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Estancia vs. Kentucky 31: Examining Leaf Nutrient Content of Established Tall Fescue

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ESTANCIA VS. KENTUCKY 31: EXAMINING LEAF NUTRIENT CONTENT OF ESTABLISHED TALL FESCUE

A Master's Thesis

Presented to

The Graduate College of

Missouri State University

In Partial Fulfillment

Of the Requirements for the Degree

Master of Science, Agriculture

By

Macie Wayne Clark

May 2021

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ESTANCIA VS. KENTUCKY 31: EXAMINING LEAF NUTRIENT CONTENT OF

ESTABLISHED TALL FESCUE

Agriculture

Missouri State University, May 2021

Master of Science

Macie Wayne Clark

ABSTRACT

In 2020 Missouri ranked third in beef cattle production and second in hay production. As part of the fescue belt of the United States, Missouri's agricultural industries rely on tall fescue (*Lolium arundinaceum* Schreb.) as a main forage crop. Two major disorders in cattle grazing tall fescue are fescue toxicosis and grass tetany. Fescue toxicosis occurs when animals consume toxic ergot alkaloids produced by an endophyte within the tall fescue plants. The most commonly grown tall fescue variety, Kentucky 31, is infected with the toxic endophyte. Varieties containing a novel non-toxic endophyte, such as Estancia, have been developed to avoid fescue toxicosis. Grass tetany is a metabolic disorder caused by low blood magnesium (Mg) in ruminants. Grazing on tall fescue with lower Mg, in the spring and fall, can elevate these symptoms. Increased leaf Mg and calcium (Ca) concentrations, along with decreased levels of leaf potassium (K), reduce the grass tetany ratio $[K/(Ca+Mg)]$, thus reducing the risk for grass tetany. The purpose of this study is to compare leaf nutrient concentrations of Kentucky 31 and Estancia varieties to determine differences in the risk for grass tetany. Established plots of Estancia and Kentucky 31 at the University of Missouri Southwest Research Center in Mount Vernon, Missouri were sampled in the fall of 2020. Upon analysis of the leaf nutrient concentrations, Estancia revealed higher leaf Ca and Mg in comparison to Kentucky 31 across all three months of the study. The increase in Ca and Mg content in Estancia produced a lower grass tetany ratio compared to that of Kentucky 31. Further research is required to determine if these varietal differences are apparent in the spring and when grown under varying soil fertility levels.

KEYWORDS: beef cattle, tall fescue, fescue toxicosis, grass tetany, toxic endophyte, novel endophyte, grass tetany ratio, magnesium

ESTANCIA VS. KENTUCKY 31: EXAMINING LEAF NUTRIENT CONTENT OF

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A Master's Thesis Submitted to the Graduate College Of Missouri State University In Partial Fulfillment of the Requirements For the Degree of Master of Science, Agriculture

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In the interest of academic freedom and the principle of free speech, approval of this thesis indicates the format is acceptable and meets the academic criteria for the discipline as determined by the faculty that constitute the thesis committee. The content and views expressed in this thesis are those of the student-scholar and are not endorsed by Missouri State University, its Graduate College, or its employees.

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INTRODUCTION

In 2020, Missouri ranked third in beef production in the United States, primarily supported by grass-based pastures throughout the state. Tall Fescue (*Lolium arundinaceum* Schreb.) is a predominant forage in these grazing systems (Missouri Ag Highlights, 2021). The ability for tall fescue to grow in poor quality soils increases its value in many regions throughout Missouri, particularly in soils containing low pH and fertility. The southern portion of Missouri's soil contains karst topography and greater rock fragments which limit the sustainable agricultural use of the land. Tall fescue is key in allowing cattle to graze on these poor quality soils, thereby aiding in nutrient recycling while creating profitable enterprises for landowners.

