

# Botanical composition of yearling steer diets in the Kansas Flint Hills

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Transl. Anim. Sci. 2018.2:S134–S138  
doi: 10.1093/tas/txy040

## INTRODUCTION

The Kansas Flint Hills have traditionally been grazed by yearling beef cattle in the summer months, when forage quality is at its peak. Early season, intensive grazing of yearling steers, or heifers on native pastures—known locally as intensive early stocking—allows producers to maximize ADG and beef production per ha with little input cost (Owensby et al., 2008). Cattle are classified as roughage eaters; a majority of their diet is comprised of graminoids with relatively few forbs or browse species (Hofmann and Stewart, 1972). Concomitantly, sericea lespedeza (SL; *Lespedeza cuneata*)—a state-recognized noxious weed—infests the Flint Hills region and competes with native plant species (Eddy and Moore, 1998).

Microhistological analysis of feces has been used to estimate the botanical composition of domestic and nondomestic animal diets for many years. It allows for microscopically aided differentiation of plant species based on epidermal and cuticular structures and provides species-specific descriptions of herbivore diets (Holechek et al., 1982). In addition, it can be used to study diets of animals in a grazing setting without disturbing natural feeding habits. In our study, 17 predominant graminoids, forbs, and forb-like plants in the

Kansas Flint Hills were examined in the diets of yearling beef steers. Our objective was to determine the botanical composition of yearling beef-steer diets grazing SL-infested native tallgrass prairie.

## MATERIALS AND METHODS

The Kansas State University Institutional Animal Care and Use Committee reviewed and approved all animal handling and animal care practices used in our experiment. All animal procedures were conducted in accordance with the Guide for the Care and Use of Animals in Agricultural Research and Teaching (FASS, 2010).

Our study was conducted at the Kansas State University Bressner Range Research Unit located in Woodson Co., KS during the growing seasons of 2015 and 2016. Eight native tallgrass pastures (31 ± 3.3 ha) infested with SL (initial basal frequency = 2.9 ± 2.74%) were burned annually in April.

Pastures were grazed by beef steers ( $n = 279$  per yr; initial BW = 264 ± 34 kg) at a relatively high stocking density (1.1 ha/steer) from 15 April to 15 July annually. Yearling beef steers were obtained from various commercial cattle growers in southeastern KS. Steers were weighed individually before grazing began each April and were assigned randomly to pastures. Steers were weighed individually again in late July.

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Received March 16, 2018.

Accepted April 14, 2018.

Four permanent 100-m transects were laid out on a north–south gradient in each pasture; ends were marked using steel posts. Steers were allowed a 14-d adaptation period before sampling began each yr. Beginning 1 May, five fresh fecal pats ( $\geq 30$  g wet weight) were collected along each transect ( $n = 20$  samples/pasture) at 2-wk intervals until 15 July (i.e., five sampling periods annually: early May, late May, early June, late June, and early July). Care was taken to avoid contamination of fecal samples with soil or vegetation. Wet fecal samples were placed in a plastic container upon retrieval and frozen ( $-20$  °C) pending processing and analysis.

Individual fecal samples were dried in a forced-air oven (55 °C; 96 h). Dried samples were ground (#4 Wiley Mill, Thomas Scientific, Swedesboro, NJ, USA) to a 1-mm particle size and composited by weight across transect and within pasture ( $n = 32$  composites per collection period). A Daisy II in vitro incubator (Ankom Technologies, Macedon, NY) was used to mix composite samples for 120 min without heat.

Plant composition and soil cover were assessed along two permanent transects in each pasture on 15 October  $\pm$  10.4 d in 2014 (i.e., pretreatment), 2015, and 2016 (i.e., posttreatment) using a modified step-point technique (Owensby, 1973; Farney et al., 2017). Transect points ( $n = 100$  per transect) were evaluated for bare soil, litter, or basal plant cover (% of total area). Plants were identified by species; basal cover of individual species was expressed as a percentage of total basal plant area.

