



# Exploring dynamics of floristic composition in Mediterranean grasslands: a case study from Sardinia, Italy

Giovanna Piga<sup>1</sup>, Maria Carmela Caria<sup>2</sup>, Nour Al Hajj<sup>1</sup>, Marco Malavasi<sup>2</sup>, Giovanni Riviuccio<sup>2</sup>, Simonetta Bagella<sup>2</sup>

<sup>1</sup> Department of Agricultural Sciences, University of Sassari, Via De Nicola, 07100, I-Sassari, Italy

<sup>2</sup> Department of Chemical, Physical, Mathematical and Natural Sciences, University of Sassari, Via Vienna, 07100, I-Sassari, Italy

Corresponding author: Maria Carmela Caria ([mccaria@uniss.it](mailto:mccaria@uniss.it))

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## Abstract

Mediterranean grasslands stand out as biodiversity hotspots due to their inclusion of diverse habitats that foster a remarkable range of biological diversity, characterized by inter-annual variability of floristic composition. This study aims to assess the inter-annual dynamics of the floristic composition in a grassland within a typical silvopastoral system in the Mediterranean basin, within the framework of rainfall variability. The study was performed on a farm representative of Mediterranean silvopastoral systems in Central-Western Sardinia (Italy). Vegetation surveys started in the spring of 2018 and concluded in the spring of 2022. They were conducted annually in the spring (S) and winter (W) seasons, using the point quadrat method along eight permanent transects. The dynamics of floristic composition were compared within and between years in terms of variability of specific percentage contribution (CSP) of each plant species. The results highlighted substantial differences in the composition of plant assemblages between the two seasons (W vs S) and between surveys within each season for all *a posteriori* comparisons. *Anthemis arvensis*, *Lolium rigidum*, *Festuca ligustica* and *Medicago polymorpha* were the main discriminating plant species between seasons. Considering the discriminant species between the four winter surveys, we found a positive effect of August rain on *Cynodon dactylon* CSP and a negative one on *Trifolium subterraneum* CSP. Considering the spring surveys, we found a positive effect of January rain on *L. rigidum* and a negative effect on *Plantago lanceolata*. The rain affected the inter-annual dynamics of floristic composition in the Mediterranean grasslands. Other factors, such as temperature, remain to be investigated. Moreover, a more extended data series may allow us to strengthen our results.

## Keywords

Climate change, false breaks, inter-annual variability, Mediterranean grasslands, seed bank, vegetation dynamics

## Introduction

Mediterranean grasslands stand out as biodiversity hotspots due to their inclusion of diverse habitats that foster a remarkable range of biological diversity, including many plant and animal species (Gigante et al. 2018; Giallonardo et al. 2019). They are secondary formations and holds cultural significance owing to their rich and ancient human history (Catorci et al. 2021). They provide several ecosystem services, including nutrient cycling, carbon sequestration, and water cycle regulation (Ribeiro et al. 2014; Seddaiu et al. 2018; Grenke et al. 2022) and agricultural goods (Unger and Jøgen 2014). However,

they are facing significant challenges, including intensive grazing management, marginal grassland abandonment (Klimek et al. 2007), and climate change (Dibari et al. 2021). These factors have led to declining surface area and grassland quality, presenting a severe conservation problem (Klimek et al. 2007).

The floristic composition and vegetation structure in grasslands result from complex interactions involving various biological and environmental factors (Puerto and Rico 1992). Livestock grazing is a crucial factor that affects vegetation structure in grasslands. The intensity and timing of grazing can significantly impact plant diversity and abundance (Ribeiro et al. 2014).