Tall fescue, the most agriculturally influential species in the *Lolium* genus, became prevalent during the 1940s through the 1960s in the United States transition zone (Sleper & West, 1996). The transition zone is defined as the zone between cool-temperate and subtropical zones. This region includes Nebraska, Kansas, Oklahoma and extends east along at the same latitudinal lines (Sleper & West, 1996). Tall fescue has ability to adapt to a "wide range of soil conditions, including flooding, drought, and infertile or shallow topsoil, tolerance of continuous grazing, high yields of forage and seed, persistence, long grazing season, compatibility with varied management practices and low incidence of pest problems" (Sleper & West, 1996). This secured the prominent use of tall fescue in the United States transition zone. Reliance on tall fescue in the beef cattle industry is prevalent, however several possible health issues that can occur in cattle need to be evaluated and managed when utilizing tall fescue.

Fescue Toxicosis

Tall fescue is not naturally resistant to pests (Marten, 1989). However, it was discovered that tall fescue is commonly infected with the endophytic fungus *Acremonium coenophialum* (Sleper & West, 1996). This symbiotic relationship with endophytic fungi and tall fescue provides the fungi a place to reside while concurrently increasing pest and drought resistance. Unlike non-infected grasses, grasses infected with endophytic fungi have a range of biotic and abiotic adaptations. These include tolerances to drought, mineral imbalance, and soil acidity resulting in the infected grasses to compete and thrive where limited resources are available (Malinowski & Belesky, 2000). The life cycle of the endophytic fungi occurs in the aerial portion of the host grass, growing from root to shoot to seed completing the cycle (Malinowski & Belesky, 2000). Ergot alkaloids are chemicals produced by the endophytic fungi within the plant. Production of these chemicals coincide directly with the growth curve of tall fescue, elevated levels occurring in late spring and fall. When ergot alkaloids are produced in high amounts in the seed heads, black ergot may become visible on the seed heads. Some ergot alkaloids are toxic to animals when consumed, causing fescue toxicosis, a disorder affecting the nervous system of the animal.

Fescue toxicosis symptoms for grazing animals include reduced rate of gain and/ or milk production, a rough hair coat, fescue foot, summer syndrome, and fat necrosis (Bush et al., 1979). Fescue foot syndrome involves soreness/ lameness in the feet, necrosis in the extremities, and can result in an arched back. These ailments can occur any time other than in the summer (Bush et al., 1979). To prevent severity of fescue toxicosis, pasture composition is important for grazing animals. When grazing on toxic endophytic fescue it is important to either have mixed pasture or supplement other feedstuffs to lower toxicity levels. Not only can fescue toxicosis

occur while feeding on infected pasture, it can also occur when feeding toxic hay. Roberts et al. (2009) monitored concentrations of the ergot alkaloid ergovaline in tall fescue hay from clipping to storage for up to 18 months to determine if concentrations were affected by moisture level at time of bailing. It was observed that ergovaline levels in low and high moisture hay decreased after cutting and continued to decrease over the curing process causing the total ergot alkaloid concentrations to continue to decline over the 18 month period, decreasing the likelihood of fescue toxicosis in animals being fed stored hay (Roberts et al., 2009).

Other ways to prevent fescue toxicosis can be done by allowing animals to graze on infected pasture during the low peak ergot alkaloid production times of year or by having a novel endophyte variety of tall fescue. When introduced into tall fescue, the novel endophyte does not contain toxic ergot alkaloids nor cause fescue toxicosis in grazing animals. Estancia is a variety of tall fescue inoculated with ArkShield, a patented novel endophytic fungi (Mountain View Seed Company, Salem, Oregon). With this relationship, Estancia is said to be resistant to disease and pests while possessing no known negative effects on grazing animals (Mountain View Seed Company, Salem, Oregon). Although Estancia with ArkShield has been shown to improve the growth and performance of grazing cattle, no research has been conducted on reducing the potential for grass tetany, another common disorder on cattle grazing tall fescue.

Grass Tetany

Grass tetany, or hypomagnesemia, is a prevalent disorder in female grazing ruminants. In cattle it typically occurs within a few weeks after calving (Bush et al., 1979). Magnesium (Mg) is an important component of the spinal fluid that surrounds the brain and spinal cord and is necessary for transmitting signals throughout the animal's body. Blood Mg levels can vary, with

average levels being approximately $2 \text{ mg} / 100 \text{ ml}$ serum (or 0.20%) for a beef cow (Bush et al., 1979). Grass tetany is attributed to low Mg levels in the blood caused by low Mg intake in the rumen. Initial symptoms include decreased appetite, isolation, and a dull coat. If untreaded this can lead into prolonged exposure symptoms such as a staggered gait, stiff manner, excitable behavior, nervousness, muscular tremors, and death (Bush et al., 1979).

