

## Changes in the plant composition of an *Amelichloa clandestina* (Hack.) grassland after non-selective grazing

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### Abstract

This study evaluated the effect of intensive non-selective grazing on the richness, diversity, and structure of a grassland dominated by *Amelichloa clandestina* (Hack.) Arriaga & Barkworth at Rancho “Los Ángeles”, Saltillo, Coahuila. Experimental plots of 600 m<sup>2</sup> were established in a 2.23 ha area, with three replicates per season. Two treatments were applied: grazed and ungrazed (control) plots, separated by electric fencing. These plots were subjected to intensive grazing 333 livestock units (LU ha<sup>-1</sup>) at the beginning of each season, beginning in the winter of 2023. In the fall of 2024, the cover of all species in five quadrats (1 m<sup>2</sup>) per plot was quantified. Richness (Margalef), diversity (Shannon-Wiener), dominance (Simpson), evenness, and floristic similarity (Sørensen-Dice) indices were calculated. Canonical correspondence analysis (CCA) was used to evaluate the relationship between moisture and compaction. ANOVA and Tukey's test were performed. Thirty-seven species grouped into 19 families were recorded, with the greatest species richness in winter (30 species; Margalef index of 4.2). Spring and summer showed the most significant similarity (80%). The species *A. clandestina*, *Ambrosia confertiflora*, and *Clematis drummondii* were associated with higher humidity in summer and autumn. In conclusion, intensive non-selective grazing, carried out in different seasons, enhanced species richness and diversity, particularly during winter, spring, and summer.

**Keywords:** ACC, *Amelichloa clandestina*, grassland, grazing, richness and diversity, stocking rate

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## Introduction

The species diversity and aboveground biomass of plant communities in grasslands are key indicators of the functioning and stability of these ecosystems, as the variety of species in plant communities directly influences the sustainability and maintenance of ecosystem services (Pennekamp *et al.*, 2018). In this sense, grazing is one of the primary practices in grasslands, impacting production, structure, species diversity, and ecosystem stability (Liu *et al.*, 2015). Furthermore, the diversity and composition of grasslands vary depending on nutrient availability and the intensity, frequency, and seasonality of grazing (Mavromihalis *et al.*, 2013). Therefore, continuous grazing can reduce perennial plant cover, increase bare soil area, and facilitate opportunistic species invasion (Reeve *et al.*, 2000). Therefore, studying changes in species diversity and biomass under different grazing regimes is essential for grassland restoration and sustainable management (Morris, 2021). Since the intensity, duration, and seasonality of grazing can substantially modify plant composition, it not only alters the physicochemical properties of the soil but also indirectly influences microbial communities (Li *et al.*, 2022). These changes are reflected in dynamics of grasslands. Positively, manipulating the duration and season of grazing can increase the abundance of native perennial species (Kahn *et al.*, 2010). A disturbance caused by grazing can increase the specific diversity of plant communities and contribute to the relative stability of these ecosystems and sustainability (Zhang *et al.*, 2020a). Likewise, the seasonal exclusion of livestock during specific periods is an essential tool for grassland management (Nie and Zollinger, 2012). Rest periods from grazing for weeks or months can promote the growth of perennial grasses (Chapman *et al.*, 2003); however, the timing of such exclusion is crucial, given that spring represents the main growth and flowering period for many grassland species. Therefore, removing grazing during this period promotes persistence and stimulates the establishment of new species, such as various native forbs and grasses (Leonard and Kirkpatrick, 2004). However, overgrazing and poor management lead to rangeland degradation, with arid and semi-arid areas being more susceptible, jeopardizing their integrity and ecosystem services (Medeiros *et al.*, 2020).

