trial. This resulted in removal of data for 5 cows (3 CON, 2 CAP; respectively).

Hay samples were taken weekly upon delivery in the pen with samples taken from a minimum of four areas in the feed row. Samples were composited and placed in a forced air oven for 72 h at 60°C. In yr 2, range samples were collected due to the placement of cows on range pastures. Range quality samples were collected monthly via hand clipping at 4 various locations throughout the pasture. Samples were then ground through a 2-mm screen of a Wiley mill and composited by month. Monthly samples were ground through a 1-mm screen of a Wiley mill and analyzed for organic matter

(OM), ash-free neutral detergent fiber (NDF_{om}), acid detergent fiber (ADF), and crude protein (CP). Analysis for NDF_{om} and ADF was conducted using the beaker method (Van Soest et al., 1991). Protein content was determined utilizing a LECO N analyzer (LECO, St. Joseph, MI).

Cow body weight (BW) and body condition score (BCS; 1 = emaciated, 9 = obese; Wagner et al., 1988) were recorded once weekly upon placement onto trial. Cow BW were taken at 0830 h, prior to hay being provided. Cows were not restricted from feed or water prior to weighing and BW were not shrunk. Two trained technicians were utilized for determination of BCS. Blood samples were taken weekly beginning 45-d postpartum via coccygeal venipuncture into serum separator vacuum tubes (Corvac, Kendall Healthcare, St. Louis, MO). Samples were centrifuged at $2,000 \times g$ at 4°C for 20 min. Serum was collected and stored at -20°C for analysis of β -hydroxybutyrate, NEFA, and serum urea nitrogen (SUN). Circulating serum β -hydroxybutyrate concentration was analyzed utilizing a colorimetric assay. A linear standard curve was determined using standards with varying known concentrations of DL- β -hydroxybutyrate acid sodium salt. Five μL of serum and 150 μL of buffer are added to each well and measured at 340 nm. Addition of 10 μL of β -hydroxybutyrate dehydrogenase was then added to each well and incubated at 37°C for 60 min. The plate was then read a second time at 340 nm. Data were first read subtracted from the second read and fitted to the linear curve to determine concentration. Cows were exposed to a fertile bull for a 45-d breeding season starting in June of each year. Pregnancy was detected via transrectal ultrasonography in October to determine reproductive performance of cows.

Calf BW was taken at birth within the first 24 h, pre-breeding, and weaning. No feed or water restriction or shrinking was applied for measurement of calf BW. Calves received a 7-way clostridial vaccine (Alpha 7, Boehringer/Ingelheim, Duluth, GA) at birth. Vaccinations for infectious bovine rhinotracheitis, bovine viral diarrhea types I and II, bovine parainfluenza virus-3, bovine respiratory syncytial virus, Mannheimia haemolytica, and Pasteurella multocida (Vista Once SQ, Merck, Kenilworth, NJ) and a 7-way clostridial vaccine (Vision 7, Merck, Kenilworth, NJ) were given at branding (late April). Bull calves were castrated at branding. Calves were weaned in October with calf BW adjusted to a 205-d age constant BW without adjusting for age of dam and sex of calf.

An acetate tolerance test (ATT) was conducted ~50 d postpartum on a subset of cows (yr 1: n = 15; yr 2: n = 12) to analyze acetate clearance as affected by GP of treatments. On the morning of the ATT, jugular catheters were inserted through which a 20% acetic acid solution was infused at 1.25 mL/kg of cow BW. Blood samples were then collected (~9 mL) -1, 0, 1, 3, 5, 7, 10, 15, 30, 60, and 90 min relative to infusion. Samples were placed in Corvac serum separator tubes, cooled, and centrifuged at 2,000 x g at 4°C for 20 min. Serum was collected and stored at -20°C for later analysis of acetate and glucose concentrations. Serum was filtered with a centrifugal filter device for 100 min at 4°C at 5,000 x g for deproteinization (Amicon Ultra-4 centrifugal device, Millipore Corp., Burlington, MA). Filtered serum was mixed at a 5:1 ratio with 25% metaphosphoric acid containing 2 g/L of 2-ethyl butyric acid as an internal standard. Acetate concentration was analyzed via gas chromatography adapted from the method of

Goetsch and Galyean (1983). The half-life of acetate was calculated as the time required for a 50% decrease from peak serum concentration (Kaneko, 1989). Serum acetate area under the curve (AUC) were calculated using the trapezoidal method.

