

The Use of Dried Distillers Grains with Solubles in Feedlot Rations for Lambs

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Summary

Dried distillers grains with solubles (DDGS) are a valuable feed resource for ruminants due to their concentrations of both protein and energy. Average daily gain (ADG) of lambs appears to improve with inclusion rate of DDGS at 20% to 30% of dietary DM. Dry matter intake likewise appears to improve when DDGS is included at rates less than 30%, with intake declining as DDGS is included at rates \geq 30%. Additionally, carcass characteristics of lambs have not been adversely affected by feeding DDGS. One of the negative connotations with feeding greater concentrations of DDGS is the risk of sulfur (S) induced polioencephalomalacia (PEM). Sulfur toxicity in lambs in research settings has not been a significant issue when the primary source of S is from DDGS, and inclusion rates of DDGS as high as 60% of dietary DM are possible with

proper management. However, regardless of research outcomes, diets exceeding 0.4% dietary S increases the risk of PEM, and caution should be encouraged when high S diets are being fed. At present, the cost and availability of ethanol co-products are limiting their use in feedlot diets to the minimum amounts needed to meet protein requirements. However, when competitively priced DDGS can be utilized to meet both protein and energy requirements for ruminants. Continued research on DDGS will be important to understand changes in product quality especially as modifications to fermentation processes and oil extraction in the ethanol industry continue to alter co-product nutrient content.

Key Words: Dried Distillers Grain With Solubles, Feedlot, Lamb, Sulfur

Introduction

The ethanol industry in the United States of America produced 36.5 million metric tons of dried distillers grains with solubles (DDGS) on average over the past three years 2018-2020 (USDA-ERS Bioenergy Statistics, 2020). Only a handful of ethanol plants are located outside the midwestern states, increasing transportation costs of DDGS for western U.S. sheep industry. However, the distribution of ethanol production makes DDGS a potential feed resource for many Midwestern sheep operations. Competitively priced DDGS can be utilized to meet both protein and energy requirements for ruminants. The evolution of ethanol production has resulted in changes to the nutrient content of DDGS. These changes have resulted in reduction in fat content and increases in protein content of some, but not all, DDGS products making feed analysis critically important in diet formulation. Dried distillers grains with solubles have been utilized in lamb feedlot diets at rates as high as 60% DM basis (Schauer et al., 2008; Neville et al., 2010; Felix et al., 2012). Due to greater concentrations of phosphorus (P) and sulfur (S) in DDGS, proper care must be taken in diet formulation to ensure animal performance and prevent urinary calculi and polioencephalomalacia (PEM; NRC, 2007). The objective of this review will be to provide an overview of research on lamb performance and carcass characteristics, as well as to address some of the perceived barriers to increasing the use of DDGS in lamb feedlot rations.

Nutrient Profile Of Dried Distillers Grains With Solubles

The nutrient content of DDGS has changed with the evolution of the ethanol production process. Traditionally, crude protein (CP), fat, and P would increase by 3-fold when comparing corn to distillers grains (Klopfenstein et al., 2008). Understanding the nutrient content of the DDGS products is critical to ensure properly balanced rations as variation in nutrient content can occur based on ethanol production processes. One of the benefits to utilizing DDGS as a primary energy source is the

low starch content. The removal of starch during the fermentation process leaves a higher fiber co-product that is safely fermented in the rumen, and potentially lessens the risk of acidosis when included at rates above 20% of dietary DM in feedlot diets (Klopfenstein et al., 2008).

With modifications to the fractionation process, and centrifugation of thin stillage, oil is now being removed resulting in a lower fat product than previously described (U.S. Grain Council). Furthermore, variation between ethanol plants and processing methods can impact fat content of DDGS with fat values ranging from 5.4% (U.S. Grain Council) to 12% (Lardy and Anderson, 2014). Previous research with lambs fed a low-fat DDGS resulted in similar performance to those fed either conventional DDGS or low-fat DDGS with added corn oil (Redding et al., 2014). Similar results from Van Emon et al. (2012) also observed lamb performance was not impacted by total dietary fat concentration (3.5 to 7.0%) in rations containing either 25% or 50% DDGS.

Dried distillers grains with solubles have an average CP content of 30.8% (NASEM, 2016) with 63% of the CP being rumen undegradable protein (Castillo-Lopes et al., 2013). As a result of new technology which resulted in improved milling and fiber separation, high-CP DDGS with CP values between 44 to 50% (U.S. Grains Council) are now available. It is important to note that not all ethanol plants utilize this technology and the actual CP content of DDGS received at each producer operation should be evaluated and actual values used to balance diets appropriately.

Mineral content of DDGS must also be considered when developing rations for feedlot lambs. Due to high P content ($0.86 \pm 0.11\%$; NASEM 2016) of DDGS additional calcium (Ca) is generally required to maintain a 2:1 Ca to P ratio in diets for prevention of urinary calculi (Schauer et al., 2005; NRC 2007). Sulfur present in DDGS should also be considered when balancing rations and will be discussed in a later section.

Other grain sources including sorghum and wheat have also been used in the ethanol production industry and may be available in the southern states and Canada, while corn is the predomi-

nant grain source used in the central United States. Wheat-DDGS contains more CP than conventional corn-DDGS (39.4% vs. 30.5%, respectively), similar ADF (15.8% vs. 13.3%), but less crude fat than conventional corn-DDGS (4.8 vs 12.1%; Curry, 2014). Cellulosic ethanol production is also producing by-products for use in livestock production, although little information on these products is currently available. Data from Lundy et al. (2015) reported that digestibility of by-products resulting from corn-fiber fermentation may be lower than those of traditional grain ethanol production in lambs.

Feed And Ration Management

One of the major management issues with feeding diets containing greater concentrations of DDGS to lambs is sorting that can occur in a textured or mixed diet. Some research has circumvented this issue by providing a finely ground diet which prevents sorting (Schauer et al., 2008; Crane et al., 2017). Totally ground rations decreased ruminal pH, below the 5.0 threshold representative of acute ruminal acidosis (Crane et al., 2017) although clinical acidosis was not observed; whereas ruminal pH decreased from 5.8 to 5.3 when lambs were provided rations including cracked corn, ground hay, and DDGS (Neville et al., 2011). In either case, the lower end of the pH reported in these studies would be indicative of either sub-acute or acute acidosis. While total mixed rations with cracked or rolled grain, DDGS, and roughage will require diligent bunk management to prevent sorting, both conventional and completely ground diets still have the potential to induce acidosis if managed inappropriately. Changes in feeding behavior could also explain the lack of visual signs of ruminal acidosis in some previous research. For example, in steers feeding behavior including number of meals and size of meals have been affected by concentrations of DDGS and corn particle size indicating potential adaptation by the animal to diet (Swanson et al., 2014) to help regulate ruminal pH. These principles have not been evaluated in lambs and further research would be needed to determine if use of fine-ground diets or traditional

particle sizes and feeding method (self-feeders vs. daily feeding) are in fact altering lamb feeding behavior.

Inclusion of wet products such as silages or liquid binders may aide in decreasing sorting issues found in DDGS rations but eliminate the use of self-feeders. Including wet or modified distillers grains with solubles instead of DDGS is another option to improve ration consistency and reduce sorting in total mixed rations provided daily. The use of modified and wet distillers grains products has not been extensively researched in lamb finishing diets. Utilizing either wet or modified distiller products will increase cost associated with transport due to low DM content, and require additional inputs associated with handling and storage, especially in warmer climates.

Research in beef cattle has demonstrated feeding roughage at conventional rates is important even in diets containing distillers grains products to optimize performance (NASEM, 2016). This topic has not been as thoroughly evaluated in lamb finishing diets, however the general practices of beef and lamb finishing management are similar, a brief review of recent literature indicates that typical roughage levels in lamb finishing diets are 10% (Huls et al., 2006; Van Emon et al., 2012) but can range from 5 to 30%. These concentrations are greater than the 6% (Hales et al., 2013) and 7.5% (May et al., 2011) roughage shown to optimize gain in beef cattle fed diets containing wet distillers grains with solubles. However, more detailed research is needed to accurately relate beef cattle data to the actual results in lamb finishing. Further, data on the use of lower quality roughage sources with various grain processing methods when fed in combination with DDGS to finishing lambs are also warranted.

Traditional roughage sources fed in combination with DDGS have included alfalfa hay (Neville et al., 2010), soybean hulls (Felix et al., 2012), or commercially manufactured pellets (Redding et al., 2014; Crane et al., 2018). Alternative roughage sources fed in combination with DDGS has also been evaluated. Cottonseed hulls have been used in lamb rations containing 40% DDGS (Whitney and Lupton, 2010). Other non-traditional roughages, including redberry juniper, have also been used as roughage

sources in DDGS based feedlot rations (Whitney et al., 2014). However, Whitney and Lupton, (2010) and Whitney et al. (2014) both expressed the need for caution related to plant secondary compound concentrations e.g., condensed tannins or volatile oils present in cottonseed hulls and redberry juniper.

Animal Performance

Dried distillers grains with solubles are typically included in rations at rates sufficient to reach protein requirements for the class of animals. This is largely a function of two factors; cost and availability. Recently, DDGS have been less competitively priced compared to corn or other energy sources, providing an economic barrier to inclusion rates beyond those needed to meet protein requirements. Costs associated with transport for many western states sheep producers has also been a barrier to further use of DDGS within the sheep industry. Recent discussion relative to an apparent decrease in overall co-product availability may further limit use of DDGS in the sheep industry. However, research has demonstrated that DDGS can be utilized in lamb finishing rations at levels up to 60% of dietary DM without significant decreases in animal performance (Schauer et al., 2006; Neville et al., 2010) allowing for DDGS to be used as the primary source of energy in finishing rations if economical.

The impacts of DDGS inclusion rate on DMI in lambs has been evaluated. Intake data from nine unique research studies utilizing DDGS in lamb feedlot rations have been summarized (Table 1). Studies summarizes had to include corn-DDGS, a control or 0% DDGS treatment, and at least one additional inclusion rate of DDGS. While not analyzed statistically, average intakes appear to increase when DDGS is included at rates less than 30% of dietary DM, while DDGS inclusion at rates $\geq 30\%$ of dietary DM appear to decrease DMI. This trend is consistent with previous reviews on DDGS in beef cattle (Klopfenstein et al., 2008). However, not all research follows this trend. Felix et al. (2012) reported no differences in DMI with increasing dietary DDGS inclusion. Schauer et al. (2008) found feeding greater concentrations of DDGS increased DMI from 1.68kg to 1.91kg, for 0% DDGS and 60% DDGS, respectively, which is contrary to most research with sheep and beef cattle. Decreased DMI was attributed to increased crude protein of the diet in lambs fed DDGS (Van Emon et al., 2012). Additional research evaluating the impacts of inclusion rates less than 20% and greater than 40% are needed to further elucidate responses at lesser and greater levels of DDGS.

The impacts of DDGS inclusion on ADG and G:F have also been evaluated and are presented in Tables 2 and 3, respectively. These studies were summa-

Table 1. Impacts of dried distillers grains with solubles (DDGS) inclusion on dry matter intake (DMI; kg) of feedlot lambs.¹

| | DDGS, % | | | | |
|----------------------------|---------|------|-------|-------|------|
| | 0 | <20 | 20-30 | 30-40 | >40 |
| Lodge et al., 1997 | 1.24 | - | - | 1.27 | - |
| Huls et al., 2006 | 1.59 | - | 1.65 | - | - |
| Schauer et al., 2006 | 1.87 | 1.92 | 1.92 | - | - |
| Schauer et al., 2008 | 1.68 | - | 1.78 | 1.83 | 1.91 |
| McKeown et al., 2010 | 1.39 | - | 1.47 | - | - |
| Neville et al., 2011 | 1.30 | - | 1.50 | 1.40 | 1.30 |
| Felix et al., 2012 | 1.48 | - | 1.56 | 1.49 | 1.47 |
| Whitney et al., 2014 | 1.47 | - | - | 1.28 | - |
| Crane et al., 2017 | 1.80 | 1.75 | 1.55 | - | - |
| Mean DMI ² , kg | 1.53 | 1.83 | 1.63 | 1.45 | 1.56 |

¹ Data utilized from published research articles utilizing corn DDGS in lamb rations. Studies included used a 0% control and at least one DDGS treatment.

² Mathematical average.

Table 2. Impacts of dried distillers grains with solubles (DDGS) inclusion on average daily gain (ADG; kg) of feedlot lambs.¹

| | DDGS, % | | | | |
|----------------------------|---------|------|-------|-------|------|
| | 0 | <20 | 20-30 | 30-40 | >40 |
| Lodge et al., 1997 | 0.28 | - | - | 0.31 | - |
| Huls et al., 2006 | 0.29 | - | 0.29 | - | - |
| Schauer et al., 2005 | 0.31 | 0.34 | - | - | - |
| Schauer et al., 2006 | 0.20 | 0.24 | 0.27 | - | - |
| Schauer et al., 2008 | 0.26 | - | 0.28 | 0.28 | 0.28 |
| McKeown et al., 2010 | 0.38 | - | 0.40 | - | - |
| Felix et al., 2012 | 0.32 | - | 0.36 | 0.31 | 0.30 |
| Whitney et al., 2014 | 0.20 | - | - | 0.23 | - |
| Crane et al., 2017 | 0.29 | 0.29 | 0.29 | - | - |
| Mean ADG ² , kg | 0.28 | 0.29 | 0.32 | 0.28 | 0.29 |

¹ Data utilized from published research articles utilizing corn DDGS in lamb rations. Studies included used a 0% control and at least one DDGS treatment.
² Mathematical average.

Table 3. Impacts of dried distillers grains with solubles (DDGS) inclusion on feed efficiency (G:F; kg of gain: kg of feed) of feedlot lambs.¹

| | DDGS, % | | | | |
|-----------------------|---------|------|-------|-------|------|
| | 0 | <20 | 20-30 | 30-40 | >40 |
| Lodge et al., 1997 | 0.23 | - | - | 0.22 | - |
| Huls et al., 2006 | 0.18 | - | 0.18 | - | - |
| Schauer et al., 2006 | 0.11 | 0.12 | 0.14 | - | - |
| Schauer et al., 2008 | 0.16 | - | 0.16 | 0.15 | 0.15 |
| McKeown et al., 2010 | 0.27 | - | 0.28 | - | - |
| Felix et al., 2012 | 0.22 | - | 0.23 | 0.21 | 0.21 |
| Whitney et al., 2014 | 0.17 | - | - | 0.18 | - |
| Crane et al., 2017 | 0.16 | 0.17 | 0.19 | - | - |
| Mean G:F ² | 0.19 | 0.15 | 0.20 | 0.19 | 0.18 |

¹ Data utilized from published research articles utilizing corn DDGS in lamb rations. Studies included used a 0% control and at least one DDGS treatment.
² Mathematical average.

ized in the same format as DMI, however only 9 and 8 research reports provided ADG and G:F, respectively. While not statistically analyzed, ADG appears to increase when DDGS is included at rates between 20 and 30% of dietary DM. Average daily gain values presented here again correspond well to improved gains found in beef cattle consuming 20 to 30% DDGS (Klopfenstein et al., 2008). Feed efficiency appears to also improve when DDGS inclusion rates are between 20 and 30%, however reported results for G:F when DDGS is included at rates less than 20% and over 40% are very limited and require additional research in feedlot lamb.

