MOLASSES AS AN ALTERNATIVE ENERGY SOURCE FOR DIETS USED IN BEEF PRODUCTION SYSTEMS

by

ANGEL ANTONIO RAUDALES QUIJADA

(Under the Direction of Francine Henry)

ABSTRACT

Commodities are a necessary purchase in cattle feeding operations; however, due to market fluctuations, costs may increase having a negative impact on the economic efficiency of such operations. Steers recently arrived at feedlot facilities are generally offered diets with elevated inclusions of forages and other commodities. These receiving diets are formulated to transition cattle to a high-grain diet. The receiving period influences performance and is an important phase prior to finishing. Stress can negatively impact cattle performance during the receiving phase, in which molasses may have the potential to mitigate stressors associated with transportation. Additionally, molasses can increase nutrient utilization by improving fiber degradation and has the potential to mitigate methane emissions. The objective of this study is to evaluate the effects of replacing soybean hulls with molasses in the receiving diets of beef steers on performance, nutrient digestibility, enteric methane emissions, blood inflammatory marker, and ruminal fermentation.

INDEX WORDS: molasses, soybean hulls, ruminal fermentation, backgrounding diets, methane emissions, stress, digestibility.

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ANGEL ANTONIO RAUDALES QUIJADA

Major Professor: Francine Henry
Committee: Darren Henry
Pedro Fontes
Lawton Stewart

Electronic Version Approved:

Ron Walcott
Vice Provost for Graduate Education and Dean of the Graduate School
The University of Georgia
December, 2023

DEDICATION

To my parents, brother, and grandparents

A.J. D.R.E.Q

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Proverbs 16:9: The heart of a man plans his way, but the Lord establishes his steps.

First, I want to thank God for his guidance and for the opportunities that he has blessed me with. I also would like to thank my advisor, Dr. Francine Henry and committee member, Dr. Darren Henry for believing in me and for being my mentors through my masters and I know for sure that both will keep being my mentors for my whole life, it was an honor and a pleasure to be part of their team. A special thanks to the interns and staff of the Animal & Dairy Science department.

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TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

Page

CHAPTER 1

INTRODUCTION AND LITERATURE REVIEW

Receiving Period of Feedlot Calves

The receiving period of newly weaned calves to a feedlot is a critical time that can impact the performance of beef cattle. Several factors will play an important role during this time period such as stress caused by transportation, weighing, sorting, and adaptation to feed bunks, waterers, and a new diet. Transportation can induce stress in cattle, which can reduce efficacy of the immune system and increase the risk of infection and diseases (Li et al., 2019). Stress can be defined as a biological response to a threat and also can be considered as an adverse effect in the environment or management that change the animal behavior(Damtew et al., 2018). Young calves are highly vulnerable to stress resulting in diarrhea, pneumonia, and fever (Damtew et al., 2018). After long transportation periods, cattle will arrive dehydrated, stressed, and weakened from this process (Van Engen & Coetzee, 2018). Due to the impact on cattle health, good management practices upon arrival are crucial to reduce the risk of diseases. The minimum amount of time of rest recommended prior to working cattle in facilities after arrival is 24 h (Felix et al., 2023). During this period cattle should be provided with clean water and long stem hay (Felix et al., 2023).Cattle that recently arrived at a feedlot will have a reduced DMI, where healthy calves will consume on average 3% of their BW, while highly stressed or unhealthy calves consume on average 1.5% or less of BW during the first 2 weeks of the receiving period (Reinhardt & Thomson, 2015). Usually, cattle arriving to the feedlot will have to adapt to higher energy diets containing grain and byproducts. The animal needs a good adaptation process due to these diets being highly digestible compared to forages. Ruminal acidosis is one of the main challenges during the receiving period due to irregular intake behavior that is mainly due to poor management (i.e, high stress environment in working facilities, transportation, and weaning times) and later an abrupt consumption of diet, by the fermentation of highly fermentable carbohydrates entering the rumen. Due to reduced ruminal pH caused by increased rate of acid production, which is faster than what the rumen wall and epithelium can absorb, incidence of ruminitis or inflammation of the epithelium may occur (Shearer, 2005). Digestive problems like acidosis can be an abundant cause of morbidity, reduced DMI, and diarrhea in confined cattle (Felix et al., 2023). Energy density is an important aspect of the nutritional requirements of recently received calves. Keeping a high forage diet such as hay with a low energy density may cause a negative impact on cattle performance, due to fiber sources being less digestible compared to other ingredients. Adding feedstuffs with greater energy density may result in increased efficiency of meeting nutritional requirements. In a study conducted by Lofgreen et al. (1975), newly-received, stressed calves were fed diets with different concentrations of net energy of gain (NEg; 0.84, 1.01 or 1.10 Mcal/kg). Cattle provided with high NEg (1.10 Mcal/kg) had greater ADG during the first weeks and also greater cumulative ADG throughout the 63 days of the study (Lofgreen et al.,1975). In a different study performed by the same authors they compared diets with 1.01, 1.10 and 1.19 Mcal/kg NEg and the diets with the two greatest NEg concentrations provided similar ADG during the first 28 d of the study (Lofgreen et al., 1980). Similarly, high stressed cattle were provided with diets containing 0.57, 0.75, and 0.90 Mcal/kg Neg (Lofgreen et al., 1980). Greater ADG from calves consuming 0.75 and 0.90 Mcal/kg NEg were observed when compared with calves fed a concentration of 0.57 Mcal/kg NEg (Lofgreen et al., 1980). Furthermore, it has been reported that cattle consuming a 75% concentrate diet had greater ADG compared with cattle consuming a native grass hay with a protein supplement (Lofgreen, 1987). However, Pritchard and Mendez (1990), reported an increase of 16% on ADG of cattle fed a high concentration of NEg (1.17 Mcal/kg) compared with a low concentration of NEg (1.01 Mcal/kg Additionally, a 7% increase in ADG with no difference in DMI was reported in calves consuming a high energy diet compared with a low energy receiving diet (Prichard et al, 1987).

Lipopolysaccharide (LPS) binding protein

Lipopolysaccharides (LPS) are an important component of the gram-negative bacteria outer membrane (Farhana et al., 2023). They are recognized for their role when they elicit a response in the host when there is a production of inflammatory cytokines, which are small proteins that play a role in cell signaling (Tucureanu et al., 2018). The LPS binding protein (LBP) binds to the lipopolysaccharide of the bacteria and interacts with a macrophage receptor that will initiate a pro-inflammatory host response (Bishara, 2012). Diverse stress factors including inflammation are going to increase the levels of LBP (Fang et al., 2023). Long transportation for up to 6 hours may increase the levels of rumen LPS in cattle (Li et al., 2019). In an experiment in which cattle where infected with gram-negative bacteria of *Mannheimia haemolytica (Pasteurella)* the LBP levels rose significantly 6 h after infection in the blood plasma, indicating that LBP may be a fast diagnostic marker for cattle sepsis (Schroedl et al., 2001). The presence of LPS in the body of the animals can elicit inflammatory responses by mammalian cells, in which free LBP accumulates during bacteria growth and lysis, increasing the concentration of LPS during sub-acute ruminal acidosis (Gressley, n.d). The basal levels of LPS binding protein in the blood of dairy cows ranged from 6 to 38 μg/ml(Bannerman et al., 2003)**.** Sub-acute ruminal acidosis increases bacterial lysis due to the changes on fermentation end products and reduced ruminal pH (Gressley, n.d).

Molasses

Molasses is the final byproduct obtained in the preparation of sucrose by different processes such as evaporation, centrifugation, and crystallization of juices coming from the sugar cane. There can be several types of molasses and in general it can be any liquid feed that contains at least a 46% of sugars (Curtin, 1983). In the literature, Madsen (1953) describes molasses as a product coming from sugar beets. Meade and Chem (1977) describe it as a product coming from the sugar cane. Molasses used for livestock diets and several other animal production areas has been recorded since the nineteenth century (Curtin, 1983). In the United States, the earliest document reporting the use of sugar cane molasses in livestock was published by Gulley and Carson (1890). The Association of American Feed Control officials (AAFCO) describes sugar cane molasses as a byproduct of the refining and processing of sugar cane for sucrose (Curtin, 1983). If the moisture content exceeds 27%, its density determined by the double dilution process must be at least 79.5^0 Brix international feed names and number 4-13-251 Sugar cane molasses (Pate, 1983). The process of obtaining molasses is divided in two forms: inedible for humans or as an edible syrup. Human edible molasses is blended with maple syrup, invert sugars or corn syrup. After processing the sugar cane and obtaining the molasses, different grades are assigned to molasses: grade A molasses, or liquor molasses and grade B massecuite. The molasses from this second process is of a lower quality and purity compared to the first molasses. Through a third process grade C massecuite is obtained, which later on separates into a third stage or grade C molasses. Grade C molasses, or blackstrap molasses, is a heavy and viscous liquid used as a supplement in the cattle industry (US Environmental Agency, 1993).

Molasses chemical composition shows wide variation as in many other industrial by-products (Palmonari et al., 2020). The sugar cane itself and its management will influence the chemical components of the molasses (Abadam, 2018). Aspects such as soil texture, climate, season, variety, process in the plant and storage will impact the quality of the molasses (Abadam, 2018). In molasses the term Brix is commonly used to determine the percentage of sucrose (Curtin, 1983). Other carbohydrates can be found on the molasses such as glucose, fructose, raffinose and other non-sugar organic materials (Curtin, 1983). Molasses does not contain a significant amount of crude protein (Palmonari et al., 2020). The nitrogenous chemicals in molasses will consist of amides, albuminoids, amino acids and other non-protein nitrogen compounds (Curtin, 1983). Sugar cane molasses can be high in minerals such as potassium, magnesium, sodium, chloride, and sulfur; carbohydrates and sugar will be the main components found in molasses (Curtin, 1983). Molasses is considered an energetic component for feed in livestock and can also be used to improve palatability in the diets (Curtin, 1983). Average nutritional composition of molasses is of 70% DM, 46% total sugars, 3% crude protein, 63% Nitrogen Free Extract, 8.1% Ash, 0.8% Calcium, 0.08% Phosphorus, 2.4% Potassium, 0.2% Sodium, 1.4% Chlorine, 0.5% Sulfur, 0.36 mg/kg Biotin; 745 mg/kg Choline; 21 mg/kg Vit. B5; 1.8 mg/kg Vit. B2; and 0.9 mg/kg Vit. B1 (Curtin, 1983). As far as energy composition, molasses contains on average 3.24 Mcal/kg digestible energy (DE), 2.82 Mcal/kg metabolizable energy (ME), 1.65 Mcal/kg NEl, 1.74 Mcal/kg NEm, 1.12 Mcal/kg Neg, and 75% TDN (NRC, 2001).

Molasses Production and Economic Impact

Molasses production in the United States was approximately of 3.5 million metric tons in the 1980´s (Curtin, 1983). Currently, molasses production has increased to up to 9.0 million metric tons, which can be attributed to an increase of the total sugar produced in the United States (Abadam, 2021). Since the 2000´s, sugarcane has accounted for a 40-45% of total sugar produced (Abadam, 2021) and production of sugar cane molasses comes from Florida, Louisiana, Texas, Hawaii, and Puerto Rico. The increase of sugar cane yields in the states of Florida and Louisiana is due to improvements on varieties of plants, harvesting, and processing technologies (Abadam, 2021). According to the Census of Agriculture (2017), the number of farms growing sugar cane has declined in the past two decades, while the area harvested has increased. The total farm numbers in the U.S has decreased from 4,714 to 4,123 in a range of 10 years within the last two decades (Abadam, 2021). Conversely, the amount of total acreage of sugar cane has increased from 704,00 in 1980 to 903,400 in 2020/2021 (Abadam, 2021). During the same period, the total short ton raw value (STRV) increased from 2.910 million to 4.251 million (Abadam, 2021). Due to the increasing and beneficial impact of the sugarcane industry, the molasses market is growing. The blackstrap molasses market size had a value of $12,889.9 million in 2019 and is estimated to increase up to $18,185.8 million by 2027 (Anil et al., 2020). Even though there is an evident increase in the blackstrap molasses and sugarcane market, during the COVID-19 pandemic the market was negatively affected. The lack of labor, global transportation, and factories not processing sugar cane are some factors that impact the economic efficiency of blackstrap molasses. In addition, the increase of production of blackstrap molasses is registering a compound annual growth rate of 5.7% from 2021 to 2027. By 2025, the animal feed sector of blackstrap molasses is likely to reach a 9.2% of compound annual growth rate (Market Data Forecast, 2023).

Effects of molasses on ruminal profile

Ruminal microorganisms. Molasses has the ability to alter the rumen microbiome. One of the most abundant species in the rumen is the *Prevotella ruminicola (*Flint et al., 1997), which can be reduced in population when molasses is being supplemented to cattle (Palmonari et al., 2023). *Prevotella ruminicola* is capable of processing a vast number of proteins and polysaccharides, having as an end product of fermentation the propionate (Betancur-Murillo et al., 2023). In addition, the inclusion of sugar in diets will not have a direct impact on some bacteria from the genus *Ruminococcus* (Palmonari et al., 2023), that are involved on the fiber digestibility and acetate production like the Ruminococcus albus (Cordero et al., 2020). Other populations such as the *Streptococcaceae* can have a greater relative abundance when molasses is included in diets (Palmonari et al., 2023), in which the species *Streptococcus bovis* is the microorganisms from the *Streptococcaceae* that increases when large fermentation of sugars occur and an increase in ruminal lactic acid (Fubini et al., 2018). *Streptococcus bovis* showed a greater presence when sugar cane molasses was included compared to a non-sugar content diet (Palmonari et al., 2023) Sugar can also increase the population of *Butyrivibrio fibrisolvens* bacteria, which allow pectins and fiber to be utilized as nutrients for the ruminants and increases levels of butyrate (Palmonari et al., 2023). In addition, the genus *Butyrivibrio* belongs to the *Lachnospiraceae* family (Palmonari et al., 2023).

Ruminal pH. Excessive intake of molasses can lead to metabolic issues such as ruminal acidosis due to a reduction on ruminal pH caused by high lactic acid levels and rapid fermentation of carbohydrates (Owens et al., 1996a). By increasing the concentration of hydrogen ions, pH decreases causing acidosis (Rusdiawan et al., 2020). Molasses is known to increase the population of *Butyrivibrio fibrisolvens* bacteria (Sun et al., 2020). Molasses has a relatively low pH content (4.8-5.5), is a highly fermentable ingredient, and if consumed in high quantities, may reduce the ruminal pH(Pate, 1983). In another study, the inclusion of a 5%, 10% and 15% of molasses in finishing diets did not affect ruminal pH (Hatch and Beeson, 1972). It has been reported that the highly fermentable sugars of molasses stimulate the rumen microflora because bacteria utilize the soluble sugars and this will improve fiber digestibility (Mordenti et al., 2021). In addition, butyrate formation generates only one H^+ whereas propionate and acetate generate 2 H^+, indicating that fermentation of sugars can promote a less drastic reduction on ruminal pH in comparison to starch, which is fermented into propionate (Mordenti et al., 2021) In a study in which different levels of molasses were supplemented to cattle, eating limpograss hay, the ruminal pH was affected regarding on the feeding time and by different concentration of inclusion of molasses in hay of 0 kg vs 0.9 kg vs 1.8 kg vs 2.7 kg per day of molasses (Abreu et al., 2022)

Volatile fatty acids profile. Molasses has a direct impact on the fermentative pathways in the rumen, which in turn affects the VFA profile (Palmonari et al., 2023). It has been observed that dietary molasses can increase total concentration of butyrate (Palmonari et al., 2023). Molasses generally increases butyrate production in the rumen, which stimulates the blood flow of the ruminal epithelium compared to other VFA (Malhi et al., 2013). An increase of molar percentages of butyrate was observed in the ruminal fluid of cattle fed alfalfa hay with an inclusion of 2.3, 4.1, or 5.1 kg of molasses (Pierson and Otterby, 1971). In another study an inclusion of 4.2% of molasses did not affect the concentration of VFA and pH (Martin and Wing, 1966). However, inclusion of up to a 15% of molasses increased the total VFA concentration of the ruminal fluid (Hatch and Beeson, 1972). In another study in which sugar was provided to cows fed hay or flaked corn the total VFA concentration also increased (Sutton, 1968). In addition, added quantities of dextrose to an artificial rumen medium increased the levels of butyrate and reduced the levels of acetate and propionate, whereas an added quantity of starch increased the levels of propionate and reduced the acetate and butyrate (Belasco, 1956). An in vitro inclusion of sugar showed an increase of bacteria *Butyvibrio fibrosolvens* which increased the levels of butyrate (Mordenti et al., 2021) One important aspect about butyrate is the ability to decrease excessive inflammation in the hindgut through the modulation of immune cells (Malhi et al., 2013). Butyrate has been studied for its anti-inflammatory effects, where it inhibits pro-inflammatory immune cells and neutrophils by reducing the production of cytokines and activation of anti-inflammatory cells (Chen & Vitetta, 2020). Butyrate also improves gut health by increasing ruminal epithelial cells growth and papillae length and width for a better absorption, which in turn can result on increased cattle performance (Malhi et al., 2013).

Effects of molasses on digestibility and dry matter intake

Despite being low in some vitamins like vitamin A and vitamin B6, molasses has a considerable amount of biotin, which can also aid on fiber degradation (Curtin, 1983). In a study in which 20 mg of biotin were supplemented to cows daily, an increase in milk production of 1-2.9 kg/hd/day was observed due to the increased fiber degradation (Chen & Vitetta, 2020). Molasses has organoleptic characteristics which increase the palatability of feed, which can help increase diet DMI (Abreu et al., 2022). The inclusion of molasses of up to 35% of the diet daily linearly increased the number of bacteria in the rumen (Foreman and Herman, 1953). Feeding molasses to cattle can improve digestion of fiber sources by increasing population of the bacteria such as *Butyrivibrio fibrisolvens and Fibrobacter succinogenes*(Alvarez & Ximena, 2007), resulting in less feed waste, as well as help maintain body condition.(Senthilkumar et al., 2016). Molasses can improve the quality of supplementation with poor quality forages due to its energy content (Abreu et al., 2022). Additionally, in cattle fed up to 1 kg of molasses daily, fiber digestibility was increased (Foreman and Herman, 1953). Foreman and Herman (1953) also suggested that molasses tended to increase the fiber digestibility of high-quality forages, and significantly increase the digestibility oh poor-quality forages.

