



## Diet selection of Raramuri Criollo and Angus x Hereford crossbred cattle in the Chihuahuan Desert<sup>☆</sup>

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### ABSTRACT

Strategies to help producers cope with unpredictable forage production associated with low and variable precipitation patterns on arid rangelands are needed, particularly if global warming trends continue as projected. One option is to identify cattle biotypes compatible with less productive shrubby landscapes. One such biotype is the Raramuri Criollo (RC) from the Copper Canyon of northern Mexico. This small-framed animal exhibits travelling behaviors that allow it to exploit vegetation at a greater distance from water, although it is unknown whether RC have dietary preferences that differ from European breeds typically raised in the southwestern U.S.A. We examined diet selection of RC vs. desert adapted Angus x Hereford (AH) crossbreds typical of the region using DNA metabarcoding to determine the proportion of plant species in fecal samples. Fecal samples were collected from 10 cows of each breed in two adjacent pastures during two seasons (growing and dormancy; four weeks per season) for three consecutive years. Dominant plant species in fecal samples of both cattle breeds were *Atriplex canescens* (four-wing saltbush), *Hoffmannseggia glauca* (hog potato), *Hopia obtusa* (vine mesquite), *Setaria leucopila* (plains bristlegrass), *Sporobolus* spp. (*S. contractus*, *S. flexuosus*, and *S. giganteus*), *Pleuraphis mutica* (tobosa), and *Bouteloua eriopoda* (black grama), which is consistent with previous studies in the region using different techniques for assessing dietary plant composition of other breeds of cattle. Only a few differences were detected between breeds. Compared to AH, fecal samples from RC tended to contain a higher proportion of mesquite and *Yucca* spp. ( $P < 0.07$ ) and less *Ephedra* spp. ( $P < 0.06$ ). The only grass species that differed between breeds was black grama ( $P < 0.05$ ), with AH fecal samples containing about twice as much as RC cows (~8% vs. 4%). This finding could have important implications for conservation of black grama in the Chihuahuan Desert.

### 1. Introduction

Rangelands in the southwestern U.S. experience low and variable precipitation patterns that often lead to variable and sparse forage production. Given the drought patterns of the past two decades and the projections for accelerated warming and drying trends in the future (Williams et al., 2020), and their adverse implications for forage production (McIntosh et al., 2019) and diet quality (i.e., shrub increases; Gherardi and Sala, 2015), identification of cattle biotypes better suited to harsh conditions may provide an option for extensive shrubby rangelands (Estell et al., 2012; Scasta et al., 2016).

Several cattle diet studies have been conducted on the Jornada and

the adjoining Chihuahuan Desert Rangeland Research Center (DCRRC) during the past 50 years using various techniques (Herbel and Nelson, 1966; Rosiere et al., 1975; Hakkila et al., 1987; Winder et al., 1996; Becerra et al., 1998). A few of those studies have compared other breeds to British breeds more typical of the region, including Santa Gertrudis (Herbel and Nelson, 1966) and Brangus (Winder et al., 1996). In addition, Becerra et al. (1998) compared Brangus, Barzona, and Beefmaster but saw no differences in botanical composition of diets among breeds. Only minor differences were detected in the other two studies. Santa Gertrudis ate more total grasses and less *Yucca* and *Salsola kali* (Russian thistle) (Herbel and Nelson, 1966). Winder et al. (1996) observed greater preference for *Sporobolus* by Brangus vs. Angus and/or Hereford

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and greater preference for *Yucca* and total shrubs than British breeds during winter. However, no heritage breeds have been compared to more traditional British breeds.

Raramuri Criollo (RC) is a cattle biotype that has undergone natural selection for the past 500 years on rugged terrain in the Copper Canyon of northern Mexico (Anderson et al., 2015), resulting in a small-framed (~350 kg adult weight) mobile animal (McIntosh et al., 2020). Our work on this biotype revealed that they travel farther per day and farther from water than Angus x Hereford (AH) crossbred cattle (Nyamuryekung'e et al., 2021; Peinetti et al., 2011; Speigal et al., 2019), and therefore may be more adapted to extensive arid rangelands with sparse forage distribution. Although anecdotal observations suggest this biotype may have a wider diet breadth (including woody species and cacti) than traditional beef breeds of European descent typically raised on arid rangelands (Anderson et al., 2015), no published information exists on diet selection by this biotype. Therefore, our objective was to compare diet selection and browsing behavior of RC and AH cattle typically found in the arid southwestern United States during both growing and dormant seasons. Our hypothesis was that RC cattle would exhibit a wider diet breadth and contain a greater proportion of shrubs in fecal samples.

## 2. Materials and methods

### 2.1. Study site

The study was conducted at the Jornada Experimental Range (JER), located in southern New Mexico (USA) in the northern part of the Chihuahuan Desert (32.6° N, 106.8° W; elevation 1300–1600 m). Long-term mean annual precipitation is 247 mm with 53 percent occurring between July and September. Average ambient temperature ranges from 36 °C in June to 13.3 °C in January. The spatially dominant ecological sites in the study pastures are shown in Fig. 1. Vegetation in the study pastures is dominated by honey mesquite (*Prosopis glandulosa* Torrey) mixed with black grama (*Bouteloua eriopoda*), dropseeds (*Sporobolus* spp.), and threeawns (*Aristida* spp.). Other common species include four-wing saltbush (*Atriplex canescens*), burrograss (*Scleropogon brevifolius*), tobosa (*Pleuraphis mutica*), broom snakeweed (*Gutierrezia sarothrae*), soap tree yucca (*Yucca elata*), and leatherweed croton (*Croton pottsii*). The two adjacent pastures (1190 and 1165 ha) used in this study each contained two watering tanks (Fig. 1). Both pastures had similar topography, soils, vegetation, and distribution of drinking water. A vegetation map depicting first, second, and third most dominant plant species is presented in Appendix Fig. 1.

