HEAT-TREATED SOYBEAN MEAL IN FORAGE-BASED CATTLE DIETS

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The Supervisory Committee certifies that this ***disquisition*** complies with North Dakota State University's regulations and meets the accepted standards for the degree of

MASTER OF SCIENCE

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ABSTRACT

Two experiments were conducted to evaluate replacing dried distillers grains with solubles (DDGS) with heat-treated soybean (TSBM) at increasing rates in forage-based diets on steer growth performance, nutrient flow, and digestibility. Experiment 1: 70 angus-based steers were utilized in a generalized randomized block design for an 85-d growing study at the NDSU Beef Cattle Research Complex. Experiment 2: five fistulated Jersey steers fitted with ruminal, duodenal, and ileal cannulas were utilized in a 4 × 5 Latin square design for 56 d to measure nutrient flow and digestibility. Diets were formulated with TSBM replacing DDGS, at 16% of diet dry matter, at 0 (TSBM0), 4 (TSBM4), 8 (TSBM8), and 12% (TSBM12) TSBM. Treatments had no effect on steer growth performance and residual carcass characteristics from the growing phase. Nitrogen, lysine, and available lysine total tract digestibility increased with increased TSBM inclusion; however, post ruminal digestion decreased.

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DEDICATION

To my Grandpa Waldner, “No one can take your education away from you.”

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LIST OF ABBREVIATIONS

˚C	Celsius
µmol/L	Micromole per liter
ADF	Acid detergent fiber
ADG	Average daily gain
BF	Backfat thickness
BW	Body weight
Ca	Calcium
cm	Centimeter
Co-EDTA	Cobalt-ethylenediamine tetraacetic acid
CP	Crude protein
d	Day
DCGF	Dried corn gluten feed
DDG	Dried distillers grains without solubles
DDGS	Dried distillers grains with solubles
DM	Dry matter
DMI	Dry matter intake
dRUP	Digestible rumen undegradable protein
EE	Ether extract
EBW	Ending body weight
FBW	Final body weight
g	Gram
g/d	Grams per day
G:F	Gain to feed
h	Hour

HCW	Hot-carcass weight
in	Inch
IU	International unit
kg	Kilogram
kg/d	Kilograms per day
KPH	Kidney, pelvic, heart fat
lb	Pound
LMA	Longissimus muscle area
MARB	Marbling
min	Minute
mL	Milliliter
mm	Millimeter
mmol	Millimole
MP	Metabolizable protein
N	Nitrogen
NDF	Neutral detergent fiber
NDSU	North Dakota State University
NEFA	Non-esterified fatty acid
NEg	Net energy for gain
NEBSBM	Non-enzymatically browned soybean meal
OM	Organic matter
P	Phosphorus
PUN	Plasma urea nitrogen
QG	Quality grade
RDP	Rumen degradable protein

RUP	Rumen undegradable protein
SB	Soybean
SBM	Soybean meal
SEM	Standard error of the mean
TDN	Total digestible nutrients
TRT	Treatment
TSBM	Heat-treated soybean meal
WCGF	Wet corn gluten feed
WDDGS	Wheat dried distillers grains with solubles
WDGS	Wet distillers grains with solubles
YG	Yield grade

1. CHAPTER 1: LITERATURE REVIEW

1.1. Introduction

Soybean meal (SBM) has become a less prominent protein source in beef cattle diets since ethanol production increased exponentially in the early 2000s due to the passage of the Clean Air Act in 1990 (Abebe, 2008). The increase in ethanol production substantially increased distillers grains production which lead to a more economical protein source than SBM. However, an increasing interest in renewable fuel will potentially cause a similar scenario for SBM. The United States Renewable Fuel Standard Program is using governmental incentives and requirements to expand biodiesel production through oilseeds (United States Environmental Protection Agency, 2024). The program has led to the potential of 13 new soybean crushing plants and the expansion of 10 which would add 750 million bushels per year of crush capacity (American Soybean Association, 2022). Therefore, an increase in SBM production is anticipated as more crushing plants begin operations. The influx of SBM production provides livestock producers with potentially another protein feedstuff to utilize in rations.

Therefore, the objectives of this literature review are to discuss processes that create soybean meal products, and analyze differences in nutrient digestibility, growth performance, and carcass characteristics of various soybean meal products and other protein feedstuffs.

1.2. Soybean Meal Products

1.2.1. Solvent-Extracted Soybean Meal

There are three major processes to create SBM from raw soybeans: 1) solvent extraction (hulled and dehulled), 2) expeller extraction, and 3) extrusion. The most common process is dehulled solvent extraction (Ishler and Varga, 2016). The solvent extraction process is similar for both hulled and dehulled SBM where soybeans are ground and hexane is applied to remove fat

(Ishler and Varga, 2016). The difference between the hulled and dehulled products is the dehulled solvent extracted SBM process removes the outer hull to create a product that has a greater crude protein (CP) concentration (52.9% CP; NASEM, 2016) and lower neutral detergent fiber (NDF; 11.3% NDF; NASEM, 2016), whereas hulled solvent extracted SBM blends toasted ground soybean hulls making it slightly lower in CP concentration (46.5% CP; NASEM, 2016) but greater in NDF concentration (18.8% NDF; NASEM, 2016) because of the fibrous content of the hulls. Due to commonality and availability, dehulled solvent extracted SBM is typically the base product available for other SBM processes. Monogastric diets include more SBM compared to ruminants (Ruiz, et al., 2020) with approximately only 9% of the total United States SBM usage being incorporated into dairy diets and approximately 0.8% in beef diets (DIS, 2018). Soybean meal is often compared to other protein feedstuffs and is considered to be the reference for protein feedstuffs due to its amino acid profile and ruminal degradation qualities (Ruiz et al., 2020). Soybean meal contains a higher concentration of lysine compared to other feedstuffs (6.16, % of CP; NASEM, 2021). The high lysine concentration in SBM provides an opportunity for cattle producers to utilize it in diets that may be deficient in lysine. As corn-based feedstuffs are limited in lysine content (Abe et al., 1997), incorporating SBM in beef cattle diets will be discussed more later. However, the SBM inclusion may boost growth performance in many different situations within the cow/calf and feedlot cattle industries.

1.2.2. Expeller Extracted Soybean Meal

Expeller extracted and extruded SBM processes are less common at soybean crushing plants. The respective products are slightly different and offer more energy and rumen undegradable protein (RUP) than solvent extraction. Like solvent extraction, the expeller extraction process begins with the soybeans being ground and dried (Lin and Kung, 1999). The process utilizes pressure (Lin and Kung, 1999) from screw presses that force soybeans through a die to remove the oil (Bhargavi et al., 2018). Heat is generated to a maximum 163˚C to initiate the Maillard reaction and to avoid negatively affecting intestinal digestion (Lin and Kung, 1999). The Maillard reaction increases RUP to 63% compared to dehulled solvent extraction at 33% of CP (NASEM, 2021). Additionally, the protein content is slightly lower for expeller SBM at 47.6% of CP (NASEM, 2021). Less oil is removed from the process which increases the energy value slightly compared to dehulled solvent extracted SBM (7.12 and 1.82% crude fat, respectively; NASEM, 2021).

1.2.3. Extruded Soybean Meal

Extrusion can be applied to both whole raw soybeans and solvent extracted SBM (Lin and Kung, 1999). Consequently, the extruded end-product needs clarification because the Beef Cattle NASEM (Extruded Soybeans; NASEM, 2016) and Dairy Cattle NASEM (Extruded Soybean Meal; NASEM, 2021) discuss only one extruded product, however, with similar reported values. For this review, whole raw soybeans that are extruded will be referred to as "extruded soybean" and extruded SBM will be referred to as the product after extrusion of SBM. For extruded soybeans, the extrusion process starts with whole beans flowing into the extruder barrel that utilizes a spiral and tapered screw to break down the soybeans into a meal which is then forced through a narrow opening (Shaver, n.d.). During the process, friction and steam are incorporated for heat treatment (AAFCO, 1997) which results in a relatively time-consuming process (1-10 tons/h; Lin and Kung, 1999). Unlike the other two soybean processes, extrusion does not remove oil during the process (Lin and Kung, 1999) and is labeled as full-fat extruded soybeans (SB). Therefore, full-fat extruded SB contains more crude fat (13.14%, NASEM, 2016) and less crude protein (44.4%; NASEM, 2016) than solvent extracted and expeller SBM products. For extruded SBM, approximately 17% moisture is added to solvent extracted SBM before entering the same extrusion process (AOAC, 1975). The final extruded SBM product decreased the protein content to 40.4% CP (NASEM, 2021) compared to solvent extracted SBM (52.6% CP; NASEM, 2021); however, the RUP content increased from 33 to 45% of CP with extruded SBM (NASEM, 2021). Extruding SBM increases the fat content compared to solvent extracted SBM (20.42% and 1.82%, respectively; NASEM, 2021).

The oil content of expeller and extruded SB and SBM products does not align with the current direction of the soybean crushing and soybean oil industry. Currently, the demand for expeller and extruded products is very minimal and almost non-existent to beef cattle producers. The increase in soybean oil demand and prices will cause both processes to adjust for more oil extraction. The extrusion process has incorporated an expeller step to press more oil out of the extruded SB product. It is commonly referred to as the extruder-expeller process and will consequently decrease the fat content to approximately 8% (Powell et al., 2011). The extruder-expeller process does decrease the fat content however, it may not be low enough to incorporate into rations as an additional rumen protected protein source at an economical price for beef cattle producers.

1.2.4. Processed Soybean Meal

The amino acid content of solvent extracted SBM makes it a feedstuff that could be used to potentially reduce ruminal degradation, increase amino acid flow to the small intestine, and enhance animal performance (Ruiz et al., 2020). There are multiple processes to heat treat soybean meal to create a product greater in RUP than dehulled solvent extracted SBM and to potentially enhance the amino acid use postruminally. Unfortunately, due to private industry product development, processes are proprietary and limited information is available on the exact processing methods. Exact processes discussed may be slightly different based on heat-treated SBM product that a company produces, however the general product concept is the same.

1.2.4.1. Non-Enzymatically Browned Soybean Meal

Non-enzymatically browned SBM (NEBSBM) combines dehulled solvent extracted SBM with a reducing sugar (xylose or glucose) and toasting for a specified period of time during the desolventizing process (Cleale et al., 1987). A Maillard reaction is induced during the process between sugar aldehyde groups and free amino groups (Hodge, 1953). The reaction protects protein from ruminal degradation yet remains available for digestion in the animal in the small intestine. Solvent extracted SBM is very degradable in the rumen (67% of CP as RDP; NASEM, 2021). After non-enzymatic browning treatment, ruminal degradation is decreased to approximately 28% of CP as RDP (AminoPlus, Ag Processing Inc., Omaha, NE and SoyPass, LignoTech USA, Rothschild, WI). Multiple studies confirm the reduced ruminal degradation (Cleale et al., 1987; Borucki Castro et al., 2007). However, there were no differences in nitrogen reaching the small intestine (Cleale et al., 1987; Ipharraguerre et al., 2005) and post ruminal digestibility (Ipharraguerre et al., 2005) compared to solvent extracted SBM in beef cattle. The relatively high digestible RUP content of solvent extracted and NESBM through in vitro disappearance (87.5 and 79.4%, respectively; Borucki Castro et al., 2007) may explain the observed lack of difference in post ruminal digestibility. Besides RUP content, both processes create similar nutrient profiles for crude protein (approximately 50%) and crude fat (approximately 1.5%; NASEM, 2021; Amino Plus; SoyPass).

1.2.4.2. Roasted Soybean Meal

Roasting SBM is another method that can be used to alter site of digestion although it is not common. The process utilizes either a drum roaster or conveyor system. The drum roaster rotates to lift the SBM through the flame (Lin and Kung, 1999), whereas, the conveyor system moves the SBM through the heating chamber (Shaver, n.d.). A unique benefit to roasting is the potential for on-farm use due to mobile roasting equipment (Lin and Kung, 1999). The uncertainty with roasted SBM is the lack of information on the optimal time and heating conditions to achieve the best results (Lin and Kung, 1999). Previous research has not comprehensively reported nutrient analysis values of roasted SBM; however, Demjanec et al. (1994) reported values for multiple experimental roasting times and discovered CP was 51.9% for all roasting times. Interestingly, the increased roasting time from 0 to 210 min increased NDF (9.2 to 67.6%, respectively) and ADF (5.8 to 22.9%, respectively) concentration. Research has shown that roasting at 130 to 145˚C increased RUP concentration by 100% (Plegge et al., 1985); however, a study from the same group indicated that roasting at 144 to 159˚C is optimal for the roasting conditions in that experiment (Plegge et al., 1982). Increased ruminal escape has been confirmed through multiple studies (Plegge et al., 1982; Plegge et al., 1985; Demjanec et al., 1994); however, only one study reported increased small intestinal N digestibility (Demjanec et al., 1994). Demjanec et al. (1994) reported a quadratic effect for N disappearance in the small intestine with increasing roasting time from 0 to 210 min at 165 ˚C using fistulated wethers. The time with the greatest disappearance was 150 min.

1.3. Digestibility of Soybean Meal Products

Solvent extracted SBM is very degradable in the rumen (70% of CP, NASEM, 2016) which led to the creation of various processing methods described above to decrease ruminal degradation. Ipharraguerre et al. (2005) evaluated solvent extracted SBM, expeller SBM, NEBSBM, and whole roasted SB in approximately 18% CP diets that are corn silage and ground shelled corn based. Total N flow to the duodenum and ruminal digestibility was not influenced by treatment; however, ruminal ammonia concentrations decreased for expeller and NEBSBM compared to solvent extracted SBM and whole raw SB. The decreased ammonia concentrations potentially suggest less ruminal N degradation for expeller SBM and NEBSBM. Feed nitrogen at the duodenum was greater for NEBSBM and whole raw SB compared to solvent extracted SBM and expeller SBM was intermediate. Although expeller and NEBSBM has increased RUP, no differences were observed for post ruminal digestibility which suggests solvent extracted SBM, expeller SBM, and NEBSBM are digested similarly in the small and large intestines. Furthermore, feed lysine flowing to the duodenum increased for NEBSBM and whole raw SB compared to solvent extracted SBM, whereas, expeller SBM was intermediate. Although ruminal digestibility was not different, total amino acid flow was increased for NESBM compared to solvent extracted and expeller SBM. Therefore, feeding NESBM resulted in increased flow of protein to the small intestine. Furthermore, Cleale et al. (1987) reported increased nonammonia N, bacterial N, and dietary N flow to the duodenum for SBM and NESBM compared to urea. Non-enzymatically browned SBM was effective in decreasing rumen degradation and increasing dietary N flow to the duodenum. Additionally, there was no difference in total tract N digestibility between all three treatments.

