Nutritional strategies for growing cattle in the Southeastern United States

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ABSTRACT

Cattle operations in the Southeastern United States are primarily cow-calf and stocker based and rely heavily on forage. As such, research regarding supplementation strategies that are specific to both the forage cultivars and management strategies that are common in the region are necessary for accurate nutritional recommendations. Additionally, further research is necessary to develop solutions to cope with the negative effects of fescue toxicosis that are common in the region. Therefore, the objectives of this dissertation are to examine supplementation strategies specific to growing cattle as well as to investigate methods of reducing ergot alkaloid intake. Corn gluten feed (**CGF**) is a common corn co-product that is used to supply supplemental energy and protein. Experiment 1 investigated the effect of supplementing CGF or corn to grazing cattle and found that in grazing situations, growing cattle that are supplemented with CGF have greater average daily gain (**ADG**) and feed efficiency compared to those supplemented with corn ($P \leq 0.03$). This improvement in ADG was without changes in subcutaneous fat accretion ($P = 0.87$), as there were no differences among treatments in fat thickness at the 12th rib ($P = 0.54$). This improvement in growth and efficiency was thought to be due to differences in the digestibility of fiber in the rumen. Therefore, experiment 2 was designed to investigate the effect of corn or CGF supplementation on apparent total tract nutrient digestibility when cattle are fed a forage-based diet. However, the results from this experiment indicated that there were no changes in neutral detergent fiber digestibility ($P > 0.19$). In fact, heifers supplemented with corn at 0.25% BW had greater acid detergent fiber digestibility compared with those supplemented with corn at the same level ($P = 0.05$). Ultimately, this resulted in animals being supplemented with corn tending to have greater

organic matter digestibility compared to those supplemented with CGF ($P = 0.10$). Therefore, the improvement in performance that is observed when grazing cattle are supplemented with CGF compared to corn does not seem to be due to improvements in fiber digestibility. Experiment 3 examined if prolonged storage of ensiled tall fescue would result in the degradation of ergot alkaloids to levels similar to that of hay. Surprisingly, there were no differences among hay or haylage samples for individual or total ergot alkaloids regardless of storage time. Ergovaline concentrations, however, declined after 30 d of storage and were similar throughout the remainder of the storage period. Although no differences were observed among treatments, the reduction of ergovaline concentrations after 30 d of storage indicate that short-term storage may help to reduce the negative effects of fescue toxicosis for the animals consuming it.

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GENERAL AUDIENCE ABSTRACT

The Southeastern United States is home to approximately 20% of the country's cattle population and is a major source of feeder cattle that enter the feedlots in the Midwest. The primary nutrient source for cattle in this region is grazeable forage, and production systems in the Southeast are primarily cow/calf or stocker based, with few forage or grain-based finishing operations. Much of the grazeable area is in a portion of the country termed the fescue belt since the primary species of grass is tall fescue. While this species has desirable forage characteristics, it is commonly infected with a fungal endophyte that produces ergot alkaloids that are toxic to livestock. These ergot alkaloids are attributed to billions of dollars of lost revenue and reduced animal welfare.

While the forage in the Southeast is of relatively good quality, many producers choose to supplement grazing cattle. This is particularly common during winter months when forage is dormant, and when desired levels of growth exceed what can be provided by forage alone. However, the source of supplemental nutrients can induce changes in the efficiency of forage digestion, and the optimal nutrient source differs when compared to grain-based diets. Therefore, it is important for producers and nutritionists in the Southeast to have access to data relevant to their region of the country. The objectives of this dissertation are to evaluate supplementation strategies relevant for the Southeast, and to evaluate a potential method of mitigating the negative effects of fescue toxicosis in harvested forage.

Corn gluten feed has previously been shown to improve performance to a greater extent compared to corn when included in a forage-based diet in a dry-lot setting but has not been investigated in grazing situations. Therefore, we investigated if this effect would be present in

grazing situations. Using growing cattle and an automated feeding system, we demonstrated that grazing cattle supplemented with corn gluten feed had greater growth performance compared to those supplemented with corn. To investigate if this was due to differences in nutrient digestibility, we then investigated how total tract digestibility was influenced by supplement type and inclusion level in forage-based diets. However, our results indicated that at relatively small levels of inclusion, total tract digestibility is not impacted by supplement type.

To investigate methods to control fescue toxicosis in livestock, we conducted an experiment to determine if ergot alkaloid concentrations in forage harvested as hay and haylage can be reduced through prolonged storage. While there were no differences in concentrations among forage types (hay and haylage), there was a beneficial effect of day for ergovaline concentrations, one of the principal toxins that is present in endophyte-infected tall fescue. This suggests that profits for livestock producers as well as animal welfare can be improved through the short-term storage of harvested tall fescue.

Collectively, these experiments demonstrate potential methods for beef producers in the Southeastern U.S. to improve growth in grazing cattle. As researchers continue to improve the understanding of the interaction between grazing livestock and supplementation programs, improved recommendations can be made regarding nutrition programs and ultimately result in a more efficient and profitable operation.

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CHAPTER I

INTRODUCTION

Beef cattle production in the Southeastern United States is primarily forage-based and dominated by cow/calf and stocker operations. As such, the Southeast is an important supplier of feeder calves to the Midwest. Much of the grazeable land in the region is planted in toxic endophyte infected tall fescue, which has been estimated to occupy over 35 million acres (Bacon and Siegel, 1988). The plant has a symbiotic relationship with a fungal endophyte, *Neotyphodium coenophialum*, which lives between the plant's cells and produces ergot alkaloids that can be toxic to livestock (Arachevaleta et al., 1989; Pedersen et al., 1990). These ergot alkaloids have been estimated to cost the industry more than $3 billion annually, with $250 million of this in the fescue-belt alone (Kallenbach, 2015). Since the discovery of the toxic effects of the forage, producers and researchers have been searching for alternatives and solutions. Much of the benefit of tall fescue is its wide range of adaptability, drought and pest tolerance, resistance to grazing, and persistence. Therefore, when researchers attempted to remove the endophyte as a solution for fescue toxicosis, the survival of the plant was greatly reduced (Bouton et al., 1993). This led to the development of novel endophyte cultivars which contain the fungal endophyte required for adequate persistence, but do not produce compounds toxic to livestock. These cultivars have stand persistence similar to those grazed on toxic endophyte infected tall fescue, but animal performance is similar to that of endophyte-free cultivars (Hopkins and Alison, 2006). Despite the clear benefits of novel endophyte cultivars, renovations costs are estimated to exceed $600/ha, and producer acceptance has been slow (Kallenbach, 2015). Common methods to mitigate the negative effects of ergot alkaloids include dilution through supplementation or inter-seeding other species in the pastures, strategic management of cattle and pastures, and through the production of hay. However, these

methods still result in ergot alkaloid intake, and the threshold at which ergot alkaloids result in toxic effects has been estimated to be less than 100 μg/kg (Liebe and White, 2018).

It is common for producers to provide supplemental feeds to growing cattle to achieve greater levels of gains and efficiency. Although the forage in the southeast is generally of good quality, increased nutrient requirements and periods of reduced forage quantity or quality usually require the provision of supplemental nutrients. Producers commonly have access to a variety of supplemental feeds including corn grain, soybean hulls, dried distillers' grains plus solubles (**DDGS**), corn gluten feed (**CGF**), and various other co-product feeds. More recently, co-product feeds such as DDGS and CGF have become increasingly more common (Klopfenstein et al., 2008). This is due in part to the reduced cost compared to corn grain, as well as the improved growth and efficiency that has been shown when feeding DDGS to cattle consuming a forage-based diet (Loy et al., 2008). Despite this, CGF has received little attention in pasture situations, though it has been shown to be superior to corn in a hay-based diet (Hardin, 2018). However, this experiment was conducted under a dry-lot setting and these results have yet to be demonstrated in a pasture with grazing cattle. Because cattle production is much different in the Southeast compared to other areas of the country, it is important for those involved in the industry to have access to research conducted in pasture-based settings that are common in this area of the country. The evaluation of various feedstuffs in grain-based diets in confinement settings offers little insight as to their value in forage-based pasture settings. This is due to large differences in the microbial populations in the rumen of animals that are fed a grain-based diet compared to a forage-based diet (Fernando et al., 2010).

The objectives of this dissertation were to investigate nutritional strategies to improve the efficiency of beef cattle production in the Southeastern U.S. More specifically, this dissertation

will investigate the incorporation of corn co-products into the rations of growing beef cattle in pasture-based settings as well as some potential methods for reducing the impact of fescue toxicosis.

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CHAPTER II

REVIEW OF LITERATURE

The cattle population in the Southeastern United States is generally characterized by smaller operations that rely heavily on pasture and comprise approximately 20% of the national inventory (Drouillard, 2018). For purposes relating to this review, the Southeast is defined as Virginia, West Virginia, Kentucky, Tennessee, North Carolina, South Carolina, Georgia, Alabama, Missouri, and Florida. Recent census reports have estimated that the total cattle inventory of this region is 15.21 million, with the majority of those being beef cows and calves (NASS, 2019). While this area of the country has few feedlots, particularly when compared to the Corn Belt, the large beef cow population produces a significant supply of feeder cattle. Once weaned, these animals are usually backgrounded or stockered before being sent to a feedlot. Therefore, the Southeast plays a crucial role in the nations beef production system.

Not surprisingly, production systems in the Southeast are structured for cow-calf and stocker production and recent reports suggest almost all of the beef cattle in the region are housed primarily on pasture (Asem-Hiablie et al., 2018). Thus, the majority of the ration is composed of fresh forage with a large concentration of fiber. As forage availability can fluctuate throughout the year and desired levels of gain can exceed what can reasonably be expected from pasture consumption alone, many producers choose to provide supplemental feeds to cattle. These supplemental feeds can vary greatly in their nutrient composition, and many producers and nutritionists choose supplements based on cost, availability, ease of handling, nutrient profile, and palatability. The differences in nutrient content, as well as interactions between the supplement and forage result in substantial differences in the actual nutritional value of a given supplement.

The purpose of this review is to describe the inclusion of various supplements in growing cattle operations in the Southeast.

PASTURES AND GRAZING SYSTEMS IN THE SOUTHEAST

There is a great deal of climate diversity in the Southeastern U.S., ranging from tropical environments in the most southern regions to temperate environments in Appalachia (Ingram et al., 2013). As a result, the forage composition of pastures can vary greatly across states and commonly include cool and warm season grasses and legumes.

Common forage types

A large portion of the Southeastern U.S. lies in the fescue belt, a portion of the U.S. where *Schedonorus arundinaceus* (Tall Fescue) is particularly prevalent. It is one of the most important and common cool-season forages in the Eastern U.S. and occupies over 35 million acres (Bacon and Siegel, 1988). The widespread adoption of this forage has largely been due to its ease of establishment, tolerance to drought and pests, and reduced management inputs (Pedersen et al., 1990). These properties have largely been attributed to a symbiotic relationship with an endophytic fungus, *Epichloe coenophilala*. The fungus grows intercellularly, and the greatest concentrations of endophyte have been observed in the leaf sheaths and seed (Bacon and Siegel, 1988). The nutritional needs of the fungus are met by nutrients in the intercellular fluid of the plant (Bacon and Siegel, 1988), and the endophyte has been shown to impart a competitive advantage to the plant through growth stimulation, improved survival, and drought tolerance (Arachevaleta et al., 1989). In addition to these advantages, however, the fungus has been shown to produce a number of ergot alkaloids, several of which are toxic to livestock and result in a negative performance response (Shelby et al., 1997). This eventually led to the development of endophyte-free and non-

toxic, novel endophyte (**NE**) varieties. Because the endophyte is involved in the adaptability and competitiveness of tall fescue, endophyte-free varieties have been shown to have poor persistence compared to endophyte infected cultivars, but no differences in competitiveness have been shown between greater and lesser ergot alkaloid producing endophytes (Hill et al., 1991). As a result of the reduced competitiveness, the percentage of endophyte infection in low-endophyte plots has been shown to increase over time, particularly after the first few years following planting (Shelby and Dalrymple, 1993). The opposite is true for the NE cultivars, which have been shown to have similar stand persistence to that of toxic endophyte cultivars (Hopkins and Alison, 2006). Despite improvements in competitiveness and survival, the endophyte has not been shown to influence forage nutritive value and endophyte infected tall fescue has a similar nutrient profile to that of other cool-season grasses (Pedersen et al., 1990).

Ryegrass (*Lolium spp.*) is another common cool-season forage in the Southeastern U.S. Annual ryegrass (*Lolium multiflorum* Lam) is well-suited for the temperate climates of the Southeast, and approximately 90% of the 1.2 million acres of annual ryegrass can be found in the region (Cosgrove et al., 1999). It is a high-quality forage that can exceed 70% TDN and 18% CP as a fresh forage (Hancock et al., 2009). While harvesting annual ryegrass hay can be difficult in early spring, it can be successfully harvested as haylage with minimal storage DM losses (McCormick et al., 1998). Although commonly used in the Northeastern U.S., perennial ryegrass (*Lolium perenne*) is not well-suited for the southern portions of the Southeast because it generally does not survive the hot summers (Hancock et al., 2009). Similar to what is seen in tall fescue, the endophyte *Neotyphodium lolli* has a symbiotic relationship with perennial ryegrass and produces ergot alkaloids, particularly Lolitrem B (Christensen et al., 1993), that result in the neurological disorder commonly referred to as perennial ryegrass staggers (Cheeke, 1995). The disease is

characterized by incoordination, staggering, head shaking, and possible collapse, and affected animals usually return to normal a short time after removal from the pasture.

Legumes are commonly interseeded into pastures in the Southeast. Clover (*Trifolium spp.*), alfalfa (*Medicago sativa L.*), and sericea lespedeza [*Lespedeza cuneata* (Dum. Cours.) G. Don.] are commonly utilized alone, or in mixed-grass pastures (Hoveland, 1986). Legumes, and clover in particular, offer several benefits when interseeded in cool-season grass pastures. In general, legumes tend to have greater levels of crude protein and minerals, as well as lesser levels of fiber compared to grasses (Lee, 2018). The greater nutritive value of legumes will increase the nutrient supply to grazing animals, and mixed pastures have been shown to result in greater ADG and weaning weights relative to tall fescue pastures (Ellis et al., 1983). In addition to greater nutritive value, legumes are capable of biological nitrogen fixation. In some cases, this can result in reduced nitrogen fertilization, although the amount of nitrogen fixed by legumes can vary greatly depending on season and stocking rate (Rouquette and Smith, 2010). As U.S. fertilizer prices have increased drastically from historical averages (Huang, 2009), this has the potential to reduce input costs and increase overall profitability. Previous data have demonstrated that lespedeza by itself is not an adequate grazing crop, as animals demonstrated reduced intake and growth compared to a mixed-grass pasture (Henson et al., 1943). Despite this, the forage has been shown to have efficacy when interseeded in tall fescue forage systems for beef cows (Scaglia et al., 2008).

Although it is an important crop in the region, alfalfa production in the Southeastern U.S. is relatively low when compared to other area of the country. Alfalfa is widely recognized as having the potential for high yield, high quality forage that can be used for pasture, hay, silage, or greenchop. Aside from the aforementioned benefits of legumes, alfalfa is highly sought after and can be a valuable cash crop. In the Southeast, approximately 80% of alfalfa is used for hay with

the remainder being used for silage (Lacefield et al., 2009). Grazing fresh alfalfa pasture can result in bloat in ruminant animals, particularly if animals are deprived of hay and allowed to graze immature alfalfa stands (Cole et al., 1943). This is largely due to the development of foam in the rumen preventing eructation, which has been attributed to a property of the leaves (Pressey et al., 1963). Small gas bubbles become trapped in the rumen fluid, which then expands to fill the rumen and prevents the normal eructation of gas. When this condition is severe and no treatment actions are taken, the animal can die suddenly due to suffocation as the enlarged rumen prevents inhalation (Majak et al., 2003). Frothy bloat can be treated through the administration of anti-foaming agents, which breaks down the foam and allows the animal to expel the gas. Therefore, ruminant producers wishing to graze fresh alfalfa should be cautious. Previous research has shown that bloat can be reduced if the plant has begun to flower, cattle are moved onto the pasture in the afternoon, grazing is uninterrupted, and through the bloat prevention additive poloxalene (Majak et al., 1995).

Warm-season grasses provide producers with the opportunity for forage growth during the hot summer months when cool-season grasses are typically dormant. Bermudagrass (*Cynodon dactylon*) and bahiagrass (*Paspalum notatum*) are common perennial warm-season grasses used in the Southeast. Bermudagrass is a high-yield forage that is well suited for grazing or hay production (Hancock et al., 2009). It is marginal in quality during the growing season, and can have crude protein (**CP**) concentrations around 15%, and neutral detergent fiber (**NDF**) concentrations around 70% in July (Hill et al., 1993). Research has shown that it can be stockpiled to extend the grazing season, though supplementation is necessary to maintain weight and body condition during winter months (Wheeler et al., 2002). Bahiagrass is lesser in quality compared to bermudagrass, with samples commonly below 51% TDN and 11% CP (Moore et al., 1991); however, it will grow in pastures with soils too drained for bermudagrass, can be used in silvopastures due to its shade

tolerance, and is tolerant to grazing and drought (Hancock et al., 2010). These grasses are particularly well-suited for the southern regions of the Southeast and can be used to provide additional forage mass during summer months when cool-season growth can be limited.

Pearl millet (*Pennisetum glaucum*) and sorghum sudangrass (*Sorghum bicolor x S. bicolor var. sudanese*) are common warm-season annuals utilized in the Southeast. Pearl millet has a long history of use for summer grazing, although the recent development of grain varieties has resulted in its use for grain production (Lee et al., 2009). Although it is lesser in quality than many of the aforementioned forages with TDN generally less than 60% and CP less than 11%, it is high yielding even under moderate drought conditions (Hancock et al., 2009). Additionally, pearl millet can be grazed under conditions that result in stress to the plant without the risk of prussic acid poisoning, though it can be high in nitrate levels. Once consumed, nitrate is converted to nitrite by microorganisms in the rumen. Nitrite is then absorbed across the rumen wall and into the bloodstream, where it oxidizes the iron in hemoglobin to form methemoglobin (Vermunt and Visser, 1987). Methemoglobin cannot act as an oxygen carrier, which ultimately induces hypoxia and eventual death in extreme cases. Sorghum sudangrass generally has a greater nutritive value than pearl millet and under ideal conditions often has greater yield, though it is more severely affected by drought (Hancock et al., 2009). While it can be grazed, care must be taken to remove animals from sorghum sudangrass pasture following cutting, drought, or frost, as these conditions can result in increased concentrations of prussic acid (Vough, 1978). When consumed in sufficient quantities, this can result in prussic acid poisoning and eventual death.

Forage quality and quantity throughout the year

Forage quality is largely dependent on the plants stage of growth. It is generally accepted that as forage maturity increases, quality decreases. This is observed analytically by a decrease in

CP and TDN, and an increase in NDF and ADF. For cool-season grasses, such as tall fescue, the greatest quality would be in the spring and fall months when the plant is in the vegetative stage. The amount of bound nitrogen and lignin in the masticate of steers consuming tall fescue pasture was greater in November and December compared to May and June (McCracken et al., 1993). As would be expected, this resulted in decreased *in vitro* organic matter digestibility of the masticate, as well as decreased organic matter intake by the steers. However, it should be noted that forage quality was moderate in all cases and CP concentrations did not decline past 13%. Similarly, Cherney et al. (1993) demonstrated a sharp decline in forage quality, as well as a sharp decrease in both the rate and extent of organic matter digestion of several species of cool-season grasses as forage matured in the summer months. Interestingly, there is evidence of differences in the rate of quality decline among different species of cool-season grasses. Collins and Casler (1990) showed that tall fescue and reed canary grass declined more rapidly with maturation than Timothy or smooth bromegrass as evidenced by *in vitro* dry matter digestibility. Similarly, Marten and Hovin, (1980) found that the rate of decline in *in vitro* dry matter digestibility and CP concentrations was greater in reed canary grass relative to tall fescue and orchard grass. As reed canary grass also had the greatest yield, it is likely that the increased growth rate led to a faster accumulation of stems which resulted in a faster rate of quality decline. As would be expected from the decreased digestibility associated with more mature forages, the digestible and metabolizable energy content of cool-season grass and legume hay was shown to sharply decline as forage maturity increases (Welch et al., 1969).

Forage quantity throughout the year is largely dependent on environmental conditions. For cool-season forages, the majority of growth occurs in the spring and fall with little growth during the summer and winter months. For example, Denison and Perry (1990) showed that tall fescue

and orchard grass have accelerating growth rates between May and June and September and October in the Appalachian Plateau, with almost no growth observed in August. While forage growth resumed in the early fall, it should be noted that this growth was considerably less than what was observed in the spring. Warm-season forages have the greatest growth during the summer months, with little growth the rest of the year. Burton et al. (1988) demonstrated that costal bermudagrass had the greatest yield in June and July, which was dependent on daylength, solar radiation, and minimum temperature.

Fescue toxicosis and its implications on beef production in the Southeastern U.S.

As previously mentioned, toxic endophyte-infected (E+) tall fescue is a major forage for beef producers in the Southeastern U.S. However, the effects ergot alkaloids have on livestock can be quite drastic and is estimated to result in over $1 billion in losses annually (Strickland et al., 2011). The negative effect ergot alkaloids have on growth has been well documented, though there are instances where reports fail to demonstrate negative effects of ergot alkaloid intake. The reasons for this vary, but are likely due to a lack of heat stress as well as animal tolerance to the toxins. Paterson et al. (1995) found that ADG across a number of experiments was reduced by 30 - 100% for steers grazing E+ tall fescue compared with low-endophyte tall fescue. Interestingly, the reduction in gain due to consumption of ergot alkaloids appears to be more significant in the spring compared to the fall (Parish et al., 2013). A number of mechanisms have been proposed to describe the reduced growth attributed to ergot alkaloid consumption. Several ergot alkaloids present in E+ tall fescue have long been known to result in vasoconstrictive effects (Solomons et al., 1989). More recent work has indicated that consumption of ergot alkaloids results in vasoconstriction of mesenteric vasculature, which may result in reduced nutrient uptake due to reduced blood flow to and from the small intestine (Egert et al., 2014). Other experiments have

shown a similar effect in the rumen, where ergot alkaloids reduced ruminal blood flow resulting in reduced VFA flux (Foote et al., 2013). Therefore, vasoconstriction of the mesenteric blood vessels may explain the reduction in digestibility (Matthews et al., 2005) and ultimately growth that is observed when cattle consume E+ tall fescue. Perhaps related to vasoconstriction is a decrease in voluntary DMI when cattle consume E+ tall fescue (Matthews et al., 2005). While control of intake can be complex, the reduced ability to dissipate heat that occurs when cattle are exposed to ergot alkaloids can result in heat stress (Aldrich et al., 1993), which is known to decrease intake. This effect is particularly apparent during summer months when poor heat dissipation is compounded by increased ambient temperatures, ultimately resulting in reduced DMI and growth. Exacerbated heat stress from ergot alkaloid consumption is likely the reason why ergot alkaloids have a greater effect on growth in the spring compared to the fall. Additionally, recent results have indicated that steers consuming E+ fescue seed had reduced concentrations of growth hormone compared to steers grazing endophyte-free seed (Bostian et al., 2016). Growth hormone is a hormone secreted by the pituitary gland that antagonizes the effects of insulin, resulting in the partitioning of nutrients to lean tissue (Bauman, 1999). Indeed, growth hormone is known to increase nitrogen retention (Davis et al., 1970) and result in increased ADG and protein deposition (Machlin, 1972). Additionally, growth hormone stimulates the production of insulin-like growth factor 1 (**IGF-1**) (Lucy, 2008), which promotes cellular growth throughout the body. Therefore, a reduction in the concentration of circulating growth hormone is likely to lead to reduced growth and lean tissue accretion.