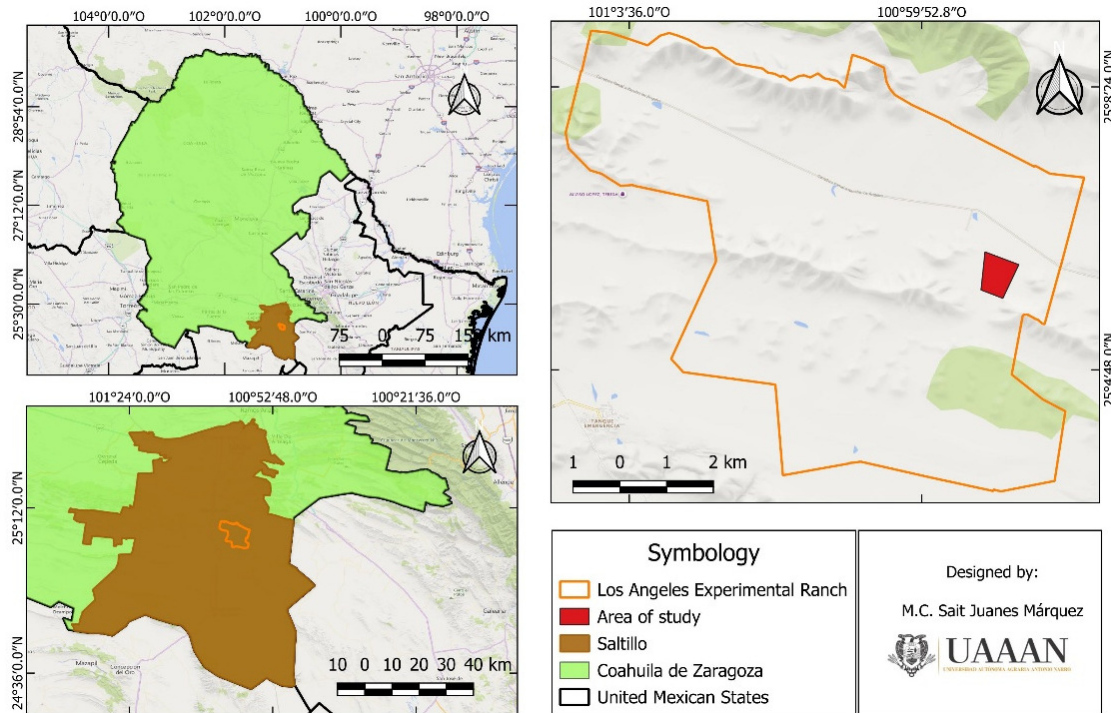
In the context of northeastern Mexico, following the abandonment of farmland, the clandestine pinegrass (*Amelichloa clandestina*) has been observed to colonize the herbaceous stratum aggressively, thanks to its high production of cleistogamous and chasmogamous seeds (Juanes-Márquez *et al.*, 2023). This species thrives in disturbed areas, scrublands, grasslands, and open pinyon pine forests, at altitudes ranging from 2,000 to 2,100 m. It is distributed in Coahuila and Nuevo León and has been introduced into West Texas (Barkworth, 1982; Villaseñor, 2016). Its presence was first documented in Kimble County in the early 1950s, spreading into the San Saba River Valley in the 1960s (Russell and Landers, 2017). This species demonstrates high adaptation, growth, and rapid spread in the region's grasslands. Therefore, a deeper understanding of the impacts of grazing throughout the different seasons is needed to design management strategies that consider the essential phenological changes of the species present. Therefore, the objective of this study was to evaluate the seasonal effect of intensive non-selective grazing on the richness, diversity, and structure of a grassland dominated by *Amelichloa clandestina*.

## Materials and Methods

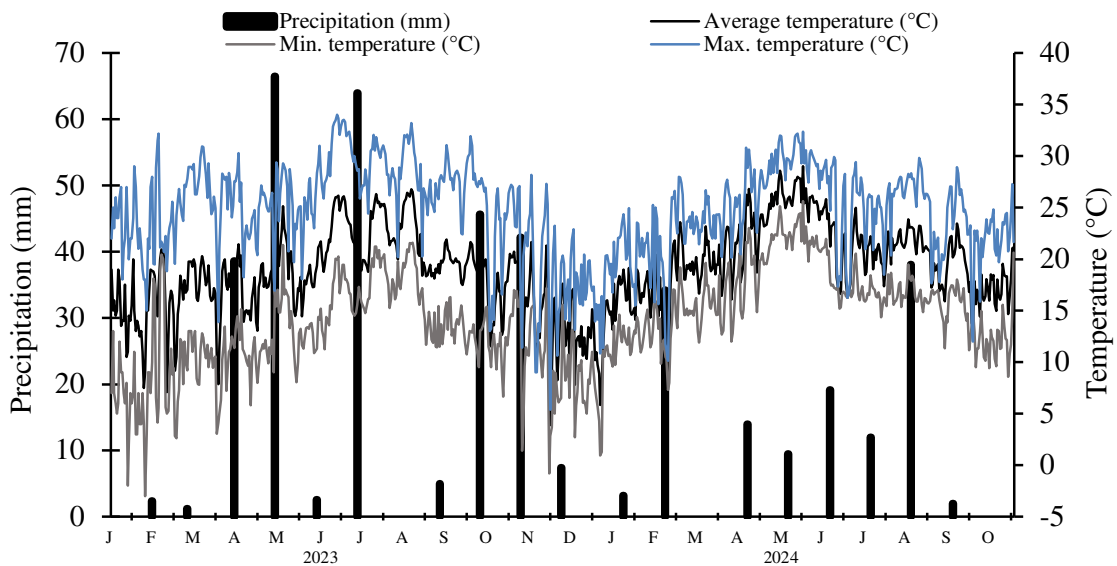
### *Study area location*

The study was conducted in a grassland dominated by *A. clandestina* in northeastern Mexico, located at the Experimental Ranch “Los Ángeles”. The area is located at coordinates 25°06'30" N and 100°59'18" W (Figure 1), at an altitude of 2,162 m. According to the Köppen classification, modified by García (2004), the climate is dry, characterized a precipitation-temperature ratio of 22.9 mm/16 °C, warm summers, and an

average temperature of 7 to 14 °C. The average annual rainfall is 350 mm, divided into May to October (87%) and November to April (13%) (Figure 2).