Borucki Castro et al. (2007) used in situ and in vitro techniques to evaluate digestibility of solvent extracted, expeller, and NESBM. Feed samples were incubated for 16 h in the rumen and then randomly assigned to either the 3-step procedure described by Calsamiglia and Stern (1995) or the mobile nylon bag technique (Hvelplund and Weisbjerg, 2000) to evaluate potential differences in post-ruminal digestion. In situ and in vitro samples were analyzed for nitrogen and amino acid concentrations from ruminal and intestinal disappearance. Unsurprisingly, after rumen incubation solvent extracted SBM was degraded to a greater extent by rumen microbes compared to expeller SBM and NESBM (70.2, 37.8, and 30.0%, respectively). Furthermore, lysine degradation followed the same pattern with solvent extracted SBM having greater ruminal degradation. Non-enzymatically browned SBM had less ruminal degradation than expeller SBM. Using the in situ method to analyze intestinal disappearance, expeller SBM and NESBM had slightly greater nitrogen and lysine digestibility. Interestingly, the in vitro method reported increased N and lysine disappearance for solvent extracted SBM compared to NESBM and expeller SBM. If taken one step further and evaluated on availability, which is the proportion of the total amino acid that is digested and absorbed (Batterham, 1992), expeller SBM and NESBM lysine concentrations are more available compared to solvent extracted SBM. The lysine composition of each SBM product was similar ranging from 2.58 to 2.99% DM. Therefore, heat-treating solvent extracted SBM was shown to decrease ruminal degradation and increase amino acid supply to the small intestine.

As mentioned previously, solvent extracted SBM is often considered the reference point when evaluating protein sources (Ruiz et al., 2020). Therefore, numerous protein feedstuffs are compared to SBM. Loerch et al. (1983) compared blood meal, meat and bone meal, dehydrated alfalfa, urea, and soybean meal. The urea treatment had the lowest feed N flow to the duodenum followed by SBM. Blood meal, a known RUP source, and dehydrated alfalfa had the greatest flow of feed amino acids. The results suggest that blood meal, meat and bone meal, and dehydrated alfalfa will increase N flow to the duodenum from feed compared to SBM and urea. Although SBM is a known rumen degradable protein source, the small RUP fraction still provides increased rumen escaped N compared to urea. Furthermore, Maxin et al. (2013) compared canola meal, high protein corn distillers grains with solubles (DDGS) and wheat dried distillers gains with solubles (WDDGS) to SBM to evaluate ruminal degradability using an in situ method. Soybean meal and WDDGS were more degraded in the rumen followed by canola meal. High protein DDGS contained the largest concentration of RUP. In terms of amino acid ruminal disappearance, canola meal and high protein DDGS consistently decreased ruminal degradation compared to SBM and WDDGS.

1.4. Cattle Growth Performance with Soybean Meal Products

1.4.1. Soybean Meal vs. Urea

Soybean meal and urea traditionally were common supplemental protein sources in beef cattle finishing rations (Milton et al., 1997a). Urea is utilized in finishing rations due to the lower cost (Milton et al., 1997b) but SBM is not often utilized because it is a relatively expensive protein source compared to urea and other protein sources such as corn distillers grains. Urea and SBM contain high concentrations of rumen degradable protein that supplies rumen microbes with N sources for microbial production (Zinn and Owens, 1983). Furthermore, unlike urea, SBM is a natural protein source that additionally provides RUP which assists in increasing the metabolizable protein content in the diet (Milton et al., 1997). Several studies have been conducted comparing SBM and urea in finishing rations with fewer studies in growing diets.

1.4.1.1. Growing Cattle Performance

A study evaluated three levels of urea (low, medium, and high) and solvent extracted SBM in isocaloric corn-based diets and with the high urea and SBM treatments to be approximately 10.8% CP. During the first 70 d, average daily gain (ADG) and gain:feed (G:F) of cattle fed the SBM treatment was greater compared to the low and medium urea levels; however, the high urea level treatment exhibited lower ADG and no difference in G:F compared to the SBM treatment. During the entire 112-d study, greater dry matter intake (DMI) than the high and low urea treatments. Additionally, steers fed SBM had greater ADG compared to all urea treatments and had greater G:F than the medium urea treatment (Thomas et al., 1984).

1.4.1.2. Finishing Cattle Performance

In ground ear corn-based diets, SBM and urea-based diets were formulated to be isonitrogenous and isocaloric and fed for 112 d. No difference in final body weight (BW) was reported; however, ADG was slightly greater for steers fed SBM compared to urea. Steers fed SBM for the first 56 d and urea for the last 56 (SBM-none) had no difference in ADG compared to calves fed SBM for the entire experiment and outperformed the urea fed treatment (Young et al., 1973). The SBM-none group showed the potential to utilize a natural protein source first until the amino acid requirements decrease and then use urea as an economical protein replacement. Milton et al. (1997) analyzed urea and solvent-extracted SBM at two nitrogen levels (1.93 and 2.25%) in approximately 80% rolled corn-based diets for 132 d. Over the entire 132 d finishing period, ADG and G:F were increased for SBM compared to urea treatments regardless of nitrogen level. Additionally, steers fed the higher nitrogen level for both SBM and urea were more efficient than the lower level. For the first 70 d of the experiment, steers fed SBM had greater ADG compared to steers fed urea which is attributed to SBM containing more RUP than urea and, therefore, increasing the MP concentration in the diet. Cosby et al. (1997) fed urea, solvent-extracted SBM, and expeller SBM for the first 84 d and the remainder of the 194-d experiment fed urea in whole corn-based diets that were formulated to be isonitrogenous. During the first 84 d, urea-supplemented steers ate less feed compared to solvent-extracted SBM and expeller SBM supplemented steers. However, no differences in DMI, ADG, and G:F were observed for the entire 194 d experiment.

Multiple studies have reported no differences or increased performance when evaluating SBM and urea. Soybean meal as a natural protein source is providing additional MP through additional RUP when the microbial RDP is met, whereas, urea does not supply any RUP. Therefore, utilizing solvent-extracted SBM in growing cattle diets will potentially increase cattle performance. Growing cattle require more available microbial protein and dietary amino acids at the small intestine (Young et al., 1973) and microbial protein production from only dietary urea may not meet those requirements. As performance did not decrease with SBM inclusion in the rations, incorporating SBM or urea will be based on economics and availability. However, there is limited recent research comparing SBM and urea largely in part due to recent ethanol production and availability of distillers grains.

1.4.2. Solvent-Extracted Soybean vs. Corn Byproducts

An increase in ethanol production in the early 2000s due to the passage of the Clean Air Act in 1990 (Abebe, 2008) substantially increased the distillers grains supply. The increased supply caused distillers grains to become a more economical protein source than SBM for cattle. Furthermore, high inclusion rates of distillers grains in feedlot rations without negative effects on performance (Schoonmaker et al., 2013) resulted in decreased utilization of SBM. However, soybean crushing capacity, similar to ethanol production in the early 2000s, is increasing due to the United States Renewable Fuel Standard Program. The program is using governmental incentives and requirements to expand biodiesel production through oilseeds (United States Environmental Protection Agency, 2023). The increase in soybean crushing capacity will potentially increase the availability of SBM for livestock producers (United Soybean Board) and potentially decrease the price of SBM. Very little recent research is available comparing SBM with corn and corn byproducts. Therefore, evaluating SBM and distillers grains as protein sources in beef cattle diets is an important part to aligning with future economical byproduct availability.

1.4.2.1. Growing Cattle Performance

An experiment with diets balanced to contain 12% CP and 64% total digestible nutrients (TDN) in corn silage-based diets that included SBM and DDGS at 12 and 22% DM, respectively, were evaluated as protein sources for ruminants. The DDGS treatment diet resulted in increased ADG compared to the SBM treatment. However, DMI and G:F were not different between SBM and DDGS fed calves (Waller et al., 1980). In contrast, Waller et al. 1980 also evaluated several supplemental nitrogen levels from urea (40 - 100% of supplement N as urea) in combination with three natural protein sources, solvent extracted SBM, DDGS, and dried distillers grains without solubles (DDG). Six 12% CP, ground corn cob-based dietary treatments were compared: 1) 100% urea, 2) 60% SBM and 40% urea, 3) 60% DDG and 40% urea, 4) 60% DDGS and 40% urea, 5) 30% DDG, 30% SBM, and 40% urea, and 6) 30% DDGS, 30% SBM, and 40% urea. Increasing the natural protein source linearly increased steer ADG and G:F. Furthermore, urea combined with DDGS and DDG had greater G:F than urea and SBM (treatments 3 and 4 vs. 2). The combination of DDG and SBM was intermediate in G:F to SBM and DDG fed separately (treatment 2 < 5 < 3). Additionally, SBM and the combination of DDGS and SBM did not perform differently.

For early weaned calves, a 99-d feeding study was conducted to evaluate 0, 30, and 60% inclusion of dried distillers grains plus solubles (DDGS) in a corn-based diet where the control diet utilized solvent extracted SBM at 17% of the diet DM. The 0% and 30% DDGS inclusion was similar in CP content (15.7%) however, the 60% diet was 21.7% CP, and the increased DDGS inclusion provided greater RUP content compared to the SBM control. Although the energy content of the diet slightly increased with DDGS inclusion, where steer ending BW, ADG, DMI, and G:F was not different (Schoonmaker et al., 2013). Furthermore, Prichard and Boggs (2006) conducted a receiving experiment with diets containing either solvent extracted SBM or DDGS in oat hay and rolled corn-based diets for 47 d. Soybean meal was included at 11.8% of diet DM and DDGS was included at 18.9% of diet DM which led to the CP content being slightly greater for the DDGS diet. (12.9 vs. 13.4% CP, respectively). There were no differences in ending BW, ADG, and G:F reported (Pritchard and Boggs, 2006). Mueller and Boggs (2011) evaluated oat silage and soybean hull-based diets, formulated to be isonitrogenous with diets containing solvent extracted SBM, DDGS, or dried corn gluten feed (DCGF). The initial 28 d did not result in ADG, DMI, and G:F differences for SBM, DDGS, and DCGF. Over the entire 52 d receiving experiment, no treatment differences in growth performance were observed (Mueller and Boggs, 2011). Firkins et al. (1985) replaced SBM and corn silage with dry corn gluten feed (DCGF), wet corn gluten feed (WCGF), and DDGS to balance CP at 11% of the diet DM in growing cattle diets. The SBM treatment had the lowest ADG and poorest G:F compared to other treatments. The energy content between all the diets was likely the main contributor to differences in performance. The SBM diet had the lowest energy content which potentially led to decreased ADG and G:F; however, no difference in ending BW was observed. Solvent extracted SBM contained the lowest crude fat content which potentially decreased ADG for SBM-fed steers (Firkins et al., 1985).

When evaluating RUP content of growing cattle diets, Wiseman (2021) evaluated replacing fine ground corn with solvent extracted SBM in corn silage-based diets. Fine ground corn, included in the diet at 18% DM, was replaced with solvent-extracted SBM at rates of 0, 4.5, 9, 13.5, and 18% diet DM. Linear increases in ADG and G:F were observed with the increased solvent-extracted SBM inclusion. Although SBM is very rumen degradable, the increased solvent extracted SBM concentration linearly increased daily RUP intake (Wiseman, 2021). The increased RUP concentration from solvent extracted SBM is likely the reason for increased performance compared to corn. The difference in protein concentration likely caused the increased response to solvent extracted SBM over corn as the CP concentration in corn is lower (52 and 8%, respectively; NASEM, 2016).

1.4.2.2. Finishing Cattle Performance

Although unconventional for commercial finishing cattle, Pittaluga et al. (2021) offered ad libitum grass hay to 120 crossbred steers (566-kg initial BW) and supplemented with concentrate mixes to determine performance between supplements containing DDGS and SBM. Additionally, DDGS was included at 15% diet DM whereas SBM was included at 8.5% diet DM in the respective concentrate mixes. Steers fed the SBM treatment were heavier and had greater G:F than the DDGS counterparts although hay intake was lesser for SBM-fed calves. However, the difference in CP between treatment diets must be noted (DDGS = 12% CP, SBM = 14% CP). Mateo et al. (2004) evaluated 20 and 40% inclusion of DDGS and wet distillers grains plus solubles (WDGS) in cracked corn-based diets with the control diet containing 10.5% SBM. The 138 and 129 d trial (year 1 and 2, respectively) detected no differences between the SBM control and 20 and 40% DDGS and WDGS inclusion rates (Mateo et al., 2004). In dry rolled corn-based diets, solvent extracted SBM, DDGS, and the combination of DDGS and SBM were balanced to 13.2% CP and evaluated in a 132-d study. Interestingly, no differences in final BW, ADG, DMI, and G:F were reported across the treatment diets (Pritchard, 2010). When Firkins et al. (1985) evaluated treatment diets containing SBM, urea, WCGF, and DCGF in corn silage-based diets at 12% CP were evaluated. Wet corn gluten feed and DCGF were included at 50% of the diet DM, whereas SBM was included at 7.8%. Calves fed SBM consumed less DM than DCGF and WCGF but more than urea. The differences in intake but not ADG resulted in DCGF having the lowest G:F (Firkins et al., 1985).

The difference in diet composition may affect the response to distillers grains or SBM inclusion in the diet. Feeding solvent extracted SBM or distillers grains may not influence growth performance in cattle fed lower energy, receiving diets. Cattle fed lower energy diets may respond greater to solvent extracted SBM inclusion, due to the increased ruminal degradation and lysine content (NASEM, 2021), especially when compared to energy feed sources. However, in higher energy diets, feeding distillers grains may result in improved growth performance compared to SBM because greater crude fat concentration (approximately 10.5% vs. 1.88%, respectively; NASEM, 2016). Corn-based diets are typically limited in lysine (Abe et al., 1997), and supplying solvent extracted SBM may balance the amino acid content in the diet and potentially improve growth performance through increasing the lysine concentration in both growing and finishing diets.

1.4.3. Solvent-Extracted Soybean Meal vs. Other Soybean Meal Products

1.4.3.1. Growing Cattle Performance

Three studies from the same research group (Plegge et al., 1983) evaluated solvent extracted and roasted SBM in medium to high energy diets fed to ~225-kg steers. All three studies had basal diets of ground corn cobs and high-moisture corn and were balanced to contain a specific CP percentage based on the study. Across all three studies, no differences in performance were detected between the inclusion of solvent extracted or roasted SBM in the diets. Due to the similar inclusion rates of solvent extracted and roasted SBM in the treatments and balanced CP content, the assumed increase in RUP from roasted SBM did not increase performance.

Two limit-fed diets containing wheat middlings or soybean hulls were offered to growing steers to evaluate the low and high inclusions of solvent extracted SBM and NESBM on steer growth performance. Both SBM products replaced dry-rolled corn, included at 12% diet DM, at 6 (low) and 12% (high) DM. There were no differences in ADG and G:F for steers supplemented solvent extracted SBM in the wheat middlings-based diet. In contrast, a linear increase was detected for ADG and G:F with the increasing level of NESBM in the wheat middlings-based diet. Feeding the soyhull based diet resulted in increased ADG and G:F with the low inclusion of both SBM products; however, growth performance did not increase at the high inclusion level (Coetzer, 2000). Coetzer (2000) additionally evaluated increasing rates of solvent extracted SBM and NESBM in high-moisture corn and cottonseed hull-based diets. Diets were balanced to achieve 40, 60, 80, and 100% of the supplemental CP from solvent extracted SBM inclusion and 15, 30, 45, and 60% of the supplemental CP from NESBM inclusion. There were no differences in ADG and G:F in cattle fed the different SBM products, however, growth performance increased with increasing inclusion levels of SBM products.