There are studies that have reported improved, or at a minimum, no negative impacts of feeding greater concentrations of DDGS on feedlot lamb performance. Van Emon et al. (2012) determined that feeding lambs 50% DDGS did not cause negative effects on feedlot performance. Contrary to the data presented by Schauer et al. (2008) and Van Emon et al. (2012), lambs fed DDGS at greater than 20% had lower ADG and G:F (Felix et al., 2012). The ADG observed by Schauer et al. (2008) were 0.26 to 0.28kg/d while those presented by Felix et al. (2012) were consistently above 0.3kg/d. It is possible that factors related to roughage source played a role

in the differences observed between research as these studies utilized a variety of roughages including hay and soybean hulls, or that the response was driven by CP content of the diets. When considering the CP content of the diets, Schauer et al. (2008) evaluated diets in excess of 20% while those of Felix et al. (2012) fell between 14.5 and 20.6%.

Future research evaluating when inclusion rate of DDGS optimizes the combination of ADG, DMI, and G:F in lambs is still warranted. The summary data presented in this manuscript appears to indicate that optimal inclusion level will be between 20% and 30% DDGS (DM basis). Further research on this topic, as well as more detailed research on the impacts of protein content of the ration, roughage source, and lamb breed type will aid in future industry recommendations.

Meat Quality

Currently, the U.S. lamb industry pays almost exclusively on carcass weight basis; therefore, we have summarized the impacts of DDGS inclusion on carcass weight (Table 4). These data were summarized in the same format as previously described for DMI, ADG, and G:F in this manuscript. As with other summaries provided in this manuscript very few studies have reported carcass data with DDGS inclusion less than 20% and greater than 40% thus more data and research are needed to draw conclusive inferences about the impacts of DDGS on hot carcass weight at these inclusion rates.

Previous research feeding DDGS to finishing lambs has demonstrated that when DDGS were provided between 0 and 20% (DM basis) no differences in carcass characteristics were found (Schauer et al., 2005). While other research has demonstrated that loin eye area was greater in lambs fed 30% DDGS (16.8 cm²) compared to lambs not feed DDGS (16.1cm²; Schauer et al., 2006). Schauer et al. (2008) found that lambs fed DDGS had increased flank streaking in lambs compared to lambs not receiving DDGS. However, Schauer et al. (2008) reported no additional benefit to flank streaking when concentrations of DDGS was increased from 20 to 40 or 60% inclusion rates. Research feeding lambs diets containing 60% DDGS has

Table 4. Impacts of dried distillers grains with solubles (DDGS) inclusion on hot carcass weight (HCW, kg) of feedlot lambs.¹

| | DDGS, % | | | | |
|----------------------------|---------|------|-------|-------|------|
| | 0 | <20 | 20-30 | 30-40 | >40 |
| Huls et al., 2006 | 33.3 | - | 33.3 | - | - |
| Schauer et al., 2005 | 29.5 | 29.0 | 29.5 | - | - |
| Schauer et al., 2006 | 31.3 | 31.8 | 31.8 | - | - |
| Schauer et al., 2008 | 30.0 | - | 32.0 | 31.0 | 31.0 |
| McKeown et al., 2010 | 25.3 | - | 25.4 | - | - |
| Felix et al., 2012 | 29.9 | - | 32.5 | 29.1 | 28.7 |
| Crane et al., 2017 | 31.0 | 31.7 | 31.1 | - | - |
| Mean HCW ² , kg | 30.0 | 30.8 | 30.8 | 30.1 | 29.9 |

¹ Data utilized from published research articles utilizing corn DDGS in lamb rations. Studies included used a 0% control and at least one DDGS treatment.

² Mathematical average.

also demonstrated improvements in flank streaking with added S (Neville et al., 2010). Previous work reported that carcasses from lambs receiving 0.43% S, in the form of elemental S, graded better than carcasses from lambs fed either 0.13 or 1.3% S (Smith et al., 1964). Replacing barley and canola meal in the diet with 20% corn-DDGS did not impact fatty acid content of subcutaneous fat (McKeown et al., 2009). However, when DDGS was included as 45% of the diet meat fatty acids concentrations including C12:0, C18:3 n-3, and PUFA n-3 were decreased while C18:2 n-6, CLA c9-t11 and CLA t9-t11 were increased when compared to lambs fed 0% DDGS (Kawecka et al., 2018). Until there is a long-term commodity-based grid marketing structure in the lamb industry feeding DDGS at greater levels should be more a concern of economics related to feed cost than that of marketing value of carcasses.

Managing Excess Sulfur

The concerns over the incidence of sulfur toxicity in ruminants while feeding DDGS has received great attention over the past few decades as high dietary S can induce PEM in ruminants. Symptoms of PEM include: impaired coordination, blindness, and seizures which can be followed by death (NRC, 2005). As a result, DDGS has been associated with onset of PEM as the S content of DDGS is typically 0.3 to 1.7% S (Buckner et al., 2011; Kim et al., 2012; Drewnoski et al., 2014). In comparison, the maximum tolerable

level of S is 0.3% and 0.5% DM basis for high-concentrate and high-roughage diets, respectively (NRC, 2005), and are still the guidelines used today in beef cattle (NASEM, 2016). It is important to note that PEM is more appropriately a sign of toxicity rather than an issue related to maximum tolerable level and further delineation of S concentrations representing toxicity are needed. Outside of the issues with PEM, high dietary S may impact dietary copper and selenium absorption in small ruminants (NRC, 2007). Much of the research conducted with lambs fed high DDGS diets has focused on either S metabolism, adaptation to S, or management practices to offset potential negative consequences of increased S intakes (Neville et al., 2011; Felix et al., 2012; Felix et al., 2014).

The relative incidence rate of PEM in ruminants is relatively low, and multiple researchers (Schauer et al., 2008; Neville et al., 2010; Felix et al., 2012) did not observe PEM with diets in containing 60% DDGS, even though the diets provided by these researchers were in excess (0.35 to 0.87% S) of the maximum tolerable level of S (0.3% S; NRC, 2005). Other researchers had high incidence of PEM in lambs fed high S (0.72% S) and low-fiber diets not containing DDGS (Krasicka et al., 1999). Ruminal pH may be a major contributing factor related to onset of PEM (Felix and Loerch, 2011) due to the increased availability of free hydrogen ions needed for form hydrogen sulfide (Gould et al., 1997; Gould 1998; Kung et al., 2000). Ruminal pH increased and ruminal

hydrogen sulfide gas concentrations decreased with increasing roughage (rNDF) in steer diets containing 32% DDGS (0.44-0.47% S; Morine et al., 2014). Research in beef cattle has further demonstrated that risk of PEM is decreased as roughage NDF increases (Nichols et al., 2013). Sulfuric acid contained within DDGS has also been suggested to decrease ruminal pH (Felix and Loerch, 2011). The interactions of roughage, grain, S concentrations, and other management decisions need to be evaluated further as they relate to the onset of acidosis and PEM.

Providing high sulfate water in conjunction with feeding DDGS can further increase risk of PEM. High sulfate water (600 mg sulfate/L) may lead to sulfur toxicity in ruminants fed high concentrate diets (NRC, 2005 and 2007). To date, feeding DDGS in combination with low sulfate water (31 and 141 mg sulfate/L, Neville et al., 2011; Schauer et al., 2008, respectively) did not result in any cases of PEM in lambs. However, feeding DDGS and providing high sulfate water would not be advisable as the occurrence of PEM would be expected to increase. In areas with high sulfate water other sources of protein with lower S content would be advisable.

One of the main practices utilized when feeding greater inclusion rates of DDGS within the feedlot industry is to include thiamin in the diet. However, the subsequent review of literature demonstrates that providing thiamin does not guarantee prevention of PEM when feeding high S diets to ruminants. Visual signs of PEM in lambs fed high S diets (0.63% S) were prevented by supplementing 243 mg thiamin/kg dietary DM (Olkowski et al., 1992). However, these same authors reported that microscopic lesions in brain tissue were not completely prevented (Olkowski et al., 1992). Lambs fed 60% DDGS (0.55% S; DM basis) receiving 142 mg-hd⁻¹-d⁻¹ of supplemental thiamin had no incidence of PEM (Schauer et al., 2008). Other work did not determine any benefits to supplementing thiamin at either 50, 100, or 150 mg-hd⁻¹-d⁻¹ compared to unsupplemented lambs fed 60% DDGS (0.74 to 0.87% S; DM basis; Neville et al., 2010).

Additional research establishing more defined concentrations of S detailing both maximum tolerable level and toxicity in sheep is warranted. When

considering the relatively tight range between requirements 0.15% S (approximated from g/d S and estimated intake; NRC, 2007) and maximum tolerable level 0.30% S (NRC, 2005); and the apparent ability of lambs to perform within expectations at levels of S well above the maximum tolerable level understanding S requirements and metabolism in lambs is critically important. Further evaluation of the impacts of ration particle size, S concentration, and ruminal pH; quantification of ruminal available S, determination of S reducing bacterial populations, and the impacts of roughage concentrations are all needed as they relate to the incorporation of DDGS in lamb rations.

Applications

In conclusion, DDGS are a versatile feed well suited for use in rations for lambs. Feeding DDGS at rates up to 60% of dietary DM are possible, however, inclusion rates of 20-30% of the ration (DM basis) may be more appropriate. Carcass quality of lambs fed DDGS fall well within acceptable limits. Proper feed management and ration balancing are needed to address potential for mineral imbalances including phosphorus and sulfur. Additional research evaluating how ration particle size impacts intake, performance, and health would provide additional insight into feeding DDGS to lambs.

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Incorporating Dried Distillers Grains with Solubles in Sheep Supplementation Programs

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Summary

In the U.S., dried distillers grains with solubles (DDGS) is a highly available coproduct of ethanol and brewery industries utilized in ruminant diets that, due to the comparable nutritional quality, is capable of substituting (partially or totally) traditional sources of protein and energy with a possibility of reducing feed costs. With the use of this product, there is the potential to increase reproductive efficiency, survivability, growth performance, and meat production and quality when a well-designed supplementation program is implemented and monitored. However, the literature has scarce papers evaluating

the effects of DDGS in sheep nutrition, notably in supplementation programs. The lack of DDGS utilization in sheep diets may be related to some challenges, such as nutritional composition variability, handling, storage, and feeding issues. In addition to addressing these challenges, an overview of the statistics and concepts applied to the ethanol and sheep industry will be presented here, as well as the criteria involved in the incorporation of DDGS into sheep supplementation programs. These latter factors include the interaction between pasture and supplement type, along with feeding and herd characteristics.

Key Words: Costs, Energy, Meat, Protein, Reproduction

Introduction

Dried distillers grains (DDGS) with solubles, which are a coproduct of the bioethanol and alcoholic beverage industries, have become a commodity for the animal feed sector in recent years. Particularly in the U.S., almost all ethanol production uses corn as a feedstock. In this regard, national ethanol production increased from 1.4 billion gallons in 1998 to 9.3 and 16.1 billion gallons in 2008 and 2018, respectively (USDA, 2019a), corresponding a 12-fold increase over those 20 years as a result of specific policies (EPA, 2019; EIA, 2019).

The dry milled ethanol production process, present in 91% of the approximately 210 biorefineries across the U.S., generates DDGS as a coproduct. Recent data (RFA, 2019) indicated the production of 37.3 million metric tons of DDGS from ethanol plants, of which one-third are exported, plus 1.0 million tons from beverage distillers. The USDA forecast for the next 10 years (USDA, 2019b) projected an increase of ethanol production. Thus, the availability of DDGS for the feed industry seems to be secure in the next decade.

Sheep flocks reached a peak of 55 million animals in 1884 (Connor et al., 1921). Since then, the numbers have declined, and the profile has been changing toward small-scale operations (USDA, 2019c). Currently, all sheep and lamb inventories total 5.2 million head (i.e., 52nd place in the world rank, proportionally 0.44% of the 1.2 billion head), with 3.0 million ewes (FAO, 2017; USDA, 2019d). Although the sheep industry accounts for less than 1% of U.S. livestock industry receipts, sheep operations are important to the economies of several states, mainly in the southern plains, mountain, and Pacific regions (USDA, 2019c). Even though the number of animals has declined in recent decades, lamb and mutton meat imports have increased, accounting for more than half of the U.S. supply (75% from Australia and 24% from New Zealand), and wool imports surpass exports. This clearly illustrates the potential and necessity of organization and improvements within the national sheep industry (USDA, 2019c; USDA, 2020).

Efficient and low-cost nutritional

programs are important to reduce the effects of seasonality and offer to the market a suite of competitive and desirable sheep products. In this context, DDGS has been utilized in ruminant diets to replace partially or totally protein supplements and energy feeds. However, there are limited studies evaluating the effects of feeding sheep DDGS compared with other species, notably the utilization of this product in supplementation programs. The lack of DDGS inclusion in sheep diets may be restrained by a series of challenges that will be discussed in this paper. In addition, this article presents opportunities and how to incorporate DDGS into sheep supplementation programs.

Nutritional and Feeding Value of Dried Distillers Grains with Solubles

Despite the many advantages of using DDGS in sheep nutrition, there is a series of challenges, starting with the variability in chemical composition. Most ethanol in the U.S. comes from corn; however, sorghum, wheat, pearl millet, barley, and others also can be used as a source of starch for fermentation, which makes a considerable difference in the final DDGS chemical composition (Liu, 2012; Pedersen et al., 2014). Focusing on corn, the differences in varieties, geographic location, growth conditions (soil type, fertilizers, weather, etc.), degree of maturity, and harvesting methods are examples of factors that can affect the chemical composition of the grain and consequently DDGS composition (Kajikawa et al., 2012; USGC, 2018).