Effects of molasses on cattle performance

Cane molasses has been used as a supplement that can improve digestibility of forage-based diets and performance of cattle (Moore et al., 1999) . In a study where zebu calves were supplemented with blocks of urea-molasses with yucca schidigera extract, had increased body weight gain and feed efficiency (Mirza et al., 2002). The addition of dried molasses to high-alfalfa silage-based diets increased milk urea nitrogen but there was no effect on animal performance (Baurhoo & Mustafa, 2014). It has been reported that inclusions of molasses between 100 to 150 g/kg of DM will not affect performance of beef steers, average daily gain and carcass weight; however, the inclusions above 200 g/kg of DM can negatively affect animal performance and carcass characteristics (de Nazaré Santos Torres et al., 2023). Body weight, carcass weight, subcutaneous fat thickness, kidney fat, pelvic fat, heart fat, and mesenteric fat of finishing steers have all been reduced with the inclusion molasses of 200 g/kg of DM in the diet; however, molasses had no effect on carcass dressing percentage, rib eye area, feed conversion ratio, blood urea nitrogen, and blood glucose (de Nazaré Santos Torres et al., 2023). Moreover, inclusion of molasses above 20% increased dry matter digestibility while decreasing NDF digestibility (de Nazaré Santos Torres et al., 2023). In contrast, when beef heifers were consuming bermudagrass hay ad libitum and supplemented with up to 2.3 kg/d of a 50:50 mixture of crude glycerol and molasses, fiber digestibility increased which led to an increased ADG (Ciriaco et al., 2016).

Soybean Hulls

Soybean hulls are the soybean seed coat and a byproduct of the soybean processing, consisting of 5 to 8% raw soybean and containing 86% structural carbohydrates (cellulose, hemicellulose, and lignin; Han et al., 2021); therefore, soybean hulls are considered a fiber source.

The bulk density of soybean hulls is relatively low as after the soybean is processed, the soybean hulls are extracted in a loose form. A low bulk density might increase the cost of transportation. Pelleting is a common practice in which the bulk density increases 3 to 3.7 times (Blasi et al., 2000). The process of pelleting does not affect DM, DMI, and NDF digestibility. (Merrill and Klopfenstein, 1985). Most of the soybean is processed by the solvent extraction procedure, in which it separates the oil and the protein-carbohydrate-fiber meal (Blasi et al., 2000). Usually, from the process of 27 kilograms of soybeans, 5 kg will result as oil and 20 kg as meal. The solvent extraction operation can be divided in three steps, the soybean preparation, oil extraction, and soybean meal.

During the stage of preparation all soybean are graded before the process starts. After passing through a screen that separates all foreign material, the soybeans are cracked with a roller which breaks it into smaller pieces. This facilitates the removal of hulls and aids to a proper flaking of the soybean. Fraction of meats and the hulls of the soybean are removed via aspiration after the crack, which later passes through a sifter and will be sort into three categories large hull and meats, small hulls and meats, and fines (Blasi et al., 2000). Pelleting helps reducing the cost of transportation of the byproduct but its inclusion in diets of cattle needs to be limited due to it being pellet and being more concentrated, which may cause some problems with urease and urea inclusion in diets. Urea is a non-protein N used as an ammonia source in ruminants, the urea is hydrolyzed by urease from rumen bacteria to produce NH_3 (Hailemariam et al., 2021). Many commercial feed companies will prefer the loose soybean hulls for an easier inclusion into their products by being a dust (Blasi et al, 2000).

Production and Economic Impact of Soybean

The soybean sector has a great impact on the US economy, averaging 115.8 billion dollars per year which is equivalent to more than 0.65 percent of the US gross domestic product (LMC International, 2019). In 2021, the total planted area reached 35.2 million hectares with an average yield of 3226 kg/ha. The total value of the US soybean crop is of 45.7 billion dollars. The US exported around 279 billion kg of soybeans, being the 51% of the total production for year 2021 (American Soybean Association, 2022). A total of 357,000 people is supported by the soybean sector in which 280,000 are paid full jobs and over 17,250 jobs at crushing and processing facilities. In the last 10 years the in the state of Georgia the total amounts of acres planted have decreased from 131,522 to 40,468 hectares planted, this is due to the prices declining below $0.35/kg. The total area increases and decreases in the state of Georgia depends on market prices (Bryant, 2021). The Census of Agriculture reported that in 2012 there were 302,963 farms raising soybeans increasing the amount since 2007 in which 279,110 were reported raising soybeans in the US. The US is the second largest producer and exporter of the world. Having this high numbers and being one of the most important producers in the world, there is a vast market for byproducts like soybean meal and soybean hulls. Prices of soybean hulls and other byproducts of the industry will be impacted by the overall soybean production, increasing, or decreasing its value on the market of commodities for cattle.

Effects of soybean inclusion in cattle diets on ruminal profile

Ruminal microorganisms. Characterization and comparison of the ruminal microbiome of dairy cows consuming three different byproducts of the soybean industry (soybean hulls, soybean meal, and raw soybean) has been performed (Arakaki et al., 2007). The concentration of cellulolytic and amylolytic anaerobic bacteria were greater when soybean hulls and soybean meal diets were fed to dairy cows; however, there was no significant difference in the overall rumen microorganism’s population (Arakaki et al., 2007). In a review where the same three byproducts were compared, an increase of microbial protein in the duodenum was reported when there was an inclusion of soybean hulls and soybean meal in the diet (Santos et al., 1998). Cellulolytic bacteria were greater in cows fed soybean hulls and soybean meal; however, there was no increment on amylolytic bacteria among the three soybean byproducts (Santos et al., 1998). These increments on the cellulolytic bacteria can be due to the soybean hulls being a fiber source and containing around 20% of cellulose (Arakaki et al., 2007). Concentrations of protozoa of the genera *Isotricha, Dasytricha, Diplodium, Ostracodinium, Epidinium, Metadinium, Eudiplodinium*, *Ophryoscolex* and *Entodinium* spp. tended to be lower when there is an inclusion of soybean hulls in the diets compared to soybean meal (Arakaki et al., 2007). A treatment × time interaction on protozoa population, in which concentrations are lower up to 6 hours after feeding when cattle are consuming soybean hulls in their diets has been reported. All treatments showed a reduction in the protozoa population at 3 h post feeding and it slightly increased at 6 h (Arakaki et al., 2007). In the same study methanogenic archaea were not detected because of the symbiotic relationship between protozoa and methanogens (Arakaki et al., 2007). However, the effect of soybean hulls decreasing the protozoa could suggest a decrease in the methanogenic population that would lead to an increase in the production of propionic acid (Hungate, 1966). This increase in propionic acid concentration is reflected on the lower acetate:propionate ratio observed in animals consuming soybean hulls (Arakaki et al., 2007). At 6 h the soybean hulls diet did not stimulate the growth of *Epidinium, Ophryoscolex, Metadinium and Eduiplodinium* microorganism genera (Arakaki et al, 2007). Other authors described that before there is an increase in vestibuliferid protozoa such as *Isotricha and Dasytrichia*, which decrease after feeding (Warner, 1966).

Ruminal pH and volatile fatty acids profile. Soybean hulls are low in lignin and the cellulose is highly digestible in the rumen, leading to increases in fermentation rates (Stein et al., 2008). They are generally used as a replacement to forage ingredients and grain commodities (Blasi et al., 2000) and will ferment differently than starch ingredients, forming acetate instead of propionate; however, soybean hulls cannot replace forages entirely because they do not contain enough physically effective fiber to stimulate rumination, saliva production, and maintain adequate ruminal pH levels between 6 and 7(Stein et al., 2008). In addition, a total replacement of starch with soybean hulls will limit the production of microbial protein and reduce the concentrations of butyrate (Blasi et al., 2000). Byproducts of the industry like soybean hulls have the ability to replace up to 30% (DM basis) of the total starch content of a diet without affecting fermentative pathways and having a negative impact on nutrient absorption and performance of dairy cows (Ipharraguerre & Clark, 2003). Research indicate that the inclusion of soybean hulls in the diets of dairy cows will have different parameters on ruminal pH, total VFA, and acetate:propionate ratio compared to soybean meal and raw soybean inclusion (Arakaki, 2007). The inclusion of soybean hulls has been reported to reduce ruminal pH and acetate:propionate ratio compared to soybean meal and raw soybean while increasing total VFA concentration, this result can be related to the higher fermentation rate of the soybean hulls compared to the other by-products (Arakaki, 2007). In vitro studies demonstrated the interaction between ruminal pH, ruminal acetate and enteric methane emissions (Russell, 1998; Lancaster et al., 2020). A decrease of up to 25% of the acetate:propionate ratio was observed, which was reported to be due to a reduction of ruminal pH and a greater amount of succinate produced by fiber digesting bacteria. Succinate is later transformed into propionate, explaining the reduction in acetate:propionate ratio (Russell, 1998). Similarly, the inclusion of soybean hulls in the diets tended to decrease ruminal pH below 6.2 for 16 hours in the post feeding period (Lancaster et al., 2020). The study consisted of comparing the effects of combining soybean hulls (0 or 30%) and calcium oxide (0 or 1%) in finishing diets containing dry distiller grains. Total VFA concentrations were greater for steers fed soybean hulls and calcium oxide rations while acetate concentration was greater for steers consuming soybean hulls compared to steers consuming diets without it (Lancaster et al., 2020). The acetate:propionate ratio was also greater for steers with an inclusion of soybean hulls in their diets. In addition, the concentrations of formate, propionate, butyrate, isobutyrate, valerate, and isovalerate did not differ with soybean hulls and calcium oxide inclusion in the diets (Lancaster et al., 2020). Soybean hulls might reduce concentration of milk fat when compared to an inclusion of soybean meal and raw soybean to the diets of dairy cows due to the reduction of the acetate:propionate ratio (Lancaster et al., 2020). A study compared the performance and rumen parameters of dairy cows consuming sugarcane varieties with or without an inclusion of soybean hulls in the diet (Lima et al., 2014a). There was no interaction between the varieties of sugar cane and inclusion of soybean hulls on feed intake and milk production; however, propionic acid concentrations were greater for cows consuming diets without the inclusion of soybean hulls (Lima et al., 2014). The inclusion of soybean hulls with sugarcane increased the acetic acid proportions compared to the other treatments (Lima et al., 2014). There was not a reduction of pH below 5.7, which can inhibit fiber degradation by inhibiting the growth and performance of fibrolytic bacteria and lead to acidosis. If the ruminal pH is below 5.8 or above 6.6 a direct impact on VFA absorption might occur, in which lower pH can increase VFA absorption (Lima., 1997). The average molar proportion of VFA in the rumen should be 73:20:7 (acetate:propionate:butyrate) for animals consuming only forage diets (Black, 1990). The inclusion of soybean hulls in the diets of dairy cows based of sugar cane had a molar proportion of 66:21:13 and dairy cows fed the same variety without the soybean hulls had a 57:33:10 (acetate:propionate:butyrate), which emphasizes the increase of acetate when there is an inclusion of soybean hulls in the diet (Lima et al., 2014). Moreover, soybean hulls are high in structural carbohydrate content that can lead to an increase in methane production due to the increase in acetate concentration. This can affect animal performance due to methanogenesis accounting for up to 18% of gross dietary energy loss (Kozloski, 2017).

Effects on cattle performance, digestibility and intake

Impacts on cattle performance have been reported when there is an inclusion of soybean hulls in corn-based diets (Ludden et al., 1995). Average daily gain of beef steers has not been affected with the inclusion of soybean hulls; however, a decrease on G:F ratio has been reported

(Ferreira et al., 2011). A reduction on average daily gain has been observed with the inclusion of soybean hulls replacing a percentage of the corn in finishing diets (Ludden et al., 1995). Data indicate that soybean hulls can be comparable in energy content to corn when it is supplemented to beef steers grazing a medium or poor-quality forage (Brown et al., 1981). In a study where cattle were fed poor quality native grass (3.7% CP) with increasing amounts of soybean hulls (0, 1, 2 and 3 kg) daily, maximum DMI of the native grass was observed when cattle were supplemented with 1 kg of soybean hulls (Martin and Hibberd, 1990). Additionally, the lowest intake of the native grass hay was observed when cattle were consuming the greatest amount of soybean hulls as a supplement, leading the authors to conclude that soybean hulls enhanced the energy status of the steers (Martin and Hibberd, 1990). Total energy intake can be similar between corn and soybean hulls when the latter is fed with a combination of poor-quality forages, despite having a significant difference in energy content. In environments where the forage is one of poor quality, the corn led to a reduction in forage intake and reduced the fiber degradation (Chan et al., 1991). The inclusion of corn in poor-quality forage diets or grazing systems can enhance the development of starch degrading microbes and reduce the fiber degrading bacteria, as a result fiber digestion can be reduced (Blasi et al., 2000). One aspect of soybean hulls is that it is low in lignin content, granting it greater digestibility compared to other fiber sources. Therefore, the energy available in diets containing soybean hulls can be compared to the ones with corn without affecting the fiber digestibility (Blasi et al., 2000).

Cows grazing on stockpiled tall fescue and tall fescue hay when pasture was limited were fed with 2 kg of soybean hulls from December to March during the winter. An approximate of 283 kg of hay per cow where saved and there was a reduction on body weight loss compared to feeding hay only (Boyles, n.d). In another study, soybean hulls were used to replace corn as a supplement provided to grazing steers. Steers were provided with either no supplementation, 2 kg of soybean hulls, or 2 kg of corn. Steers consuming either corn or soybean hulls had a similar ADG of 0.99 kg compared to steers that had no supplementation (Boyles, n.d). Soybean hulls can have the same efficiency of gain and impact on performance when fed as a creep supplement to beef steers (Boyles, n.d). Soybean hulls can negatively impact performance of feedlot steers if it replaces corn completely in finishing diets. Soybean hulls appear to be more beneficial if its supplemented to growing steers that are grazing or supplemented with hay only. This can help producers to decide which ingredient will be more adequate depending on the market price of commodities (Boyles, n.d). The use of soybean hulls can be an alternative energy source for feeding growing steers, replacing corn and other grains. Feeding high starch ingredients to cattle grazing or consuming forages as the main diet can negatively impact intake and can reduce fiber digestion (Chase and Hibberd, 1987). Energy supplements (1.36 kg/animal/d of rolled corn, ground soybean hulls, or whole soybean hulls) were compared with no supplementation on performance of cattle grazing smooth brome grass for 138 d during the summer (Bittner, 2012). Results indicate that there was no difference between the energetic supplements; however, cattle that received supplementation had greater average daily gain compared to cattle grazing without any supplement (Bittner, 2012). Similarly, when the effects of supplementing soybean hulls vs corn-soybean meal to grazing cattle were evaluated, no differences were observed on average daily gain when supplements were provided at 0.5% (DM) of BW; however, when supplements were provided at 0.8% to 1.0% (DM) of BW, greater average daily gain was reported for the soybean hulls treatment (Anderson et al., 1988).

Summary

Molasses is a byproduct from the sugar cane industry with a great potential for improving cattle performance and digestibility of high forage diets. The sugar cane industry is expanding in the US, which can generate a greater surplus of blackstrap molasses to be used in the animal industry. That can provide alternatives to energetic commodities benefiting beef cattle operations. Studies indicate that molasses fermentation in the rumen alters the microbiome increasing populations of *Butyrivibrio fibrisolvens,* which can lead to increased fiber degradation and butyrate production. Butyrate production may benefit the ruminal papillae growth and help reduce gut inflammation potentially caused by stress factors such as transportation. Lipopolysaccharides binding protein can be used as an inflammation marker and plays an important role in the relationship between LPS and gram-negative bacteria cell wall. Lastly, improvement on DM digestibility and animal performance has been observed when molasses is supplemented to cattle consuming high-forage diets. However, molasses is a highly fermentable ingredient, which can cause a reduction on ruminal pH, potentially leading to ruminal acidosis if over consumed. Soybean hulls is a byproduct from the soybean industry that is extensively used in the cattle industry. Being a fiber source with a low content of lignin makes soybean hulls more fermentable and digestible than other fiber sources. By being mainly composed of structural carbohydrates, the production of acetate can be greater in cattle consuming diets with an inclusion of soybean hulls compared to ingredients composed of sucrose and sugars, leading to the possibility of increasing enteric methane emissions. Some studies have shown a similar ADG of grazing cattle supplemented with soybean hulls compared to corn but had a negative impact on performance if corn was replaced by soybean hulls in finishing diets. Therefore, it has been suggested that soybean hulls have a great potential to improve performance of cattle consuming a high roughage diet. The nutritional aspects of these two commodities and their availability for producers can help to develop alternative strategies that can increase the ability of farms in the state of Georgia to retain ownership throughout the finishing phase of the beef industry.

References

Abreu, D., Dubeux, J. C. B., Queiroz, L. D., Jaramillo, D., Da Silva Santos, E. R., van Cleef, F., Vela-Garcia, C., DiLorenzo, N., & Ruiz-Moreno, M. (2022). Supplementation of Molasses-Based Liquid Feed for Cattle Fed on Limpograss Hay. Animals, 12(17). https://doi.org/10.3390/ani12172227

Alvarez, T., & Ximena, M. (2007). Dietary effect on rumen microbial protein synthesis and the rumen bacterial community: A review of the literature. University of Queensland. https://espace.library.uq.edu.au/view/UQ:158677/n02content_Alvarez.pdf?dsi_version=5aba9188d5e3b30e0ba465d0fe27c9d6

Baurhoo, B., & Mustafa, A. (2014). Short communication: Effects of molasses supplementation on performance of lactating cows fed high-alfalfa silage diets. Journal of Dairy Science, 97(2), 1072–1076. https://doi.org/10.3168/jds.2013-6989

Betancur-Murillo, C. L., Aguilar-Marín, S. B., & Jovel, J. (2023). Prevotella: A Key Player in Ruminal Metabolism. In Microorganisms (Vol. 11, Issue 1). MDPI. https://doi.org/10.3390/microorganisms11010001

Bittner, C. J. (2012).” Evaluation of Soybean Hulls and Feed Additives in Finishing Beef Diets” https://digitalcommons.unl.edu/animalscidiss/61

Blasi, D. A., Titgemeyer, E. C., Drouillard, J., & Steve I. Paisley. (2000). Kansas State University. https://bookstore.ksre.ksu.edu/pubs/mf2438.pdf

Bryant, C. (2021). Soybean Production in Georgia. https://grains.caes.uga.edu/content/dam/caes-subsite/grains/docs/soybean/2021-Soybean-Production-Guide.pdf

Chen, J., & Vitetta, L. (2020). The role of butyrate in attenuating pathobiont-induced hyperinflammation. Immune Network, 20(2). https://doi.org/10.4110/in.2020.20.e15

Ciriaco, F. M., Henry, D. D., G Mercadante, V. R., Schulmeister, T. M., Ruiz-Moreno, M., Lamb, G. C., DiLorenzo, N., Foran, M., Jones, D., Nowell, C., Wood, C., & Thomas, D. (2016). Effects of molasses and crude glycerol combined in a liquid supplement on ruminal fermentation in beef steers consuming bermudagrass hay 1,2. J. Anim. Sci, 94, 3851–3863. https://doi.org/10.2527/jas2016-0491

Damtew, A., Erega, Y., Ebrahim, huseen, Tsegaye, S., & Msigie, D. (2018). The Effect of long Distance Transportation Stress on Cattle: a Review. Biomedical Journal of Scientific & Technical Research, 3(3). https://doi.org/10.26717/bjstr.2018.03.000908