1.4.3.2. Finishing Cattle Performance

Solvent Extracted SBM, expeller SBM, and urea were evaluated in steam flaked corn-based diets to heifers. Heavier BW was measured for expeller SBM compared to urea, whereas, solvent-extracted SBM was intermediate to expeller and urea in final BW. There were no differences in DMI and G:F, however, like final BW, ADG followed a similar pattern. Expeller SBM fed calves gained more than urea while solvent extracted SBM was intermediate (Walker et al., 2006).

1.4.4. Non-Enzymatically Browned Soybean Meal vs. Corn Byproducts

Both DDGS and NESBM are high in protein (30.79 and ~50%, respectively) and RUP (67.93 and ~72%, respectively), however the fat content of DDGS is greater than NEBSBM (10.73 and ~1.5%, respectively) due to the increased oil content of DDGS (NASEM, 2021; Amino Plus; SoyPass). Non-enzymatically browned SBM provides greater lysine content whereas DDGS offers a greater methionine content. Typically, lysine and methionine are the first limiting amino acids depending on the diet (Hussein and Berger, 1995; Wessels et al., 1997). Therefore, feeding a combination of DDGS and NESBM may enhance performance by balancing lysine and methionine in the diet, however the energy density of the diet must be similar to distinguish growth differences between amino acid balance rather than from increased energy from DDGS.

1.4.4.1. Growing Cattle Performance

Partially replacing 9% of DDGS with NESBM resulted in lighter ending BW and decreased G:F in diets consisting of 35% dry rolled corn, 28% corn silage, and 20% WCGF (Heiderscheit and Hansen, 2020). The authors attribute the heavier BW for DDGS to increased NEg in the diet. Additionally, the CP content of the NESBM diet was greater than the DDGS diet (14.6 and 13.0%, respectively) and the increased CP did not make up the difference in performance. In brome hay-based diets, two levels of WDGS (20 and 35%) were evaluated with NESBM replacing WDGS at 0, 30, and 60% in each respective WDGS level. No differences for NESBM level were detected for growth performance. A WDGS × NESBM level interaction was observed with a linear increase in DMI as NESBM replaced WDGS but only at the 35% inclusion level. Additionally, due to the decreased energy content with the NESBM replacement, a tendency for a decreased linear effect was observed at the high level of 35% WDGS for G:F (Spore et al., 2021).

The previous two studies compared two protein sources similar in nutritional content, whereas Wiseman (2021) replaced fine ground corn with NESBM to evaluate increasing MP in corn silage-based diets. Fine ground corn (included at 18% diet DM) was replaced with NESBM at the same inclusion levels as stated previously with SBM. There were linear increases in ending BW, ADG, and G:F with the increase in NESBM. Additionally, calculated RUP intake linearly increased with the increase in NESBM inclusion level.

1.4.4.2. Finishing Cattle Performance

Steers fed corn-based diets containing DDGS, included at 8.0% of the diet, and NESBM partially replaced 6.7% diet DM of the 8.0% DDGS inclusion during the 175 d finishing period. Steers fed DDGS had heavier ending BW, however, this response could be attributed to the heavier weights at the beginning of the finishing period. Although daily NE_g was greater for the DDGS treatment group and the CP content was greater for the NESBM calves, there were no differences in ADG, DMI, and G:F (Heiderscheit and Hansen, 2020).

Non-enzymatically browned SBM potentially will have a larger impact in receiving and growing cattle than finishing cattle. Increased RUP to growing cattle has been shown to improve growth performance (Wiseman, 2021). The large RUP concentration in NESBM could potentially increase performance especially compared to energy feed sources. Additionally, incorporating NESBM in corn-based diets to balance amino acids and increase the lysine availability in the small intestine may improve growing cattle performance.

1.5. Carcass Characteristics with Soybean Meal Inclusion

Young et al. (1973) reported heavier hot carcass weights (HCW) for steers fed solvent extracted SBM for the entire 112 d experiment and steers fed SBM for the first 56 d and urea for the last 56 d (SBM-none) compared to urea supplementation even through final BW was no different after the 112-d experiment. Similarly, SBM-fed steers had heavier HCW compared to urea and increased longissimus muscle area (LMA). The authors attribute the increased HCW and LMA to greater calculated dietary MP concentrations for the SBM treatment compared to urea (Milton et al., 1997). Steers fed urea, solvent extracted SBM, and expeller SBM for 84 d and urea for the remainder of the feeding period had no differences in HCW, backfat (BF), LMA, kidney, pelvic, heart (KPH) fat, yield grade, and marbling (MARB; Cosby and Stanton, 1997). Feeding ad libitum hay intake with DDGS or SBM concentrate mixes did not result in differences in dressing percentage (DP), HCW, LMA, BF, and MARB (Pittaluga et al., 2021). However, a study evaluating SBM at 10.5% diet DM compared to 20 and 40% DDGS and WDGS inclusion rates resulted in lesser 12th rib BF and lower yield grade compared to DDGS and WDGS treatments (Mateo et al., 2004). Heifers receiving ractopamine supplementation and supplemented with urea, solvent extracted, or expeller SBM did not experience differences in carcass characteristics; however, KPH was lower for solvent extracted SBM fed heifers compared to urea and expeller SBM treatment groups. Ractopamine supplementation resulted in lower KPH for expeller SBM compared to urea and solvent extracted SBM (Walker et al., 2006). Research completed on steers fed diets containing either DDGS or NESBM for 180 d did not result in any differences in carcass characteristics. The difference in d 180 final BW did not translate to increased HCW for DDGS fed steers (Heiderscheit and Hansen, 2020). In corn-based finishing diets that evaluated solvent extracted SBM, DDGS, and the combination of DDGS and SBM for 132 d, no differences in carcass characteristics were observed (Pritchard, 2010).

In studies evaluating SBM and SBM products, many researchers did not report differences in carcass characteristics compared to DDGS and urea, and none observed poorer carcass characteristics. Therefore, individuals marketing finished cattle on a dressed or grid-based system should not have reservations with cattle fed different SBM products. Ultimately, utilizing any of the SBM products in beef cattle growing or finishing rations will be based on availability and economics of local feedstuffs.

1.6. Thesis Objectives

The objectives of this thesis are to evaluate replacing DDGS with TSBM at increasing inclusion levels to analyze potential differences in growth performance, residual carcass characteristics, nutrient flow and digestibility. Furthermore, MP, N, lysine, and available lysine are focal points that are discussed in forage-based diets.

1.7. Literature Cited

AAFCO. 1997. 1997 Official Publication. Pp2666. AAFCO, Atlanta, GA.

Abe, M., T. Iriki, and M. Funaba. 1997. Lysine deficiency in postweaned calves fed corn and corn gluten meal diets. J. Anim. Sci. 75:1974-1982. doi:10.2527/1997.7571974x.

Abebe, M. 2008. History of ethanol. Journalism & Mass Communications: Student Media. 6. https://digitalcommons.unl.edu/journalismstudent/6.

American Soybean Association. 2022. Soybean processing growth is crushing it. Economist's Angle: Soybean Processing Growth Is Crushing It - American Soybean Association (soygrowers.com).

Association of Official Agricultural Chemists. 1975. Official Methods of Analysis. 12th ed. AOAC. Washington, DC.

Batterham, E. S. 1992. Availability and utilization of amino acids for growing pigs. Nutrition Research Reviews. 5:1-18. doi:10.1079/NRR19920004.

Bhargavi, G., P. Nageswara Rao, and S. Renganathan. 2018. Review of the extraction methods of crude oil from all generation biofuels in last few decades. IOP Conf. Ser.: Mater. Sci. Eng. 330 012024. doi: 10.1088/1757-899X/330/1/012024.

Borucki Castro, S. I., L. E. Philip, H. Lapierre, P. W. Jardon, and R. Berthiaume. 2007. Ruminal degradability and intestinal digestibility of protein and amino acids in treated soybean meal products. J. Dairy Sci. 90:810-822. doi: 10.3168/jds.S0022-0302(07)71565-5.

Calsamiglia, S. and M. D. Stern. 1995. A three-step in vitro procedure for estimating intestinal digestion of protein in ruminants. J. Anim. Sci. 73:1459-1465. doi:10.2527/1995.7351459x.

Coetzer, C. M. 2000. Manipulation of ruminal protein degradability. ProQuest Dissertations and Theses Global.

Cosby, N. T. and T. L. Stanton. 1997. Performance of feedlot steers supplemented with natural protein or urea during the early finishing phase. Prof. Anim. Sci. 13:124-128. doi:10.15232/S1080-7446(15)31864-7.

Demjanec, B. N. R. Merchen, J. D. Cremin, Jr., C. G. Aldrich, and L. L. Berger. 1995. Effect of roasting on site and extent of digestion of soybean meal by sheep: I. Digestion of nitrogen and amino acids. J. Anim. Sci. 73:824-834. doi: 10.2527/1995.733824x.

Decision Innovation Solutions. 2018. 2018 Soybean meal demand assessment – United States; United Soybean Board.

Firkins, J. L., L. L. Berger, and G. C. Fahey, Jr. 1985. Evaluation of wet and dry distillers grains and wet and dry corn gluten feeds for ruminants. J. Anim. Sci. 60.3:847-860. doi:10.2527/jas1985.603847x.

Heiderscheit, K. J., and S. L. Hansen. 2020. Effect of rumen-protected lysine on growth performance, carcass characteristics, and plasma amino acid profile in feedlot steers. Transl. Anim. Sci. 2020.4:1-9. doi: 10.1093/tas/txaa128.

Hodge, J. E. 1953. Chemistry of browning reactions in model systems. J. Agric. Food Chem. 1:928.

Hussein, H. S. and L. Berger. 1995. Feedlot performance and carcass characteristics of Holstein steers as affected by source of dietary protein and level of ruminally protected lysine and methionine. J. Anim. Sci. 73:3503-3509. doi:10.2527/1995.73123503x.

Hvelplund, T. and M. R. Weisbjerg. 2000. In situ techniques for the estimation of protein degradability and postrumen availability. Pages 233-258 in Forage Evaluation in Ruminant Nutrition. D. I. Givens, E. Owen, R. F. E. Axford, and H. M. Omed, ed. CABI Publishing, Wallingford, Oxfordshire, UK. doi:10.1019/97/9780851993447.0233.

Ipharraguerre, I. R., J. H. Clark, and D. E. Freeman. 2005. Rumen fermentation and intestinal supply of nutrients in dairy cows fed rumen-protected soy products. J. Dairy Sci. 88:2879-2892. doi: 10.3168/jds.S0022-0302(05)72969-6.

Lin, C. and L. Kung. 1999. Heat treated soybeans and soybean meal in ruminant nutrition. Tech. Bull. Am. Soybean Assoc. and United Soybean Board, St. Louis, MO.

Loerch, S. C., L. L. Berger, S. D. Plegge, G. C. Fahey, Jr. 1983. Digestibility and rumen escape of soybean meal, blood meal, meat and bone meal and dehydrated alfalfa nitrogen. J. Anim. Sci. 57:1037-1047. doi:10.2527/jas1983.5741037x.

Mateo, K. S., K. E. Tjardes, C. L. Wright, T. J. Tanya, and B. D. Rops. 2004. Evaluation of feeding varying levels of wet distillers grains with solubles as compared to dry distillers grains with solubles to finishing steers. South Dakota Beef Report. Paper 4. http://openprairie.sdstate.edu/sd_beefreport_2004/4.

Maxin, G. D. R. Ouellet, and H. Lapierre. 2013. Ruminal degradability of dry matter, crude protein, and amino acids in soybean meal, canola meal, corn, and wheat dried distillers grains. J. Dairy Sci. 96:5151-5160. doi:10.3168/jds.2012-6392.

Milton, C. T., R. T. Brandt, Jr., and E. C. Titgemeyer. 1997a. Effects of dietary nitrogen source and concentration in high-grain diets on finishing steer performance and nutrient digestion. J. Anim. Sci. 1997. 75:2813-2823. doi:10.2527/1997.75102813x.

Milton, C. T., R. T. Brandt, Jr., and E. C. Titgemeyer. 1997b. Urea in dry-rolled corn diets: finishing steer performance, nutrient digestion, and microbial protein production. J. Anim. Sci. 75:1415-1424. doi:10.2527/1997.7551415x.

Mueller, C. J. and D. L. Boggs. 2011. Use of soybean hulls with or without corn by-product protein sources in feedlot backgrounding diets. Prof. Anim. Sci. 27:228-234. doi:10.15232/S1080-7446(15)30478-2.

National Academies of Sciences, Engineering, and Medicine. 2016. Nutrient requirements of beef cattle: eighth revised edition. Washington, DC: The National Academies Press. doi: 10.17226/19014.

National Academies of Sciences, Engineering, and Medicine. 2021. Nutrient requirements of dairy cattle: eighth revised edition. Washington, DC: The National Academies Press. doi: 10.17226/25806.

Pittaluga, A. M., C. Clark, and A. E. Relling. 2021. Effect of protein source and nonroughage NDF content in finishing diets of feedlot cattle fed free-choice hay on growth performance and carcass characteristics. Transl. Anim. Sci. 2021.5:1-7. doi:10.1093/tas/txab224.

Plegge, S. D., L. L. Berger, and G. C. Fahey, Jr. 1982. Effect of roasting on utilization of soybean meal by ruminants. J. Anim. Sci. 55:395-401. doi:10.2527/jas1982.552395x.

Plegge, S. D., L. L. Berger, and G. C. Fahey, Jr. 1983. Performance of growing and finishing steers fed roasted soybean meal. J. Anim. Sci. 1983.57:1374-1382. doi:10.2527/jas1983.5761374x.

Plegge, S. D., L. L. Berger, and G. C. Fahey, Jr. 1985. Effect of roasting temperature on the proportion of soybean meal nitrogen escaping degradation in the rumen. J. Anim. Sci. 61:1211-1218. doi: 10.2527/jas1985.6151211x.

Powell, S., V. D. Naranjo, D. Lauzon, T. D. Bidner, L. L. Southern, and C. M. Parsons. 2011. Evaluation of an expeller-extruded soybean meal for broilers. J. Appl. Poult. Res. 20:353-360. doi:10.3382/japr.2010-00305.

Pritchard, R. H., 2010. Use of de-oiled dried distillers grains as a source of crude protein in finishing cattle diets. South Dakota Beef Report. Paper 3. http://openprairie.sdstate.edu/sd_beefreport_2010/3.

Pritchard, R. H. and D. L. Boggs. 2006. Use of DDGS as the primary source of supplemental crude protein in calf receiving diets. South Dakota Beef Report. Paper 3. http://openprairie.sdstate.edu/sd_beefreport_2006/3.

Ruiz, N., C. M. Parsons, H. H. Stein, C. N. Coon, J. E. van Eys, and R. D. Miles. 2020. A review: 100 years of soybean meal. A historical look at the soybean and its use for animal feed. Feedstuffs. https://www.feedstuffs.com/news/review-100-years-soybean-meal.

Schoonmaker, J. P., M. C. Claeys, and R. P. Lemenager. 2013. Effect of increasing distillers grains inclusion on performance and carcass characteristics of early-weaned steers. J. Anim. Sci. 2013.91:1784-1790. doi:10.2527/jas2011-5075.