### 2.2. Animals

The study was approved by the New Mexico State University Institutional Animal Care and Use Committee (Protocol #2015-021). Ten RC and 10 AH cows (4 years or older) were randomly selected from Jornada herds each period, with no cows used in consecutive periods. Cows grazed in two adjacent pastures for 28 days in summer 2015 (July–August), winter 2015–2016 (January–February), summer 2016 (August–September), winter 2016–2017 (January–February), summer 2017 (August–September), and winter 2017–2018 (January–February). Each breed grazed each pasture 14 days per period and cows were then switched to the reciprocal pasture. Cows were nursing in summer (calves <2 wk old at the beginning of each summer period) and dry in winter. Breeds grazed pastures separately and were switched to adjacent pastures at the mid-point of each period (end of week 2) to assure both groups were exposed to the same pasture but without social interaction between breeds. Prior to the study and between periods, cows grazed native range similar to the experimental pastures as part of the main herds (RC and AH herds maintained separately and supplemented as needed to maintain body condition). Sampling dates for the growing season (i.e., summer periods) varied due to interannual variation in precipitation patterns. Pastures were not grazed except during the two 4-

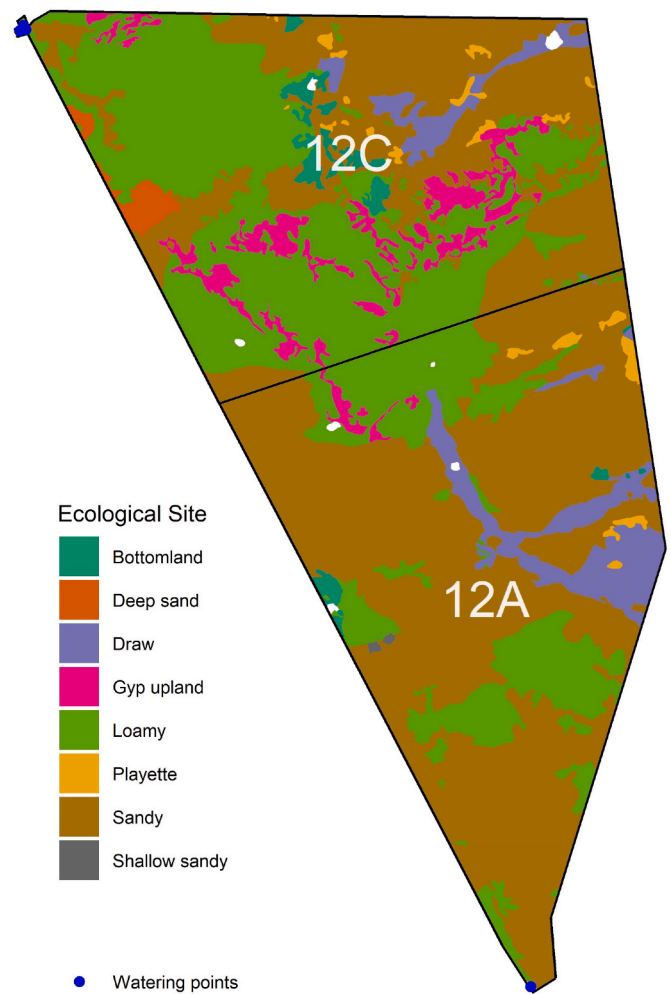


Fig. 1. Location of water tanks and dominant ecological sites in the study pastures.

week periods each year. Breeds were randomly assigned to pasture each year to address potential confounding of forage availability. Consequently, AH grazed pasture 12A first in year 1 and 2 and RC were first in year 3. In addition, stocking rates were ~106–108 ha/AUM (well below the recommended stocking rate) and grazing intensity was estimated to be light to conservative based on visual estimates of key forage species use, which should have minimized the possibility of differences in forage availability between breeds.

### 2.3. Sampling and analysis

Fecal samples (rectal grab samples) were collected from each cow on day 14 in each pasture in both seasons all three years. After switching to the alternate pastures, cows were sampled again on day 28, resulting in 240 samples over the study. Samples were frozen for subsequent analysis. Dormant samples from year 2 were inadvertently dried (60 °C). However, preliminary tests indicated no difference in results (number of hits) from frozen and dried fecal samples. Fecal samples were analyzed for plant DNA using DNA metabarcoding with chloroplast gene *trnL* primers by a commercial laboratory (Jonah Ventures) as described by Craine et al. (2015, 2016).

Relative concentrations of Exact Sequence Variants (ESVs) were identified that represent a group of taxa with similar *trnL* sequences. Sequences subjected to Exact Sequence Variants analysis yielded 3,106 unique ESVs. Taxa were identified using the reference library each ESV was known to represent. Because the reference library included global

taxa, we confined the reference library to taxa included in the *Pocket Guide to the Flora of the Jornada Plain* (Allred, 2011) to identify taxa that could plausibly exist in the study pastures. We also collected specimen vouchers for 20 prevalent species not represented in the trnL library for inclusion in the reference library, and resolved ESVs based on results of the 20 plant samples we submitted for trnL analysis. This process allowed us to resolve 1,450 ESVs to an individual species, 788 to an individual genus, 810 to an individual family, and 47 to a family group. We could not resolve 11 ESVs observed in fecal samples to any taxa. The 1,450 ESVs resolved to a single species spanned only 87 unique plant species. Twenty-seven species mapped to a single ESV, but most mapped to more than one. Species with the highest number of distinct ESVs included black grama (74), *Hoffmannseggia glauca* (hog potato; 74), *Hopia obtusa* (vine mesquite; 101), *Echinochloa colona* (119), and four-wing saltbush (247). Because of the possible one-to-many, many-to-one, and many-to-many relationships between ESVs and plant taxa, we chose not to analyze ESV diversity as a response. Rather, our strategy was to resolve individual ESVs to the finest taxonomic resolution possible. The ESV data contained 67 plant taxa with an overall mean of 1% or greater of total ESVs. ESVs were summed to produce a total ESV count per taxonomic group for each animal within pasture, season and year, and then divided by the total ESV count for that animal to calculate relative abundance of that taxonomic group in the diet. The relative abundance of an ESV is assumed to scale with the proportion of protein derived from a given species, given the relationship between chloroplast density in leaves and plant protein concentrations (Craine, 2021).

#### 2.4. Statistical Analysis

We analyzed the effect of breed, season, year, and their interactions on the relative proportions of plant species in fecal samples. For taxa with multiple ESVs (e.g., four-wing saltbush), this approach assumes multiple ESVs representing the same taxa represent either genotypic variation or sequencing error. Therefore, adding these counts together serves to quantify the total number of reads for a given taxa more accurately rather than inflate their presence in the diet. We assigned each ESV to a functional group (grass, forb, shrub, or unknown) and calculated the proportion of total ESV counts for analysis. Fitting a statistical model to the data proved difficult for most taxa due to the high variability between individual cows, pastures, seasons, and years. We opted to sum species and species groups to genus level for analysis in some cases. Sixteen species or genera and four functional groups were present in sufficient amounts in all periods to analyze. Mixed-effects models were used to separately analyze these groups with breed, season, year, and their interactions as fixed effects. Random effects were pasture, pasture nested in year\*season, period\*breed\*pasture nested in year\*season, and cow ID nested in breed\*year\*season. We used R version 4.0.2 for data processing and graphics (Wickham et al., 2019; R Core Team, 2020) and the MIXED procedure in SAS 9.4 software for model fitting (SAS Institute Inc., Cary, NC, USA). Main effects and interactions analyzed included breed, season, year, breed\*season, breed\*year, season\*year, and breed\*season\*year.