Dakota Beef, S., & Pritchard, R. (1987). Open Praire: Open Public Research Access Institutional Repository and Information Exchange Feeding High Grain Diets to Steer Calves with Different Feedlot Arrival Weights Recommended Citation "Feeding High Grain Diets to Steer Calves with Different Feedlot Arrival Weights." http://openprairie.sdstate.edu/sd_beefreport_1987http://openprairie.sdstate.edu/sd_beefreport_1987/12

de Nazaré Santos Torres, R., Bertoco, J. P. A., de Arruda, M. C. G., de Melo Coelho, L., Paschoaloto, J. R., Neto, O. R. M., Ezequiel, J. M. B., Almeida, M. T. C., Chardulo, L. A. L., & Baldassini, W. A. (2023). Effects of molasses supplementation on animal performance and carcass parameters of beef cattle: a meta-analysis. Tropical Animal Health and Production, 55(3), 143. https://doi.org/10.1007/s11250-023-03573-9

Food and Agricultural Industry. (1997). Sugarcane Processing. https://www3.epa.gov/ttnchie1/ap42/ch09/final/c9s10-1a.pdf

Felix, T. (2023). Transitioning Calves: From Pasture to Feedlot. https://extension.psu.edu/media/wysiwyg//extensions/catalog_product/db4a13155a0d49d38e50b837f6252252/f/i/figure-1-dirty-water-

Ferreira, E. M., Pires, A. V., Susin, I., Mendes, C. Q., Gentil, R. S., Araujo, R. C., Amaral, R. C., & Loerch, S. C. (2011). Growth, feed intake, carcass characteristics, and eating behavior of feedlot lambs fed high-concentrate diets containing soybean hulls. Journal of Animal Science, 89(12), 4120–4126. https://doi.org/10.2527/jas.2010-3417

Gressley, T. F. (2014). Inflammatory Responses to Sub-Acute Ruminal Acidosis. https://animal.ifas.ufl.edu/apps/dairymedia/RNS/2014/2014_proceedings.pdf#page=42

Han, L., Song, H., Fu, L., Li, J., Yang, L., & Liu, H. (2021). Effect of extraction method on the chemical profiles and bioactivities of soybean hull polysaccharides. Food Science and Nutrition, 9(11), 5928–5938. https://doi.org/10.1002/fsn3.2483

Hatch, C. F., & Beeson, W. M. (1972). Effect of Different Levels of Cane Molasses on Nitrogen and Energy Utilization in Urea Rations for Steers. Journal of Animal Science, 35(4). https://academic.oup.com/jas/article/35/4/854/4667132

Ipharraguerre, I. R., & Clark, J. H. (2003). Soyhulls as an alternative feed for lactating dairy cows: A review. In Journal of Dairy Science (Vol. 86, Issue 4, pp. 1052–1073). American Dairy Science Association. https://doi.org/10.3168/jds.S0022-0302(03)73689-3

Lancaster, N. A., Muegge, C. R., Carvalho, J. R. R., Lopes, R. C., Narumiya, R. S., Pinese, F., Baird, A. N., & Schoonmaker, J. P. (2020). Effect of calcium oxide and soybean hull addition to feedlot diets containing dried distillers grains and corn stover on steer performance, carcass characteristics, and digestibility. Translational Animal Science, 4(3). https://doi.org/10.1093/tas/txaa105

Li, F., Shah, A. M., Wang, Z., Peng, Q., Hu, R., Zou, H., Tan, C., Zhang, X., Liao, Y., Wang, Y., Wang, X., Zeng, L., Xue, B., & Wang, L. (2019a). Effects of land transport stress on variations in ruminal microbe diversity and immune functions in different breeds of cattle. Animals, 9(9). https://doi.org/10.3390/ani9090599

Lima, M. L. P., Simili, F. F., de Medeiros, M. I. M., Neto, G. B., Ribeiro, E. G., & de Paz, C. C. P. (2014a). Performance and rumen parameters of crossbred dairy cows fed two sugarcane varieties combined or not with soybean hulls. Revista Brasileira de Zootecnia, 43(12), 654–661. https://doi.org/10.1590/S1516-35982014001200005

LMC International. (2019). The Economic Impact of U.S. Soybean and End Products on the U.S. Economy. In LMC International. https://www.nopa.org/wp-content/uploads/2020/03/LMC_U.S.-Soy-Economic-Impact-Study-for-USB_FINAL-31320.pdf

Ludden, P. A., Cecava, M. J., & Hendrix, K. S. (1995). The Value of Soybean Hulls as a Replacement for Corn in Beef Cattle Diets Formulated With or Without Added Fat1/'. https://academic.oup.com/jas/article/73/9/2706/4632894

Malhi, M., Gui, H., Yao, L., Aschenbach, J. R., Gäbel, G., & Shen, Z. (2013). Increased papillae growth and enhanced short-chain fatty acid absorption in the rumen of goats are associated with transient increases in cyclin D1 expression after ruminal butyrate infusion. Journal of Dairy Science, 96(12), 7603–7616. https://doi.org/10.3168/jds.2013-6700

Martel, C. A., Titgemeyer, E. C., Mamedova, L. K., & Bradford, B. J. (2011). Dietary molasses increases ruminal pH and enhances ruminal biohydrogenation during milk fat depression. Journal of Dairy Science, 94(8), 3995–4004. https://doi.org/10.3168/jds.2011-4178

Mirza, I. H., Khan, A. G., Azim, A., & Mirza, M. A. (2002). Effect of Supplementing Grazing Cattle Calves with Urea-molasses Blocks, with and without Yucca schidigera Extract, on Performance and Carcass Traits. Asian-Australian Journal of Animal Science, 15(9). https://www.animbiosci.org/upload/pdf/15_207.pdf

Moore, J. E., Brant, M. H., Kunkle, W. E., & Hopkins, D. I. (1999). Roche/ASAS Foundation Beef Cattle Nutrition Symposium: Forage Supplementation and Grazing Effects of Supplementation on Voluntary Forage Intake, Diet Digestibility, and Animal Perf ormance1t2. https://academic.oup.com/jas/article/77/suppl_2/122/4653431

Mordenti, A. L., Giaretta, E., Campidonico, L., Parazza, P., & Formigoni, A. (2021). A review regarding the use of molasses in animal nutrition. In Animals (Vol. 11, Issue 1, pp. 1–17). MDPI AG. https://doi.org/10.3390/ani11010115

Subcommittee on Dairy Cattle Nutrition, Committee on Animal Nutrition, Board on Agriculture and Natural Resources, & National Research Council. (2001). Nutrient Requirements of Dairy Cattle: Seventh Revised Edition, 2001 (7th ed.). https://doi.org/10.17226/9825

Owens, F. N., Secrist, D. S., Hill, W. J., & Gill, D. R. (1996). Bud Britton Memorial Symposium on Metabolic Disorders of Feedlot Cattle. In J. Anim. Sci (Vol. 76). https://academic.oup.com/jas/article-abstract/76/1/275/4625198

Palmonari, A., Cavallini, D., Sniffen, C. J., Fernandes, L., Holder, P., Fagioli, L., Fusaro, I., Biagi, G., Formigoni, A., & Mammi, L. (2020). Short communication: Characterization of molasses chemical composition. Journal of Dairy Science, 103(7), 6244–6249. https://doi.org/10.3168/jds.2019-17644

Palmonari, A., Federiconi, A., Cavallini, D., Sniffen, C. J., Mammi, L., Turroni, S., D'Amico, F., Holder, P., & Formigoni, A. (2023). Impact of Molasses on Ruminal Volatile Fatty Acid Production and Microbiota Composition In Vitro. Animals, 13(4). https://doi.org/10.3390/ani13040728

Pate, F. M. (1983). Molasses in Beef Nutrition. National Feed Ingredients Association. https://rcrec-ona.ifas.ufl.edu/media/rcrec-onaifasufledu/pdf/Molasses-in-Beef-Nutrition.pdf

Reinhardt, C., & Thomson, D. U. (2015). Nutrition of Newly Received Feedlot Cattle. In Veterinary Clinics of North America - Food Animal Practice (Vol. 31, Issue 2, pp. 283–294). W.B. Saunders. https://doi.org/10.1016/j.cvfa.2015.03.010

Rusdiawan, A., Mar'atus Sholikhah, A., & Prihatiningsih, S. (2020). The Changes in pH Levels, Blood Lactic Acid and Fatigue Index to Anaerobic Exercise on Athlete After NaHCO 3 Administration. In Malaysian Journal of Medicine and Health Sciences (Vol. 16, Issue SUPP16).

Schroedl, W., Fuerll, B., Reinhold, P., Krueger, M., & Schuett, C. (2001). Acute phase marker LPS in cattle. Journal of Endotoxin Research, 7(1). https://pubmed.ncbi.nlm.nih.gov/11521082/

Senthilkumar, S., Suganya, T., Deepa, K., Muralidharan, J., & Sasikala, K. (n.d.). Supplementation of Molasses in Livestock Feed. www.ijset.net

Shearer DVM, J., & Extension Veterinarian, D. (2005). Rumen Acidosis, Heat Stress and Laminitis (Vol. 11). https://cales.arizona.edu/extension/dairy/conference/proceedings/2005/shearer.pdf

Stein, H. H., Berger, L. L., Drackley, J. K., Fahey, G. F., Hernot, D. C., & Parsons, C. M. (2008). Nutritional Properties and Feeding Values of Soybeans and Their Coproducts.

Sun, X. Q., Wang, Y. P., Wei, R. Y., Chen, B., & Zhao, X. (2020). Effects of replacing starch with three sugars in a concentrate and forage diet on in vitro rumen fermentation, fatty acid composition and related bacteria. Animal Production Science, 60(9), 1173–1182. https://doi.org/10.1071/AN18287

Tucureanu, M. M., Rebleanu, D., Constantinescu, C. A., Deleanu, M., Voicu, G., Butoi, E., Calin, M., & Manduteanu, I. (2018). Lipopolysaccharide-induced inflammation in monocytes/macrophages is blocked by liposomal delivery of Gi-protein inhibitor. International Journal of Nanomedicine, 13, 63–76. https://doi.org/10.2147/IJN.S150918

Van Engen, N. K., & Coetzee, J. F. (2018). Effects of transportation on cattle health and production: A review. In Animal Health Research Reviews (Vol. 19, Issue 2, pp. 142–154). Cambridge University Press. https://doi.org/10.1017/S1466252318000075

CHAPTER 2

EFFECTS OF REPLACING SOYBEAN HULLS WITH MOLASSES DURING THE RECEIVING PERIOD OF FEEDLOT CALVES[1]

[1]Angel A. Raudales-Quijada, Darren D. Henry, Pedro L. P. Fontes, Samir Burato, Jhones O. Sarturi, and Francine M. Ciriaco. To be submitted to *Journal of Animal Science*

Abstract

An experiment was conducted to evaluate the effects of replacing soybean hulls with molasses in the receiving diets of weaned feedlot steer-calves on growth performance, nutrient digestibility, enteric methane emissions, and a blood inflammatory marker. Thirty-six growing Angus crossbred steers (296 ± 12 kg body weight) were used in a generalized randomized block design in two periods (18 steers per period) with two dietary treatments. Diets consisted of dry matter (DM basis): 1) 46% corn silage, 25% dried distillers grains with solubles, 27% molasses, 0.5% limestone, and 1.5% vitamins and mineral packet (MOL) and 2) 46% Corn silage, 22% dried distillers grains with solubles, 30% soybean hulls, 0.5% limestone and 1.5% vitamins and mineral packet (SBH). Each experimental period consisted of 44 d, wherein both periods, on d -2, -1 and 0, steers were weighed and the average was considered initial body weight. Steers were stratified by initial body weight and randomly assigned to individual pens, which were randomly assigned to one of the two treatments (MOL or SBH; n = 18 steers/treatment). Dietary treatments started on d 0. Intermittent body weight measurements were recorded on d 7, 14, 21, 28, and 35. Final body weight was considered the average of two consecutive weights recorded on d 41 and 42. Blood samples were collected on days corresponding to body weight measurements. Feed intake data was collected. From d 19 to 24 for the first period and from d 17 to 22 for the second period, breath samples were collected, using the sulfur hexaflouride technique, to determine enteric methane production, and feed and feces were collected to determine apparent total tract nutrient digestibility. Data were analyzed using the MIXED procedure of SAS with steer as the experimental unit. The model included the fixed effect of treatment and the random effect of period. Initial (P = 0.82) and final body weights (P = 0.41) were not affected by treatment. Additionally, no effects of treatment were observed on dry matter intake (P = 0.30), average daily gain (P = 0.23) or gain to feed ratio (G:F) (P = 0.43). Grams of CH_4 per day (P = 0.24), per kg of OMI (P = 0.13), per kg of MBW (P = 0.20), or per kg of ADG (P = 0.90) were not affected by treatment; however grams of CH_4 per kg of OMD (P = 0.02), grams of CH_4 per kg of NDF intake (NDFI; P = <.0001), and grams of CH_4 per kg of NDF digested (NDFD; P = <.0001) were affected by treatment, where there was a reduction for MOL on grams CH_4 per kg of OMD and a reduction for SBH on grams CH_4 per kg of NDFI and NDFD. Concentration of LPS binding protein had no treatment effect (P = 0.90). Digestibility and nutrient intake was affected by treatment for NDF and ADF in which intake of NDF (P = <0.01) and ADF (P = <0.01) were greater for SBH and digestibility of NDF (P = <0.01) and ADF (P = <0.01) were lower for MOL. The data indicate that an inclusion of 27% of molasses (DM basis) can replace soybean hulls as an energy source in feedlot receiving diets without affecting animal performance but reducing the fiber digestibility. Grams of CH_4 per kg of organic matter digested (OMD) can be reduced with a 27% inclusion of molasses (DM basis), but grams of CH_4 per kg of NDFI or NDFD can increase. In addition, the concentrations of LPS binding protein as an inflammatory blood marker will not be impacted by the inclusion of molasses in feedlot receiving diets.

Introduction

In the cattle industry, feed typically accounts for up to 40-60% of the total costs associated with the operation (UNL Beef, 2017). Forage can be one of the most expensive dietary ingredients when considering the cost per unit of energy (Z. K. Smith, 2021). During the entrance to the feedlot, cattle are generally offered diets with greater inclusions of roughage with receiving diets being formulated with the purpose of getting cattle started on total mixed rations with moderated gains as they transition to final, high-grain finishing diets. During the receiving phase metabolic issues related to stress, such as gut inflammation, may occur due to the impact of weaning and transportation to new facilities. To help mitigate this stress, highly-fermentable fiber byproducts are often used to aid in transitioning cattle from a roughage based diet to the finishing ration. Soybean hulls is a byproduct that consists of the seed coat of the soybean (Gnanasambandam & Proctor, 1999) and is a common energy source used in the beef industry. Market fluctuations lead producers to look for alternative byproducts that can enhance or maintain cattle performance. Molasses, a byproduct of the sugar cane industry (Curtin, 1983), can increase nutrient utilization due to improved fiber degradation, which in turn can be reflected in improved cattle performance (Ciriaco et al., 2015). Moreover, the inclusion of molasses in cattle diets has been reported to increase butyrate production in the rumen (Ciriaco et al., 2021)

It was hypothesized that steers consuming a diet containing molasses would have greater fiber digestibility leading to greater growth performance and that, due to the fermentative pathways of molasses leading to greater butyrate concentration, enteric methane emissions would be reduced. Additionally, it was hypothesized that steers would have lesser gastro-intestinal tract dysfunctions due to positive impacts of butyrate on gut health, which would lead to fewer inflammatory responses. Therefore, the objective was to determine the effects of replacing soybean hulls with molasses in the receiving diets of feedlot weaned steer-calves on growth performance, apparent total tract nutrient digestibility, enteric methane emissions, and a blood inflammatory marker.

Materials and Methods

The animal use protocol (AUP # A2022 11-016-Y1-A0) for all procedures and working techniques involving animals in this study was approved by the University of Georgia Institutional Animal Care and Use Committee.

Experimental Design, Animals, and Treatments

The study was conducted from January 2023 to Abril 2023 at the University of Georgia Metabolism Complex located in Tifton, Georgia (31°29'42.3"N 83°31'43.3"W). A total of 36 growing (weaned) Angus crossbred steers (296 ± 12 kg initial body weight (IBW)) were used in a generalized randomized block design in two periods (18 steers per period). Steers used were part of the university's herd and, at weaning, had been administered vaccinations against clostridial diseases (One Shot Ultra 7; Zoetis, Parsippany, NJ), infectious bovine rhinotracheitis, bovine virus diarrhea Types 1 and 2, parainfluenza 3, and respiratory syncytial virus (Bovi-Shield GOLD 5; Zoetis) and a dose of a broad spectrum anthelmintic (Valbazen Suspension; Zoetis). During the experimental period (d 0 to 42), two diets were provided and they were considered treatment. Diets consisted of (DM basis): 1) 46% corn silage, 25% dried distillers grains with solubles, 27% molasses, 0.5% limestone, and 1.5% vitamins and mineral packet (**MOL**) and 2) 46% corn silage, 22% dried distillers grains with solubles, 30% soybean hulls, 0.5% limestone, and 1.5% vitamins and mineral packet (**SBH**), these diets were formulated using the NRC and with similar isocaloric and isonitrogenous proportions. Diets were mixed in a truck mounted total mixed ration (TMR) mixer (5100 Jaylor; Ontario, Canada). Diets were individually weighed and delivered by hand to each steers' feed bunk at 1100h. The experiment consisted of two 44-d periods and procedures described were performed in the same way for both periods.

Transportation and Data Collection Procedures

On d -2 blood samples were collected and steers were weighed and provided with a common diet consisting of (DM basis) 79% corn silage, 19% dried distillers grains, 0.5% limestone, and 1.5% mineral. On d -1 steers were weighed and immediately transported via truck and trailer for 12 h from 0800 to 2000 h (6 h in one direction then 6 h back to the facility) to induce stress experienced by calves when shipped to another location, as is standard in the beef industry. Steers were provided bermudagrass hay (Tifton 85) the evening they returned to the facilities.

On d 0, blood samples were collected and steers were weighed, stratified by initial body weight and randomly assigned to individual pens, which were randomly assigned to one of the two treatments (MOL or SBH; n = 9 steers/treatment). The average of d -2, -1, and 0 weights was considered initial body weight. Dietary treatments started on d 0. Other blood collections and body weight measurements were performed on d 7, 14, 21, 28, 35, 41, and 42. Final body weight was considered the average of weights recorded on d 41 and 42. Blood samples were collected via jugular venipuncture into 10-mL evacuated tubes containing K-EDTA (BD Vacutainer, Franklin Lakes, NJ). Tubes were inverted 10 times, immediately placed on ice, and subsequently centrifuged for 15 min at 2,400 × g at 4° C. Plasma was then transferred to labeled polypropylene tubes (12 × 75 mm; Fisherbrand; Thermo Fisher Scientific Inc., Waltham, MA) and stored at −20 °C for further analysis of LBP

Feed bunks were evaluated visually using the clean bunk management approach and feed intake data was collected by measuring dry matter of feed offered and orts refused (Rusche, 2023). Weekly, bunks were cleaned, orts weighed, and orts samples were collected. Samples of dietary ingredients were collected weekly for DM analysis and for adjusting the diet content.

Diet samples were collected weekly for further analysis (Table 2.1) and for DM content which was used for adjusting the intake as fed.