Shaver, R. n.d. Soy protein alternatives for lactating dairy cows. https://shaverlab.dysci.wisc.edu/wp-content/uploads/sites/204/2015/04/soyprotein.pdf

Spore, T. J., Z. E. Carlson, K. H. Wilke, G. E. Erickson, T. J. Klopfenstein, J. C. MacDonald, and A. K. Watson. 2021. Effects of replacing wet distillers grains with supplemental SoyPass in forage-based growing cattle diets. Transl. Anim. Sci. 2021.5:1-10. doi:10.93/tas/txab117.

Thomas, E. E., C. R. Mason, and S. P. Schmidt. 1984. Relation of feedlot performance and certain physiological responses to the metabolizable protein and urea content of cattle diets. J. Anim. Sci. 58:1285-1291. doi:10.2527/jas1984.5851285x.

Titgemeyer, E. C., N. R. Merchen, and L. L. Berger. 1988. Estimation of lysine and methionine requirements of growing steers fed corn silage-based or corn-based diets. J. Dairy Sci. 71:421-434. doi: 10.3168/jds.S0022-0302(88)79572-7.

United States Environmental Protection Agency, 2024. Renewable Fuel Standard Program. Renewable Fuel Standard Program | US EPA.

Walker, D. K., E. C. Titgemeyer, J. S. Drouillard, E. R. Loe, B. E. Depenbusch, and A. S. Webb. 2006. Effects of ractopamine and protein source on growth performance and carcass characteristics of feedlot heifers. J. Anim. Sci. 2006. 84:2795-2800. doi:10.2527/jas.2005-614.

Waller, J. T. Klopfenstein, and M. Poos. 1980. Distillers feeds as protein sources for growing ruminants. J. Anim. Sci. 51:1154-1167. doi:10.2527/jas1980.5151154x.

Wessels, R. H., E. C. Titgemeyer, G. St. Jean. 1997. Effect of amino acid supplementation on whole-body protein turnover in Holstein steers. doi:10.2527/1997.75113066x.

Wiseman, A. R. 2021. Evaluation of novel feed ingredients on finishing and growing cattle performance and intake modeling for growing cattle. ProQuest Dissertations and Theses Global.

Young, A. W., J. A. Boling, and N. W. Bradley. 1973. Performance and plasma amino acids of steers fed soybean meal, urea, or no supplemental nitrogen in finishing rations. J. Anim. Sci. 36:803-808. doi:10.2527/jas1973.364803x.

Zinn, R. A., and F. N. Owens. 1983. Site of protein digestion in steers: Predictability. J. Anim. Sci. 56:707-716. doi:10.2527/jas1983.563707x.

2. CHAPTER 2: THE EFFECTS OF REPLACING DRIED DISTILLERS GRAINS WITH SOLUBLES WITH HEAT-TREATED SOYBEAN MEAL IN FORAGE-BASED DIETS ON STEER PERFORMANCE, NUTRIENT FLOW, AND DIGESTIBILITY

2.1. Abstract

The objectives of the study were to evaluate increasing metabolizable protein (MP) and lysine concentrations on growth performance, nutrient flow, and digestibility. Experiment 1: 70 Angus-based steers (initial BW = 298 ± 16 kg) were utilized in a generalized randomized block design for an 85-d growing study at the NDSU Beef Cattle Research Complex. Steers were stratified into three weight blocks. Experiment 2: five ruminally, duodenally, and ileally fistulated Jersey steers were utilized in a 4 x 5 Latin square design to measure nutrient digestibility and flow at the NDSU Animal Nutrition and Physiology Center. Heat-treated soybean meal (TSBM) replaced dried distillers grains plus solubles (DDGS) in the diet at 0 (TSBM0), 4 (TSBM4), 8 (TSBM8), and 12% (TSBM12) of the diet DM. Diets were modeled to supply excess MP, whereas lysine was predicted to be sufficient only in the 12% treatment. In experiment 1: individual intakes were measured daily using an automated feed system. After experiment 1, steers were commingled into a single pen, finished on a common diet for 150 d, and shipped to a commercial abattoir to collect carcass characteristics. Data were analyzed using the MIXED procedure of SAS with steer as the experimental unit and fixed effects of treatment and block for experiment 1 and individual animal within period with the fixed effect of treatment and period and random effect of animal for experiment 2. In the growing experiment, dietary treatment did not influence ($P \geq 0.27$) ending BW, average daily gain, dry matter intake, and gain:feed. However, on d 85, plasma urea nitrogen increased linearly ($P = 0.01$) with TSBM suggesting increased N intake and ammonia concentration in the rumen. Residual carcass

characteristics from experiment 1 were not different ($P \geq 0.16$) for hot-carcass weight, longissimus muscle area, backfat thickness, marbling, or calculated yield grade. In experiment 2, post-ruminal digestibility of nitrogen, total lysine, and available lysine decreased linearly ($P = 0.01$). Therefore, the Maillard reaction potentially decreased nutrient availability in the intestines. In conclusion, growth performance was not improved when replacing a proportion of DDGS with TSBM in growing cattle diets even when supply of RUP and lysine was increased. Intestinal digestibility may be impaired with the inclusion of TSBM products in growing cattle diets.

2.2. Introduction

Dried distillers grains plus solubles (DDGS) is a common feedstuff for producers in the Midwest due to its high concentrations of protein (31% CP, 68% RUP; NASEM, 2016) and energy, availability, and competitive price. Corn-based diets are typically limiting in the essential amino acid (AA) lysine (NASEM, 2016). It has been hypothesized that supplementing soybean meal (SBM) could satisfy lysine requirements for growing cattle (Merchen and Titgemeyer, 1992). The United States Renewable Fuel Standard Program is driving the expansion of biodiesel production from oilseeds through governmental incentives and requirements (United States Environmental Protection Agency, 2024). Therefore, soybean crushing plants are being built in North Dakota and surrounding states to keep pace with the increasing demand. The crushing process extracts soybean oil and produces feed byproducts such as SBM and soybean hulls. Soybean meal is high in protein (53% CP, 29% RUP; NASEM, 2016), is highly rumen degradable (Titgemeyer et al., 1989), and contains greater concentrations of lysine than other feed ingredients, including corn grain or corn DGS. The high ruminal degradability of SBM limits the flow of dietary AA, specifically lysine, to the small intestine (Borucki Castro et al., 2007). High ruminal degradability of dietary proteins can be problematic during high production or rapid growth periods, as microbial protein alone may not meet amino acid requirements (Orskov et al., 1970; Chalupa et al., 1974; Clark et al., 1987). Therefore, incorporating rumen bypass protein into growing cattle diets has been shown to improve gain:feed (G:F; Zinn and Owens, 1993).

An additional process to conventional SBM, non-enzymatic browning, creates a product with reduced ruminal degradation and can result in improved growth performance as compared to SBM (Coetzer, 2000). Non-enzymatic browning utilizes conventional solvent-extracted SBM and a reducing sugar to initiate a Maillard reaction during the desolventizing-toasting step to create heat-treated soybean meal (TSBM; Cleale et al., 1987). Heat-treated soybean meal increases dietary escape protein (Cleale et al., 1987) and amino acid flow to the small intestine (Borucki Castro et al., 2007). The increased contribution of rumen undegradable protein (RUP) to the small intestine can result in greater metabolizable protein (MP) availability to the animal (NASEM, 2016). Supplying additional lysine in beef cattle diets is well-researched through rumen-protected lysine products and has been shown to linearly increase G:F as the inclusion rate increases (Veloso et al., 2016; Montano et al., 2019), whereas less is understood on the effects of including TSBM in beef cattle diets. Spore et al. (2021) studied replacing wet distillers grains included at 20 and 35% of the diet (dry matter basis [DM]) with TSBM at increased rates (0, 30, and 60% DM, respectively) in forage-based growing beef cattle diets and observed decreased G:F. The authors attributed this effect to the decreased energy content of TSBM compared to wet distillers grains and not changes in intestinal lysine supply.

The current study evaluated the effects of increasing concentrations of lysine and metabolizable protein by feeding heat-treated soybean meal in forage-based growing cattle diets on 1) growth performance and 2) nutrient flow and site of digestion. The hypothesis was as TSBM replaces more DDGS, growth performance will improve due to increased metabolizable protein and intestinal supply of essential amino acids. Additionally, replacing TSBM with DDGS at increasing levels will increase lysine and nitrogen availability and digestibility. The objectives of the study were to evaluate replacing DDGS with TSBM at increasing levels and the subsequent effects on metabolizable protein, and nitrogen and lysine availability in forage-based diets.

2.3. Materials and Methods

The procedures used in the experiment were approved by the North Dakota State University Institutional Animal Care and Use Committee (protocol #20220036 and #20220078) which comply with the Guide for the Care and Use of Agricultural Animals in Research and Teaching.

2.3.1. Experiment 1: Cattle Performance

Seventy Angus-based steers (initial BW = 298 ± 16 kg) were utilized in an 85-d generalized randomized block design with animal being the experimental unit. All steers were sourced from the Central Grasslands Research and Extension Center near Streeter, North Dakota and transported to the North Dakota State University Beef Cattle Research Complex in Fargo, North Dakota for the study. Upon arrival, steers were provided ad libitum access to water and feed consisting of corn silage, oat hay, and a dry meal supplement in a monoslope barn with drylot access. A two-week training period acclimated steers to the facility and to the automated feed intake monitoring system (Insentec Roughage Intake Control, Hokofarm B. V., Marknesse, The Netherlands). During the two-week training period, six steers were removed due to not acclimating to the automated feed intake monitoring system. Throughout the experiment, 16 d of individual intake data were removed from analysis due to frigid cold weather hindering the effectiveness of the automated feed intake monitoring system. When the system was shut down, all steers received a common diet of corn silage, oat hay, and a dry meal supplement.

Dietary treatments of TSBM (AminoPlus, Ag Processing Inc., Omaha, NE) replaced DDGS at levels of 0 (TSBM0), 4 (TSBM4), 8 (TSBM8), and 12% of diet DM (TSBM12; Table 1). Diets also contained 44% corn silage, 37% oat hay, and 3% dry meal supplement (DM basis). The empirical solutions model of the Beef Cattle Nutrient Requirements Model (version 1.0.37.15; NASEM, 2016) was used to formulate for increased metabolizable protein and lysine supply as the inclusion of TSBM increased in the diet. Lysine requirements in the formulated diets were predicted to be deficient for TSBM0, TSBM4, and TSBM8 treatments (-6.23, -3.09, and -0.24 g/d, respectively) and in excess for TSBM12 treatments (2.80 g/d). Whereas, the metabolizable protein requirement was predicted to be adequate for all treatments (40.5, 68.9, 89.8, and 117.5 g/d, respectively). Metabolizable energy was predicted to be limiting gain for all four treatments.

Table 1. Experimental diets and nutrient composition of experiment 1: Cattle Performance

Item	TSBM0	TSBM4	TSBM8	TSBM12
Ingredient, % DM				
Corn silage	44	44	44	44
Oat hay	37	37	37	37
DDGS[1]	16	12	8	4
TSBM[2]	0	4	8	12
Supplement[3]	3	3	3	3
Nutrient Composition				
DM, % As-fed	46.9	46.9	46.9	46.9
CP, % DM	12.6	13.1	13.6	14.1
Fat, % DM	2.6	2.4	2.3	2.1
NDF, % DM	52.2	51.3	50.5	49.6
ADF, % DM	29.7	29.4	29.1	28.9
Starch, % DM	17.0	16.9	16.8	16.7
Metabolizable protein balance[4], g/d	40.5	68.9	89.8	117.5
Lysine balance[4], g/d	-6.23	-3.09	-0.24	2.80
NEg, Mcal/kg[5]	0.95	1.01	0.98	1.02

[1] Dried distillers grains with solubles.

[2] Heat-treated soybean meal (AminoPlus, Ag Processing Inc., Omaha, NE).

[3] Supplement formulated to provide 22.9 g/ton monensin (Rumensin 90, Elanco Animal Health). Supplement contained 1.62% fine ground corn, 1.00% limestone, 0.30% salt, 0.05% beef trace mineral, 1.50 IU of vitamin A, 0.27 IU of vitamin D, 0.03 IU of vitamin E per kilogram on a dry matter basis.

[4] Calculated utilizing the empirical solutions model of the Beef Cattle Nutrient Requirements Model 2016 (version 1.0.37.15; NASEM, 2016).

[5] Calculated using the NRC, 1996 equations.

Prior to the initiation of the study, steers were limit-fed a common diet containing 40% corn silage, 57% oat hay, and 3% dry meal supplement (DM basis) at 1.8% BW for five days to minimize gut fill variation followed by three days of weighing (Watson et al., 2013). The average of the consecutive 3-d weights served as the initial BW. Steers were blocked by initial body weight (BW) into light (initial BW = 280 ± 6 kg), medium (initial BW = 296 ± 4 kg), and heavy (initial BW = 317 ± 8 kg) blocks and assigned randomly to treatments. After completion of the study, steers were limit-fed and the 3-d weighing process was repeated to measure ending BW. On d 0, steers were implanted with 80 mg of trenbolone acetate and 16 mg of estradiol (Revalor-IS, Merck Animal Health, Summit, NJ). Steers received 22 mL of Pyrethroid (Clean-

Up II, Elanco Animal Health, Greenfield, IN) on d 56 for the control and prevention of lice. Body weights were determined every 28 d to monitor interim performance.

After the study was completed, steers were transported to the NDSU Central Grasslands Research and Extension Center. For 150 d, steers received a common diet of corn, DDGS, and liquid supplement. Corn silage was utilized for the first approximately 121 d and then replaced with hay for the remainder of the feeding period. Steers were transported to a commercial abattoir to collect hot-carcass weight (HCW), longissimus muscle area (LMA), backfat thickness (BF), marbling score (MARB), quality grade (QG), and yield grade (YG).

Blood was collected every 28 d via jugular venipuncture with an 18-gauge x 3.8-cm needle (Greiner Bio-One, Monroe, North Carolina) and into two vacuum tubes (10 mL/tube). One tube contained lithium heparin (plasma) and one tube contained a clot activator (serum; BD Vacutainer, Becton, Dickinson and Company, Franklin Lakes, NJ). Blood was centrifuged at 3,000 x g for 10 minutes at 4˚C. Plasma and serum samples were then stored at -20˚C. Plasma samples were analyzed for glucose, plasma urea nitrogen (PUN), and non-esterified fatty acids (NEFA). Glucose was analyzed using the Infinity Glucose Hexokinase Liquid Stable Reagent (Fisher Diagnostics, Middletown, VA). The QuantiChrom Urea Assay Kit (BioAssay Systems, Hayward, CA) was used to determine BUN and NEFA samples were analyzed utilizing Fujifilm HR Series NEFA-HR 2 (Fujifilm Healthcare Americas Corporation, Lexington, MA).