Approximately 65% of total basal vegetation cover on pastures used in our experiment was composed of the following forage species: big bluestem (*Andropogon gerardii*), little bluestem (*Schizachyrium scoparium*), switchgrass (*Panicum virgatum*), Indian grass (*Sorghastrum nutans*), blue grama (*Bouteloua gracilis*), side-oats grama (*Bouteloua curtipendula*), buffalo grass (*Bouteloua dactyloides*), sedges (*Carex* spp.), purple prairie-clover (*Dalea purpurea*), lead-plant (*Amorpha canescens*), dotted gayfeather (*Liatris punctata*), heath aster (*Symphotrichum ericoides*), SL (*Lespedeza cuneata*), Baldwin's ironweed (*Vernonia baldwinii*), Western ragweed (*Ambrosia psilostachya*), annual broomweed (*Amphiachyris dracunculoides*), and common ragweed (*Ambrosia artemisiifolia*). Reference standards for each above-named plant species were prepared using methods described by Holechek et al. (1982). Each standard sample was derived by hand-clipping 10 to 20 individual plants from a homogeneous stand of each plant type. Fruiting culms were discarded, whereas leaves, flowers, and vegetative stems were dried in a forced-air oven

(55 °C; 96 h). Dried samples were ground to a 1-mm particle size using a cyclone-style sample mill (model no. 80335R, Hamilton Beach, Glen Allen, VA).

Composite fecal samples and reference standards were prepared for microhistological analyses using methods described by Holechek et al. (1982), as adapted Preedy et al. (2013). A small amount (approximately 0.5 to 1 g) of sample material was placed into a beaker. Samples were soaked individually overnight in 50% EtOH (v/v). Ethanol was decanted after soaking and samples were homogenized and washed with deionized H<sub>2</sub>O over a No. 200 US-standard sieve. Samples were then soaked in a 0.05-M NaOH solution for 20 min and again washed with deionized H<sub>2</sub>O for an additional 5 min through a No. 200 US-standard sieve.

Composite fecal samples and reference standards were placed on slides (five slides per composite fecal sample and three slides per reference standard) using a dissecting needle. Two to three drops of Hertwig's solution were applied and slides were held 2 to 5 s over a propane flame to dry. Slides were not permanently mounted, as the addition of glass covers slips and Hoyer's solution decreased visibility of plant fragments.

Sample slides and reference-standard slides were viewed with a compound microscope (DC5-163, Thermo Fisher Scientific, Asheville, NC) at 100 $\times$  magnification. The microscope was equipped with a digital camera; 20 randomly selected slide fields from each fecal-composite sample slide and each reference-standard slide were photographed digitally and stored (Preedy et al., 2013).

Observers of microscopically photographed images were trained using methods similar to those described by Holechek and Gross (1982). Observers viewed photographs of reference-standard slides until establishing familiarity with the epidermal and cuticular characteristics of each plant species. Observers were able to view reference-standard slide photos simultaneously with fecal-sample slide photographs for reference. Individual plant fragments on each sample-slide field of view were counted and identified by plant species. The total number of fragments of each plant species on a given slide were converted to frequency of occurrence (i.e., [total of individual species  $\div$  total of all species] \* 100; Holechek and Vavra, 1981). Plant fragment prevalence in fecal-sample slide fields was assumed to be equivalent to prevalence in fecal samples and equivalent, on a percentage basis, to botanical composition of the diets grazed by beef steers (Sparks and Malechek, 1968). Plant fragments that were not among the 17 range-plant species for which

reference standards were prepared were classified as either unidentified graminoids or unidentified forbs.

Steer growth performance data were analyzed as a completely randomized design using a mixed model (SAS Inst. Inc., Cary, NC). Class variables included animal, pasture, and year. The model contained a term for pasture only and animal within pasture and year was used as a random term. Least squares means were considered different when protected by a significant *F*-test ( $P \leq 0.05$ ).

Mean basal cover percentages, standard deviations, minimum basal covers, and maximum basal covers for bare soil, litter, total basal vegetation, graminoids, forbs, shrubs, and individual plant species were calculated using the PROC MEANS procedure (SAS Inst. Inc., Cary, NC). Values were summarized across pastures and yr of the experiment.