Apparent total tract digestibility

To determine apparent total tract digestibility of dry matter (DM), organic matter (OM), crude protein (CP), and fiber components (NDF and ADF), feed, orts, and fecal samples were collected during four consecutive days. Feed and orts were collected from each individual bunk from d 19 to 22. Fecal samples were collected from d 20 to 23 twice daily at 0800 and 1600 h via rectal grab or from the ground, immediately after defecation. Feed, orts, and fecal samples were frozen at -20°C for further analysis. At the end of the experiment, samples were dried at 55°C in a forced-air oven for 72 h and ground in a Wiley mill (Arthur H. Thomas Co., Philadelphia, PA) to pass a 2-mm screen. Samples were composited within steer to determine concentrations of nutrients and internal digestibility marker. Indigestible NDF (**iNDF**) was used as an internal marker to determine apparent total tract digestibility of nutrients.

Enteric Methane Emissions

Enteric CH_4 emissions were measured using the SF_6 tracer technique as described by Henry et al. (2020). Briefly, on d 7, steers were dosed, intraruminally, using a balling gun with a brass permeation tube containing approximately 3.3 g of SF_6 with an average release rate of 5.7 mg/d. Each collection canister was evacuated (63.5 cmHg) to ensure continuous collection of samples for 24 h. Collection of breath samples for analysis of CH_4 were performed daily for five consecutive days, starting at least 10 days after placement of permeation tubes. Four collection canisters were used to determine ambient CH_4 and SF_6 concentrations daily. Only steers with at least three successful days of collection and measurement were considered in the final analysis, in which one of the steers spitted out the permeation tube. That experimental unit was not considered for CH_4 analysis.

Laboratory Analysis

To determine sample DM, feed, fecal and orts samples were weighed (0.5 g) into ceramic crucibles and dried in a forced-air oven at 100°C for 16 h. After weighing, crucibles were placed in a muffle furnace at 600°C for 4 h before returning to a 100°C forced-air oven. Hot, ashed samples were weighed and used to calculate OM. For NDF, dried and ground feed, orts, and fecal samples were weighed in duplicate in F57 bags (Ankom Technology, Macedon, NY) and analyzed in an Ankom^{200} Fiber Analyzer using heat-stable α-amylase and sodium sulfite. Subsequent ADF analysis was performed sequentially (Van Soest et al., 1991). Total N of feed, orts, and fecal samples were determined by combustion using a LECO TruMac N analyzer (LECO, St. Joseph, MI) following official method 992.15 (AOAC, 1995). Crude protein was calculated by multiplying the N concentration by 6.25. Concentration of iNDF in feed and fecal samples were determined as described by (Cole et al., 2011) with modifications by (Krizsan & Huhtanen, 2013). Samples were placed in F57 bags and incubated for 288 h in the rumen of a ruminally-cannulated steer grazing Alfalfa-bermudagrass mixed pastures. After incubation, samples were rinsed, air-dried, and analyzed for NDF concentration as described above, without the use of α-amylase. Methane and SF_6 concentrations in breath samples were analyzed by gas chromatography (Trace 1310 Greenhouse Gas Analyzer, Thermo Scientific, Waltham, MA). For SF_6, an electron capture detector (350°C) and a packed column (TG-Packed column Hayesep D, mesh size 80/100, length 2.0 m, ID 2.0 mm, OD 3.2 mm; Thermo Scientific) were used. Methane was analyzed using a flame ionization detector (250°C) and a packed column (TG-Packed column Porapak Q, mesh size 80/100, length 0.5 m, ID 2.0 mm, OD 3.2 mm; Thermo Scientific).

Temperature of injectors and columns were 80 and 60° C, respectively for both gases. Carrier gases were N for SF_6 and CH_4, respectively. Concentration of the inflammatory marker LBP in plasma was determined by a third-party laboratory by ELISA (Creative Bio-Labs, Shirley, NY).

Calculations

Apparent total tract digestibility of DM, OM, CP, NDF, and ADF were calculated as follows:

Nutrient digestibility (%) = 100 – {100 × [(iNDF concentration in feed /iNDF concentration in feces) × (nutrient concentration in feces/nutrient concentration in feed)]}.

Emissions of CH_4 produced by steers were determined in relation to the SF_6 tracer gas captured in the collection canisters. The following equation was used to quantify CH_4 production:

$$Q_{CH_4} = Q_{SF_6} \times ([CH_4]_\gamma - [CH_4]_\beta) \div ([SF_6]_\gamma - [SF_6]_\beta)$$

in which Q_{CH_4} is CH_4 emissions per animal (g/d), Q_{SF_6} is SF_6 release rate (mg/d), $[CH_4]_\gamma$ is the concentration of CH_4 in the animals collection canister, $[CH_4]_\beta$ is the concentration of CH_4 in the ambient canisters, $[SF_6]_\gamma$ is the concentration of SF_6 in the animals collection canister, and $[SF_6]_\beta$ is the concentration of SF_6 in the ambient collection canister.

Statistical analysis

All data were analyzed as a generalized randomized block design with repeated measures for LBP data. The MIXED procedure of SAS was used and steer was considered the experimental unit. The model for all non-repeated data included the fixed effect of treatment and the random effect of period (block). For CH_4 emissions analysis, the data from one steer was removed due to the steer likely losing his permeation tube as his SF_6 emissions were below detection limits. Therefore, for CH_4 emissions, $n = 17$ and $n = 18$ per treatment, for SBH and

MOL, respectively. For LBP data, steer was considered the subject and the covariance structure chosen was first order autoregressive based on the smallest Akaike information criterion. Concentrations of LBP on d -2 were used as a covariate. The model included the fixed effect of treatment, day, and their interactions. Period and steer within treatment were included as random effects, with the latter used to designate the denominator degrees of freedom. One steer's data was removed from the LBP analysis due to extremely high values (>1000 ng/dL). Therefore, for data analysis of LBP, treatments have different number of experimental units (n = 18 for MOL and n = 17 for SBH). Significance was declared at $P \leq 0.05$ and tendencies were considered when $0.05 < P \leq 0.10$.

Results and Discussion

Animal Performance

Animal performance is presented in Table 2.2. Steers total DMI (P = 0.30), final BW (P = 0.41), ADG (P = 0.23), and G:F (P = 0.43) did not differ between treatments. Although only numerically, steers consuming SBH had greater total DMI (8.11 kg) compared to steers consuming MOL (7.79 kg). This impact might be due to the high levels of sulfur in the molasses diet (Table 2.1). The maximum tolerable amount of sulfur for diets containing 40% forage is 0.45% of the total DM (White, 2017). In a study in which the impact of different concentrations of sulfur (0.12, 0.31 and 0.46%) in diets of finishing steers were analyzed, decreased performance was reported from steers consuming diets with 0.31 or 0.46% of sulfur content (Pogge etal., 2013). The performance of cattle consuming bahiagrass hay treated with CaO and 10% molasses has been reported to not be affected; however, the authors attributed the lack of performance to the low quality of the forage (Ciriaco et al., 2021). Similar to the current experiment, when molasses was included in the diets of finishing beef steers at 4.5%, 9% and 13.5% of the diet DM, there were no effects on the DMI compared to a dry supplement with 0% inclusion of molasses (T. L. Felix et al., 2018). Moreover, Royes et al., (2001) reported greater performance from steers consuming stargrass hay and supplemented with 2.8 kg of soybean hulls compared to steers supplemented 2.8 kg of molasses. Similar to our results, in a study in which performance of dairy cows was evaluated, when cows were fed two different sugar cane varieties with or without soybean hulls, DMI was similar between groups (Lima et al., 2014). Homem et al. (2019) reported that an inclusion of 10 or 20% soybean hulls in finishing diets of sheep did not alter the DMI. Moreover, there was no difference in DMI when different percentages of soybean hulls (6.37, 12.88, and 25.99%) was included in corn-based diets of sheeps; however, the lowest DMI was from the group with no inclusion of soybean hulls (Russell et al., 2016).

In the present experiment, no differences were observed on final BW (P = 0.41) of steers. Similarly, Ciriaco et al. (2015) reported no effect on final BW of heifers that were supplemented with up to 2.3 kg/d of a 50:50 mixture of molasses and crude glycerol. In another study, where bahiagrass hay was treated with 10% molasses, no effect on final BW was observed (Ciriaco et al., 2021). Likewise, there was no difference in BW of dairy cows with or without an inclusion of molasses (0, 2.5 and 5%) in diets composed of alfalfa or corn silage-based diets (Broderick & Radloff, 2004). N In a meta-analysis, it was emphasized that the inclusion of molasses between 100 to 150 g/kg of DM in straw hay diets had no effect on performance of beef steers; however, inclusions above 200 g/kg of DM can have a negative impact on cattle performance (de Nazaré Santos Torres et al., 2023). In a trial performed by Lancaster et al. (2020), BW did not differ among finishing steers fed with or without soybean hulls with (0 or 30% in DM inclusion) or without and inclusion of CaO.   No differences in ADG of (P = 0.23) steers were observed between treatments. Similarly, de Nazaré Santos Torres et al., (2023) reported that there was no treatment effect on ADG with an inclusion of molasses from 20 to 30% on straw hay-based diets. In contrast, there was a linear increase in ADG of heifers consuming bahiagrass hay and supplemented with up to 2.3 kg/d of a 50:50 molasses and crude glycerol mixture (Ciriaco et al., 2015). Stateler et al., (1995) reported an increase in ADG when 2.05 kg DM/d of molasses was supplemented to cattle consuming a forage-based diet. The inclusion of molasses seems to improve the performance of cattle grazing or being fed only a roughage source. An inclusion of only 10% of molasses in finishing diets also did not have an effect on ADG (Crawford et al., 1978). With a 20% inclusion of molasses on barley diets, no effect on ADG of heifers was observed, compared to medium or low inclusions of molasses between 5-15% (Lofgreen, 1965). Greater ADG was reported by (Asimwe et al., 2015) on beef steers fed hay, rice polishing or maze meal, with an inclusion of 47% molasses and hominy feed compared to concentrates with maize meal and rice polishing. Ferreira et al. (2011) reported that lambs had a similar ADG when cracked corn was replaced with soybean hulls up to 45%. Boyles reported a similar ADG of 0.9 kg for cattle grazing tall fescue and supplemented with 1.8 kg of soybean hulls or corn, while cattle not supplemented had an ADG of 0.68 kg.

In the current study, G:F was not affected by treatment ($P = 0.43$). Similarly, supplementation with up to 2.3 kg/d of a 50:50 mixture of molasses and crude glycerol had no effect on G:F of growing heifers consuming bermudagrass hay (Ciriaco et al., 2015). Conversely, steers supplemented with 1.4% or 2.8% of molasses (DM basis) had a lower G:F compared to steers supplemented 1.4% or 2.8% of soybean hulls or corn (Royes et al., 2001). Moreover, in a study in which soybean hulls were supplemented with or without an inclusion of CaO in finishing diets no effects on G:F were observed (Lancaster et al., 2020).

Blood Inflammatory Marker LPS Binding Protein

Concentration of the blood inflammatory marker LBP is presented in Figure 2.1. No effect of treatment was observed for LBP concentrations (P = 0.90), indicating that under the conditions of the present experiment, the inclusion of molasses in the diet of backgrounding steers did not impact the concentrations of LBP. In a study evaluating the effects of liquid vs dry molasses on LBP concentrations before and after transportation of beef steers, a tendency for lower concentrations of LBP after transportation was reported for cattle supplemented with liquid molasses with a 19% reduction in the concentrations of LPS (between 40 to 50 µg/mL); being from 8 to 38 µg/mL the concentrations for healthy animals (Silva et al., 2023). Li et al. (2019) reported an increase in the concentrations of LBP for a group of Simmental crossbred (from 14.61 ng/mL to 17.56 ng/mL) and yak cattle (13.25 ng/mL to 17.54 ng/mL) after a 6 hour transportation in a trailer, indicating that long transportation times can in fact be considered a stressor. Chen and Vitetta, (2020) reported that butyrate has the ability to control proliferation of pathogens by increasing the mucosal cell barrier, inhibiting pro-inflammatory immune cells as well as reducing pro-inflammatory cytokines.

Apparent Total Tract Digestibility

Apparent total tract digestibility of nutrients is presented in Table 2.3. No effect of treatment was observed for intake of DM (P = 0.95), OM (P = 0.59), or CP (P = 0.87); however, as expected, due to the chemical composition of soybean hulls, there was a treatment effect on intake of NDF (P < 0.01), and ADF (P < 0.01) in which both were greater for SBH. Digestibility of DM tended (P = 0.10) to be greater for steers consuming MOL while, no difference between treatments was observed on digestibility of OM (P = 0.17) and CP (P = 0.49). However, there was an effect of treatment on digestibility of NDF (P < 0.01) and ADF (P < 0.01), in which both were greater for steers consuming SBH. A liquid feed supplementation based on molasses (0.9, 1.8 and 2.7 kg/d) has been reported to not affect intake of DM and OM from cattle consuming limpograss hay (Abreu et al., 2022). In another study, the inclusion of molasses in beef steer diets (100, 150 and 200 g/kg of DM) had no effect on DM and OM intake (de Nazaré Santos Torres et al., 2023). In addition, DM and OM digestibility were reduced in a study in which soybean hulls were included in corn-based diets of feedlot lambs (Ferreira et al., 2011). Similar results were reported in a study in which molasses fed at 20% of diet DM to beef steers consuming stargrass or bluestem hay, increased the digestibility of OM and decreased fiber digestibility (Kalmbacher et al., 1994; Royes et al., 2001). Similar to this study, the inclusion of soybean hulls in corn-based diets with or without enzymes did not affect the digestibility of DM, OM and CP; however, it did affect NDF and ADF digestibility, reducing the digestibility as soybean hulls increased (Russell et al., 2016). In addition, the inclusion of soybean hulls with or without CaO on finishing diets of beef steers did not affect the DM, OM, and CP intake; however, a similar result was reported for fiber digestibility in which it was greater for treatments with an inclusion of soybean hulls (Lancaster et al., 2020). In contrast to this study, the inclusion of up to 2.3 kg/d of a 50:50 mixture of molasses and crude glycerol linearly increased total tract NDF digestibility of growing heifers consuming bermudagrass hay (Ciriaco et al., 2015).

Enteric methane emissions

Enteric CH_4 emissions are presented in Table 2.4. There was no effect on grams of CH_4 produced per day (P = 0.24), per kg of organic matter intake (OMI; P = 0.13), per kg of metabolic body weight (MBW; P = 0.32), or per kg of ADG (P = 0.90). The term emission intensity is referred as the CH_4 produced per unit of animal production (Hristov et al., 2013), because there is no effect of treatment on cattle performance, we did not observe any impact on emission intensity. However, there was an effect of treatment for g of CH_4 produced per kg of organic matter digested (OMD; $P = 0.02$) in which MOL promoted a reduction of gram of CH_4 produced per kg of OMD when compared to SBH. Although the organic matter digested (OMD) was only numerically greater for MOL (Table 4), it could potentially reflect the reduction of grams of CH_4 produced per kg of OMD for the MOL treatment. Additionally, there was an effect of treatment on grams of CH_4 produced per kg of NDF digested (NDFD; $P < 0.01$) and NDF intake (NDFI; $P < 0.01$), in which both were greater for MOL. Even though MOL had a lower fiber content, SBH was more digestible in terms of NDF which is reflected in the lower grams of CH_4 produced per kg of NDFD and NDFI. Molasses has the ability to alter the ruminal fermentation profile by increasing the concentrations of butyrate at the expense of acetate (Palmonari et al., 2023). Methanogens are anaerobic archaea that grow by producing CH_4 gas (Buan, 2018). When one mole of acetate is produced, one mole of CO_2 and one mole of H_2 is produced as well, which later can potentially be utilized by methanogens to produce methane. Different from acetate, when one mole of butyrate is produced, one mole of H_2 is utilized and one mol of CO_2 is produced, theoretically reducing substrates for methanogenesis. In addition, its is been said that there is a growing desire to reduce the environmental of CH_4 via enteric fermenatino (Henry et al., 2020). Molasses also has the ability to aid the microbial growth of bacteria that plays a role in the fiber degradation and butyrate production, such as *Butyrivibrio fibrisolvens* (Palmonari et al., 2023). In other studies, an inclusion of molasses in an in vitro trial, had a lower concentration of *Methanobacteriaceae* compared to no inclusion of molasses (Palmonari et al., 2023). In contrast to this study, dairy cows fed a diet with an inclusion of molasses had greater daily CH_4 production compared to dairy cows consuming diets with wheat inclusion; however, the authors attributed the greater methane production to the fact that cows consuming the wheat diet had a reduced feed intake during the days of sample collection (Børsting et al., 2020).

Conclusions

There is a potential for replacement of soybean hulls with molasses in backgrounding diets without having negative impacts on performance of cattle. The inclusion of 27% molasses on a DM basis to the diets of backgrounding cattle can reduce the fiber digestibility increasing the grams of CH_4 produced per kg of NDFD and NDFI. However, molasses can potentially reduce grams of CH_4 produced per kg of OM digested by slightly increasing OM digestibility, this due to the molasses being a highly fermentable ingredient with high amounts of sugar. Future studies designed to evaluate cattle performance consuming similar diets used in the present study should be conducted.

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References

Abreu, D., Dubeux, J. C. B., Queiroz, L. D., Jaramillo, D., Da Silva Santos, E. R., van Cleef, F., Vela-Garcia, C., DiLorenzo, N., & Ruiz-Moreno, M. (2022). Supplementation of Molasses-Based Liquid Feed for Cattle Fed on Limpograss Hay. Animals, 12(17). https://doi.org/10.3390/ani12172227

Alvarez, T., & Ximena, M. (2007). Dietary effect on rumen microbial protein synthesis and the rumen bacterial community: A review of the literature [University of Queensland]. https://espace.library.uq.edu.au/view/UQ:158677/n02content_Alvarez.pdf?dsi_version=5aba9188d5e3b30e0ba465d0fe27c9d6

American Soybean Association. (2022). A Reference Guide to Important Soybean Facts & Figures. Soystats 2022. https://soygrowers.com/wp-content/uploads/2022/06/22ASA-002-Soy-Stats-Final-WEB.pdf

Arakaki, L. C., Gaggiotti, M., Gallardo, M., Valtorta, S., Conti, G., Quaino, O., Carrillo, Jo., Gregoret, R., Kudo, H., & Dehorty, B. (2007). The Effects of Feeding Soybean Meal, Soybean Hulls and Raw Soybean Seed on Rumen Microbial Populations and Ruminal Fermentation in Grazing Dairy Cows.

Asimwe, L., Kimambo, A. E., Laswai, G. H., Mtenga, L. A., Weisbjerg, M. R., Madsen, J., & Mushi, D. E. (2015). Growth performance and carcass characteristics of Tanzania Shorthorn Zebu cattle finished on molasses or maize grain with rice or maize by-products. Livestock Science, 182, 112–117. https://doi.org/10.1016/j.livsci.2015.11.001

Bannerman, D. D., Paape, M. J., Hare, W. R., & Eun, J. S. (2003). Increased levels of LPS-binding protein in bovine blood and milk following bacterial lipopolysaccharide challenge.