On d ~60 d, milk production was determined using a modified weigh-suckle-weigh method described by Waterman et al. (2006) on a subset of cows (yr 1: n = 16; yr 2: n = 30). The day prior to measuring milk production, cows were separated from calves by 1000 h and allowed to suckle at 1700 h before being separated again. The next day cows were milked utilizing a portable milking machine (Porta-Milker, Coburn Company Inc., Whitewater, WI). Ten minutes prior to milking, cows were administered an injection of oxytocin (Vedo Inc., St. Joseph, MO) intramuscularly to facilitate milk letdown. Milking started at 0630 h and initiation of milking was recorded for individual cows. Cows were milked until machine pressure ceased to extract additional fluid and milk weight was recorded for calculation of 24-h production. An aliquot was collected and sent to (Heart of America DHIA, Kansas City, MO) for milk protein, butterfat, lactose, solids non-fat (SNF), and milk urea nitrogen (MUN) analysis.

Data were analyzed as a randomized block design using the MIXED procedure (SAS Inst. Inc., Cary, NC, USA). Cow served as experimental unit with supplemental treatment, year, and cow age set as fixed effects. Interactions which were not significant were removed from the model. Cow BW, BCS, β -hydroxybutyrate, and NEFA concentrations were analyzed as repeated measures with date of collection serving as a repeated factor with an autoregressive covariate structure. Acetate half-lives were estimated for each animal by regressing the logarithmically transformed acetate

concentrations over time (Kaneko, 1989). Area under the curves were determined for acetate and glucose using the trapezoidal summation method. Significance level was set at $P \leq 0.05$.

Results and Discussion

Cow BW did not differ ($P \geq 0.55$; Table 3) between supplemental treatments when reported at pre-calving, initiation of supplement, pre-breeding, end of supplementation, and weaning. In agreement, other studies reported no difference in cow BW with increasing GP in diets (Endecott et al., 2012; Mulliniks et al., 2011). Cow BCS were not influenced ($P \geq 0.58$) by postpartum supplementation treatments, prior to calving, at initiation of supplementation, pre-breeding or ending of supplementation.

At weaning, cow BW was not impacted ($P = 0.39$; Table 3) by supplementation treatments. However, cows fed CON supplement tended ($P = 0.10$) to have a greater BCS at weaning than their counterparts fed CAP. Pregnancy rate in yr 1 was not different ($P = 0.75$) between postpartum supplementation treatment. The lower pregnancy rate average of 74% observed in the current study is likely due to the harsh winter weather conditions observed in the winter of 2018 – 2019 leading up to calving season. This agrees with Waterman et al. (2006) who observed no difference in pregnancy rate between cows supplemented with protein supplement with and without calcium propionate. At pre-breeding, calf BW were not influenced ($P = 0.40$; Table 4) by supplemental treatments of dam. Calf pre-weaning ADG and 205-d adjusted calf BW did not differ ($P = 0.92$) by supplementation treatments of dam. Previous studies (Mulliniks

et al., 2011; Endecott et al., 2012) also reported no difference in calf weaning BW with increasing levels of GP supplementation to dam.

Circulating non-esterified fatty acid and β -hydroxybutyrate concentrations can be used to identify negative energy balance or ketosis as they indicate the mobilization of fat stores for energy in the form of ketones (Wathes et al., 2007). Cows fed CAP had a tendency ($P = 0.07$; Table 5) to have lower circulating serum β -hydroxybutyrate concentration. Patton et al. (2004) supplemented lactating dairy cows with calcium propionate and reported a numerical decrease in β -hydroxybutyrate compared to cows receiving no supplementation. In agreement, Mulliniks et al. (2011) reported a decrease in β -hydroxybutyrate with increasing GP supplementation. In contrast, Endecott et al. (2012) reported no differences in circulating serum β -hydroxybutyrate concentrations in young range cows fed increasing levels of GP. The decreased β -hydroxybutyrate concentrations would suggest that increasing GP in the current study improves acetate utilization, resulting in decreased production of ketones (Endecott, 2006). In contrast, postpartum supplemental treatments did not influence ($P = 0.63$) circulating serum NEFA concentrations. These results agree with other studies who reported no impact of increasing GP on serum NEFA concentrations (Waterman et al., 2006; Mulliniks et al., 2011; Endecott et al., 2012).