Additionally, differences in processing technologies among biorefineries influence the chemical composition, ruminal degradation, and nutritive value of DDGS (Jie et al., 2013; De Boever et al., 2014; Lee et al., 2016). Furthermore, new engineering technologies have been implemented that can affect DDGS composition, including 1) corn fiber separation for cellulosic ethanol production, 2) enhanced corn oil extraction methods, and 3) production of high protein (>40%) coproducts (USGC, 2018). The removal of extra corn oil, for instance, affects the nutritional profile of DDGS, primarily by reducing the crude

fat content, normally reducing the energy and increasing protein contents. As a result, the DDGS composition and nutritive value differ not only among plants but also among years of production from the same plant or even among batches (Belyea et al., 2010).

Thus, it is recommended to complete a chemical analysis of DDGS prior to use, because the actual values often differ from standard references (e.g., NRC) (Liu, 2012). Table 1 presents DDGS chemical composition data from corn ethanol plants.

The chemical composition of DDGS makes it a peculiar feed. It carries the potential to be included in animal diets to replace traditional sources of protein (e.g., soybean meal, cotton seed meal) and energy (e.g., corn, sorghum) due to the crude protein (CP) content (from 27 to 33%) and energy concentration (89.7% of total digestible nutrients, TDN; Nuez-Ortín and Yu, 2009), equivalent to 130% of the energy value of corn (Klopfenstein, 1996), as well as lower relative costs (Alshdaifat and Obeidat, 2019). The DDGS contains less CP and digestible protein but a higher TDN than cotton seed meal (Hoffman and Baker, 2011). Compared to sorghum grain, DDGS has lower non-fiber carbohydrates and higher fat, NDF, ADF, and CP contents (Trujillo et al., 2016).

For small ruminant production systems, the potential exists for DDGS to replace traditional and, most of the time, more expensive feeds (McEachern et al., 2009; Whitney and Braden, 2010). Furthermore, DDGS contains high levels of bypass protein (from 43 to 63% of total protein; Pecka-Kielb et al., 2017) and sulfur (0.55%), both of which have been shown to enhance growth and animal fiber production (Castro-Pérez et al., 2013). Additionally, DDGS utilization reduces acidosis, a frequent issue in high-grain diets (Klopfenstein, 1996), by virtue of the proportion of NDF (40.5%). However, DDGS is deficient in lysine, which is the first limiting amino acid; therefore, it is necessary to combine DDGS with some feed rich in lysine, such as soybean meal (SBM, Todorov et al., 2013).

No papers were found comparing dry lot with grazing situation in terms of lysine deficiency in sheep. There are works showing response to lysine supple-

Table 1. Chemical composition of dried distillers grain with solubles.

| Component | Average | Minimum | Maximum | CV (%) | Sources |
|------------|---------|---------|---------|--------|----------------------------|
| DM (%) | 90.9 | 88.9 | 92.2 | 2.4 | 1, 5, 7, 9, 10 |
| CP (%) | 35.3 | 26.9 | 33.1 | 5.6 | 1, 2, 3, 5, 6, 7, 8, 9, 10 |
| Fat (%) | 14.3 | 10.8 | 13.7 | 7.2 | 2, 3, 5, 6, 7, 8, 10 |
| NDF (%) | 40.5 | 26.6 | 58.9 | 23.5 | 1, 2, 3, 6, 7, 8, 9, 10 |
| ADF (%) | 14.8 | 7.00 | 23.7 | 30.4 | 1, 3, 5, 6, 7, 8, 9, 10 |
| Ash (%) | 4.63 | 2.00 | 6.70 | 31.9 | 1, 2, 3, 6, 8, 10 |
| Starch (%) | 5.23 | 5.10 | 5.50 | 3.6 | 2, 5, 10 |
| P (%) | 0.78 | 0.68 | 0.90 | 10.4 | 3, 4, 8, 10 |
| Ca (%) | 0.11 | 0.03 | 0.29 | 102.2 | 3, 4, 10 |
| S (%) | 0.55 | 0.33 | 0.84 | 39.4 | 5, 6 |

- 1 Cromwell et al. (1993).
- 2 Lodge et al. (1997).
- 3 Spiehs et al. (2002)
- 4 Batal and Dale (2003).
- 5 Belyea et al. (2004).
- 6 Belyea et al. (2010).
- 7 Kajikawa et al. (2011).
- 8 Olukosi and Adebisi (2013).
- 9 Gauthier et al. (2019).
- 10 Ranathunga et al. (2019).

mentation in dairy cows fed DDGS (French et al., 2010). However, the availability of additional methionine and lysine may not increase milk production in ewes, because increased methionine and lysine availability is preferentially used for plasma low density lipoprotein synthesis, resulting in changes in the saturated fatty acid profile of milk (McCoard et al., 2016). Lynch et al. (1991) did not find differences in intake, body weight (BW) or N balance when supplemented ewes in dry lot with lysine (diets had low and moderate CP levels). For ruminants receiving forage-based diets, deficiencies of amino acids are unlikely to be severe, unless the diets are based on grass silages (Titgemeyer and Loest, 2001).

The high-fat content of DDGS (14.3%) works well for supplementation programs, where DDGS enters a relatively low proportion of the total diet, and fat increases the energy value of the ration and increases feed efficiency (i.e., gain:feed). On the other hand, DDGS can be an issue when used in larger proportions, for instance, in fattening feedlot diets. A fat content over 8% can depress ruminal fiber digestion, reduce intake, and suppress performance, mainly when forage is the primary source of nutrients (Palmquist, 1994; Pezzanite

et al., 2010). The phosphorus (P) content in DDGS is high (0.78%). This characteristic can be an economic advantage, considering this element is the third most expensive nutrient in a diet, although P presence in excreta has been an environmental concern due to its pollutant potential (Liu, 2011). The calcium (Ca) content of DDGS varies considerably. In a given supplementation strategy, the mineral concentration of the supplement after mixing DDGS with other ingredients should be considered for the mineral formulation to meet the nutritional requirements of a specific flock and maintain an adequate balance among the minerals, notably the Ca:P ratio.

Dried distillers grains with solubles is known for its substantial sulfur (S) concentration. Sulfuric acids are commonly used in the dry-grind ethanol process to keep pH levels conducive to yeast cells that, in addition to natural corn and yeast sulfur concentrations, result in a byproduct that may contain considerable levels of this mineral. This can lead to concerns regarding pH, limitations in feed intake, and animal health (i.e., polioencephalomalacia) (Uwituzi et al., 2011; Morrow et al., 2013; Drenowski et al., 2014). In some parts of the U.S., such as rangelands in the West,

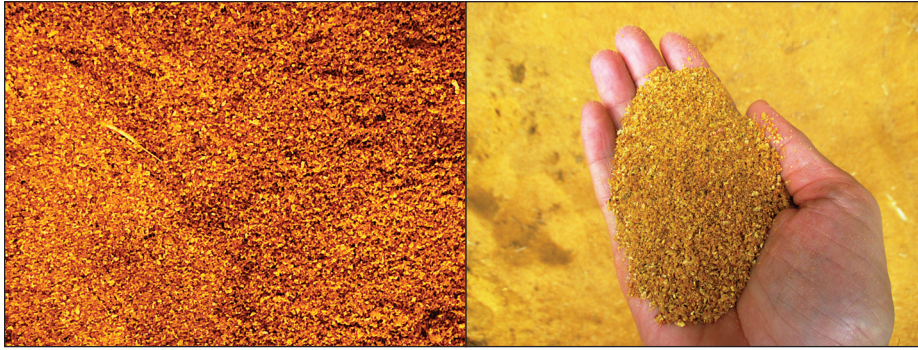
drinking water also can be a major source of sulfur, increasing the risk of the aforementioned problems. Thus, caution is recommended. Water tests determining S level can help the nutritionist to calculate how much DDGS could be used as supplement. Increased water consumption when animals are fed diets with high levels of S (Neville et al., 2011) can aggravate the concern if the water contains considerable levels of S.

For ruminants consuming high-concentrate diets, the maximum tolerable level of S is 0.3%, according to the NRC (2005). However, works using DDGS in feedlot lamb diets with levels of S up to 0.8% have not reported any related problems (Neville et al., 2010; Neville et al., 2011; Morrow et al., 2013). In fact, the proportion of concentrate in supplementation programs normally is lower than in finishing feedlot rations, notably for ewes.

Lambs fed diets with more than 0.6% of S are at risk of developing polioencephalomalacia (Morrow et al., 2013). To prevent this disease in sheep, some research groups have suggested the administration of thiamine when 25 to 50% DDGS is included in the diet (Pezzanite et al., 2010); however, Neville et al. (2010) stated that thiamine did not appear to be necessary when DDGS is less than 60% of the diet.

The nutritive characteristics (i.e., high energy, CP, P, and S) and low acquisition cost make DDGS attractive for feeding sheep on rangelands and pastures. However, DDGS, like any feed source, may contain mycotoxins depending on, among other factors, the levels in the original grain feedstock, because there is very little degradation of mycotoxins during ethanol production (Liu, 2011). Mycotoxins, which are secondary metabolites produced by fungi (i.e., molds), are very stable molecules. The ingestion, skin contact, or inhalation of these fungal metabolites can cause illness or even death by mycotoxicosis (Gallo et al., 2015). Although ruminants are less susceptible than nonruminant species, mycotoxins can affect sheep health in many ways (Mostrom and Jacobsen, 2011). Indeed, an assessment of the prevalence and levels of mycotoxins in DDGS in the U.S. generally found concentrations below the FDA regulations, which can fall well

Figure 1. Color variation of dried distillers grain with solubles.



Quadros, D.G. San Angelo, TX, 2019.

Quadros, D.G. Champaign-Urbana, IL, 2014.

below any harmful concentration when DDGS is blended with other ingredients to mix the total ration (Zhang et al., 2009).

Color can vary in DDGS from light to dark and yellowish to reddish (Figure 1). The variations in grain and methods used among ethanol plants, plus the complex interactions of many factors during the process within a plant, lead to great variations in the color score (Jie et al., 2013). Both particle size and color had little correlation with the composition of DDGS (Liu, 2008). Lightness or darkness also did not seem to be related to chemical composition, except for ADF and ADIN, in which darkness is associated with increased concentrations of these constituents (Cromwell et al., 1993).

Opportunities

Reducing Feeding Costs

The increase in feed costs has challenged profit margins in all livestock systems (Johnson, 2016). Thus, it is imperative to search for alternatives to offset feeding costs without sacrificing the performance and quality of the final products (Tjardes, 2002). Dried distillers grains with solubles has gained popularity as feedstuffs for animal nutrition because of availability, nutritive value, and costs. More than a source of energy, DDGS has a considerable density of protein, that is the most expensive fraction of a diet (Sahin et al., 2013). Given the substitutability of DDGS for corn and SBM, DDGS prices are significantly related to these feedstuffs (Langemeier, 2020). In the past, DDGS used to be

approximately 70 to 90% of the price of corn (Griffin et al., 2012). However, recent studies (Langemeier, 2020; Dennis and Erickson, 2021) showed prices of DDGS more expensive than corn 70% of the time in the last 5 years. Conversely, DDGS prices have always been below SBM. From 2007 to 2020, on average, SBM prices were 129% higher than DDGS (Langemeier, 2020). The costs of grazing forages also have increased. Therefore, supplementing DDGS in grazing situations may be profitable through increased market weight and decreased feeding costs (Morris et al., 2006; Griffin et al., 2012).

The feasibility of a supplementation program depends on both the cost of additional average daily gain (ADG) and the value of the additional ADG. However, the net return varies with DDGS cost and BW (Jerkins et al., 2009). Furthermore, the economic analysis of a supplementation program must measure the additional return and break-even on supplemental feed cost (Tjardes, 2002; Gadberry et al., 2010).

The use of DDGS likely will reduce feed costs, but the unitary cost (e.g., \$US per pound) is relative to the amount produced, which is related to animal performance. For instance, if DDGS in the diet improved growth performance and the animal could be fed for fewer days, it would result in decreased costs. Even if higher inclusions of DDGS decreased feeding values but still resulted in comparable or better performance than a corn-based diet, it may be economically advantageous because of decreased input costs (Erickson et al., 2012).

As the literature evaluating DDGS for sheep supplementation is very scarce,

many of our assumptions had to be taken from beef cattle. We found that DDGS has become a viable resource for supplementing growing cattle consuming forage-based diets. In the supplementation model based on pastures or hay (i.e., winter roughage supplementation), the introduction of DDGS has been economically viable, with a greater net return, probably because its inclusion in the diet increased performance and reduced the forage demand due to the higher content of nutrients in the DDGS compared to roughages such as forage or hay (Morris et al. 2006, Klopfenstein et al. 2008; Griffin et al. 2012).

The most prominent source of regional variation in DDGS utilization is the spatial dependence in its price relative to competing for feed ration inputs. Feed prices reflect supply and demand conditions and other region-specific factors unique to a market. Least-cost rations are formulated with the most cost-effective combination of inputs while meeting minimum nutritional requirements. The DDGS price and nutrient content determine its feeding value within least-cost ration formulas, and the volume of DDGS utilized in the diet is contingent upon its value concerning competing feed inputs (Johnson, 2016). When compared to commercial pellets, cubes, blocks, and tubs, the differential in favor of mixing a supplement on-farm with DDGS or to use it as a unique concentrate supplement might be economically attractive, despite the intensification in labor and extra feeding trough management. As DDGS prices fluctuate at the time of the purchase, extension agents and producers should evaluate local markets to compare its price with feedstuffs such as corn, sorghum grain, SBM, and CSM.

To obtain an accurate DDGS cost, one should not only consider the feed cost but also the final delivery cost, computing any additional costs associated with freight or storage. Hence, the size of the load and the distance of the supplier have an enormous influence on the final cost. Hauling small loads will increase the cost of the operation per ton of DDGS delivered. Shipping 24-ton loads per truck for short distances (up to 250 miles) is more common and cost-effective (Dooley and Martens, 2008).

In this context, for an owner of a

small sheep operation to reduce the costs of storage and mixing, one clever solution is to gather a group of neighbors, form a cooperative, and share a commodity barn and mixer. Mixing feed on site can result in substantial financial savings because it allows the use of mixtures with cheaper and regional ingredients, such as low-quality roughages as a source of fiber (e.g., hay, crop wastes), DDGS, minerals, and, depending on the category, grains to increase energy, and other protein feeds such as cotton seed meal.

Increasing Reproductive Efficiency and Lamb Production

It is known that nutrition before breeding and during gestation has a significant effect on the fertility of ewes and the development and survival rates of the resulting lambs (Robinson et al., 2006). The practice of increasing nutrient intake before and during breeding (“flushing”) can increase conception rates by increasing the number of eggs and the embryo survival rate (Shad et al., 2011). The utilization of DDGS in “flushing” supplementation can improve ovulation and fertility due to the high amounts of easily fermentable carbohydrates (energy) that provide fast access to glucose for follicular development as well as bypass protein, leaving large amounts of amino acids available to produce protein-based hormones such as growth hormone and insulin-like growth factor 1 (IGF-1) (Erdogan et al. 2018). In addition, the risk of acidosis in utilizing short-term feeding schemes with DDGS is lower than with other traditional sources of energy, such as corn and sorghum, due to the greater NDF content (Buckner et al., 2008). In rams, the inclusion of DDGS in their diets did not negatively impact reproductive traits (Crane et al., 2018).