### 3. Results and discussion

#### 3.1. Dominant plant species in fecal samples

The top 20 plant taxa (% mean  $\pm$  SD) by season, year, and across the entire study are presented in Table 1. Over all three years of the study, the highest percentage of ESVs in fecal samples was four-wing saltbush. This shrub was consumed in greater amounts during winter than summer and was the predominant ESV in winter samples. Four-wing was the dominant ESV in year 2 and ranked fourth in years 1 and 3. The second most dominant species overall was hog potato; it was the dominant species in summer and was consistently present in all three years (Table 1). Vine mesquite, the third most dominant species, was

**Table 1**

Dominant plant species in fecal samples as a percentage of total ESVs (mean  $\pm$  SD, ranking for each category in parentheses) by season and year pooled across cattle breeds.

Taxa	Total	Season		Year		
		Winter	Summer	1	2	3
Atriplex canescens <sup>s</sup>	13.04 $\pm$ 15.27 (1)	24.74 $\pm$ 13.18 (1)	1.34 $\pm$ 3.86 (13)	9.53 $\pm$ 8.11 (4)	17.25 $\pm$ 19.68 (1)	12.34 $\pm$ 16.67 (4)
Hoffmannseggia glauca <sup>f</sup>	11.24 $\pm$ 17.27 (2)	0.07 $\pm$ 0.11 (58)	22.42 $\pm$ 18.73 (1)	8.78 $\pm$ 16.15 (6)	7.28 $\pm$ 11.73 (3)	17.67 $\pm$ 22.57 (1)
Hopia obtusa <sup>s</sup>	10.14 $\pm$ 12.39 (3)	15.19 $\pm$ 14.99 (2)	5.09 $\pm$ 6.39 (5)	11.34 $\pm$ 10.18 (3)	6.55 $\pm$ 9.15 (4)	12.54 $\pm$ 17.16 (3)
Setaria leucopila <sup>s</sup>	7.08 $\pm$ 10.73 (4)	10.35 $\pm$ 12.94 (4)	3.80 $\pm$ 7.06 (7)	15.92 $\pm$ 13.50 (1)	3.85 $\pm$ 7.44 (8)	1.47 $\pm$ 15) (15)
Euphorbia spp. <sup>fs</sup>	6.97 $\pm$ 12.06 (5)	0.04 $\pm$ 0.12 (69)	13.91 $\pm$ 14.12 (2)	3.31 $\pm$ 4.32 (8)	4.75 $\pm$ 7.34 (7)	12.87 $\pm$ 18.57 (2)
Sporobolus spp. <sup>s</sup>	6.20 $\pm$ 11.07 (6)	0.88 $\pm$ 1.13 (17)	11.52 $\pm$ 13.90 (3)	13.55 $\pm$ 16.39 (2)	2.63 $\pm$ 3.03 (10)	2.43 $\pm$ 5.69 (7)
Pleuraphis mutica <sup>s</sup>	5.94 $\pm$ 7.44 (7)	6.09 $\pm$ 7.02 (6)	5.78 $\pm$ 8.14 (4)	7.71 $\pm$ 8.72 (7)	7.89 $\pm$ 8.75 (2)	2.21 $\pm$ 2.35 (8)
Bouteloua eriopoda <sup>s</sup>	5.90 $\pm$ 8.42 (8)	10.83 $\pm$ 9.58 (3)	0.97 $\pm$ 1.86 (19)	9.43 $\pm$ 12.80 (5)	6.17 $\pm$ 5.50 (5)	2.12 $\pm$ 2.86 (9)
Ephedra spp. <sup>s</sup>	3.49 $\pm$ 5.84 (9)	6.69 $\pm$ 6.96 (5)	0.29 $\pm$ 0.62 (35)	0.37 $\pm$ 0.75 (28)	3.79 $\pm$ 3.32 (9)	6.32 $\pm$ 8.95 (5)
Boerhavia intermedia <sup>f</sup>	2.37 $\pm$ 5.70 (10)	0.03 $\pm$ 0.07 (78)	4.71 $\pm$ 7.48 (6)	0.13 $\pm$ 0.25 (34)	5.11 $\pm$ 8.33 (6)	1.87 $\pm$ 4.77 (11)
Prosopis glandulosa <sup>s</sup>	2.16 $\pm$ 3.22 (11)	3.43 $\pm$ 4.07 (7)	0.89 $\pm$ 1.28 (21)	1.27 $\pm$ 1.15 (12)	0.71 $\pm$ 0.94 (27)	4.50 $\pm$ 4.73 (6)
Croton pottsii <sup>f</sup>	1.29 $\pm$ 2.81 (12)	0.38 $\pm$ 0.84 (27)	2.19 $\pm$ 3.74 (8)	0.76 $\pm$ 1.46 (18)	2.59 $\pm$ 4.47 (11)	0.51 $\pm$ 0.97 (26)
Aristida spp. <sup>s</sup>	1.18 $\pm$ 1.87 (13)	1.41 $\pm$ 2.16 (8)	0.94 $\pm$ 1.60 (20)	1.41 $\pm$ 2.37 (10)	1.99 $\pm$ 1.95 (13)	0.13 $\pm$ 0.30 (46)
unknown Asteraceae <sup>fs</sup>	1.15 $\pm$ 1.36 (14)	1.18 $\pm$ 1.42 (11)	1.11 $\pm$ 1.35 (16)	0.63 $\pm$ 0.35 (19)	1.37 $\pm$ 1.61 (19)	1.44 $\pm$ 1.70 (16)
Sphaeralcea coccinea <sup>f</sup>	1.09 $\pm$ 1.54 (15)	0.51 $\pm$ 0.92 (22)	1.67 $\pm$ 1.84 (12)	1.08 $\pm$ 1.77 (14)	0.67 $\pm$ 1.09 (29)	1.52 $\pm$ 1.76 (14)
Zinnia grandiflora <sup>f</sup>	1.05 $\pm$ 3.61 (16)	0.18 $\pm$ 0.55 (36)	1.91 $\pm$ 5.03 (10)	2.28 $\pm$ 6.10 (9)	0.60 $\pm$ 1.53 (32)	0.26 $\pm$ 0.68 (33)
Echinochloa colona <sup>s</sup>	1.04 $\pm$ 1.56 (17)	1.27 $\pm$ 1.63 (9)	0.81 $\pm$ 1.53 (27)	0.48 $\pm$ 0.55 (26)	1.94 $\pm$ 2.43 (14)	0.70 $\pm$ 0.64 (22)
Oenothera suffrutescens <sup>f</sup>	1.02 $\pm$ 1.78 (18)	0.09 $\pm$ 0.21 (51)	1.95 $\pm$ 2.16 (9)	0.79 $\pm$ 1.16 (17)	0.22 $\pm$ 0.49 (45)	2.05 $\pm$ 2.61 (10)
Krameria lanceolata <sup>f</sup>	0.90 $\pm$ 2.95 (19)	<0.01 $\pm$ 1.49 (149)	1.80 $\pm$ 4.05 (11)	0.19 $\pm$ 0.38 (32)	0.88 $\pm$ 2.47 (24)	1.63 $\pm$ 4.59 (13)
unknown Poaceae <sup>s</sup>	0.86 $\pm$ 0.75 (20)	0.91 $\pm$ 1.00 (16)	0.81 $\pm$ 0.40 (26)	0.89 $\pm$ 0.37 (16)	1.15 $\pm$ 1.19 (21)	0.54 $\pm$ 0.24 (25)