Supplementation treatment did not influence ($P = 0.18$; Table 6) acetate AUC. However, cows receiving CAP tended ($P = 0.09$) to have a longer acetate half-life than those receiving CON. However, this result does contradict the tendency for decreased circulating β -hydroxybutyrate concentrations for cows supplemented with CAP. In

contrast, Mulliniks et al. (2011) reported acetate half-life to decrease with increasing GP of the diet. In addition, Cronjé et al. (1991) reported increased rate of acetate clearance when providing GP in the form of protein and propionate. Results from the current study did not support our hypothesis that increasing level of GP would improve acetate utilization. In agreement, Sanchez et al. (2014) reported greatest numerical acetate half-life values when supplementing with increasing levels of GP. Endecott et al. (2012) reported no effect of calcium propionate in acetate half-lives. Therefore, it is necessary to recognize that varying forms of precursors are not equally used in gluconeogenesis which may explain why CON had a tendency for decreased acetate half-life compared to CAP.

No differences ($P = 0.39$; Table 7) in 24-h milk yield were found between CAP and CON. Similar results were reported by others (Patton et al., 2004; Mulliniks et al., 2011) with no difference in 24-h milk production with increasing supplementation of GP. Milk fat, protein, lactose, and SNF content were not affected ($P > 0.05$; Table 7) by supplemental treatments. In agreement, Mulliniks et al. (2011) reported no differences in milk constituents with increasing levels of supplemented GP. Cows fed CAP had a tendency ($P = 0.06$) to have a lower milk urea nitrogen concentration than their counterparts fed CON. Milk urea nitrogen concentration is associated with the ratio of protein and energy intake and can be associated with efficiency of protein metabolism (Oltner et al., 1985). High concentrations of milk urea nitrogen are associated with excess protein, excess rumen degradable protein (RDP), or inadequate energy affecting efficiency of protein utilization (Jonker et al., 1998). This may indicate in the current

study that the addition of calcium propionate improved the protein efficiency of dietary protein.

Implications

Postpartum supplementation strategies did not influence cow BW or BCS after calving. Addition of calcium propionate to a protein supplement resulted in a decrease of β -hydroxybutyrate concentration indicating lower ketone production suggesting improved energy efficiency with increasing GP. However, this improvement did not result in improvements in pregnancy rate.

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Table 3.1. Nutrient profile¹ (as-fed basis) of protein supplements with and without calcium propionate fed in 2019 and 2020

	CON	CAP ²
Dry Matter, %	90.4	90.4
Crude Protein, %	29.8	29.0
Non-protein Nitrogen, %	4.15	4.00
RUP ³ , (% CP)	39.7	39.7
RDP ⁴ , (% CP)	60.4	60.4
Crude Fat, %	4.64	4.50
Crude Fiber, %	6.01	7.0
Zinc, ppm	147.0	147.0
Copper, ppm	32.7	32.0
Manganese, ppm	86.1	86.0
Vitamin A, IU/kg	22,750	22,026

¹Nutrient analysis provided from Farmers Ranchers Co-op, Ainsworth, NE.

²CAP = 40 g of calcium propionate (Kemin Industries, Inc., Des Moines, IA) added to supplement.

³Rumen undegradable protein as a % of crude protein.

⁴Rumen degradable protein as a % of crude protein.

Table 3.2. Feed analysis for forages fed 2019 – 2020

	Hay (Range ¹) Quality	
	2019	2020
Organic Matter, %	91.2	93.1 (93.3)
NDF _{om} ² , %	74.2	74.1 (80.1)
Acid Detergent Fiber, %	45.0	43.6 (36.7)
Crude Protein, %	6.96	--

¹Quality sample for range was taken as cows were allowed to graze range as the season progressed.

²NDF_{om} = ash-free neutral detergent fiber.

Table 3.3. Supplementation effects on cow body weight (BW), body condition score (BCS), and reproductive performance for 2- and 3-yr-old postpartum cows receiving grass hay and fed protein supplement with and without calcium propionate in 2019 and 2020

	CON	CAP ¹	SEM	<i>P</i> -value
Pre-calving cow BW, kg	466.0	461.2	6.45	0.57
Initial cow BW ² , kg	426.0	424.3	6.49	0.85
Initial cow BCS ²	5.25	5.28	0.05	0.65
Pre-breeding cow BW, kg	422.8	418.0	6.76	0.59
Pre-breeding BCS	5.08	5.05	0.05	0.58
Ending cow BW ³ , kg	414.0	408.8	6.72	0.55
Ending cow BCS ³	5.17	5.18	0.04	0.89
Weaning Cow BW ⁴ , kg	442.3	428.3	11.60	0.39
Weaning Cow BCS ⁴	5.12	4.93	0.08	0.10
Pregnancy Rate ⁴ , %	76.0	72.0	0.09	0.75

¹ CAP = 40 g of calcium propionate (Kemin Industries, Inc., Des Moines, IA) added to supplement.

²Initial = initiation of supplementation.

³Ending = cease of supplementation period

⁴Weaning cow BW and cow BCS represent yr 1 data only.