Maternal nutritional regimens during different periods of gestation affect fetal development (Ford et al., 2007). Supplementation is expected to have a significant impact on enhancing fetal growth and survivability. For instance, high percentages of cereals increase rumen propionic acid, which is transformed into glucose in the liver and stimulates insulin secretion, which may increase the availability of nutrients required by the uterus for gestation (Harmon 1992; Radunz et al., 2011). Supple-

mentation with DDGS can not only benefit the fetus but also have a strong positive effect on ewe BW and body condition score (BCS) (Van Emon et al., 2014; Torreão et al. 2014). In addition, gestational feed costs may be significantly reduced for ewes fed DDGS (Radunz et al., 2011). It is worth a reminder that an inappropriate dietary regimen, along with the presence of two or more fetuses, may cause toxemia of pregnancy and other metabolic disorders (Sigurðsson, 1991; Van Saun, 2000).

In sheep flocks grazing in rangelands, maternal undernutrition is common and has been associated with low BW, low vigor, and high mortality in neonatal lambs, mainly in twin or triplet births (Mellor and Stafford, 2004; Ford et al., 2007). Concentrate supplementation enhances lamb strength and locomotor ability, avoiding a delay in consuming colostrum, which could increase the morbidity and mortality of lambs, especially in extensive systems (Pedernera et al., 2018). Moreover, providing ewes with a high-energy supplement during the final stage of pregnancy greatly increases the amount and viscosity of colostrum, which increases lamb survivability, particularly in those bearing twin lambs, in response to the circulating concentrations of hormones and metabolites that influence the provision of glucose and lactose synthesis (Banchero et al., 2004).

Undeniably, milk is essential for lamb survival and growth (Gently, 2010). Ewes consuming DDGS in their diets increased milk production due to the greater digestibility when DDGS replaced parts of barley grain and SBM in a conventional ration (Alshdaifat and Obeidat, 2019). Additionally, ewe milk yield has a positive effect on the lamb growth rate (Morgan et al., 2007).

Utilization of DDGS in creep-feeding supplements, which can increase weaning weights, thus helping the lambs attain feedlot weights faster, as well as in diets for drylot lambs on the ranch, are seen as opportunities for producers. In these situations, DDGS can replace (partially or totally) traditional and more expensive feed ingredients such as corn, sorghum, SBM, and cotton seed meal, reducing feeding costs without compromising growth performance and feed efficiency (Huls et al., 2006; Todorov et al., 2013; Crane et al., 2018; Hodges et al.,

2020). Consequently, DDGS is indicated for growing and fattening lamb diets, constituting an economical and palatable protein and energy feed ingredient (Sahin et al., 2013).

Gastrointestinal nematode infections negatively impact the health and performance of infected animals and the economic results of sheep production systems, notably on rangelands and pastures (Mavrot et al., 2015). Nutrition can affect the ability of the animal to contain, overcome, and cope with the consequences of parasitism (Coop and Kyriazakis, 2001). Supplementation with dietary protein can enhance the expression of acquired resistance and increase resilience to the pathogenic effects of major nematode parasites of the abomasum (e.g., *Haemonchus contortus* and *Teladorsagia circumcincta*) and small intestine (*Trichostrongylus colubriformis*) in young and mature sheep (Van Houtert et al., 1995; Steel et al. 2003; Turner et al., 2016). Protein-supplemented animals produced higher plasma levels of parasite-specific immunoglobulin A (IgA), which is the major immunologic mechanism of inhibiting worm development and regulating worm numbers (Stear et al. 1995; Strain and Stear 2001; Steel et al. 2003). Supplementation of grazing lambs with DDGS increased growth performance and reduced anthelmintic applications and the risk of anemia due to internal parasites (Felix et al., 2012).

Challenges

Handling and Storage

Compared to other feedstuffs, DDGS has some intrinsic physical and chemical properties that affect handling and storage, such as the propensity for poor flowability (i.e., the relative movement of bulk of particles), bridging (Figure 2), and caking (when macroparticles are incapable of independent translations) (Ganesan et al., 2008a; USGC, 2018). Particle agglomeration and caking during transportation results in increasing costs related to break the bridges, worker safety issues, vehicle damage, and economic losses (Bhadra et al., 2017).

Many factors affect the physical properties of DDGS: moisture content (which also influences microbial growth

Figure 2. Dried distillers grain with solubles (DDGS) “bridging up” in a commodity shed.



Quadros, D.G. Champaign-Urbana, IL, 2014.

and consequently feed safety); humidity (the hygroscopic properties of DDGS can lead to bridging, caking, and reduced flowability during transport and storage); temperature (the most drastic is freezing of the moisture to form ice bridges); pressure (the bulk may be subjected to compaction due to vibration); and solubles and fat contents (Ganesan et al., 2007; Ganesan et al., 2008a; Clementso and Ileleji, 2010; USGC, 2018). Regarding the fat content, the extraction of corn oil from DDGS has become a common practice in the corn ethanol industry. Low-oil DDGS had a lower average particle size and a narrower particle size distribution than regular DDGS, which indicates a higher probability of compaction but more uniformity of handling (Bhadra et al., 2017). Additionally, particle size and particle size distribution play significant roles in flowability and other properties, such as bulk density, angle of repose (angle between the horizontal and the slope of a heap of granular material dropped from some elevation), and compressibility (Ganesan et al., 2008a). Because of variability, the bulk density of DDGS ranges from 365 to 590 kg/m³ (Bhadra et al. 2009; Clementso and

Ileleji, 2010; USGC, 2018).

To overcome some of the handling and storage challenges of DDGS, anticaking and flowability agents have been researched, and adapted-design feeders have been developed. Flow conditioners and anticaking agents (e.g., calcium carbonate, zeolite) have been used as additives at low concentrations (up to 2%) to keep steady or increase the flow rate (Ganesan et al., 2008a), although some works (Ganesan et al., 2008b; Johnston et al., 2009) have found no advantage to use them for preventing flowability and caking issues. Pelleting is another approach that a few ethanol plants have attempted to improve bulk density and flowability; however, the additional costs related to infrastructure and equipment requirements, additional storage space, and labor have constrained this practice (USGC, 2018).

The material and design of storage bins and feeders also can interfere with DDGS flowability (Hilbrands et al., 2016). Furthermore, the storage bin ought to be designed for optimal feed flow to avoid bridging and caking issues. As a dry feed, DDGS requires relatively minimal storage facilities. Delivery, storage, and loading areas should be pro-

tected from wind or moisture. In practice, storage areas (old sheds or high-ground sites walled off with large hay bales) can be surfaced with hardened clay, gravel, blacktop, or cement and covered with tin sheets and/or heavy tarps. Caution should be taken when storing dry coproducts in upright bins due to settling and bridging. On the farm, the bulk storage of DDGS can be made in 22.7kg (50-lb) sacks, 2-ton bags, or commodity barns (Figure 3).

Feeding

Supplement is subjected to animal sorting. Sheep are known by their sorting capacity (more than cattle and less than goats or deer), which, together with other physiological adaptations, is very important for species adaptation in different environments, including harsh conditions (Preston and Leng, 1987; Van Soest, 1994). When in a heterogeneously mixed-species pasture (e.g., rangelands), they can exercise diet selection by choosing the most nutritive plants and parts of the plants by using unique foraging styles, learned perception, and prehensile capabilities (McFarland et al., 1992; Bartolomé et al., 1998; Pittarello et al., 2017). The ability to select certain portions of plants is extended to mixed rations to some degree, which may result in an unbalanced intake of nutrients (not consuming enough or overconsumption), reduction of the nutritive value of the ration, alteration of rumen fermentation, increased risks of rumen disorders (e.g., acidosis), and ultimately affects digestion efficiency and production (Miller-Cushon and DeVries, 2017; Sari et al., 2018). Processing (e.g., grinding) and mixing feed ingredients are common practices attempting to prevent sorting by animals and ensure that daily nutritional requirements will be satisfied (Zinn, 2004).

Another practical concern related to supplementation, notably in rangelands, is nontarget species consumption, including wildlife (e.g., hogs, birds, deer, bison, and raccoons) and other livestock species (e.g., cattle and goats). Livestock and wildlife may compete for feed resources (Ranglack et al., 2015; Schieltz and Rubenstein, 2016). Before choosing the supplementation management option, sheep raisers and managers should be aware of the potentially signif-

Figure 3. Bulk storage of dried distillers grains with solubles (DDGS) in 22.7kg (50-lb) sacks (a), 2-ton bags (b), or compartmentalized commodity sheds (c).



Quadros, D.G.
San Angelo, TX, 2020

Quadros, D.G.
San Angelo, TX, 2020

Quadros, D.G. Bell, FL, 2013

icant loss of supplemental feed by non-target animals (Lambert Jr. and Demarais, 2001). Feed consumption by other species will increase costs, reduce feed supply, and compromise the supplement plan and feasibility.

An additional challenge that can affect the predictability of sheep supplementation results is weather, i.e., wind, rain, and snow. Strong winds are typical in the U.S. central plains. Part of the roughage and other light ingredients can simply be blown by the wind. Rain can increase the moisture content, change the consistency, and reduce the intake of a given feed. The type and placement of feed troughs also can interact with rain influence. For instance, if the feeder is in a location favorable to water accumulation, the site easily can turn into a muddy area, and the normal trampling effects on soil density surrounding the feeder will increase the possibility of water accumulation and mud that can reduce supplement consumption and be a concern to animal health and welfare.

The feeder type (e.g., open, covered, autofeeder) varies in the degree of protection against the weather. All feeder types (grain, hay, and mineral) should be built off the ground to avoid sheep getting their feet inside or even laying in them. Additionally, the way the supplement is presented influences the susceptibility to wind and rain; as an illustra-

tion, mixed feeds are more impacted than pellets, blocks, or tubs.

A long distance between the feeder and the water trough can negatively affect feed intake, but in rangelands, they are normally close to each other to facilitate inspection and management.

The feeders need space for all sheep and/or lambs to be fed at once. Hand feeding and self-feeding should allow a minimum of 1.5 linear feet and 6 to 8 inches of trough space per sheep, respectively. Self-feeding allows more flexibility in managing time and labor, while hand feeding allows you to control the intake and inspect the animals more effectively (Craddock and Yeaman, 2012).

The effectiveness of a supplementation program can be affected by the individual intake variation, which is increased by excessive trough space, limited supplement allowance, self-fed supplements, feed and feed delivery equipment neophobia, and individual feeding of supplements (Bowman and Sowell, 1997).

Self-feeders can reduce labor. Intake limiters can be utilized to regulate the consumption. Sodium chloride (salt, NaCl) is commonly used as a limiter of supplement intake. Nel (1985) recommended the inclusion of salt in 25% of the supplement to restrict the intake by sheep. Other options of intake limiters

used for ruminant production are ammonium chloride, ammonium sulfate, calcium hydroxide, urea, animal fat, among others (Schauer et al., 2004; Sugg, 2013). However, works testing intake limiters for sheep are scarce. If amount of DDGS or a mixed supplement effectively consumed cannot be controlled by intake limiters, self-feeders will be more suitable for finishing diets where maximum feed intake is required to get sheep to a marketable weight.

Automated feeding systems have been developed to reduce labor, control the amount supplement, and optimize flexibility. However, there is still lacking validation works testing them in the field.

Sheep Supplementation

In rangelands, native shrubs mixed with grasses may provide adequate nutrient levels for grazing sheep production, except for copper, which is deficient in most months of the year, especially during fall and winter (Ramirez, 2003). However, forage may be insufficient to meet the nutrient requirements of ewes during late gestation and early lactation, therefore concentrate supplementation at a rate of 1% of BW is recommended to enhance the BCS of ewes and birth weight, survivability, and growth rate of lambs (Chaturvedi et al., 2003). Supple-

mentation is a strategic practice often intended to improve reproductive performance (“flushing”). For instance, short-term changes in protein supplementation around the time of mating, especially postmating, can have a beneficial effect on the estrus nonreturn rate, lambing rate to the first estrus, and litter size in ewes grazing on rangelands (Ocak et al., 2006).

Lambs commence to consume some forage in the pasture after the first couple of weeks and can be weaned onto solid food at an early age (Michalk and Saville, 1979). According to these authors, offering an exclusive differentiated supplement for lambs (creep feeding) may be used to increase lamb growth and reduce the competition of young lambs with their mothers since ewes will increase their liveweight at the expense of lamb growth. In addition, preweaning nutrition and gains might have a significant influence on postweaning performance and finishing liveweight (Bhatt et al., 2009).

Independent of the breed, supplementation is a key strategy for finishing lambs grazing on semiarid rangelands because the concentrate level is reflected in better growth performance and feed efficiency (Santra et al., 2002). Supplemented lambs had higher carcass yield, dressing percentage, and loin eye area than those managed on extensive rangelands only (Karim et al., 2007). Additionally, the inclusion of concentrate in the lamb diet improved the sensory quality of the meat, which was related to its effect on lowering the intensity of undesirable odors and flavors (strange, rancid, and acid), generating a higher intensity of typical lamb aroma and producing higher tenderness (Resconi et al., 2009). In addition, supplemental feeding can alleviate grazing pressure and conse-

quently maintain long-term grassland productivity, avoiding overgrazing and desertification (Bösing et al., 2014).

To implement a well-planned supplementation program in rangelands or established pastures, the first consideration is forage availability. If the forage is insufficient to attend the demand or it is unavailable (which becomes more of a dry lot scenario), hay, silage, baleage, or agricultural wastes should be provided.

Then, to recommend the proportion of DDGS and other ingredients in the feed mixture and the amount offered, the breed, size (weight), purpose (wool, meat), category of the herd (ewes, rams, lambs, weaners, wethers, hoggets, muttons), physiological state (pregnancy, lactation, flushing), and objective must be determined, because the animals will vary in terms of their nutritional requirements.

Interaction of Pasture Supplement

The interaction between the forage available (i.e., rangeland, grasslands, grass-legume mixed pasture, silage, hay, byproducts) and the supplement should be considered when a supplementation program is planned.