The percentages of bare soil, litter cover, total basal vegetation cover, grass basal cover, forb basal cover, shrub basal cover, and basal cover of individual plant species were analyzed as a completely randomized design using a mixed model (SAS Inst. Inc., Cary, NC). Class variables were pasture, transect, and year. The model contained a term for pasture only and transect within pasture was used as a random term. Least squares means were considered different when protected by a significant *F*-test ( $P \leq 0.05$ ).

Diet composition data were analyzed as a completely randomized design using the PROC MIXED procedure of SAS (SAS Inst. Inc., Cary, NC). Class variables included pasture, transect, period, and yr. The model contained terms for pasture, period, and the 2-way interaction. Transect within pasture and year and period within pasture and year were considered random effects. Pasture effects on selection patterns for each of the 17 plant-species standards, unknown grass plants, and unknown forb plants were not detected ( $P \geq 0.09$ ); moreover, period  $\times$  pasture effects on diet selection patterns were also not detected ( $P \geq 0.11$ ). Therefore, period sums of squares were partitioned using preplanned orthogonal polynomial contrasts ( $n = 4$ ). Period effects for the highest-order, significant contrast ( $P \leq 0.05$ ) are discussed.

Kulczynski's Similarity Index (**KSI**;  $((2c_i) / (a_i + b_i)) * 100$ , where  $a_i$  is the % basal cover of component  $i$ , and  $b_i$  is the % of component  $i$  selected by an herbivore, and  $c_i$  is the lesser of  $a_i$  and  $b_i$ ) was used to evaluate yearling-steer diet selection patterns in relation to botanical composition of pastures. For the purposes of our analysis, we assumed that KSI values  $\geq 80\%$  indicated little or no discrimination (i.e., selection patterns were very similar to plant availability), that KSI values between 21% and 79% indicated

moderate discrimination, and that KSI values  $\leq 20\%$  indicated either strong preference for or avoidance of individual plant species. When KSI values were  $\leq 20\%$ , preference and avoidance were distinguished from one another by comparing the proportion of the specific plant in yearling-steer diets with basal cover of the specific plant on pastures.

## RESULTS AND DISCUSSION

Initial BW, final BW, and ADG of steers were not influenced ( $P \geq 0.22$ ) by pasture (data not shown). In general, steer growth performance was excellent during the term of our experiment (mean ADG =  $1.4 \pm 0.32$  kg); therefore, we concluded that any potential differences in forage composition between native tallgrass pastures used in our experiment were insufficient to influence steer BW gain.

Proportions of total graminoids, big bluestem, little bluestem, switchgrass, Indian grass, blue grama, buffalo grass, sedges, total forb and forb-like plants, purple prairie-clover, leadplant, dotted gayfeather, heath aster, Baldwin's ironweed, annual broomweed, and common ragweed were not different ( $P \geq 0.06$ ) between pastures (data not shown). Conversely, there were differences ( $P < 0.01$ ) in proportions of unidentified graminoids, SL, and western ragweed between pastures, whereas dotted gayfeather was not detectable in our analysis of pasture forage composition.

No pasture or period  $\times$  pasture effects were detected ( $P \geq 0.09$ ) in selection of total graminoids, total forb or forb-like plants, or any of the 17 reference standards selected for microhistological analyses; however, period effects ( $P \leq 0.03$ ) in selection were detected for little bluestem, switchgrass, Indian grass, buffalo grass, sedges, unidentified graminoids, leadplant, and unidentified forbs (data not shown). Therefore, period effects on selection of individual plant species or groups of plant species were characterized using orthogonal polynomial contrasts (Table 1). The proportions of total graminoids and total forbs and forb-like plants in the diets of grazing steers were not influenced ( $P = 0.25$ ) by sampling period and were interpreted to indicate steer diets were strongly dominated by graminoids ( $\geq 88.4\%$  of diets).