Aside from the negative effect on growth, ergot alkaloid consumption is implicated with reduced animal welfare and health. Perhaps one of the most significant is the effect ergot alkaloid consumption has on incidence of heat stress in cattle. Animals consuming ergot alkaloids through

the diet have been shown to have elevated core body temperatures compared to those not consuming ergot alkaloids when ambient temperatures exceed the thermoneutral zone (Al-Haidary et al., 2001). Rhodes et al. (1991) demonstrated that blood flow to the rib skin is reduced by ergot alkaloids, resulting in greater inefficiency at dissipating heat. This can be observed in the skin temperature during heat stress, which has been shown to be depressed in animals consuming ergot alkaloids due to reduced blood flow and ultimately heat transfer (Gadberry et al., 2003). In addition to reduced blood flow, heat stress is thought to be exacerbated by a rough hair coat in the summer. Cattle consuming ergot alkaloids have been shown to have thicker hair coats in the summer, which contributes to the overall increase in core body temperature and respiration rate (Nihsen et al., 2004). Researchers have assumed this was due primarily to the inability to efficiently shed winter coats, however, more recent research has challenged this idea. Aiken et al. (2011) clipped and bleached hair on steers consuming E+ tall fescue in June and July to determine the effect of ergot alkaloids on hair growth rate. The authors found that the majority of hair regrowth grew in the summer months and to excessive lengths. Therefore, it is possible that rough hair coats are due to an inability to regulate hair growth rather than an inability to shed the winter coat. Aside from its implications in heat stress, vasoconstriction from the consumption reduces blood flow to the extremities. During times of extreme cold during the winter, this reduces the animal's ability to maintain thermal homeostasis in the extremities, which can result in lameness and necrosis of the hooves.

At present, few data are available regarding the effect of ergot alkaloid consumption on immune function. Poole et al. (2019) observed that steers with chronic exposure to low to moderate concentrations of ergovaline had increased concentrations of peripheral pro- and anti-

inflammatory cytokines. This was indicative of increased immune responsiveness, and when combined with other stressful factors such as weaning, may predispose animals to disease.

Endophyte-infected tall fescue has been and continues to be a serious economic problem for producers in the Southeast. In the early 1990s, Hoveland (1993) estimated that E+ tall fescue resulted in greater than $600 million in losses across the U.S. After taking into account the equine and small ruminant industries, this value was estimated to exceed $1 billion in 2011 (Strickland et al., 2011), and $3 billion in 2015 (Kallenbach, 2015). Researchers have pushed pasture renovation to NE cultivars, but producer acceptance has been slow due to costs associated with renovation, the inability to use land being renovated, and uncertainty of persistence (Caldwell et al., 2013). Anderson et al. (2019) reported that beef cows grazing E+ tall fescue had a 5% reduced calving rate and 9% reduced pregnancy rate following artificial insemination compared to cows consuming NE infected tall fescue. Assuming all calves born were weaned and a price of $3.09/kg for a 272 kg steer calf, this equates to a $42 loss per cow exposed. This results in greater than $360 million of losses in reproductive efficiency alone across the approximately 8.6 million beef cows and replacement heifers in the Southeast. The reduction in ADG observed when growing animals consume E+ tall fescue can vary dramatically, but researchers have proposed a 39 g to 47 g reduction for every 10% increase in endophyte infection rate (Liebe and White, 2018). Assuming a 50% infection rate, a 100-d grazing duration, and a 47 g reduction in growth, grazing stocker cattle on E+ tall fescue can cost producers 23.5 kg in lost weight gain per animal. With a sale price of $3.00/kg for a 363 kg steer, greater than $120 million are lost annually across the approximately 1.7 million head of growing cattle in the Southeast. Therefore, it is likely that the total economic losses attributed to fescue toxicosis exceed $500 million annually for producers in the Southeastern United States.

Mitigating the negative effects of tall fescue

The most effective way to cope with the effects of tall fescue toxicosis is to replace all pastures affected by the toxic endophyte, but this is expensive and is not usually a practical solution. Because of this, current recommendations focus on managing pastures and animals to limit ergot alkaloid intake. Ergot alkaloid concentrations have been shown to be considerable greater in the seed heads compared to the leaf blades (Rottinghaus et al., 1991). Therefore, producers can limit ergot alkaloid intake by controlling the amount of seed heads in E+ fescue pastures (Kallenbach, 2015). By chemically suppressing seed heads in E+ tall fescue, grazing steers had greater ADG and serum prolactin concentrations compared to those grazing untreated pastures (Aiken et al., 2012). Some of this improvement is likely due to improvements in forage nutritive value when the plant is kept in the vegetative stage, as treated pastures were shown to have greater CP, water soluble carbohydrates, and *in vitro* digestible dry matter concentrations.

Other strategies to mitigate the negative effects of ergot alkaloids on animal performance include management strategies to limit ergot alkaloid intake during critical periods in the production cycle. Ergot alkaloids in tall fescue have been shown to vary considerably throughout the year (Rottinghaus et al., 1991; Kallenbach et al., 2003), and coupling periods of reduced ergot alkaloid concentrations with critical periods such as breeding can improve performance. For example, Caldwell et al. (2013) showed that calving rate can be nearly doubled by rotating spring calving cows to NE tall fescue pastures 28 d prior to the breeding season and grazing them there for 4 to 6 weeks, and calving rates were similar to spring calving cows grazing NE pastures year-round. However, this management strategy was much less beneficial for fall calving cows, which in general, had greater calving rates compared to spring calving cows. Therefore, producers who have no grazeable NE tall fescue pastures may benefit through the use of fall calving programs,

and those who have a small area of NE pastures can prioritize its utilization to the breeding season to improve calving rates without incurring significant costs associated with the renovation of all pastures.

Perhaps one of the least disruptive to management strategies already in place is ergot alkaloid degradation through hay production. In many cases, beef cattle producers already have practices in place to harvest forage as hay to then feed during the winter months. Ergot alkaloids are relatively unstable molecules and are sensitive to acids, bases, light and air (Garner et al., 1993). During the curing process in hay production, clipped grass is exposed to sunlight as it dries. Research has shown that during this time, the levels of TEA and ergovaline decline tremendously (Roberts et al., 2009). In fact, ergot alkaloid concentrations continue to decline during storage, but to a much lesser degree. Because the majority of this decline occurs during the curing process, it is likely that the largest driver of ergot alkaloid degradation is the exposure to sunlight. During Spring when weather conditions are variable, producers may opt to ensile harvested forage to preserve forage quality rather than waiting until weather conditions are suitable for hay production, when the forage is more mature. However, because the forage is dried to a much lesser extent compared to hay, ensiled forage is exposed to considerably less sunlight. Previous reports have demonstrated that ergot alkaloid concentrations do not decline to the same extent in ensiled tall fescue compared to hay (Roberts et al., 2002; Roberts et al., 2014). Because of this, current recommendations are not to ensile E+ tall fescue. However, these reports have not investigated the decline of ergot alkaloids during the storage period after ensiling. Research has shown that various anaerobic microorganisms are capable of detoxifying various ergot alkaloids (Perumbakkam et al., 2006). Combined with reduced pH compared to hay, it is possible that increased storage time will lead to greater ergot alkaloid degradation over time.

In addition to the degradation of ergot alkaloids during hay curing, ammoniation of hay will further reduce concentrations (Roberts et al., 2002; Craig et al., 2011; Roberts et al., 2011). Steers that were fed ammoniated Kentucky-31 tall fescue straw had improved feed intake, ADG, and serum prolactin concentrations compared to those fed Kentucky-31 straw (Kallenbach et al., 2006). However, ammonia will improve the quality of poor-quality forage by breaking down bonds between lignin and cellulose, so it is likely that some of the improvement noted during this experiment was due to increased nutrient digestibility rather than reduced ergot alkaloid content. Nevertheless, the reduced ergovaline concentration in the ammoniated straw combined with greater serum prolactin concentrations of steers suggest that a reduction in fescue toxicity likely played a role.

SUPPLEMENTATION OF GRAZING BEEF CATTLE

Common supplement sources for producers in the Southeastern U.S.

Producers in the Southeast have access to a variety of supplements to encompass a wide variety of feeding situations. Because the forage species generally tend to be moderate to high in quality, energy is usually the first limiting nutrient in beef cattle production systems in the Southeast. However, some instances such as winter feeding, exceptionally poor-quality forage, or increased levels of production may require protein supplementation. Traditionally corn has been the principal source of supplemental energy. It is high in energy, easily digestible, and accessible for most producers. However, price increases in the early- to mid-2000s that coincided with the expansion of corn processing industries led to a shift toward the use of co-product feeds as energy supplements. Particularly in the Southeast, the presence of nearby corn sweetener plants provides many producers with cheap, plentiful access to corn gluten feed (CGF). Corn gluten feed is a fibrous product that is high in energy, moderate in CP, and in many cases, is a cheaper method of

providing supplemental energy to grazing cattle. Similar to CGF, dried distillers grains plus solubles (DDGS) may be an available supplement source for producers. As a co-product of ethanol production, it is high in energy in the form of fiber, and moderate in CP. However, DDGS may not be a viable supplement for all producers as the majority of ethanol plants are in the Midwest and trucking costs may be cost-prohibitive in some cases. It is common for many local co-operative associations to produce a blended supplement of several ingredients, usually composed of a blend of corn, CGF, DDGS, and soybean hulls. For many producers, these blends may be the easiest method of supplementation.

Growing cattle supplementation

Much research has been conducted evaluating the supplement type and inclusion level in growing cattle diets. Unsurprisingly, the optimum type and level depend largely on particular production practices. As previously mentioned, corn is a common energy supplement for growing cattle and has long been shown to improve ADG when supplemented to grazing animals (Hess et al., 1996; Pavan and Duckett, 2008). However, research has shown that fibrous energy sources produce similar improvements in performance compared to grain supplementation when fed to cattle consuming a forage-based diet. Anderson et al. (1988) demonstrated that soybean hulls have an energy value equal to corn when supplemented to grazing steers. Literature is mixed regarding the effect of soybean hull supplementation on forage intake, as some data show minimal impact on hay DMI (Martin and Hibberd, 1990), while others show a decrease in forage intake (Richards et al., 2006). In contrast, corn grain supplementation was shown to result in a substantial decrease in forage DMI when supplemented to grazing cattle (Pavan and Duckett, 2008), which negatively impacts pasture utilization. As soybean hulls contain energy in the form of highly digestible fiber, ruminal fermentation dynamics may not be negatively impacted through supplementation and

allow for more efficient use of forage. This was shown by Richards et al. (2006) who supplemented steers consuming freshly clipped tall fescue. Animals supplemented with soybean hulls had greater total tract organic matter disappearance and flow of nitrogen to the duodenum. Additionally, rumen pH was not changed through supplementation, which is desirable because a decrease in rumen pH can reduce the efficiency of fiber digestion. This could partially explain the minimal impact on forage intake, as impaired fiber digestion would result in slower passage rate and therefore reduced intake. Supplementation with similar fibrous energy sources such as wheat middlings has also been shown to improve weight gain, though it should be noted that forage intake was significantly depressed (Heldt et al., 1998). In many cases, supplementation through fibrous energy sources results in a greater improvement in gain compared to starch sources for cattle consuming a forage-based diet (Gadberry et al., 2015).

Although the protein content of forage is usually sufficient in many of the forages utilized in the Southeast, it may become limiting during winter months. Recent research has shown that ADG can be improved in heifers grazing tall fescue from November to January when supplemental protein is provided (Lyons et al., 2016). Towards the end of December CP concentrations dropped below 10% and may have been limiting growth, which would explain the improvement in growth with supplementation. Although it cannot be determined from their experiment, it is possible that the improvement in growth was due to an increase in the supply of rumen undegradable protein (RUP). Poor to medium quality forages have the potential to have growth-limiting concentrations of RUP, and supplementation with a source of RUP will increase the flow of protein and ultimately amino acids to the small intestine. MacDonald et al. (2007) demonstrated a slight improvement in growth when heifers consuming smooth bromegrass (13% CP) were supplemented with increasing

amounts of RUP. Despite this, a meta-analysis showed that supplemental protein, in general, does not improve ADG when cattle graze tall fescue (Gadberry et al., 2015).

The optimal level of supplementation for grazing cattle depends largely on the type of supplement and base diet. However, considerations should be given to maximize pasture utilization and not depress forage intake to the point that the supplement is the largest component of the diet. Russell et al. (2016) showed that the optimal inclusion of corn was around 16% of DM in a forage-based diet. When corn was included in greater concentrations, a decrease in diet digestibility was accompanied with a decrease in ADG. This is likely the result of negative interactions between the starch in corn and microbial populations in the rumen. Therefore, producers wishing to feed grain should ensure that they are not providing it at a level that will negatively impact forage digestion and utilization. Martin and Hibberd (1990) fed soybean hulls to beef cows at levels up to 3 kg/d without negative impacts to forage intake and digestibility. However, Scaglia (2017) reported that the efficiency of supplementation was greater for animals receiving 0.5% BW soybean hulls compared to those receiving 1% BW soybean hulls. Corn co-products such as DDGS have been successfully incorporated into grazing diets in concentrations greater than 1% BW, or 45% of DM (Griffin et al., 2012). However, it should be noted that the rate at which DDGS replaced forage (substitution rate) was not constant with varying inclusion rates. As DDGS was incorporated into the diet in greater concentrations the substitution rate also increased. Therefore, greater levels of co-product supplementation may suppress forage intake to the point that cost of gain begins to increase and cattle become less economically efficient.

Use of corn co-products as supplements for beef cattle

As previously mentioned, the increased use of corn grain for ethanol production resulted in greater corn grain prices as well as increased availability of co-products. Nearby corn sweetener

plants in the Southeast provide producers easy access to CGF which is high in energy and moderate in CP. During the processing of corn grain for corn sweetener the majority of starch and sugars are removed, and the resulting co-product contains the majority of its energy in the form of fiber. Despite the increased use of CGF as an energy supplement, relatively little work has been conducted evaluating its use in forage-based production systems. Hardin (2018) showed that CGF supplementation increased ADG beyond that of corn in forage-based diets, and that CGF had a greater energy value than corn with respect to improvements in animal performance. Additionally, the energy value relative to corn increased as the level of supplementation increased from 0.15% BW to 0.96% BW. However, it should be noted that the greatest energy value occurred when CGF was supplemented at 0.15% BW, indicating that the animals utilized the supplement most efficiently at lower levels of inclusion. It should be noted that this experiment was conducted in a drylot with cattle fed mixed-grass hay, and the nutritive value of forage fed in this experiment may underrepresent what would be typical in a grazing situation. Zumbaugh et al. (unpublished) reported that cattle grazing stockpiled tall fescue and supplemented with CGF had greater ADG and efficiency compared to those supplemented with corn. Forage nutritive value in this experiment was typical for cool-season grass pastures in the Southeastern U.S., with moderate levels of energy and protein.

Dried distillers grains, a co-product of ethanol production, are also commonly fed to growing beef cattle. Loy et al. (2008) demonstrated that supplementing DDGS to growing cattle consuming a forage-based diet resulted in greater ADG and efficiency compared to corn. The addition of corn gluten meal to corn diets did not improve animal performance when fed at 0.21% BW, indicating that the improvement in gain is not explained by the increased CP content of DDGS compared to corn. However, performance was improved by the addition of CP when supplemented

at 0.81% BW, which could indicate that the greater CP requirement for growth resulted in a situation that CP became limiting in the corn supplemented animals. Corrigan et al. (2009) showed a linear increase in growth when DDGS was supplemented from 0.25% BW to 1% BW in a forage-based diet. Interestingly, the level of condensed distillers solubles added to the DDGS influenced the optimal rate of inclusion. A quadratic effect of DDGS inclusion was observed when condensed distillers solubles composed 22% of DDGS, and ADG was maximized when supplemented at 0.5% BW. The decreased ADG and efficiency observed when cattle were supplemented at levels greater than 0.5% BW could be explained by the increased fat content, resulting in negative interactions with forage digestibility. Dried distillers grains plus solubles have been shown to improve growth compared to soybean hulls when fed to cattle consuming forage-based diets (Engel et al., 2008). Therefore, the improved growth resulting from supplementation with DDGS rather than corn cannot be attributed to the fiber content alone. It is likely that the increased growth is a result of a combination of highly digestible fiber, increased CP content, and increased levels of fat.

Because of the nature of the wet and dry-milling processes, considerable between and within-plant variation can be observed in the nutrient profile of co-products. Buckner et al. (2008) reported that the variation in DDGS fat content varied as much as 4.5% with a plant and as much as 13.1% between plants. The sulfur content varied considerably more, with a coefficient of variation as large as 8% within a plant and 36% between plants. Because of the large variation, it is important to have actual nutrient content analyzed prior to inclusion in a ration. This is particularly important for sulfur because co-products tend to be greater in this mineral. Combined with large variation in actual sulfur content, greater levels of inclusion in the diet can result in polioencephalomalacia (PEM), or sulfur toxicity. While the upper level of sulfur tolerance is greater in cattle consuming a forage-based diet compared to a grain-based diet, large levels of

sulfates in the water combined with greater inclusion of co-products in the diet can lead to a situation where dietary sulfur levels exceed the maximum tolerable level and result in PEM.

It is common for local cooperative groups to offer a pre-mixed commodity blend. While the ingredient makeup will vary depending on location and access to feedstuffs, these blends typically contain a mixture of corn, CGF, DDGS, soybean hulls, and molasses. Because of the variability in composition among various blends, it is difficult to characterize these products as a class. However, past research has indicated that feeding a blend of co-products results in positive performance effects. In an experiment utilizing feedlot cattle, Loza et al. (2010) indicated that a 1:1 blend of wet CGF and wet DGS can replace corn up to 75% of DM without negative effects on growth. However, the blend outperformed the corn-based control when fed between 25% DM and 50% DM indicating that benefits may subside when fed at levels greater than this. Interestingly, it appears that there is a synergistic effect on digestibility when feeding a mixture of soybean hulls and DDGS. Smith et al. (2017) fed cows diets consisting of hay, soybean hulls, DDGS, or a mixture of soybean hulls and DDGS and measured total tract digestibility. Cows fed the mixture of soybean hulls and DDGS had greater DM, organic matter, NDF, and ADF digestibility compared to either the hay control or supplemented with soybean hulls or DDGS fed alone, indicating that the mixture imparts positive associative effects on digestion. It is possible that the improvement in digestibility is due to increased RDP supply in conjunction with increased amounts of highly digestible fiber, allowing the microbial degradation of feedstuffs to be more efficient, though additional research is needed to validate this.

Creep feeding and self-feeding

Creep feeding is a method of providing supplemental nutrients to pre-weaned calves while excluding cow access. This is done to increase calf growth rate and weaning weights and result in

greater revenue at sale. To be effective in promoting weight gain, a creep feed must supply nutrients that are limiting in the diet. Loy et al. (2002) found that energy was the first limiting nutrient in the nursing calf's diet. Although this experiment was conducted in North Dakota with native range forage, it is likely that the same applies in the Southeast since forage CP concentrations in the Southeast are usually considerably greater. Because calves receiving creep feed are grazing and have a forage-based diet, supplementing greater levels of concentrate has a negative impact on fiber digestibility, and can result in decreased efficiency (Udén, 1984). Fibrous creep feeds, however, do not alter ruminal organic matter digestion and tend to increase apparent total tract organic matter digestibility (Soto-Navarro et al., 2004). This could explain the similar levels of ADG of calves supplemented with soybean hulls compared to corn, despite calves fed soybean hulls having reduced levels of supplement intake (Faulkner et al., 1994). Previous research has demonstrated that although calf ADG is improved when creep feed is provided *ad libitum,* feed efficiency is maximized when calves are limit fed (Faulkner et al., 1994). Therefore, it is advantageous to prevent overconsumption of creep feed to maximize the return on feed costs. A number of alternative feedstuffs have been successfully incorporated into creep feeding programs. Gelvin et al. (2004) demonstrated that a combination of field peas and wheat middlings can improve ADG in nursing calves without impacting milk or forage organic matter intake or digestibility. Similarly, Reed et al. (2006) demonstrated that DDGS can be incorporated into a creep feed up to 50% without negative effects on growth, efficiency, intake, or digestion compared to a soybean meal-based feed.

Principal drawbacks of creep feeding include potentially reduced pasture utilization by calves and increased feed costs. As previously mentioned, the former is of particular concern when high-starch feeds are used due to the reduction in forage digestibility and efficiency. However,

Shike et al. (2007) reported reduced returns for calves not creep fed compared to calves fed a starch and fiber-based creep feed, which were similar. Additionally, the authors reported that creep feeding ultimately improved carcass value, which may provide additional incentive to creep feed for producers who retain ownership of weaned calves. In contrast, Meteer et al. (2013) reported that although calves that were not creep fed had reduced ADG, the reduced feed costs resulted in an overall improved profitability for the cow-calf producer. This is likely the result of a small margin in ADG between the control and creep fed calves, resulting in a situation where the cost of additional gain is greater than the value. This suggests that creep feeding is most beneficial for producers who have relatively small weaning weights when calves are not supplemented but may not be profitable for calves which are grazing high quality forage and have relatively large weaning weights on forage alone. Additionally, creep feeding has been shown to negatively impact the reproductive performance of replacement heifers, and therefore may not be suitable for breeding animals (Martin et al., 1981). Sexten et al. (2004) showed that creep fed replacement heifers had a marked reduction in milk production, which may be due to an increase in the number and size of adipocytes in the udder (Prichard et al., 1989).