Journal of Dairy Science, 86(10), 3128–3137. https://doi.org/10.3168/jds.S0022-0302(03)73914-9

Baurhoo, B., & Mustafa, A. (2014). Short communication: Effects of molasses supplementation on performance of lactating cows fed high-alfalfa silage diets. Journal of Dairy Science, 97(2), 1072–1076. https://doi.org/10.3168/jds.2013-6989

Betancur-Murillo, C. L., Aguilar-Marín, S. B., & Jovel, J. (2023). Prevotella: A Key Player in Ruminal Metabolism. In Microorganisms (Vol. 11, Issue 1). MDPI. https://doi.org/10.3390/microorganisms11010001

Bittner, C. J. (2012). Evaluation of Soybean Hulls and Feed Additives in Finishing Beef Diets https://digitalcommons.unl.edu/animalscidiss/61

Blasi, D. A., Titgemeyer, E. C., Drouillard, J., & Steve I. Paisley. (2000). Soybean Hulls. Kansas State University. https://bookstore.ksre.ksu.edu/pubs/mf2438.pdf

Børsting, C. F., Brask, M., Hellwing, A. L. F., Weisbjerg, M. R., & Lund, P. (2020). Enteric methane emission and digestion in dairy cows fed wheat or molasses. Journal of Dairy Science, 103(2), 1448–1462. https://doi.org/10.3168/jds.2019-16655

Boyles, S. (1999). SOYBEAN HULLS. https://agnr.osu.edu/sites/agnr/files/imce/pdfs/Beef/SoybeanHulls.pdf

Broderick, G. A., & Radloff, W. J. (2004). Effect of molasses supplementation on the production of lactating dairy cows fed diets based on alfalfa and corn silage. Journal of Dairy Science, 87(9), 2997–3009. https://doi.org/10.3168/jds.S0022-0302(04)73431-1

Bryant, C. (2021). Soybean Production in Georgia. https://grains.caes.uga.edu/content/dam/caes-subsite/grains/docs/soybean/2021-Soybean-Production-Guide.pdf

Buan, N. R. (2018). Methanogens: Pushing the boundaries of biology. In Emerging Topics in Life Sciences (Vol. 2, Issue 4, pp. 629–646). Portland Press Ltd. https://doi.org/10.1042/ETLS20180031

Chen, J., & Vitetta, L. (2020). The role of butyrate in attenuating pathobiont-induced hyperinflammation. Immune Network, 20(2). https://doi.org/10.4110/in.2020.20.e15

Ciriaco, F. M., Henry, D. D., G Mercadante, V. R., Schulmeister, T. M., Ruiz-Moreno, M., Lamb, G. C., DiLorenzo, N., Foran, M., Jones, D., Nowell, C., Wood, C., & Thomas, D. (2016). Effects of molasses and crude glycerol combined in a liquid supplement on ruminal fermentation in beef steers consuming bermudagrass hay 1,2. J. Anim. Sci, 94, 3851–3863. https://doi.org/10.2527/jas2016-0491

Ciriaco, F. M., Henry, D. D., Mercadante, V. R. G., Schulmeister, T., Ruiz-Moreno, M., Lamb, G. C., & Dilorenzo, N. (2015). Effects of different levels of supplementation of a 50:50 mixture of molasses:crude glycerol on performance, Bermuda grass hay intake, and nutrient digestibility of beef cattle. J. Anim. Sci, 93, 2428–2438. https://doi.org/10.2527/jas2015-8888

Ciriaco, F. M., Henry, D. D., Sanford, C. D., Canal, L. B., Dubeux, J. C. B., & Dilorenzo, N. (2021). Performance of growing beef cattle consuming bahiagrass hay treated with calcium oxide and molasses. Translational Animal Science, 5(4). https://doi.org/10.1093/tas/txab195

Cole, N. A., McCuistion, K., Greene, L. W., & McCollum, F. T. (2011). Effects of concentration and source of wet distillers grains on digestibility of steam-flaked corn-based diets fed to finishing steers1. The Professional Animal Scientist, 27(4), 302–311. https://doi.org/10.15232/S1080-7446(15)30493-9

Curtin, L. V. (1983). Molasses-General Considerations. National Feed Ingredients Association. https://rcrec-ona.ifas.ufl.edu/media/rcrec-onaifasufledu/pdf/Molasses---General-Considerations.pdf

Damtew, A., Erega, Y., Ebrahim, huseen, Tsegaye, S., & Msigie, D. (2018). The Effect of long Distance Transportation Stress on Cattle: a Review. Biomedical Journal of Scientific & Technical Research, 3(3). https://doi.org/10.26717/bjstr.2018.03.000908

Dakota Beef, S., & Pritchard, R. (1987). Open PRAIRIE: Open Public Research Access Institutional Repository and Information Exchange Feeding High Grain Diets to Steer Calves with Different Feedlot Arrival Weights Recommended Citation "Feeding High Grain Diets to Steer Calves with Different Feedlot Arrival Weights." http://openprairie.sdstate.edu/sd_beefreport_1987http://openprairie.sdstate.edu/sd_beefreport_1987/12

de Nazaré Santos Torres, R., Bertoco, J. P. A., de Arruda, M. C. G., de Melo Coelho, L., Paschoaloto, J. R., Neto, O. R. M., Ezequiel, J. M. B., Almeida, M. T. C., Chardulo, L. A. L., & Baldassini, W. A. (2023). Effects of molasses supplementation on animal performance and carcass parameters of beef cattle: a meta-analysis. Tropical Animal Health and Production, 55(3), 143. https://doi.org/10.1007/s11250-023-03573-9

Food and Agricultural Industry. (1997). Sugarcane Processing. https://www3.epa.gov/ttnchie1/ap42/ch09/final/c9s10-1a.pdf

Felix, T. (2023). Transitioning Calves: From Pasture to Feedlot. https://extension.psu.edu/media/wysiwyg//extensions/catalog_product/db4a13155a0d49d38e50b837f6252252/f/i/figure-1-dirty-water-

Felix, T. L., Long, C. J., Stierwalt, M. R., Carvalho, P. H. V., & Blalock, H. M. (2018). Effects of increasing dietary level of a commercial liquid supplement on growth performance and carcass characteristics in feedlot steers. Translational Animal Science, 2(2), 216–222. https://doi.org/10.1093/tas/txy009

Ferreira, E. M., Pires, A. V., Susin, I., Mendes, C. Q., Gentil, R. S., Araujo, R. C., Amaral, R. C., & Loerch, S. C. (2011). Growth, feed intake, carcass characteristics, and eating behavior of feedlot lambs fed high-concentrate diets containing soybean hulls. Journal of Animal Science, 89(12), 4120–4126. https://doi.org/10.2527/jas.2010-3417

Flint, J. H., & Stewart, C. S. (1999). Bacteroides and prevotella. In C. Batt & M. Lou Tortorello (Eds.), Encyclopedia of Food Microbiology (Second Edition).

Gressley, T. F. (2014). Inflammatory Responses to Sub-Acute Ruminal Acidosis. https://animal.ifas.ufl.edu/apps/dairymedia/RNS/2014/2014_proceedings.pdf#page=42

Hailemariam, S., Zhao, S., He, Y., & Wang, J. (2021). Urea transport and hydrolysis in the rumen: A review. In Animal Nutrition (Vol. 7, Issue 4, pp. 989–996). KeAi Communications Co. https://doi.org/10.1016/j.aninu.2021.07.002

Han, L., Song, H., Fu, L., Li, J., Yang, L., & Liu, H. (2021). Effect of extraction method on the chemical profiles and bioactivities of soybean hull polysaccharides. Food Science and Nutrition, 9(11), 5928–5938. https://doi.org/10.1002/fsn3.2483

Hatch, C. F., & Beeson, W. M. (1972). Effect of Different Levels of Cane Molasses on Nitrogen and Energy Utilization in Urea Rations for Steers. Journal of Animal Science, 35(4). https://academic.oup.com/jas/article/35/4/854/4667132

Henry, D. D., Ciriaco, F. M., Araujo, R. C., Fontes, P. L. P., Oosthuizen, N., Rostoll-Cangiano, L., Sanford, C. D., Schulmeister, T. M., Dubeux, J. C. B., Lamb, G. C., & DiLorenzo, N.

(2020). Effects of bismuth subsalicylate and encapsulated calcium-ammonium nitrate on enteric methane production, nutrient digestibility, and liver mineral concentration of beef cattle. Journal of Animal Science, 98(8). https://doi.org/10.1093/jas/skaa234

Homem, A. C., Nocera, B. F., Faleiros, L. F., Almeida, M. T. C., Paschoaloto, J. R., Perez, H. L., D'Áurea, A. P., & Ezequiel, J. M. B. (2019). Partial replacement of corn by soybean hulls in highgrain diets for feedlot sheep. Pesquisa Agropecuaria Brasileira, 54. https://doi.org/10.1590/S1678-3921.PAB2019.V54.00029

Ipharraguerre, I. R., & Clark, J. H. (2003). Soyhulls as an alternative feed for lactating dairy cows: A review. In Journal of Dairy Science (Vol. 86, Issue 4, pp. 1052–1073). American Dairy Science Association. https://doi.org/10.3168/jds.S0022-0302(03)73689-3

Kalmbacher, R. S., Brown, W. F., & Pate, F. M. (1994). Effect of Molasses-Based Liquid Supplements on Digestibility of Creeping Bluestem and Performance of Mature Cows on Winter Range'. https://academic.oup.com/jas/article/73/3/853/4632796

Krizsan, S. J., & Huhtanen, P. (2013). Effect of diet composition and incubation time on feed indigestible neutral detergent fiber concentration in dairy cows. Journal of Dairy Science, 96, 1715–1726.

Lancaster, N. A., Muegge, C. R., Carvalho, J. R. R., Lopes, R. C., Narumiya, R. S., Pinese, F., Baird, A. N., & Schoonmaker, J. P. (2020). Effect of calcium oxide and soybean hull addition to feedlot diets containing dried distillers grains and corn stover on steer performance, carcass characteristics, and digestibility. Translational Animal Science, 4(3). https://doi.org/10.1093/tas/txaa105

Li, F., Shah, A. M., Wang, Z., Peng, Q., Hu, R., Zou, H., Tan, C., Zhang, X., Liao, Y., Wang, Y., Wang, X., Zeng, L., Xue, B., & Wang, L. (2019). Effects of land transport stress on variations in ruminal microbe diversity and immune functions in different breeds of cattle. Animals, 9(9). https://doi.org/10.3390/ani9090599

Lima, M. L. P., Simili, F. F., de Medeiros, M. I. M., Neto, G. B., Ribeiro, E. G., & de Paz, C. C. P. (2014). Performance and rumen parameters of crossbred dairy cows fed two sugarcane varieties combined or not with soybean hulls. Revista Brasileira de Zootecnia, 43(12), 654–661. https://doi.org/10.1590/S1516-35982014001200005

LMC International. (2019). The Economic Impact of U.S. Soybean and End Products on the U.S. Economy. In LMC International. https://www.nopa.org/wp-content/uploads/2020/03/LMC_U.S.-Soy-Economic-Impact-Study-for-USB_FINAL-31320.pdf

Ludden, P. A., Cecava, M. J., & Hendrix, K. S. (1995). The Value of Soybean Hulls as a Replacement for Corn in Beef Cattle Diets Formulated With or Without Added Fat1/'. https://academic.oup.com/jas/article/73/9/2706/4632894

Malhi, M., Gui, H., Yao, L., Aschenbach, J. R., Gäbel, G., & Shen, Z. (2013). Increased papillae growth and enhanced short-chain fatty acid absorption in the rumen of goats are associated with transient increases in cyclin D1 expression after ruminal butyrate infusion. Journal of Dairy Science, 96(12), 7603–7616. https://doi.org/10.3168/jds.2013-6700

Mansfield, H. R., & Stern, M. D. (1994). Effects of Soybean Hulls and Lignosulfonate-Treated Soybean Meal, on Ruminal Fermentation in Lactating Dairy Cows. Journal of Dairy Science, 77(4), 1070–1083. https://doi.org/10.3168/jds.S0022-0302(94)77042-9

Martel, C. A., Titgemeyer, E. C., Mamedova, L. K., & Bradford, B. J. (2011). Dietary molasses increases ruminal pH and enhances ruminal biohydrogenation during milk fat depression. Journal of Dairy Science, 94(8), 3995–4004. https://doi.org/10.3168/jds.2011-4178

Mirza, I. H., Khan, A. G., Azim, A., & Mirza, M. A. (2002). Effect of Supplementing Grazing Cattle Calves with Urea-molasses Blocks, with and without Yucca schidigera Extract, on Performance and Carcass Traits. Asian-Australian Journal of Animal Science, 15(9). https://www.animbiosci.org/upload/pdf/15_207.pdf

Moore, J. E., Brant, M. H., Kunkle, W. E., & Hopkins, D. I. (1999). Roche/ASAS Foundation Beef Cattle Nutrition Symposium: Forage Supplementation and Grazing Effects of Supplementation on Voluntary Forage Intake, Diet Digestibility, and Animal Perf ormance1t2. https://academic.oup.com/jas/article/77/suppl_2/122/4653431

Mordenti, A. L., Giaretta, E., Campidonico, L., Parazza, P., & Formigoni, A. (2021). A review regarding the use of molasses in animal nutrition. In Animals (Vol. 11, Issue 1, pp. 1–17). MDPI AG. https://doi.org/10.3390/ani11010115

Subcommittee on Dairy Cattle Nutrition, Committee on Animal Nutrition, Board on Agriculture and Natural Resources, & National Research Council. (2001). Nutrient Requirements of Dairy Cattle: Seventh Revised Edition, 2001 (7th ed.). https://doi.org/10.17226/9825

Owens, F. N., Secrist, D. S., Hill, W. J., & Gill, D. R. (1996). Bud Britton Memorial Symposium on Metabolic Disorders of Feedlot Cattle. In J. Anim. Sci (Vol. 76). https://academic.oup.com/jas/article-abstract/76/1/275/4625198

Palmonari, A., Cavallini, D., Sniffen, C. J., Fernandes, L., Holder, P., Fagioli, L., Fusaro, I., Biagi, G., Formigoni, A., & Mammi, L. (2020). Short communication: Characterization of molasses chemical composition. Journal of Dairy Science, 103(7), 6244–6249. https://doi.org/10.3168/jds.2019-17644

Palmonari, A., Federiconi, A., Cavallini, D., Sniffen, C. J., Mammi, L., Turroni, S., D'Amico, F., Holder, P., & Formigoni, A. (2023). Impact of Molasses on Ruminal Volatile Fatty Acid

Production and Microbiota Composition In Vitro. Animals, 13(4). https://doi.org/10.3390/ani13040728

Pate, F. M. (1983). Molasses in Beef Nutrition. National Feed Ingredients Association. https://rcrec-ona.ifas.ufl.edu/media/rcrec-onaifasufledu/pdf/Molasses-in-Beef-Nutrition.pdf

Pogge, D. J., Lonergan, S., Russell, J., Loy, D., & Spurlock, M. (2013). The effect of supplemental vitamin C on performance, antioxidant capacity, carcass characteristics, and meat quality of steers fed high sulfur finishing diets [Dissertation]. Iowa State University.

Reinhardt, C., & Thomson, D. U. (2015). Nutrition of Newly Received Feedlot Cattle. In Veterinary Clinics of North America - Food Animal Practice (Vol. 31, Issue 2, pp. 283–294). W.B. Saunders. https://doi.org/10.1016/j.cvfa.2015.03.010

Royes, J. B., Brown, W. F., Martin, F. G., & Bates, D. B. (2001). Source and level of energy supplementation for yearling cattle fed ammoniated hay 1. In J. Anim. Sci (Vol. 79). https://academic.oup.com/jas/article/79/5/1313/4625952

Rusche, W. (2023, August 11). Feed Bunk Management. https://extension.sdstate.edu/feed-bunk-management

Rusdiawan, A., Mar’atus Sholikhah, A., & Prihatiningsih, S. (2020). The Changes in pH Levels, Blood Lactic Acid and Fatigue Index to Anaerobic Exercise on Athlete After NaHCO 3 Administration. In Malaysian Journal of Medicine and Health Sciences (Vol. 16, Issue SUPP16).

Russell, J. R., Sexten, W. J., & Kerley M. S. (2016). soybean hull and enzyme incusion. Journal of Animal Science.

Schroedl, W., Fuerll, B., Reinhold, P., Krueger, M., & Schuett, C. (2001). Acute phase marker LPS in cattle. Journal of Endotoxin Research, 7(1). https://pubmed.ncbi.nlm.nih.gov/11521082/

Senthilkumar, S., Suganya, T., Deepa, K., Muralidharan, J., & Sasikala, K. (2016). Supplementation of molasses in livestock feed. International Journal of Sciece, Environment, 5(3). www.ijset.net

Shearer, J. (2005). Rumen Acidosis, Heat Stress and Laminitis. 11. https://cales.arizona.edu/extension/dairy/conference/proceedings/2005/shearer.pdf

Silva, B. C., Godoi, L. A., Supapong, C., Bitsie, B., Valadares Filho, S. C., & Schoonmaker, J. P. (2023). Effect of a molasses-based liquid supplement on gastrointestinal tract barrier function, inflammation, and performance of newly received feedlot cattle before and after a transport stress. Journal of Animal Science, 101. https://doi.org/10.1093/jas/skac295

Spears, J. W., Lloyd, K. E., & Fry, R. S. (2011). Tolerance of cattle to increased dietary sulfur and effect of dietary cation-anion balance. Journal of Animal Science, 89(8), 2502–2509. https://doi.org/10.2527/jas.2010-3265

Stateler, D. A., Kunkle, W. E., & Hammond, A. C. (1995). Effect of Protein Level and Source in Molasses Slurries on the Performance of Growing Cattle Fed Hay During Winter 1. In Range Sciences. https://academic.oup.com/jas/article-abstract/73/10/3078/4632623

Stein, H. H., Berger, L. L., Drackley, J. K., Fahey, G. F., Hernot, D. C., & Parsons, C. M. (2008). Nutritional Properties and Feeding Values of Soybeans and Their Coproducts.

Sun, X. Q., Wang, Y. P., Wei, R. Y., Chen, B., & Zhao, X. (2020). Effects of replacing starch with three sugars in a concentrate and forage diet on in vitro rumen fermentation, fatty acid composition and related bacteria. Animal Production Science, 60(9), 1173–1182. https://doi.org/10.1071/AN18287

Tucureanu, M. M., Rebleanu, D., Constantinescu, C. A., Deleanu, M., Voicu, G., Butoi, E., Calin, M., & Manduteanu, I. (2018). Lipopolysaccharide-induced inflammation in monocytes/macrophages is blocked by liposomal delivery of Gi-protein inhibitor. International Journal of Nanomedicine, 13, 63–76. https://doi.org/10.2147/IJN.S150918

Van Engen, N. K., & Coetzee, J. F. (2018). Effects of transportation on cattle health and production: A review. In Animal Health Research Reviews (Vol. 19, Issue 2, pp. 142–154). Cambridge University Press. https://doi.org/10.1017/S1466252318000075

Van Soest, P. J., Robertson, J. B., & Lewis, B. A. (1991). Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. Journal of Dairy Science, 74(10), 3583–3597. https://doi.org/10.3168/jds.S0022-0302(91)78551-2

White, J. (2017). Cattle can suffer from too much sulfur. https://todaysfarmermagazine.com/mag/livestock/1303-cattle-can-suffer-from-too-much-sulfur#:~:text=The%20National%20Academy%20of%20Sciences,may%20result%20in%20sulfur%20toxicity.