Grazing on lush spring and fall growth of tall fescue can increase susceptibility to grass tetany. Rapid leaf growth may cause lower Mg concentrations in the leaf tissue and specific organic acids produced by the plant may chelate Mg and remove it from the ruminant tissue (Sleper, 1979). Concentrations of other minerals in the animals diet, such as nitrogen (N), potassium (K), and calcium (Ca) may reduce Mg absorption by the animal as well (Sleper, 1979). In addition to Mg, other macronutrients such as Ca and K, in tall fescue have been found to influence grass tetany potential in grazing ruminants. When K levels increase and both Ca and Mg decrease in the leaf tissue, the imbalance results in increased potential for grass tetany. This has led to the utilization of the grass tetany ratio $(K/(Ca+Mg))$ by agronomists and landowners. The grass tetany ratio is calculated using concentrations (meq) of these macronutrients, a ratio of above a 2.2 (cmol_c kg⁻¹) indicates elevated susceptibility of grass tetany in the grazing ruminant (Marten, 1989). This increases the possibility for the grazing ruminant to develop the previously listed symptoms.

The grass tetany ratio of tall fescue has been found to be greater during periods of rapid growth. This typically occurs in the spring and fall. Potassium levels tend to either increase or slightly decrease in concentration in leaf tissue, while Mg levels tend to greatly decrease (McClain & Blevins, 2007). Because of these tendences, the grass tetany ratio can fluctuate greatly during these rapid growth periods.

To avoid grass tetany, Mg can be supplemented into the diet of tall fescue-based grazing systems. Ways to supplement Mg in the diet are to feed Mg mineral, utilize mixed pastures with legumes and other grasses, feed high Mg forages, and manage forages for greater leaf Mg such as with P fertilization. Although, adding mineral to the diet is a direct method of increasing Mg, mineral supplements are expensive and not palatable (Sleper, 1979). It has been determined to be more economical to incorporate mixed pasture systems and high Mg forages.

Mineral content in varieties of tall fescue is important in determining how much feed and supplement should be incorporated into the ruminant diet to prevent deficiencies. Hill & Guss (1976) reported mineral accumulation appeared to be under genetic control and breeding for certain mineral contents should be possible. Sleper et al. (1977) also focused on reducing grass tetany in cattle grazing tall fescue through breeding. The objectives for their work were to determine the genetic variation, heritability of the grass tetany ratio, and examined the interrelationships between the nutrients in tall fescue. Sleper et al. (1977) found reducing the grass tetany ratio in tall fescue samples were high in heritability and mass/ recurrent selection should be effective in improving the ratio. Sleper et al. (1977) also found that during periods of growth, leaf Mg levels increased at a slower rate than Ca levels and impacted the day to day variation of the grass tetany ratio more so than temperature changes affecting the leaf K levels. It was concluded that breeding is one way to change Ca, Mg and K, but there are other factors that need to be assessed to understand why mineral content changes from day to day (Sleper et al., 1977). Additional factors can alter mineral content in tall fescue leaves, such as fertilization, temperature, and availability of water.

Reinbott & Blevins (1994) observed how fertilization for N, P, K, and Mg as well as root temperature can effect Mg, Ca, and K levels in the leaves during the spring months when grass

tetany is prevalent. Reinbott & Blevins (1994) found that regardless of soil temperature leaf Mg and Ca concentrations were higher after P fertilization in field-grown tall fescue. Lower amounts of P were needed to raise the leaf Mg concentrations than to raise the leaf Ca concentrations. Reinbott & Blevins (1997) recognized that to see a change in leaf Ca and Mg after P application, adequate soil Mg may be required in field grown tall fescue. In both experiments, Reinbott & Blevins (1994, 1997) found that higher P treatments resulted in lower grass tetany ratios, and this was consistent with an earlier study (Reinbott & Blevins 1991). Reinbott & Blevins (1994) also observed that Mg fertilization of tall fescue was more effective when P fertilization was also applied. This concludes that when correcting grass tetany, both P and Mg fertilization need to be evaluated. Reinbott & Blevins (1997) concluded that producers should monitor soil P levels and maintain adequate P in pastures to maintain Mg, Ca and P quality and forage production. A study conducted by Lock et al. (2002) also concluded that by fertilizing tall fescue pastures with P, producers can improve the uptake of Mg in the forage to help prevent the susceptibility of grass tetany in beef cattle.