Previous research has shown that the value of creep feeding depends on the timing and duration of supplementation. Calves that were creep fed for 56 d or 84 d had improved ADG and supplement efficiency, while calves that were creep fed for 28 d showed no improvement over a non-creep fed control (Tarr et al., 1994). Supplemental gain was most efficient for calves creep fed for 56 d, indicating a possible quadratic effect of duration on efficiency. In heifers, ADG has been shown to linearly increase with creep feeding duration to 84 d, although subcutaneous fat accretion was influenced quadratically (Buskirk et al., 1996). Therefore, producers wishing to

creep feed should carefully evaluate their current production system and design a feeding program to maximize efficient gain.

Frequency of supplementation

The costs associated with feeding animals comprise a significant proportion of total operation costs. Therefore, reducing the frequency that grazing animals are provided supplement has received attention as a method to reduce input costs. Although the majority of research has been conducted in the context of beef cows, growing cattle have received some attention. McIlvain and Shoop (1962) reported no differences in gain among steers fed cottonseed cake daily or every 3 d while grazing winter range pastures in either the winter or summer. Similarly, supplementing steers consuming poor-quality hay with cottonseed meal every 12, 24, or 48 h improved intake, *in-situ* NDF and ADF digestibility, and growth compared to the unsupplemented control, but the frequency of supplementation did not result in an improvement (Hunt et al., 1989). In fact, steers supplemented every 24 h had lower ADG compared to those supplemented every 12 h and 48 h. More recent research has indicated that supplementing steers consuming medium quality hay with a soybean hull and CGF blend clearly improves performance, but there were no differences in nutrient digestibility (Drewnoski and Poore, 2012), growth or intake between animals supplemented daily compared to every other day (Drewnoski et al., 2014).

Interestingly, the steers that were supplemented every other day had increased plasma IGF-1 concentrations compared to both animals supplemented daily and those not supplemented (Drewnoski et al., 2014). Somewhat conflicting results were reported by Moriel et al. (2020) in which heifers receiving daily concentrate supplementation while grazing bahiagrass pastures had both greater ADG and plasma IGF-1 concentrations compared to those supplemented 3 times weekly. However, these animals were *Bos indicus* influenced and it is possible that differences in

sex, breed, and pasture composition resulted in conflicting results. In fact, in a separate experiment *Bos indicus* heifers supplemented with a fibrous supplement daily observed similar improvements with regards to both growth and plasma IGF-1 concentrations compared to those supplemented every 3 d (Cooke et al., 2008).

With regards to forage intake, Loy et al. 2007 showed that there were no differences in hay intake among heifers supplemented daily or every other day. However, these animals were maintained in confinement and their response to supplementation frequency may be different than in a grazing situation. Farmer et al. (2001) demonstrated a linear increase in forage intake with decreasing supplementation frequency. Despite the mixed results, it is likely that more frequent and greater levels of supplementation will decrease forage DMI in grazing cattle.

The results of previous research suggest, in the case of *Bos taurus* growing cattle, that there is little difference in growth performance when supplementing daily or every other day. Therefore, producers wishing to reduce input costs may achieve some success by reducing the frequency of supplement delivery. This practice may also promote greater forage DMI, which is generally the cheapest nutrient source for grazing livestock.

ASSOCIATIVE EFFECTS AND THEIR IMPLICATIONS

Positive associative effects

Indirect effects that enhance the efficiency of digestion are known as positive associative effects and can arise in a number of feeding situations. Most commonly, these are the result of modulation of the rumen microbiome through the provision of specific substrates. Because the majority of energy available to the ruminant animal is in the form of volatile fatty acids (VFA), changes in the ability of bacteria to produce these can have profound impacts on growth and efficiency. Hart

(1987) demonstrated that digestion can be improved when low levels (< 30% of DM) of sorghum grain are added to a silage-based diet. When sorghum grain was included in greater amounts a subsequent decrease in DM, organic matter, and NDF digestibility was noted. A similar effect was observed in which substituting soybean hulls for corn grain increased NDF digestibility when corn inclusion was less than 20% of DM, but caused a reduction when included in greater concentrations (Russell et al., 2016). This effect is likely the result of providing highly digestible energy sources that facilitate microbial growth and digestion of feeds, without resulting in a major shift of microbial populations. This is supported by Hoover and Stokes (1991), who stated that the rate of digestion of carbohydrates is the major factor controlling the energy available for microbial growth. Additionally, *in vitro* fermentation data show that the addition of corn to a soybean hull-based diet at levels of 20% or less result in a slight increase in pH and the ratio of acetate to propionate (Russell et al., 2016). Based on those data, it is unlikely that low levels of starch inclusion in forage-based diets induce a decrease in cellulolytic bacteria populations that would inhibit fiber digestion. For cattle producers in the Southeast, supplementing small amounts of grain to grazing cattle can help maximize the efficiency and digestion of fresh forage.

Positive associative effects can be observed in situations when degradable intake protein (DIP) is supplemented to animals consuming poor-quality forage. In most cases, poor quality forage is deficient in nutrients for both the animal and rumen microbes, which results in decreased microbial efficiency. This effect was demonstrated by Koster et al. (1996) by ruminally infusing steers consuming poor-quality hay with casein (90% DIP). The steers were noted to have greater organic matter intake and true ruminal digestibility of organic matter and NDF when casein was infused into the rumen. However, the improvement in digestibility was variable as the effect of DIP was cubic, and the authors noted diminishing returns in increased intake when casein was

infused at levels greater than 540 g/d. It should be noted that this experiment utilized hay that was severely deficient (< 2%) in CP. It is unlikely that even the most mature forages in the Southeast would ever have CP depressed to this point. However, a similar experiment demonstrated a linear improvement in intake as well as total tract organic matter and NDF digestibility with increasing levels of DIP in the diet when steers were fed mature (4.3% CP) forage sorghum hay (Mathis et al., 2000). However, steers fed Bermudagrass (8.2% CP) or Bromegrass (5.9% CP) did not elicit the same response to increasing levels of DIP. Determining a minimum threshold to elicit a beneficial response is difficult without the proportions of protein fractions. Ultimately, the reduced ruminal digestibility is driven by a deficiency in DIP rather than CP. Crude protein values can certainly provide useful information, but there are cases in which the majority of protein is in the form of undegradable intake protein (UIP), for example corn gluten meal which is 60 - 70% UIP. Additionally, heat damage could result in an increase in bound protein that is unavailable to both the rumen microbes as well as the animal. Forages naturally contain some bound protein, though unless significant heat damage has occurred this amount is generally small. Because forages in the Southeast are typically moderate to high in CP throughout the year, it is unlikely that grazing cattle would exhibit improved digestion through DIP supplementation. However, in cases when poor-quality hay or pasture is being fed and protein becomes limiting, it may be beneficial to provide supplemental UIP to increase the flow of amino acids to the small intestine.

Negative associative effects

Negative associative effects can arise in certain situations in which digestibility, growth, and efficiency are negatively impacted due to the interactions between feeds. One of particular interest to beef producer in the Southeast is the interaction between moderate to high levels of starch in forage-based diets. Grain supplementation has been shown to decrease fiber digestibility

in ruminants (Faulkner et al., 1994; Garcés-Yépez et al., 1997). Specifically, this effect seems to occur when grain is supplemented between 16% and 30% of DM (Russell et al., 2016). This negative associative effect is due to shifts in the rumen microbial population. As ruminants which are adapted to a forage-based diet are fed increasing amounts of grain, the proportion of bacteria which utilize starch as their primary substrate increases and the proportion of bacteria which utilize fiber as their primary substrate decreases. This was shown by Fernando et al. (2010) who demonstrated that grain consumption by cattle previously fed forage resulted in a decrease in the population of *Fibrobacter succinogenes* and the proportion of *Fimicutes/Bacteroidetes*. *Fibrobacter* spp. has been previously identified as the major cellulolytic bacteria present in the rumen, and *Fibrobacter succinogenes* was the most abundant of several cellulolytic bacteria measured by Koike and Kobayashi (2001). Additionally, Fernando et al. (2010) reported that the ratio of *Firmicutes/Bacteroidetes* is greater in forage-fed cattle compared to grain-fed cattle, which may be due to differences in the efficiency of energy substrate utilization among the bacterial species in the two phyla. These changes demonstrate how the rumen microbial population undergoes a dramatic shift when nutrient substrates are changed. The net result is that the animal becomes increasingly adapted to efficiently digest starch and less adapted to efficiently digest fiber. From a production standpoint, this can be observed by the decreased ADG and efficiency when forage-fed cattle are supplemented with corn at 30% of DM (Russell et al., 2016).

Although unlikely in pasture-based production systems, diets with large amounts of unprotected lipids can cause a disruption in ruminal fermentation and result in decreased feed intake, digestibility, and efficiency. The addition of approximately 10% tallow results in a substantial decline in ruminal DM, NDF, and ADF digestibility (Jenkins and Palmquist, 1984). Practical recommendations for forage-fed cattle range from a maximum inclusion of 2% to 4% of

DM (National Academies of Sciences and Medicine, 2016). At this level total tract digestibility appears to be unchanged, although a substantial decrease in ruminal digestion was noted (Jenkins and Fotouhi, 1990). This is thought to be due to limited hindgut fermentation, which can lessen the decrease in digestibility when not at maximum capacity (Jenkins, 1993). Dietary fat is less detrimental to the ruminal digestion of non-structural carbohydrates (Jenkins, 1993), and as such, cattle fed grain-based diets can tolerate dietary fat concentrations up to 10% of DM (National Academies of Sciences and Medicine, 2016). These effects are thought to be caused by either the direct antimicrobial effects of lipids on rumen microbes, or through the coating of feed particles which prevents either bacterial attachment to the feed or the contact between feed particles and bacterial enzymes (Jenkins, 1993). Regardless, supplementing rumen-protected fats have been shown as a method to provide greater concentrations of supplemental energy without disturbing rumen digestion kinetics (Bayourthe et al., 1993).

DILUTING ERGOT ALKALOID INTAKE THROUGH SUPPLEMENTATION

Background and relevance

Some researchers have stated that the negative effects of E+ tall fescue can be mitigated through the use of supplemental feeds (Strickland et al., 2011; Kallenbach, 2015). It is thought that consumption of supplement will decrease dietary concentrations of ergot alkaloids, and thereby improve animal performance as a result. Ultimately, this effect is the result of a reduced proportion of forage in the diet due to inclusion of the supplement. As ADG improves as the concentration of ergot alkaloids in the diet decreases (Liebe and White, 2018), producers can realize improved growth from both the decreased ergot alkaloid concentration and the greater plane of nutrition due to supplementation.

Evidence of a dilution effect and subsequent improvement in performance

Despite reports suggesting a dilution effect, many of the experiments these claims are based on are confounded by the greater plane of nutrition for animals in the supplement group. For example, Forster et al. (1993) observed an increase in ADG when steers grazing E+ tall fescue were supplemented with either corn or rice bran. While it is possible that part of the performance response can be explained by decreased dietary ergot alkaloid concentration, the design of the experiment makes it impossible determine how much of this is due to toxin dilution. Similarly, Stokes et al. (1988) noted an improvement in ADG when cattle consuming E+ tall fescue were supplemented with corn, but found no corresponding increase in serum prolactin concentrations. Additionally, there was no interaction between supplementation and endophyte status on ADG or prolactin, suggesting that no dilutive effect was observed. Likewise, Poole et al. (2019) noted that serum prolactin concentrations were significantly reduced for animals consuming E+ tall fescue compared to endophyte free tall fescue despite protein supplementation. Serum prolactin is a common biomarker for exposure to ergot alkaloids in cattle, and reduced concentrations of serum prolactin are associated with greater intake of ergot alkaloids. Although this effect has been debated, prolactin has been shown to promote the production and secretion of milk in cattle (Lacasse et al., 2016). Therefore, it is likely that the decreased milk production that is associated with intake of ergot alkaloids is due to the reduction in prolactin concentrations. Carter et al. (2010) reported that steers fed pelleted soybean hulls at 2.3 kg/d had greater than 2-fold increase in serum prolactin concentrations compared to those grazing E+ tall fescue without supplementation. The interpretation that this is due to dilution of toxin is somewhat complicated by the reportedly greater forage DMI of steers consuming the soybean hulls relative to those grazing. However, forage DMI was estimated by the reduction in apparent forage mass as measured with a rising plate meter and

may not accurately reflect individual animal DMI. Therefore, it is still plausible that the greater prolactin intake is due to a reduction in toxin intake.

Effect of dilution on animal welfare

If ergot alkaloid concentrations can be diluted through supplement feeding it is possible that the animals will be able to dissipate heat easier, ultimately leading to reduced heat stress and improved welfare. Carter et al. (2010) noted a reduction in the percentage of animals with a rough hair coat and an increase in the percentage of animals with a sleek hair coat when they were supplemented with pelleted soybean hulls compared to those only consuming the forage. Although temperature data were not collected in this experiment, the reduced proportion of rough hair coats and increased proportion of sleek hair coats may lead to a greater ability to dissipate heat, and ultimately reduced heat stress. Other experiments have shown that providing supplemental seaweed extract decreases core body temperature and respiration rate in cattle that are heat stressed, though this is unlikely to be a dilution effect as the extract was provided in small quantities (Spiers et al., 2004). Therefore, it is unclear the effect of ergot alkaloid dilution on improving animal welfare, though it is possible that dilution will reduce the incidence of heat stress during the summer months due to the reduced proportion of rough hair coats.

CONCLUSIONS

The Southeastern U.S. is an important region for the U.S. beef industry, providing approximately 20% of the nation's cow herd and is a major source of feeder cattle. Production systems are characterized as predominantly pasture-based with a substantial portion of cattle grazed on E+ tall fescue. While the nutritive value of this forage is generally good, ergot alkaloids can be toxic to grazing livestock and result in reduced performance, stress, and in some cases

health problems. While it can be difficult to estimate the exact economic impact ergot alkaloids have on beef production, it is no doubt substantial, and likely leads to greater than $500 million in lost revenue each year in this region alone.

Supplemental feeding can be a valuable strategy to improve growth and efficiency in cattle raised in the Southeast. In times of poor forage quality or low quantity, such as during the winter or summer, supplemental feeds can fill the gap between nutrients provided and what is required for the desired level of growth. Optimal supplement choice will vary greatly depending on availability, cost, and basal diet type. For most production systems in the Southeast, cattle will graze throughout the year and their basal diet will be forage. In most cases, the protein content of forages will be sufficient to meet the requirement for most levels of gain. However, if animals are grazed on very mature forage or if poor-quality warm season grasses make up the majority of the diet, greater levels of growth may require protein supplementation. In these cases, the energy content of the diet is likely limiting, and a protein and energy supplement such as DDGS or CGF may make the most economical sense to supplement. If animals are grazing cool-season grasses, energy is likely the first limiting nutrient in most cases. Supplementation with fibrous energy supplements is usually the most beneficial for grazing cattle due to the interaction between the feeds and rumen physiology. The fiber present in the feed is easily degraded in the rumen to produce energy, and the fiber does not cause a decrease in rumen pH and fiber digestibility that is commonly seen when starch-based supplements are fed. This results in greater overall diet digestibility, which oftentimes results in improved feed efficiency and greater profits. The role of supplementation in combating the negative effects of ergot alkaloids has long been theorized, but relatively little research is available to conclusively determine its effect. It is clear that supplementation improved performance when cattle graze E+ tall fescue, however, a clear link

between supplementation, reduced ergot alkaloid intake, and improved performance has yet to be shown.

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CHAPTER III

Determining the feeding value of corn gluten feed relative to corn when supplemented to beef calves grazed on stockpiled tall fescue pastures

ABSTRACT

The objective of this experiment was to compare the performance response of supplementing corn gluten feed (**CGF**) or corn to calves grazing stockpiled tall fescue pastures over a 70-d treatment period. Forty-four Angus × Simmental calves (238 ± 4 kg) were stratified across 6 treatments by BW and sex in a 2 × 3 factorial arrangement. Calves were supplemented either cracked corn (n = 16) or CGF (n = 18). Calves were housed in 2 groups and supplemented individually using the SmartFeed Pro feeding system (C-Lock Inc., Rapid City, SD) with supplement DMI programmed at 0.59, 0.87, or 1.13% BW. Calves which failed to train to the feeding system (n = 10) were treated as an un-supplemented control. Weight and flesh condition score (**FCS**) on a scale from 1 to 9 were recorded every 14-d, and ultrasound 12th rib fat thickness (**FT**) was measured on d 0 and 70. Data were analyzed with PROC GLM in SAS with supplement type as a main effect, sex as a fixed effect, and group as a random effect. Individual supplement DMI was included in the model as a covariate rather than a main effect due to large variation between programmed and actual DMI. Significance was defined at $P \leq 0.05$ and means were separated using LSD. Initial BW, FCS, and FT were similar ($P \geq 0.12$) among treatments. No differences ($P \geq 0.25$) were observed in supplement DMI throughout the experiment. No differences ($P \geq 0.64$) were observed in ADG and supplement G:F through 14 d. Through d 28 to 70, CGF-fed calves had greater ($P \leq 0.03$) ADG and supplement G:F relative to those fed corn. Final FCS was greater ($P < 0.01$) for CGF-supplemented calves relative to those supplemented corn, though FT was not different ($P = 0.87$).

These data indicate that supplementing CGF rather than corn yields a greater performance response in beef calves grazing stockpiled tall fescue.

INTRODUCTION

The expansion of the ethanol industry in the early 2000s increased the price and volatility of corn and increased the availability of corn co-products. Corn co-products have traditionally been utilized as a protein source, but increases in corn prices resulted in increased interest in their use as energy supplements. Since then, research has shown that co-products such as dried distillers' grains plus solubles (**DDGS**) and corn gluten feed (**CGF**) have a greater energy value than corn when included in grain-based rations (Loe et al., 2006; Buckner et al., 2007; Klopfenstein et al., 2008). Despite this, relatively little work has been done regarding the inclusion of co-products in forage-based rations. Because of major differences in basal diet type, it is likely that the energy value and resulting performance of animals will be different in forage-based diets compared to grain-based diets. While starch-based energy supplements have been the principal supplemental energy source for grazing cattle, they have been shown to negatively impact fiber digestion, intake, and pasture utilization (Anderson et al., 1988; Russell et al., 2016). This ultimately results in decreased growth efficiency and pasture utilization. This is likely why, in general, supplementing grazing cattle with fibrous energy sources results in greater improvements in gain compared to grain sources, despite the apparent lower energy content (Gadberry et al., 2015). Previous research has demonstrated that cattle consuming a forage-based diet and supplemented with DDGS have greater ADG compared to those supplemented with corn (Loy et al., 2008). This improvement is likely due in part to differences in energy source (starch vs. fiber); however, these differences cannot be attributed to the fiber content alone as DDGS supplementation has been shown to improve growth beyond that of soybean hulls (Engel et al., 2008). Despite previous work regarding the effect of DDGS supplementation for cattle consuming forage-based rations, few data are available regarding CGF supplementation. In a dry-lot setting with mixed-grass hay, CGF

supplementation has been shown to increase ADG compared to corn, although no differences were noted in feed efficiency (Hardin, 2018). As cattle in the Southeastern U.S. are primarily managed in pastures, additional data are necessary to determine if similar results are observed under grazing conditions. Therefore, the objectives of this experiment were to characterize the performance response of growing cattle supplemented with corn or CGF when grazing tall fescue pastures.

MATERIALS AND METHODS

All animal procedures were approved by the Virginia Tech Institutional Animal Care and Use Committee (Protocol #: 18-009).

Animals, diets, and management

Forty-four 8 mo old Angus × Simmental beef steers and heifers (239 ± 14 kg) were stratified by body weight and sex and allotted to one of six treatments in a 2 × 3 factorial arrangement. Treatments consisted of individual supplementation of either cracked corn or pelleted corn gluten feed (**CGF**) fed at 0.59% BW, 0.87% BW, or 1.13% BW while grazing fescue-mixed grass pasture over a 70-d period from November 16, 2018 until January 25, 2019. Weather data were collected from the National Oceanic and Atmospheric Administration from the weather station at the Virginia Tech Airport (Table 3.1). The failure of 10 calves to successfully train to the feeding system after the start of the trial resulted in a total of 16 animals allotted to the corn treatments and 18 animals allotted to the CGF treatments. Supplement inclusion levels were formulated using the Beef Cattle Nutrient Requirements Model (BCNRM; National Academies of Sciences, Engineering, and Medicine, 2016) to meet nutrient requirements for 0.68, 0.96, or 1.25 kg gain/day assuming a daily forage dry matter intake (DMI) of 1.8% BW using the BCNRM.

Supplement DMI was updated every 2 weeks to account for changes in BW. Actual nutrient composition of supplements are shown in Table 3.1.

Calves were allocated into 2 groups housed in one of 2 pastures that were stockpiled in the Fall and were rotated to new pastures on d 31 (December 17) before forage mass was determined to be limiting by visual appraisal. The pastures were approximately 2.25 ha with a stocking rate of approximately 10 calves/ha. Group size was held constant during the experiment. The total ergot alkaloid concentrations of the pastures were 1,025 ± 553 µg/kg prior to rotating animals and 86 ± 119 µg/kg after rotating animals. Mixed grass hay was fed to both groups from d 24 to d 31 (December 10 to December 17) due to snow, and from d 55 (January 10) until the conclusion of the treatment period due to limited available forage mass. Calves had *ad libitum* access to water and a commercially available vitamin and mineral premix (Southern States Cooperative, Richmond VA) throughout the treatment period. The mineral was 14.6% Ca, 1.9% P, 7.8% Na, 11.8% Mg, 0.8% K, 323 ppm Cu, 26.0 ppm Se, 1,299 ppm Zn, 1,243 ppm Mn, 59 ppm I, 49 ppm Co, 220,242 IU/kg Vit A, 56,879 IU/kg Vit D, and 551 IU/kg Vit E.