Table 3.4. Supplementation effect on gains and calf BW for calves from dams being supplemented protein with and without calcium propionate

	CON	CAP ¹	SEM	<i>P</i> -value
Pre-breeding calf BW, kg	66.0	67.8	1.60	0.40
Pre-weaning ADG ² , kg	0.801	0.799	0.017	0.92
205-d Calf BW ² , kg	164.3	163.8	3.49	0.92

¹ CAP = 40 g of calcium propionate (Kemin Industries, Inc., Des Moines, IA) added to supplement.

²Weaning performance was reported for 2019 only; weaning calf BW was adjusted to a common 205-d BW.

Table 3.5. Supplement effects on acetate tolerance test in 2019 for 3-yo cows receiving grass hay and protein supplement with and without calcium propionate

	CON	CAP ¹	SEM	<i>P</i> -value
Acetate AUC ²	186.0	213.8	14.24	0.18
Acetate Half-life, min	45.9	63.4	7.10	0.09

¹ CAP = 40 g of calcium propionate (Kemin Industries, Inc., Des Moines, IA) added to supplement.

²AUC = area under the curve.

Table 3.6. Supplement effects on serum metabolites in 2- and 3-yo cows receiving grass hay and protein supplement with and without calcium propionate

	TRAD	CAP ¹	SEM	<i>P</i> -value
β-hydroxybutyrate, μmol/L	185.5	166.1	7.83	0.07
NEFA ³ , μmol/L	124.9	129.3	4.81	0.36

¹ CAP = 40 g of calcium propionate (Kemin Industries, Inc., Des Moines, IA) added to supplement.

²Days postpartum

³NEFA = non-esterified fatty acids

Table 3.7. Supplementation and year effects on milk production of 2- and 3-yo-cows receiving postpartum RUP supplementation with or without calcium propionate

	TRAD	CAP ¹	SEM	<i>P</i> -value
Total Milk Yield, kg/d	4.25	4.60	0.40	0.39
Constituents, %				
Butterfat	2.75	2.58	0.16	0.45
Protein	2.72	2.63	0.07	0.23
Lactose	5.33	5.36	0.06	0.55
SNF ²	8.89	8.82	0.10	0.52
MUN ³	18.78	17.31	0.79	0.06

¹ CAP = 40 g of calcium propionate (Kemin Industries, Inc., Des Moines, IA) added to supplement.

²SNF = solids non-fat.

³MUN = milk urea nitrogen.

CHAPTER IV: Impact milk production on cow reproductive performance and calf
growth

T. M. King[§], J. A. Musgrave[§], J. C. MacDonald^{*}, R. N. Funston[§], and J. T. Mulliniks[§]

^{*}Department of Animal Science, University of Nebraska-Lincoln, Lincoln, NE 68583

[§]West Central Research and Extension Center, University of Nebraska, North Platte, NE
69101

Abstract:

As cow-calf producers focus on greater weaning weights, selection for increased production parameters including milk production and weaning weight have become prevalent. However, increased cow-calf production may not be captured due to environmental conditions and resource availability. A retrospective analysis was conducted to model the impact of milk production on utilizing data collected from a March calving herd ($n = 348$) from 2000 to 2018 in the Nebraska Sandhills. Cow body weight (BW) was collected in June, July, September, November, and January. Milk yield was determined at this time utilizing the weigh-suckle-weigh technique. A subset of calves ($n = 87$) entered a feedlot upon weaning and were fed a high-concentrate diet until slaughter. The objective of this study was to determine the impact milk production has on subsequent cow reproductive performance and calf performance throughout the pre- and post-weaning phases. Cow body weight (BW) and cow age increased ($P < 0.01$) average milk production throughout the lactation period. Pregnancy rate and subsequent calf birth date were not influenced ($P \geq 0.43$) by level of milk production. Increasing

dam milk production resulted in greater ($P < 0.05$) calf pre-weaning ADG and adjusted 205-d calf weaning BW. In addition, dam milk production increased ($P < 0.01$) steer progeny final live calf BW and hot carcass weight (HCW). Additionally, dam milk production tended to positively influence ($P = 0.06$) carcass yield grade. However, quality grade, marbling score, ribeye area, and backfat were not impacted ($P \geq 0.18$) by dam milk production. Calf performance from this study indicated for each additional kg of milk production calf weaning BW increased 6.3 kg. The greater adjusted 205-d calf weaning BW was maintained through the feeding period resulting in greater final live BW and HCW.