If adequate P is not maintained in tall fescue pastures, invasive species that are prevalent in low P soils can, over time, replace the tall fescue with a lower quality forage that provides little grazing benefit, except in early spring (Dustman & van Landingham, 1930). Blevins et al. (2018) found a combination of lime and P fertilization helped limit invasive species, like broomsedge, in tall fescue pastures. Lime applications can also improve soil and leaf nutrient contents to aid in lowering the grass tetany ratio. Hamilton et al. (2012) found calcitic lime applications to increase leaf Ca and decrease leaf K of Kentucky 31 grown in southwest Missouri. Dolomitic lime additionally increased leaf Mg, resulting in both lime applications lowering the grass tetany ratio (Hamilton et al., 2012).

Stockpiling is used by beef producers in the mid-west to extend the grazing period from fall into winter. This reduces the costs of winter feeding. The ability to stockpile tall fescue in the winter months is due to its hardy nature and capability to maintain growth and quality in cooler temperatures (McClain & Blevins, 2009). Although stockpiling is economic, tall fescue in the late winter months may have lower leaf nutrient contents. McClain & Blevins (2009) found stockpiled tall fescue had a grass tetany ratio above 2.2 from October through January in southwest Missouri. Phosphorus fertilization in early fall did not have an effect on leaf Ca concentrations. Calcium is immobile and remains constant during the late fall and winter. However, it was determined that P fertilization increases leaf Mg in stockpiled tall fescue (McClain & Blevins, 2007). Although poultry litter and P fertilization can be helpful in increasing leaf nutrient content of stockpiled tall fescue, knowing the soil nutrient content is important before applying fertilizer.

Current Study

This study evaluated two different cultivars of tall fescue, Kentucky 31 and Estancia. Kentucky 31 is the major toxic endophyte cultivar grown in the Midwest due to survivability in harsh environmental conditions. Estancia is a novel endophyte cultivar of tall fescue and can also survive in such conditions. It has not been determined if leaf Mg content or the grass tetany ratio is different among these two cultivars. This study was conducted to determine if there is a difference in leaf nutrient content between established fields of Kentucky 31 and Estancia tall fescue.

MATERIALS AND METHODS

The sampling for this study was conducted at the University of Missouri Southwest Research Center in Mount Vernon, Missouri (Figure 1) on 30 ft by 40 ft plots of Kentucky 31 and Estancia tall fescue established in September 2012 (Kenyon, 2017). The plots of eight tall fescue varieties were established in a four blocks with four replications of each variety randomized within blocks (Figure 2). This project involved the sampling of the four Kentucky 31 plots and the four Estancia plots (Figure 2). These plots have been grazed by cattle in a rotational grazing system since establishment, and periodically fertilized with poultry litter applications. The most recent poultry litter application was in August 2020 at the rate of two tons per acre.

Figure 1. Aerial view of the tall fescue plots located on University of Missouri Southwest Research Center in Mount Vernon, Missouri.

Figure 2. Plot map of tall fescue cultivars with Kentucky 31 and Estancia plots outlined in red (Kenyon, 2017).