Supplement intake was monitored and controlled using the SmartFeed Pro feeding system (C-Lock inc., Rapid City, SD). Each trailer contained two stainless steel feed bins suspended on two load cells. The outside of the trailer is equipped with a radio frequency identification (RFID) reader directly adjacent to a gate with a locking mechanism that controls access to the feed bin. The metal frame surrounding the gate is designed such that only one animal can access the feed bin at a time. Each animal was assigned a unique RFID ear tag (Allflex, USA, Dallas, TX) that operated at a frequency of 134.2 KHz, and an individual daily intake limit was set on the feeding system. In brief, as an animal which has not consumed the allotted amount came in close proximity of the RFID reader, the gate would unlock, and the animal would have access to the feed. The two

load cells under the feed bin would determine the difference in feed mass at the beginning and end of each meal event and calculate a daily sum. Once the animal had consumed the allotted amount of feed, and actuator would close the door and the gate would no longer unlock for that animal that day. The daily feed quota would then be reset at midnight, and feed intake data was uploaded to the cloud. Each pasture was equipped with one SmartFeed trailer that each contained two feed bins. Each feed bin was assigned a unique feed type and animals were given access to a single feed bin. Therefore, animals from each treatment were comingled in each of the pastures.

Calves were trained to eat from the SmartFeed trailers over a 28-d period prior to the start of the experiment. In order to reduce competition and facilitate consumption, calves were divided to two groups of approximately twenty-eight. Because it was anticipated that some calves would fail to train, a total of fifty-seven calves were utilized for training. During the first seven days of the training period, a group was housed in a 320 m^2 paddock containing a SmartFeed trailer. Calves were provided *ad libitum* access to water and a training feed. The training feed contained soybean hulls, cottonseed meal, and 13% salt to limit intake to approximately 2.3 kg/day. Calves were locked in the paddock during the day and provided access to a pasture in the afternoon and throughout the night. After the majority of animals were observed to be consistently eating out of the feeders, gates to the paddock were left open and the feed restriction gate was gradually closed until at the highest setting. The group was then moved into a new pasture with a SmartFeed trailer and intake was limited to 2.3 kg/head/day. This process was repeated for both groups. Approximately 5 d prior to the start of the experiment, calves and trailers were moved to a single lot near the experimental pastures and were fed corn at a rate of 2.3 kg/head/day. A total of 13 calves failed to adequately train to the feeding system and were removed from the experiment. After the experiment began an additional 10 calves failed to adequately consume their allotted

amount of feed and were used as an un-supplemented control treatment. The feed intake for these calves was programmed to be 0 on d 14 to prevent them from consuming supplement.

Animal measurements

Animals were weighed on 2 consecutive days at the beginning and end of the experiment, with interim BW recorded once every 14 d of the treatment period. Interim ADG was calculated individually each time interim BW was recorded. During weighing, calves were restrained in a squeeze chute and a flesh condition score (FCS) was assigned based on visual appraisal and brief palpation. Scores were assigned by 2 trained personnel to the nearest 0.5 score on a 1 to 9 scale. A score of 1 indicated a severely emaciated animal and a score of 9 indicated an animal with excessive fat deposits. At the beginning and end of the experiment, ultrasonography was utilized to determine ultrasound 12^{th} rib fat thickness (**FT**). Briefly, hair was clipped to a depth ≤ 0.25 cm on the animal's right side from the shoulder to the hook bone and vegetable oil was liberally applied to the clipped area. Measurements were taken in a transverse orientation between the 12^{th} and 13^{th} ribs, approximately 10 cm distal to the midline. Three to 7 measurements were taken per animal and the 3 measurements within 0.07 cm were averaged. Measurements were taken with an Aloka SSD-500 (Wallingford, CT) B-110 mode instrument equipped with a UST-5011 3.5 MHz general purpose transducer array. The distance function was used to measure the fat thickness

Supplement DMI was measured daily for each animal by the SmartFeed Pro system. Daily intake was then averaged for the periods 0 to 14 d, 0 to 28 d, 0 to 42 d, 0 to 56 d, and 0 to 70 d and converted to a DM basis using the average % DM of each supplement over the 70-day feeding period. There was a 5 d period in the middle of the experiment when the ropes that function to close the door broke on two feed units, which resulted in unreliable measurements. As such, these

values were excluded from the average for each calf assigned to those feeders. The average BW of each calf was calculated for each period and used to calculate the ADFI on a % BW basis.

Supplement gain to feed (**G:F**) was calculated on an individual basis by dividing ADG by average daily supplement DMI for each period on a kg basis. G:F relative to the control was calculated by dividing additional ADG relative to the control (ADG for each period less the mean ADG of control group) by supplement intake for each period.

Mean ADG was calculated for the un-supplemented control group at each time point. The difference in ADG for the un-supplemented control group relative to the control group was then calculated on an individual basis by subtracting the mean ADG of the control group from each animal's ADG during that period.

Forage and supplement analysis

Forage grab samples were collected from each pasture from 20 points to obtain a representative sample along with feed samples approximately every 2 wk for analysis of nutritive value. A portion of the forage samples collected for nutritive value were frozen at -20 °C and sent to Agrinostics Inc. for analysis of total ergot alkaloids. Samples of corn and CGF were dried for 24-h at 105 °C for determination of DM, with these values being used to update supplement intake levels. Additional forage and feed samples were dried at 55 °C for at least 72-h, ground to pass through a 1-mm screen, and analyzed for crude protein (**CP**), neutral detergent fiber (**NDF**), acid detergent fiber (**ADF**), ether extract (**EE**), and ash. Crude protein was analyzed by measuring the nitrogen content using an Elementar Vario EL Cube (Elementar Americas Inc., Ronkonkoma, NY) and multiplying by 6.25. An Ankom 200 fiber analyzer (ANKOM, Macedon, NY) was used to determine NDF and ADF following the manufacturers recommendations. Ether extract was

measured using the Ankom XT10 following the manufactures recommendations (ANKOM, Macedon, NY). Analysis of CP, ADF, and EE were conducted using Association of Official Analytical Chemists (1990) methods 990.03, 973.18, and 920.39, respectively. Neutral detergent fiber was measured using the methods described by Goering and Van Soest (1970). Ash was determined by placing a 0.5 g sample in a furnace at 600 °C for 24 h. Total digestible nutrients (**TDN**) was then calculated for pasture samples using equation 3.1 (SGS-AgriFood Labs, Guelph, ON):

$$\boldsymbol{TDN, \% = 98.625 - (ADF, \%DM * 1.048)} \textbf{Eq. 3.1}$$

Forage mass

Forage mass (**FM**) was determined approximately weekly using a rising plate meter (NZ Agriworks Ltd., Feilding, NZ), with 10 readings taken randomly throughout each pasture. Within each sampling date and pasture, forage under the rising plate was clipped to ground level at 3 spots after taking a rising plate meter reading and dried at 105 °C for at least 24 h. Data from each pasture was compiled and used to develop a regression equation to relate the rising plate meter reading to FM. The derived equation is displayed in equation 3.2 and had an R^2 of 0.57:

$$\boldsymbol{Forage\ Mass\ \left(\frac{kg}{ha}\right) = 114.05 * RPM\ Reading + 2321.9} \quad \textbf{Eq. 3.2}$$

Cost of gain

Cost of gain was calculated using actual prices paid for corn and CGF as well as individual supplement DMI. The mean overall ADG of the control group was subtracted from the overall ADG for each animal to calculate additional ADG from supplementation. Average daily gain and average supplement DMI were then multiplied by 70 to return total gain and total supplement

intake over the course of the experiment. Total feed costs were calculated by multiplying supplement DMI by the cost per kg of the supplement on a dry matter basis. Cost of gain was then calculated by dividing total gain by total supplement cost and is expressed as kg of gain per $. This was done to properly account for the negative gain relative to the control that some animals experienced.

Statistical analysis

Animal data were analyzed by ANOVA in SAS 9.4 (SAS Institute Inc., Cary, NC), with significance defined at $P \leq 0.05$. This experiment was originally designed as a 2 × 3 factorial arrangement with main effects of supplement type and supplement DMI. Individual variation in daily supplement DMI was greater than anticipated and resulted in a continuum of intakes rather than 3 distinct levels. As a result, supplement DMI as a percent of BW was included in the model as a covariate rather than a main effect. The model included the fixed effects of supplement and sex, the random effect of pasture group, and the supplement DMI as a percent of BW as a covariate. The model for cost of gain included the fixed effects of supplement and sex, and the random effect of pasture group, without including supplement DMI as a % of BW as a covariate. When ANOVA revealed a significant difference, means were separated using Fisher's LSD. The interaction between supplement type and supplement DMI was not significant at any point and was removed from the model. Forage mass and nutritive value data were analyzed by repeated measures using PROC MIXED in SAS 9.4. The model included the fixed effects of date and pasture group, and the repeated subject was pasture using the spatial power covariance structure. This covariance structure was chosen because the data was not equally spaced throughout time, and it was anticipated that measurements that are spaced further away in time would be less correlated than

those spaced closer together in time. This covariance structure also resulted in the lowest AICc and BIC values.

RESULTS AND DISCUSSION

Forage measurements

This experiment was conducted during the late fall and early winter months when cool-season grasses would typically be dormant. As would be expected during this time, forage nutritive value decreased throughout the experiment (Figure 3.1). Neutral detergent fiber increased (P = 0.03) throughout the experiment and EE decreased (P = 0.03) throughout the experiment. Acid detergent fiber tended to increase (P = 0.06), which resulted in a tendency for TDN to decrease (P = 0.06), as TDN was calculated using ADF content. No differences (P = 0.21) in CP were noted, with CP concentrations remaining moderate by the conclusion of this experiment (12%, DM).

A decrease (P < 0.01) in FM over time was observed over time as animals consumed the available forage (Figure 3.2). Calves were rotated once during the experiment when FM was estimated to be approximately 4,000 kg/ha using the rising plate meter. At the conclusion of this experiment FM was approximately 3,500 kg/ha.

Animal performance

Calf BW, ADG, supplement DMI, and G:F are shown in Table 3.2. Initial and final BW were not different ($P \geq 0.49$) among supplement types. Average daily gain was not different (P = 0.64) between treatments through the first 14 d but was greater ($P \leq 0.01$) for calves fed CGF compared to those fed corn throughout the rest of the 70-d experiment. Supplement G:F followed a similar trend to that of ADG. Through 14 d there was no difference (P = 0.80) in supplement G:F among treatments though calves fed CGF were more efficient ($P \leq 0.02$) than those

supplemented with corn throughout the remainder of the experiment. Final BW was likely not different among treatments due to variation in individual BW. It is not surprising that ADG was not different during the first 14 d due to the short time period animals were on treatments at this point in the experiment. However, on d 28 animals supplemented with CGF gained faster than those supplemented with corn. Previous literature has shown that supplementation with a different corn co-product, DDGS, results in greater ADG than corn in a forage-based ration (Loy et al., 2008). A meta-analysis of similar experiments revealed a quadratic effect of DDGS or wet distillers grains plus solubles inclusion level on ADG, with an optimal inclusion level of around 30% of DM (Klopfenstein et al., 2008). Although work regarding the inclusion of CGF in forage-based diets is limited, Hardin (2018) showed that CGF supplementation results in greater ADG compared to corn when fed to steers consuming a hay-based diet and maintained in a dry lot. Despite the improved ADG, no differences were noted in supplement or overall G:F. However, it was reported by Hardin (2018) that steers were able to consume hay from other bunks, which likely added considerable variation to intake measurements, making comparisons difficult. Somewhat conflicting results were reported for steers fed silage-based rations in which the addition of CGF resulted in decreased ADG and G:F compared to DDGS and corn/soybean meal (Segers et al., 2013). However, interpretation of their results is complicated by the basal diet type of corn silage, as well as the greater than normal proportion of heat damaged protein in the CGF supplement. The results of the current experiment suggest that the improvements in ADG noted by Hardin (2018) are observed when CGF is supplemented to grazing cattle.

While this experiment did not investigate possible mechanisms for the improvement in performance, previous research has suggested that the nutrient profile of the supplement may play a large role. Grain supplementation has been shown to decrease fiber digestibility in ruminants

consuming a forage-based ration (Faulkner et al., 1994; Garcés-Yépez et al., 1997), and corn has been shown to exert this negative effect when supplemented at levels between 16% and 30% DM (Russell et al., 2016). The large differences in energy substrate between the forage and grain result in a shift in the microbial profile in the rumen, which reduces the animal's efficiency at digesting the fiber in the forage. Given that the bulk of the nutrients consumed by animals in the current experiment was in the form of forage, it is possible that corn supplementation reduced ruminal fiber digestibility, thus reducing animal performance.

Supplement DMI was not different ($P \geq 0.28$) among treatments at any point throughout the experiment (Table 3.2). This was anticipated as supplement DMI was designed to be similar across supplement types.

Throughout the experiment, supplemented animals generally had greater ADG compared to the control (Table 3.3), with the exception of the corn-fed calves on day 28. By the conclusion of the treatment period, calves supplemented with CGF gained approximately 0.3 kg/d more than un-supplemented calves ($P < 0.01$), and as would be expected from the ADG data, this difference was greater for calves fed CGF compared to corn. Similar to the supplement G:F data, cattle supplemented with CGF had a greater ($P \leq 0.03$) G:F relative to the control compared to cattle supplemented with corn after d 14 (Table 3.2). In fact, by d 70 cattle supplemented with CGF had nearly twice as great G:F relative to the control compared to those supplemented with corn. These data suggest that, overall, animal ADG was not limited by the CP content of the forage, as calves supplemented with the low-protein corn had greater levels of ADG compared to those which were un-supplemented.

When actual nutrient content of the forage and supplements, and cattle weights were entered into the BCNRM model to predict performance, calves fed corn and CGF were predicted

to gain 0.96 and 1.06 kg/d, respectively. While the model predicted cattle fed CGF to have greater ADG compared to those fed corn, the relative difference in gain was considerably underestimated (0.10 kg/d estimated vs. 0.17 kg/d actual). Similar results were obtained by Hardin, 2018, although the magnitude of difference was substantially greater in the current experiment. Additionally, observed performance in the current experiment was significantly lower than what was predicted. This is likely a combination of the effects of colder than anticipated weather, snow, and significantly more mud than what was expected which likely increased maintenance energy requirements and decreased performance. In typical production systems animals experiencing increased environmental challenges would commonly be fed a greater amount of supplement to meet increased maintenance energy requirements; however, we held supplement DMI as a percent of BW constant throughout the experiment to avoid potentially confounding effects. For both groups, ADG declined throughout the duration of the experiment (Figure 3.3), which is likely the result of a combination of worsening environmental conditions and declining forage nutritive value.

Body composition

Initial FCS and FT were not different ($P \geq 0.11$) among treatments at the start of the experiment. Surprisingly, calves supplemented with CGF had greater ($P < 0.01$) FCS compared to those supplemented with corn by d 70. This is in contrast to the results of Hardin, 2018, who reported no differences in FCS over a 56-d experiment. However, the relatively large number of experimental units along with a potential bias for assigning larger, heavier weight cattle a greater FCS could explain why cattle supplemented with CGF had greater FCS than those supplemented with corn. This is supported by the FT data, which revealed no differences ($P = 0.87$) between

calves supplemented with CGF compared to those supplemented with corn through d 70. Therefore, it is unlikely that supplementation with CGF had any effect on subcutaneous fat accretion. This is unsurprising, as these animals were relatively young and would be primarily increasing lean tissue mass rather than fat mass at this age.

Supplement economics

Over the course of this experiment calves supplemented with CGF had greater gain per unit of feed cost ($P < 0.01$; Table 3.3). The supplements utilized in the current experiment cost \$0.24/kg (\$214.78/907 kg) for corn and \$0.20/kg (\$182.94/907 kg) for CGF on a DM basis. The decreased cost of CGF relative to corn combined with the greater ADG of animals supplemented with CGF resulted in a greater than 2-fold increase in growth per dollar. It should be noted, however, that calculated gain per dollar does not include pasture or hay costs. Despite this, forage intake would likely be similar among treatments given there were no differences in supplement intake. Similar results were obtained by Hardin (2018), who demonstrated that steers supplemented with CGF had greater returns on supplement cost compared to those supplemented with corn at 0.54 and 0.96% BW.

Conclusions

The results from the current experiment suggest that supplementing grazing beef cattle with CGF results in a greater performance response and is more economical compared to corn supplementation. This was observed through increased ADG and improved supplemental feed efficiency, without changes in subcutaneous fat accretion when cattle were supplemented with CGF. While the current experiment did not investigate possible reasons for this, previous research

has suggested that this may be due, in part, to associative effects and the resulting improved digestive efficiency, as well as synergistic interactions between nutrient sources in the supplement. For producers with access to CGF, this is advantageous since CGF can usually be purchased at a lower cost than corn. Taken together, this can result in decreased cost per unit of gain and ultimately a more profitable operation.

Table 3.1: Average maximum[1] and minimum[2] temperature and precipitation[3] over the 70-d treatment period.[4]

	0 to 14 d	14 to 28 d	28 to 42 d	42 to 56 d	56 to 70 d
Maximum temperature (°C)	6.8	5.6	7.3	8.7	1.8
Minimum temperature	-3.3	-3.5	-2.3	-0.1	-6.3
Precipitation (mm)	51.8	42.2	61.7	32.3	51.3
Cumulative precipitation (mm)[5]	1,201	1,244	1,305	1,338	1,389

[1] Average of the average daily maximum temperature for each day.
[2] Average of the average daily minimum temperature for each day.
[3] Total precipitation throughout each period.
[4] Data provided by the National Oceanic and Atmospheric Administration. Data are from the period of November 15, 2018 to January 25, 2019.
[5] Cumulative precipitation since January 1, 2018.

Table 3.2: Nutrient composition of feedstuffs supplemented to cattle grazing stockpiled tall fescue pastures over a 70-d treatment period.

		Analyzed Concentration, % DM[1]				
	DM, % As Fed	**CP**	**NDF**	**ADF**	**EE**	**Ash**
Cracked Corn	86.2	9.0	11.8	2.8	5.1	2.1
Corn Gluten Feed	90.3	21.1	38.1	8.7	2.4	7.1
Hay	81.7	11.2	70.1	40.6	1.9	6.9

[1] DM = dry matter; CP = crude protein; NDF = neutral detergent fiber; ADF = acid detergent fiber; EE = ether extract.

Table 3.3: Effect of supplement type and supplement DMI[1] on body weight (BW), average daily gain (ADG), and flesh condition score (FCS) of calves grazing fescue pasture and supplemented with either corn or corn gluten feed (CGF) over a 70-day treatment period.[2]

	Supplement Type[3]			*P* - value
	Corn	**CGF**	**SEM**	**Supplement**
BW, kg				
Initial	240	236	6.3	0.68
Final	272	280	8.0	0.49
ADG, kg/d				
0 to 14 d	1.05	1.13	0.13	0.64
0 to 28 d	0.73[b]	0.99[a]	0.07	<0.01
0 to 42 d	0.71[b]	0.92[a]	0.05	<0.01
0 to 56 d	0.56[b]	0.76[a]	0.04	<0.01
0 to 70 d	0.46[b]	0.63[a]	0.04	<0.01
Supplement DMI, %BW				
0 to 14 d	0.42	0.50	0.05	0.28
0 to 28 d	0.58	0.61	0.04	0.68
0 to 42 d	0.63	0.64	0.04	0.72
0 to 56 d	0.66	0.69	0.04	0.54
0 to 70 d	0.66	0.71	0.05	0.40
Supplement G:F				
0 to 14 d	1.141	1.084	0.168	0.80
0 to 28 d	0.522[b]	0.661[a]	0.043	0.02
0 to 42 d	0.449[b]	0.565[a]	0.028	<0.01
0 to 56 d	0.337[b]	0.432[a]	0.025	<0.01
0 to 70 d	0.277[b]	0.349[a]	0.021	0.02
G:F relative to the control[5]				
0 to 14 d	0.153	0.240	0.163	0.69
0 to 28 d	-0.066[b]	0.095[a]	0.045	0.01
0 to 42 d	0.047[b]	0.178[a]	0.032	<0.01
0 to 56 d	0.082[b]	0.193[a]	0.033	0.02
0 to 70 d	0.088[b]	0.173[a]	0.027	0.03

[a-b] Means within a row not sharing a common superscript differ significantly ($P \leq 0.05$).

[1] Supplement DMI was calculated as the average of daily supplement intakes throughout the designated feeding period as measured by the SmartFeed Pro feeding system divided by the average body weight during that period on an individual basis.

[2] Values are means of 16 replicates (corn) or 18 replicates (CGF).

[3] Supplements were formulated to be fed at 0.59% BW, 0.87% BW, and 1.13% BW for corn and CGF.

[4] Supplement = main effect of supplement type.