Table 2.1. Analyzed chemical composition of receiving diets with an inclusion of molasses or soybean hulls.

Item	MOL[1]	SBH[2]
DM	58.36	59.46
OM, %	91.63	94.08
CP, %	15.2	14.9
NDF, %	22.9	38.4
ADF, %	12.0	25.4
TDN, %	79	75
NEm, Mcal/kg	1.97	1.85
NEg, Mcal/kg	1.32	1.21
Digestible Energy, Mcal/kg	3.57	3.39
Metabolizable Energy, Mcal/kg	3.17	2.98
Calcium, %	0.87	0.81
Phosphorus, %	0.56	0.52
Magnesium, %	0.39	0.35
Potassium, %	2.22	1.34
Sodium, %	0.13	0.12
Sulfur, %	0.48	0.21

Analyzed by a commercial laboratory sing a wet chemistry package (Dariy One, Itahace, NY)
[1]MOL: 46% corn silage, 25% dried distillers grains with solubles, 27% molasses, 0.5% limestone, and 1.5% vitamins and mineral packet
[2]SBH: 46% corn silage, 22% dried distillers grains with solubles, 30% soybean hulls, 0.5% limestone, and 1.5% vitamins and mineral packet

Table 2.2. Effects of the inclusion of molasses or soybean hulls in the diet on the performance of backgrounding beef steers.

	Treatment[1]			
Item	MOL	SBH	SEM[2]	*P*-value
Initial BW[3], kg	294	293	3.1	0.82
Final BW[4], kg	366	372	7.2	0.41
ADG, kg	1.70	1.86	0.100	0.23
G:F	0.21	0.22	0.010	0.43
DMI, kg	7.79	8.11	0.236	0.30

[1]MOL: 46% corn silage, 25% dried distillers grains with solubles, 27% molasses, 0.5% limestone, and 1.5% vitamins and mineral packet; SBH: 46% corn silage, 22% dried distillers grains with solubles, 30% soybean hulls, 0.5% limestone, and 1.5% vitamins and mineral packet
[2] Standard error of the mean (SEM) n = 18 steers/treatment
[3]Initital BW was the average of BW recorded on d -1, d -2 (before 12 h transportation for inducing stress) and d 0 (12 h after transportation and before starting the dietary treatments)
[5]Final BW was the average of the BW recorded on d 41 and d 42

Table 2.3. Effects of the inclusion of molasses or soybean hulls in the diet on nutrient intake and total tract digestibility of backgrounding beef steers.

	Treatment[1]			
Item	MOL	SBH	SEM[2]	*P*-value
4-d intake, kg/d				
DM	8.12	8.15	0.529	0.95
OM	7.44	7.66	0.485	0.59
CP	1.36	1.34	0.112	0.87
NDF	2.36	3.72	0.253	< 0.01
ADF	1.10	2.13	0.142	< 0.01
Digestibility, %				
DM	72.75	70.37	1.236	0.10
OM	73.60	71.68	1.213	0.17
CP	65.64	66.92	1.321	0.49
NDF	54.93	64.45	1.219	< 0.01
ADF	55.38	64.31	1.261	< 0.01

[1]MOL: 46% corn silage, 25% dried distillers grains with solubles, 27% molasses, 0.5% limestone, and 1.5% vitamins and mineral packet; SBH: 46% corn silage, 22% dried distillers grains with solubles, 30% soybean hulls, 0.5% limestone, and 1.5% vitamins and mineral packet
[2]n = 18 steers/treatment

Table 2.4. Effects of the inclusion of molasses or soybean hulls in the diet on enteric CH_4 production of backgrounding beef steers.

Item[3]	Treatment[1]			
	MOL	SBH	SEM[2]	*P*-value
CH_4 emissions, g/d	190.86	204.78	8.434	0.24
CH_4 emissions, g/kg OMI	25.72	27.69	2.063	0.13
CH_4 emissions, g/kg OMD	35.79	39.46	2.333	0.02
CH_4 emissions, g/kg MBW	2.53	2.67	0.100	0.32
CH_4 emissions, g/kg NDFI	81.06	57.36	6.433	< 0.01
CH_4 emissions, g/kg NDFD	156.59	92.22	10.605	< 0.01
CH_4 emissions, g/kg ADG	114.53	113.76	5.196	0.90

[1]MOL: 46% corn silage, 25% dried distillers grains with solubles, 27% molasses, 0.5% limestone, and 1.5% vitamins and mineral packet; SBH: 46% corn silage, 22% dried distillers grains with solubles, 30% soybean hulls, 0.5% limestone, and 1.5% vitamins and mineral packet
[2] MOL: n = 18 steers; SBH: n = 17 steers
[3]CH_4 was determined from the average of at least 3 24-h period of breath sample collection. OMI, OM intake; OMD, OM digested; MBW, metabolic BW; NDFI, NDF intake; NDFD, NDF digested; ADG; average daily gain.

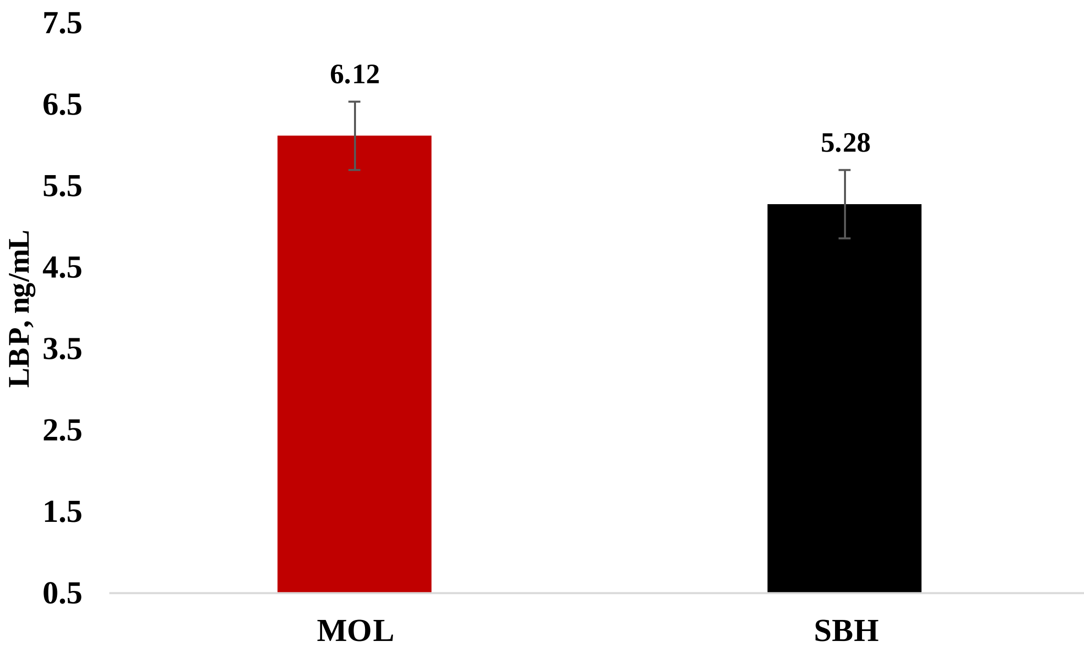

Figure 2.1. Effects of the inclusion of molasses or soybean hulls in the diet on concentrations of LPS binding protein (LBP) of backgrounding beef steers. MOL (*n* = 18 steers): 46% corn silage, 25% dried distillers grains with solubles, 27% molasses, 0.5% limestone, and 1.5% vitamins and mineral packet; SBH (*n* =17): 46% corn silage, 22% dried distillers grains with solubles, 30% molasses, 0.5% limestone, and 1.5% vitamins and mineral packet. No difference between treatments (*P* = 0.90). Error bars represent the SE of treatment means.

CHAPTER 3

EFFECTS OF REPLACING SOYBEAN HULLS WITH MOLASSES ON RUMINAL FERMENTATION PROFILE OF BEEF STEERS CONSUMING A BACKGROUNDING DIET[1]

[1]Angel A. Raudales-Quijada, Darren D. Henry, Pedro L. P. Fontes, Samir Burato, Jhones O. Sarturi, and Francine M Ciriaco. To be submitted to *Journal of Animal Science*

Abstract

An experiment was conducted to evaluate the effects of replacing soybean hulls with molasses in backgrounding diets of beef steers on ruminal fermentation profile and total dry matter intake. Six Angus crossbred steers (586 ± 85 kg body weight) were used in a cross-over design with two diets (treatments; DM basis): 1) 46% corn silage, 25% dried distillers grains with solubles, 27% molasses, 0.5% limestone, and 1.5% vitamins and mineral packet (MOL) and 2) 46% corn silage, 22% dried distillers grains with soluble, 30% soybean hulls, 0.5% limestone and 1.5% vitamins and mineral packet (SBH). The experiment consisted of 38 days split into two periods with a 14-d adaptation in each period and a 7-d washout between periods. On d 15 of each period, ruminal fluid samples were collected every 3 h post feeding for 24 h. Data were analyzed as a cross-over design with repeated measures using steer as the experimental unit. Steers total DMI was not affected by treatment ($P = 0.25$). Ruminal pH was not affected by treatment ($P = 0.53$) or treatment × time interaction ($P = 0.31$). A treatment × time interaction was observed for concentrations of NH_3-N ($P = 0.02$), where at 3 ($P < 0.01$) and 6 h ($P = 0.05$) post-feeding, it was greater for MOL. There was an effect of treatment on concentration of butyrate ($P < 0.01$), where it was greater for MOL. A treatment × time interaction was detected for acetate ($P = 0.02$), propionate ($P = 0.04$), BCVFA ($P < 0.01$), valerate ($P < 0.01$), caproate ($P = 0.02$), and total VFA ($P = 0.05$) concentrations. No treatment × time interactions were observed on major VFA molar proportions ($P \geq 0.24$); however, there was an effect of treatment on the molar proportions of acetate ($P < 0.01$), butyrate ($P < 0.01$), and BCFA ($P = 0.02$). Data indicate that a 27% inclusion of molasses in the backgrounding diets of beef steers can shift the molar proportion of VFA increasing butyrate at the expense of acetate, which can be beneficial to improve gut health. The daily fluctuations in ruminal NH_3-N suggest that molasses can benefit

the microbial growth to aid in fiber digestion. Lastly, soybean hulls can be replaced with molasses in backgrounding diets without negatively affecting ruminal pH and total dry matter intake.

Introduction

Soybean hulls are considered a fiber source consisting mainly of structural carbohydrates with a low lignin content (Boyles) and fiber is mainly fermented into acetate in the rumen. When one mole of acetate is produced, there is also the production of one mole of CO_2 and one mole of H_2, which are substrates used by methanogens for CH_4 production. Due to its highly fermentable nature, molasses, can aid in fiber degradation by increasing bacterial growth and altering the ruminal VFA profile, which is shifted towards greater concentrations of butyrate (Ciriaco et al., 2015). In the production of one mole of butyrate, one mole of H_2 is utilized and one mole of CO_2 is produced, which can directly impact enteric CH_4 emissions due to decreased concentrations of H_2 (Palmonari et al., 2023). Additionally, molasses can alter ruminal pH by being a highly fermentable ingredient (Curtin, 1983). Excessive consumption of molasses can lead to increased production of volatile fatty acids, which can decrease ruminal pH and lead to acidosis (Owens et al., 1996). Therefore, it was hypothesized that the fermentative pathways of molasses would lead to greater butyrate production, while soybean hulls would be mainly fermented into acetate in the rumen. The objective was to determine the effects of replacing soybean hulls with molasses on ruminal fermentation profile and total dry matter intake of beef steers consuming a backgrounding diet.

Materials and Methods

The animal use protocol (AUP # A2022 12-014-Y1-A0) for all procedures and working techniques involving animals in this study was approved by the University of Georgia Institutional Animal Care and Use Committee.

Experimental Design, Animals, and Treatments

The study was conducted from February – April 2023 at the University of Georgia Tucker Barn located in Tifton, Georgia (31°29'49.4"N 83°31'38.6"W). A total of 6 Angus crossbred steers (586 ± 85 kg initial BW) were used in a cross-over design with a 7-d washout period between two experimental periods. During the experimental periods, two diets were provided and they were considered treatment. In Period 1, steers were randomly assigned to one of the two treatments: 1) 46% corn silage, 25% dried distillers grains with solubles, 27% molasses, 0.5% limestone, and 1.5% vitamins and mineral packet (**MOL)** or 2) 46% corn silage, 22% dried distillers grains with solubles, 30% pelleted soybean hulls, 0.5% limestone, and 1.5% vitamins and mineral packet (**SBH**). In Period 2, steers previously receiving MOL received SBH and vice-versa. In each period, d 0 to 14 were for adaptation to the diets and d 15 for a 24-h collection of ruminal fluid. During the washout period, all steers consumed a corn silage-based diet without molasses or soybean hulls (79% corn silage, 19% dried distillers grains with solubles, 0.5% limestone, and 1.5% vitamins and mineral packet). Diets were mixed using a truck mounted vertical mixer (Jaylor 5100; Ontario, Canada). Steers were fed individually and diet was weighed in buckets and delivered by hand to the feed bunks. Feed bunks were evaluated visually using the bunk management approach to allow between 3 to 5% of feed remaining in the feed bunk daily and feed intake data was collected by measuring dry matter of feed offered and orts refused. Bunks were cleaned, orts weighed, and orts samples collected once a week. Samples of dietary ingredients were grabbed weekly for DM analysis and for adjusting the diet content based on the most recent DM. Diet samples were collected weekly for further nutritional analysis and for DM content which was used for adjusting the intake as fed.

Ruminal Fluid Collection

All procedures for collecting data and samples were performed in an identical manner for both periods. On d 15 of each period, ruminal fluid samples were collected before feeding and every 3 h post-feeding for 24 h. Before sample collection, ruminal contents were thoroughly mixed by hand and a representative sample of digesta was collected from three different sites (cranial, ventral, and caudal). Samples were strained through four layers of cheesecloth and ruminal fluid pH was immediately measured using a manual pH meter (FiveEasy Plus FP20 benchtop pH/mV meter, Mettler-Toledo, LLC, Columbus, OH) before altering the sample. Two 12-mL samples of ruminal fluid were transferred into 15-mL conical tubes and 120 µL of a 20% (vol/vol) sulfuric acid solution were added to each tube to stop fermentation and preserve the sample. Samples were immediately placed on ice and subsequently frozen and stored at -20°C for further analysis of NH_3-N and VFA.

Laboratory Analysis

Concentrations of NH_3-N in the ruminal fluid were measured following the phenol-hypochlorite technique as described by Broderick and Kang (1980), with modifications as described by Ciriaco et al., (2016). Briefly, ruminal fluid samples were centrifuged at 10,000 × g for 15 minutes at 4°C (Avanti J-E, Beckman Coulter Inc.). One milliliter of phenol reagent was added into 12 × 75 mm borosilicate disposable culture tubes (Fisherbrand; Thermo Fisher Scientific Inc.,Waltham, MA) followed by 20 µL of ruminal fluid supernatant. After vortexing, 0.8 mL of a hypochlorite solution was added and mixed. Tubes were covered with marbles and placed in a 95°C water bath for 5 minutes. After cooling, 0.2 mL of each sample were transferred into wells of a flat-bottom 96-well plate. Absorbance was read on a plate reader (BioTek Epoch Microplate Reader, Agilent Technologies, Santa Clara, CA) at 620 nm.

Concentrations of VFA were measured in a liquid-liquid solvent extraction using ethyl acetate as described by Ruiz-Moreno et al. (2015) and Ciriaco et al. (2016). Briefly, ruminal fluid samples were centrifuged at 10,000 × g for 15 minutes at 4°C. Two milliliters of the supernatant were mixed with 0.4 mL of meta-phosphoric (25% wt/vol) and crotonic acid (2 g/L; internal standard) solution into 12 × 75 mm polypropylene tubes. Samples were frozen overnight, thawed, and centrifuged for 15 minutes at 10,000 × g at 4°C. Supernatant was transferred into 12 × 75 mm borosilicate tubes and mixed with ethyl acetate at a 2:1 (ethyl acetate: rumen fluid) ratio for extraction. Tubes were shaken vigorously and a 5-min rest was allowed for separation. The ethyl acetate fraction (top layer) was transferred to 9-mm glass vials (Fisherbrand; Thermo Fisher Scientific Inc.) and capped. Samples were analyzed with a gas chromatograph (Trace 1310, Thermo Scientific, Waltham, MA) using a flame ionization detector (250°C) and a capillary column (110 °C, TG-WAXMS, length 30 m, ID 0.25 mm, film thickness 0.25 µm, Thermo Scientific).

Statistical Analysis

Data were analyzed as a cross-over design using the MIXED procedure of SAS with steer within period as the experimental unit. Ruminal fermentation parameters (VFA, NH_3-N, and pH) were analyzed as repeated measures. The model included the fixed effects of treatment, time, their interaction, order, and period. Steer within treatment was included in the model as a random effect to designate the denominator degrees of freedom. Steer within period was considered the subject, and the covariance structure chosen was first order autoregressive based on the smallest

Akaike information criterion value. For total dry matter intake data, the model included the fixed effects of treatment, order, and period. Significance was declared at $P \leq 0.05$ and tendencies were considered when $0.05 < P \leq 0.10$.

Results and Discussion

Total dry matter intake

Total DMI (Table 3.1) was not affected by treatment ($P = 0.25$); however, steers fed the SBH diet had numerically greater intake (13.34 kg) compared to the MOL group (11.71 kg). The numerical difference might be due to the greater content of sulfur (0.48%) in the diet with inclusion of molasses (Table 2.2). White (2017), reported that a sulfur content of 0.45% or greater on diets containing at least 40% forage can have a negative impact on the DMI and performance. Spears et al. (2011), reported that steers consuming corn silage-based diets with a 0.12, 0.30 or 0.46% of sulfur had no effect on DMI; however, when steers were switched to a corn-based diet the DMI decreased as sulfur concentrations increase.

Ruminal pH, NH_3 -N and VFA

Ruminal pH (Table 3.1), was not affected by treatment × time ($P = 0.32$) or treatment ($P = 0.53$), but there was an effect of time (data not shown; $P < 0.01$). Regardless of treatment, the lowest ruminal pH was observed at 6 hours after feeding (5.50), with the highest before feeding (0 h) and 24 h after feeding (6.61). These results are not surprising as both molasses and soybean hulls can be highly fermentable: molasses by its high content of sugars and soybean hulls being low in lignin content. Similar to our results, an inclusion of molasses from 26 – 35% had no effect on ruminal pH (de Nazaré Santos Torres et al., 2023). In a study in which a 50:50 mixture of crude glycerol and molasses was being supplemented to cattle eating bermudagrass hay, a treatment × time interaction was reported in which at 3 h, and up to 6 h post feeding pH was decreased for cattle supplemented with 2.3 kg/d of the mixture; however, pH did not drop below 6.32 (Ciriaco et al., 2016). Mantin and Wing (1966) reported that pH was not affected by low levels (4.2, 8.4 and 12.6% on DM basis) of molasses included in corn-based diets of beef steers. In addition, no treatment × time interactions were reported on ruminal pH when soybean hulls were included in diets with inclusion of sugar cane (Lima et al., 2014).