Key Words: beef cow, calf performance, milk production

Introduction

Livestock producers have tended to select for increased output traits like milk production and growth to increase productivity. Even with the increased selection for greater calf growth potential, some regions in the United States have seen a plateau in calf body weight (BW) at weaning (Lalman et al., 2019). When focusing on reaching maximum potential of these output traits, it is important to consider the multitude of variables that impact a production system. With increased milk production, nutrient requirements for cows become increased (Ferrell and Jenkins, 1984; Montaña-Bermudez et al., 1990), which may not be met if range and forage availability for grazing is already limited at meeting lactation demands.

Historically, weaning weight and milk production have been associated with a positive relationship with greater milk production resulting in heavier calves at weaning (Clutter and Nielsen, 1987; Abdelsamei et al., 2005). In contrast, others have only reported the benefit of increased milk production improving calf performance within the first 60 d after birth (Clutter and Nielsen, 1987; Ansotegui et al, 1991; Edwards et al., 2017). Gleddie and Berg (1968) reported the correlation between average daily gain (ADG) of calves and milk yield estimates increased between the first and second month and continued to decrease thereafter as forage consumption increased. The reliance on milk for dietary energy can result in increased calf BW at peak lactation (Edwards et al., 2017), but benefits of increased milk production may decrease as stage of lactation increases. Our hypothesis was that increasing milk production would negatively impact cow reproductive performance while increasing calf gains in the Nebraska Sandhills. Therefore, the objective of this study was to determine the impact milk production has on subsequent cow reproductive performance and calf performance throughout the pre- and post-weaning phases.

Materials and Methods

All animal care and management procedures were reviewed and approved by the University of Nebraska Institutional Care and Animal Use Committee (IACUC approval number 1474).

Data were collected between the years 2000 – 2018 from the March calving herd at the University of Nebraska Gudmundsen Sandhills Laboratory (Whitman, NE). Cows (n=348; ~20/yr) utilized were Husker Reds (5/8 Red Angus and 3/8 Simmental) and were

2 to 11 y of age (Table 1). The herd from which the data was collected was utilized as the Nebraska Ranch Practicum herd. Cows were maintained in the practicum herd and have data points from multiple years dependent on their pregnancy score and temperament due to the routine handling of the herd. Cows that failed to become pregnant were removed from the herd. In year 2000 and 2015 to 2018, cows were assigned to one of two grazing treatments: meadow or range. From years 2001 to 2014, all cows were grazed on upland range.

Cow Management

Cow body weight (BW) and body condition score (BCS; 1 = emaciated, 9 = obese; Wagner et al., 1988) were recorded in June, July, September, November, and January. Body condition scores were recorded by two trained technicians.

Milk production was estimated using the weigh-suckle-weigh technique (Green et al., 1991) in June, July, September, and November. Data collected from the month of June was utilized for pre-breeding variables. Calves were separated from cows by 1000 h and allowed to suckle at 1700 h before being separated again. Feed and water were restricted for cows and calves overnight. Calf BW were taken at 0700 h the following morning at which time cows and calves were paired up, allowing calves to suckle. Upon completion of suckling period (not exceeding 30 minutes), calves were weighed again. Difference in calf BW was calculated and used to extrapolate for milk production over 24 hr.

Cows were exposed for natural service for a 45-d breeding season. Bulls were Husker Red (~5/8 Red Angus, 3/8 Simmental) with moderate growth potential. Five days after bull turn out, cows received a single intramuscular shot of prostaglandin F_{2α} (25 mg; Lutalyse, Zoetis, Parsippany, NJ). Bull-to-cow ratio was 1:20. Transrectal ultrasonography was used each September for detection of pregnancy to determine reproductive performance of cows. Cows received 0.454 kg/d of a distillers-based range cube (32% CP (DM)) for 30 to 45-d beginning January 1 each year. Meadow hay was provided at a rate of 13.0 kg/hd/d in the winter months.

Pre-Weaning Calf Management

At birth, calves received a 7-way clostridial vaccine (Alpha 7, Boehringer/Ingelheim, Duluth, GA) and calf BW was recorded. Calves received vaccinations for infectious bovine rhinotracheitis, bovine viral diarrhea types I and II, bovine parainfluenza virus-3, bovine respiratory syncytial virus, Mannheimia haemolytica, and Pasteurella multocida (Vista Once SQ, Merck, Kenilworth, NJ) and a 7-way clostridial vaccine (Vision 7, Merck, Kenilworth, NJ) at branding. Bull calves were castrated at branding (late April). Calf BW were recorded in June, July, September, and November. Calves were weaned from September through December depending on forage availability. Due to the differences in weaning dates, calf BW at weaning was adjusted to a 205-d age constant BW without adjusting for age of dam and sex of calf. At weaning one vaccination of Vista Once SQ (Merck, Kenilworth, NJ) and 7-way clostridial vaccine with somnus (Vision 7 Somnus, Merck, Kenilworth, NJ) was given with the second dose of Vista Once SQ given 14 d later.