Inter-annual variability of floristic composition is an intrinsic feature of Mediterranean grasslands. Understanding this variability is crucial for the sustainable management of pastures and ensuring they provide nutritious resources for livestock effectively. The most prominent climatic factors shaping grasslands are the presence of intense summer droughts, unpredictable autumn rains and significant intra- and inter-annual fluctuations in precipitation (Fernández-Moya et al. 2011), particularly during the spring and summer months (Catorci et al. 2021). These conditions result in substantial differences in productivity, species composition, and overall diversity of these ecosystems from one year to the next (Carmona et al. 2015). The timing and variability of rainfall within a year influence the growth and performance of plant species (Suttle et al. 2007). Annual species, which evolved specific adaptations to survive under such conditions, dominate these communities. These species spend the summer months in seed form, effectively avoiding the harsh conditions of high temperatures and water scarcity during this period. Autumn is a critical period for the renewal of plant life in these ecosystems: the first rains initiate germination and regrowth of many species.

Some models suggest that regions experiencing a significant increase in temperature accompanied by a simultaneous decrease in precipitation are particularly vulnerable to biodiversity loss. In such areas, there is a likelihood of an increase in less palatable drought-resistant species (Tardella et al. 2016), and drought is considered the main factor controlling forage quality and palatability (Dibari et al. 2021). Additionally, research has demonstrated that grasslands with a high level of biodiversity are less susceptible to the adverse effects of drought events, showing a more rapid recovery following periods of drought. Indeed, biodiversity plays a crucial role in enhancing the resilience of plant communities to extreme weather events and contributes to the overall stability of ecosystems (Tardella et al. 2016).

Climate conditions in the Mediterranean basin have been experiencing a trend toward increased aridity in recent decades (Kafle and Bruins 2009). This shift towards drier conditions is expected to persist, with projections indicating a potential 20% decrease in total annual precipitation by 2050. Furthermore, inter-annual variation in rainfall will increase, leading to years of exceptionally wet conditions alternating with years of drought in the Mediterranean region. These fluctuations in precipitation can pose significant challenges for various sectors, including agriculture, water supply, and the stability of ecosystems. As a result, it is becoming increasingly crucial to develop adaptation strategies and comprehensive plans to address the impacts of these changing climatic conditions (Golodets et al. 2013).

This study aims to assess the inter-annual dynamics of the floristic composition in a grassland within a typical silvopastoral system in the Mediterranean basin within the framework of rainfall variability.

Surveys were carried out over five years twice each year: the first in winter, following the annual vegetative growth before the winter vegetative dormancy period, and the second in spring, at the peak of the vegetation season. This seasonal sampling approach allows for comprehensively assessing inter and intra-annual variability in floristic composition, capturing vegetation dynamics at two critical points in the year and providing valuable data for understanding fluctuations in plant communities within the Mediterranean silvopastoral system.

## Methods

### Study area

The study site is located within a private farm in Central-western Sardinia, Italy (40°8'N, 8°35'E) with an elevation of 500 m above sea level (Fig. 1), characterized by calcifugal, meso-supramediterranean cork oak series, specifically *Viola dehnhardtii-Quercus suberis*  $\Sigma$  (Bacchetta et al. 2009). The underlying geological substrate consists of Plio-Pleistocene and Oligo-Miocene volcanic rocks. The landscape in this region is diverse and includes various land uses associated with agro-silvopastoral production activities.

This farm serves as a representative example of Mediterranean silvopastoral systems. Livestock grazing on the farm comprises two breeds: Charolais and Sardo-Modicani. The animals' diet combines direct grazing on available forage in permanent grasslands and hay produced



**Figure 1.** Study site location.

in the farm. Additionally, external feed supplements are used to meet the nutritional needs of the animals, with feeding strategies adapted to the seasonal availability of grazing herbage and the specific dietary requirements of the animals at different physiological stages.

The farmer employs a vertical transhumance system, which involves the seasonal movement of livestock from mountainous areas to valley regions. Grazing primarily occurs in winter and spring, following a continuous scheme with a stocking rate of 1.5 LSU (Livestock Unit) per hectare per year (Frongia 2021).