Rangelands are very significant in the American sheep production context. Rangeland is a land on which indigenous vegetation (predominately grasses, grass-like plants, forbs, or shrubs) is managed as a natural ecosystem (Mitchell, 2000). All rangelands present extreme spatial and temporal variability. Spatial variability occurs at scales ranging from the plant part to the regional level, while temporal variability ranges from a few seconds to a few years, resulting in a mosaic of patches characterized by fluctuations in forage quality and availability (O’Reagain and Schwartz, 1995; O’Rea-

gain and McMeniman, 2002). Above-ground net primary production (ANPP) varies among years in response to inter-annual precipitation variability in mesic and semiarid rangelands, while in desert rangelands, ANPP does not respond as much to precipitation pulses because plant growth is limited by inherently low leaf area and plant density (Poley et al., 2013). Conversely, animals raised in rangelands require a relatively constant intake of nutrients to satisfy the requirements of metabolism, growth, and reproduction (O’Reagain and Schwartz, 1995). Thus, seasonal deficiencies in nutrients (protein and/or energy) are frequent in both arid and high elevation rangelands (DelCurto et al., 2000), and supplementation is required to mitigate nutrient deficiencies and the effects of plant secondary metabolites (PSM) toxicity (Kawas et al. 2010).

When low-quality roughages are not limited in quantity, protein is generally the most beneficial supplemental nutrient (DelCurto et al., 2000). Forage CP levels below 7%, typical in the dry season (Table 2), result in decreased forage intake due to decreased ruminal microbial activity, which reduces digestibility and the rate at which the particles pass throughout the rumen (passage rate) (Poppi and McLennan, 1995).

Therefore, for sheep consuming dry pastures, the first limiting nutrient is usually rumen-degradable protein (RDP), especially in summer rainfall regions. Once RDP deficiencies are corrected, energy and possibly undegraded dietary protein (UDP) usually become the next limiting nutrients (O’Reagain and McMeniman, 2002). In contrast to protein supplements, energy supplements may decrease both the intake and digestibility of high-fiber low-quality forage; however, when the availability of

Table 2. Crude protein (CP) and dry matter digestibility (DMD) of North American rangelands in wet and dry seasons.

| Rangeland | Species | Forage type ¹ | Wet season | | Dry season | |
|-----------------------|---------------|--------------------------|------------|---------|------------|---------|
| | | | CP (%) | DMD (%) | CP (%) | DMD (%) |
| Temperate shrubland | Mixed grasses | G | 7.0 | 53.0 | 5.0 | 42.0 |
| | Mixed shrubs | S | 11.1 | 55.0 | 7.0 | 41.0 |
| Subtropical shrubland | Mixed grasses | G | 8.0 | 44.0 | 5.0 | 31.0 |
| | Forbs | F | 19.0 | 59.0 | 11.0 | 53.0 |

¹ G, grass; S, shrub; F, forb

Source: Adapted from O’Reagain and McMeniman (2002).

low-quality forage is limiting, energy supplementation becomes a viable alternative (DelCurto et al. 2000). In some cases, however, there is no effect or even increased utilization of grazed forage with energy supplementation, mainly when lower levels of supplementation are offered (Caton and Dhuyvetter, 1997).

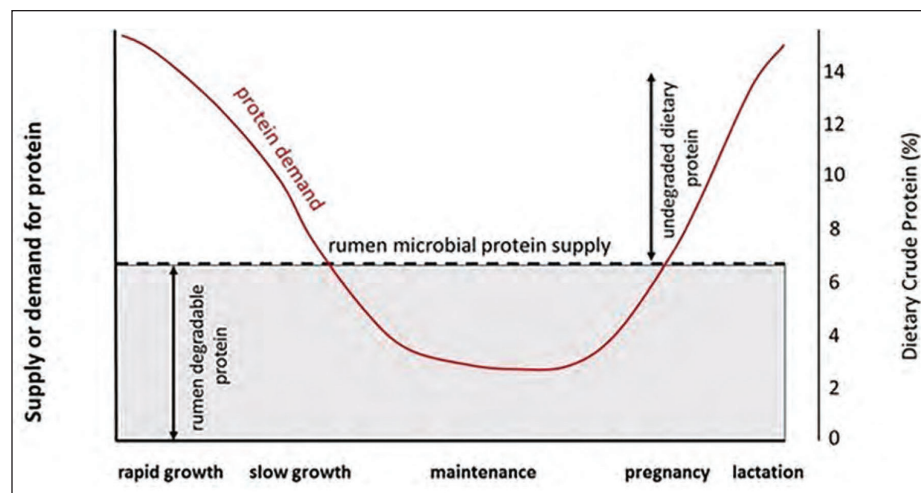
Feed Characteristics and Herd Category

To adjust the proportion of the ingredients in a supplement, the herd category and the physiological stage of the animals should be considered as well as the season of the year. Under extensive grazing conditions, supplementation programs to overcome seasonal nutrient deficiencies usually need to be low in cost and targeted at reproducing or young growing sheep. In summer rainfall regions, spring lambing is the preferred option, and autumn-joined ewes in later stages of pregnancy or lactating, frequently grazing RDP-deficient pastures, should be supplemented. Ewes in late pregnancy and lactating, grazing dry pastures with low protein concentrations, may respond to UDP and RDP supplementation. The utilization of energy supplements to maintain liveweight or support pregnancy and lactation is usually suitable for the beginning of drought conditions (O'Reagain and McMeniman, 2002).

Microbial proteins are synthesized from RDP or nonprotein nitrogen. Microbial protein is adequate for maintenance, slow growth, early pregnancy, and low milk production. Rapid growth, late pregnancy, and high milk yields (early lactation) require dietary protein that escapes ruminal degradation (Abbott, 2018), as shown in Figure 4.

Feeding ewes diets with higher TDN and CP during late gestation affects the performance of lambs at weaning (Torreão et al., 2014). Most likely, a greater concentration of metabolizable protein during the last third of gestation supports the improvement of offspring growth performance due to enhanced placental or mammary gland function (Van Emon et al., 2014). The supplementation of ewes during pregnancy results in a higher BCS at weaning, most likely because they can deposit more body fat reserves during lactation. At the end of pregnancy, the nutrient

Figure 4. Theoretical relationship between protein demand and protein supply in grazing sheep.



Source: Adapted from Kempton et al. (1978) by Abbott (2018).
[Used by permission of Dr. Kym Abbott, 2021].

demand increases, as 60% of fetal growth occurs during this period. At this stage, an inappropriate dietary regimen, along with the presence of two or more fetuses, may cause toxemia during pregnancy (Van Saun, 2000).

The energy intake of lactating ewes has important effects on the volume of milk produced, milk energy yield per day, and lamb growth (Wilson et al., 1971). During lactation, nutrition deserves more attention because ewes can be in three distinct phases of nutritional requirements. First, and this usually occurs during the first weeks postpartum, the ewe has a negative energy balance because milk production is increasing and intake has not yet reached its peak; thus, the animal mobilizes body reserves. Second, the energy balance was zero, as milk production declined, and females reached the peak dry matter intake. Finally, in the third phase, when the energy balance is positive, body reserves are replenished (Tedeschi et al., 2010).

When the goal is to obtain early spring lambs, supplemented ewes increased BW during the lactation period, while without supplementation, they lost weight, mainly among the ewes suckling lambs that were not creep-fed (Jordan and Gates, 1961). Increasing lamb performance from birth to weaning results in postweaning feed efficiency and reduces feeding costs when considering the entire production cycle of sheep meat (Galvani et al., 2014). Early creep

feeding showed a positive effect on BW gain and facilitated the transition from monogastric to a ruminant, buffering the weight loss of the lambs after the milk production peak of their dams (Abou Ward, 2008; Martínez et al., 2015). In addition, the supplementation of lambs by creep feeding can reduce the dependence on anthelmintic treatment (Melo et al., 2017).

In a supplementation program, it is important to consider the protein-energy relationships because animal performance depends mainly on the supply of amino acids (AAs) and energy-yielding substrates delivered to the tissues (Poppi and McLennan, 1995). There is evidence (Archibeque et al., 2008) that DDGS, as a supplement for forage-fed sheep, can improve the absorption of AAs. One step before that, however, is to provide substrates for rumen microbiota growth. Indeed, utilizing DDGS to formulate supplements for sheep is an opportunity to deliver rumen degradable (30–50%) and undegradable (50–70%) protein as well as a considerable concentration of energy and fiber (McEachern et al., 2009; Belyea et al., 2010; Castillo-Lopez et al., 2013; Gao et al., 2015). Urea is a low-cost alternative to be added to the supplement to increase CP and RDP contents (Golluscio et al. 1998). However, positive responses to urea supplements are more variable in grazing sheep than in cattle, partly because of sheep grazing selectivity.

Therefore, higher-quality diet intake provides enough RDP to support rumen fermentation. Furthermore, there is a large individual variability in supplement intake that can result in urea toxicity in some animals and variation in performance (Dove, 2002). Dietary supplementation with DDGS is safer than urea and increases rumen ammonia and total volatile fatty acid (VFA) concentrations in sheep (Radev, 2012).

Depending on the type and quantity of supplement offered and the quality of the forage available, the voluntary forage intake (VFI) can be reduced (substitutive effect), increased (complementary effect), or unaltered (additive effect) (Dove 2002; Kawas et al. 2010). Concentrate feeding stimulated the intake of low-quality forages, was additive with a medium-quality forage, and reduced the intake of a high-quality forage (Huston et al., 1988). Supplements increased VFI when the forage total digestible nutrients (TDN):CP ratio was greater than 7 (N deficit) (Moore et al., 1999).

The DDGS is classified as a protein feed. The true protein supplements also include plant protein sources, such as grain legumes (e.g., lupins, vetches), pulses (e.g., peas, fava beans), oilseeds and oilseed meals (e.g., whole cottonseeds, cottonseed meal, SBM, sunflower meal), plus animal protein sources, such as fish meal (O'Reagain and McMeniman, 2002). However, due to its energy content, DDGS also can replace partially or, depending on the animal category, totally, the energy high-carbohydrate feeds such as corn, sorghum grain, millet, barley, wheat, and oats. Research has shown (Wysocka et al., 2015) that DDGS is a source of inexpensive and highly available proteins. Most of the studies were conducted in feedlot, though. A recent review (Neville et al., 2021) provided an overview of research on growth performance and carcass characteristics and addressed some of the perceived barriers to increasing the use of DDGS in lamb feedlot rations.

Dietary additives (e.g., buffers, anabolic hormones, feed enzymes, synthetic amino acids, essential oils, and microorganisms) have been used to improve animal performance and efficiency, prevent certain diseases, and preserve feeds (Azzaz et al., 2015). The mode of action of feed additives is generally to manipulate the rumen fermentation environ-

ment, bring improvements in ruminant nutrition by increasing feed conversion efficiency and productivity, stabilize rumen pH to reduce acidosis risk, increase DMI, reduce methanogenesis, enhance rumen development and stability during dietary transitions, reduce pathogen load and shedding, improve meat quality, and buffer against dietary health risks (e.g., mycotoxins) (Frater, 2014).

The most important and widely used feed additives in ruminant diets are ionophore antibiotics (e.g., monensin, lasalocid, laidlomycin propionate, salinomycin, and narasin), but feed enzymes, probiotics (live microbial feed supplements), buffering agents, methane inhibitors, and many other additives are used depending on the situation (Mackie et al., 2002; Azzaz et al., 2015). Another additive that has been studied is polyethylene glycol. This substance can reduce the antinutritional effects of condensed tannins and improve the feeding value of many plants in a rangeland (Narvaez et al., 2011; Bailey et al., 2019). However, the effects of the inclusion of these additives in supplements for ruminants in rangelands or fed high-forage diets on animal performance or efficiency vary considerably (Huston et al., 1990; Kunkle et al., 2000; Piñeiro-Vázquez et al., 2009; Nagpal et al., 2015). Therefore, in a commercial sheep operation, their applicability must be carefully analyzed, case by case, by the nutritionist.

5.3 How to Incorporate DDGS into Sheep Supplementation Programs

The most economical way to incorporate DDGS into the sheep supplementation program at the ranch level is to purchase DDGS in bulk from a commodity broker, then store and mix feed on site

and feed each category of animal according to the nutrient requirements and performance objectives. In addition, the size of the business in terms of the number of animals, infrastructure, and investment capacity should be considered during the process of decision-making regarding the best management practices for DDGS in the production system.

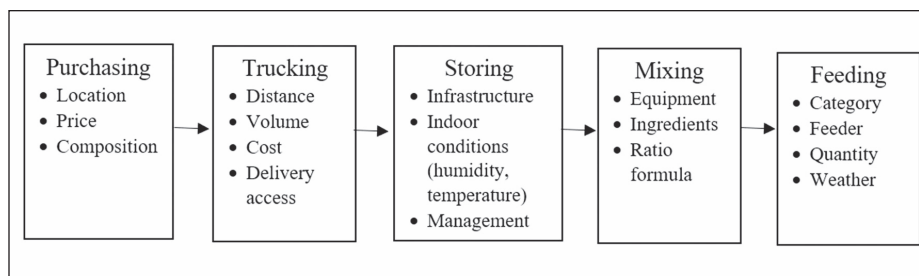
Figure 5 is a diagram that shows the process of incorporating DDGS in a sheep supplementation program, including purchasing, trucking, storing, mixing, and feeding.

Very limited studies were conducted studying DDGS in sheep supplementation programs. From a practical standpoint, DDGS can be included in supplementation programs for all sheep categories and physiological phases, such as ewes (breeding, gestation, lactation), rams, and lambs (preweaning and postweaning).

Ely et al. (1991) evaluated the utilization of 1/3 of the diet of DDGS vs. SBM to feed ewes with twin lambs from 14 to 56 days post-partum (using fescue-hay based diets) and discovered that ewes fed DDGS lost less weight and produced greater total milk fat per day. Distillers grain with solubles can be used as a protein supplement to low-quality forage, most likely during mid-gestation or when ewes are not pregnant (Pezzanite et al., 2010). According to these authors, levels of 0.5% to 1.0% of BW daily of DDGS can be fed to ewes consuming low quality forages, and during late gestation and lactation, DDGS can be used as a source of protein or energy depending on forage quality.

According to Held (2006), ewes fed a DDGS supplemented diet produced 16.5% more milk fat per day. Their lactation study, evaluating the use of DDGS to replace 2/3 of the corn, resulted in a

Figure 5. Diagram of how to incorporate dried distillers grain with solubles in a sheep supplementation program.



12% improvement in reared lamb growth for ewes nursing triplets. They also discovered that ewes fed DDGS had greater BCS at parturition and at weaning than those fed corn or haylage rations.