<sup>s</sup> = grass, <sup>s</sup> = shrub, <sup>f</sup> = forb.

consistently high across years, especially in winter. The fourth most abundant species overall, *Setaria leucopila* (plains bristlegrass), was a major component in both summer and winter, and was the most dominant plant species in year 1 samples. The sixth, seventh, and eighth

highest percentages overall were grasses: *Sporobolus* spp. (*S. contractus*, *S. flexuosus*, and *S. giganteus*), tobosa, and black grama (Table 1). *Sporobolus* spp. were the third most dominant species in summer samples but were present in only trace amounts in winter diets. Tobosa was in the top

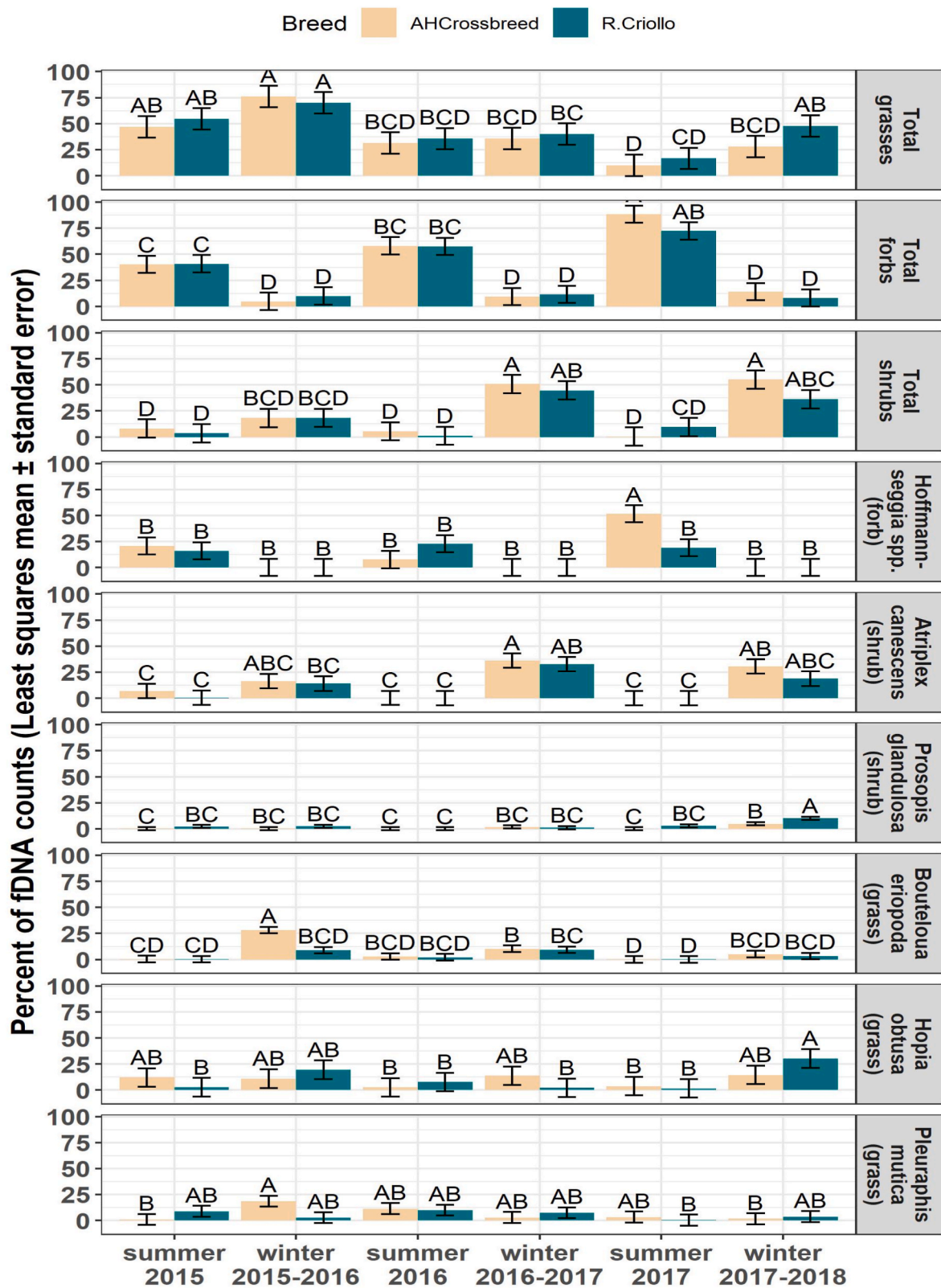


Fig. 2. Percentage of ESV counts (Least square means ± SE) for plant functional groups (forb, grass, shrub) and selected dominant plant species by season and year for Angus x Hereford and Raramuri Criollo cows. Means with the same letter do not differ ( $P > 0.05$ ).

10 species present in both winter and summer and in all three years. Black grama was high in winter samples and was present in only trace amounts in summer collections, but was a dominant component all three years. *Ephedra* spp. ranked ninth overall and were an important component of winter diets. Other common species above 1% of the total were honey mesquite, leatherweed croton, and threeawns. Mesquite and threeawns were more prevalent diet components in winter, while leatherweed ranked higher during summer.

Across breed, forb intake was greater in summer and shrub intake was higher in winter ( $P < 0.05$ ; Fig. 2). Grass intake showed no clear relationship with season, but generally decreased across years (Fig. 2). The decrease in grass consumption from year 1 to year 3 did not differ between breeds in either winter or summer ( $P > 0.05$ ), but over time, grass intake based on visual patterns appeared to decline while forb intake increased during summer. During winter, patterns were less pronounced, though grass intake decreased and shrub intake increased in the first two years. In year 3, breeds diverged during winter, with RC consuming more grass and AH consuming more shrubs. Several differences were detected in the consumption of life forms or individual species within season and/or year. Some of these differences may be

related to differences in seasonal rainfall. Precipitation patterns (Fig. 3) varied among seasons and years, and drought conditions were evident during much of the three-year study. Thus, carryover effects from year to year may have differentially affected availability of certain forage species and functional groups given that some species are more drought tolerant. Seasonal differences in foraging behavior may also have been influenced by nutritional demands due to physiological state (lactating vs. dry) of the cows.