**Figure 1.** Geographic location of the study area at Experimental Ranch “Los Ángeles”, Saltillo, Coahuila, Mexico. Prepared by the authors (Sait Juanes Márquez)



**Figure 2.** Average daily maximum, average, minimum temperature, and accumulated precipitation from January 2023 to October 2024, climate conditions during the study period

### *Methodology*

The experiment was conducted in a 2.23 ha area divided into experimental units of 600 m<sup>2</sup> (10 x 60 m). Two treatments were applied: grazed and ungrazed (control) plots, separated by electric fencing. The study was carried out from January 2023 to October 2024 under a seasonal rotation scheme (spring, summer, autumn, and winter). At the beginning of each season, intensive non-selective grazing was applied (Acocks, 1966) with a stocking rate of 333 livestock units (LU ha<sup>-1</sup>), adjusted according to forage availability (Aranda *et al.*, 2023). Measurements were taken in October 2024, after 22 months of treatment, reflecting the cumulative effects of the four seasons.

### *Variables evaluated*

During the rainy season, five 1 m<sup>2</sup> quadrats (1 x 1 m) were established in each experimental unit, where the cover of the present species was measured, and the species present, cover, and height were identified (Arévalo *et al.*, 2021). The recorded species were organized at the family, genus, and species levels according to the taxonomic nomenclature reported on the Missouri Botanical Garden's tropicos.org website (2024) to obtain botanical composition. Direct species subsequently estimated species richness counts in each census. From this, the Margalef richness index, Simpson index, Shannon-Wiener diversity index, and Evenness index were derived (Moreno, 2001). Likewise, their migratory and successional status was consulted on the CONABIO Mexican weeds website (2024). To assess floristic similarities between grazing seasons, the presence and absence of species were recorded in the four seasons. These data were used to construct a dendrogram.

### *Data analysis*

The study was conducted under a randomized block design, with three replicates per treatment. Treatments consisted of the grazing practices carried out at the beginning of each season, as well as control area. The indices were determined using PAST software version 4.10 (Toto *et al.*, 2023). The Sørensen-Dice similarity index was used to determine floristic similarity using MVSP statistical software version 3.22. A dendrogram was generated using the UPGMA (Unweighted Pair Group Method with Arithmetic Mean) clustering method. An analysis of variance (ANOVA) was performed to evaluate the effect of grazing in each season. The analysis was conducted using JMP 14 PRO statistical software, and means were compared using the Tukey test, with a significance level of  $\alpha = 0.05$ . In addition to analysing vegetation distribution based on soil variables and evaluating changes in species composition, data were collected on the species with the highest cover values in the sample plots. These data were processed using multivariate ordination techniques, specifically Canonical Correspondence Analysis (CCA), with CANOCO version 5 software for Windows (Ter-Braak and Šmilauer, 2002). The environmental variables considered included soil ecological parameters such as moisture and compaction at 2.5, 5, and 7.5 cm depths.

## **Results**

### *Plant composition*

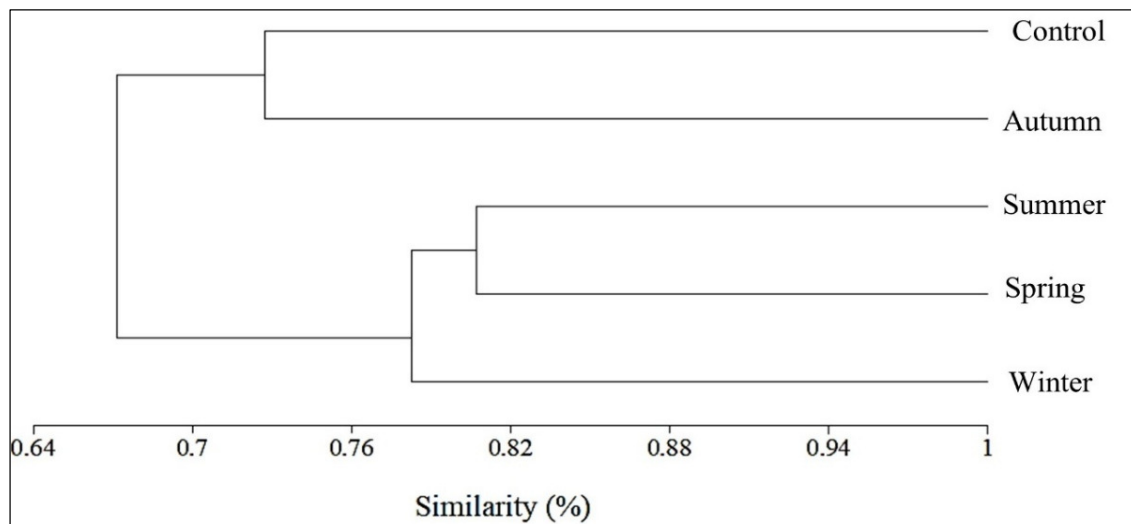
Table 1 presents the species richness of the prominent families and genera. The families with the greatest species richness are Asteraceae, with 10 species (27% of the total flora), Poaceae with nine species (24%), Fabaceae with three species (8%), and Lamiaceae, also with three species (8%). The most species-rich genera are Bouteloua, Euphorbia, and Panicum, each with two species (5% of the total flora). Herbs are the life form with the most incredible richness, with 28 species (76%), while Cactaceae are represented by one species (3%).