Selection of big bluestem, dotted gayfeather, heath aster, SL, western ragweed, annual broomweed, and common ragweed were also not influenced ( $P \geq 0.07$ ) by sampling period. Conversely, steer selection of little bluestem decreased ( $P < 0.01$ ) linearly with advancing season, whereas selection of switchgrass, Indian grass, and Baldwin's ironweed increased ( $P \leq 0.04$ ) linearly with advancing season.

**Table 1.** Botanical composition of yearling-steer diets in the Kansas Flint Hills: orthogonal polynomial contrasts by period

Item	Botanical composition (% of diet DM)						P value*			
	Early May	Late May	Early June	Late June	Early July	SEM	Lin.	Quad.	Cubic	Quartic
Total graminoids	91.1	90.2	88.4	90.1	90.3	1.77	0.68	0.25	0.89	0.37
<i>Andropogon gerardii</i>	21.4	23.9	23.3	18.4	20.5	2.51	0.21	0.45	0.08	0.40
<i>Schizachyrium scoparium</i>	27.0	23.6	19.7	16.0	16.9	1.76	<0.01	0.07	0.21	0.76
<i>Panicum virgatum</i>	11.5	13.4	15.7	17.0	18.0	1.53	<0.01	0.52	0.85	0.82
<i>Sorghastrum nutans</i>	9.8	11.1	14.0	15.1	14.1	1.87	<0.01	0.19	0.36	0.78
<i>Bouteloua gracilis</i>	9.7	9.0	7.4	9.3	10.5	1.13	0.46	0.02	0.93	0.21
<i>Bouteloua curtipendula</i>	3.6	3.4	1.4	3.1	2.4	0.81	0.16	0.24	0.72	0.02
<i>Bouteloua dactyloides</i>	5.8	3.4	4.1	6.8	4.7	0.92	0.62	0.29	<0.01	0.33
<i>Carex</i> spp.	1.9	2.0	2.4	3.4	2.9	0.35	<0.01	0.44	0.03	0.28
Unidentified graminoids	0.3	0.4	0.3	0.9	0.3	0.19	0.33	0.08	0.02	0.02
Total forb and forb-like	8.9	9.8	11.6	9.9	9.7	1.77	0.68	0.25	0.89	0.37
<i>Dalea purpurea</i>	1.8	2.0	3.0	2.8	2.1	0.54	0.26	0.05	0.27	0.43
<i>Liatris punctata</i>	0.4	0.8	0.6	0.6	0.5	0.20	0.75	0.16	0.21	0.29
<i>Amorpha canescens</i>	0.1	0.2	<i>tr</i>	0.1	<i>tr</i>	0.07	0.09	0.17	0.18	<0.01
<i>Symphotrichum ericoides</i>	<i>tr</i>	<i>tr</i>	<i>tr</i>	0.1	<i>tr</i>	0.03	0.73	0.30	0.19	0.07
<i>Lespedeza cuneata</i>	<i>tr</i>	<i>tr</i>	<i>tr</i>	0.1	<i>tr</i>	0.04	0.58	0.69	0.44	0.10
<i>Vernonia baldwinii</i>	1.0	1.6	2.2	2.0	2.2	0.59	0.04	0.35	0.77	0.57
<i>Ambrosia psilostachya</i>	2.4	2.0	2.3	2.1	2.3	0.54	0.94	0.58	0.83	0.54
<i>Amphiachyris dracunculoides</i>	0.2	0.1	0.2	0.2	0.2	0.08	0.45	0.39	0.36	0.56
<i>Ambrosia artemisiifolia</i>	1.2	0.9	1.0	0.9	1.0	0.30	0.56	0.41	0.90	0.58
Unidentified forbs	1.8	2.0	2.4	1.1	1.3	0.21	<0.01	<0.01	<0.01	<0.01

\*P values associated with linear (lin.), quadratic (quad.), cubic, and quartic single-degree-of-freedom orthogonal polynomial contrasts.