Soil Sampling

The majority of the plots are planted on Hoberg silt loam, 2 to 5 percent slopes (Fineloamy, siliceous, active, mesic Oxyaquic Fragiudalfs) and Gerald silt loam, 0 to 2 percent slopes (Fine, mixed, active, mesic Aeric Fragiaqualfs) (Figure 3). On September 4, 2020 one soil sample from each Kentucky 31 and Estancia plot were collected. Each sample consisted of approximately 12 cores randomly selected within each plot to a depth of one to two inches. Sampling depth was limited on this date due to the dryness of the soil surface. On November 13, 2020 three soil samples comprised of approximately 12 cores each, were taken to a 6 in. depth from each established Kentucky 31 and Estancia plot. Soil samples were air-dried, ground to pass a 2mm screen, and analyzed in the Missouri State University Darr College of Agriculture research lab for pHs, Bray I P, Ca, Mg and K. Calcium, Mg and K were extracted using the ammonium acetate extraction method (Nathan et al., 2012) and determined using Atomic Absorption/ Flame Emission Spectrophotometry (Agilent Technology, 200 Series AA, Santa

Clara, California, USA). Soil P was extracted using the Bray I extraction method (Nathan et al., 2012) and measured using a GENESYS™ 30 Visible Spectrophotometer (ThermoFisher Scientific, Waltham, Massachusetts, USA).

Figure 3. Map of soil types at study site (70006- Creldon silt loam, 1-3% slopes; 70012- Hoburg silt loam, 2-5% slopes; 70045- Keeno gravely silt loam, 3-8% slopes; 73031- Gerald silt loam 0- 2% slopes) (Web Soil Survey, USDA NRCS).

Leaf Tissue Sampling

Three fescue leaf tissue samples, composed of approximately 20 most recently collared leaves each, were randomly collected from each plot on September 4, October 2, and November 13, 2020 (Figure 4). Samples were placed in a forced air oven (Cascade TEK, Cornelius, Oregon, USA) and dried at 55 °C. Dried fescue samples were finely ground using a modified coffee grinder and stored in plastic bags at room temperature. Leaf tissue samples were weighed between 0.245-0.255 g and digested in 5 mL of trace nitric acid using a MARS 6 Microwave Digestion System (CEM Corp., Matthews, North Carolina, USA). The CEM Plant Materials digestion protocol was used to completely digest the tissue (CEM Corp., Matthews, North Carolina, USA). The samples were filtered through Cytiva Whatman qualitative filter paper, grade CFP1, and brought up to a final volume of 25 mL with Nanopure DI water and stored in capped 50 mL polypropylene tubes.

Figure 4. Tall fescue tiller, the circle identifying the collar on the leaf that was chosen for sampling.

Phosphorus of the leaf tissue (% by weight) was determined colorimetrically by diluting the digests 1:20 with Nanopure DI water .One mL of the dilution was combined with 4 mL of working solution, vortexed, and then allowed to process for 20 min. A standard curve was created with premade plant digest background standards of 0, 0.5, 1, 2.5, 5 ppm P set at a wavelength of 660 nm mixed with the working solution. Samples were measured using a GENESYS™ 30 Visible Spectrophotometer (ThermoFischer Scientific, Waltham, Massachusetts, USA).

Leaf contents (% by weight) of Ca, Mg, and K were determined using Atomic Absorption/ Flame Emission Spectrophotometry (Agilent Technology, 200 Series AA, Santa Clara, California, USA). The grass tetany ratio was then determined in milliequivalents per kg of dry matter of the nutrients in plant tissue using the formula: $([K]/39) \div [([Ca]/20) + ([Mg]/12.1)]$ (Kemp and t'Hart, 1957).

Statistical Analysis

This study utilized a mixed block design. The model included variety and harvest dates as fixed factors, and block as a random factor. This model tested for significance of macronutrient levels between the varieties across harvest dates as well as interactions between variety and harvest date using Mixed Effects Model in MiniTab (Penn State University, Pennsylvania, USA, 2021). Harvest and variety were considered significant when means differed at $P \le 0.05$, means separated by Fisher's LSD pairwise comparison.