Radunz et al. (2011), comparing winter-feeding systems with haylage, limit-fed corn, or limit-fed DDGS (~1.2 lbs/day), reported the heaviest BW of ewes at parturition when DDGS was fed. Ewes fed corn and DDGS had greater BCS at parturition than haylage, and at weaning, ewes fed DDGS had greater BCS than those fed corn or haylage rations. Body weight of lambs at birth tended to be heavier from ewes fed corn and DDGS compared to haylage, but there was no effect of ewe gestation diet on lamb weaning weight. Body composition of lambs at birth, ewe milk production, as well as preweaning lamb growth rate and mortality were not affected by feeding program.

Van Emon et al. (2015), testing diets with different levels of metabolizable protein contain up to 43% of DDGS, observed DDGS supplementation during the last period of gestation had a strong positive effect on ewe BW and BSC, but minimal effect on lamb birth weight and development after birth.

Erdogan et al. (2018) found DDGS can be included as protein source in pregnancy rations up to 15% of DM to obtain reproductive performance outcomes equal to or exceeding those obtained with SBM. They also reported no significant differences in BW or BCS among the groups fed DDGS or SBM at either the start or the end of the flushing period, with no significant effect on lamb weight at birth.

Alshdaifat and Obeidat (2019), testing a diet with 50:50 roughage:concentrate ratio, with up to 30% of DDGS and approximately 2.4kg/day (5.3 lbs/day, 30% of DDGS) for nursing ewes for 8 wk, obtained increased milk production and no effects on milk composition with the increasing of DDGS in the diets, whilst being cost effective.

Knowing that supplemental feed costs is a significant factor in profitability and sustainability of rangeland sheep production systems, especially during winter months and periods of drought, we believe that 60% DDGS based supplement can be used for ewes with posi-

tive effect on pregnant ewe productivity, lambing rate, and health, and lamb weaning weight feed, while costs can be reduced by 30% compared to commercial pelleted supplements. For rams, based on the work of Crane et al. (2018), it expected no negative effects in the reproductive traits due to increasing DDGS in the diet at levels up to 45%. For lambs preweaning, creep-feeding at 1% increased ADG and minimized forage quality fluctuation (Santos et al., 2018). When used as a feedstuff for growing/finishing lambs, DDGS can be fed at a level of 25% to 50% of the diet dry matter (Pezzanite et al., 2010). After studying 2.5% of BW of DDGS for finishing lambs on pasture, Felix et al. (2012) observed DDGS-supplemented lambs had greater ADG (double) compared to the lambs that had not received supplementation, and DDGS supplementation reduced the percentage of lambs requiring treatment for internal parasites.

Final Remarks

Dried distillers grains with solubles is undoubtedly a great source of protein, energy, and other nutrients in ruminant diets, with possible cost-benefit advantages when compared with other traditional feed ingredients. Therefore, its inclusion in sheep supplementation programs is recommended, since the interactions among pasture, supplement, feeding characteristics, and herd category have been used to align the supplementation plan with the expected and measured reproductive efficiency and growth performance.

Currently, U.S. research efforts have focused on the effects of DDGS on ruminal fermentation, methane production, digestion, N balance, and animal performance. Another line of research is about the effects of drying and other DDGS manufacturing processes on RDP and RUP and the post-ruminal digestibility of RUP. Additionally, some research groups have evaluated how color can be used as an indicator of the nutritional quality of DDGS.

Upcoming U.S. research may evaluate how novel corn ethanol conversion processes can affect the uniformity and nutritional value of DDGS, for instance, high-protein and reduced-oil DDGS. Food safety has increasingly assumed a

key role in ruminant production systems. Consequently, methods to minimize the risks of spoilage during transit and storage have been developed, and research to understand how corn growing conditions can affect aflatoxin and other mycotoxins in DDGS has been conducted. Another trend is to study the potential solutions to overcome flowability problems, such as the effects of particle size, temperature when loading, moisture content, the proportion of solubles added to the grains, and the number of times DDGS has been handled and unloaded during transit.

Compared to beef and dairy cattle, DDGS has been insufficiently studied for small ruminants. In parallel, the producer's assistance is deficient in this field. Thus, more research and extension are necessary to develop feasible models of including DDGS in sheep diets to increase yield and reduce costs, contributing to amplifying the popularity and market of sheep products, notably lamb meat and processed meat products.

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Case Study: Effects of Supplemental Leucine in Milk Replacer on Pre- and Post-weaning Growth, Carcass Characteristics, Serum Amino Acids, and Visceral Tissue Mass in Lambs¹

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Summary

The objective was to determine the effects of leucine supplementation during the pre-weaning period on growth performance until slaughter. Nineteen commercial Dorset ram lambs (5.07 ± 0.15 kg) were used. Leucine was added to milk replacer at 0 (control) or 2.9% of DM and provided to lambs for ad libitum intake for 42 d using LAC-TEK automated milk feeders. Lambs were then fed a corn-based finishing diet and slaughtered in two blocks based on final body weight (BW). Data were analyzed using the MIXED and GLM procedure of SAS. Leucine supplementation increased ADG ($P = 0.007$) during the pre-weaning period. Because milk replacer intake was not measured during the pre-weaning period, it is difficult to conclude if effects of Leu on pre-weaning growth were influenced by differences in milk replacer intake. Final BW and ADG during the finishing period were not affected by pre-weaning Leu supplementation. Mass of the reticulorumen

tended ($P = 0.09$) to be greater in lambs supplemented with Leu pre-weaning but no other tissue masses were affected ($P \geq 0.39$) by pre-weaning Leu supplementation. Hot carcass weight and 12th rib fat thickness were unaffected by treatment. Thickness at the body wall was greater ($P = 0.05$) in lambs supplemented with Leu pre-weaning. Longissimus area, yield grade, quality grade, and percent boneless, closely trimmed retail cuts were not different between treatments. Results suggest that supplemental Leu to lambs fed milk replacer via automated feeders during the pre-weaning period increases growth in the pre-weaning period, especially in low birth weight lambs, without negatively affecting lamb performance in the finishing period. Additionally, Leu supplementation to lambs fed milk replacer may be useful to increase ADG of lighter weight lambs in the pre-weaning period.

Key Words: Developmental Programming, Leucine, Milk Replacer, Neonatal, Sheep

Introduction

Leucine is an essential amino acid (AA) and is required in diets to meet physiological needs, although in typical diets it is not believed to be limiting for production (Wu, 2009). Leucine has numerous effects on metabolism (Wu, 2009; Dodd and Tee, 2012; Millward, 2012) and is the primary AA signal for increasing muscle protein synthesis (Pedroso et al., 2015). Leucine supplementation to pre-weaned pigs has been shown to increase skeletal muscle protein synthesis (Escobar et al., 2010), mass of the longissimus dorsi, and BW (Columbus et al., 2015). Data are limited on the effects of supplemental Leu in pre-weaned lambs.

Lambs from twin or triplet births and twin lambs with a greater range in birth weights have been reported to have lower survivability during the pre-weaning period (Borg et al., 2007; Miller et al., 2010; Juengel et al., 2018; Notter et

al., 2018). Removing lambs with lower birth weights and rearing with milk replacer may increase lamb survivability and production later in life. Chai et al. (2018) reported that removing a twin lamb from its dam after 10, 20, or 30 days and reared on milk replacer until 60 days of age resulted in greater ADG than their siblings that remained with their dam. Additionally, Soberon et al. (2012) reported that increasing ADG during the pre-weaning period in Holstein heifers increased milk production during the first lactation suggesting pre-weaning programming of productivity later in life.

The objectives of this study were to determine the effects of supplemental Leu to lambs fed milk replacer using an automated feeding system during the pre-weaning period on ADG and serum AA during the pre-weaning period, DMI and ADG during the finishing period, visceral organ masses, and carcass characteristics of lambs. We hypothesized

that supplemental Leu to lambs fed milk replacer via automatic feeders would increase ADG during the pre-weaning period, ADG and gain:feed during the finishing period, and the cutability of carcasses.

Materials and Methods

Animals, Facilities, and Experimental Design

All procedures involving the use of animals were approved by the North Dakota State University (NDSU) Institutional Animal Care and Use Committee. Nineteen (n = 10 control, n = 9 Leu) neonatal fall-born ram lambs (5.07 ± 0.15 kg; twin-born n = 16, triplet-born n = 3) predominately of Dorset breeding were used. Lambs remained with ewes (2.8 ± 0.4 years old) for 12 h post-birth, so that lambs received colostrum, and then were randomly allotted to either a control milk replacer (Shepherd's Choice, Premier1 Supplies, Washington, IA, USA; n = 10; 8 twin lambs and 2 triplet lambs or the control milk replacer with 2.9% (DM basis) added Leu (n = 9; 8 twin lambs and 1 triplet lamb). Lambs were housed at the NDSU Sheep Unit and were fed using automated LAC-TEK Stainless 61450 milk dispensers (Biotic Industries, Inc., Bell Buckle, TN, USA; 1 per treatment;) which allowed for continuous ad libitum consumption of milk replacer. LAC-TEK machines were calibrated to deliver one part milk replacer (Table 1) and four parts heated water before the initiation of the experiment.

Lambs were allowed access to milk replacer for 42 d to provide a sufficient length of time to observe effects on growth performance. Individual intake of milk replacer was not measured making it difficult to conclude if treatment effects were because of the effects of Leu on milk replacer intake or other physiological effect(s). Water and starter feed, consisting of a creep feed and chopped alfalfa hay (Table 2), were provided for ad libitum intake when individual lambs reached 14 d of age. A partition within each pen was used to divide lambs that were less than 14 d of age from older lambs to reduce competition for the nipple feeders (nipple feeders separated along partition allowing a single LAC-TEC feeder to be used per treatment). On d 42, lambs were weaned and

Table 1. Dietary composition and nutrient concentrations of milk replacer and milk replacer supplemented with leucine (DM basis)¹

| Ingredient | Milk replacer | Milk replacer + leucine |
|-----------------------------|---------------|-------------------------|
| Milk replacer, % | 100 | 97.1 |
| Supplemental Leu | 0 | 2.9 |
| Nutrient Composition | | |
| Ash | 6.15 | 5.98 |
| Fat | 16.0 | 15.5 |
| CP | 24.4 | 26.6 |
| Ca | 0.990 | 0.962 |
| P | 0.728 | 0.707 |
| AA | | |
| Glu | 3.95 | 3.84 |
| Leu | 2.41 | 5.25 |
| Asp | 2.36 | 2.29 |
| Lys | 2.21 | 2.15 |
| Thr | 1.68 | 1.63 |
| Pro | 1.67 | 1.62 |
| Val | 1.58 | 1.54 |
| Ile | 1.53 | 1.49 |
| Ser | 1.25 | 1.21 |
| Ala | 1.14 | 1.11 |
| Phe | 0.977 | 0.949 |
| Tyr | 0.850 | 0.826 |
| Arg | 0.850 | 0.826 |
| Met | 0.648 | 0.630 |
| Gly | 0.573 | 0.557 |
| His | 0.563 | 0.547 |
| Cys | 0.467 | 0.454 |
| Trp | 0.446 | 0.433 |

¹ AA = amino acid; CP = crude protein; DM = dry matter.

removed from pre-weaning pens, comingled with weaned lambs from both treatments, and provided ad libitum access to water, creep feed, and chopped alfalfa hay.

After all lambs were weaned, lambs were moved to the NDSU Animal Nutrition and Physiology Center and penned in groups of four or five (0.91×2.4 m pens) in a temperature-controlled room (14°C) on Tenderfoot flooring (Tandem Product, Inc., Minneapolis, MN) for 39 ± 3.6 d. Lambs were penned in groups until individual lambs reached approximately 30 kg BW (backgrounding period; from weaning until beginning of finishing period) or 12 weeks of age, and then were penned individually (0.91×1.2 m pens) for the remainder of the experiment (finishing period) to monitor daily feed intake. Creep feed and chopped alfalfa hay were provided for ad libitum intake and lambs were transitioned to a finishing diet (Table 2) over 14 d by feeding (DM basis) 75% of the backgrounding diet and 25% of the finishing diet for 5 d, 50% of the backgrounding diet and 50% of the finishing diet for 4 d, and 25% of the backgrounding diet and 75% of the finishing diet for 5 d. The finishing diet consisted of 90% pellets and 10% chopped alfalfa hay (DM basis) and was formulated to meet or exceed nutritional requirements for growing lambs gaining 300 g/d (NRC, 2007). Lambs were provided the finishing diet at 5% of BW to ensure ad libitum intake, and feeding amounts were adjusted every 14 d based on lamb weight.

The five heaviest lambs from each treatment were selected for slaughter after $68 (\pm 3.4)$ days of feeding the finishing diet and all remaining lambs were slaughtered after $96 (\pm 3.6)$ days of feeding the finishing diet. Lambs were slaughtered in two groups to increase uniformity in carcasses between lambs and because of constraints in slaughter capacity. The five heaviest lambs from each treatment were selected for the first day of slaughter to assure similar days on feed between treatment groups and because BW is a more objective measure than visual assessment of fatness or degree of finish. Hot carcass weight (HCW) and dressing percent was determined after slaughter. After a 24-h chill, carcasses were knife-ribbed between the

Table 2. Nutrient composition of feeds provided to lambs (DM basis)¹

| Nutrient composition | Creep | | Finishing | |
|----------------------|---------------------|-------|---------------------|-------|
| | Pellet ² | Hay | Pellet ³ | Hay |
| DM | 89.1 | 89.7 | 88.0 | 87.7 |
| Ash | 6.87 | 8.80 | 4.31 | 9.33 |
| CP | 21.8 | 14.0 | 18.2 | 16.7 |
| EE | 3.38 | 0.77 | 3.18 | 0.815 |
| NDF | 13.5 | 57.7 | 11.1 | 52.4 |
| ADF | 7.70 | 40.3 | 3.27 | 37.9 |
| Ca | 0.767 | 1.04 | 0.686 | 1.08 |
| P | 0.434 | 0.295 | 0.330 | 0.284 |

¹ CP = crude protein; DM = dry matter; EE = ether extract.

² Creep pellet consisted of (DM basis) corn (46.6%), soybean meal (30%), beet pulp (19%), limestone (1.5%), urea (0.10%), and trace mineral salt supplement (2.8%).

³ Finishing pellet consisted of (DM basis) corn (86.1%), soybean meal (9.6%), urea (1.65%), limestone (1.1%), and trace mineral salt supplement (1.58%).

12th and 13th rib. Carcasses were evaluated for longissimus muscle area (LMA), fat thickness at the 12th rib (BF), body wall thickness (BWT), yield grade, leg score, flank streaking, and quality grade by trained personnel. Standard USDA grading procedures were used to derive a calculated yield grade. Percent of boneless, closely trimmed retail cuts (%BCTRC) was calculated, as described by Savell and Smith (2000).