There was no effect of treatment ($P = 0.41$) on ruminal NH_3 -N concentrations (Table 6); however, a treatment × time interaction ($P = 0.02$) was observed, in which steers consuming MOL had greater concentrations of NH_3-N at 3 ($P < 0.01$) and 6 ($P = 0.05$) hours post feeding (Fig. 3.1). For both treatments the peak concentration of NH_3-N is at 3 hours post feeding. Steers in this project were fed once a day and having their highest intake during the first hours of the post feeding time, which may explain a peak in NH_3-N concentrations at 3 h post feeding and then decreasing for the rest of the day. Ciriaco et al., (2016) observed linear decrease on ruminal NH_3-N concentrations after liquid feed supplementation (up to 2.3 kg/d of 50:50 molasses:crude glycerol), which may be related to the bacteria utilizing the NH_3-N to increase its population. In this study, the decrease in ruminal NH_3-N concentration after 3 h post feeding in both treatments could be due to an increase in ruminal microbial growth since molasses is a source of highly-fermentable carbohydrates that can enhance the development of bacteria like *Butyrivibrio fibrisolvens* (Palmonari et al., 2023), while soybean hulls can enhance the development of *Fibrobacter succinogenes* (Arakaki et al., 2007). A plausible explanation for the peak in NH_3-N in the first post feeding hours could be because of the diurnal intake pattern for cattle fed once a day, in which intake will be higher in the first hours after feeding time.

There was a treatment effect (Table 3.2) on butyrate ($P < 0.01$) concentration; however, no treatment × time interaction was observed ($P = 0.32$). A treatment × time interaction was detected for acetate (*P* = 0.02), propionate (*P* = 0.04), branched chained volatile fatty acids (BCVFA) (*P* < 0.01), valerate (*P* < 0.01), caproate (*P* = 0.02), and total VFA (*P* = 0.04) concentrations. There was a tendency for a greater total VFA concentrations at 3 h (*P* = 0.08) and at 6 h (*P* = 0.02) and 9 h (*P* = 0.01) post-feeding for SBH compared to MOL. In a study in which corn was replaced by soybean hulls in the diets of dairy cows, the total concentrations of VFA increased when 5% of the concentrate was substituted with soybean hulls (Mansfield & Stern, 1994). Lancaster et al. (2020) reported an increase in total VFA concentration in steers fed a finishing diet with the inclusion of soybean hulls (30% of DM) with or without an inclusion of CaO. Similar to this study, an inclusion of 5% on DM basis of molasses reduced the total VFA concentrations and molar proportions of propionate in dairy cows (Martel et al., 2011b).There was an effect of treatment (Table 3.3) on the molar proportions of acetate (*P* < 0.01), butyrate (*P* < 0.01) and BCVFA (*P* = 0.02; Table 8), while a treatment × time interaction was observed for valerate (*P* < 0.01). The increase in molar proportion of butyrate at the expense of acetate for the steers consuming MOL is in accordance with most of the published literature when there is an inclusion of molasses in the diets of beef cattle (Ciriaco et al., 2021). Increased molar proportions of butyrate are shown when bacteria such as *Butyrivibrio Fibrisolvens* increased in population (Palmonari et al., 2023). Moreover, Palmonari et al. (2023) reported a correlation between the proliferation of *Butyrivibrio Fibrisolvens* and *Streptococcus bovis* with the increased molar proportions of butyrate and the inclusion of molasses in the diets. In another study, a decrease in molar proportions of acetate and BCVFA was observed, whereas molar proportions of butyrate and valerate increased linearly as the level of a 50:50 mixture of molasses and crude glycerol increased up to 2.3 kg/d (Ciriaco et al., 2015). In addition, a greater acetate concentration was reported in cattle consuming diets with an inclusion of soybean hulls

(Lancaster et al., 2020). Arakaki et al., (2007) also reported greater acetate concentrations in cattle that were supplemented with soybean hulls compared to soybean meal and raw soybean. The inclusion of soybean hulls increased the proliferation of microorganisms that produce acetate and ferment cellulose and hemicellulose such as *Fibrobacter succinogenes and Ruminococcus albus* (Russell et al., 1992).

Conclusions

In the present study, the ruminal VFA profile was shifted towards greater butyrate concentrations when molasses is included and to greater acetate concentrations when soybean hulls is included in backgrounding diets. In addition, the peaks in NH_3-N concentrations at 3 h post feeding suggest that feed consumption was probably happening immediately and finished shortly after feed was provided, while the decrease in concentrations past 3 h may be due to an increase in microbial growth populations. The results observed in the present study suggest that soybean hulls can potentially be replaced with molasses without a negative effect on ruminal pH

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References

Abreu, D., Dubeux, J. C. B., Queiroz, L. D., Jaramillo, D., Da Silva Santos, E. R., van Cleef, F., Vela-Garcia, C., DiLorenzo, N., & Ruiz-Moreno, M. (2022). Supplementation of Molasses-Based Liquid Feed for Cattle Fed on Limpograss Hay. Animals, 12(17). https://doi.org/10.3390/ani12172227

Alvarez, T., & Ximena, M. (2007). Dietary effect on rumen microbial protein synthesis and the rumen bacterial community: A review of the literature [University of Queensland]. https://espace.library.uq.edu.au/view/UQ:158677/n02content_Alvarez.pdf?dsi_version=5ab a9188d5e3b30e0ba465d0fe27c9d6

American Soybean Association. (2022). A Reference Guide to Important Soybean Facts & Figures. Soystats 2022. https://soygrowers.com/wp-content/uploads/2022/06/22ASA-002-Soy-Stats-Final-WEB.pdf

AOAC. (1995). Official Method of Analysis (16th ed.). Association of Official Analytical Chemists.

Arakaki, L. C., Gaggiotti, M., Gallardo, M., Valtorta, S., Conti, G., Quaino, O., Carrillo, Jo., Gregoret, R., Kudo, H., & Dehorty, B. (2007). The Effects of Feeding Soybean Meal, Soybean Hulls and Raw Soybean Seed on Rumen Microbial Populations and Ruminal Fermentation in Grazing Dairy Cows.

Asimwe, L., Kimambo, A. E., Laswai, G. H., Mtenga, L. A., Weisbjerg, M. R., Madsen, J., & Mushi, D. E. (2015). Growth performance and carcass characteristics of Tanzania Shorthorn Zebu cattle finished on molasses or maize grain with rice or maize by-products. Livestock Science, 182, 112–117. https://doi.org/10.1016/j.livsci.2015.11.001

Bannerman, D. D., Paape, M. J., Hare, W. R., & Eun, J. S. (2003). Increased levels of LPS-binding protein in bovine blood and milk following bacterial lipopolysaccharide challenge. Journal of Dairy Science, 86(10), 3128–3137. https://doi.org/10.3168/jds.S0022-0302(03)73914-9

Baurhoo, B., & Mustafa, A. (2014). Short communication: Effects of molasses supplementation on performance of lactating cows fed high-alfalfa silage diets. Journal of Dairy Science, 97(2), 1072–1076. https://doi.org/10.3168/jds.2013-6989

Betancur-Murillo, C. L., Aguilar-Marín, S. B., & Jovel, J. (2023). Prevotella: A Key Player in Ruminal Metabolism. In Microorganisms (Vol. 11, Issue 1). MDPI. https://doi.org/10.3390/microorganisms11010001

Bittner, C. J. (2012). Evaluation of Soybean Hulls and Feed Additives in Finishing Beef Diets https://digitalcommons.unl.edu/animalscidiss/61

Blasi, D. A., Titgemeyer, E. C., Drouillard, J., & Steve I. Paisley. (2000). Soybean Hulls. Kansas State University. https://bookstore.ksre.ksu.edu/pubs/mf2438.pdf

Børsting, C. F., Brask, M., Hellwing, A. L. F., Weisbjerg, M. R., & Lund, P. (2020). Enteric methane emission and digestion in dairy cows fed wheat or molasses. Journal of Dairy Science, 103(2), 1448–1462. https://doi.org/10.3168/jds.2019-16655

Boyles, S. (1999). Soybean Hulls https://agnr.osu.edu/sites/agnr/files/imce/pdfs/Beef/SoybeanHulls.pdf

Broderick, G. A., & Radloff, W. J. (2004). Effect of molasses supplementation on the production of lactating dairy cows fed diets based on alfalfa and corn silage. Journal of Dairy Science, 87(9), 2997–3009. https://doi.org/10.3168/jds.S0022-0302(04)73431-1

Bryant, C. (2021). Soybean Production in Georgia. https://grains.caes.uga.edu/content/dam/caes-subsite/grains/docs/soybean/2021-Soybean-Production-Guide.pdf

Buan, N. R. (2018). Methanogens: Pushing the boundaries of biology. In Emerging Topics in Life Sciences (Vol. 2, Issue 4, pp. 629–646). Portland Press Ltd. https://doi.org/10.1042/ETLS20180031

Chen, J., & Vitetta, L. (2020). The role of butyrate in attenuating pathobiont-induced hyperinflammation. Immune Network, 20(2). https://doi.org/10.4110/in.2020.20.e15

Ciriaco, F. M., Henry, D. D., G Mercadante, V. R., Schulmeister, T. M., Ruiz-Moreno, M., Lamb, G. C., DiLorenzo, N., Foran, M., Jones, D., Nowell, C., Wood, C., & Thomas, D. (2016). Effects of molasses and crude glycerol combined in a liquid supplement on ruminal fermentation in beef steers consuming bermudagrass hay 1,2. J. Anim. Sci, 94, 3851–3863. https://doi.org/10.2527/jas2016-0491

Ciriaco, F. M., Henry, D. D., Mercadante, V. R. G., Schulmeister, T., Ruiz-Moreno, M., Lamb, G. C., & Dilorenzo, N. (2015). Effects of different levels of supplementation of a 50:50 mixture of molasses:crude glycerol on performance, Bermuda grass hay intake, and nutrient digestibility of beef cattle. J. Anim. Sci, 93, 2428–2438. https://doi.org/10.2527/jas2015-8888

Ciriaco, F. M., Henry, D. D., Sanford, C. D., Canal, L. B., Dubeux, J. C. B., & Dilorenzo, N. (2021). Performance of growing beef cattle consuming bahiagrass hay treated with calcium oxide and molasses. Translational Animal Science, 5(4). https://doi.org/10.1093/tas/txab195

Cole, N. A., McCuistion, K., Greene, L. W., & McCollum, F. T. (2011). Effects of concentration and source of wet distillers grains on digestibility of steam-flaked corn-based diets fed to finishing steers1. The Professional Animal Scientist, 27(4), 302–311. https://doi.org/10.15232/S1080-7446(15)30493-9

Curtin, L. V. (1983). Molasses-General Considerations. National Feed Ingredients Association. https://rcrec-ona.ifas.ufl.edu/media/rcrec-onaifasufledu/pdf/Molasses---General-Considerations.pdf

Damtew, A., Erega, Y., Ebrahim, huseen, Tsegaye, S., & Msigie, D. (2018). The Effect of long Distance Transportation Stress on Cattle: a Review. Biomedical Journal of Scientific & Technical Research, 3(3). https://doi.org/10.26717/bjstr.2018.03.000908

Dakota Beef, S., & Pritchard, R. (1987). Open PRAIRIE: Open Public Research Access Institutional Repository and Information Exchange Feeding High Grain Diets to Steer Calves with Different Feedlot Arrival Weights Recommended Citation "Feeding High Grain Diets to Steer Calves with Different Feedlot Arrival Weights." http://openprairie.sdstate.edu/sd_beefreport_1987http://openprairie.sdstate.edu/sd_beefreport_1987/12

de Nazaré Santos Torres, R., Bertoco, J. P. A., de Arruda, M. C. G., de Melo Coelho, L., Paschoaloto, J. R., Neto, O. R. M., Ezequiel, J. M. B., Almeida, M. T. C., Chardulo, L. A. L., & Baldassini, W. A. (2023). Effects of molasses supplementation on animal performance and carcass parameters of beef cattle: a meta-analysis. Tropical Animal Health and Production, 55(3), 143. https://doi.org/10.1007/s11250-023-03573-9

Food and Agricultural Industry. (1997). Sugarcane Processing. https://www3.epa.gov/ttnchie1/ap42/ch09/final/c9s10-1a.pdf

Felix, T. (2023). Transitioning Calves: From Pasture to Feedlot. https://extension.psu.edu/media/wysiwyg//extensions/catalog_product/db4a13155a0d49d38e50b837f6252252/f/i/figure-1-dirty-water-

Felix, T. L., Long, C. J., Stierwalt, M. R., Carvalho, P. H. V., & Blalock, H. M. (2018). Effects of increasing dietary level of a commercial liquid supplement on growth performance and carcass characteristics in feedlot steers. Translational Animal Science, 2(2), 216–222. https://doi.org/10.1093/tas/txy009

Ferreira, E. M., Pires, A. V., Susin, I., Mendes, C. Q., Gentil, R. S., Araujo, R. C., Amaral, R. C., & Loerch, S. C. (2011). Growth, feed intake, carcass characteristics, and eating behavior of feedlot lambs fed high-concentrate diets containing soybean hulls. Journal of Animal Science, 89(12), 4120–4126. https://doi.org/10.2527/jas.2010-3417

Flint, J. H., & Stewart, C. S. (1999). Bacteroides and prevotella. In C. Batt & M. Lou Tortorello (Eds.), Encyclopedia of Food Microbiology (Second Edition).

Gressley, T. F. (2014). Inflammatory Responses to Sub-Acute Ruminal Acidosis. https://animal.ifas.ufl.edu/apps/dairymedia/RNS/2014/2014_proceedings.pdf#page=42

Hailemariam, S., Zhao, S., He, Y., & Wang, J. (2021). Urea transport and hydrolysis in the rumen: A review. In Animal Nutrition (Vol. 7, Issue 4, pp. 989–996). KeAi Communications Co. https://doi.org/10.1016/j.aninu.2021.07.002

Han, L., Song, H., Fu, L., Li, J., Yang, L., & Liu, H. (2021). Effect of extraction method on the chemical profiles and bioactivities of soybean hull polysaccharides. Food Science and Nutrition, 9(11), 5928–5938. https://doi.org/10.1002/fsn3.2483

Hatch, C. F., & Beeson, W. M. (1972). Effect of Different Levels of Cane Molasses on Nitrogen and Energy Utilization in Urea Rations for Steers. Journal of Animal Science, 35(4). https://academic.oup.com/jas/article/35/4/854/4667132

Henry, D. D., Ciriaco, F. M., Araujo, R. C., Fontes, P. L. P., Oosthuizen, N., Rostoll-Cangiano, L., Sanford, C. D., Schulmeister, T. M., Dubeux, J. C. B., Lamb, G. C., & DiLorenzo, N. (2020). Effects of bismuth subsalicylate and encapsulated calcium-ammonium nitrate on enteric methane production, nutrient digestibility, and liver mineral concentration of beef cattle. Journal of Animal Science, 98(8). https://doi.org/10.1093/jas/skaa234

Homem, A. C., Nocera, B. F., Faleiros, L. F., Almeida, M. T. C., Paschoaloto, J. R., Perez, H. L., D'Áurea, A. P., & Ezequiel, J. M. B. (2019). Partial replacement of corn by soybean hulls in highgrain diets for feedlot sheep. Pesquisa Agropecuaria Brasileira, 54. https://doi.org/10.1590/S1678-3921.PAB2019.V54.00029

Ipharraguerre, I. R., & Clark, J. H. (2003). Soyhulls as an alternative feed for lactating dairy cows: A review. In Journal of Dairy Science (Vol. 86, Issue 4, pp. 1052–1073). American Dairy Science Association. https://doi.org/10.3168/jds.S0022-0302(03)73689-3

Kalmbacher, R. S., Brown, W. F., & Pate, F. M. (1994). Effect of Molasses-Based Liquid Supplements on Digestibility of Creeping Bluestem and Performance of Mature Cows on Winter Range'. https://academic.oup.com/jas/article/73/3/853/4632796

Krizsan, S. J., & Huhtanen, P. (2013). Effect of diet composition and incubation time on feed indigestible neutral detergent fiber concentration in dairy cows. Journal of Dairy Science, 96, 1715–1726.

Lancaster, N. A., Muegge, C. R., Carvalho, J. R. R., Lopes, R. C., Narumiya, R. S., Pinese, F., Baird, A. N., & Schoonmaker, J. P. (2020). Effect of calcium oxide and soybean hull addition to feedlot diets containing dried distillers grains and corn stover on steer performance, carcass characteristics, and digestibility. Translational Animal Science, 4(3). https://doi.org/10.1093/tas/txaa105

Li, F., Shah, A. M., Wang, Z., Peng, Q., Hu, R., Zou, H., Tan, C., Zhang, X., Liao, Y., Wang, Y., Wang, X., Zeng, L., Xue, B., & Wang, L. (2019). Effects of land transport stress on variations in ruminal microbe diversity and immune functions in different breeds of cattle. Animals, 9(9). https://doi.org/10.3390/ani9090599

Lima, M. L. P., Simili, F. F., de Medeiros, M. I. M., Neto, G. B., Ribeiro, E. G., & de Paz, C. C. P. (2014). Performance and rumen parameters of crossbred dairy cows fed two sugarcane varieties combined or not with soybean hulls. Revista Brasileira de Zootecnia, 43(12), 654–661. https://doi.org/10.1590/S1516-35982014001200005

LMC International. (2019). The Economic Impact of U.S. Soybean and End Products on the U.S. Economy. In LMC International. https://www.nopa.org/wp-content/uploads/2020/03/LMC_U.S.-Soy-Economic-Impact-Study-for-USB_FINAL-31320.pdf

Ludden, P. A., Cecava, M. J., & Hendrix, K. S. (1995). The Value of Soybean Hulls as a Replacement for Corn in Beef Cattle Diets Formulated With or Without Added Fat1/’. https://academic.oup.com/jas/article/73/9/2706/4632894

Malhi, M., Gui, H., Yao, L., Aschenbach, J. R., Gäbel, G., & Shen, Z. (2013). Increased papillae growth and enhanced short-chain fatty acid absorption in the rumen of goats are associated with transient increases in cyclin D1 expression after ruminal butyrate infusion. Journal of Dairy Science, 96(12), 7603–7616. https://doi.org/10.3168/jds.2013-6700

Mansfield, H. R., & Stern, M. D. (1994). Effects of Soybean Hulls and Lignosulfonate-Treated Soybean Meal, on Ruminal Fermentation in Lactating Dairy Cows. Journal of Dairy Science, 77(4), 1070–1083. https://doi.org/10.3168/jds.S0022-0302(94)77042-9

Martel, C. A., Titgemeyer, E. C., Mamedova, L. K., & Bradford, B. J. (2011). Dietary molasses increases ruminal pH and enhances ruminal biohydrogenation during milk fat depression. Journal of Dairy Science, 94(8), 3995–4004. https://doi.org/10.3168/jds.2011-4178

Mirza, I. H., Khan, A. G., Azim, A., & Mirza, M. A. (2002). Effect of Supplementing Grazing Cattle Calves with Urea-molasses Blocks, with and without Yucca schidigera Extract, on Performance and Carcass Traits. Asian-Australian Journal of Animal Science, 15(9). https://www.animbiosci.org/upload/pdf/15_207.pdf

Moore, J. E., Brant, M. H., Kunkle, W. E., & Hopkins, D. I. (1999). Roche/ASAS Foundation Beef Cattle Nutrition Symposium: Forage Supplementation and Grazing Effects of Supplementation on Voluntary Forage Intake, Diet Digestibility, and Animal Perf ormance1t2. https://academic.oup.com/jas/article/77/suppl_2/122/4653431

Mordenti, A. L., Giaretta, E., Campidonico, L., Parazza, P., & Formigoni, A. (2021). A review regarding the use of molasses in animal nutrition. In Animals (Vol. 11, Issue 1, pp. 1–17). MDPI AG. https://doi.org/10.3390/ani11010115

Subcommittee on Dairy Cattle Nutrition, Committee on Animal Nutrition, Board on Agriculture and Natural Resources, & National Research Council. (2001). Nutrient Requirements of Dairy Cattle: Seventh Revised Edition, 2001 (7th ed.). https://doi.org/10.17226/9825

Owens, F. N., Secrist, D. S., Hill, W. J., & Gill, D. R. (1996). Bud Britton Memorial Symposium on Metabolic Disorders of Feedlot Cattle. In J. Anim. Sci (Vol. 76). https://academic.oup.com/jas/article-abstract/76/1/275/4625198

Palmonari, A., Cavallini, D., Sniffen, C. J., Fernandes, L., Holder, P., Fagioli, L., Fusaro, I., Biagi, G., Formigoni, A., & Mammi, L. (2020). Short communication: Characterization of molasses chemical composition. Journal of Dairy Science, 103(7), 6244–6249. https://doi.org/10.3168/jds.2019-17644

Palmonari, A., Federiconi, A., Cavallini, D., Sniffen, C. J., Mammi, L., Turroni, S., D'Amico, F., Holder, P., & Formigoni, A. (2023). Impact of Molasses on Ruminal Volatile Fatty Acid Production and Microbiota Composition In Vitro. Animals, 13(4). https://doi.org/10.3390/ani13040728

Pate, F. M. (1983). Molasses in Beef Nutrition. National Feed Ingredients Association. https://rcrec-ona.ifas.ufl.edu/media/rcrec-onaifasufledu/pdf/Molasses-in-Beef-Nutrition.pdf

Pogge, D. J., Lonergan, S., Russell, J., Loy, D., & Spurlock, M. (2013). The effect of supplemental vitamin C on performance, antioxidant capacity, carcass characteristics, and meat quality of steers fed high sulfur finishing diets [Dissertation]. Iowa State University.