Dietary DM was determined weekly by sampling ingredients and oven-drying at 60˚C for 48 h to adjust diets to supply the correct amounts of each feed ingredient in the mixed diet. Weekly corn silage, DDGS, TSBM, and supplement samples were freeze-dried (SP VirTis Genesis Pilot Freeze Dryer, ATS Automation Tooling Systems, Inc., Warminster, PA). Oat hay weekly samples were oven-dried at 60˚ C for 48 h. After grinding through a 1-mm screen using a

Wiley mill (Thomas Scientific, Swedesboro, NJ), all samples were composited into 4-week intervals on an equal weight basis. Composited ingredient samples were analyzed for DM (AOAC, 2010; method 934.01), organic matter (OM; AOAC, 2010; method 942.05), crude protein (CP; LECO Corporation, St. Joseph, MI), neutral detergent (NDF) and acid detergent fiber (ADF; Ankom Technology, Macedon, NY, 1998), ether extract (EE; AOAC, 2010; method 920.39), starch (Herrera-Saldana and Huber, 1989), calcium (Ca; AOAC, 2010; method 968.08), and phosphorus (P; AOAC, 2010; method 965.17).

2.3.1.1. Statistical Analysis

Data were analyzed utilizing the MIXED procedure of SAS (SAS 9.4, SAS Inst. Inc., Cary, NC). Growth performance and feed intake were analyzed with treatment (n = 4) and block (weight group; n = 3) as fixed effects. Data were evaluated and considered significant at $P \leq 0.05$. Orthogonal contrasts utilized were DDGS vs. TSBM (TSBM0 vs. TSBM4, TSBM8, TSBM12), and linear and quadratic effects of TSBM inclusion. Model residual plots were utilized to ensure mixed procedure assumptions were met, and necessary outliers were removed. Two steers were identified as having data for all traits that were outliers compared to other steers (one from TSBM4 and one from TSBM8) and, therefore, were removed from the dataset.

2.3.2. Experiment 2: Nutrient Flow and Digestibility

Five fistulated Jersey steers (initial BW 288 ± 22 kg) fitted with ruminal, duodenal, and ileal cannulas were utilized in a 4 × 5 Latin square design at the Animal Nutrition and Physiology Center in Fargo, North Dakota. The 4-period study, totaling 56 d, consisted of 9 d of adaptation and 5 d of collection (d 0 to 4) per period to determine nutrient flow and total tract digestibility when partially replacing DDGS with TSBM in forage-based growing cattle diets.

Dietary treatments are the same as described in Experiment 1. Diets also contained 43.5% corn silage, 36.5% grass hay, and 4% dry meal supplement (DM basis; Table 2). Chromic oxide was incorporated into the diet daily at 0.25% DM as a digesta marker. Diets were mixed several times weekly and stored in a cooler at 7˚C to maintain freshness.

Table 2. Experimental diets and nutrient composition of experiment 2: Nutrient Flow and Digestibility

Item	TSBM0	TSBM4	TSBM8	TSBM12
Ingredient, % DM				
Corn silage	43.5	43.5	43.5	43.5
Grass hay	36.5	36.5	36.5	36.5
DDGS[1]	16.0	12.0	8.0	4.0
TSBM[2]	0.0	4.0	8.0	12.0
Supplement[3]	3.75	3.75	3.75	3.75
Chromic oxide	0.25	0.25	0.25	0.25
Nutrient Composition				
DM, % As-fed	42.5	42.4	42.5	42.3
CP, % DM	16.0	16.6	17.1	17.7
Fat, % DM	1.6	1.5	1.4	1.3
NDF, % DM	53.3	52.6	51.8	51.1
ADF, % DM	27.5	27.1	26.7	26.3
Starch, % DM	10.4	10.3	10.2	10.2
Lysine, % DM	0.55	0.63	0.70	0.78
Available lysine[4], % DM	0.48	0.55	0.63	0.70

[1] Dried distillers grains with solubles.

[2] Heat-treated soybean meal (AminoPlus, Ag Processing Inc., Omaha, NE).

[3] Supplement formulated to provide 22.9 g/ton monensin (Rumensin 90, Elanco Animal Health). Supplement contained 1.62% fine ground corn, 1.00% limestone, 0.30% salt, 0.05% beef trace mineral, 1.50 IU of vitamin A, 0.27 IU of vitamin D, 0.03 IU of vitamin E per kilogram on a dry matter basis.

[4] Analyzed utilizing the 1-Fluoro-2,4-dinitrobenzene reaction at the University of Missouri Agricultural Experiment Station Chemistry Laboratory.

2.3.2.1. Sample Collections

Diets were fed at 0800 daily. Orts were weighed daily for tracking daily individual intake. During the collection period, orts were weighed and subsampled at 0730 h and frozen at -20˚C. Orts were composited and analyzed within each period.

Each steer was fitted with a fecal collection bag during the collection period. Feces were weighed at 0700 and 1900. The sample was mixed by hand to achieve a homogenous mixture and 2% was subsampled on a wet basis. Subsamples were composited for each period and frozen at -20˚C until analysis.

Co-EDTA was prepared utilizing the method described by Uden et al. (1980) and dosed into the rumen at 0600 h on d 0. Rumen fluid was collected (~120 mL) via the rumen cannula with a metal strainer equipped with a hand suction pump at 0600 (before Co-EDTA administered), 0800 (diets delivered), 1000, 1200, 1400, 1600, and 1800 h timepoints. After each timepoint, samples were frozen at -20˚C for ammonia (Broderick and Kang, 1980) and Co-EDTA concentration analysis (Uden et al., 1980).

Twelve duodenal and ileal digesta sampling timepoints were used across three days to represent every other hour in a 24 h cycle: d 1 – 0300, 0900, 1500, 2100; d 2 – 0100, 0700, 1300, 1900; d 3 – 0500, 1100, 1700, 2300. Approximately 200 mL of chyme was collected at each timepoint and stored in a -20˚C freezer. Samples were thawed and composited (on an equal weight basis) for each animal and period.

Rumen contents were collected (4 kg) at 1400 h on d 4 via the rumen cannula. Samples were collected from multiple locations in the rumen beneath the rumen mat. Contents were mixed with 1 L of 0.9% NaCl solution, blended for 5 minutes (Ninja Blender, Needham, Massachusetts, USA), and strained through 4 layers of cheesecloth. The sample was frozen at -20˚C until bacterial isolation.

2.3.2.2. Laboratory Analysis

Dietary DM was determined weekly through sampling ingredients and oven-drying at 60˚ C for 48 h (Grieve SB-350, The Grieve Corporation, Round Lake, IL, USA). Ingredient samples were collected before each mixing session during the collection period for diet analysis. All ingredients, fecal, duodenal and ileal samples were freeze-dried (SP VirTis Genesis Pilot Freeze

Dryer, ATS Automation Tooling Systems, Inc., Warminster, PA). Ort samples were oven-dried at 60˚C for 48 h. Ingredient, ort, and fecal samples were ground through a 1-mm screen utilizing a Wiley mill (Thomas Scientific, Swedesboro, NJ). After ingredient grinding, the weighted average for days associated per mixing session were composited by period. Duodenal, ileal, and bacterial isolate samples were ground utilizing a mortar and pestle until a uniform consistency was obtained.

Bacterial isolate samples were centrifuged at 500 x g for 20 minutes to remove protozoa and feed particles. The particle free supernatant was centrifuged at 30,000 x g for 20 minutes for bacteria collection. Isolated bacteria were frozen at -20˚C prior to freeze drying (SP VirTis Genesis Pilot Freeze Dryer, ATS Automation Tooling Systems, Inc., Warminster, PA) and analysis. Rumen bacterial isolates were analyzed for DM (AOAC, 2010; method 934.01), OM (AOAC, 2010; method 942.05), CP (LECO Corporation, St. Joseph, MI), lysine availability (AOAC, 2006; method 975.44), amino acid concentrations (AOAC, 2006; 982.20), and purines (Zinn and Owens, 1986).

Ingredients, orts, feces, duodenal, and ileal samples were analyzed for DM (AOAC, 2010; method 934.01), OM (AOAC, 2010; method 942.05), CP (LECO Corporation, St. Joseph, MI), NDF and ADF (Ankom Technology, Macedon, NY, 1998), starch (Herrera-Saldana and Huber, 1989), and chromic oxide (Fenton and Fenton, 1979). The chromic oxide recovery averaged 65.7% and varied from 44 to 95%. Therefore, recovered chromic oxide from the feces was assumed to equal chromic oxide intake for calculating nutrient flow and disappearance. Ingredient, duodenal and ileal digesta, and fecal samples for amino acid content (AOAC, 2006; 982.20), and lysine availability (AOAC, 2006; method 975.44) were analyzed at the University of Missouri Agricultural Experiment Station Chemistry Laboratory. Feed ingredients, orts, and feces were also analyzed for EE (AOAC, 2010; method 920.39). Calcium (AOAC, 2010; method 968.08) and phosphorus (AOAC, 2010; method 965.17) were analyzed on feed ingredients.

To analyze for ruminal degradation and digestible RUP, the DDGS and TSBM were subjected to a modified three-step in vitro procedure using a 12-h incubation (Gargallo et al., 2006) to analyze for ruminal degradation and digestible RUP. Samples from the in situ and in vitro (Daisy ^{II} Incubator, Ankom Technology, Macedon, NY) portions of the procedure were analyzed for DM (AOAC, 2010; method 934.01), OM (AOAC, 2010; method 942.05) and CP (LECO Corporation, St. Joseph, MI).

2.3.2.3. Statistical Analysis

Data were analyzed using the MIXED procedure of SAS (SAS 9.4, SAS Inst. Inc., Cary, NC). Nutrient digestibility and intake with treatment (n=4) and period (n=4) as fixed effects and steer (n=5) as a random effect. The experimental unit is steer within period. Data were considered significant at $P \leq 0.05$ for treatment, DDGS vs. TSBM, linear and quadratic orthogonal contrasts.

2.4. Results and Discussion

2.4.1. Experiment 1: Cattle Performance

Increasing metabolizable protein and lysine concentrations through TSBM supplementation did not affect ($P \geq 0.27$) ending body weight (EBW), average daily gain (ADG), dry matter intake (DMI), and gain:feed (G:F; Table 3). Similar to our results, no differences in ending BW, ADG, DMI, and G:F were observed in brome hay-based diets when replacing wet distillers grains with solubles (WDGS) with TSBM at increased inclusion rates (Spore et al., 2021). In contrast, Heiderscheit and Hansen (2020) utilized dry rolled corn-based diets for a 56-d growing study and reported heavier EBW and increased G:F for steers offered only DDGS compared to a diet that replaced DDGS with TSBM. These steers then transitioned onto corn-based finishing diets for 124 d to evaluate DDGS and DDGS replaced with TSBM. No differences in ADG, DMI, and G:F were reported, however, final BW was different due to the heavier EBW in the growing period (Heiderscheit and Hanson, 2020). Like the current study, Spore et al. (2021) and Heiderscheit and Hansen (2020) reported increased dietary CP concentrations when TSBM replaced DDGS. As growth performance did not improve with TSBM replacing DDGS, metabolizable energy limiting gain may impact growth performance greater than the CP concentration. Our nutrient analysis found DDGS to contain 5.38% fat compared to TSBM at 0.93%, therefore, explaining the decrease in fat content as a potential source for differences in the energy content in treatment diets.

Table 3. Experiment 1: growth performance of growing cattle on forage-based diets with heat-treated soybean meal

	Treatments[1]					P - value			
Item	TSBM0	TSBM4	TSBM8	TSBM12	SEM	TRT	DDGS vs. TSBM	Linear	Quadratic
Steers, n	16	18	17	17	--	--	--	--	--
Initial BW, kg	298	298	297	298	1.6	0.99	0.95	0.95	0.95
Ending BW, kg	378	384	380	384	4.3	0.68	0.39	0.52	0.84
ADG[2], kg/d	0.95	1.02	0.96	1.01	0.046	0.57	0.31	0.45	0.80
DMI[3], kg/d	7.7	7.6	7.6	7.5	0.17	0.90	0.59	0.46	0.92
Gain:Feed	0.123	0.134	0.127	0.135	0.005	0.27	0.14	0.19	0.84

[1] Dietary percent of heat-treated soybean meal (TSBM) replacing a proportion of 16% dried distillers grains plus solubles in the diet; TSBM0: 0% heat-treated soybean meal, TSBM4: 4% heat-treated soybean meal, TSBM8: 8% heat-treated soybean meal, TSBM12: 12% heat-treated soybean meal.

[2] Average daily gain.

[3] Dry matter intake.

Furthermore, the previous two studies replaced DDGS with TSBM, two known protein sources. When replacing fine ground corn with TSBM at increasing rates in corn-silage growing based diets, linear increases in ending BW, ADG, G:F, and RUP were reported (Wiseman, 2021). Additionally, replacing a RDP supplement (primarily fine ground corn) with a RUP supplement (combination of TSBM and concentrated corn gluten meal) at increasing levels in corn silage-based growing diets for 83 d, linearly increased EBW, ADG, and G:F (Oney et al., 2019). The increasing MP balances from deficient to in excess suggests increasing RUP concentrations should improve growth performance.

In the present study, MP was modeled to be in excess for all four treatments, as well as increase with increasing inclusion of TSBM due to TSBM containing greater RUP content. A plateau in daily gain is anticipated when the MP requirement is attained (Wilkerson et al., 1993). As there were no differences in growth performance in the present study, the additional MP did not improve performance and was likely plateaued. However, as TSBM replaced DDGS, the modeled NEg of the diets decreased, which would suggest decreased ADG due to energy limiting gain in all treatment diets. Even though the modeled NEg was decreasing, growth performance was not different among treatments and could be attributed to excess MP being utilized as energy. Therefore, potentially improving ADG greater than expected for TSBM supplemented steers.

Similar to MP, once the lysine requirement is met and no longer limiting, overfeeding will not improve growth performance (Klemesrud et al., 2000a; Klemesrud et al., 2000b). In the current study, dietary lysine increased as TSBM replaced DDGS in all four treatments. However, no differences in performance were experienced when the predicted lysine balance increased from deficient to excess. Klemesrud et al. (2000b) reported 2.56 g/d of supplemental lysine optimized growth performance when evaluating 9 treatments that ranged from 0 to 12 g/d of rumen-protected lysine in corn-based finishing diets. Ending BW, ADG, and G:F linearly increased with two levels of rumen-protected lysine compared to the control in steam-flaked corn based diets fed to Holstein calves (Montano et al., 2019). Additionally, the control and low inclusion of rumen-protected lysine treatment were predicted to be deficient whereas the high inclusion of rumen-protected lysine treatment was in excess for the lysine requirement. Several studies have evaluated increased dietary lysine flowing to the duodenum and reported improved growth performance when using various rumen protected lysine products (Klemesrud et al., 2000b; Veloso et al., 2016; Montano et al., 2019). Increasing dietary lysine that escapes ruminal degradation should exhibit improved growth performance in growing cattle if metabolizable lysine limits growth. Therefore, the source of dietary lysine may dictate its effectiveness on improving growth performance. The Maillard reaction used to increase RUP and lysine content of TSBM may also be detrimental to the bioavailability of the lysine. Awawdeh et al. (2007) used a chick growth assay to evaluate lysine availability and reported a decrease in lysine availability with a treated SBM product due to the reactions from the Maillard reaction with the epsilon amino group of lysine. There is limited research regarding Maillard reaction on SBM products and the subsequent performance and digestion effects on ruminants.

Due to the increasing lysine concentrations from deficient to in excess from TSBM0 to TSBM12, respectively, increased growth performance was assumed to occur with the substitution of TSBM because the predicted lysine requirement would be met. Growth performance was not different across treatments therefore concluding: 1) lysine was not the first limiting amino acid, 2) the lysine requirement was met for all treatments or 3) the lysine in the TSBM was not bioavailable to the animal due to excessive heating (Awawdeh, et al., 2007).