## Data collection

Vegetation surveys were conducted using the "point quadrat" method (Bullock 1996). The survey areas were determined by eight permanent transects measuring 50 m long, randomly located in 2018. Along each transect, points were marked at regular intervals of 1 m, and all plant species that came into contact with a needle at these points were recorded to assess the frequency occurrence of each species (Daget and Poissonet 1971). In addition, since the less frequent species are likely missed using this method, a complete list of plant species within a buffer zone of 1 meter to the right and 1 m to the left of the transect line was compiled (Verdinelli et al. 2022).

These transects' starting and ending points were recorded using a Garmin Montana 610 handheld GPS device, allowing for precise mapping of their positions.

Plant nomenclature and life form associated with each species follow <https://dryades.units.it/floritaly/>.

Vegetation surveys started in the spring of 2018 (survey 0) and concluded in the spring of 2022 (survey 8). The surveys were conducted annually in two seasons: spring (S) and winter (W) (Table 1).

Meteorological data for the years 2017–2022 (daily Tmax, Tmin, and rainfall) were provided by ARPAS (Agenzia regionale per la protezione dell'Ambiente della Sardegna) from the meteorological station of Macomer, located 20 km from the farm. The study period showed inter-annual rainfall fluctuations in total quantity and monthly distribution. Annual rain varied between 1221 mm (2018) and 752 mm (2020), and the rainiest month was November 2021 (239 mm). The average annual Tmax ranged between 19.8°C (2018) and 20.6°C (2021); the warmest month was August 2012 (32.5°C). The average yearly Tmin went between 10.4°C (2017) and 11.0°C (2018), and the coldest month was February 2018 (3.1°C).

## Data analysis

The dynamics of floristic composition were evaluated within and between years in terms of variability of specific percentage contribution (CSP) of each plant species.

**Table 1.** Year and season of each survey.

Season	Year	Survey
Spring	2018	0
Winter	2018	1
Spring	2019	2
Winter	2019	3
Spring	2020	4
Winter	2020	5
Spring	2021	6
Winter	2021	7
Spring	2022	8

The frequency of occurrence of each plant species ( $FS_i$ ) recorded along each transect was converted into  $CSP_i$  using the formula:

$$CSP_i = \frac{FS_i}{\sum FS_i} 100$$

All species exclusively recorded within the buffer area were assigned a CSP of 0.3 (Pittarello et al. 2019). The CSP of each species for each transect  $\times$  season was used to build a species/cover matrix for data analysis.

To calculate the dissimilarity between pairs of samples, the Bray–Curtis dissimilarity measure was applied to square-root transformed data, resulting in a distance matrix. Non-parametric multidimensional scaling (nMDS) was employed as the ordination method to visualize differences in the composition of plant assemblages (Clarke and Gorley 2006). The floristic composition of each sample was compared among season (W vs S) and years of surveys (T) using permutational multivariate analysis of variance (PERMANOVA) (Anderson 2001), using a nested design, with T nested in W or S. Taxa responsible for differences among samples in different surveys were identified by similarity percentages for species contributions analysis (SIMPER) (Clarke 1993). Species that contributed at least 5% dissimilarity for any comparisons were considered significant discriminators.

The multivariate statistical analyses were carried out using the PRIMER software package version 7, with the PERMANOVA add-on developed by Anderson et al. (2008). Finally, the trajectory between surveys were overlaid on the nMDS plot to represent the intra-annual distances between plant assemblages in the two seasons (De Cáceres et al. 2019).

To evaluate the possible effects of rain on the CSP of the taxa responsible for the inter-annual differences in the composition of plant assemblages in winter, we considered the rainfall before the winter surveys, i.e., August to November, as single months or cumulated. To evaluate the possible effects of rain on the CSP of the taxa responsible for the inter-annual differences in the composition of plant assemblages in spring, we considered the rainfall before the spring surveys, i.e., February to May, as a single month or cumulated. A linear model was adopted to describe the relationships between CSP values of the taxa responsible for the inter-annual differences in the composition of plant assemblages and rainfall as a covariate.