Unidentified graminoids and forbs were detected in only small amounts (i.e., 0.3% to 0.9% of graminoid fragments and 1.1% to 2.4% of forb or forb-like plant fragments) in yearling beef-steer diets; therefore, the 17 standards that we chose for microhistological characterization of diets were sufficient to allow other researchers evaluating beef cattle diets in the tallgrass prairie region to describe a large majority of diet components. Similar microhistological standards were useful for identifying >90% of plant fragments in beef cattle diets in related experiments (Aubel et al., 2011; Preedy et al., 2013).

The significance of period effects on the appearance of remaining microhistological reference standards in the diets of yearling steers was less clear. Proportions of blue grama in steer diets appeared to be greater (quadratic effect –  $P = 0.02$ ) in May, late June, and early July when compared with early June. Conversely, purple prairie-clover appeared with greatest (quadratic effect –  $P = 0.05$ ) frequency in yearling-steer diets in early June compared with other sampling periods. Proportions of buffalo grass and sedges in yearling-steer diets responded cubically ( $P \leq 0.03$ ) to advancing season, with the greatest selection of both forage types occurring in late June. Proportions of side-oats grama, unidentified graminoids, leadplant, and

unidentified forbs in yearling-steer diets responded quartically ( $P \leq 0.02$ ) to advancing season, with peak selection occurring in early May, late June, late May, and early June, respectively.

Sericea lespedeza and heath aster appeared to be the least-preferred plant species by yearling beef steers among the 17 reference standards that were evaluated microhistologically, resulting in only one period (i.e., late June) in which more than trace amounts of either plant were detected in steer fecal material. Significant heath aster consumption by beef cows grazing tallgrass prairie has been documented recently (Aubel et al., 2011; Preedy et al., 2013), leading us to speculate that steers avoided heath aster from lack of grazing experience. Avoidance of SL by confined beef cattle consuming tallgrass prairie hay (Eckerle et al., 2011) or by grazing beef cows (Preedy et al., 2013) is well documented. Yearling steers likely learned to avoid SL early during the grazing period during each year of our experiment. We speculated that consumption of SL and the condensed tannins therein were unlikely to have caused a negative postingestive feedback response related to a dearth of ruminally available N because consumed amounts were miniscule. Eckerle et al. (2011) reported that beef cows likely developed a flavor-related aversion to SL before a general ruminal malaise occurred. Condensed tannins are astringent

in nature and may be perceived by some herbivores as having a bitter flavor (Provenza et al., 1990). We concluded that steers avoided SL because of the astringent flavor associated with condensed tannins, rather than because of any detrimental effects of condensed tannins on ruminal N metabolism.

Prior research indicated that microhistological evaluation of plant fragments in feces may overestimate graminoid consumption and underestimate forb consumption (Lewis, 1994). Conversely, relatively large proportions of forbs were detected in the diets of yearling beef steers in our experiment. This was also true in reports dealing with beef-cow diets by Aibel et al. (2011) and Preedy et al. (2013).

In spite of limits to detection of highly digestible plant parts, fecal microhistology has several distinct advantages over alternative techniques for characterizing the botanical composition of herbivore diets: analyses of animal feces does not require animal sacrifice or surgical alteration; the number of samples collected is limited only by analytical cost and time; the technique allows for little interaction between researcher and animal and does not interfere with normal grazing habits and movements; and it is thought to reflect a broader spectrum of eating behavior compared with alternative sampling sites because it represents a greater number of grazing bouts (Holechek et al., 1982; McInnis et al., 1983).

### IMPLICATIONS

Yearling beef cattle grazing native tallgrass pastures selected diets of fairly consistent composition during the early May to early July time frame. Yearling-steer diets were strongly dominated by grasses (i.e.,  $\geq 88.4\%$ ); however, a significant proportion of forb or forb-like plants were also detected in steer diets (i.e., 8.9% to 11.6%). In cases where temporal trends were noted in selection of individual plant species, we speculated that those changes were driven by small, coincident changes in availability of alternative diet choices or by learned forage preferences or aversions. Contrary to our original hypothesis, we developed evidence to support the idea that even relatively inexperienced grazers can exhibit strong positive or negative discrimination in diet-component selection. In particular, yearling beef steers avoided sericea lespedeza.

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