2.4.1.1. Blood Parameters

There were no treatment differences (*P* > 0.31) for d 0 glucose, NEFA, PUN concentrations (data not presented) and d 85 glucose and NEFA (Table 4). Plasma urea nitrogen was influenced (*P* < 0.01) by TSBM inclusion in the diet and linearly increased with the increased replacement rate of DDGS on d 85. Differences in PUN are largely attributed to the increase in dietary CP with increasing dietary TSBM, which has been documented by others (Vasconcelos et al., 2009; Jennings et al., 2018). Furthermore, TSBM0 and TSBM4 (7.60 and 8.41 µmol/L, respectively) are below the concentration (approximately 8.5 µmol/L PUN) that is correlated with a plateau in rumen ammonia concentration (Vercoe, 1969; Reynolds and Kristensen, 2008). Therefore, ammonia concentration potentially exceeded ruminal microbial requirements (Satter and Slyter, 1974) for TSBM8 and TSBM12 and was converted to urea in the liver. Increased nitrogen excretion has been shown to occur at a PUN concentration of approximately 7 µmol/L in feedlot steers (Preston et al., 1965; Preston et al., 1978; Hammond, 1983). All treatments with TSBM resulted in steers having greater than approximately 7 µmol/L and potentially resulted in greater amounts of urea excretion. Additionally, energy was predicted to be limiting growth in all treatment diets and decreased slightly with increased TSBM inclusion; therefore, the separation between nitrogen and energy concentrations in the rumen increased when the CP content increased with each treatment diet (Hammond, 1996). In a similar study, a linear increase in PUN was reported when replacing TSBM with WDGS at similar inclusion rates to the current study (Spore et al., 2021). Additionally, PUN concentrations increased from the low WDGS level to the high WDGS level due to the increase in CP concentration in the diets. Heiderscheit and Hansen (2020) reported at the end of a 179-d finishing period, TSBM-fed calves had greater PUN concentration than the DDGS group and could be attributed to the greater CP concentration in the TSBM diet.

Table 4. Blood characteristics of growing cattle on forage-based diets with heat-treated soybean meal on day 85

	Treatments[1]					*P* - value			
Item	TSBM0	TSBM4	TSBM8	TSBM12	SEM	TRT	DDGS vs. TSBM	Linear	Quadratic
Steers, n	16	18	17	17	--	--	--	--	
Glucose, mmol/L	4.71	4.60	4.81	4.70	0.080	0.33	0.91	0.65	0.99
PUN[2], mmol/L	7.60	8.41	9.77	10.68	0.370	0.01	0.01	0.01	0.89
NEFA[3], µmol/L	538	535	560	474	44.3	0.53	0.77	0.39	0.34

[1] Dietary percent of heat-treated soybean meal (TSBM) replacing a proportion of 16% dried distillers grains plus solubles in the diet; TSBM0: 0% heat-treated soybean meal, TSBM4: 4% heat-treated soybean meal, TSBM8: 8% heat-treated soybean meal, TSBM12: 12% heat-treated soybean meal.

[2] Plasma urea nitrogen.

[3] Non-esterfied fatty acid.

2.4.1.2. Carcass Characteristics

There were no differences ($P \geq 0.16$) in final body weight (FBW), hot-carcass weight (HCW), longissimus muscle area (LMA), backfat thickness (BF), marbling score (MARB), and calculated yield grade (YG) on residual effects during the growing phase (Table 5). Similarly, Heiderscheit and Hansen (2020) concluded there was no difference between feeding DDGS control and replacing DDGS with TSBM during the growing and finishing phase of steers fed corn-based diets. No differences in carcass characteristics were observed with increasing levels of metabolizable protein when comparing steam-flaked corn-based diets with a control, high inclusion rate of rumen degradable protein (SBM), and high inclusion rate of RUP (corn gluten meal; Samuelson et al., 2023). There is very little carcass data related to TSBM and more studies need to be completed to evaluate any potential effects.

Table 5. Carcass characteristics of growing cattle on forage-based diets with heat-treated soybean meal

	Treatments[1]					*P* - value			
Item	TSBM0	TSBM4	TSBM8	TSBM12	SEM	TRT	DDGS vs. TSBM	Linear	Quadratic
Steers, n	16	18	17	17	--	--	--	--	
HCW[2], kg	362	368	362	372	6.2	0.62	0.49	0.44	0.76
LMA[3], cm^2	82.1	81.4	83.9	84.5	1.69	0.50	0.57	0.21	0.69
Backfat, cm	1.29	1.34	1.17	1.26	0.08	0.47	0.73	0.46	0.82
Marbling[4]	414	413	388	414	14.7	0.49	0.59	0.25	0.99
Yield grade[5]	3.23	3.37	3.02	3.17	0.11	0.16	0.73	0.28	0.99

[1] Dietary percent of heat-treated soybean meal (TSBM) replacing a proportion of 16% dried distillers grains plus solubles in the diet; TSBM0: 0% heat-treated soybean meal, TSBM4: 4% heat-treated soybean meal, TSBM8: 8% heat-treated soybean meal, TSBM12: 12% heat-treated soybean meal.

[2] Hot carcass weight.

[3] Longissimus muscle area.

[4] Marbling score scale: 300 = select; 400 = low choice; 500 = average choice.

[5] USDA, 2017

2.4.2. Experiment 2: Nutrient Flow and Digestibility

2.4.2.1. Nutrient Intake and Flow

There were no differences ($P \geq 0.14$) in DM, OM, N, NDF, ADF, and starch intake across treatments. However, treatments were different ($P = 0.01$) in total lysine intake due to the increased lysine concentration of TSBM compared to DDGS (3.10 and 1.18%, respectively; Table 6). The difference in total lysine intake between TSBM and DDGS resulted in linear effects of TSBM ($P = 0.01$) and DDGS vs. TSBM inclusion ($P = 0.01$) differences. All feed ingredients were analyzed for available lysine concentration using the 1-Fluoro-2,4-dinitrobenzene reaction (AOAC, 2006; method 975.44), therefore, the available lysine for DDGS and TSBM are 90.7% and 94.8%, respectively, of the total lysine value. Similar to total lysine, available lysine detected treatment differences ($P = 0.01$) that observed linear ($P = 0.01$) and DDGS vs. TSBM inclusion ($P = 0.01$) effects. An amino acid analysis completed by Borucki Castro (2007), reported a lysine value of 2.89% (% of DM) on the same TSBM product utilized in the present study. Available lysine is the portion of lysine that is capable of being digested and absorbed for efficient utilization (Batterham, 1992). Therefore, the increase in available lysine from TSBM compared to DDGS should increase available lysine digestibility and increase digestibility compared to total lysine.

Table 6. Nutrient values for dried distillers grains with solubles (DDGS) and heat-treated soybean meal (TSBM)

Item	DDGS	TSBM
Lysine, % DM	1.18	3.10
Available lysine[1], % DM	1.07	2.94
RDP[2,5], % CP	36.89	22.07
RUP[3,5], % CP	63.10	77.90
dRUP[4,5], % RUP	80.40	76.70

[1] Analyzed utilizing the 1-Fluoro-2,4-dinitrobenzene reaction at the University of Missouri Agricultural Experiment Station Chemistry Laboratory.

[2] Rumen degradable protein.

[3] Rumen undegradable protein.

[4] Digestible rumen undegradable protein.

[5] Modified three-step in vitro procedure adapted from Gargallo et al., 2006.

Nutrient flow to the duodenum, ileum, and feces resulted in no treatment differences ($P \geq 0.15$) for DM, OM, N, NDF, ADF, starch, total lysine and available lysine. However, DM flow to the duodenum tended ($P = 0.06$) to linearly decrease with increased TSBM inclusion. Based on diet formulation and nutrient analysis, flow differences were not expected for DM, OM, NDF, ADF, and starch. Furthermore, results from the experiment suggest that increases in RUP through TSBM supplementation did not result in increased N, total lysine and available lysine flow to the small intestine. Total N and nonmicrobial N flow to the duodenum was not different across treatments ($P \geq 0.28$; Table 7). Therefore, microbial N flow did not observe treatment differences ($P = 0.83$) when replacing DDGS with TSBM, suggesting that microbial protein synthesis was not limited by the decreasing levels of RDP in the diets. However, it has been reported that decreasing the RDP concentration in diets decreases microbial N flow (Reynal and Broderick, 2005). Additionally, the increasing RUP concentration in the diets, when TSBM replaced DDGS, did not elicit an increase of N at the duodenum. As there were no differences in N intake, microbial N flow and total N at the duodenum, the similarity in RUP (% of CP) concentrations between DDGS and TSBM (63.11 and 77.93%, respectively) may explain why there was not an increase in N at the duodenum when replacing DDGS with TSBM. Larger differences in RUP concentrations between feedstuffs may elicit greater differences in microbial and nonmicrobial N flow to the duodenum. A study evaluating soybean meal (high RDP) and a combination of corn gluten meal and blood meal (high RUP) as supplemental protein sources reported increased microbial N flow with SBM compared to the combination of corn gluten meal and blood meal (Cecava et al., 1991). However, the combination of corn gluten meal and blood meal increased nonmicrobial N at the duodenum compared to SBM. Furthermore, the evaluation of different energy and protein feedstuffs varying in RUP content, suggested increased N at the duodenum for sources that are less degraded in the rumen (Zinn et al., 1981; Cecava et al., 1988).

Table 7. Nitrogen (N) intake and site of digestion in steers fed forage-based diets with heat-treated soybean meal

	Treatments[1]					*P* - value			
Item	TSBM0	TSBM4	TSBM8	TSBM12	SEM	TRT	DDGS vs. TSBM	Linear	Quadratic
N intake, g/d	205	222	230	238	16.16	0.14	0.04	0.03	0.63
Duodenal flow, g/d									
Feed	111	108	115	103	8.77	0.68	0.81	0.61	0.51
Bacterial	55	53	56	52	3.87	0.35	0.58	0.55	0.54
Total	165	161	172	155	10.45	0.28	0.66	0.43	0.31
Ileal flow, g/d	78	78	78	73	5.20	0.40	0.62	0.21	0.30
Fecal excretion, g/d	71	69	68	68	5.53	0.80	0.37	0.37	0.74
Digestibility									
True ruminal, % of N intake	46.0	49.1	47.7	57.1	2.9	0.07	0.12	0.03	0.28
Apparent intestinal, % of N intake	46.3	43.7	48.1	36.5	3.0	0.04	0.24	0.05	0.10
Apparent intestinal, % of N entering duodenum	57.3	57.4	60.9	56.3	1.2	0.02	0.32	0.86	0.02
Small intestinal, % of N intake	42.9	39.9	42.7	34.3	2.5	0.07	0.15	0.04	0.25
Large intestinal, % of N intake	3.3	3.8	5.4	2.2	1.4	0.30	0.70	0.73	0.12
Total tract, % of N intake	65.6	68.2	69.8	71.6	1.4	0.03	0.01	0.01	0.70

[1] Dietary percent of heat-treated soybean meal (TSBM) replacing a proportion of 16% dried distillers grains plus solubles in the diet; TSBM0: 0% heat-treated soybean meal, TSBM4: 4% heat-treated soybean meal, TSBM8: 8% heat-treated soybean meal, TSBM12: 12% heat-treated soybean meal.

Total and available lysine at the duodenum was not different ($P \geq 0.34$) across treatments even though intake increased when TSBM replaced DDGS (Tables 8 and 9). Furthermore, nonmicrobial and microbial total and available lysine did not result in treatment differences ($P \geq 0.83$), suggesting the replacement of DDGS with TSBM did not decrease total and available lysine degradability. Similarly, comparing solvent extracted SBM and TSBM observed no difference in total and available lysine flow at the duodenum (Mansfield and Stern, 1994; Ipharraguerre et al., 2005). Santos et al. (1984) reported no difference in lysine flow to the duodenum for solvent extracted SBM and DDGS because of the greater RDP of SBM, even though lysine intake was greater for SBM. Furthermore, DDGS had greater lysine flow at the terminal ileum than SBM suggesting lysine from SBM was more available in the small intestine. Similarly, Borucki Castro et al. (2007) reported decreased lysine intestinal disappearance for TSBM compared to solvent extracted SBM using an in vitro method (87.8 and 97.8%, respectively).

Table 8. Lysine intake and site of digestion in steers fed forage-based diets with heat-treated soybean meal

	Treatments[1]					*P* - value			
Item	TSBM0	TSBM4	TSBM8	TSBM12	SEM	TRT	DDGS vs. TSBM	Linear	Quadratic
Lysine intake, g/d	43	51	58	65	3.87	0.01	0.01	0.01	0.75
Duodenal flow, g/d									
Feed	14	17	18	14	5.18	0.89	0.67	0.98	0.46
Microbial	39	37	40	40	4.60	0.83	0.99	0.65	0.79
Total	53	54	57	54	3.50	0.50	0.38	0.52	0.28
Ileal flow, g/d	22	22	21	20	1.65	0.29	0.35	0.09	0.42
Fecal excretion, g/d	17	17	19	17	1.58	0.26	0.49	0.96	0.11
Digestibility									
True ruminal, % of lysine intake	66.6	65.0	66.7	80.3	9.3	0.58	0.69	0.29	0.41
Apparent intestinal, % of lysine intake	83.5	74.7	71.5	56.7	4.5	0.01	0.01	0.01	0.43
Apparent intestinal, % of lysine entering duodenum	58.6	59.5	62.8	62.1	2.0	0.14	0.09	0.04	0.57
Small intestinal, % of lysine intake	71.8	65.8	65.5	51.2	4.5	0.02	0.03	0.01	0.37
Large intestinal, % of lysine intake	11.6	8.9	6.0	5.4	1.5	0.01	0.01	0.01	0.37
Total tract, % of lysine intake	61.0	66.0	67.7	74.4	2.1	0.01	0.01	0.01	0.66

[1] Dietary percent of heat-treated soybean meal (TSBM) replacing a proportion of 16% dried distillers grains plus solubles in the diet; TSBM0: 0% heat-treated soybean meal, TSBM4: 4% heat-treated soybean meal, TSBM8: 8% heat-treated soybean meal, TSBM12: 12% heat-treated soybean meal.

Table 9. Available Lysine[1] intake and site of digestion in steers fed forage-based diets with heat-treated soybean meal

	Treatments[2]					*P* - value			
Item	TSBM0	TSBM4	TSBM8	TSBM12	SEM	TRT	DDGS vs. TSBM	Linear	Quadratic
Lysine intake, g/d	37	46	52	58	3.49	0.01	0.01	0.01	0.62
Duodenal flow, g/d									
Feed	12	15	16	12	4.79	0.89	0.62	0.88	0.47
Microbial	36	34	37	37	4.31	0.83	0.99	0.64	0.78
Total	48	49	52	49	3.12	0.34	0.27	0.30	0.28
Ileal flow, g/d	19	18	18	17	1.47	0.33	0.22	0.09	0.72
Fecal excretion, g/d	15	15	15	15	1.76	0.90	0.52	0.75	0.53
Digestibility									
True ruminal, % of lysine intake	67.7	67.0	68.5	81.1	9.5	0.60	0.65	0.29	0.45
Apparent intestinal, % of lysine intake	89.5	75.9	76.1	57.6	5.7	0.01	0.01	0.01	0.60
Apparent intestinal, % of lysine entering duodenum	61.0	62.4	66.1	65.3	2.2	0.12	0.06	0.03	0.48
Small intestinal, % of lysine intake	78.4	67.0	70.0	54.7	5.0	0.01	0.01	0.01	0.39
Large intestinal, % of lysine intake	11.0	5.9	5.6	2.9	2.4	0.02	0.01	0.01	0.43
Total tract, % of lysine intake	61.0	65.8	69.8	74.0	2.6	0.01	0.01	0.01	0.87

[1] Analyzed utilizing the 1-Fluoro-2,4-dinitrobenzene reaction at the University of Missouri Agricultural Experiment Station Chemistry Laboratory.