## Results

### Dynamics of floristic composition

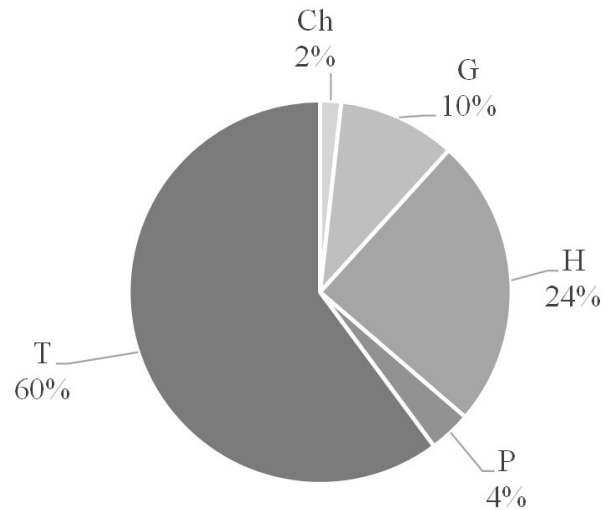
In total, 179 plant species were identified, with 166 present in spring and 102 in winter. The most abundant species belonged to the Fabaceae family (36%), followed by Poaceae (32%) and Asteraceae (11%). In terms of biological forms, Therophytes were strongly dominant, followed by Hemicryptophytes (Fig. 2).

The results obtained from nMDS analysis (Fig. 3) highlighted substantial differences in the composition of plant assemblages between the two seasons (W vs S) and between surveys (0-8) within each season for all *a posteriori* comparisons, as confirmed by the PERMANOVA analysis (Pperm=0.001).

As identified by SIMPER, the main discriminating plant species between seasons were four (Fig. 4). The most abundant species in winter was *Anthemis arvensis*, and in spring *Lolium rigidum*. The analysis highlighted the marked seasonality, showing higher values of frequency in spring of *Festuca ligustica* and *Medicago polymorpha* among the other discriminating species.

Poaceae and Fabaceae were more abundant in spring, while Asteraceae cover was similar in the two seasons (Fig. 5).

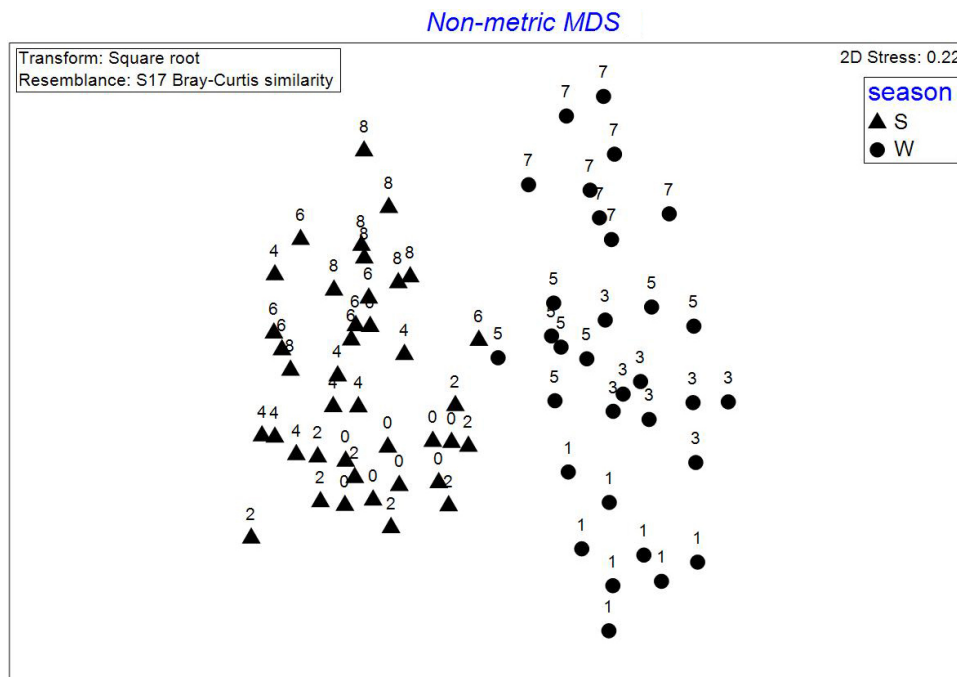
Comparing the four winter surveys, the main discriminating species, as identified by SIMPER, were five (Fig. 6). Among them *A. arvensis*, *Cynodon dactylon* and *T. subterraneum* showed a high CSP variability year by year while *Bromus hordeaceus* and *L. rigidum* were more stable.