### 3.2. Comparison with previous diet research

Key forage species present in fecal samples from this study are in general agreement with what others have found on the Jornada or the adjacent CDRRC. [Herbel and Nelson \(1966\)](#) examined diets of Hereford and Santa Gertrudis cows in a three-year study on the Jornada and reported that overall, the key species consumed by these cattle breeds were black grama in winter and tobosa in summer. About 50–60% of cattle diets were perennial grasses, with a seasonal range of 35–72% (highest in summer and fall). Important dietary species in that study were mesa dropseed, black grama (during winter), *Sporobolus airoides*

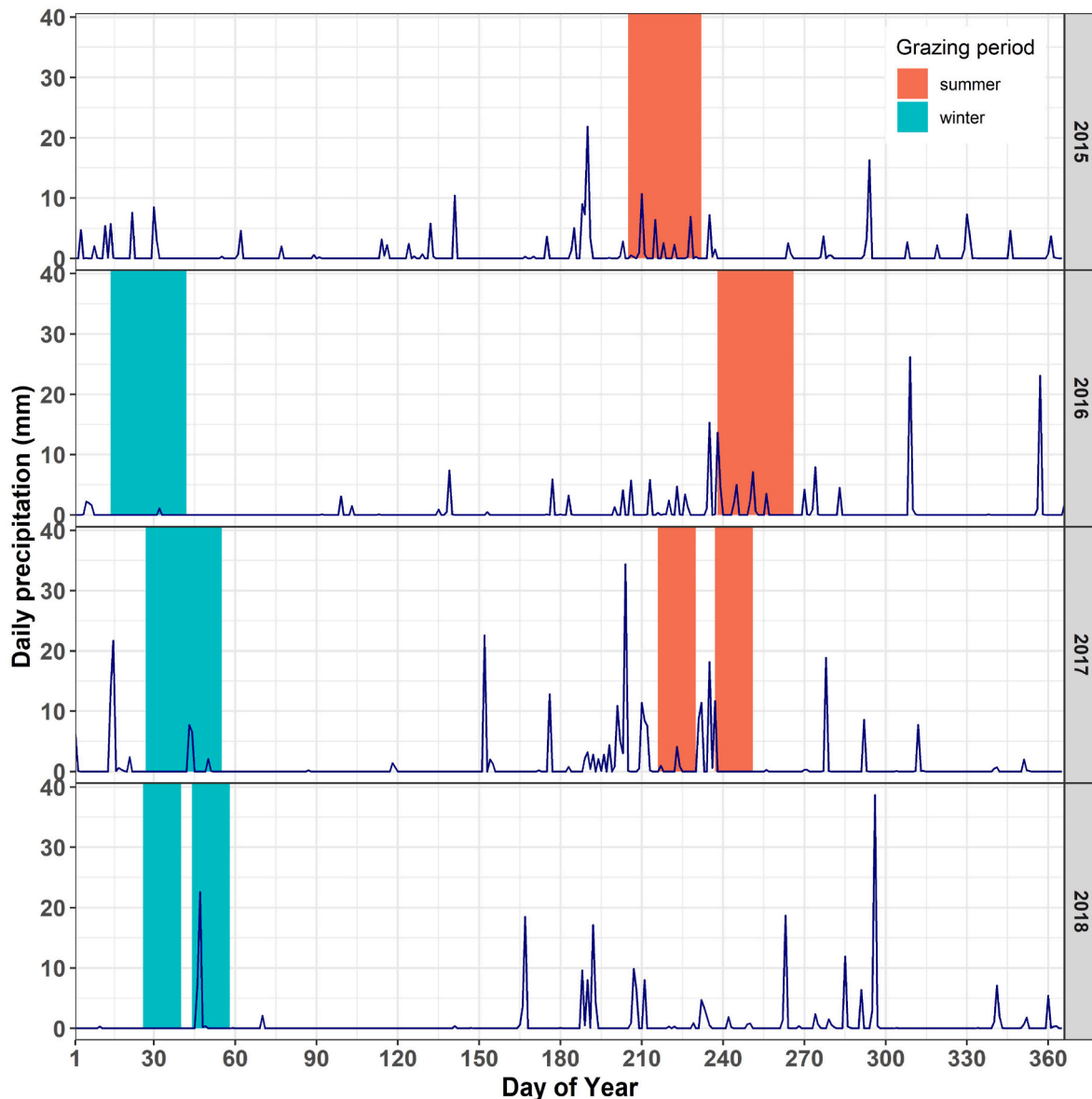


Fig. 3. Daily precipitation during the study in relation to individual study periods.

(alkali sacaton, in spring), burrograss (throughout year), *Aristida longisetata* (red threeawn), and tobosa (in summer). Leatherweed croton (12%) and *Psilostrophe tagetinae* (wooly paperflower, as high as 16%) were consistently important perennial forbs. Shrubs were generally not important diet components except yucca during winter and spring and *Ephedra trifurca* (nearly 5% in fall). Mesquite and four-wing saltbush were both below 1% of the diet. Rosiere et al. (1975) examined diets of Hereford steers in a year-long study on the CDRRC and reported diet composition across seasons to be approximately 45% grasses, 32% forbs, and 19% shrubs. Grass intake was highest in summer, lowest in spring, and intermediate in fall and winter. The most prevalent grass was mesa dropseed, while black grama was a moderate dietary component (0–20%). *Aristida adsonensis* (sixweeks threeawn) was an important component of summer and fall diets. Forb consumption was generally highest in spring and summer, with Russian thistle the most dominant forb, especially in fall and winter. Leatherweed croton and wislizenus spectaclepod were also low but consistent dietary components. Shrub intake was low except in spring, when about 70% of the diet consisted of soaptree yucca, although some *Ephedra* use (0–8%) was observed in fall and winter. Mesquite was found only in one summer collection (primarily pods but some leaves), and four-wing saltbush was not detected in their samples. Hakkila et al. (1987) examined diets of Hereford x Brangus crossbred steers on the Jornada in a year-long study and found that grasses made up over 60% of the diet and over 70% in four of their six collection periods. Mesa dropseed was the predominant species, comprising well over half of all grass intake in four of the six collections, with vine mesquite the dominant grass in the other two collection periods. Across the study, black grama was only 2% of the total diet and was present in only trace amounts in all but the first collection when it was 8% of the diet. During two seasons (spring and fall), forbs and shrubs were collectively over 60% of the diet. Leatherweed croton and *Descurainia pinnata* (tansy mustard; spring only) were important forbs and honey mesquite (11%) and soaptree yucca (8%) were the most important shrub species consumed by cattle across seasons. Most of the mesquite consumption was fruits in late summer and fall and dropped leaves in spring. Four-wing saltbush constituted 3% of the diet, and almost all of this was during spring. Winder et al. (1996) examined Brangus, Hereford, and Angus cow diets on the CDRRC during three seasons and found mesa dropseed and threeawns to be the most common grass components in the fall, with forbs comprising over 50% of the diet during this time, and leatherweed croton being nearly all of the forb component. Total grasses in the winter diet ranged from 84 to 96%, with black grama being the dominant winter forage, ranging from 61% for Brangus to 85% for Angus. Shrub consumption was low during this period except for Brangus, which consumed about 12% yucca. Summer diets were 55–75% grasses, dominated by mesa dropseed. Croton was again present in high amounts, ranging from 13 to 40%, depending on breed. Becerra et al. (1998) examined diets of Barzona, Brangus, and Beefmaster cows in a year-long study on the CDRRC and reported that grass intake ranged from over 90% in winter to a low of 62% in summer. Mesa dropseed, threeawns, and black grama were the primary species consumed. Forb consumption ranged from 28% in summer to 6% in winter, with leatherweed croton the dominant forb in all seasons (highest in summer, lowest in winter). Across the study, shrubs constituted 8% of the diet. Mesquite was the primary shrub consumed, ranging from 1% in winter to over 10% in late summer, while yucca and ephedra were minor shrub components.