In the study area, across all seasonal grazing sites, a total of 37 species grouped into 19 botanical families were recorded (Table 3).

**Table 1.** Most representative families and genera of the grassland flora are dominated by Mexican needlegrass (*Amelichloa clandestina*) in the study site in northeastern Mexico

Family	Genera (species)	Genera	Species
Asteraceae	10 (10)	Bouteloua	2
Poaceae	7 (9)	Euphorbia	2
Fabaceae	3 (3)	Panicum	2
Lamiaceae	3 (3)	Amaranthus	1
Malvaceae	3 (3)	Ambrosia	1
Convolvulaceae	2 (2)	Amelichloa	1
Solanaceae	2 (2)	Anoda	1
Verbenaceae	2 (2)	Argemone	1
Chenopodiaceae	1 (1)	Asphodelus	1
Amaranthaceae	1 (1)	Buddleja	1
Asphodelaceae	1 (1)	Chamaesaracha	1
Brassicaceae	1 (1)	Chenopodium	1
Cactaceae	1 (1)	Cirsium	1

Figure 3 identifies two main groups based on the treatments applied to the *Amelichloa clandestina* grassland. The Sorensen similarity index showed significant differences between species across different seasons ( $p < 0.05$ ). Intensive non-selective grazing notably impacted the species composition and the grouping of treatments in the *Amelichloa clandestina* grassland in other seasons. The first group includes control treatment and those corresponding to the autumn season. The second group comprises treatments during spring, summer, and winter. The low similarity in species composition separated these groups, differentiating them from the rest of the studied areas.

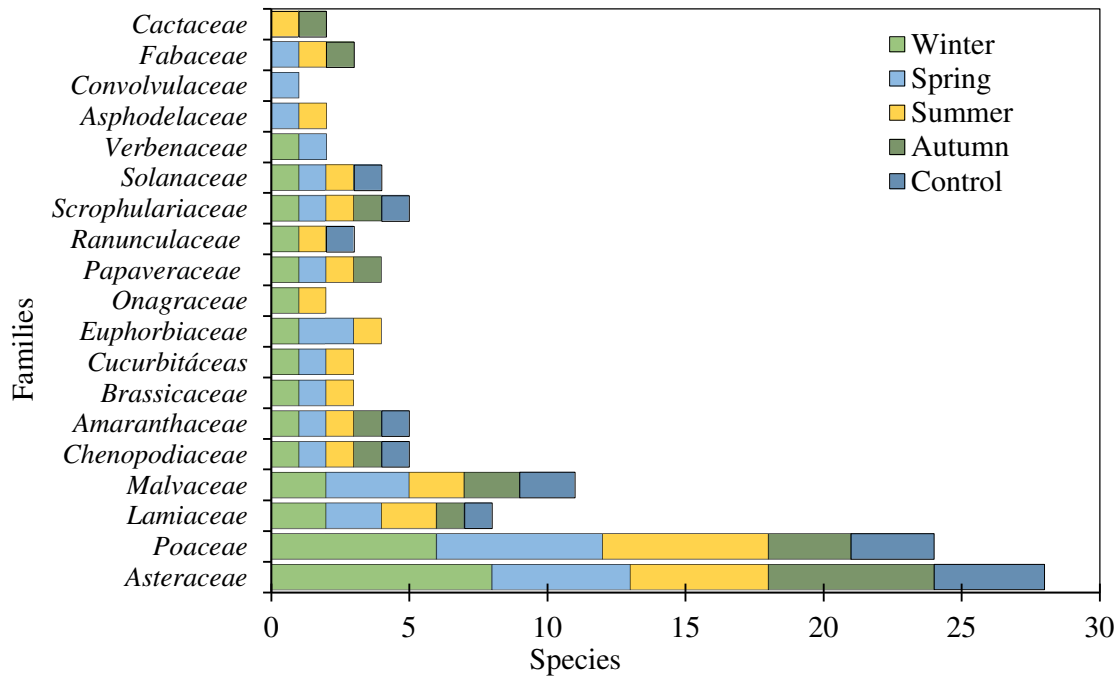


**Figure 3.** Floristic similarity coefficient (Sorensen-Dice; UPGMA grouping method), between four seasons after intensive non-selective cattle grazing in an *Amelichloa clandestina* grassland in northeastern Mexico

A similarity analysis identified several groups among the samples: General Group (67% similarity), including the four seasons and the control. Witness-Autumn Subgroup (72%): Formed by the witness and

autumn within the general group. Winter-spring-summer subgroup (78%): Composed of winter, spring, and summer. The highest Similarity Group (80%) comprises spring and summer, presenting the highest similarity. This grouping indicates that the spring and summer seasons share more similar characteristics.