[5] Calculated as ADG, kg divided by ADFI, kg

Table 3.4: Effect of supplement type and supplement DMI[1] on the difference in average daily gain (ADG) relative to the average of an un-supplemented control group and cost of gain of calves grazing fescue pasture and supplemented with either corn or corn gluten feed (CGF) over a 70-day treatment period.[2]

	Supplement Type[3]			*P* – value
	Corn	**CGF**	**SEM**	**Supplement**
Difference, kg/d[4]				
0 to 14 d	0.21	0.29	0.13	0.64
0 to 28 d	-0.09[b]	0.17[a]	0.07	<0.01
0 to 42 d	0.10[b]	0.31[a]	0.05	<0.01
0 to 56 d	0.17[b]	0.37[a]	0.04	<0.01
0 to 70 d	0.17[b]	0.34[a]	0.04	<0.01
Cost of gain (kg gain/$)[5]				
0 – 70 d	0.24[b]	0.71[a]	0.33	<0.01

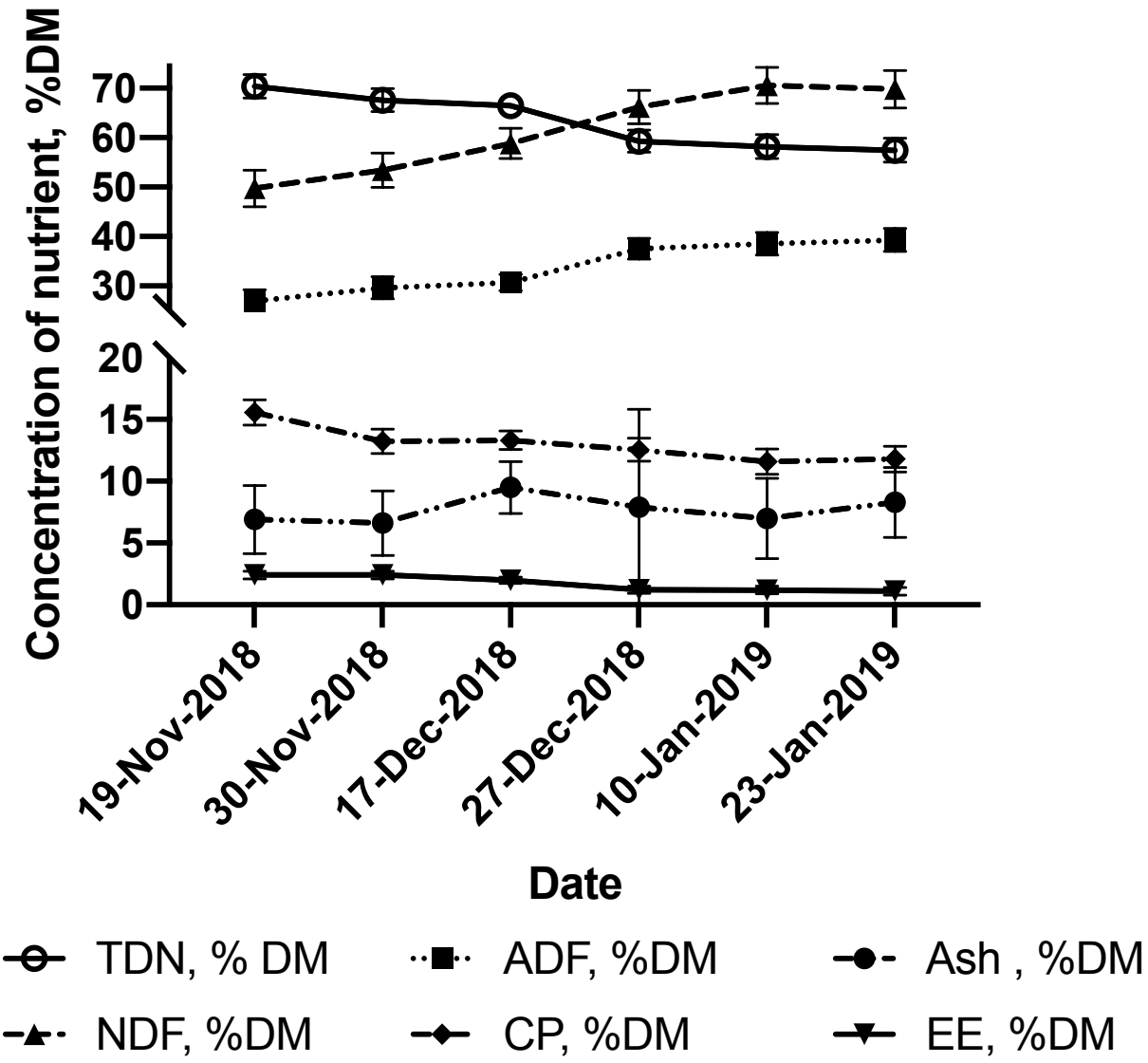
[a-b] Means within a row not sharing a common superscript differ significantly ($P \leq 0.05$).
[1] Supplement DMI was calculated as the average of daily supplement intakes throughout the designated feeding period as measured by the SmartFeed Pro feeding system divided by the average body weight during that period on an individual basis.
[2] Values are means of 16 replicates (corn), 18 replicates (CGF), or 10 replicates (control).
[3] Supplements were formulated to be fed at 0.59% BW, 0.87% BW, and 1.13% BW for corn and CGF.
[4] Difference was calculated on an individual basis as the ADG of each calf during the period minus the group average ADG of the control group for that period.
[5] Calculated as additional gain relative the un-supplemented control over the 70-d period divided by total supplementation cost on an individual basis. Supplement costs were $0.20/kg DM for CGF and $0.24/kg DM for corn.

Table 3.5: Effect of supplement type and Supplement DMI[1] on fat thickness at the 12th rib of calves grazing fescue pasture and supplemented with either corn or corn gluten feed (CGF) over a 70-day treatment period.[2]

	Supplement[3]			*P* – value
	Corn	**CGF**	**SEM**	**Supplement**
12th rib fat thickness, cm[4]				
Day 0	0.23	0.23	0.01	0.72
Day 70	0.21	0.21	0.01	0.87
FCS[5]				
Initial	4.53	4.89	0.16	0.11
Final	4.95	5.49	0.14	<0.01

[1] Supplement DMI was calculated as the average of daily supplement intakes throughout the designated feeding period as measured by the SmartFeed Pro feeding system divided by the average body weight during that period on an individual basis.
[2] Values are means of 16 replicates (corn) or 18 replicates (CGF)
[3] Supplements were formulated to be fed at 0.59% BW, 0.87% BW, and 1.13% BW for corn and CGF.
[4] Fat thickness was measured at the 12th rib via ultrasonography.
[5] Flesh condition score was assigned on a scale of 1 to 9. A score of 1 indicates severely emaciated and a score of 9 indicates excessive fat.

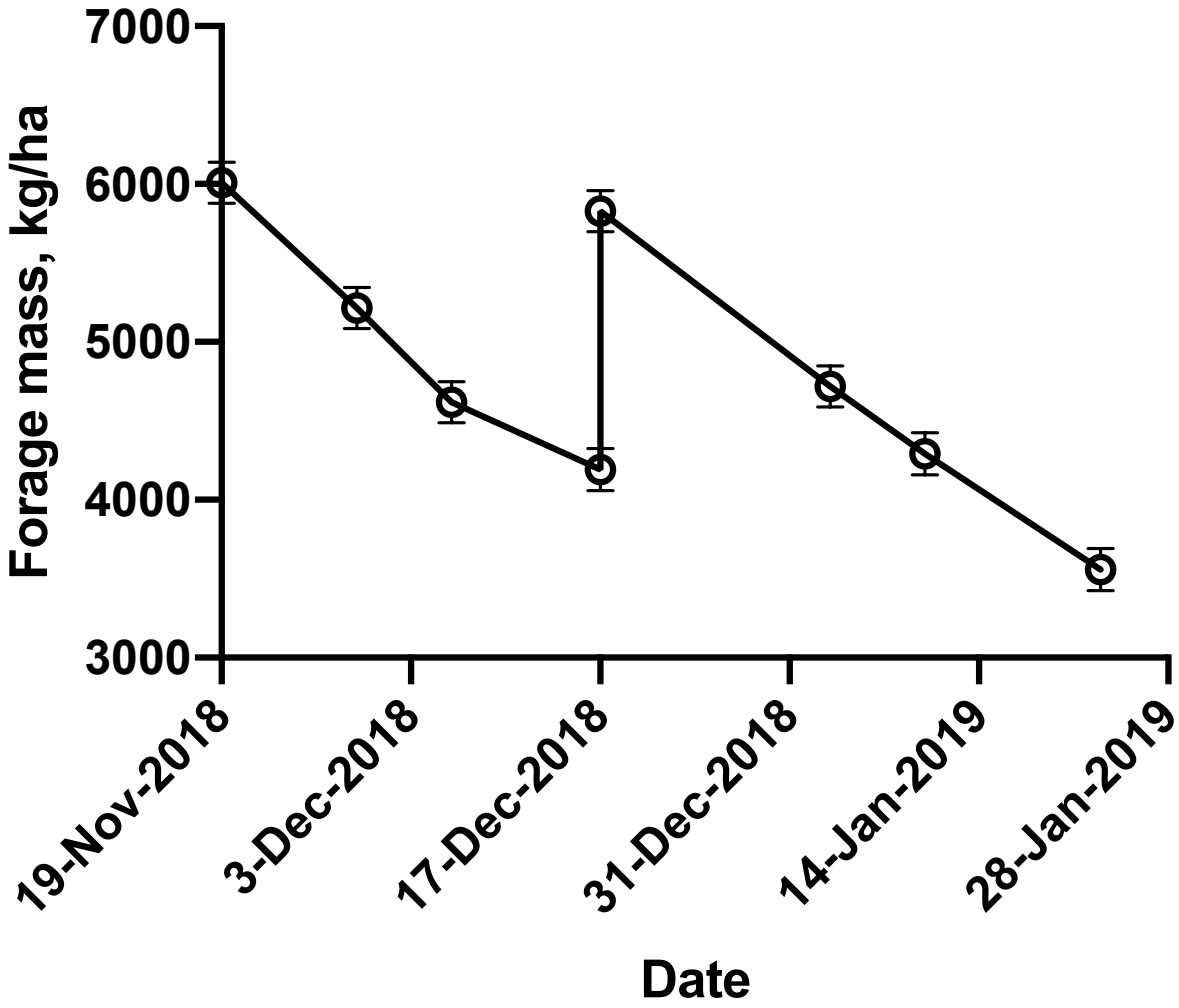
Figure 3.1: Nutritive value of stockpiled tall fescue forage grazed by beef cattle over a 70-d treatment period.[1,2]



[1] Values are means ± SEM for all pastures on a given day.

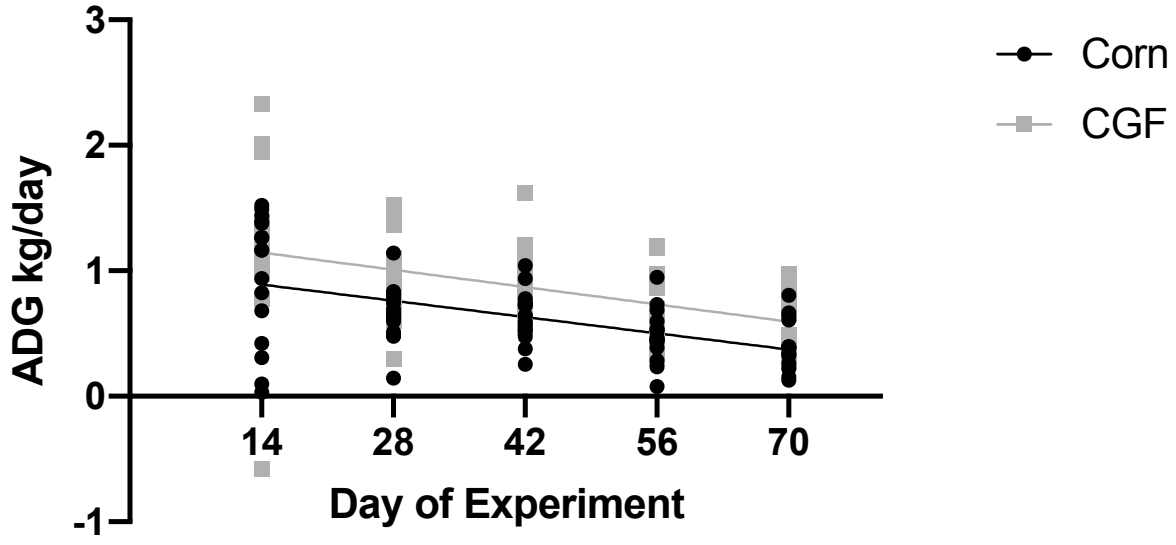
[2] $P < 0.05$ for NDF and EE. $P = 0.06$ for ADF and TDN over time. $P \geq 0.21$ for Ash and CP over time.

Figure 3.2: Forage mass of stockpiled tall fescue forage grazed by beef cattle over a 70-d treatment period.[1,2]



[1] Values are mean ± SEM for all pastures on a given day. Pastures were rotated on December 17, 2018.
[2] $P < 0.01$ for Date.

Figure 3.3: Cumulative average daily gain (ADG) from d 0 of calves grazing stockpiled tall fescue and supplemented with either corn or corn gluten feed (CGF) over a 70-d treatment period.



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CHAPTER IV

Determination of nutrient digestibility in calves supplemented with various levels of corn gluten feed when consuming forage-based rations

ABSTRACT

The objective of this experiment was to determine the effect of supplementing various levels of corn or corn gluten feed (**CGF**) on apparent total tract nutrient digestibility in cattle consuming a forage-based ration. Twelve purebred Angus and Simmental heifers were stratified across treatments by BW and breed in a 2 x 2 factorial arrangement across 2 periods (n = 6/treatment). Treatments included either corn or CGF supplemented at 0.25% BW or 0.75% BW with *ad libitum* access to alfalfa hay, with no animal receiving the same diet in both periods. Each period lasted 21 d, with animals being housed as a group during the first 10 d and individually thereafter. During the final 11 d of each period, 9.5 g of TiO_2 was provided via a bolus daily as an indigestible marker. Fresh alfalfa hay and supplement were provided to each animal daily, and feed offered and refused were recorded daily while animals were housed individually. During the final 3 d of each period, fecal samples were obtained directly from the rectum of each animal every 6 h, adjusted forward by 2 h each day to obtain a sample representative of each 2 h period of a 24 h cycle. Samples were pooled by animal within period and analyzed for neutral detergent fiber (NDF), organic matter (OM), and crude protein (CP). Data were analyzed using lm in R, and the model included the fixed effects of supplement type, level, the interaction between supplement type and level, breed, and period. Significance was defined at $P \leq 0.05$ and means were separated using LSD. No differences ($P \geq 0.28$) in ADG or feed efficiency were noted among treatments. Supplement intake was greater ($P < 0.01$) for animals supplemented at 0.75% BW compared to 0.25% BW, and hay intake was reduced ($P = 0.01$) for animals supplemented at 0.75% BW compared to 0.25% BW. Total tract NDF, ADF, and OM digestibility did not differ among treatments, but OM and ADF digestibility

tended to be greater (P = 0.10 and P = 0.06, respectively) for animals supplemented with corn compared to CGF. Crude protein digestibility was not influenced by supplement type (P = 0.46) but tended to be greater (P = 0.07) for animals supplemented at 0.25% BW compared to 0.75% BW. The results of this experiment indicate that at supplement levels below 0.75% BW, supplementing cattle with CGF does not result in an improvement in apparent total tract digestibility compared to corn.

INTRODUCTION

Previous research has demonstrated that supplementing cattle consuming a forage-based diet with corn gluten feed (**CGF**) results in a greater performance response when compared to corn supplementation in both a dry lot setting (Hardin, 2018) and in a pasture (Chapter III). At present, few data are available which characterize the mechanisms behind the observed performance responses. Previous research has demonstrated that increasing the amount of starch-based energy sources in forage-based diets results in a substantial reduction in fiber, and ultimately organic matter, digestibility (Faulkner et al., 1994; Garcés-Yépez et al., 1997; Russell et al., 2016). This is largely due to changes in the rumen microbial population, which subsequently results in changes in digestive efficiency. As starch concentration is increased in the diet, the increased concentrations of starch favor the growth of amylolytic bacteria and the proportion of fibrolytic bacteria decreases (Fernando et al., 2010). This ultimately results in a decreased ability to digest the fiber in forage and reduces growth and efficiency. Supplementation with fibrous feedstuffs, however, have been shown to impart beneficial effects on ADG, intake, and efficiency when fed to grazing cattle (Anderson et al., 1988; Martin and Hibberd, 1990; Richards et al., 2006). In fact, supplementation with soybean hulls has been shown to improve organic matter and nitrogen digestibility in forage-fed cattle compared to un-supplemented animals (Richards et al., 2006)

Since the majority of the starch present in corn co-products is removed during processing, the majority of the energy present in these feedstuffs is in the form of fiber. Therefore, it is likely that they will exhibit similar effects as fibrous supplements such as soybean hulls. Indeed, DDGS have been shown to increase the apparent total tract digestibility of both organic matter and NDF when supplemented at levels up to 1.2% BW in forage-based diets (Leupp et al., 2009). In fact, a

linear increase in organic matter and NDF digestibility was observed, which is likely due to the greater digestibility of the fiber in DDGS compared to forage.

Despite work investigating the effect of DDGS on total tract digestibility in cattle consuming forage-based diets, few data are available relating to CGF supplementation. Hannah et al. (1990) showed that CGF supplementation linearly increased apparent total tract NDF digestibility in heifers fed alfalfa haylage. However, much of their data focused on greater levels of CGF supplementation (> 40% of the diet). While the authors did find that supplementing CGF at 60% of DM resulted in greater NDF digestibility compared to corn, dietary supplement inclusion focused on moderate (20% of intake) or large (60% of intake) levels of inclusion. Many pasture-based growing strategies in the Southeastern U.S. rely on low to moderate inclusion of supplements in the diet, which likely influence the rumen microbial population differently.

Therefore, the objectives of this experiment were to evaluate the impact of supplementing either corn or CGF at 0.25% or 0.75% BW to heifers consuming an alfalfa hay-based diet on growth, intake, and apparent total tract digestibility.

MATERIALS AND METHODS

All animal procedures were approved by the Virginia Tech Institutional Animal Care and Use Committee (Protocol #: 19-087).

Animals, diets, and management

Twelve purebred Angus (n = 5) or Simmental (n = 7) heifers (16.8 ± 0.9 mo; 53.9 ± 40.0 days pregnant; 445 ± 46.8 kg at start of period 1; 467 ± 42.1 kg at start of period 2) were stratified by weight and breed and allotted to 1 of 4 treatments in a 2 x 2 factorial arrangement across 2 periods, resulting in a total of 6 experimental units for each treatment. Treatments consisted of

either corn or CGF supplemented at 0.25% BW or 0.75% BW, and *ad libitum* access to square-baled alfalfa hay. Each period lasted for 21 d, with 4 d between each period, and no animal received the same diet in both periods. At the beginning of each period, animals were adapted to their supplement at a rate of 25% of their daily supplement intake for that period per day over a total of 4 d until each animal was consuming their allotted supplement. Animals were housed as a group with *ad libitum* access to alfalfa hay and brought into individual stalls daily for feeding during the first 10 d of each period. From d 11 until the end of each period, animals were housed individually in stalls and fed their supplement once daily at approximately 0800. Stalls were cleaned and fresh alfalfa hay was provided daily. During the last 11 d of each period, animals were given a daily 9.5 g bolus of TiO_2 (Thermo Fisher Scientific, Waltham, MA) as an external marker. External marker was dosed by restraining animals in a squeeze chute each morning and using a balling gun to administer the marker in a single, size #10 gelatin capsule (Torpak Inc., Fairfield, NJ).

Animal measurements

Animals were weighed over 2 consecutive days at the beginning and end of each period, and the final weight of period 1 was used as the beginning weight of period 2. During the final 11 d of each period, all feed provided (hay and supplement) was weighed before feeding, and feed refusals were weighed back daily before feeding. All animals consumed the entirety of their supplement each day, and hay intake was managed to result in approximately 5 kg of refusals each day. Samples of refused hay were taken throughout the 11 d that animals were housed individually, weighed, and dried for 24 h at 105 °C to calculate refusal DM. The period average for each animal was then used to calculate hay refusals on a DM basis.

Fecal samples were collected from each animal during the final 3 d of each period. Briefly, animals were restrained in a squeeze chute and samples were taken directly from the rectum and placed into plastic cups. Lubricant was not used during collection to avoid potential contamination. Samples were collected every 6 h over the 3-d period, which was adjusted forward by 2 h each day to obtain a sample representative of every 2 h of a 24 h cycle. All fecal samples were stored at -20 °C until to analysis of titanium concentrations.

Feed analysis

Samples of each feed were collected once during each period. A bale core attached to a drill was used to obtain a single hay core from approximately 20 bales to obtain a representative sample. Subsamples of each were dried at 105 °C for 24 h to determine DM. The remaining sample was dried at 55 °C for at least 72 h, ground to pass through a 1 mm screen using a Wiley mill (Thomas Wiley Mill, Thomas Scientific, Swedesboro, NJ), and analyzed for crude protein (**CP**), neutral detergent fiber (**NDF**), acid detergent fiber (**ADF**), and ash. Crude protein was analyzed in duplicate by measuring the nitrogen content using an Elementar Vario EL Cube (Elementar Americas Inc., Ronkonkoma, NY) and multiplying by 6.25. An Ankom 200 fiber analyzer (ANKOM, Macedon, NY) was used to determine NDF and ADF following the manufacturers recommendations. Analysis of CP, ADF, and EE were conducted using Association of Official Analytical Chemists (1990) methods 990.03, 973.18, and 920.39, respectively. Neutral detergent fiber was measured using the methods described by Goering and Van Soest (1970). Ash was determined in duplicate by placing a 0.5 g sample in a furnace at 600 °C for 24 h.

Nutrient digestibility

Fecal samples were dried to a constant weight at 55 °C, ground to pass through a 1 mm screen using a Wiley mill, composited by animal within period, and analyzed for CP, NDF, ADF, and organic matter (**OM**). Organic matter was calculated in duplicate as the amount of sample lost during ashing.

Fecal samples were analyzed in duplicate for titanium concentration. Briefly, approximately 0.5 g of sample was ashed for 24 h at 600 °C. Approximately 10 mL of concentrated sulfuric acid was added to each crucible and the samples were boiled at 337 °C for 60 min. Samples were then filtered through Whatman 541 filter paper (Whatman, Inc, Buckinghamshire, UK) and diluted to 50 mL. Titanium concentration was determined using inductively coupled plasma atomic emission spectroscopy at 336.12 nm at the Virginia Tech Soil Science Laboratory (Blacksburg, VA). Titanium concentrations in the diet were calculated using the average weight of the TiO_2 bolus (accounting for the molecular weight of oxygen) and the average daily DMI over the final 11 d of each period. Apparent total tract digestibility was then calculated for CP, NDF, and OM using equation 4.1.

$$\boldsymbol{Digestibility, \% = 100 - \left(100 * \frac{Ti\ \%, feed}{Ti\ \%, feces} * \frac{\%\ nutrient\ feces}{\%\ nutrient\ feed}\right)} \quad \textbf{Eq. 4.1}$$

Statistical analysis

Animal within period was considered the experimental unit for all analyses. Data were analyzed using lm in R (R Foundation for Statistical Computing, Vienna, Austria) with significance defined at $P \leq 0.05$ and trends defined as $0.05 < P \leq 0.10$. Supplement type, supplement level, the interaction between supplement type and supplement level, period, and breed were included in the model as a fixed effects. Treatment means are reported as least squares means ± SEM.

RESULTS AND DISCUSSION

Growth performance

Growth performance data are shown in Table 4.3. No significant interactions were noted for ADG or G:F ($P \geq 0.28$). Neither supplement type nor level had an effect ($P \geq 0.11$) on ADG or G:F. While previous reports have shown that animals consuming a forage based ration and supplemented with CGF have greater ADG and G:F compared to those supplemented with corn (Hardin, 2018), the current experiment was not designed to measure differences in growth performance. The relatively low number of experimental units (n = 6 per treatment) combined with the short duration of each period make it difficult to capture differences in growth between the treatments. This is supported by the data in the previous chapter, as differences in ADG and G:F were not realized until animals had been on treatments for 28 d.

For all treatments, the concentration of crude protein in the diet was high (Table 4.2) and above the animal's nutrient requirements (Table 4.3). Previous data have shown that including excessive amounts of crude protein in the diet increases maintenance energy requirements due to the energy costs of nitrogen disposal (Jennings et al., 2018). However, this effect was only observed when animals were fed at twice their maintenance intake and there were no differences in performance. Therefore, it is unlikely that the excessive levels of CP in the current experiment influenced growth performance.