Sample Collection

Lambs were weighed after birth and every 7 d until weaning. After weaning, lambs were weighed every 14 d through the backgrounding and finishing periods and 2 consecutive days before slaughter. Blood samples were collected via jugular venipuncture on d 1, 21, and 42 of the pre-weaning period at 1200 h for serum metabolite and AA concentration analyses. Blood samples were allowed to clot for 30 min at room temperature before being placed on ice and transferred to the laboratory. Serum was harvested by centrifugation ($3,000 \times g$ at 4°C) for 20 min, transferred to microcentrifuge tubes, and stored at -20°C until subsequent analysis. Samples of milk replacer (mixed with water), creep feed, chopped alfalfa hay, and finishing diet were sampled weekly. At slaughter, contents of the digestive tract were emptied and trimmed from the mesentery, and the mass and length of the small intestine, and mass of the reticulorumen, abomasum, omasum, colon, cecum, liver,

pancreas, spleen, visceral fat, and kidneys were recorded.

Sample analysis

Feed samples were thawed at room temperature and subsequently dried (60°C) in a forced-air oven for 48 h before being ground to pass a 1-mm screen. Feed samples were analyzed (AOAC, 1990) for DM, OM, N, and ether extract (EE). The techniques of Van Soest et al. (1991) were used to quantify neutral detergent fiber (NDF) and acid detergent fiber (ADF) non-sequentially. Crude protein concentration was calculated as $6.25 \times \text{N}$. Amino acid concentration of milk replacer was analyzed by high performance liquid chromatography after acid hydrolysis (AOAC, 1990). To assure accurate and precise analyses in our laboratory, samples are run in duplicate to ensure acceptable coefficients of variation and forage and concentrate control samples are run periodically to ensure consistency over time.

Serum glucose concentration was measured using the hexokinase/glucose-6-phosphate dehydrogenase method (Farrance, 1987) using the Infinity Glucose hexokinase kit (Thermo Trace, Louisville, KY, USA). Serum urea concentration was measured (Jung et al., 1975) using the QuantiChrom Urea Assay Kit (BioAssay Systems, Hayward, CA, USA). Serum free AA concentrations were analyzed by reversed phase ultra-performance liquid chromatogra-

phy after pre-column derivatization of AA with 6-aminoquinolyl-N-hydroxy-succinimidyl carbamate (Salazar et al., 2012; Lemley et al., 2013) and using an ethylene bridged hybrid C18 column (2.1 × 150 mm; 1.7 μm; Waters Corp., Milford, MA, USA).

Statistical Analysis

Pre-weaning and backgrounding data were analyzed as a completely randomized design. Lamb pre-weaning weight, serum AA, and serum metabolites were analyzed using the MIXED procedure in SAS (SAS 9.4, SAS Institute Inc., Cary, NC, USA) with repeated measures. The experimental unit was lamb. However, because lambs were offered milk replacer from a single feeder, it may be considered pseudoreplication rather than true replication. Because individual milk replacer intake was not measured, it is difficult to conclude if treatment effects were because of the effects of Leu on intake of milk replacer or some other physiological effect(s). The repeated effect was day, and the model included effect of day and treatment and their interaction. The REG procedure of SAS was used to examine how Leu supplementation influenced the relationship between birth weight and pre-weaning ADG. Tissue mass, initial and final finishing weight, and ADG during the finishing period were analyzed using the GLM procedure in SAS. The experimental unit was lamb, and the model included effects of treatment; days on feed during the backgrounding period was used as a covariate for finishing period data. Carcass characteristics data were analyzed as a randomized complete block design (slaughter date; n = 2 blocks). The experimental unit was lamb, and the model included effects of block and treatment. Significance was declared at $P \leq 0.05$ and tendency at $0.05 < P \leq 0.10$.

Results

Birth weight did not differ between treatments (Figure 1A). A treatment × week interaction was observed ($P < 0.01$) for pre-weaning BW with BW greater ($P \leq 0.04$) in lambs supplemented with Leu pre-weaning from day 7 until weaning. Average daily gain of lambs during the pre-weaning period (Figure 1B) was greater in lambs supple-

Figure 1. Body weight (kg; A; pooled SEM = 0.563) and average daily gain (ADG, kg/d; B) of lambs during the pre-weaning period. Week 0 corresponds to lamb weight at birth. Effects for body weight were treatment × week ($P < 0.01$), treatment ($P < 0.01$), and week ($P < 0.01$), and effect for ADG was treatment ($P < 0.01$). * indicates significance between treatments within day.

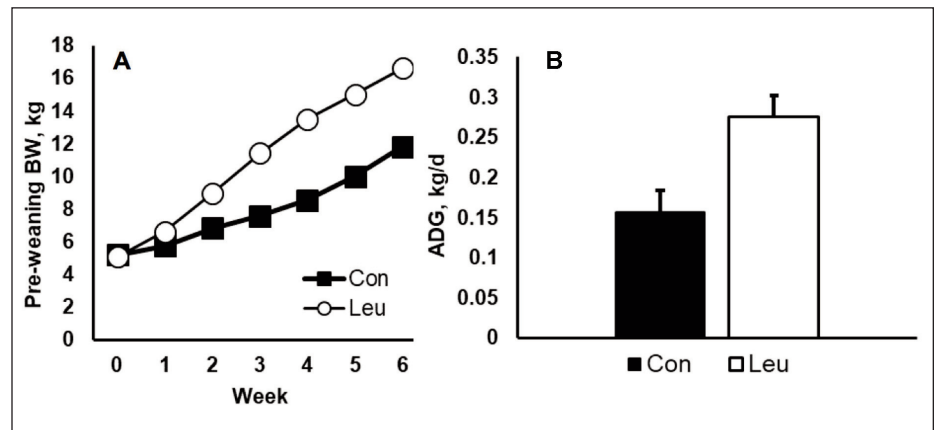
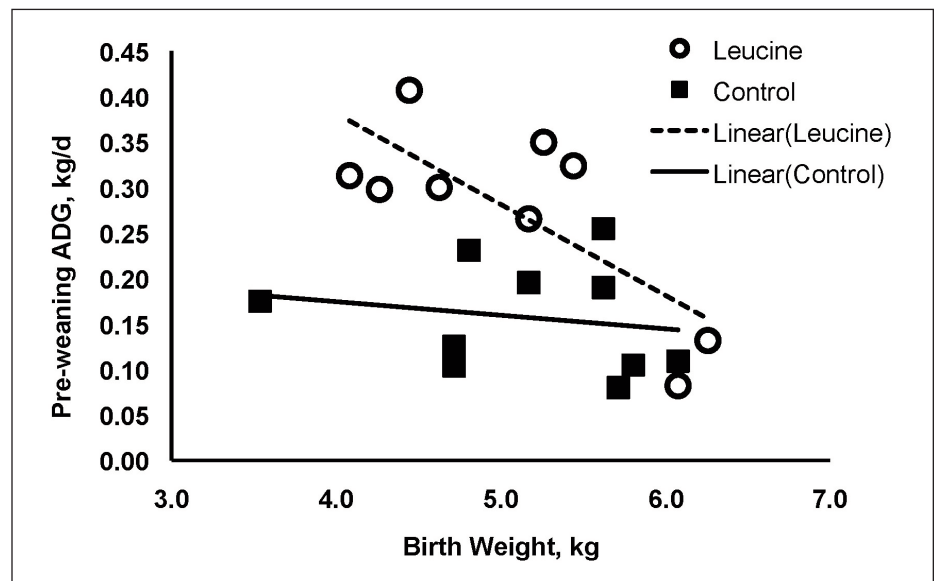


Figure 2. Linear regression of birth weight and pre-weaning average daily gain (ADG) for lambs during the pre-weaning period in Leu-supplemented and control lambs. Solid square and solid line are control, open circle and dashed line are leucine supplemented lambs. Linear regression of birth weight and pre-weaning ADG in control lambs: $y = -0.0149x + 0.2343$, $r^2 = 0.04$ ($P = 0.60$). Linear regression of birth weight and pre-weaning ADG in lambs supplemented with Leu pre-weaning: $y = -0.1001x + 0.7818$, $r^2 = 0.56$ ($P = 0.02$).



mented with Leu pre-weaning than control lambs ($P < 0.01$). In lambs supplemented with Leu pre-weaning, ADG decreased as lamb birth weight increased ($r^2 = 0.56$; $P = 0.02$; Figure 2) but in control lambs birth weight was not associated with ADG ($r^2 = 0.04$; $P = 0.60$).

No day × treatment interactions were observed for serum AA concentrations (Supplementary Table 1). Serum Leu concentrations were greater ($P <$

0.01) in lambs supplemented with Leu pre-weaning (Table 3). Lambs supplemented with Leu pre-weaning had greater ($P = 0.03$) serum Asp concentrations than control lambs. Serum concentrations of Arg, His, Ile, Lys, Met, Phe, Thr, Val, total EAA (essential AA), Ala, Asn, Gln, Glu, Gly, Pro, Ser, Tyr, total NEAA (non-essential AA) and total AA were not affected by treatment. A day effect was observed ($P < 0.01$) for

Table 3. Serum AA profile of lambs fed milk replacer with or without supplemental leucine during the pre-weaning period¹

| Item, μM | Treatment | | SEM | P-value |
|---------------------------|-----------|---------|-------|---------|
| | Control | Leucine | | |
| Arg | 255 | 255 | 22.2 | 0.98 |
| His | 144 | 143 | 9.97 | 0.91 |
| Ile | 84.7 | 94.7 | 10.23 | 0.50 |
| Leu | 167 | 308 | 23.6 | <0.01 |
| Lys | 160 | 180 | 10.8 | 0.22 |
| Met | 35.5 | 44.9 | 4.60 | 0.17 |
| Phe | 87.1 | 89.1 | 5.80 | 0.81 |
| Thr | 369 | 438 | 33.0 | 0.16 |
| Val | 294 | 281 | 22.4 | 0.67 |
| Total EAA | 1679 | 1911 | 101.8 | 0.13 |
| Ala | 233 | 240 | 9.7 | 0.61 |
| Asn | 88.6 | 95.5 | 5.73 | 0.40 |
| Asp | 13.0 | 15.2 | 0.69 | 0.03 |
| Gln | 161 | 151 | 12.8 | 0.60 |
| Glu | 240 | 270 | 18.4 | 0.27 |
| Gly | 517 | 476 | 28.7 | 0.32 |
| Pro | 224 | 234 | 15.9 | 0.67 |
| Ser | 135 | 150 | 8.1 | 0.20 |
| Tyr | 99.4 | 104.0 | 7.5 | 0.69 |
| Total NEAA | 1737 | 1732 | 46.7 | 0.94 |
| Total AA | 3416 | 3643 | 137.9 | 0.26 |
| Metabolites, mg/dL | | | | |
| Urea | 19.0 | 21.4 | 1.09 | 0.13 |
| Glucose | 94.1 | 107.0 | 4.46 | 0.06 |

¹ AA = amino acids; EAA = essential amino acids; NEAA = non-essential amino acids

serum concentrations of His, Lys, Met, Phe, Val, total EAA, Asp, Gln, Glu, Pro, and Tyr where serum AA concentrations increased over the experimental period (Supplementary Table 1). Serum concentrations of His, Ile, Thr, Ala, Asn, Gly, Ser, total NEAA, and total AA decreased ($P < 0.01$) from d 1 to 21, then increased on d 42. Serum concentrations of Arg increased ($P < 0.01$) from d 1 to 21, then decreased on d 42.

There was no effect of treatment on serum urea N but a day effect was observed ($P < 0.01$) where serum urea N decreased from d 1 to 21, then increased on d 42 (Supplementary Table 1). Serum glucose concentrations tended to be greater ($P = 0.06$) in lambs supplemented with Leu pre-weaning than control lambs (Table 3). A day effect was observed ($P = 0.02$) for serum glucose, where concentrations increased from d 1 to 21, then decreased on d 42 (Supplementary Table 1).

Days on feed for the backgrounding period was not affected by treatment

(Table 4) although Control lambs had numerically 24% greater (43 vs 36 days, respectively, $P = 0.11$) days on feed than Leu lambs. Control lambs had greater weight gain ($P = 0.01$) and ADG ($P = 0.05$) during the backgrounding period

Table 4. Backgrounding and finishing performance of lambs fed milk replacer with or without supplemental leucine during the pre-weaning period¹

| Backgrounding | Treatment | | SEM | P-value |
|--------------------------|-----------|---------|--------|---------|
| | Control | Leucine | | |
| Days on feed | 43.3 | 34.8 | 3.59 | 0.11 |
| Weight gain, kg | 13.7 | 9.8 | 0.98 | 0.01 |
| ADG, g | 320 | 284 | 12.2 | 0.05 |
| Finishing | | | | |
| Initial finishing BW, kg | 27.7 | 30.1 | 0.78 | 0.05 |
| Final finishing BW, kg | 56.3 | 59.3 | 1.50 | 0.18 |
| ADG, g | 374 | 374 | 14.1 | 0.99 |
| Days on feed | 81.1 | 82.5 | 1.47 | 0.53 |
| Daily feed intake, kg | 1.46 | 1.40 | 0.078 | 0.65 |
| Gain:feed | 0.284 | 0.282 | 0.0272 | 0.66 |
| Age at slaughter, d | 179 | 181 | 1.69 | 0.39 |

¹ ADG = average daily gain; BW = body weight.

compared to lambs supplemented with Leu during the pre-weaning period. Lambs supplemented with Leu pre-weaning were transitioned to the finishing diet at heavier weights than control lambs ($P = 0.05$). For the finishing period, final BW, ADG, days on feed, average DMI, gain:feed, and age at slaughter were not affected by Leu supplementation.

Supplemental Leu pre-weaning did not affect small intestinal length or mass at slaughter (Table 5). Mass of the reticulorumen tended ($P = 0.09$) to be greater in lambs supplemented with Leu pre-weaning. Supplemental Leu pre-weaning did not affect mass of the abomasum, omasum, colon, cecum, liver, pancreas, spleen, visceral fat, or kidneys on an absolute or on a g tissue/kg BW basis.

Hot carcass weight, dressing percent, and back fat at the 12th rib was not affected by treatment (Table 6). Body wall thickness was greater ($P = 0.05$) in lambs supplemented with Leu pre-weaning. Longissimus dorsi area, yield grade, percent of boneless closely trimmed retail cuts, leg score, flank streakings, and quality grade were not affected by treatment.