The diversity analysis revealed that the Asteraceae family was the most dominant in the plots, recording eight species during winter. This family was followed by the Poaceae family, which identified six species in winter, spring, and summer. The Malvaceae family showed its most excellent richness in spring, with three species recorded. The other families were represented by a single species in the floristic composition of each season (Figure 4).



**Figure 4.** Number of species per botanical family after cattle grazing with a heavy stocking rate at the beginning of each season on an *Amelichloa clandestina* grassland in northeastern Mexico

#### *Species diversity*

Grazing had a significant effect on species diversity ( $p < 0.05$ ). Winter, spring, and summer exhibited greater species richness with 30, 29, and 28 species, respectively, compared to autumn, which had 15 species (Table 2; Table 3). Likewise, according to the Margalef wealth index, winter, spring, and summer presented the highest indices with values of 4.2, 4.3, and 4.0, respectively, followed by autumn with 2.6. Considering the Shannon diversity index ( $H'$ ), summer presented the highest index with 2.18, followed by winter and autumn with 2.02 and 1.90, respectively, while the lowest indices were observed in spring (1.88).

However, with the Simpson diversity ( $1-D$ ) and evenness ( $Evenness e^{H/S}$ ) indices, no significant differences were found ( $p > 0.05$ ), with average values of 0.80 and 0.32 for these indices. However, winter, spring, and summer grazing favoured greater plant species diversity and evenness than autumn and control.

**Table 2.** Richness and diversity indices after grazing with a high stocking rate at the beginning of each season in an *Amelichloa clandestina* grassland in northeastern Mexico

Season	Indices	Standard error	P-value
	Specific richness Taxa S		
Winter	30 a	1.63	<.0001*
Spring	29 a		
Summer	28 a		
Autumn	18 b		
Control	15 c		
	Individual		
Winter	785 a	33.9	<.0001*
Spring	623 d		
Summer	689 b		
Autumn	681 c		
Control	404 e		
	Margalef richness index		
Winter	4.2 a	0.233	<.0001*
Spring	4.3 a		
Summer	4.1 a		
Autumn	2.6 b		
Control	2.3 c		
	Shannon H diversity index		
Winter	2.02 ab	0.037	0.0075*
Spring	1.88 b		
Summer	2.18 a		
Autumn	1.90 ab		
Control	1.80 b		
	Simpson 1-D diversity index		
Winter	0.80 a	0.022	0.977
Spring	0.78 a		
Summer	0.82 a		
Autumn	0.82 a		
Control	0.79 a		
	e^H/S Evenness Index		
Winter	0.26 a	0.029	0.1468
Spring	0.22 a		
Summer	0.31 a		
Autumn	0.39 a		
Control	0.42 a		

Within column, means with different letters differ ( $\alpha = 0.05$ )

**Table 3.** List and existence of species by season after intensive non-selective grazing in an *Amelichloa clandestina* grass in northeastern Mexico