[2] Dietary percent of heat-treated soybean meal (TSBM) replacing a proportion of 16% dried distillers grains plus solubles in the diet; TSBM0: 0% heat-treated soybean meal, TSBM4: 4% heat-treated soybean meal, TSBM8: 8% heat-treated soybean meal, TSBM12: 12% heat-treated soybean meal.

2.4.2.2. Nutrient Digestibility

Apparent ruminal digestibility of DM, OM (Table 10), and NDF was not influenced (*P* ≥ 0.16) by treatment. However, a tendency (*P* = 0.07) for a treatment effect was observed for true ruminal digestibility of N (Table 7). A linear increase in true ruminal N digestibility was observed, suggesting RDP of TSBM is greater than DDGS which contradicts values found in the current study (22.07 and 36.89% RDP, respectively). The RDP values are similar to previous research on DDGS (Castillo-Lopez et al., 2013) and TSBM (Borucki Castro et al., 2007; Cleale et al., 1987). Feeding protein sources with greater RUP shifts the site of digestion from the rumen to the small intestine to increase amino acid availability in the small intestine. However, replacing DDGS with TSBM linearly decreased (*P* = 0.05) apparent post ruminal N digestibility. In contrast, a study comparing TSBM with solvent extracted SBM reported no differences in post-ruminal N digestion (Ipharraguerre et al., 2005). Increasing TSBM inclusion levels would be expected to potentially shift site of digestion from the rumen to the small intestine. However, there were no differences in nonmicrobial-nonammonia N at the duodenum suggesting the site of digestion was not different among treatments. Nitrogen digestibility in the small intestine tended to detect treatment differences (*P* = 0.07) and decreased linear (*P* = 0.04) effects with increased DDGS inclusion. The slightly greater digestible RUP (dRUP) value for DDGS potentially increased N digestibility in the small intestine (80.43% and 76.69%, respectively) or other amino acids were potentially affected by the Maillard reaction.

Table 10. Organic matter (OM) intake and site of digestion in steers fed forage-based diets with heat-treated soybean meal

	Treatments[1]					*P* - value			
Item	TSBM0	TSBM4	TSBM8	TSBM12	SEM	TRT	DDGS vs. TSBM	Linear	Quadratic
OM intake, kg/d	7.09	7.40	7.45	7.51	0.541	0.79	0.35	0.39	0.69
Duodenal flow, kg/d									
Feed	1.99	1.96	2.11	1.76	0.21	0.57	0.84	0.50	0.37
Bacterial	1.00	0.93	1.01	1.00	0.12	0.92	0.88	0.83	0.78
Total	2.98	2.89	3.16	2.76	0.21	0.19	0.73	0.44	0.21
Ileal flow, kg/d	2.46	2.51	2.50	2.41	0.19	0.87	0.92	0.70	0.50
Fecal excretion, kg/d	2.19	2.16	2.25	2.12	0.17	0.57	0.89	0.69	0.43
Digestibility									
Apparent ruminal, % of intake	57.9	59.8	56.3	63.3	2.3	0.16	0.44	0.18	0.23
Apparent intestinal, % of intake	11.3	10.6	13.0	8.5	1.7	0.28	0.75	0.40	0.23
Apparent intestinal, % of duodenum	26.7	25.9	28.7	23.3	3.0	0.58	0.82	0.54	0.40
Small intestinal, % of intake	7.3	5.9	9.3	4.3	2.2	0.45	0.74	0.57	0.42
Large intestinal, % of intake	3.9	4.7	3.5	4.2	1.2	0.85	0.88	0.90	0.97
Total tract, % of intake	69.2	70.4	69.3	71.9	1.3	0.35	0.34	0.20	0.54

[1] Dietary percent of heat-treated soybean meal (TSBM) replacing a proportion of 16% dried distillers grains plus solubles in the diet; TSBM0: 0% heat-treated soybean meal, TSBM4: 4% heat-treated soybean meal, TSBM8: 8% heat-treated soybean meal, TSBM12: 12% heat-treated soybean meal.

True ruminal total and available lysine digestibility observed large standard errors (SEM) of 9.3 and 9.5%, respectively, which potentially precluded measurement of a treatment response ($P \geq 0.58$). Post ruminal digestibility linearly ($P = 0.01$) decreased with increased TSBM inclusion for total and available lysine. Furthermore, small intestinal total and available lysine digestibility was affected by treatment ($P \geq 0.01$) and linearly ($P = 0.01$) decreased when DDGS was replaced by TSBM. However, using an in vitro method for analyzing lysine intestinal digestibility, Borucki Castro et al. (2007) reported 87.8% intestinal digestibility for TSBM. Dried distillers grains with solubles observed an 83.9% intestinal lysine digestibility under similar procedural conditions (Mjoun et al., 2010). Interestingly, Borucki Castro et al. (2007) compared solvent extracted SBM and TSBM intestinal lysine digestibility and reported decreased digestibility for TSBM compared to solvent extracted SBM in an in vitro setting. The current study and Borucki Castro et al. (2007) potentially observed decreased lysine availability in the small intestine due to excessive heating during the non-enzymatic browning process. Lysine is susceptible to heat damage due to its ϵ-amino group reacting with reducing sugars and becoming nutritionally unavailable to the animal (Kim et al., 2012); consequently, intestinal digestibility is negatively impacted (van Barneveld et al., 1995). No difference in total and available lysine flow between treatments in the current study, therefore, decreasing total and available lysine availability in the small intestine compared to DDGS. It should be noted of the slight numerical increases in digestibility values for available lysine compared to total lysine, likely due to the 1-Fluoro-2,4-dinitrobenzene reaction potentially accounting for lysine in a form ready for digestion. However, more experiments analyzing amino acid flow with partial replacement of a feedstuff in the live animal compared to in situ and in vitro methods must be completed to determine amino acid availability.

There were no treatment differences for apparent total tract digestibility ($P \geq 0.32$) for DM, OM, NDF, ADF, and starch. Nitrogen, total and available lysine were influenced by treatment ($P \geq 0.01$) in apparent total tract digestibility across treatments. Additionally, apparent total tract digestibility was linearly influenced by TSBM inclusion ($P \geq 0.01$) and was less in the comparison of DDGS treatment vs TSBM treatments for N, total and available lysine. Although no differences in nutrient flow were experienced, differences in total tract digestibility for N indicate the influence of N concentration in the diet and the respective concentrations of RDP and RUP. In contrast, a study evaluating increasing levels of high-protein corn coproduct that replaced TSBM reported no differences in CP digestibility (Carroll et al., 2023). Several studies have reported no differences in total tract N digestibility when comparing solvent extracted SBM and TSBM (Cleale et al., 1987; Ipharraquerre et al., 2005; Reynal and Broderick, 2005). Although N total tract digestibility increased, total and available lysine total tract digestibility decreased suggests other amino acids are potentially increasing with the increased TSBM inclusion level.

The authors note the negative and variable duodenal and ileal NDF and ADF values (Table 11). The duodenal flow of NDF and ADF flow was lower compared to ileal NDF and ADF. Therefore, negative and variable post-ruminal digestibility is not plausible. Multiple studies have reported similar findings (Ansia et al., 2020; Panah et al., 2020) and attribute the unanticipated values to potentially unrepresentative samples collected from duodenal and ileal cannula and the use of chromic oxide as a digesta marker (Firkins et al., 1986; Olijhoek et al., 2016).

Table 11. Neutral detergent fiber (NDF), acid detergent fiber (ADF) and starch intake and site of digestion in steers fed forage-based diets with heat-treated soybean meal

	Treatments[1]					*P* - value			
Item	TSBM0	TSBM4	TSBM8	TSBM12	SEM	TRT	DDGS vs. TSBM	Linear	Quadratic
NDF, kg/d									
Intake	4.27	4.40	4.39	4.31	0.32	0.96	0.68	0.90	0.61
Duodenal flow	1.33	1.26	1.22	1.26	0.13	0.72	0.30	0.41	0.44
Ileal flow	1.70	1.77	1.74	1.75	0.16	0.95	0.62	0.77	0.76
Fecal excretion	1.56	1.53	1.59	1.46	0.11	0.24	0.48	0.23	0.29
Digestibility									
Apparent ruminal, % of NDF intake	69.1	70.7	72.6	70.8	2.3	0.71	0.36	0.45	0.44
Apparent intestinal, % of NDF intake	-6.0	-6.5	-10.1	-4.7	1.9	0.22	0.54	0.99	0.10
Apparent intestinal, % of NDF entering duodenum	-21.5	-22.2	-36.8	-16.8	7.0	0.24	0.61	0.99	0.14
Small intestinal, % of NDF intake	-8.9	-11.5	-12.6	-11.8	3.1	0.77	0.33	0.42	0.52
Large intestinal, % of NDF intake	3.0	5.0	2.7	7.1	2.2	0.43	0.42	0.29	0.57
Total tract, % of NDF intake	63.3	64.2	62.4	66.2	2.2	0.55	0.65	0.41	0.46
ADF, kg/d									
Intake	2.20	2.73	2.27	2.25	1.65	0.94	0.56	0.74	0.64
Duodenal flow	0.81	0.73	0.72	0.72	0.07	0.21	0.04	0.08	0.25
Ileal flow	0.96	0.99	0.99	1.01	0.09	0.93	0.56	0.58	0.88
Fecal excretion	0.93	0.89	0.98	0.93	0.07	0.15	0.83	0.36	0.81

Table 11. Neutral detergent fiber (NDF), acid detergent fiber (ADF) and starch intake and site of digestion in steers fed forage-based diets with heat-treated soybean meal (continued)

	Treatments[1]					*P* - value			
	TSBM0	TSBM4	TSBM8	TSBM12	SEM	TRT	DDGS vs. TSBM	Linear	Quadratic
Digestibility									
Apparent ruminal, % of ADF intake	63.3	67.1	68.3	68.1	2.2	0.30	0.08	0.11	0.34
Apparent intestinal, % of ADF intake	-5.8	-7.5	-12.5	-9.6	2.2	0.09	0.05	0.04	0.19
Apparent intestinal, % of ADF entering duodenum	-18.4	-23.7	-39.6	-30.3	6.7	0.08	0.05	0.04	0.17
Small intestinal, % of ADF intake	-6.9	-11.4	-12.5	-13.3	3.8	0.53	0.17	0.19	0.59
Large intestinal, % of ADF intake	1.4	7.6	-0.3	7.8	6.1	0.61	0.56	0.64	0.86
Total tract, % of ADF intake	57.5	59.5	55.4	58.4	2.8	0.59	0.89	0.89	0.83
Starch, kg/d									
Intake	0.82	0.85	0.86	0.86	0.06	0.89	0.45	0.51	0.71
Fecal excretion	0.05	0.05	0.05	0.04	0.004	0.87	0.55	0.43	0.94
Digestibility									
Total tract, % of starch intake	94.3	94.5	94.6	94.9	0.4	0.66	0.39	0.24	0.84

[1] Dietary percent of heat-treated soybean meal (TSBM) replacing a proportion of 16% dried distillers grains plus solubles in the diet; TSBM0: 0% heat-treated soybean meal, TSBM4: 4% heat-treated soybean meal, TSBM8: 8% heat-treated soybean meal, TSBM12: 12% heat-treated soybean meal.

2.4.2.3. Rumen Ammonia Concentration

Rumen ammonia concentrations observed treatment, DDGS vs. TSBM, and linear differences ($P = 0.01$) suggesting N degradation in the rumen increased when TSBM replaced DDGS at increasing levels (Table 12). Increased ammonia concentration could be attributed to the increased supply of N, however, increasing RUP has shown to decrease ammonia concentrations. Cecava et al. (1991) reported decreased ammonia concentration when evaluating solvent extracted SBM (source high in RDP) and a combination of corn gluten meal and blood meal (sources high in RUP). Furthermore, studies comparing solvent extracted SBM and TSBM reported increased ammonia concentrations with solvent extracted SBM (Calsamiglia et al., 1995; Stanford et al., 1995). In the present study, as TSBM replaces DDGS, RDP is assumed to decrease, however, no difference in microbial and feed N at the duodenum suggests the additional RUP from TSBM was degraded in the rumen. This contradicts our in situ finding that showed DDGS contained a greater concentration of RDP compared to TSBM.

Table 12. Ammonia concentrations and rumen liquid dilution rate in steers fed forage-based diets with heat-treated soybean meal

	Treatments[1]					*P* - value			
Item	TSBM0	TSBM4	TSBM8	TSBM12	SEM	TRT	DDGS vs. TSBM	Linear	Quadratic
Ammonia, mmol	4.90	6.06	7.08	7.28	0.54	0.01	0.01	0.01	0.14
Dilution Rate, %/h	10.3	9.1	10.2	10.3	0.01	0.58	0.59	0.69	0.37

[1] Dietary percent of heat-treated soybean meal (TSBM) replacing a proportion of 16% dried distillers grains plus solubles in the diet; TSBM0: 0% heat-treated soybean meal, TSBM4: 4% heat-treated soybean meal, TSBM8: 8% heat-treated soybean meal, TSBM12: 12% heat-treated soybean meal.

2.5. Implications

Increasing calculated MP and metabolizable lysine flow to the small intestine did not improve growth performance in growing steers fed forage-based diets. The observed lack of difference in growth performance due to increased lysine concentrations may suggest that the lysine requirement was met or was not the first limiting amino acid. However, due to both MP and lysine increasing when TSBM replaces DDGS, it is difficult to distinguish between MP and lysine because of the lack of differences in growth performance. Replacing DDGS with TSBM linearly increased total and available lysine intake because of differences in lysine concentration between DDGS and TSBM. The lack of differences in total N flow at the duodenum suggests that both DDGS and TSBM have similar RUP concentration. However, potentially unrepresentative samples could also contribute to the lack of differences with nutrient flow values. Nitrogen, total, and available lysine total tract digestibility linearly increased with TSBM inclusion suggesting increased availability from TSBM compared to DDGS. However, decreased post-ruminal digestibility signifies decreased nutrient availability from TSBM. The use of TSBM in growing cattle diets will be based on TSBM availability and feed costs. Likely, TSBM could be incorporated at low inclusion rates to increase N in growing diets.

2.6. Literature Cited

AOAC, 2006. Official Methods of Analysis. 18th Edition, Association of Official Analytical Chemists, Gaithersburg, MD.

AOAC, 2010. Official Methods of Analysis. 18th Edition. Association of Official Analytical Chemists, Washington, DC.