Given the high plane of nutrition for all diets (≥ 66% TDN and ≥ 16% CP) and relatively small differences between diets, it is not surprising the no differences in growth performance were observed among treatments. As these animals were yearling heifers that were nearing mature body weight, these diets exceeded the animal's nutrient requirements. Since nutrients were provided in excess of requirements, small changes in the amount of energy or protein are unlikely to influence

growth. Additionally, it should be noted that growth was over-predicted when the diets utilized in the current experiment were entered into the BCNRM. Therefore, it is likely that the oversupply of nutrients is greater than what is shown in Table 4.3.

There was a significant effect of period in which animals in period 1 had greater ($P = 0.05$) ADG compared to period 2. This is likely the result of the animal's growth curve slowing, as the animals would have been closer to their mature BW at the end of period 1.

Intake

Feed intake data are shown in Table 4.3. No significant interactions were noted for supplement or hay intake ($P \geq 0.22$). Supplement intake was not different ($P = 0.66$) among supplement types but was greater ($P < 0.01$) for animals supplemented at 0.75% BW compared to those supplemented at 0.25% BW. This is not surprising as treatments were designed such that the corn and CGF groups would have a similar level of intake with distinct differences between supplement level. Hay intake did not differ ($P = 0.65$) among supplement types, however, animals supplemented at 0.75% BW consumed less hay ($P = 0.01$) compared to those supplemented at 0.25% BW. Past research has demonstrated that increasing levels of supplement in the diet decreases forage intake in a variety of supplements (Adams, 1985; Hannah et al., 1990; MacDonald et al., 2007; Pavan et al., 2007). For example, total DMI was shown to increase quadratically while forage DMI decreased quadratically when feeding DDGS to cattle consuming a forage-based diet (Griffin et al., 2012). In the current experiment hay intake decreased by 1 kg for a 2.34 kg increase in supplementation, or a substitution rate of approximately 0.43 kg of hay per 1 kg of supplement. These data agree to the results published by Griffin et al. (2012) who

showed that greater levels of inclusion of DDGS linearly increased the substitution rate, with forage intake decreasing by approximately 0.4 kg per kg of DDGS when included at 3 kg.

While the rate at which forage is replaced by supplement in the ration lends itself well to increasing overall DMI and therefore ADG in grazing cattle, it is important to remember that this rate has been shown to linearly increase as supplement inclusion in the diet increases. Because of this, providing excessive levels of supplement to grazing cattle not only increases the overall cost of the ration, but likely also reduces pasture utilization by depressing forage intake.

There was an effect of period on hay intake in which animals consumed greater ($P < 0.01$) amounts of hay in period 2 compared to period 1. This is likely the result of reduced NDF content in period 2 compared to period 1, as greater quantities of fiber will limit the animal's intake.

Nutrient digestibility

Apparent total tract digestibility data are shown in Table 4.4. No significant interactions between supplement type and level were noted for the digestibility of OM, CP, or NDF ($P \geq 0.13$), although there was a significant ($P = 0.05$) interaction for ADF digestibility. This was particularly surprising for NDF digestibility. We hypothesized that low levels of corn supplementation would have minimal impact on, or even improve, NDF digestibility, and that greater levels of corn supplementation would impair it, while CGF supplementation would improve NDF digestibility at both levels. This hypothesis was based on the results reported by Russell et al. (2016) who showed that NDF digestibility decreases quadratically as corn supplementation is increased in a forage-based ration, as well as Leupp et al. (2009) who reported that NDF digestibility increased linearly with greater levels of DDGS supplementation. However, there are several differences between these experiments and the current experiment that could explain the lack of interaction.

While the diets utilized by Russell et al. (2016) were fiber-based, the fibrous feedstuff included in the diets was pelleted soybean hulls. Additionally, the rations were fed as a TMR with a variety of ingredients rather than a simple, two component diet. In their experiment, the decrease in NDF digestibility appeared when the concentration of corn in the diet was between 16 and 31% of DM. In the current experiment the proportion of supplement in the ration was approximately 9% for the cattle fed at 0.25% BW and 25% for the cattle fed at 0.75% BW. It is possible that the large differences in the basal diets between the current experiment and what was used in the experiment conducted by Russell et al. (2016) led to differences in the inclusion rate of corn that results in negative associative effects. Additionally, while no significant differences were reported for the interactive effect, the geometric means may suggest that differences could have been realized had a third treatment of greater inclusion been used in the experiment, or had the experiment utilized more animals per treatment.

To our surprise, no differences ($P = 0.20$) among supplement types were noted for NDF digestibility. While little data are available comparing the effect of corn to CGF supplementation on NDF digestibility, Hannah et al. (1990) reported that bulls supplemented with CGF at 60% of DMI had greater NDF digestibility compare to those supplemented corn in an alfalfa haylage-based ration. This treatment was most similar to the cattle supplemented CGF at 0.75% BW, and it is possible that corn supplementation results in a greater improvement in NDF digestibility at lower levels. For example, Russell et al. (2016) reported nearly a 6% increase in apparent total tract digestibility of steers supplemented 8% corn compared to those receiving no corn in a forage-based diet. In contrast, Leupp et al. (2009) reported a much more modest improvement when DDGS were supplemented. Therefore, it appears that the additional energy provided by corn when supplemented at low levels does not impair NDF digestibility. Additionally, it is possible that the

levels that we fed the supplements at were not great enough to result in negative associative effects that could be captured by total tract digestibility.

Interestingly, ADF digestibility was greater for animals supplemented with corn compared to CGF at 0.25% BW, but this difference disappeared at the 0.75% BW level. Russell et al. (2016) demonstrated a quadratic effect of corn level supplementation on ADF digestibility, with levels below 16% of DM improving ADF digestibility and greater levels reducing it. The improvement in ADF digestibility at low corn inclusion levels reported by Russell et al. (2016) may explain why ADF digestibility was greater for cattle supplemented with corn at the 0.25% BW but not the 0.75% BW in the current experiment.

Total tract ADF digestibility tended ($P = 0.06$) to be greater for animals supplemented with corn compared to CGF. However, this is due to the improvement observed at the 0.25% BW level rather than the 0.75% BW level.

The lack of interaction between supplement type and interaction for the digestibility of OM agrees with the results published by Russell et al. (2016) and Leupp et al. (2009). In both experiments, supplementation with corn (Russell et al., 2016) and DDGS (Leupp et al., 2009) resulted in a linear increase in OM digestibility. Therefore, it is unlikely that negative associative effects in the rumen impact OM digestibility in the way that they have been shown to impact fiber digestibility.

Apparent total tract CP digestibility was not affected ($P = 0.46$) by supplement type and OM digestibility tended ($P = 0.10$) to be greater for cattle supplemented with corn compared to CGF. Although there were no differences in NDF or CP digestibility, corn contains the majority of its energy in the form of highly digestible non-fiber carbohydrates. Therefore, it is likely that

the greater concentration of non-fiber carbohydrates ultimately resulted in greater OM digestibility for animals supplemented with corn compared to CGF.

Supplement level did not affect ($P \geq 0.63$) NDF, ADF, or OM digestibility but tended ($P = 0.07$) to reduce CP digestibility for animals supplemented at 0.75% BW. This is thought to be due to the greater digestibility of protein in alfalfa relative to either supplement. Previous literature has stated that the apparent total tract CP digestibility of alfalfa hay can range from between 70% (Horner et al., 1985) to nearly 80 – 90% (Robinson, 1998). Therefore, replacing greater amounts of the forage with supplement will ultimately reduce CP digestibility if the CP present in the supplement is less digestible than that of the forage. However, in most cases beef cattle are not fed alfalfa as their sole forage source, so it is likely that this effect would be reduced if the animals were provided a grass hay with less digestible CP.

There was a significant effect of period ($P < 0.01$) for NDF, ADF, and CP digestibility. These effects are thought to be because of the relatively large differences in the nutritive value of the alfalfa hay between the periods.

Conclusions

Contrary to our hypothesis, the results of this experiment do not demonstrate an improvement in fiber digestibility when animals are supplemented with CGF compared to corn below 0.75% BW, or 25% of DM. While this does not explain the reason for the improvement in performance that has been observed when cattle consuming a forage-based diet are supplemented with CGF compared to corn, these results suggest that it is not related to total tract fiber digestibility at levels below 25% of DM. Therefore, cattle producers who supplement grazing

cattle can supplement animals with the more inexpensive, more accessible feed without negatively impacting total tract fiber digestibility.

Table 4.1: Composition of feedstuffs fed to heifers receiving *ad libitum* hay and corn or corn gluten feed (CGF) at 0.25% or 0.75% BW over two 21-d periods.

		Analyzed Concentration, % DM[1]			
	DM, %	**CP**	**NDF**	**ADF**	**Ash**
Alfalfa Hay					
Period 1	89.8	16.0	51.2	35.5	7.2
Period 2	92.7	19.8	37.3	22.3	7.0
CGF[2]	88.6	22.3	34.6	9.7	7.2
Cracked Corn[2]	85.9	8.4	15.0	2.3	1.6

[1] DM = dry matter; CP = crude protein; NDF = neutral detergent fiber; ADF = acid detergent fiber.
[2] Values are from samples that were composited across periods.

Table 4.2: Nutrient composition of diets fed to heifers receiving *ad libitum* hay and corn or corn gluten feed (CGF) at 0.25% or 0.75% BW over two 21-d periods.

	Analyzed Concentration, % DM[1, 2]				
Treatment	NDF	ADF	CP	OM	TDN[3]
Corn, 0.25% BW	41.1	26.5	17.8	93.3	67.4
CGF, 0.25% BW	43.4	27.1	19.0	92.8	66.7
Corn, 0.75% BW	35.4	22.2	16.2	94.2	70.8
CGF, 0.75% BW	41.9	24.2	19.5	92.8	69.1

[1] Nutrient values are the averages across all animals in a given treatment using analyzed nutrient content and intake data.

[2] DM = dry matter; CP = crude protein; NDF = neutral detergent fiber; ADF = acid detergent fiber.

[3] Calculated using the TDN content of individual feedstuffs and animal intake. The TDN content of corn grain and CGF were obtained from the values published by the NRC (2016). The TDN content of the alfalfa hay was calculated from ADF concentrations using the equation: TDN, % = 88.875 – (0.812 * ADF) and averaged across periods.

Table 4.3: Nutrient requirements and supply of heifers receiving *ad libitum* hay and corn or corn gluten feed (CGF) at 0.25% or 0.75% BW over two 21-d periods.

	Maintenance Requirements[1]		**Growth Requirements[1,2]**		**Supply[1]**		**Balance, %**	
	ME, Mcal/d	MP, g/d	ME, Mcal/d	MP, g/d	ME, Mcal/d	MP, g/d	ME	MP
Corn, 0.25% BW	11.54	365.1	18.88	478.9	30.51	887.3	100.3	105.1
CGF, 0.25% BW	11.57	365.1	19.31	484.9	30.97	928.4	100.3	109.2
Corn, 0.75% BW	11.42	365.1	24.93	636.4	33.21	970.7	100.2	106.3
CGF, 0.75% BW	11.43	365.1	22.76	578.4	34.21	1094.1	100.3	116.0

[1] Determined by entering animal and feed characteristics in the 2016 BCNRM. Requirements are requirements for maintenance.

[2] The model over-predicted growth at an average of 1.97 kg/d across treatments. Average observed ADG was 0.80 kg/d.

Table 4.4: Initial body weight (IBW), final body weight (FBW**),** average daily gain, intake, and gain:feed of purebred heifers fed *ad libitum* alfalfa hay and supplemented with either corn or corn gluten feed (CGF) at 0.25% or 0.75% BW.[1]

	IBW kg	**FBW kg**	**ADG[2] kg/d**	**Hay intake[2] kg/d**	**Supplement intake kg/d**	**G:F[2]**
Corn, 0.25% BW	448	464	0.77	11.4	1.12	0.061
CGF, 0.25% BW	462	473	0.52	11.7	1.14	0.043
Corn, 0.75% BW	455	473	0.85	10.8	3.42	0.060
CGF, 0.75% BW	468	490	1.06	10.2	3.51	0.079
SEM	19.6	18.5	0.22	0.38	0.11	0.017
P - value	0.98	0.82	0.31	0.22	0.74	0.28
Main effect of supplement type						
Corn	451	468	0.81	11.1	2.27	0.060
CGF	465	481	0.79	10.9	2.32	0.061
SEM	13.7	13.0	0.15	0.26	0.08	0.012
P - value	0.49	0.48	0.92	0.65	0.66	0.96
Main effect of supplement level						
0.25% BW	455	468	0.64	11.5[a]	1.13[b]	0.052
0.75% BW	461	482	0.95	10.5[b]	3.47[a]	0.069
SEM	14.0	13.3	0.16	0.27	0.08	0.012
P - value	0.86	0.55	0.11	0.01	<0.01	0.21

[a-b] Means within the same column not sharing a common superscript differ significantly ($P \leq 0.05$)

[1] n = 6 per treatment for the interactive term.

[2] A significant effect of period was observed ($P \leq 0.05$).

Table 4.5: Apparent total tract nutrient digestibility of purebred heifers fed *ad libitum* alfalfa hay and supplemented with either corn or corn gluten feed (CGF) at 0.25% or 0.75% BW.[1]

	NDF digestibility[2] %	ADF digestibility[2] %	CP digestibility[2] %	OM digestibility %
Corn, 0.25% BW	70.5	64.6[a]	74.8	77.6
CGF, 0.25% BW	64.7	54.4[b]	73.5	73.1
Corn, 0.75% BW	66.1	58.5[ab]	68.0	74.4
CGF, 0.75% BW	66.2	58.6[ab]	72.1	74.2
SEM	2.15	2.53	1.87	1.4
P - value	0.19	0.05	0.16	0.13
Main effect of supplement type				
Corn	68.3	61.5	71.4	76.0
CGF	65.5	56.5	72.8	73.6
SEM	1.51	1.78	1.31	0.98
P - value	0.20	0.06	0.46	0.10
Main effect of supplement level				
0.25% BW	67.6	59.5	74.1	75.4
0.75% BW	66.1	58.5	70.0	74.3
SEM	1.54	1.81	1.34	1.0
P - value	0.66	0.96	0.07	0.63

[a-b] Means within the same column not sharing a common superscript differ significantly ($P \leq 0.05$)

[1] n = 6 per treatment for the interactive term.

[2] A significant effect of period was observed ($P \leq 0.05$).

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CHAPTER V
Determining the rate of ergot-alkaloid degradation in toxic endophyte infected tall fescue hay and haylage during long term storage

ABSTRACT

The objectives of this experiment were to determine changes in the nutritive value and ergot alkaloid concentrations of endophyte-infected tall fescue hay and haylage during a 180-d storage period. A field containing Kentucky-31 tall fescue was clipped in late June and allowed to dry in the field. Forage was sampled from the windrow of cut forage in 6 location blocks, and treatments included haylage and hay. Samples of each treatment were taken in all 6 blocks. The dry matter of the windrow was measured every 2 h after clipping, and haylage samples were taken when the dry matter of the windrow reached 50%. Hay samples were taken when the dry matter of the windrow reached 80%. Seven subsamples of each treatment within block were chopped to 1.91 cm in length and vacuum sealed in oxygen-excluding bags. The bags were then opened at 30 d intervals over a 180-d period and samples were taken for analysis of pH, nutritive value, and individual ergot alkaloid concentrations. Treatment within block on each storage day was considered the experimental unit. Data were analyzed in SAS using the MIXED procedure, and fixed effects included treatment, day, and the treatment by day interaction. Location block was included in the model as a random effect. As expected, pH was decreased for haylage compared to hay at all time points ($P < 0.01$). Dry matter was greater ($P < 0.01$) for hay compared to haylage and did not change over time ($P = 0.37$). Crude protein did not differ by treatment ($P = 0.25$) but was different by day ($P = 0.06$), but this difference was small and likely of no biological significance. Neutral detergent fiber values were greater ($P < 0.01$) for hay compared to haylage and declined during storage ($P < 0.01$). Few differences were observed for ADF concentrations, despite an interaction in which hay was greater than haylage on day 30 while haylage was greater than hay on day 120

($P < 0.01$). Total ergot alkaloid concentrations did not differ by treatment ($P = 0.61$), but ergovaline concentrations declined ($P < 0.01$) during storage. Collectively, these results indicate minimal differences in nutritive value and ergot alkaloid concentrations between hay and haylage during storage, and that ergovaline concentrations decline during storage.

INTRODUCTION

Tall fescue (*Lolium arundinaceum*) is an important cool-season forage grown in the Southeastern U.S. It has historically been a popular forage in the region due to its wide-range of adaptability, and tolerance to cold, shade, drought, and grazing pressure. This tolerance is due largely to a symbiotic relationship between the plant and a fungal endophyte, *Neotyphodium coenophialum* (Arachevaleta et al., 1989; Pedersen et al., 1990). This fungus lives between the plant's cells and produces ergot alkaloids, several of which are toxic to livestock (Pirelli et al., 2016). Symptoms of toxicosis include a rough hair coat, vasoconstriction, elevated body temperature, and reduced milk production, feed intake, and growth (Roberts and Andrae, 2004). Vasoconstriction and rough hair coats can exacerbate heat stress in the summer months, further decreasing dry matter intake (**DMI**) and weight gain (Paterson et al., 1995). Losses associated with fescue toxicosis have been estimated to result in losses greater than $1 billion in 2011 (Strickland et al., 2011) and $3 billion in 2015 (Kallenbach, 2015), and it is estimated that greater than 25% of the U.S. beef cattle population are exposed to E+ tall fescue (Bussard and Aiken, 2012).

Researchers have proposed a number of alternatives to mitigate economic and performance losses associated with fescue toxicosis. These solutions include diluting the ergot alkaloid concentration in the diet with legumes or supplemental feeds, preventing the accumulation of seed heads in pastures, managing grazing systems to minimize ergot alkaloid intake, and renovation of pastures (Kallenbach, 2015). However, with the exception of complete renovation, these solutions do not eliminate ergot alkaloid intake and are therefore not ideal solutions. Ergovaline, the principal ergot alkaloid responsible for decreased performance, has been shown to negatively affect performance at concentrations below 100 μg/kg (Liebe and White, 2018). While pasture renovation removes the toxic forage from animals' diets, it is costly, and adoption of this practice

has not been widespread. Caldwell et al., 2013 found that partial pasture renovation along with grazing E+ tall fescue during non-critical periods in the cow-calf production cycle was beneficial, but producers must still set aside a portion of their pasture land to accomplish this.

Previous research has demonstrated that ergot alkaloid concentrations are greater in fresh forage compared to hay (Norman et al., 2007), and concentrations decline during the curing and storage process (Roberts et al., 2009). This decline is largely due to exposure to light, as ergot alkaloids are sensitive to degradation when exposed to UV radiation and heat (Garner et al., 1993). However, experiments investigating this effect in ensiled tall fescue have not noted the same level of decline. Roberts et al. (2014) noted that although there was a decline in ergovaline concentrations after 100 d of ensiling, TEA concentrations actually increased. Therefore, current recommendations advise against the ensiling of tall fescue as haylage or baleage due to the lack of decline in ergot alkaloid concentrations. However, the results reported by Roberts et al. (2014) only measured ergot alkaloid levels 100 d post-harvest, and in many cases, producers would not be feeding the forage until much later. Therefore, it is possible that the results of their experiment do not accurately capture the true levels of ergot alkaloids at time of feeding.

The production of ensiled tall fescue provides producers with the benefit of optimal harvest time in the mid to late spring, when weather conditions tend to be wet relative to early summer. Currently, producers are left with delaying harvest until conditions are favorable for hay production, which results in less nutritious, mature forage, and can reduce the total yield of hay fields through fewer cuttings. If ensiled tall fescue can be successfully made and managed to mitigate the impacts of fescue toxicosis, producers could harvest forage to balance for yield and quality rather than being as reliant on weather conditions. This can result in needing less purchased forages during the late fall and winter months, ultimately improving profits.

The objectives of this experiment were to characterize ergovaline and TEA levels in toxic endophyte-infected (**E+**) tall fescue that was made into hay or ensiled over a 180-d storage period. This storage period was selected to build on the results of previous research and mimic a storage period more similar to typical forage production systems. Additional objectives were to characterize changes in nutritive value during storage.

MATERIALS AND METHODS

Experimental design

This experiment was designed as a randomized complete block design with 2 treatments, hay or haylage. An established toxic endophyte-infected tall fescue field at the Virginia Tech College Farm (Blacksburg, VA) was randomly divided into 6 location blocks and mowed in late June 2019. Samples from each block were taken when the DM of the windrow reached approximately 50% (haylage) and 85% (hay). Each sample was then divided into 7 subsamples that were stored for increasing amounts of time to represent storage over a 180 d period.

Forage harvest

This experiment was conducted in 2019 in Blacksburg, VA (37.22 °N, -80.46 °W) on Groseclose and Poplimento silt loam soils with a 2 to 7% slope. Forage was harvested by the Virginia Tech College Farm (Blacksburg, VA). The pasture was an established pasture used for hay production that had no N applied to it that year. Forage species composition was 75.7% grass, 19.7% clover, 1.6% thistle, and 3.0% weed and was determined using the step-point method described by Evans et al. (1957), with a total of 300 observations. The forage mass of the field at the time of cutting was determined by clipping three 0.25 m^2 quadrats to ground level and drying

at 105 °C for at least 24 h. Additional forage grab samples were taken at the time of clipping, frozen at -20 °C, and shipped to Agrinostics (Agrinostics Ltd., Watkinsville, GA) for analysis of TEA concentrations by ELISA. The initial forage mass was approximately 5,655 ± 1528 kg/ha and TEA concentrations were 2,038 μg/kg.

An approximately 0.57 ha section of pasture was cut at the location on June 23, 2019. The field was blocked into 6 randomly selected locations that were marked. The following morning, the DM of the forage was measured approximately every 2 h using a microwave oven. Subsamples were taken from each location and composited. A 50 g subsample of the composited forage was then placed in the microwave and dried to a constant weight. Haylage samples were taken the following morning from each location when the DM content reached approximately 50%. Hay samples were taken from the same 6 locations on June 27, when the DM content of the forage reached approximately 80%. The forage was tedded once between the collection of haylage and hay samples, and it was noted that there was cloud cover during the afternoon of June 24 and during the day on June 25. Samples from each block were placed into large trash bags and frozen at -20 °C immediately after collection, prior to processing on June 27.