Species	Seasons					Family	Life cycle
	Wi	Sp	Su	Au	C		
<i>Amaranthus blitoides</i> S. Watson	✓	✓	✓	✓	✓	Amaranthaceae	Annual
<i>Ambrosia confertiflora</i> D.C	✓			✓		Asteraceae	Perenne
<i>Amelichloa clandestina</i> (Hack.) Arriaga & Barkworth	✓	✓	✓	✓	✓	Poaceae	Perenne
<i>Anoda cristata</i> (L.) Schlecht.	✓	✓	✓	✓	✓	Malvaceae	Annual
<i>Argemone echinata</i> G.B. Ownbey	✓	✓	✓	✓		Papaveraceae	Annual
<i>Asphodelus fistulosus</i> L.	-	✓	✓	-	-	Asphodelaceae	Perenne
<i>Bouteloua dactyloides</i> (Nutt.) Columbus	✓	✓		-	-	Poaceae	Perenne
<i>Buddleja scordioides</i> Kunth	✓	✓	✓	✓	✓	Scrophulariaceae	Perenne
<i>Chenopodium album</i> L.	✓	✓	✓	✓	✓	Chenopodiaceae	Perenne
<i>Clematis drummondii</i> Torr. & A. Gray	✓		✓	-	✓	Ranunculaceae	Perenne
<i>Convolvulus equitans</i> Benth.	-	✓		-	-	Convolvulaceae	Perenne
<i>Cucurbita foetidissima</i> Kunth	✓	✓	✓	-	-	Cucurbitaceae	Perenne
<i>Disakisperma dubium</i> (Kunth) P.M. Peterson & N. Snow	✓	✓	✓	✓	✓	Poaceae	Perenne
<i>Dyssodia papposa</i> (Vent.) Hitchc.	✓	✓	✓	✓	✓	Asteraceae	Annual
<i>Eragrostis mexicana</i> (Hornem.) Link	✓	✓	✓	-	✓	Poaceae	Annual
<i>Eruca vesicaria</i> (L.) Cav	✓	✓	✓	-	-	Brassicaceae	Annual
<i>Euphorbia exstipulata</i> Engelm.	✓	✓		-	-	Euphorbiaceae	Annual
<i>Euphorbia serrula</i> Engelm.		✓	✓	-	-	Euphorbiaceae	Annual
<i>Gaura coccinea</i> Pursh	✓		✓	-	-	Onagraceae	Perenne
<i>Glandularia bipinnatifida</i> Nutt	✓	✓		-	-	Verbenaceae	Annual
<i>Grindelia oxylepis</i> Greene	✓	✓	✓	✓		Asteraceae	Perenne
<i>Helianthus laciniatus</i> A. Gris	✓	✓	✓	✓	✓	Asteraceae	Perenne
<i>Hoffmannseggia glauca</i> (Ortega) Eifert	-	✓	✓	✓	-	Fabaceae	Perenne
<i>Hopia obtusa</i> (Kunth) Zuloaga & Morrone	✓		✓	-	-	Poaceae	Perenne
<i>Marrubium vulgare</i> L.	✓	✓	✓	-	-	Lamiaceae	Perenne
<i>Muhlenbergia torreyi</i> (Kunth) Hitchc. ex-Bush	-		✓	-	-	Poaceae	Perenne
<i>Opuntia rastrera</i> F.A.C. Weber	-		✓	✓	-	Cactaceae	Perenne
<i>Panicum ballii</i> Vasey	✓	✓	✓	✓	-	Poaceae	Perenne
<i>Parthenium incanum</i> Kunth	✓	✓	✓	✓	✓	Asteraceae	Perenne
<i>Salvia reflexa</i> Hornem	✓	✓	✓	✓	✓	Lamiaceae	Annual
<i>Sanvitalia angustifolia</i> Engelm. ex A. Gray)	✓	✓	✓	✓	✓	Asteraceae	Annual
<i>Setaria leucopila</i> (Scribn. & Merr.) K. Schum.	-	✓	-	-	-	Poaceae	Perenne
<i>Sida abutifolia</i> Mill.	✓	✓	-	-	-	Malvaceae	Perenne
<i>Solanum elaeagnifolium</i> Cav.	✓	✓	✓	-	✓	Solanaceae	Perenne
<i>Solidago velutina</i> DC.	✓					Asteraceae	Perenne
<i>Sphaeralcea angustifolia</i> (Cav.) G. Don	-	✓	✓	✓	✓	Malvaceae	Perenne
<i>Thelesperma simplicifolium</i> (A. Gray) A. Gray	✓	-	-	-	-	Asteraceae	Perenne

Wi=Winter, Sp=Spring, Su=Summer, Au= Autumn, C=Control

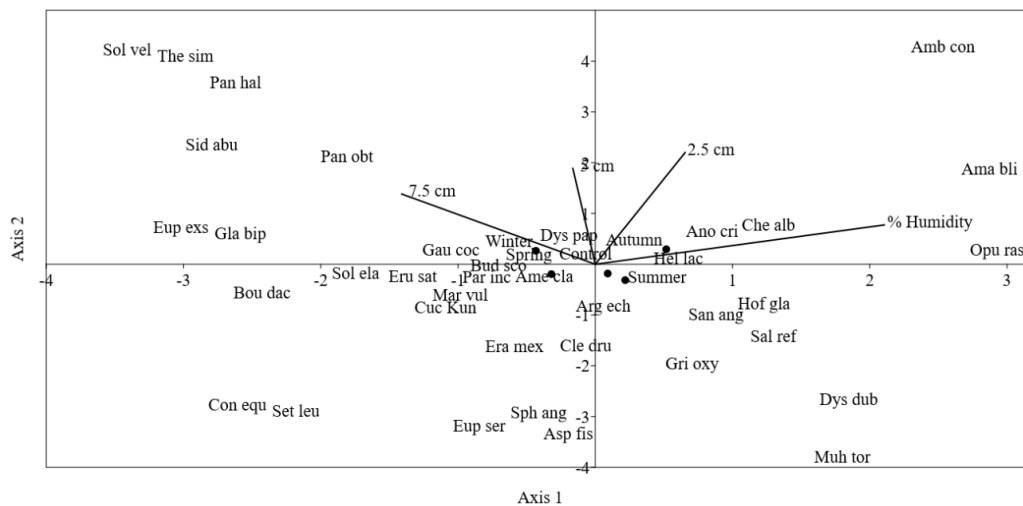
The environmental variables in the model explained 80% of this variation. This response suggests that environmental variables are significantly associated with species distribution. CCA (Table 4) generated four axes representing environmental gradients. The first axis (eigenvalue: 0.137) explained 39% of the modelled variation, while the second axis (eigenvalue: 0.0853) raised the cumulative explained variation to 63.2%. Together, these axes explained more than 60% of the relationship between species and the environment. Axes 3 and 4 contributed in a smaller proportion to the total variance. The canonical correlations between the axes and the environmental variables are high (1.0 and 0.96 for axes 1 and 2, respectively), indicating a strong association. These results support the interpretation of environmental gradients as key determinants of species distribution.