Nearly all of the plant species appearing in the above studies were important constituents of diets of cows in our study. The most significant deviation between previous studies and our study was the strong signal for four-wing saltbush in our samples. In contrast, four-wing was a minor component of cattle diets in the five studies on the Jornada and CDRRC, while grasses were present in consistently high concentrations in those studies (Becerra et al., 1998; Hakkila et al., 1987; Herbel and Nelson, 1966; Rosiere et al., 1975; Winder et al., 1996). The distribution of life forms in cattle fecal samples in our study was approximately 40%

grasses, 35% forbs and 20% shrubs, with black grama representing about 6% of fecal samples (Table 2). In contrast, total grasses were typically higher in the previous studies, accompanied by lower percentages of shrubs and forbs. With the exception of Rosiere et al. (1975) who reported 45% grass in cattle diets, all the other reports indicated a range of >55% to nearly 80% total grasses in cattle diets (Becerra et al., 1998; Hakkila et al., 1987; Herbel and Nelson, 1966; Winder et al., 1996). Black grama was an important component of total grasses in some studies (Becerra et al., 1998; Rosiere et al., 1975) but not others (Hakkila et al., 1987). The ~6% black grama observed in the present study fell approximately midway across all studies. Shrub intake was typically low in the previous studies (Becerra et al., 1998; Herbel and Nelson, 1966; Rosiere et al., 1975), although yucca was approximately 70% of the spring diet in the latter study. It is difficult to make direct comparisons among studies because of differences in seasons examined in different studies. For example, Hakkila et al. (1987) observed >60% combined shrubs plus forbs in spring and fall, and Winder et al. (1996) noted over 50% forbs in fall diets.

Discrepancies among studies may be due to several factors. Some of the studies examined only diet composition during only one year and most examined at least four periods (seasons) per year. Seasonal and annual precipitation in this region is extremely variable (McIntosh et al., 2019). It is important to note these studies were conducted from >20 to over 50 years ago. Multiple drought cycles and their impacts on existing vegetation undoubtedly affect species composition on these native rangelands. McIntosh et al. (2019) examined long-term data on the CDRRC from 1967 to 2018 and found that perennial grass production in the second half of the time series decreased by 43% compared to the first interval. The reduction was accompanied by lower (~19%) and more variable precipitation and ~1 °C greater mean maximum and mean ambient temperature during the latter interval. Also, the previous studies used several different breeds of cattle of varying ages, physiological stages and sex and multiple techniques were used to determine botanical composition of cattle diets, including visual diet scans and microhistological analyses of esophageal or fecal samples. Microhistological analysis is commonly used to determine diet botanical composition of livestock based on fecal forage fragments and accounts for differential digestibility of various plant species (Holechek et al., 1982).

Although DNA barcoding is a relatively new technique and few

**Table 2**

Effect of cattle breed on percentage of plant species in fecal samples of Angus x Hereford crossbred (AH) and Raramuri Criollo (RC) cows grazing arid rangeland.

Taxa	Means		Standard Error		P-value
	AH	RC	AH	RC	
<i>Aristida</i> spp. <sup>g</sup>	1.29	1.13	0.47	0.47	0.71
<i>Atriplex canescens</i> <sup>s</sup>	15.04	11.00	3.41	3.41	0.31
<i>Bouteloua eriopoda</i> <sup>s</sup>	7.85	3.96	1.56	1.56	<0.05
<i>Echinochloa colona</i> <sup>s</sup>	1.07	1.01	0.70	0.70	0.92
<i>Ephedra</i> spp. <sup>s</sup>	4.74	2.29	0.72	0.72	0.05
<i>Hoffmannseggia glauca</i> <sup>f</sup>	13.40	9.73	3.35	3.35	0.45
<i>Hopia obtusa</i> <sup>s</sup>	9.61	10.66	6.14	6.14	0.80
<i>Pleuraphis mutica</i> <sup>g</sup>	6.44	5.48	2.22	2.22	0.75
<i>Prosopis glandulosa</i> <sup>s</sup>	1.23	3.05	0.62	0.62	0.06
<i>Scleropogon brevifolius</i> <sup>s</sup>	0.13	0.18	0.15	0.15	0.70
<i>Setaria leucopila</i> <sup>s</sup>	4.84	9.36	2.36	2.36	0.20
<i>Solanum</i> spp. <sup>f</sup>	0.12	0.25	0.10	0.10	0.36
<i>Sphaeralcea coccinea</i> <sup>f</sup>	0.59	1.60	0.45	0.46	0.14
<i>Sporobolus</i> spp. <sup>g</sup>	4.75	8.93	2.38	2.38	0.24
<i>Yucca</i> spp. <sup>s</sup>	0.06	0.80	0.27	0.27	0.07
<i>Zinnia grandiflora</i> <sup>f</sup>	2.05	0.04	1.07	1.07	0.20
Forb	35.83	33.41	3.37	3.38	0.62
Grass	37.99	44.04	5.66	5.66	0.29
Shrub	23.00	18.88	3.76	3.77	0.43
Unknown	3.17	3.53	0.91	0.92	0.79

<sup>g</sup> = grass, <sup>s</sup> = shrub, <sup>f</sup> = forb.

reports exist comparing it to other methods, King and Schoenecker (2019) examined feral horse diets using microhistology and DNA barcoding. Fecal samples from horses contained mostly grasses with both techniques, but the grass species present differed between the two techniques and the proportion of forbs was much lower with microhistology than DNA metabarcoding. Also, the amount and timing of precipitation in the Chihuahuan Desert is extremely variable within and among years and timing of grazing relative to precipitation likely impacted forage species available for consumption. Though DNA metabarcoding is a relatively new technology, that fact that the most prevalent dietary species generally align with previous studies on similar rangelands using a variety of techniques suggests the method identifies species that would be expected to be present in fecal samples from cattle grazing these pastures. As mentioned earlier, some of the differences could be due to protein concentration of various species. The above studies all reported botanical composition of the diet on a dry matter or organic matter basis. Relative amounts of ESV counts per plant species in fecal DNA reflect relative amounts of dietary protein from those species (Craine et al., 2016). However, counts can be scaled to biomass intake if protein content of species of interest is known (Craine et al., 2016). Thus, if an animal consumes equal amounts of two species, but one species has twice the protein of the other, it is expected that the high-protein species would have twice the number of reads as the low-protein species. This is further complicated by the fact that protein content of different parts of a given plant and/or the same plant at different times of the year can vary. As a general rule, plants on Chihuahuan Desert rangelands contain less protein during dormancy than the growing season, and forbs and shrubs typically have a higher protein content than grasses (Nelson et al., 1970). Moreover, the proportion of

ESVs for a given taxonomic unit to the total ESVs in that sample should not be influenced by water content, negating the need to report estimates of species percentage in the diet on a dry matter basis.