RESULTS

Leaf Nutrient Content

Leaf nutrient content varied between varieties of tall fescue and between harvest dates. Leaf Ca significantly differed between Kentucky 31 and Estancia, as well as between the harvests (Table 1). In all three harvests, leaf Ca was greater in Estancia leaves than Kentucky 31 leaves (Figure 5). Across harvests, leaf Ca declined from October to November in both varieties (Figure 5). Magnesium content was significantly different between varieties and harvests (Table 2). Leaf Mg declined each month from September through November (Figure 6). As with Ca, leaf Mg content was significantly greater in Estancia than in Kentucky 31 in all three harvests (Figure 6). Leaf K content showed no significant difference between varieties (Table 3, Figure7). Between harvests however, leaf K was different in each harvest, being greatest in September and lowest in October (Table 3, Figure 7). Similar to K, leaf P content was different in each harvest, with the greatest concentration occurring in September, and lowest of the three harvests in October (Table 4, Figure 8). Between varieties, Estancia was significantly greater than Kentucky 31 in P in the October harvest only(Figure 8).

Term	DF Num DF Den F-Value P-Value			
Var	1.00	9.00	14.55	$0.004*$
Har	2.00	9.00	24.53	$0.000*$
Var*Har	2.00	9.00	0.00	0.995

Table 1. Leaf Ca ANOVA where variety (Var) and harvest (Har) are fixed effects. P-values with an $*$ indicate significance (P<0.05).

Figure 5. Percent leaf Ca of Estancia and Kentucky 31 across three harvests in fall of 2020. Values are means \pm SE, n= 12. Significantly different (p<0.05, Fisher's LSD pairwise comparison) values within a harvest are indicated with an *, and values not followed by the same capital letters are significantly different across harvest dates.

Figure 6. Percent leaf Mg of Estancia and Kentucky 31 across three harvests in fall of 2020. Values are means \pm SE, n= 12. Significantly different (p<0.05, Fisher's LSD pairwise comparison) values within a harvest are indicated with an *, and values not followed by the same capital letters are significantly different across harvest dates.

Table 3. Leaf K ANOVA where variety (Var) and harvest (Har) are fixed effects. P-values with an $*$ indicate significance (P<0.05).

Term	DF Num DF Den F-Value P-Value			
Var	1.00	3.00	1.27	0.341
Har	2.00	6.00	54.50	$0.000*$
Var*Har	2.00	6.00	0.70°	0.535

Figure 7. Percent leaf K of Estancia and Kentucky 31 across three harvests in fall of 2020. Values are means \pm SE, n= 12. Significantly different (p<0.05, Fisher's LSD pairwise comparison) values within a harvest are indicated with an *, and values not followed by the same capital letters are significantly different across harvest dates.

Table 4. Leaf P ANOVA where variety (Var) and harvest (Har) are fixed effects. P-values with an $*$ indicate significance (P<0.05).

Term	DF Num DF Den F-Value P-Value			
Var	1.00	57.00	4.44	$0.040*$
Har	2.00	6.00	97.19	$0.000*$
Var*Har	2.00	57.00	0.72	0.490

Figure 8. Percent leaf P of Estancia and Kentucky 31 across the three harvests in fall of 2020. Values are means \pm SE, n= 12. Significantly different (p<0.05, Fisher's LSD pairwise comparison) values within a harvest are indicated with an *, and values not followed by the same capital letters are significantly different across harvest dates.

Grass Tetany Ratio

From September through November, leaf nutrient content of Ca, Mg, K, and P in both Estancia and Kentucky 31 declined (Figures 5, 6, 7, and 8). Estancia exhibited at least 0.03% greater leaf Ca levels and 0.05% greater leaf Mg levels than Kentucky 31 across all harvests (Figures 5 and 6). With no difference between the varieties in leaf K (Figure 7), the differences in leaf nutrient concentrations resulted in differences in the grass tetany ratios (Table 5). Estancia exhibited 0.13 to 0.3 lower grass tetany ratios compared to those in Kentucky 31, with the greatest difference occurring in November (Table 5, Figure 9).

Term			DF Num DF Den F-Value P-Value	
Var	1.00	3.00	11.64	$0.042*$
Har	2.00	6.00	41.18	$0.000*$
Var*Har	2.00	6.00	1.53	0.291

Table 5. Grass tetany ratio ANOVA where variety (Var) and harvest (Har) are fixed effects. Pvalues with an $*$ indicate significance (P<0.05).