Processing

Sample processing occurred over a 3-d period. A total of 7 subsamples for each treatment within block were created to measure changes over 6 months. A 300 g (wet) subsample for haylage, and 150 g subsample for hay were chopped to 1.91 cm using a commercial lettuce chopper (Vollrath, Sheboygan, WI) and sealed in a plastic food saver bag using a vacuum sealer (Weston Brands LLC, Southern Pines, NC). Bags from each block (both haylage and hay treatments) were placed into light excluding plastic totes and stored indoors at room temperature. The room

averaged 21 °C ± 1.3 °C and 33.9% ± 7.7% humidity. Initial samples (d 0) were immediately frozen at -20 °C after sealing. Bags from each treatment within block were opened every 30 d from June until December to measure changes during storage over a 180-d period (samples processed after 0, 30, 60, 90, 120, 150, and 180 d of storage).

Sampling procedure

One sample from each treatment within block was opened on 30-d intervals. If processing could not occur that day, samples were stored at -20 °C until they could be processed. Each bag was split into 3 subsamples of either 100 g (haylage) or 50 g (hay). Measurement of pH was determined on one of these subsamples according to the procedures outlined by Roberts et al. (2014). Briefly, 25 g of wet forage was weighed and placed into a food processor with 250 mL of nanopure water. The sample was then ground using the food processor for approximately 60 sec, and the homogenate was filtered through 2 layers of cheese cloth into a beaker. A Fisher Scientific accumet AE150 benchtop pH meter (Thermo Fisher Scientific, Waltham, MA) was then used to record the pH of the homogenate. This process was then repeated, and the duplicate reading was averaged for each sample. The pH meter was calibrated once using a 2-point curve with pH standard solution according to the manufacturer's recommendations prior to sample processing. A second subsample was then weighed, placed into an aluminum pan, and dried at 55 °C for at least 72 h for analysis of nutritive value. The final subsample was then stored at -20 °C until analysis of ergot alkaloids.

Nutrient analysis

Following drying, samples were weighed to determine dry matter (**DM**) and ground to pass through a 1 mm screen using a Wiley mill (Thomas Wiley Mill, Thomas Scientific, Swedesboro, NJ), and analyzed for crude protein (**CP**), neutral detergent fiber (**NDF**), acid detergent fiber (**ADF**), and ash. Crude protein was analyzed in duplicate by measuring the nitrogen content using an Elementar Vario EL Cube (Elementar Americas Inc., Ronkonkoma, NY) and multiplying by 6.25. An Ankom 200 fiber analyzer (ANKOM, Macedon, NY) was used to determine NDF and ADF following the manufacturers recommendations. Analysis of CP, ADF, and EE were conducted using Association of Official Analytical Chemists (1990) methods 990.03, 973.18, and 920.39, respectively. Neutral detergent fiber was measured using the methods described by Goering and Van Soest (1970).

Analysis of ergot alkaloids

Forage samples held for analysis of ergot alkaloids were frozen at -80 °C for at least 12 h prior to freeze-drying over a 10-d period. Freeze-dried samples were then shipped to the Veterinary Medical Diagnostic Laboratory at the University of Missouri (Columbia, MO) for the analysis of ergovaline and Clavicep's ergot alkaloids concentration by high-performance liquid chromatography (**HPLC**) with fluorescence detector using the procedures of Rottinghaus et al. (1991) with modifications reported by Hill et al. (1993) and Rottinghaus et al. (1993), respectively.

Freeze-dried ground samples (5.0 g) were extracted using 100 mL chloroform and 5 mL of aqueous 0.1 M sodium hydroxide overnight. The extract (20 mL) was filtered through Whatman PS-1 filter paper, and 10 mL of filtrate was passed through an Ergosil cleanup column to remove pigments with 1.5 mL of chloroform/acetone (80/20) followed by 3 mL of petroleum ether. Ergovaline and ergot alkaloids were eluted from the cleanup column with 2 mL methanol and

passed through an M-224 column to remove any remaining pigments if necessary. Samples were then analyzed by Hitachi HPLC with Model 5110 pump, Model 5210 autosampler, Model 5440 fluorescence detector, and Hitachi Chromaster software on a microcomputer. The HPLC column was a Phenomenex Luna C18 column (100 x 4.6 mm, 3 µm, PN: 00D-4521-E0) with a Phenomenex guard cartridge (PN: KJ0-4282) and C18 guard column (4.0 x 3.0, PN: AJ0-4287). The mobile phase was acetonitrile and water with 20% ammonium carbonate with an injection volume of 20 µL at a flow rate of 1.0 mL/min at room temperature. The total run time for ergovaline and ergot alkaloids was approximately 12 min and 22 min per sample, respectively. Total ergot alkaloids are defined as the sum of the ergovaline, ergosine, ergotamine, ergocornine, ergocryptine, and ergocrystine concentrations, and other ergot alkaloids are defined as the sum of ergocornine, ergocryptine, and ergocrystine concentrations.

Statistical analysis

Statistical analysis was performed using SAS (SAS Institute Inc., Cary, NC) with significance defined at $P \leq 0.05$ and trends defined as $0.05 < P \leq 0.10$. This experiment was analyzed as a repeated measures design using the MIXED procedure. The model included the fixed effects of treatment, sampling day, and the interaction, and the random effect of location. Day was considered the repeated effect, and treatment within block was the repeated subject with the compound symmetry covariance structure. This covariance structure was chosen as the within-subject correlation was expected to be constant over time. Pairwise comparisons of simple main effects were conducted using the SLICE statement when there was a significant interaction.

RESULTS AND DISCUSSION

pH

Data for pH are shown in in Figure 5.1. The pH of haylage was lower ($P < 0.01$) than that of hay and declined to approximately 4.5 during storage. This pH was similar to that reported by Roberts et al. (2014) for high moisture tall fescue silage, as well as those reported by Turner et al. (1993) for tall fescue ensiled using mini silos. As expected, the pH of hay did not experience a similar decline. These data suggest that we were successful in ensiling of the haylage treatment and adequately drying of the hay treatment.

Nutritive value

Dry matter and CP data are shown in Figure 5.2 and NDF and ADF data are shown in Figure 5.3. As would be expected by the experimental design, DM was significantly greater ($P < 0.01$) for hay compared to haylage and did not differ by day ($P = 0.37$). Since the storage bags were completely sealed and in a closed system, no changes in moisture content were anticipated throughout the storage period.

There was no treatment by day interaction for NDF, but Surprisingly, NDF was greater ($P < 0.01$) for hay compared to haylage and there was an effect of day ($P < 0.01$). The windrows were tedded once between the collection of haylage and hay samples, and it is possible that this resulted in some leaf losses that increased the proportion of fiber in hay compared to haylage. Since the pasture had a relatively high proportion of clover (20%), the relative fragility and faster drying of the clover leaves is likely what led to this leaf loss. The decrease in NDF with time is likely the result of microbial degradation of fiber. Desta et al. (2016) reported that structural carbohydrate concentrations decline throughout storage when napier grass is ensiled. As would be expected, this effect was increased through the addition of fibrolytic enzymes in both the results reported by Desta et al. (2016), as well similar results reported by Dehghani et al. (2012).

The ADF concentration data are somewhat less clear, as there was a treatment by day interaction ($P < 0.01$). The ADF concentration of hay was greater than that of haylage on day 30, but the opposite was true on day 120. No other differences among treatments were noted. Because of the lack of differences among treatments on other days, as well as the conflicting results on day 30 and 120, there were likely no biologically significant differences in ADF concentrations. Similarly, the results reported by Desta et al. (2016) showed little differences in the ADF concentration of ensiled napier grass over a 90-d period. Previous work with ensiled wheat straw has indicated that there is an apparent decrease in NDF concentrations over time, primarily being driven by a reduction in hemicellulose concentrations with little change in cellulose or ADF concentrations (Singh et al., 1996). Combined with the results of the current experiment, it is likely that changes in fiber concentration during the storage of forage are due primarily to the degradation of hemicellulose, with minimal impact on cellulose.

There was no treatment by day interaction ($P = 0.78$) nor effect of treatment ($P = 0.25$) for CP. However, there was to be an effect of day ($P = 0.03$). Previous research has shown that CP values are not affected by storage time nor ensiling for various species of ensiled forage (Singh et al., 1996; Snyman and Joubert, 1996; Polan et al., 1998). In the current experiment the differences across time are relatively small. Therefore, it is likely that this difference has little biological significance. In fact, given the methods of analyzing CP and the design of the current experiment it would be surprising for there to be differences in CP concentration. The standard method of analyzing CP is to measure the concentration of nitrogen in the sample and estimate the CP content from the average nitrogen content of amino acids (AOAC, 1995). Given that the forage is sealed in a closed system, there should not be any mass transfer into or out of the system. Therefore, it is possible that CP is being utilized and metabolized by bacteria, resulting in nitrogen taking a

different form, but the mass of nitrogen should remain constant. It would then be assumed that analysis of nitrogen content would remain similar regardless of protein metabolism, resulting in similar CP content.

Individual ergot alkaloids

Ergovaline concentrations for each treatment over time are shown in Figure 5.4 panel A, and the main effect of day is shown in Figure 5.4 panel B. Although there were no differences among treatments ($P = 0.96$), there was an effect of day ($P < 0.01$) in which ergovaline declined after 60 d of storage and then plateaued. These data agree with the results reported by Roberts et al. (2014) who measured ergovaline concentrations in tall fescue at baling and after 100 d of ensiling. However, Turner et al. (1993) found no differences in ergovaline concentrations in tall fescue mini silos over 60 d. Given that there were no treatment effects, these data show that ergovaline concentrations decline in harvested E+ tall fescue regardless of moisture content at baling. However, it should be noted that the ergovaline concentrations in both treatments at the conclusion of this experiment were greater than 100 µg/kg. Liebe and White (2018) demonstrated that the threshold of ergovaline concentrations to induce clinical tall fescue toxicosis is approximately 60 µg/kg. Therefore, these forages would still likely induce fescue toxicosis when fed to animals despite the decline in ergovaline concentrations. Additionally, the lack of change in ergovaline concentrations after 60 d suggests that the majority of the decrease occurs during the initial ensiling period.

No differences were noted for ergosine or ergotamine for the effects of treatment, day, or the interaction of treatment and day (Figure 5.5). Ergovaline has long been associated with fescue toxicosis and is generally considered the principal alkaloid that induces clinical symptoms (Tor-

Agbidye et al., 2001). Because of this, much of the research regarding managing tall fescue toxicosis in harvested forage has focused on ergovaline and total ergot alkaloid concentrations (Turner et al., 1993; Tor-Agbidye et al., 2001; Roberts et al., 2002; Roberts et al., 2009; Roberts et al., 2014). However, other ergot alkaloids such as ergotamine have been demonstrated to induce physiological responses, such as vasoconstriction (Foote et al., 2011). Data from the current experiment demonstrate that although they were present in relatively large concentrations, there were no effects of treatment or ensiling time ($P > 0.14$) on ergosine and ergotamine concentrations.

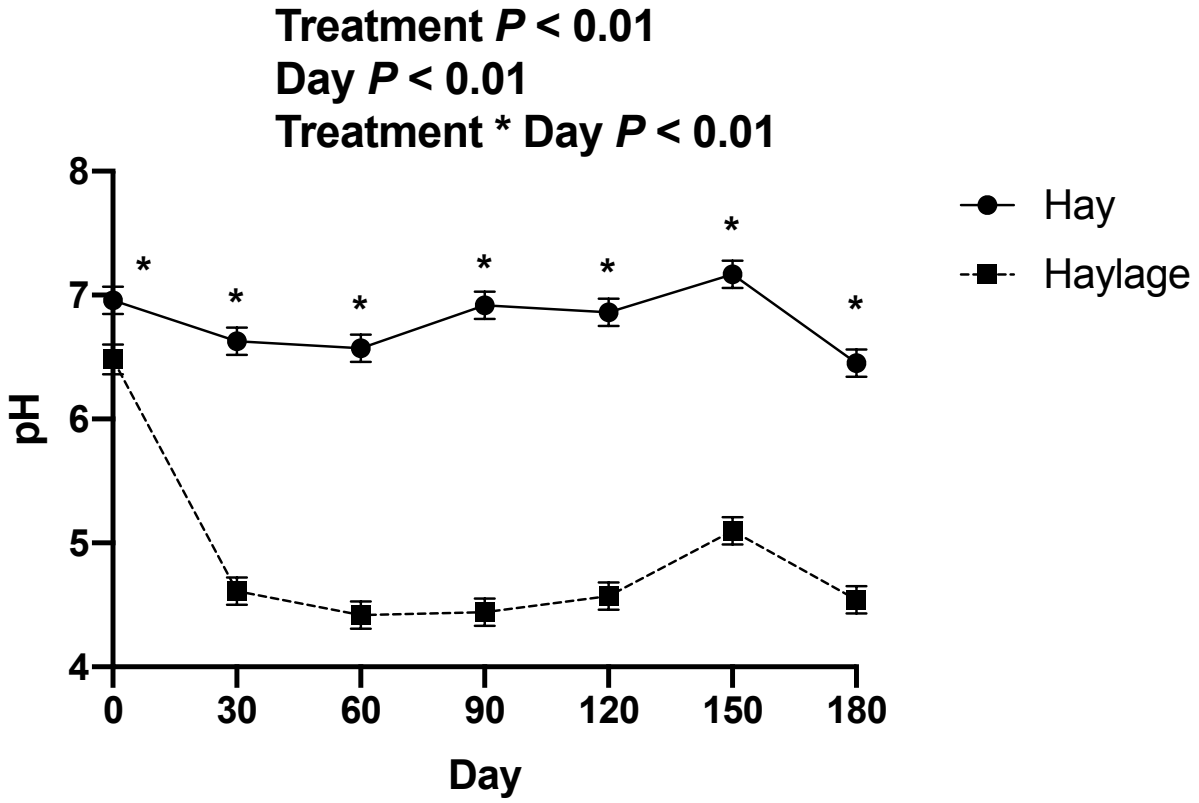
Interestingly, there tended to be an interaction and effect of treatment for other ergot alkaloids ($P = 0.10$ and $P = 0.09$, respectively). Other ergot alkaloids tended to be greater in haylage compared to hay on d 180 but were similar prior to that. This was surprising given that there were no differences noted for TEA, ergovaline, ergosine, or ergotamine. However, it should be noted that the alkaloids included in this measurement were present at concentrations of approximately 100 μg/kg. Therefore, differences present in these compounds could easily be masked by the aforementioned compounds. Although they are present in relatively low concentrations, ergocornine, ergocryptine, and ergocristine have all been shown to induce vasoconstriction of the right ruminal artery *in vivo* (Foote et al., 2011). Therefore, it is possible that the increased concentrations of these alkaloids on day 180 in haylage relative to hay could result in negative effects on animal performance.

Surprisingly, there was no significant interaction nor effect of treatment or day ($P = 0.32$) for TEA concentrations. Previous data have shown that TEA concentrations slowly decline in stored hay (Roberts et al., 2009), and at greater moisture levels, even increase in ensiled fescue (Roberts et al., 2014). However, it should be noted that the high moisture treatment used by Roberts et al. (2014) had considerably more moisture (68% moisture) than the haylage treatment in the

current experiment, and it is possible that the increase in TEA levels was due to the greater moisture content as the authors did not observe the same effect when moisture content was approximately 40%. Additionally, it was expected that TEA concentrations would be lower in hay compared to haylage since hay production has been shown to reduce TEA levels, particularly when compared to ensiled tall fescue (Roberts et al., 2002; Roberts et al., 2009). In fact, TEA levels were numerically similar at d 0 to what was measured in the pasture prior to harvest. However, it should be noted that initial pasture TEA levels were determined by ELISA while TEA levels in the harvested forage were determined using HPLC. The ELISA method detects the presence of a lysergic acid functional group, and is therefore non-specific and accounts for a greater range of compounds compared to HPLC (Schnitzius et al., 2001). Furthermore, it has been suggested that the degradation of ergovaline does not alter the ergoline ring that is targeted by the ELISA (Roberts et al., 2014). Therefore, TEA concentrations would likely remain unchanged if measured using ELISA despite a decline in ergovaline concentrations. Additionally, it should be noted that overall TEA concentration means as analyzed by HPLC for hay and haylage were 1,681 µg/kg and 1,806 µg/kg, respectively, which is numerically lower than that recorded for the pasture prior to harvest. Since ergot alkaloids are sensitive to light (Garner et al., 1993), changes in cloud cover or light intensity will result in changes in the rate of TEA decline during curing. Therefore, it is expected that there is considerable variation across experiments investigating TEA levels during curing due to fluctuations in weather. Additionally, the heat generated during the fermentation process in large round bales would be much greater than what would have been generated in the current experiment. Nevertheless, the results of the current experiment did not suggest that producing hay provides a reduction in TEA concentrations compared to haylage.

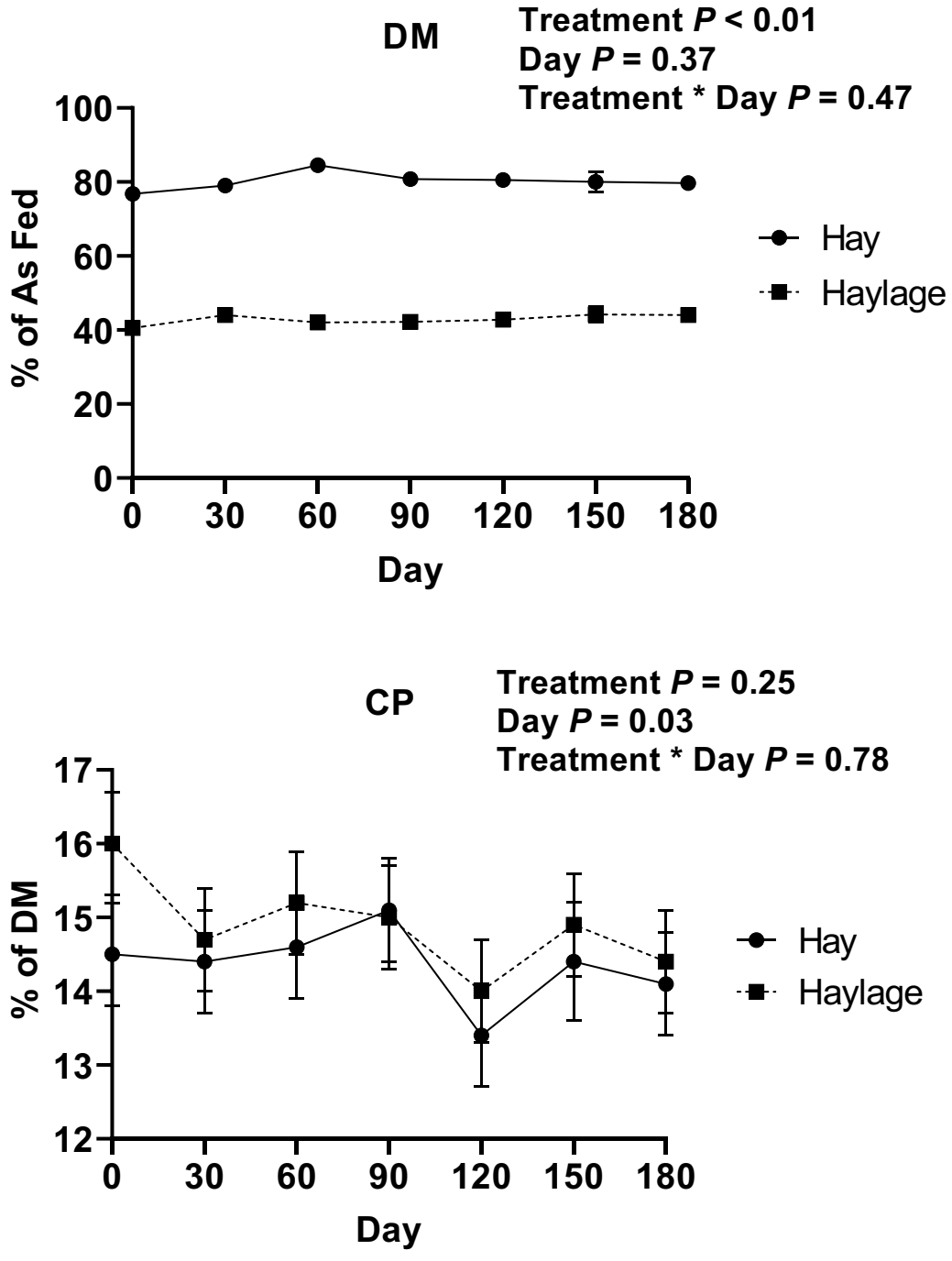
In conclusion, the results of this experiment show little differences between hay and haylage in mini-silos over a 180 d period. Minimal differences in nutritive value were noted across treatments, with the exception of reduced NDF concentrations in haylage relative to hay. No significant treatment effects or interactions were observed for ergot alkaloid concentrations. However, ergovaline concentrations declined in both treatments during short term storage. This reduction occurred after 60 d of storage and coincides with the drop in pH during the initial period of ensiling. In most production settings harvested forage would be fed after a much longer storage period, so this benefit can likely be obtained through minimal operational changes. While additional data are necessary to validate this in production settings where the forage would be in large round bales that are stored outside and exposed to sunlight and temperature fluctuations, these data suggest that producers harvesting E+ tall fescue can utilize haylage production when needed to optimize quality and yield without sacrificing ergot alkaloid degradation that has been observed in hay.