Based on the report by Nelson et al. (1970), crude protein (CP) concentrations for important perennial grasses in our study are approximately 4.5–7% (black grama), 4–10.5% (threeawn), 4.5–9% (mesa dropseed), and 4–9% (alkali sacaton). Black grama CP was more consistent across seasons while the other species were lowest during dormancy (Nelson et al., 1970). Thus, grasses, especially during dormancy, might be underestimated by the DNA technique. Leatherweed croton was an important component of our cattle diets that contains about 8.5% CP in dormancy and over 16% during the growing season (Nelson et al., 1970). Thus, it would be expected to be inflated in summer vs. winter diets, and overestimated relative to grasses with lower CP content. In the Nelson et al. study, the only shrub consumed consistently was yucca (~9–11% CP), though mature four-wing salt-bush, mesquite leaves/beans, and Mormon tea were eaten occasionally. Those species contained ~13, 13, and 9% CP, respectively, and would again be expected to be overestimated relative to grass concentration in samples from this study.

### 3.3. Breed differences

Breed differences in diet selection (the main focus of this study) should not be influenced by CP content of forages consumed by cattle, since both breeds were exposed to the same plants in the same pastures during the same years within two weeks of each other. Sixteen plant genera or species groups were present in cattle fecal samples in sufficient quantity in all sampling periods to analyze by breed across time (Fig. 4).

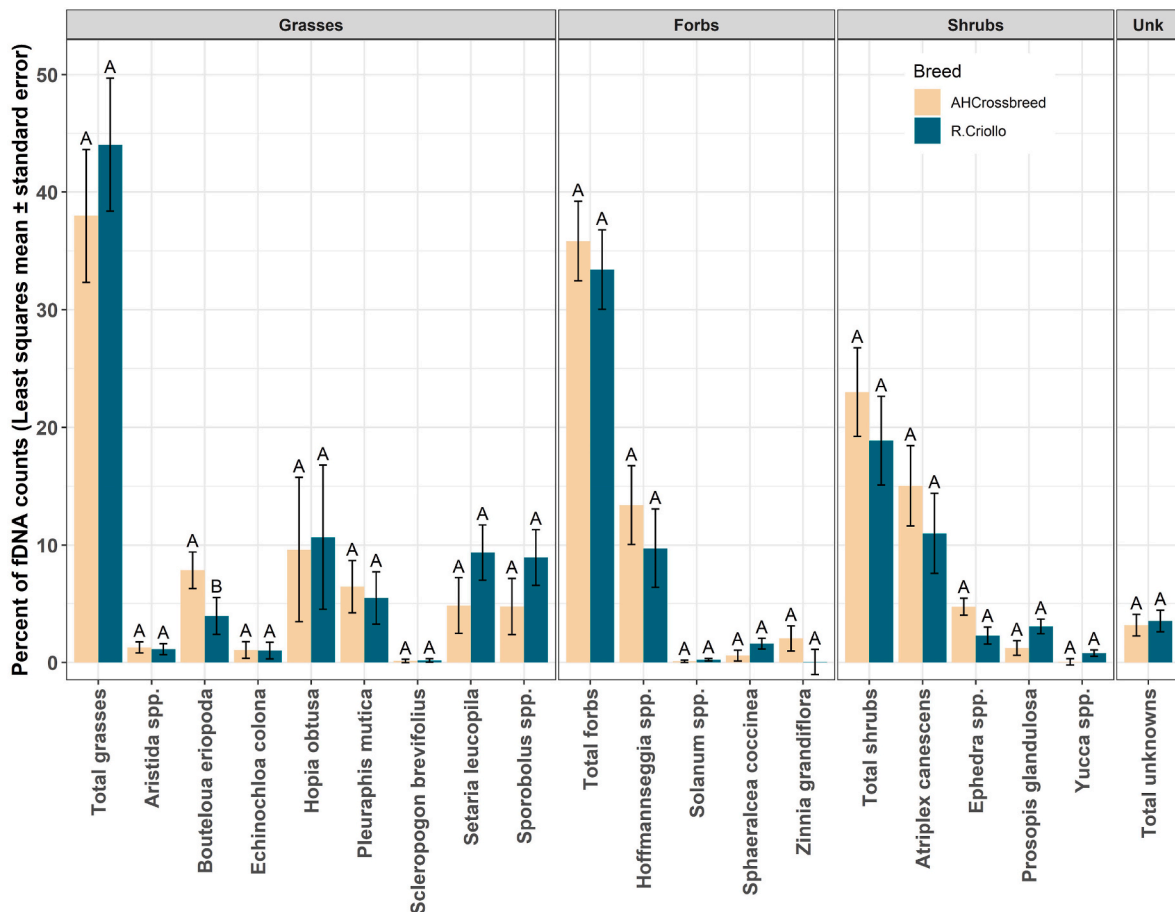


Fig. 4. Effect of breed on percentage of DNA counts from 16 dominant plant species and four functional groups in fecal samples from Angus x Hereford and Raramuri Criollo cows grazing arid rangeland.

Only black grama content differed ( $P < 0.05$ ) between breeds, with AH fecal samples containing about twice as much as RC cows (Table 2). Although AH consumed numerically more black grama during winter all three years, the difference was particularly evident in year 1 (Fig. 2). In addition, RC cows tended to eat more mesquite ( $P < 0.06$ ) and *Yucca* spp. ( $P < 0.07$ ) and less *Ephedra* spp. ( $P < 0.06$ ) than AH (Table 2). No functional group categories (grass, shrub, forb, unknown) differed between breeds. Though mean differences between breeds were numerically large for some other species (e.g., four-wing saltbush, hog potato, plains bristlegrass, and dropseeds), no statistical differences were detected. When pooled by functional group (Fig. 4), no breed differences were detected, although RC consumed numerically more grass and less shrubs and forbs than AH (Table 2), in contrast to our expectations.