Post-Weaning Calf Management

A subset of calves (total n = 87; Table 2) were held in a drylot on *ad libitum* hay for 2 weeks postweaning and then shipped to a feedlot at the West Central Research and Extension Center (North Platte, NE). Upon arrival at the feedlot all steer calves were implanted with 14 mg of estradiol benzoate and 100 mg trenbolone acetate (Synovex Choice, Zoetis, Parsippany, NJ). Adapted over a 21-d period, calves were finished on a diet containing 48% dry rolled corn, 40% wet corn gluten feed, 7% ground grass hay, and 5% supplement. Calves were slaughtered at a commercial abattoir (Tyson Fresh Meats, Lexington, NE) when estimated to visually have 1.27 cm backfat (BF) and carcass data were collected 24 h post slaughter.

Statistical Analysis

Data collected throughout the lactation period were averaged (June – November) and used as variables in the regression analysis. When analyzing pre-breeding data, June cow BW, milk yield, and calf BW were used. Cow age and cow BW were included in the models as fixed effects due to their significant ($P < 0.01$) impacts on milk production. Julian date of birth and sex of calf did not significantly ($P \geq 0.08$) impact average milk yield and were removed from the average model. Julian date of birth did affect ($P = 0.02$) pre-breeding milk yield and was included in the model. Milk production was included as a fixed effect in the cow reproductive and calf growth models. Sex of calf and Julian date of birth were included in the calf growth and carcass characteristic regression models. Normal distribution was assumed for all models except for cow pregnancy rate which was analyzed as a binomial distribution. Year and cow served as

random effects in all models. Significance level was set at an $\alpha \leq 0.05$. All data were analyzed using R (R Core Team, 2017).

Results and Discussion

Cow Performance

Average milk production throughout the lactation period was positively influenced by cow BW and cow age ($P < 0.001$; Table 3). Every additional 100-kg increase in cow BW resulted in a 0.9 kg increase in milk production, which is greater than reported by McMorris and Wilton (1986) of 0.3 kg increase in milk production per every 100-kg increase of cow BW. In agreement, Vaz et al. (2016) reported cows with a greater milk yield to have a heavier BW at weaning. However, as days in lactation progressed, the nutrient demand of the greater milk production diluted the BW difference between the high milk producing and low milk producing groups due to the increased BW loss in the high milk producing group. In dairy cows, milk yield is increased in heavier or larger cows and the interval to peak milk yield is shorter (Roche et al., 2007). This increase in milk yield and decrease to peak lactation would increase the availability of nutrients from milk providing greater growth potential in the initial stage of the calf's life. Evaluating milk production at pre-breeding, average days postpartum would place the cows close to 60 d which is considered peak lactation. At this time, cow BW had a greater ($P < 0.01$) influence on milk production at peak lactation compared to the average influence across the lactation period.

In addition to cow BW, cow age has been shown to impact milk production within the first three lactations and plateau after that (Clutter and Nielsen, 1987). Furthermore, milk production has also been shown to decrease after 6 to 8 years of age (Lush and Shrode, 1950; Boggs et al., 1980). The current study reported an increase ($P < 0.001$; Table 3) of 0.02 kg in milk production per year of cow age. This result could be due to the young average age of the herd (~ 4 yr) which agrees closely to results by Lubritz et al. (1989) who reported increasing milk production as age increased in cows from 2 to 5 years of age. Rutledge et al. (1971) showed that the influence of age of dam on milk production resulted in an indirect impact on calf weaning BW.

A decrease in reproductive efficiency in young cows has been reported by others due to the metabolic demand caused by lactation (Mulliniks et al., 2013; Hobbs et al., 2017). Neither cow pregnancy rate nor subsequent calving date were impacted ($P \geq 0.43$; Table 4) by milk production. This agrees with results from McMorris and Wilton (1986) who reported no influence of milk production on gestation length and Berry et al. (2003) who reported no difference between milk yield and interval to first service. However, Edwards et al. (2017) reported a decrease in pregnancy rate in cows producing the greatest milk production (~12 kg / d) at peak lactation. Similar results were reported in dairy cows with the greatest milk yield by 100 d postpartum having lower pregnancy rates after the first service compared to their lower milk producing counterparts (Buckley et al., 2007). Negative energy balance is often observed during early lactation due to the demands of milk production and can cause a decrease in fertility. Butler (2000) observed an inverse relationship between milk production and fertility with decreasing fertility

observed as milk production increased. These results are in contrast to our current study, however the average milk production, throughout the data collection period (June to November), of 6.22 ± 1.85 kg per d may have not provided enough variance to detect a difference. At peak lactation, the average milk yield was 6.82 ± 2.41 kg per d for the current study. Furthermore, when considering the milk yield at peak lactation, the lack of differences in pregnancy rate were similarly reported by Edwards et al. (2017) who observed no impact of milk production on pregnancy rate in low (~ 6.6 kg) and moderate (~ 9.0 kg) producing cows.