Reinhardt, C., & Thomson, D. U. (2015). Nutrition of Newly Received Feedlot Cattle. In Veterinary Clinics of North America - Food Animal Practice (Vol. 31, Issue 2, pp. 283–294). W.B. Saunders. https://doi.org/10.1016/j.cvfa.2015.03.010

Royes, J. B., Brown, W. F., Martin, F. G., & Bates, D. B. (2001). Source and level of energy supplementation for yearling cattle fed ammoniated hay 1. In J. Anim. Sci (Vol. 79). https://academic.oup.com/jas/article/79/5/1313/4625952

Rusche, W. (2023, August 11). Feed Bunk Management. https://extension.sdstate.edu/feed-bunk-management

Rusdiawan, A., Mar'atus Sholikhah, A., & Prihatiningsih, S. (2020). The Changes in pH Levels, Blood Lactic Acid and Fatigue Index to Anaerobic Exercise on Athlete After NaHCO 3

Administration. In Malaysian Journal of Medicine and Health Sciences (Vol. 16, Issue SUPP16).

Russell, J. R., Sexten, W. J., & Kerley M. S. (2016). soybean hull and enzyme incusion. Journal of Animal Science.

Schroedl, W., Fuerll, B., Reinhold, P., Krueger, M., & Schuett, C. (2001). Acute phase marker LPS in cattle. Journal of Endotoxin Research, 7(1). https://pubmed.ncbi.nlm.nih.gov/11521082/

Senthilkumar, S., Suganya, T., Deepa, K., Muralidharan, J., & Sasikala, K. (2016). Supplementation of molasses in livestock feed. International Journal of Sciece, Environment, 5(3). www.ijset.net

Shearer, J. (2005). Rumen Acidosis, Heat Stress and Laminitis. 11. https://cales.arizona.edu/extension/dairy/conference/proceedings/2005/shearer.pdf

Silva, B. C., Godoi, L. A., Supapong, C., Bitsie, B., Valadares Filho, S. C., & Schoonmaker, J. P. (2023). Effect of a molasses-based liquid supplement on gastrointestinal tract barrier function, inflammation, and performance of newly received feedlot cattle before and after a transport stress. Journal of Animal Science, 101. https://doi.org/10.1093/jas/skac295

Spears, J. W., Lloyd, K. E., & Fry, R. S. (2011). Tolerance of cattle to increased dietary sulfur and effect of dietary cation-anion balance. Journal of Animal Science, 89(8), 2502–2509. https://doi.org/10.2527/jas.2010-3265

Stateler, D. A., Kunkle, W. E., & Hammond, A. C. (1995). Effect of Protein Level and Source in Molasses Slurries on the Performance of Growing Cattle Fed Hay During Winter 1. In Range Sciences. https://academic.oup.com/jas/article-abstract/73/10/3078/4632623

Stein, H. H., Berger, L. L., Drackley, J. K., Fahey, G. F., Hernot, D. C., & Parsons, C. M. (2008). Nutritional Properties and Feeding Values of Soybeans and Their Coproducts.

Sun, X. Q., Wang, Y. P., Wei, R. Y., Chen, B., & Zhao, X. (2020). Effects of replacing starch with three sugars in a concentrate and forage diet on in vitro rumen fermentation, fatty acid composition and related bacteria. Animal Production Science, 60(9), 1173–1182. https://doi.org/10.1071/AN18287

Tucureanu, M. M., Rebleanu, D., Constantinescu, C. A., Deleanu, M., Voicu, G., Butoi, E., Calin, M., & Manduteanu, I. (2018). Lipopolysaccharide-induced inflammation in monocytes/macrophages is blocked by liposomal delivery of Gi-protein inhibitor. International Journal of Nanomedicine, 13, 63–76. https://doi.org/10.2147/IJN.S150918

Van Engen, N. K., & Coetzee, J. F. (2018). Effects of transportation on cattle health and production: A review. In Animal Health Research Reviews (Vol. 19, Issue 2, pp. 142–154). Cambridge University Press. https://doi.org/10.1017/S1466252318000075

Van Soest, P. J., Robertson, J. B., & Lewis, B. A. (1991). Methods for dietary fiber, neutral detergent fiber, and nonstarch polysaccharides in relation to animal nutrition. Journal of Dairy Science, 74(10), 3583–3597. https://doi.org/10.3168/jds.S0022-0302(91)78551-2

White, J. (2017). Cattle can suffer from too much sulfur. https://todaysfarmermagazine.com/mag/livestock/1303-cattle-can-suffer-from-too-much-sulfur#:~:text=The%20National%20Academy%20of%20Sciences,may%20result%20in%20sulfur%20toxicity.

Table 3.1. Effects of the inclusion of molasses or soybean hulls in backgrounding diets on total dry matter intake and ruminal fermentation profile of beef steers.

	Treatment[1]			*P*-value[3]		
Item	MOL	SBH	SEM[2]	TRT	TIME	TRT x TIME
DMI, kg/d	11.71	13.35	0.939	0.25	-	-
Ruminal pH	5.98	6.05	0.076	0.53	< 0.01	0.31
NH_3 – N, m*M*	3.24	2.75	0.393	0.40	< 0.01	0.02

[1]MOL: 46% corn silage, 25% dried distillers grains with solubles, 27% molasses, 0.5% limestone, and 1.5% vitamins and mineral packet; SBH: 46% corn silage, 22% dried distillers grains with solubles, 30% soybean hulls, 0.5% limestone, and 1.5% vitamins and mineral packet
[2]*n* = 6 steers/treatment
[3]Observed significance levels for treatment (TRT), TIME, and their interaction (TRT × TIME).

Table 3.2. Effects of the inclusion of molasses or soybean hulls in backgrounding diets on ruminal VFA concentrations of beef steers.

	Treatment[2]			*P*-value[4]		
Item[1]	MOL	SBH	SEM[3]	TRT	TIME	TRT x TIME
VFA, m*M*						
Acetate	45.68	52.66	2.065	0.04	< 0.01	0.01
Propionate	13.96	15.43	0.582	0.11	< 0.01	0.04
Butyrate	15.64	11.83	0.732	0.006	< 0.01	0.32
BCVFA[5]	0.56	0.86	0.064	0.01	0.01	< 0.01
Valerate	1.47	1.29	0.106	0.27	< 0.01	< 0.01
Caproate	0.098	0.064	0.012	0.08	< 0.01	0.01
Total VFA, m*M*	77.43	82.16	3.301	0.34	< 0.01	0.04
A:P[6]	3.33	3.49	0.070	0.14	< 0.01	0.50

[1]Ruminal fluid was collected every 3 h for 24 h.
[2]MOL: 46% corn silage, 25% dried distillers grains with solubles, 27% molasses, 0.5% limestone, and 1.5% vitamins and mineral packet; SBH: 46% corn silage, 22% dried distillers grains with solubles, 30% soybean hulls, 0.5% limestone, and 1.5% vitamins and mineral packet
[3]*n* = 6 steers/treatment
[4]Observed significance levels for treatment (TRT), TIME, and their interaction (TRT × TIME).
[5] BCFVA = branched chain VFA: isobutyrate + isovalerate + 2 methylburyrate
[6]Acetate to propionate ratio

Table 3.3. Effects of the inclusion of molasses or soybean hulls in backgrounding diets on ruminal VFA molar proportions of beef steers.

	Treatment[2]			*P*-value[4]		
Item[1]	MOL	SBH	SEM[3]	TRT	TIME	TRT x TIME
VFA, mol/100mol						
Acetate	59.54	64.65	0.514	< 0.01	< 0.01	0.43
Propionate	18.01	18.70	0.295	0.13	< 0.01	0.61
Butyrate	19.70	13.95	0.446	0.01	< 0.01	0.24
BCVFA[5]	0.74	1.07	0.076	0.01	< 0.01	0.86
Valerate	1.86	1.53	0.102	0.05	< 0.01	<0.01
Caproate	0.12	0.07	0.014	0.05	< 0.01	0.05

[1]Ruminal fluid was collected every 3 h for 24 h.
[2]MOL: 46% corn silage, 25% dried distillers grains with solubles, 27% molasses, 0.5% limestone, and 1.5% vitamins and mineral packet; SBH: 46% corn silage, 22% dried distillers grains with solubles, 30% soybean hulls, 0.5% limestone, and 1.5% vitamins and mineral packet
[3]n = 6 steers/treatment
[4]Observed significance levels for treatment (TRT), TIME, and their interaction (TRT × TIME).
[5] BCFVA = branched chain VFA: isobutyrate + isovalerate + 2 methylburyrate

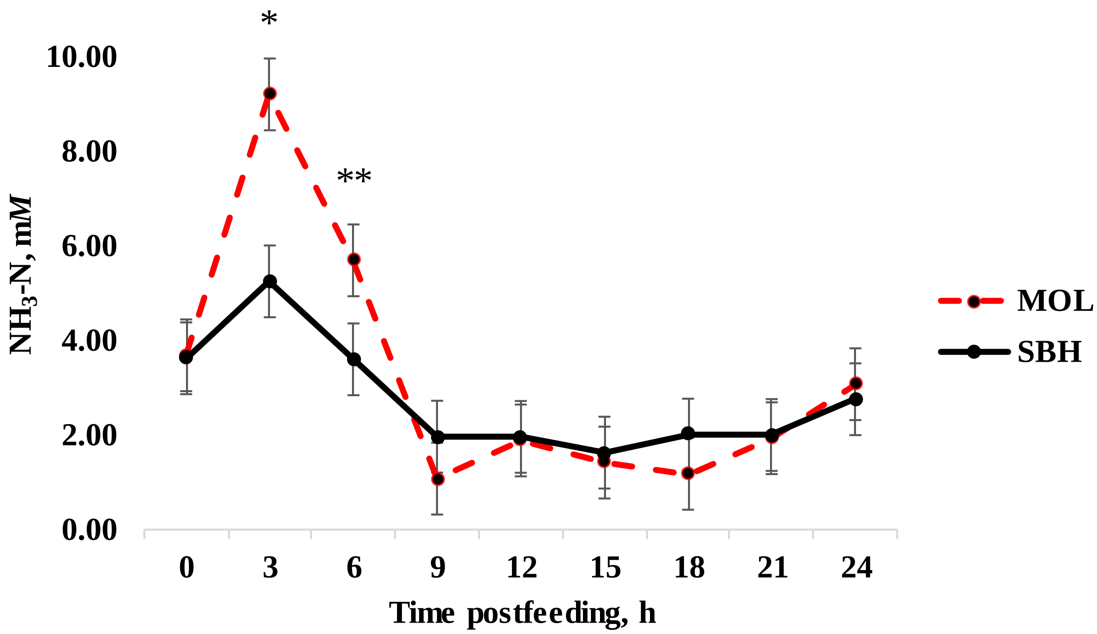

Figure 3.1. Effects of the inclusion of molasses or soybean hulls in backgrounding diets on ruminal on NH_3-N concentrations of beef steers. MOL: 46% corn silage, 25% dried distillers grains with solubles, 27% molasses, 0.5% limestone, and 1.5% vitamins and mineral packet; SBH: 46% corn silage, 22% dried distillers grains with solubles, 30% soybean hulls, 0.5% limestone, and 1.5% vitamins and mineral packet.A treatment × time interaction post-feeding was observed ($P = 0.02$). $n = 6$ steers/treatment. Error bars represent the SEM for treatment × time interaction. * ($P < 0.01$); ** ($P = 0.05$)

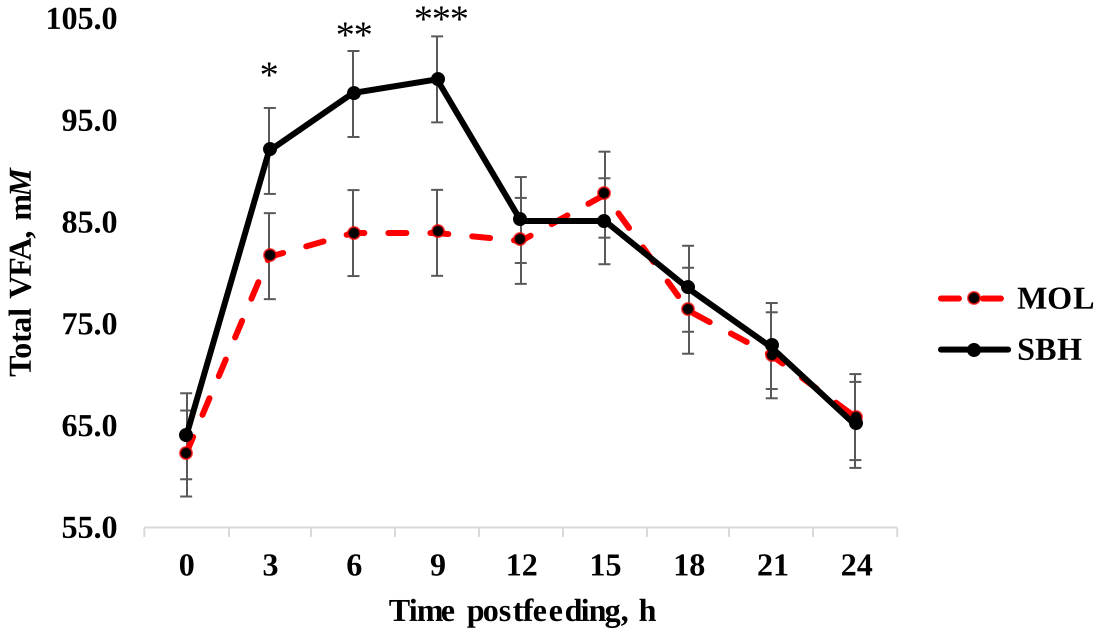

Figure 3.2. Effects of the inclusion of molasses or soybean hulls in backgrounding diets on ruminal total VFA concentration of beef steers. MOL: 46% corn silage, 25% dried distillers grains with solubles, 27% molasses, 0.5% limestone, and 1.5% vitamins and mineral packet; SBH: 46% corn silage, 22% dried distillers grains with solubles, 30% soybean hulls, 0.5% limestone, and 1.5% vitamins and mineral packet. A treatment × time interaction post-feeding was observed (P = 0.05). n = 6 steers/treatment. Error bars represent the SEM for treatment × time interaction. * (P = 0.09); ** (P = 0.02); *** (P = 0.01)

CHAPTER 4

IMPLICATIONS AND RECOMMENDATIONS

The results from these experiments indicate that molasses can be an option for replacing soybean hulls in backgrounding diets without negative impacts on beef cattle performance. The availability of commodities like soybean hulls can be impacted by the production of the soybean industry. This may cause an increase in the cost of the byproduct, making it more expensive for the producer and increasing the most important cost on a cattle operation, that is the cost of feeding. As well, the cost of molasses is going to be impacted by the production of sugar cane and its availability on the market. By having similar results on performance, the producers will have the ability to choose which of these two commodities can be utilized in the diets of backgrounding cattle and not increasing the cost of feed. Similar DM intake, OM intake and CP intake were observed in this study between both treatments. As expected, NDF and ADF intake were greater for steers consuming a diet with an inclusion of soybean hulls due to the high content of structural carbohydrates in the soybean hulls. However, even though there is a greater content of fiber in the backgrounding diets with an inclusion of soybean hulls, a 27% inclusion of molasses on a DM basis to the backgrounding diets can reduce the fiber digestibility. These results also indicate that total CH_4 produced daily was not reduced by the inclusion of molasses in the diets; however, there was a numerical reduction in the molasses treatment. In addition, when CH_4 produced was calculated per kilogram of organic matter digested, a reduction from steers fed a backgrounding diet with a 27% inclusion of molasses was observed. In addition, when CH_4 produced per kilogram of NDF digested and NDF intake is calculated, it was reduced for steers consuming a backgrounding diet with an inclusion of soybean hulls. Also, the inclusion of 27% molasses can shift the ruminal VFA profile, increasing the molar proportion and concentration of butyrate at the expense of acetate. Concentrations of NH_3-N were greater at 3 h post feeding for steers fed a diet with the inclusion of molasses and the pH was not be negatively impacted when 27% of molasses was included in the diets compared to an inclusion of soybean hulls. Since the COVID pandemic began, there has been a greater demand of locally raised beef thought the US. In addition, there is an increased interest from producer on retaining the ownership of the calves throughout the finishing phase of the beef industry. Having the option to choose between two commodities that will have similar effects on performance, can benefit the producer to fulfill the objective of finishing its own calves inside his cattle operation.