Ansia, I., H. H. Stein, C. Brøkner, and J. K. Drackley. 2020. Short communication: A pilot study to describe duodenal and ileal flows of nutrients and to estimate small intestine endogenous protein losses in weaned calves. J. Dairy Sci. 103:9102-9109. doi:10.3168/jds.2020-18281.

Awawdeh, M. S., E. C. Titgemeyer, J. S. Drouillard, R. S. Beyer, and J. E. Shirley. 2007. Ruminal degradability and lysine bioavailability of soybean meals and effects on performance of dairy cows. J. Dairy Sci. 90:4740-4753. doi:10.3168/jds.2007-0210.

Batterham, E. S. 1992. Availability and utilization of amino acids for growing pigs. Nutrition Research Reviews. 5:1-18. doi:10.1079/NRR19920004.

Borucki Castro, S. I., L. E. Phillip, H. Lapierre, P. W. Jardon, and R. Berthiaume. 2007. Ruminal degradability and intestinal digestibility of protein and amino acids in treated soybean meal products. J. Dairy Sci. 90:810-822. doi:10.3168/jds.S0022-0302(07)71565-5.

Broderick, G. A. and J. H. Kang. 1980. Automated simultaneous determination of ammonia and total amino acids in ruminal fluid and in vitro media. J. Dairy Sci. 63:64-75. doi:10.3168/jds.S0022-0302(80)82888-8.

Calsamiglia, S., M. D. Stern, and J. L. Firkins. 1995. Effects of protein source on nitrogen metabolism in continuous culture and intestinal digestion in vitro. J. Anim. Sci. 73:1819-1827. doi:10.2527/1995.7316819x.

Carroll, A. L., D. L. Morris, M. L. Jolly-Beithaupt, K. J. Herrick, A. K. Watson, and P. J. Kononoff. 2023. Energy and nitrogen utilization of lactating diary cattle fed increasing inclusion of a high-protein processed corn coproduct. J. Dairy Sci. 106:8809-8820. doi:10.3168/jds.2023-23360.

Castillo-Lopez, E., T. J. Klopfenstein, S. C. Fernando, and P. J. Kononoff. 2013. In vivo determination of rumen undegradable protein of dried distillers grains with solubles and evaluation of duodenal microbial crude protein flow. J. Anim. Sci. 91:924-934. doi:10.2527/jas2012-5323.

Cecava, M. J., N. R. Merchen, L. L. Berger, and G. C. Fahey, Jr. 1988. Effects of dietary energy level and protein source on site of digestion and duodenal nitrogen and amino acid flows in steers. J. Anim. Sci. 1988. 66:961-974. doi:10.2527/jas1988.66496x.

Cecava, M. J., N. R. Merchen, L. L. Berger, R. L. Mackie, and G. C. Fahey, Jr. 1991. Effects of dietary energy level on protein source on nutrient digestion and ruminal nitrogen metabolism in steers. J. Anim. Sci. 69:2230-2243. doi:10.2527/1991.6952230x.

Chalupa, W. 1975. Rumen bypass and protection of proteins and amino acids. J. Dairy Sci. 58:1198-1218. doi:10.3168/jds.S0022-0302(75)84697-2.

Clark, J. H., M. R. Murphy, and B. A. Crooker. 1987. Supplying the protein needs of dairy cattle from by-product feeds. J. Dairy Sci. 70:1092-1109. doi:10.3168/jds.S0022-0302(87)80116-9.

Cleale IV, R. M, R. A. Britton, T. J. Klopfenstein, M. L. Bauer, D. L. Harmon, and L. D. Satterlee. 1987. Induced non-enzymatic browning of soybean meal. II. Ruminal escape and net portal absorption of soybean protein treated with xylose. J. Anim. Sci. 65:1319-1326. doi:10.2527/jas1987.6551319x.

Coetzer, C. M. 2000. Manipulation of ruminal protein degradability. ProQuest Dissertations and Theses Global.

Fenton, T. W. and M. Fenton. 1979. An improved procedure for the determination of chromic oxide in feed and feces. Can. J. Anim. Sci. 59:631-634. doi:10.4141/cjas7909-081.

Firkins, J. L., L. L. Berger, N. R. Merchen, G. C. Fahey, Jr., and D. R. Nelson. 1986. Effects of feed intake and protein degradability on ruminal characteristics and site of digestion in steers. J. Dairy Sci. 69:2111-2123. doi:10.3168/jds.S0022-0302(86)90643-9.

Gargallo, S., S. Calsamiglia, and A. Ferret. 2006. Technical note: A modified three-step in vitro procedure to determine intestinal digestion of proteins. J. Anim. Sci. 84:2163-2167. doi:10.2527/jas.2004-704.

Hammond, A. C. 1983. The use of blood urea nitrogen concentration as an indicator of protein status in cattle. Bov. Pract. 18:114-118. doi:10.21423/bovine-vol1983no18p114-118.

Hammond, A. C. 1996. Update on BUN and MUN as a guide for protein supplementation in cattle. Pages 45-54 in Proc. 7th Annu. Rumin. Nutr. Symp., Univ. Florida, Gainesville, FL.

Heiderscheit, K. J. and S. L. Hansen. 2020. Effect of rumen-protected lysine on growth performance, carcass characteristics, and plasma amino acid profile in feedlot steers. Transl. Anim. Sci. 4:1-9. doi:10.1093/tax/txaa128.

Herrera-Saldana, R. and J. T. Huber. 1989. Influence of varying protein and starch degradabilities on performance of lactating cows. J. Dairy Sci. 72:1477-1483. doi:10.3168/jds.S0022-0302(89)79257-2.

Ipharraguerre, I. R., J. H. Clark, and D. E. Freeman. 2005. Rumen fermentation and intestinal supply of nutrients in diary cows fed rumen-protected soy products. J. Dairy Sci. 88:2879-2892. doi:10.3168/jds.S0022-0302(05)72969-6.

Jennings, J. S., B. E. Meyer, P. J. Guiroy, and N. A. Cole. 2018. Energy costs of feeding excess protein from corn-based by-products to finishing cattle. J. Anim. Sci. 96:653-669. doi:10.1093/jas/sky021.

Kim, J. C., B. P. Mullan, and J. R. Pluske. 2012. Prediction of apparent, standardized, and true ileal digestible total and reactive lysine contents in heat-damaged soybean meal samples. J. Anim. Sci. 90:137-139. doi:10.2527/jas53819.

Klemesrud, M. J., T. J. Klopfenstein, and A. J. Lewis. 2000a. Metabolizable methionine and lysine requirements of growing cattle. J. Anim. Sci. 78:199-206. doi:10.2527/2000.781199x.

Klemesrud, M. J., T. J. Klopfenstein, R. A. Stock, A. J. Lewis, and D. W. Herold. 2000b. Effect of dietary concentration of metabolizable lysine on finishing cattle performance. J. Anim. Sci. 78:1060-1066. doi:10.2527/2000.7841060x.

Mansfield, H. R. and M. D. Stern. 1994. Effects of soybean hulls and lignosulfonate-treated soybean meal on ruminal fermentation in lactating diary cows. J. Dairy Sci. 77:1070-1083. doi:10.3168/jds.S0022-0302(94)77042-9.

Merchen, N. R. and E. C. Titgemeyer. 1992. Manipulation of amino acid supply to the growing ruminant. J. Anim. Sci. 70:3238-3247. doi:10.2527/1992.70103238x.

Mjoun, K. K. F. Kalscheur, A. R. Hippen, and D. J. Schingoethe. 2010. Ruminal degradability and intestinal digestibility of protein and amino acids in soybean and corn distillers grains products. J. Dairy Sci. 93:4144-4154. doi:10.3168/jds.2009-2883.

Montaño, M. F., J. O. Chirino, B. C. Latack, J. Salinas-Chavira, and R. A. Zinn. 2019. Influence of supplementation of growing diets enriched with rumen-protected methionine and lysine on feedlot performance and characteristics of digestion in holstein steer calves. Applied Animal Science 35:318-324. doi:10.15232/aas.2019-01843.

National Academies of Sciences, Engineering, and Medicine. 2016. Nutrient requirements of beef cattle, eighth revised edition. Washington, DC: The National Academies Press. doi:10.17226/19014.

National Academies of Sciences, Engineering, and Medicine. 2021. Nutrient requirements of dairy cattle, eighth revised edition. Washington, DC: The National Academies Press. doi:10.17226/25806.

Olijhoek, D. W., A. L. F. Hellwing, M. Brask, M. R. Weisbjerg, O. Højberg, M. K. Larson, J. Dijkstra, E. J. Erlandsen, and P. Lund. 2016. Effect of dietary nitrate level on enteric methane production, hydrogen emission, rumen fermentation, and nutrient digestibility in dairy cows. J. Dairy Sci. 99:6191-6205. doi:10.3168/jds.2015-10691.

Oney, C. R., J. K. Gramkow, F. Henry Hilscher, G. E. Erickson, J. C. MacDonald, T. J. Klopfenstein, and A. K. Watson. 2019. Corn silage rumen undegradable protein content and response of growing calves to rumen undegradable protein supplement. Transl. Anim. Sci. 3:51-59. doi:10.1093/tas/txz014.

Orskov, E. R. 1970. Nitrogen utilization by the young ruminant. Page 20 in Proc. 4th Nutr. Conf. Feed Manuf. H. Swan and D. Lewis, ed. Univ. Nottingham, Churchill, London, Engl.

Panah, F. M., S. Lashkari, A. L. F. Hellwing, M. Larsen, and M. R. Weisbjerg. 2020. Effects of toasting and decortication of oat on nutrient digestibility in the rumen and small intestine and on amino acid supply in dairy cows. J. Dairy Sci. 103:1481-1499. doi:10.3168/jds.2019-17142.

Preston, R. L., F. Byers, and K. R. Stevens. 1978. Estrogenic activity and growth stimulation in steers fed varying protein levels. J. Anim. Sci. 46:541-546. doi:10.2527/jas1978.462541x.

Preston, R. L., D. D. Schnakenberg, and W. H. Pfander. 1965. Protein utilization in ruminants. I. Blood urea nitrogen as affected by protein intake. J. Nutr. 86:281-288.

Reynal, S. M. and G. A. Broderick. 2005. Effect of dietary level of rumen-degraded protein on production and nitrogen metabolism in lactating dairy cows. J. Dairy Sci. 88:4045-4064. doi:10.3168/jds.S0022(05)73090-3.

Reynolds, C. K. and N. B. Kristensen. 2008. Nitrogen recycling through the gut and the nitrogen economy of ruminants: an asynchronous symbiosis. J. Anim. Sci. 86(E. Suppl.):E293-E305. doi:10.2527/jas.2007-0475.

Samuelson, K. L., M. E. Hubbert, E. R. Oosthuysen, and C. A. Löest. 2023. Effects of dietary protein concentration and degradability on performance, carcass characteristics, net energy utilization, and metabolizable protein balance of finishing beef heifers receiving 0 or 400 mg of ractopamine hydrochloride. Applied Animal Science. 39:56-68. doi:10.15232/aas.2022-02351.

Santos, K. A., M. D. Stern, and L. D. Satter. 1984. Protein degradation in the rumen and amino acid absorption in the small intestine of lactating dairy cattle fed various protein sources. J. Anim. Sci. 58:244-255. doi:10.2527/jas1984.581244x.

Satter, L. D. and L. L. Sylter. 1974. Effect of ammonia concentration on rumen microbial protein production in vitro. Br. J. Nutr. 32:199-208. doi:10.1079/BJN19740073.

Spore, T. J., Z. E. Carlson, K. H. Wilke, G. E. Erickson, T. J. Klopfenstein, J. C. MacDonald, and A. K. Watson. 2021. Effects of replacing wet distillers grains with supplemental SoyPass in forage-based growing cattle diets. Transl. Anim. Sci. 5:1-10. doi:10.1093/tas/txab117.

Stanford, K., T. A. McAllister, Z. Xu, M. Pickard, and K. J. Cheng. 1995. Comparisons of lignosulfonate-treated canola meal and soybean meal as rumen undegradable protein supplements for lambs. Can. J. Anim. Sci. 75:371-377. doi:10.4141/cjas95-056.

Titgemeyer, E. C., N. R. Merchen, and L. L. Berger. 1989. Evaluation of soybean meal, corn gluten meal, blood meal and fish meal as sources of nitrogen and amino acids disappearing from the small intestine of steers. J. Anim. Sci. 67:262-275. doi:10.2527/jas1989.671262x.

Udén, P., P. E. Colucci, and P. J. Van Soest. 1980. Investigation of chromium, cerium and cobalt as makers in digesta. Rate of passage studies. J. Sci. Food Agric. 31:625-632. doi:10.1002/jsfa.2740310702.

USDA. 2017. United states standards for grades of carcass beef. United states department of agriculture, agricultural marketing service, livestock, poultry, and seed program. Washington, DC.

United States Environmental Protection Agency, 2024. Renewable Fuel Standard Program. Renewable Fuel Standard Program | US EPA.

van Barneveld, R. J., E. S. Batterham, and D. C. Skingle. 1995. The effect of heat on amino acids for growing pigs. 4. Nitrogen balance and urine, serum, and plasma composition of growing pigs fed on raw or heat-treated field peas (*Pisum sativum*). Br. J. Nutr. 73:259-273. doi:10.1079/BJN19950028.

Vasconcelos, J. T., N. A. Cole, K. W. McBride, A. Gueye, M. L. Galyean, C. R. Richardson, and L. W. Greene. 2009. Effects of dietary crude protein and supplemental urea levels on nitrogen and phosphorus utilization by feedlot cattle. J. Anim. Sci. 87:1174-1183. doi:10.2527/jas.2008-1411.

Veloso, V. A., C. L. Van Bibber-Krueger, and J. S. Drouillard. 2016. Ruminally-protected lysine (SafeGain) improves performance of growing beef cattle. Kansas Agricultural Experiment Station Research Reports: Vol. 2: Iss. 1. doi:10.4148/2378-5977.1174.

Vercoe, J. E. 1969. The transfer of nitrogen from the blood to the rumen in cattle. Aust. J. Agric. Res. 20:191-197.

Watson, A. K., B. L. Nuttelman, T. J. Klopfenstein, L. W. Lomas, and G. E. Erickson. 2013. Impacts of limit-feeding procedure on variation and accuracy of cattle weights. J. Anim. Sci. 91:5507-5517. doi:10.2527/jas2013-6349.

Wiseman, A. R. 2021. Evaluation of novel feed ingredients on finishing and growing cattle performance and intake modeling for growing cattle. ProQuest Dissertations and Theses Global.

Zinn, R. A. and F. N. Owens. 1986. A rapid procedure for purine measurement and its use for estimating net ruminal protein synthesis. Can. J. Anim. Sci. 66:157-166. doi:10.4141/cjas86-017.

Zinn, R. A. and F. N. Owens. 1993. Ruminal escape protein for lightweight feedlot calves. J. Anim. Sci. 71:1677-1687. doi:10.2527/1993.7171677x.

Zinn, R. A., L. S. Bull, and R. W. Hemken. 1981. Degradation of supplemental proteins in the rumen. J. Anim. Sci. 52:857. doi:10.2527/jas1981.524857x.