**Table 4.** Canonical correspondence analysis

Statistic	Axis 1	Axis 2	Axis 3	Axis 4
Proper values	0.137	0.085	0.057	0.071
Explanatory variation (cumulative)	39.0	63.2	79.6	100
Pseudocanonical correlation	1.0	0.96	0.98	000
Adjusted variation explained (cumulative)	48.96	79.43	100	-

Permutation Test Results: pseudo-F=1.3, P=0.274

Figure 5 shows species distribution along two principal axes and environmental variables such as humidity, soil compaction at different depths, and seasons. *Muhlenbergia torreyi*, *Opuntia rastrera*, *Ambrosia confertiflora*, *Clematis drummondii*, *Helianthus laciniatus*, and *Amelichloa clandestina* were grouped towards the axis of highest humidity, mainly in summer and autumn. Species such as *Hopia obtusa*, *Gaura coccinea*, *Solidago velutina*, *Thelesperma simplicifolium*, and *Panicum hallii* were associated with soil compaction (2.5, 5, and 7.5 cm), associated with winter. On the other hand, during spring, *Setaria leucopila*, *Bouteloua dactyloides*, *Euphorbia serrula*, *Euphorbia exstipulata*, *Glandularia bipinnatifida*, and *Sida abutifolia* stood out. The control showed the lowest presence of species, including *Hoffmannseggia glauca*, *Asphodelus fistulosus*, and *Sphaeralcea angustifolia*; therefore, it can be recognized that the distribution of the species is influenced by seasonal factors such as temperature, precipitation, and edaphic conditions.



**Figure 5.** Canonical Correspondence Analysis of the species arrangement after cattle grazing at the beginning of the season in an *Amelichloa clandestina* grassland, concerning humidity soil compaction at different depths (2.5, 5, and 7.5 cm)

The arrows indicate the direction of each variable's most significant contribution, while the species' location reflects their association with these conditions.

## Discussion

The recorded flora (Table 3) represents 1.2% of the flora reported for Coahuila (Villarreal-Quintanilla, 2001). According to Rzedowski (1992), grasslands and shrublands host approximately 6,000 species, equivalent to 20% of Mexico's total flora; of these, 0.61% are found in the *Amelichloa clandestina* grassland, indicating low species richness. The species richness observed in the grassland is lower than that reported by Juanes-Márquez *et al.* (2024), who recorded 74 species in a study conducted in the same area. Arévalo *et al.*, (2021) reported 70 species

Previous studies in Argentine grasslands have shown that 10-year grazing exclusion changes floristic composition, diversity, and vegetation structure (Nai-Bregaglio *et al.*, 2002). These findings demonstrate that the grazing season significantly influences the similarity and diversity coefficient of the plant community. In this study, two main groups were formed: (1) control and autumn, and (2) spring, summer, and winter. However, the similarity of autumn with the control in the first group could be related to more recent grazing, indicating a shorter time between this event and the evaluation. This suggests that grazing intensity and timing are key factors influencing plant community structure, independently or interacting with seasonal characteristics (Amezaga *et al.*, 2004). The trend observed in the Sorensen similarity index showed a decrease in the number of shared species between the grazed plant community and the control (ungrazed area). This pattern reflects the influence of grazing on vegetation structure and diversity. Therefore, when managing grasslands, the potential effects of grazing on plant communities and their ecosystem functions must be considered, so that more effective management and grazing strategies can be designed (Chen *et al.*, 2024). In arid grasslands, characterised by shallow soils, steep slopes, and extreme droughts, species composition is more stable (Meier *et al.*, 2021). This stability, linked to water and nutrient scarcity, limits ecological succession, maintaining a relatively constant composition (Meier and Partzsch, 2018).

In contrast, with greater moisture availability, semi-arid grasslands favour nutrient uptake and more extensive vegetation cover (Ellenberg and Leuschner, 2010). This promotes successional processes, including establishing new species and gradually transforming the plant community. The floristic composition in semi-arid environments such as the *Amelichloa clandestina* grassland is more dynamic. These patterns are associated with seasonal conditions, grazing, and climate interaction (Petřík *et al.*, 2011).

In this regard, Rolnik and Olas (2021) mention that the Asteraceae family is known for dominating spaces thanks to its reproductive strategy, which confers competitive advantages. Similarly, the Poaceae family plays an essential role in grazing ecosystems, where their rapid regeneration after grazing contributes to their persistence (Proulx and Mazumder, 1998). The dominance of Asteraceae and Poaceae in specific seasonal periods (Proulx and Mazumder, 1998) can be explained by the impact of grazing on the floristic composition, which increases species diversity in nutrient-rich ecosystems, while in poor ecosystems, it can limit species regeneration.