Figure 5.1: pH of toxic endophyte-infected tall fescue hay or haylage over increasing storage time.[1]



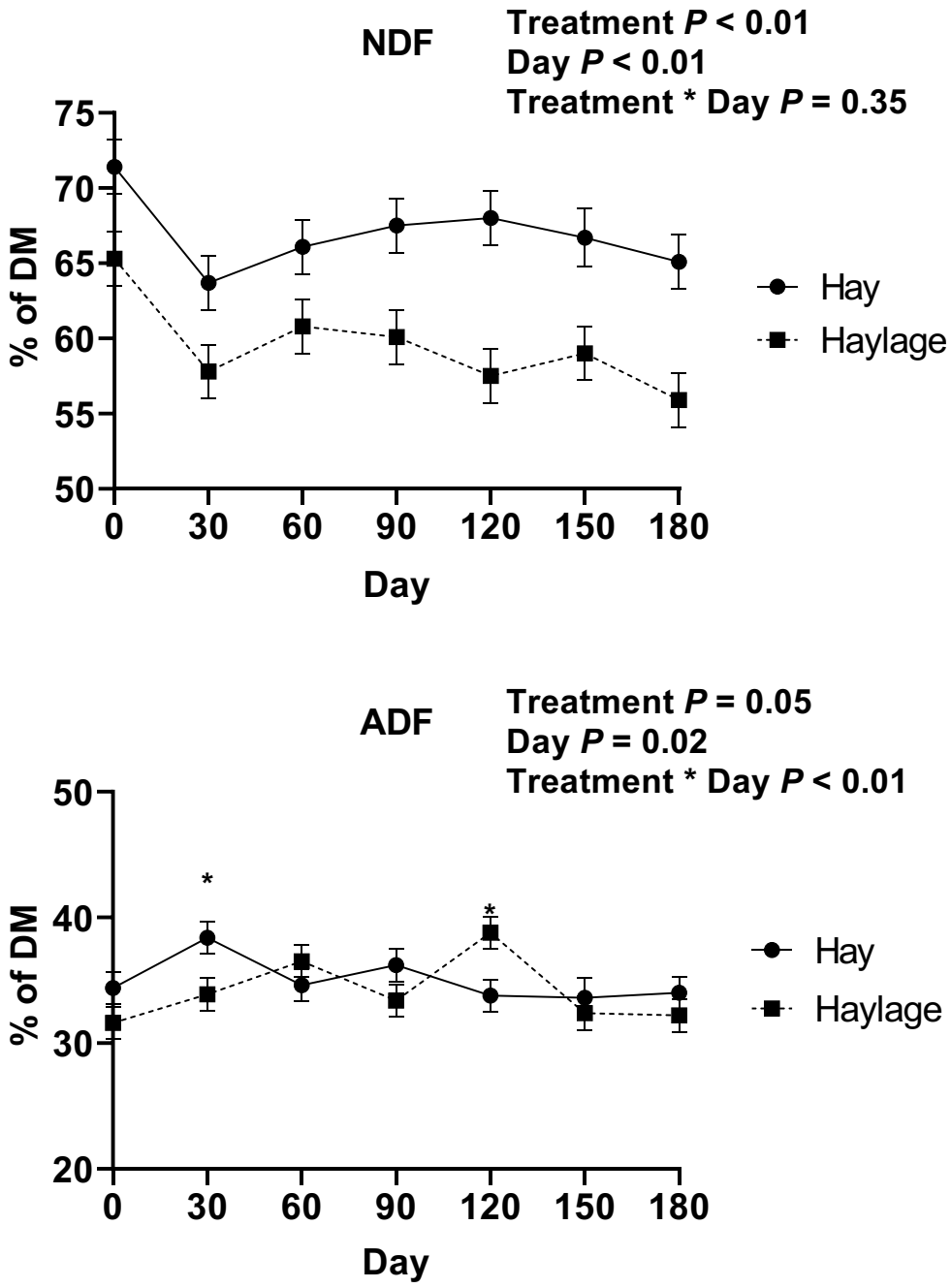
[1] Values are means of n = 6 experimental units per treatment on each day. The experimental unit is considered a mini silo of hay or haylage on the given day.
[*] Treatments differ within the given day ($P \leq 0.05$)

Figure 5.2: Dry matter (DM) and crude protein (CP) of toxic endophyte-infected tall fescue hay or haylage over increasing storage time.[1]



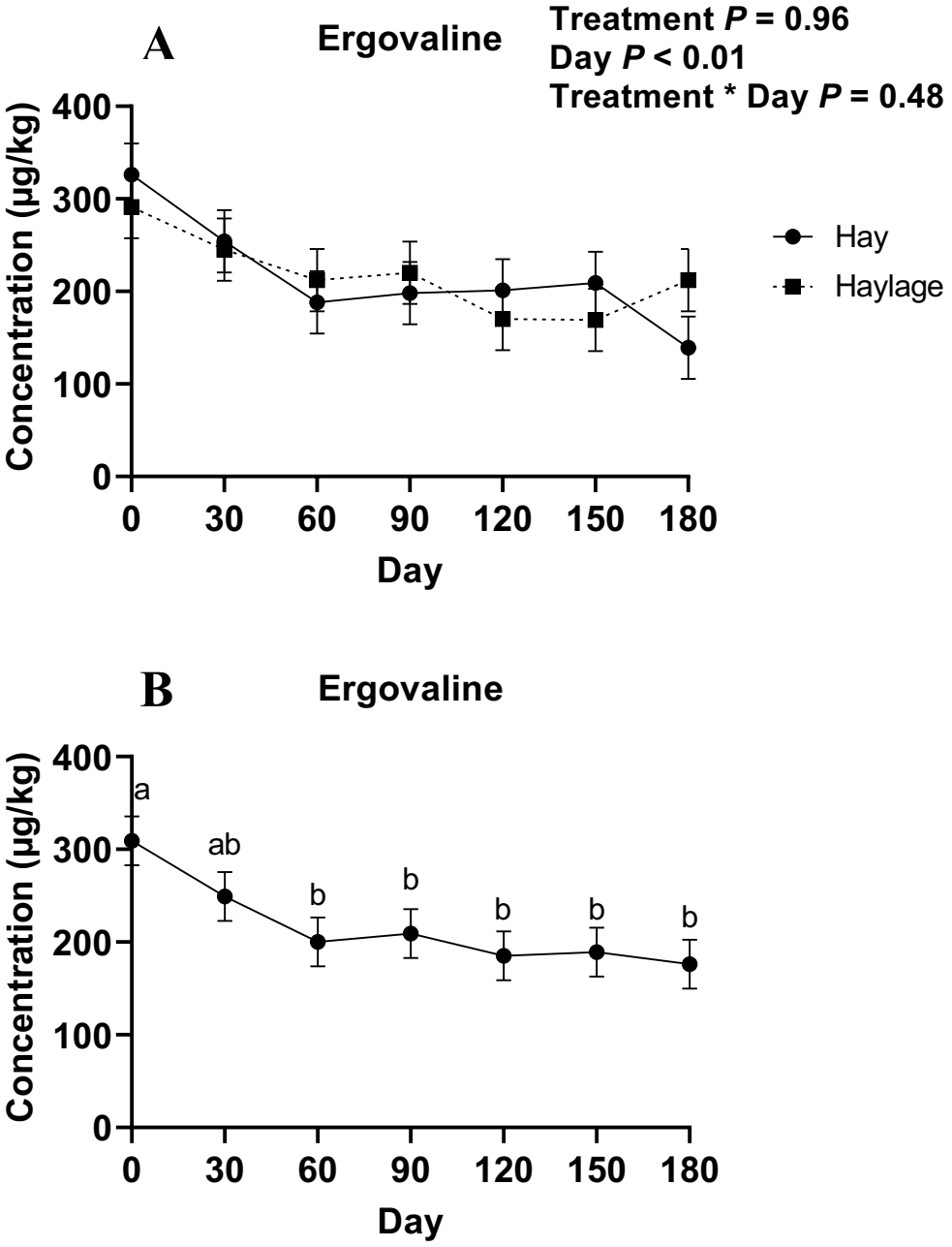
[1] Values are means of n = 6 experimental units per treatment on each day. The experimental unit is considered a mini silo of hay or haylage on the given day.

Figure 5.3: Neutral detergent fiber (NDF) and acid detergent fiber (ADF) of toxic endophyte-infected tall fescue hay or haylage over increasing storage time.



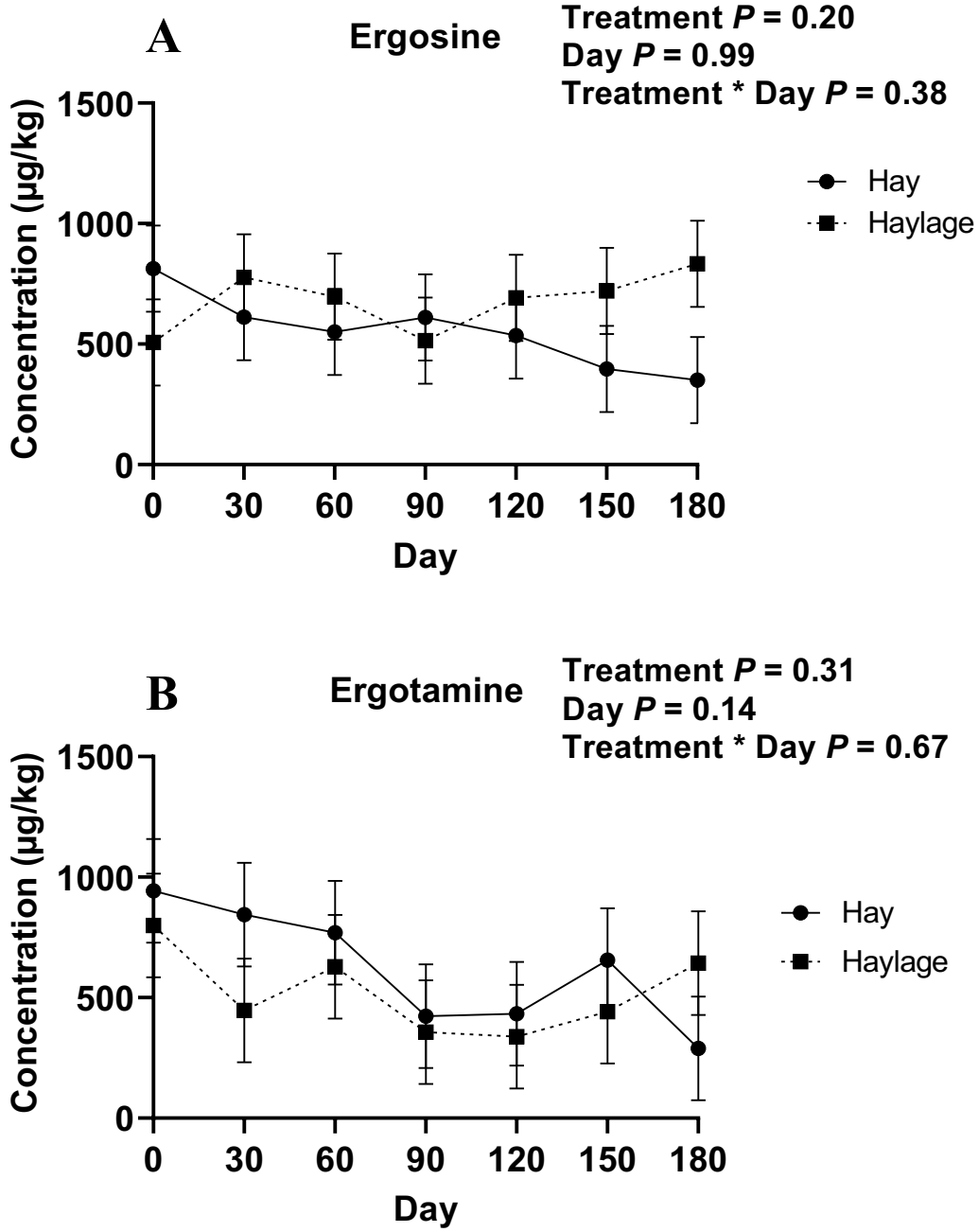
[1] Values are means of n = 6 experimental units per treatment on each day. The experimental unit is considered a mini silo of hay or haylage on the given day.
[*] Treatments differ within the given day ($P \leq 0.05$)

Figure 5.4: Concentrations of ergovaline in toxic endophyte-infected tall fescue hay or haylage over increasing storage time (**A**).[1] The main effect of day on the concentrations of ergovaline throughout the ensiling period are shown in panel **B**.[2]



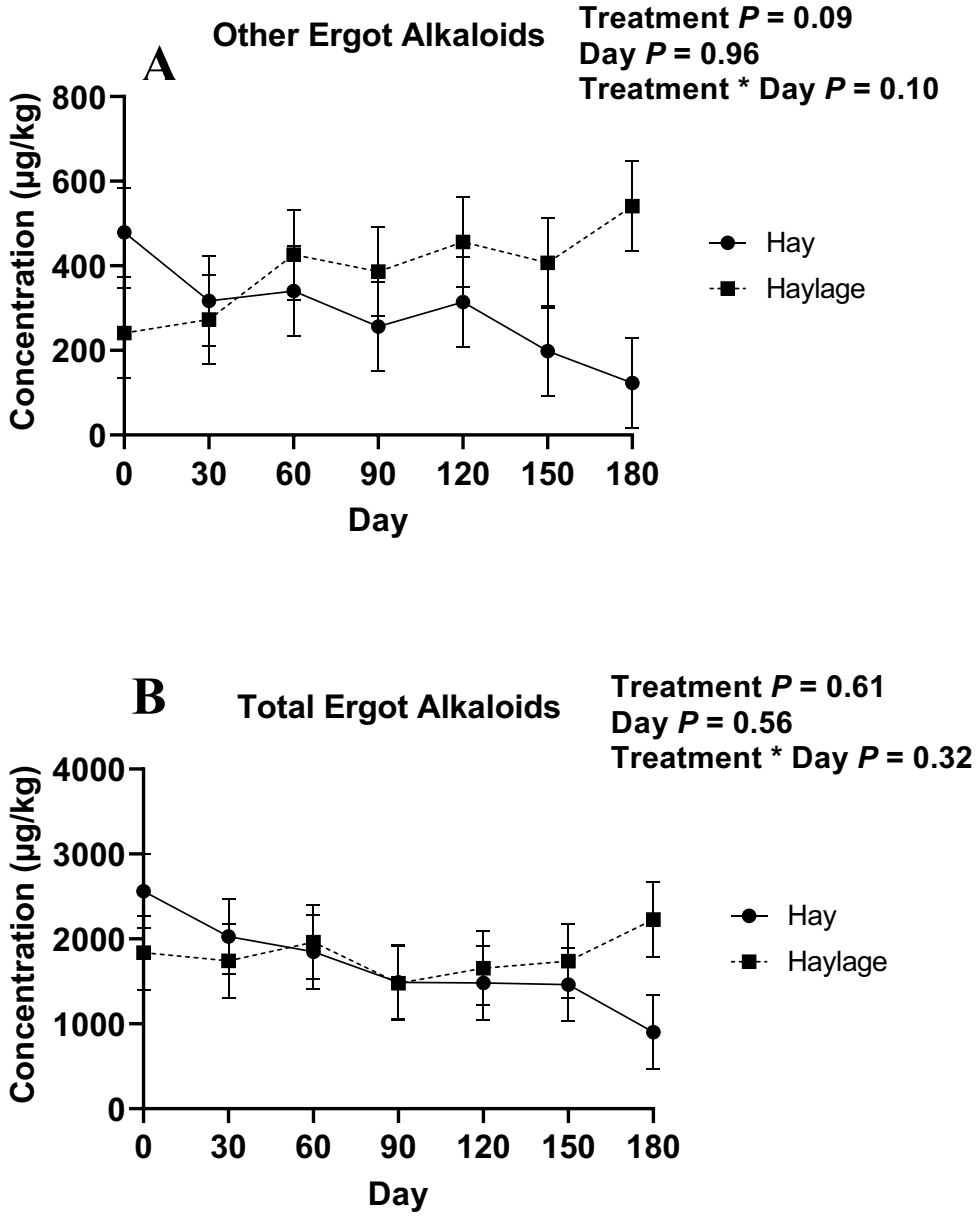
[a-b] Values not sharing a common superscript differ significantly ($P \leq 0.05$).
[1] Values are means of n = 6 experimental units per treatment on each day. The experimental unit is considered a mini silo of hay or haylage on the given day.
[2] Values are means of n = 12 experimental units (n = 6 hay; n = 6 haylage) on each day.

Figure 5.5: Concentrations of ergosine (**A**) and ergotamine (**B**) in toxic endophyte-infected tall fescue hay or haylage over increasing storage time.[1]



[1] Values are means of n = 6 experimental units per treatment on each day. The experimental unit is considered a mini silo of hay or haylage on the given day.

Figure 5.6: Other ergot alkaloids[1] (ergocornine, ergocryptine, and ergocrystine; **A**) and Total ergot alkaloids[2] (**B**) in toxic endophyte-infected tall fescue hay or haylage over increasing storage time.[3]



[1] Other ergot alkaloids are defined as the sum of ergocornine, ergocryptine, and ergocrystine concentrations analyzed using HPLC.
[2] Total ergot alkaloids are defined as the sum of the ergovaline, ergosine, ergotamine, ergocornine, ergocryptine, and ergocrystine concentrations analyzed using HPLC.
[3] Values are means of n = 6 experimental units per treatment on each day. The experimental unit is considered a mini silo of hay or haylage on the given day.

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CHAPTER VI
Epilogue

Nutritional strategies for growing cattle in the Southeastern U.S. have continued to evolve as new research becomes available and producer access to feedstuffs changes. In most cases producers will need to provide supplemental energy or a combination of supplemental energy and protein since many of the cool-season forage species in the region will generally first become limiting in energy. Since the increase in corn prices in the mid-2000s, there has been an increase in the inclusion of co-product and by-product feeds in cattle supplements. As a result, there has been increased research interest in incorporating these feedstuffs into pasture supplementation programs. By further understanding the interaction between supplemental feeds and forages in the Southeastern U.S., recommendations made to producers regarding their supplementation programs can be improved.

Experiment 1 was designed to investigate whether the improved growth response of cattle supplemented with CGF compared to corn reported by Hardin (2018) would be maintained with grazing animals. These data indicate that supplementation with CGF results in greater growth performance compared to corn when supplemented to grazing cattle. This indicates that producers can obtain considerably greater returns on their feed investment, as CGF can be purchased at a lower price than corn in many situations. It should be noted, however, that ADG was considerably lower than what was anticipated. In most production settings, the level of supplementation would have been increased as temperatures decreased, and the animal's maintenance energy requirements increased. A constant level of supplementation as a percent of BW was maintained throughout this experiment, and growth declined in the later months as a result. Additionally, supplements were not formulated to be isonitrogenous so animals receiving the CGF supplement had greater crude protein supply relative to those supplemented with corn. While the forage in this experiment was

relatively good quality, it is possible that the cattle supplemented with corn were limited by CP. However, the additional growth of cattle supplemented with corn relative to the un-supplemented control group suggests that this effect may be minimal. Regardless, the increased CP content of CGF relative to corn should be considered when designing a supplementation strategy. Additionally, there was considerably more variation in individual daily supplement intake than was anticipated. This prevented us from investigating the interaction between supplement source and level as originally designed, and instead resulted in daily supplement intake being included in the model as a covariate. Large individual variation in supplement intake has previously been reported with SmartFeed trailers (Reuter et al., 2017). It is likely that this variation is similar to what would be observed when animals are supplemented using a common feed bunk in pastures and is likely due to competition with other animals for bunk space and time as well as daily variation in forage intake. Therefore, it is unlikely that animals can be fed precisely enough to design experiments with small differences in programmed feed intake when group fed. Researchers that desire to control supplement intake in grazing situations should consider larger gaps in programmed feed intake if the animals are group fed or consider utilizing a more controlled individual feeding method.

While experiment 1 demonstrated that supplementing CGF results in a greater performance response relative to corn, it did not elucidate the reason this occurs. Therefore, experiment 2 was designed to determine if this effect was due to changes in nutrient digestibility. However, results of this experiment failed to show a negative effect on digestibility when corn was supplemented, and no differences were observed in growth performance. In fact, at the low level (0.25% BW) heifers supplemented with corn had greater ADF digestibility compared to those supplemented with CGF. This was surprising as previous work with DDGS had shown the opposite. The

inclusion levels were chosen based on a previous report showing the effects of corn grain supplementation on digestibility in a fiber-based ration (Russell et al., 2016). However, the large differences in fiber source (soybean hulls vs. alfalfa hay) may have resulted in the rumen profile being impacted differently at the same inclusion level of corn. Therefore, additional data are necessary to determine if this effect is observed at greater supplement inclusion levels. Furthermore, additional data regarding the impact of varying levels of corn and CGF supplementation on ruminal fiber digestibility will help to solidify the interpretation of total tract digestibility data. However, these results suggest that there is no beneficial effect on digestibility by supplementing CGF rather than corn at levels below 0.75% BW.

Previous research has shown that ergot alkaloids present in tall fescue decline during curing and throughout storage in hay but not in haylage (Roberts et al., 2009; Roberts et al., 2014). Therefore, experiment 3 was designed to measure the decline of ergot alkaloids in both fescue hay and haylage throughout a 180-d storage period. Because ergovaline has been recognized as the principal alkaloid responsible for fescue toxicosis, we were particularly interested in how ergovaline concentrations changed throughout storage. There were no differences in ergot alkaloid concentrations among treatments, nor was the treatment by day interaction significant. This was surprising given that concentrations of total ergot alkaloids and ergovaline have previously been shown to be lower in hay compared to haylage. However, since ergot alkaloid degradation in hay is influenced by weather conditions, particularly sunlight, the differences between the current experiment and previous results could be due to the cloud cover experienced during and after clipping. If this is the case, producers may not obtain benefit from making hay compared to haylage under cloudy weather conditions. However, despite the lack of differences among treatments there was a beneficial effect of storage time as ergovaline concentrations decreased over time. This

effect was realized after 60 d of storage and was maintained throughout the remainder of the experiment. Therefore, ergovaline concentrations in harvested forage can be reduced easily by producers through short term storage.

While the results of this experiment indicated a beneficial effect of increased storage time on decreased ergovaline concentrations, care must be taken when utilizing the results for producer recommendations. The experiment was conducted utilizing mini silos in a controlled environment to reduce variation from large differences in weather conditions throughout the year, which is very different from conditions in a production environment. Differences in exposure to sunlight, ambient temperatures, and temperature fluctuation during the fermentation process may induce changes in the rate and extent of ergot alkaloid degradation, particularly in the ensiled forage. Therefore, additional data are necessary under production conditions to verify the effects of ensiling time on ergot alkaloid concentrations. Ideally, these data would include similar repeated measures sampling to capture changes over time, but practical limitations may limit investigators to only a few samples to capture this.

Collectively, these experiments demonstrate methods to improve performance of growing beef cattle in pasture-based production systems. Producers wishing to supplement grazing cattle can do so with greater growth returns by supplementing CGF compared to corn. The reasons for this are likely due to a multitude of factors including differences in CP content and potential changes in ruminal digestion, as apparent total tract fiber digestibility was not improved in heifers supplemented with CGF compared to corn, regardless of inclusion level. While the data regarding changes in ergot alkaloids during the storage of hay and haylage made from E+ tall fescue are surprising and more complicated than what was expected, producers may realize improvements in ergovaline concentration by storing harvested E+ tall fescue for at least 30 d post-harvest. This can

likely be implemented without changes to current operations, as forage is typically harvested in the Spring and fed in the Winter when pasture forage mass becomes limiting. Continued research in both supplementation strategies and fescue ergot alkaloid management will allow Southeastern beef producers to improve growth and returns.

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