Some notable differences were observed for breed within season by year. Breed  $\times$  season  $\times$  year effects were detected ( $P < 0.05$ ) for eight genera or species groups, with most of those differences occurring in only one season/year combination. Breed differences were detected for *Zinnia grandiflora* in summer 2015 and hog potato in summer 2017, with AH fecal samples containing greater concentrations than RC fecal samples in both cases ( $P < 0.05$ ). Also, AH cows consumed more *Ephedra* spp. and RC consumed more mesquite during winter 2017 ( $P < 0.05$ ). Most of the shrub consumption by both breeds was four-wing saltbush (Fig. 2). Intake of black grama was substantially higher for AH during winter 2015, while RC consumed more plains bristlegrass during the same period ( $P < 0.05$ ). *Yucca* spp. differed among breeds in winter 2016 and *Aristida* spp. differed in both summer and winter of 2016 ( $P < 0.05$ ). No differences were observed between breeds within season/year combinations for forbs, grasses, or shrubs despite large numerical differences in some cases (Fig. 2).

Though little published information regarding diet selection of criollo cattle is available, anecdotal information suggests that RC would be expected to consume more shrubs and a wider breadth of plant species than breeds of European descent. Lack of observed differences among plant functional groups is counter to our hypothesis, given that RC are considered heavy browsers by many. However, a behavior study by Koppa (2007) reported that RC spent more time in tobosa and mixed grasses vegetation classifications than AH on 10 or more of the 15 days examined in the same pastures, while AH spent more time on black grama. This study was conducted in October and November when tobosa would be expected to be low palatability, yet RC spent up to 42% of their day in tobosa areas. In contrast, preliminary findings of Duni et al. (2021) at the CDRRC adjacent to the Jornada using a visual scan method indicated no difference in browsing frequency for RC vs. Brangus cows during summer but RC browsed more than Brangus during spring (~65% vs. 30% browsing frequency, respectively). In a companion behavior study conducted at the same time as our study, Nyamuryekung'e et al. (this issue) monitored animal movement and vegetation

use of these same animals. During winter, AH preferred pixels with highest black grama density (and avoided other grasses; i.e., threeawns, dropseeds) while RC strongly avoided black grama during dormancy (and moderately avoided other grasses). In the summer, when tobosa is generally most palatable, both breeds strongly preferred areas with a medium to high density of tobosa, while neither breed preference was related to tobosa pixels in winter. These findings support both our observations regarding breed differences and seasonal differences in consumption of black grama and tobosa.

#### 4. Conclusions

Our results generally did not support our hypothesis that RC have a wider diet breadth or consume more shrubs than AH cows. However, black grama, a critical grass species of ecological and conservational importance, was present in lower concentration in fecal samples from RC cows. These results suggest this heritage breed may potentially have utility for managing black grama rangelands, presumably because they spend more time foraging on other species of less conservational concern.

#### CRediT authorship contribution statement

**R.E. Estell:** Conceptualization, Writing – review & editing. **S. Nyamuryekung'e:** Data Collection, Writing – review & editing. **D.K. James:** Statistical Analysis, Writing – review & editing. **S. Spiegel:** Writing – review & editing. **A.F. Cibils:** Writing – review & editing, Conceptualization. **A.L. Gonzalez:** Conceptualization, Data Collection. **M.M. McIntosh:** Data Collection, Writing – review & editing. **K. Romig:** Data Collection.

#### Declaration of competing interest

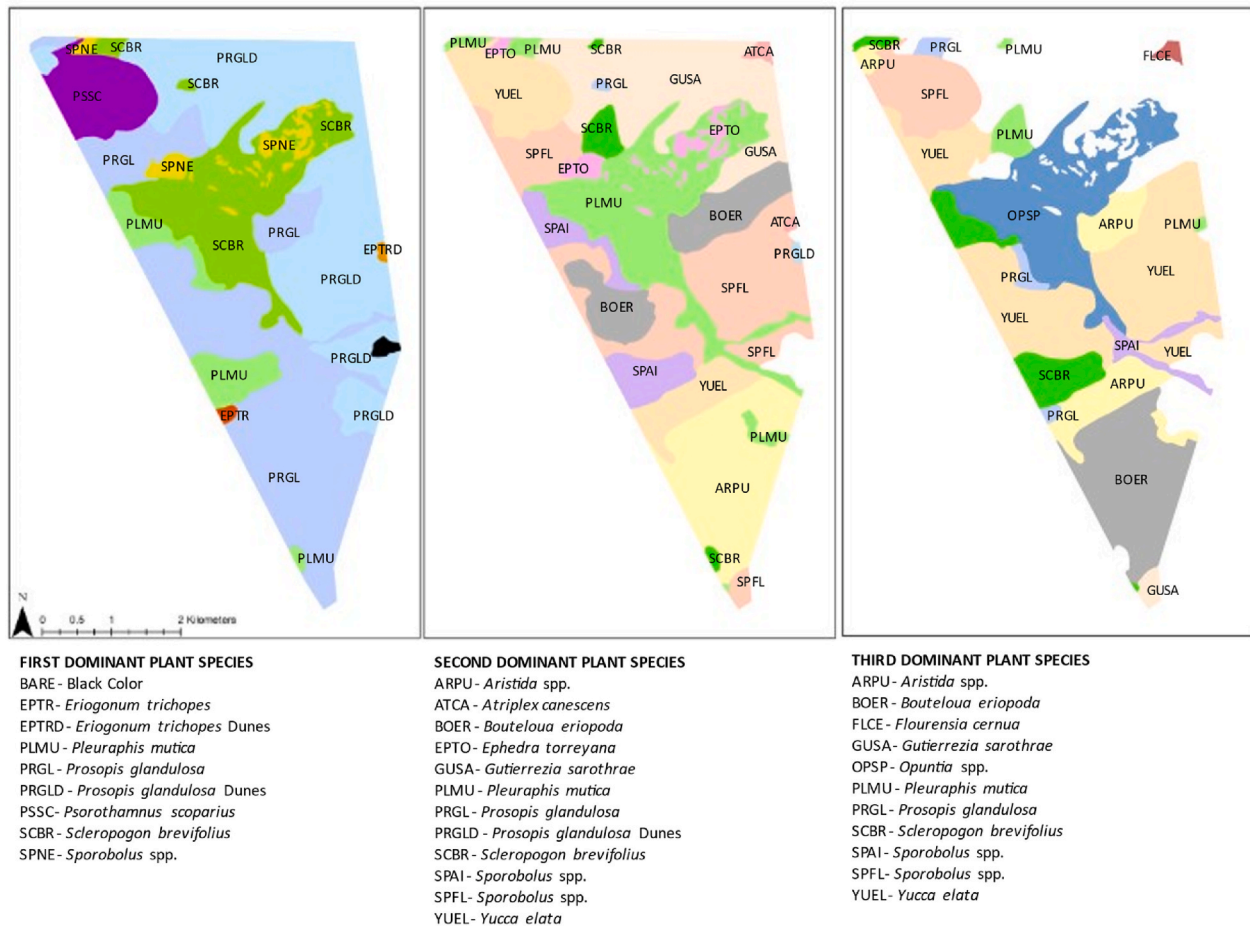
The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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#### Appendix





Appendix Fig. 1. Vegetation map showing first, second, and third most dominant plant species in study pastures.

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