Figure 9. Grass tetany ratios of Estancia and Kentucky 31 based off leaf nutrient content across the three harvests in fall of 2020. Values are means \pm SE, n= 12. Significantly different (p<0.05, Fisher's LSD pairwise comparison) values within a harvest are indicated with an *, and values not followed by the same capital letters are significantly different across harvest dates.

Soil Analysis

In September, soil samples were taken at one to two inch depths. This represented the nutrient availability of the topsoil and showed low variability across plots (Table 6). November soil samples were taken to a depth of six inches resulting in normal pHs and available nutrient levels for a grazed pasture in southwest Missouri. Plots 2 and 4 showed significantly higher available P compared to the other plots. while P was greater in plot 4 and lower in plot 5 (Table 7).

	Plot Variety pHs			Ca Mg K P $(\#/A)$ $(\#/A)$ $(\#/A)$ $(\#/A)$		
	KY31 5.72 2689			699	370	19
3	KY31 5.95 2794 904				397	25
5	KY31 5.73 2465				665 289	22
7	KY31 5.59 2401				630 225	16
2	EST	5.68	2669	794	331	15
4	EST 5.89 2628			824 481		33
6			EST 5.64 2563		655 243	15
8			EST 5.51 2700	722 357		20

Table 6. Soil test results from the September 2020 harvest. Values are means, n= 3.

	Plot Variety pHs		Ca Mg K P $(\#/A)$ $(\#/A)$ $(\#/A)$ $(\#/A)$	
$\mathbf{1}$	KY31 5.42 2476 612 279			25
3 ⁷	KY31 5.68 2467 625 278			36
5 ⁵	KY31 5.50 2190 523 174			$12*$
$7\degree$	KY31 5.48 2301 541 187			25
2			EST 5.48 2745 718 339* 23	
$4\overline{ }$			EST 5.71 2023 578 346* 45*	
6			EST 5.56 2380 509 191	23
8	EST 5.12 2245		527 224	37

Table 7. Soil test results from the November 2020 harvest. Values are means, n= 3. Significantly different (p<0.05, Fisher's LSD pairwise comparison) values within a column are indicated with an *.

DISCUSSION

The decline of leaf nutrients through the fall followed the same trend as previous Kentucky 31 tall fescue studies conducted at the MU Southwest Research Center in Mount Vernon, Missouri (McClain & Blevins, 2009). With the loss of nutrients from fall going into winter, Estancia shows higher levels of Ca and Mg and similar levels of K than Kentucky 31. Resulting in an improved grass tetany ratio in Estancia compared to Kentucky 31. The grass tetany ratios were below the 2.2 threshold for susceptibility for grass tetany. However, these data indicate that cattle grazing on Estancia in the fall may have less potential risk for developing grass tetany than when grazing Kentucky 31.

Further testing in April and May of 2021 is needed to determine if the differences in leaf nutrient content between Kentucky 31 and Estancia continue into the spring. Rapid growth in spring increases the grass tetany ratio and risk for the disorder in grazing animals, any improvements in the ratio could help reduce grass tetany occurrences.

In this study, the September soil sampling depth was limited by dry soil conditions. Therefore, the results of these samples were indicative of the topsoil fertility, not the entire six inch depth. Only the November soil sample data should be used as representation of soil fertility levels at the study site. As leaf nutrient content of tall fescue is depended on soil fertility levels (Blevins et. al., 2018., Hamilton et al., 2012., Lock et. al., 2002., McClain & Blevins, 2007., Reinbott & Blevins, 1991, 1994, 1997), it is important to monitor differences between varieties across multiple soil fertility levels. Future research should include established tall fescue plots of both varieties grown in various soil fertility conditions.

CONCLUSION

This study found Estancia tall fescue leaves to have greater leaf Ca and Mg levels and improved grass tetany ratio during the fall than Kentucky 31 tall fescue leaves. Continued sampling of these plots in the spring and summer, as well as over more than one year, will provide more information to determine if Estancia will reduce the susceptibility of grass tetany on grazing animals compared to those grazing Kentucky 31. Additionally, the data provided by this study will support further efforts to identify how soil fertility, especially soil P fertilization, and other environmental conditions affect the differences of leaf nutrients between Estancia and Kentucky 31.

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