According to the study by Osem *et al.* (2002), species richness can increase under grazing conditions due to the increased availability of soil resources and the simultaneous reduction in competition resulting from eliminating larger, more palatable species. Conversely, in the absence of grazing, species richness decreases due to increased competition for light and the displacement of smaller, less competitive species (Huston, 1994). Meanwhile, Borer *et al.* (2014) demonstrate that the presence of grazing can limit excessive shading by controlling the growth of dominant species, thereby favouring the coexistence of smaller plants and reducing competition for canopy light. Therefore, the absence of herbivores increases competition for light and nutrients, which can lead to a decrease species richness. Similarly, the growth and composition of grassland plant communities have been reported to be strongly influenced by grazing intensity (Zhang *et al.*, 2020b). These responses can also be explained by combining other factors, such as herbivore selectivity, species palatability, and vegetation diversity (Msadek *et al.*, 2022). According to Bullock and Pakeman (1997), controlled intensive grazing increases the total number of plant species, especially favouring the presence of

herbs and grasses, while shrubs tend to proliferate in areas with lower grazing intensity. López *et al.* (2017) reported a positive effect of horse grazing on diversity in northern Spain, favouring the presence of several rare species of high conservation value (*C. filipendulum*, *G. pneumonanthe*, *S. tinctoria*, and *S. humilis*), which increased in abundance and frequency with grazing.

Furthermore, plant species richness and diversity in Galicia increased in the *Genista scorpius*-dominated understory under grazing (Rigueiro-Rodríguez *et al.*, 2012). Likewise, grazing generated significant changes in the plant community when intensive non-selective grazing was carried out, increasing the number of species (Table 3). This effect was evident in all seasons, possibly due to greater nutrient availability caused by a greater number of animal excreta due to a high grazing load (Zanella *et al.*, 2021). During grazing, livestock feeding and trampling cause changes in the quantitative characteristics of grassland plant communities (Zhang *et al.*, 2020b). Increased soil nutrient content can alter the competitive balance and facilitate the presence of specific species (Klinerová and Dostál, 2020).

Furthermore, livestock manure represents a significant vector of plant dispersal regarding the quantity of seeds and the number of species transported (Zanella *et al.*, 2021). Species diversity is a fundamental indicator of grassland community structure and is closely related to ecosystem functioning (Bleicher *et al.*, 2020). Thus, grazing controls grassland plant communities' structural characteristics and species diversity (Milchunas and Lauenroth, 1993). Several studies indicate that species diversity can increase under an adequate grazing system (Hobbs and Huenneke, 1992), and several indicators of community structure also reach maximum values under these conditions (Milchunas *et al.*, 1988).

These results suggest that the species community is primarily organised around one or two dominant environmental gradients, reflected in the first axes of the CCA. This type of pattern is common in ecological studies, where key environmental factors (water, temperature, nutrients, or seasonality) determine species composition (Lolila *et al.*, 2023). For example, in a study of tree communities in Europe, 36% of the variation in biomass fractions was attributed to species genetic diversity, while 64% was due to environmental variability (Veresoglou and Peñuelas, 2019). The association of species such as *Muhlenbergia torreyi* and *Amelichloa clandestina* with the wettest seasons (summer and autumn) is consistent with research highlighting that water availability is critical for species growth (Xu *et al.*, 2015). As observed in winter-associated species, soil compaction and depth significantly influence species composition. Compaction increases mechanical strength, limits root penetration, and water availability, favouring stress-tolerant species (Rosenberg, 1964). The effects of compaction at different depths (2.5, 5, and 7.5 cm) indicate that soil physical conditions dictate water retention and nutrient flux, affecting species adaptation, especially during dry periods (Zhu and Lin, 2011). However, certain grasses thrive in compacted soils due to stronger root adaptations (Vanderburg *et al.*, 2020). Species in the Poaceae family may have roots capable of penetrating dense layers and accessing water retained in deeper strata (Nippert and Knapp, 2007). Thus, the distribution of Poaceae in the diagram suggests that they find a competitive advantage under conditions of winter compaction where moisture can be maintained at greater depths. The control species show the lowest species diversity; research in poorly managed grasslands indicates that the absence of moderate disturbances (controlled grazing or prescribed burning) reduces biodiversity and favours the dominance of a few species (Chaturvedi *et al.*, 2018). Environmental factors, such as variability in soil moisture and compaction, are crucial in species richness and community structure, as observed in arid and semi-arid ecosystems (Munhoz *et al.*, 2008).

## Conclusions

Seasonal grazing in the *A. clandestina* grassland altered its structure, increasing species richness and diversity, particularly in winter, spring, and summer. This was associated with increased species richness and a reduction in the aerial cover and height of *A. clandestina*, which facilitated the establishment of other species.

Seasonal grazing management impacts grassland structure. A close relationship exists between the environmental variables (humidity and compaction) and species composition, indicating that the selected factors reflect important ecological patterns. Seasonal grazing with rest periods is recommended to reduce *A. clandestina* dominance, increase diversity, and ensure grassland sustainability through soil monitoring

### Authors' Contributions

Conceptualization: SJ-M, PA-V; Data curation: SJ-M; Funding acquisition: PA-V; Investigation: PA-V, MM-B, MC-Z, SJ-M; Methodology: PA-V, SJ-M, JAE-D; Project administration: PA-V; Supervision: PA-V, MM-B, AP-S; Writing - original draft: SJ-M, PA-V; Review and editing: PA-V, SJ-M.

All authors read and approved the final manuscript.

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### Conflict of Interests

The authors declare there are no conflicts of interest related to this article.

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