Discussion

Supplemental Leu has been shown to increase muscle mass and growth in piglets (Escobar et al., 2010; Columbus et al., 2015). Additionally, increasing ADG of Holstein heifers during the pre-weaning period increased milk produc-

Table 5. Tissue mass of lambs fed milk replacer with or without supplemental leucine during the pre-weaning period¹

| Item | Treatment | | SEM | P-value |
|-----------------|-----------|---------|--------|---------|
| | Control | Leucine | | |
| Small intestine | | | | |
| Length, m | 23.6 | 24.4 | 0.63 | 0.40 |
| g | 576 | 595 | 34.1 | 0.70 |
| g/kg BW | 10.4 | 9.98 | 0.628 | 0.66 |
| Reticulorumen | | | | |
| g | 1052 | 1169 | 45.5 | 0.09 |
| g/kg BW | 19.0 | 19.5 | 0.67 | 0.61 |
| Abomasum | | | | |
| g | 151 | 162 | 8.9 | 0.40 |
| g/kg BW | 2.71 | 2.73 | 0.164 | 0.92 |
| Omasum | | | | |
| g | 116 | 127 | 17.4 | 0.67 |
| g/kg BW | 2.13 | 2.13 | 0.309 | 0.99 |
| Colon | | | | |
| g | 425 | 449 | 38.2 | 0.66 |
| g/kg BW | 7.68 | 7.48 | 0.621 | 0.82 |
| Cecum | | | | |
| g | 48.9 | 57.6 | 6.90 | 0.39 |
| g/kg BW | 0.880 | 0.989 | 0.1310 | 0.57 |
| Liver | | | | |
| g | 965 | 1006 | 49.8 | 0.57 |
| g/kg BW | 17.4 | 16.9 | 0.88 | 0.68 |
| Pancreas | | | | |
| g | 65.0 | 59.8 | 5.97 | 0.55 |
| g/kg BW | 1.16 | 1.01 | 0.092 | 0.27 |
| Spleen | | | | |
| g | 90.5 | 98.0 | 4.07 | 0.21 |
| g/kg BW | 1.62 | 1.61 | 0.072 | 0.96 |
| Visceral fat | | | | |
| g | 2928 | 3014 | 250.0 | 0.81 |
| g/kg BW | 52.2 | 50.2 | 3.62 | 0.71 |
| Kidneys | | | | |
| g | 131 | 129 | 7.5 | 0.84 |
| g/kg BW | 2.34 | 2.18 | 0.129 | 0.39 |

¹ BW = body weight.

tion up to the third lactation (Soberon et al., 2012). However, data are limited on effects of supplemental Leu to lambs fed milk replacer via automated feeders during the pre-weaning period on pre- and post-weaning growth. Therefore, our objectives were to evaluate the effects of Leu supplemented pre-weaning on pre- and post-weaning growth of lambs, serum AA, visceral organ mass, and carcass characteristics.

In the current study, pre-weaning supplemental Leu to lambs fed for ad libitum intake using an automated feeders increased BW and ADG in the pre-weaning period, suggesting that Leu supplementation in pre-weaning lambs increases growth up until weaning at 42

days of age. Because lambs were group-fed and individual milk replacer intake was not measured, it is difficult to conclude if effects of Leu supplementation were mediated through changes in Leu intake or other physiological effect(s). However, it is unlikely that the magnitude of the observed positive effects on ADG could be mediated solely by differences in DMI. Future research is needed to monitor milk replacer intake in lambs supplemented with Leu. Our results differ from results of Mao et al. (2019) who reported no effects of Leu on BW or ADG in Hu lambs up to 30 d of age. Reasons for the discrepancy in results could be because lambs from the Mao et al. (2019) experiment nursed their dams

until 5 d of age, Leu supplementation started at 11 d of age which resulted in the supplementation period of only 19 d, and lambs were limit-fed. This may suggest that Leu needs to be supplemented earlier in life and/or for a longer time period to elicit effects on growth during the pre-weaning period.

Interestingly, the stronger relationship between birth weight and ADG in light-weight lambs supplemented with Leu in the current study suggests that increasing Leu concentration in milk replacers may be beneficial for lambs with low birth weights. Although it is commonly assumed that lambs with lower birth weights have decreased ADG during the pre-weaning period, Greenwood et al. (1998) reported that lambs with low birthweight have the capacity to grow at similar rates than heavier lambs when fed milk replacer. Also, Wardrop (1968) reported a positive relationship between birth weight and pre-weaning ADG in female but not in male lambs. However, previous research has shown that low birth weight twin or triplet lambs and twin lambs with a greater range of birth weights had lower incidences of survivability in the pre-weaning period (Miller et al., 2010; Juengel et al., 2018). Removing lambs with low birth weights or lambs from triplet births from ewes and feeding with milk replacer supplemented with Leu could result in increased ADG and survivability. Further research is needed to confirm or refute the role that Leu supplementation may have on growth of low birth weight lambs.

As expected, serum Leu concentration was greater in lambs supplemented with Leu pre-weaning. Our results are similar to those of Nair et al. (1992) and Cao et al. (2018) who reported an increase in plasma Leu in humans and calves when supplemented with Leu. Supplemental Leu during the pre-weaning period had minimal effects on serum concentrations of other AA, suggesting that Leu was not inhibiting uptake or utilization of other AA, or that other AA were not limiting for growth in lambs supplemented with Leu pre-weaning. Supplemental Leu did increase serum Asp concentrations in lambs supplemented with Leu pre-weaning. These results contradict those of Zheng et al. (2019) and Mao et al. (2019) who reported increases in serum Met, Thr,

Table 6. Carcass characteristics of lambs fed milk replacer with or without supplemental leucine during the pre-weaning period¹

| Item | Treatment | | SEM | P-value |
|-----------------------------------|-----------|---------|-------|---------|
| | Control | Leucine | | |
| Hot carcass weight, kg | 30.10 | 31.90 | 0.89 | 0.31 |
| Dressing percent | 54.0 | 53.3 | 0.51 | 0.33 |
| 12th rib back fat, mm | 9.70 | 11.1 | 0.922 | 0.34 |
| Body wall thickness, cm | 2.84 | 3.16 | 0.087 | 0.05 |
| Longissimus area, cm ² | 15.8 | 15.6 | 0.75 | 0.20 |
| Yield grade | 4.10 | 4.79 | 0.315 | 0.22 |
| % BCTRC | 44.7 | 43.6 | 0.47 | 0.12 |
| Leg score ² | 2.20 | 2.04 | 0.278 | 0.27 |
| Flank streaking ³ | 2.50 | 2.07 | 0.360 | 0.37 |
| Quality grade ⁴ | 1.90 | 1.76 | 0.198 | 0.45 |

¹ BCTRC = Boneless closely trimmed retail cuts

² Leg scores on scale 1-4; 1 = Low Choice, 2 = Average Choice, 3 = High Choice, 4 = Low Prime

³ Flank scores on scale 1-4; 1 = Slight, 2 = Small, 3 = Modest, 4 = Moderate

⁴ Quality grade on scale 1-3; 1 = High Choice, 2 = Low Prime, 3 = Average Prime

His, EAA, Gly, and Ser and increases in plasma concentrations of Ile and decreases in plasma concentrations of Ala and Met from calves and lambs, respectively, receiving supplemental Leu. These differences between studies could be because of differences in feeding management. Lambs in the current study were allowed ad libitum access to milk replacer, whereas calves from Zheng et al. (2019) and lambs from Mao et al. (2019) were meal-fed two or three times each day, respectively. El-Kadi et al. (2012) reported that continuous enteral delivery of milk replacer to piglets has limited effects on plasma AA, whereas bolus enteral delivery of milk replacer increased plasma AA concentrations. Boutry et al. (2013) also reported that bolus feedings increase EAA and NEAA concentrations in piglets compared to piglets that had a continuous orogastric infusion of milk replacer. It is possible that the lack of response in most of the serum AA is because of the continuous access to feed for lambs in the current study.

In the current study, lambs supplemented with Leu pre-weaning had a tendency for greater serum glucose concentrations. Leucine is unlikely to directly affect glucose production as it is a ketogenic AA (Voet and Voet, 2008). These results were not expected as Leu is known to increase insulin sensitivity in humans, which helps regulate circulat-

ing glucose levels (Zanchi et al., 2012). Previous research has reported no effects of supplemental Leu on glucose concentrations in pre-weaned piglets, calves, and lambs (Manjarín et al., 2016; Cao et al., 2018; Mao et al., 2019). Jarrett et al. (1964) reported that as lambs develop a functional rumen, glucose concentrations in the blood decreases. Although unlikely because control lambs had lower ADG, lambs on the control milk replacer could have had greater ruminal development, leading to lower serum glucose concentration than lambs supplemented with Leu pre-weaning.

In this study, an increase in total weight gain and ADG during the backgrounding period was observed in lambs fed the control milk replacer pre-weaning. The lambs likely experienced compensatory growth during this period, as at weaning they were 29% lighter than lambs supplemented with Leu pre-weaning. Similarly, Greenwood and Café (2007) reported that calves with lower weaning weights had greater ADG during the backgrounding period, when fed until a common age or weight, but ADG during the finishing period was unaffected. However, further research is needed to better understand how pre-weaning Leu-supplementation influences post-weaning growth in different growing and finishing systems.

During the finishing period, initial BW of lambs supplemented with Leu

pre-weaning was greater than control lambs; this was likely because Leu lambs reached the target weight more rapidly than control lambs. However, final weight of lambs supplemented with Leu pre-weaning was not different than control lambs. Lambs supplemented with Leu pre-weaning in this experiment attained a heavier weight at weaning, and numerically started the finishing period after a shorter backgrounding period (36 vs. 43 d) than control lambs. This resulted in a numerical increase in days on feed for the finishing period for lambs supplemented with Leu pre-weaning as lambs within both treatments were slaughtered after a common time on feed on one of two dates. Gain:feed often decreases as animals mature and deposit more fat than protein (Ferrell, 1988). The greater body wall fat thickness observed in lambs supplemented with Leu pre-weaning could suggest greater fat deposition later in the finishing period. More efficient feed conversion during the finishing period may have been observed in lambs supplemented with Leu pre-weaning if slaughtered at a common body fatness rather than common days on feed. This could suggest that the resulting increase in ADG during the pre-weaning period in lambs supplemented with Leu pre-weaning may be beneficial in reducing the number of days lambs spend on feed and attain market weight, without negatively affecting carcass yield and quality.

In conclusion, supplemental Leu to lambs fed milk replacer with automated feeders during the pre-weaning period increased lamb ADG by 75% at weaning and increased weaning weight by 5 kg, but the increased ADG did not persist through the finishing period. Because individual milk replacer intake was not measured, it is difficult to conclude if effects of Leu on ADG were influenced by differences in milk replacer intake. Additionally, Leu supplementation to lambs fed milk replacer may be useful to increase ADG of lighter weight lambs in the pre-weaning period, and produce lambs for harvest at a similar compositional endpoint in fewer days for finishing. Future research is needed to monitor milk replacer intake in lambs supplemented with Leu and to further study the potential role that Leu supplementation may have on growth of low birth weight lambs.

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Supplementary Material

Supplementary Table S1. Serum AA and metabolites of lambs fed milk replacer with or without supplemental leucine during the pre-weaning period¹

| AA, μ M | Treatments | | | | | | SEM ¹ | P-value | | |
|---------------------------|------------|------|------|---------|------|------|------------------|---------|-------|------------------|
| | Control | | | Leucine | | | | Day | Trt | Day \times Trt |
| | 1 | 21 | 42 | 1 | 21 | 42 | | | | |
| Arg | 153 | 371 | 240 | 176 | 338 | 252 | 33.5 | <0.01 | 0.98 | 0.80 |
| His | 295 | 67.1 | 71.2 | 273 | 73.2 | 82.1 | 17.3 | <0.01 | 0.91 | 0.61 |
| Ile | 96.8 | 77.1 | 80.2 | 115 | 67.5 | 102 | 14.8 | 0.01 | 0.50 | 0.26 |
| Leu | 226 | 147 | 127 | 334 | 337 | 253 | 37.4 | 0.13 | <0.01 | 0.31 |
| Lys | 142 | 140 | 198 | 158 | 168 | 213 | 20.9 | 0.08 | 0.22 | 0.93 |
| Met | 49.5 | 28.7 | 28.4 | 54.4 | 35.7 | 44.7 | 7.21 | 0.01 | 0.17 | 0.70 |
| Phe | 136 | 71.1 | 54.1 | 139 | 70.3 | 57.6 | 8.62 | <0.01 | 0.81 | 0.91 |
| Thr | 604 | 197 | 306 | 570 | 472 | 272 | 67.3 | <0.01 | 0.16 | 0.11 |
| Val | 459 | 222 | 202 | 498 | 175 | 169 | 32.4 | <0.01 | 0.67 | 0.54 |
| Total EAA | 2309 | 1368 | 1359 | 2379 | 1807 | 1547 | 176 | <0.01 | 0.13 | 0.57 |
| Ala | 394 | 141 | 162 | 425 | 133 | 161 | 15.8 | <0.01 | 0.61 | 0.57 |
| Asn | 145 | 49.9 | 71.2 | 151 | 66.5 | 69.1 | 10.3 | <0.01 | 0.40 | 0.68 |
| Asp | 17.7 | 10.0 | 11.2 | 24.2 | 11.0 | 10.5 | 1.31 | <0.01 | 0.03 | 0.06 |
| Gln | 62.3 | 20.9 | 17.6 | 22.3 | 22.0 | 18.5 | 20.6 | <0.01 | 0.60 | 0.28 |
| Glu | 471 | 132 | 118 | 542 | 116 | 152 | 25.5 | <0.01 | 0.27 | 0.10 |
| Gly | 537 | 319 | 697 | 471 | 363 | 592 | 52.2 | <0.01 | 0.32 | 0.36 |
| Pro | 439 | 108 | 124 | 433 | 127 | 141 | 20.8 | <0.01 | 0.67 | 0.94 |
| Ser | 241 | 68.1 | 95.0 | 251 | 85.2 | 113 | 11.5 | <0.01 | 0.20 | 0.96 |
| Tyr | 193 | 54.9 | 50.4 | 213 | 53.0 | 45.7 | 10.61 | <0.01 | 0.69 | 0.72 |
| Total NEAA | 2580 | 1082 | 1550 | 2524 | 1164 | 1508 | 96.0 | <0.01 | 0.94 | 0.80 |
| Total AA | 4888 | 2450 | 2909 | 4902 | 2971 | 3055 | 231.4 | <0.01 | 0.26 | 0.50 |
| Metabolites, mg/dL | | | | | | | | | | |
| Urea | 24.6 | 17.9 | 14.5 | 27.2 | 15.9 | 21.1 | 1.87 | <0.01 | 0.13 | 0.08 |
| Glucose | 82.6 | 101 | 98.5 | 93.7 | 126 | 101 | 7.43 | 0.02 | 0.06 | 0.14 |

¹ AA, amino acids; EAA, essential amino acids; NEAA, non-essential amino acids.