Pre-Weaning Calf Performance

Increases in pre-breeding calf BW, adjusted 205-d calf weaning BW, and pre-weaning ADG were reported due to milk production. Pre-breeding calf BW was increased ($P < 0.05$; Table 5) by 1.13 kg for every additional kg increase in milk production. Pre-weaning ADG increased ($P < 0.01$) by 0.03 kg/d for every additional kg increase in milk production. Milk production has been shown to produce a 71.3% variance in calf ADG (Gleddie and Berg, 1968) or 66% variation in 8-month calf BW (Neville, 1962) with the greatest impact on variance reported at 60 d postpartum. While the initial 60-d postpartum is the most important, Neville (1962) reported a kg of milk was worth the same in months 7 and 8 postpartum as it is in 5 and 6 months postpartum. Additionally, as the nutrition plane of forage grazed improved, more milk was needed to produce a kg of calf gain in the later months. Beal et al. (1990) identified a correlation between individual milk production and preweaning calf growth, supporting the increase that was reported in pre-weaning ADG in the current study. This was reflected in

adjusted 205-d calf weaning BW increase ($P < 0.01$) of 6.32 kg of calf BW for every additional kg increase of milk production, which is slightly lower than the gain of 7.89 kg reported by Mulliniks et al. (2020). In contrast, Edwards et al. (2017) reported no differences in calf BW after ~d 58 postpartum, which may be due to differences in forage quality consumed by the suckling calves. After 60 d of age, calf pre-weaning ADG has been shown not to be different between dams with differing milk production levels (Clutter and Nielsen, 1987; Ansotegui et al, 1991; Edwards et al., 2017). However, in agreement with the current study, others have shown increased dam milk production results in greater calf BW at 205-d adjusted weaning (Clutter and Nielsen, 1987; Minick et al., 2001). Dams with a lower milk yield produced calves that averaged 10% less kg calf BW per 100 kg of cow BW at birth and 16.1% less kg of calf BW per 100 kg of cow BW at weaning (Vaz et al., 2016). The BW advantage was maintained in calves from heavier milk producing dams over their counterparts from dams with lower milk production. When evaluating the ratio of calf BW weaned to cow BW, dams with a higher ratio tended to have a greater milk production (Williams et al., 2018).

Post-Weaning Performance

Carcass characteristics after a finishing period has been shown not to be influenced by dam milk production (Lewis et al., 1990). In the current study, dam milk production had no impact ($P \geq 0.18$; Table 6) on backfat thickness or marbling score in progeny. Additionally, quality grade and ribeye area were not influenced ($P \geq 0.49$) by increasing dam milk production. However, yield grade tended ($P = 0.06$) to increase with increasing dam milk production. In agreement, Clutter and Nielsen (1987) reported

greater cutability in calves from dams with lower milk production. While, Clutter and Nielsen (1987) detected marbling scores to be greater in calves from high milk producing dams, the current study observed no difference in backfat or marbling score. Final live calf BW after the finishing phase increased ($P < 0.01$) by 10.6 kg for every additional kg increase in milk production. Davis et al. (1985) suggested that increasing milk production in lower producing Hereford cows would have improved the efficiency of post-weaning performance in their progeny. In contrast, Clutter and Nielsen (1987) reported that calves from dams with greater milk production lost their gain efficiency that was observed in pre-weaning upon entering the feedlot. The change in nutrition plane upon entering the feedlot often allows for a period of compensatory growth observed in calves from dams with lower milk production. The current study also observed HCW ($P < 0.01$) to increase with an additional 6.65 kg of HCW for every additional kg increase in milk production. These increases could be due to the impact of increased milk production on calf weaning BW resulting in heavier calves entering the feedlot. Lewis et al. (1990) reported increased weights of calves entering the post-weaning period from dams with higher-milk production. While numerically higher at slaughter, no significant differences were observed between calves from dams with low-, moderate-, and high-milk producing dams (Lewis et al., 1990). In addition, Clutter and Nielsen (1987) suggest that 63% of the weaning weight advantage was observed through slaughter. However, feedlot ADG was not impacted ($P = 0.47$) by dam milk production. In agreement with the current study, Abdelsamai et al. (2005) reported similar feedlot ADG, but the greater weaning BW calves consumed more milk and had decreased days on feed.

No differences due to milk production on ADG were reported by Lewis et al. (1990), however feed intake increased as milk production increased.

Implications

Results from the current study would suggest that greater cow BW will increase milk production, and selection for increased growth in the cowherd will indirectly increase milk production. Dam milk production had a positive influence on calf pre-weaning growth and BW with additional gains of 0.02 kg/d and 6.32 kg of additional weaning weight with every kg increase in average milk production. Therefore, it is important to consider the role milk intake has when striving to achieve greater calf weaning BW. Even with the greatest impact being reported in the first 60-d, milk production continued to be an important factor on calf growth throughout the pre-weaning phase. The greater BW at weaning in the offspring of dams with greater milk production, produced an advantage that tended to be maintained throughout the feeding period to produce steers with greater final live BW and HCW.

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Table 4.1. Demographics of cows utilized for data collection from 2000 – 2018 for average lactation period and pre-breeding season (June)

Measurement	Lactation Period Average ¹			Pre-breeding Average ²		
	Minimum	Maximum	Mean	Minimum	Maximum	Mean
Cow Age, yr	2	11	3.56	--	--	--
Cow BW, kg	283	856	455	263	819	425
Cow BCS	4.00	7.00	5.29	4.00	7.00	5.20
Milk Yield, kg/d	1.45	12.14	5.80	0.36	14.2	6.81
Julian Calving Date	53	123	79.5	--	--	--
Calf Birth BW, kg	22.7	52.7	35.2	--	--	--

¹Lactation period average accounts for June – November.

²Pre-breeding average is based on data collected in June.

Table 4.2. Number of steers entering the feedlot at West Central Research and Extension Center (North Platte, NE)

Year	Number of Calves
2009	9
2011	10
2012	10
2015	21
2016	21
2017	16

Table 4.3. Regression coefficient estimates used to determine the increase of cow demographics on milk yield (kg)

Measurement	Estimate ¹	SEM	P-value
<i>Average Milk Yield</i>			
Cow Age	0.02	0.07	< 0.001
Average Cow BW, 100 kg	0.91	0.17	< 0.001
<i>Pre-breeding Milk Yield</i>			
Julian Date of Birth	0.02	0.01	0.018
Cow Age	0.29	0.10	0.003
Average Cow BW, 100 kg	1.06	0.23	< 0.001

¹Estimates provide the increase or decrease response in the measured variable for every additional increase in fixed effect.

Table 4.4. Impact of milk production on cow demographics and reproductive performance

	Estimate	SEM	<i>P</i> -value
Avg Cow BW, kg	17.2	1.67	< 0.01
Avg BCS	0.03	0.01	< 0.01
Pregnancy Rate	0.003	0.35	0.99
Subsequent calving date	0.38	0.48	0.43

¹Estimates provide the increase or decrease response in the measured variable for every additional 1 kg increase in milk production.

Table 4.5. Regression coefficients used to estimate the increase on pre-weaning calf performance per kg increase of milk production

Measurement	Estimate ¹	SEM	<i>P</i> -value
Pre-breeding calf BW, kg	1.59	0.34	< 0.001
Pre-weaning ADG, kg/d	0.03	0.004	< 0.001
Adj. 205-d calf BW, kg	6.07	0.67	< 0.001

¹Estimates provide the increase or decrease response in the measured variable for every additional 1 kg increase in milk production.

Table 4.6. Regression coefficients used to estimate the increase on post-weaning calf performance and carcass characteristics per kg increase of milk production

Measurement	Estimate ¹	SEM	P-value
<i>Feedlot Live Performance</i>			
Feedlot ADG, kg/d	0.02	0.02	0.96
Final Live Calf BW, kg	10.6	3.51	< 0.01
<i>Carcass Characteristics</i>			
Hot Carcass Weight, kg	6.65	2.21	< 0.01
Quality Grade ²	-0.017	0.025	0.49
Yield Grade	0.105	0.055	0.06
Ribeye Area, cm	0.028	0.246	0.91
Marbling Score	2.37	5.98	0.69
Backfat, cm	0.042	0.030	0.18

¹Estimates provide the increase or decrease response in the measured variable for every additional 1 kg increase in milk production.

²Quality grade was assigned numerical values with 1 = Prime, 2 = Choice, etc.