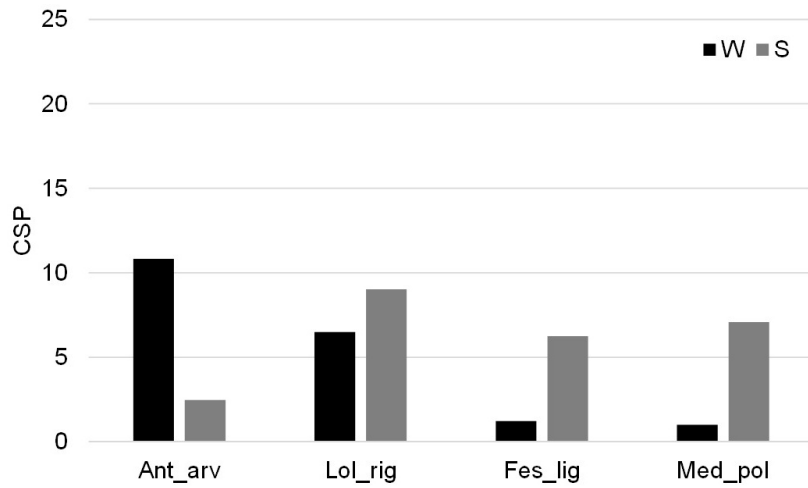
**Figure 2.** Biological spectrum (Ch=Chamaephytes; G=Geophytes; H=Hemicryptophytes; P=Phanerophytes; T=Therophytes).

Comparing the five spring surveys, the main discriminating species identified by SIMPER, were *Avena barbata*, *Plantago lanceolata*, *F. ligustica*, *L. rigidum*, *M. polymorpha* and *T. subterraneum* (Fig. 7). Some of them, such as *M. polymorpha* and *P. lanceolata*, showed a very high CSP variability year by year.

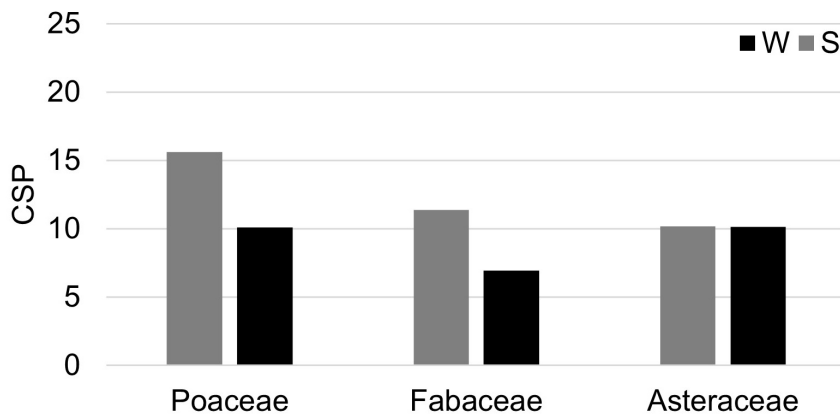
The trajectories analysis showed a more linear trend and greater distances between elevations in the winter season than in the spring season (Fig. 8).



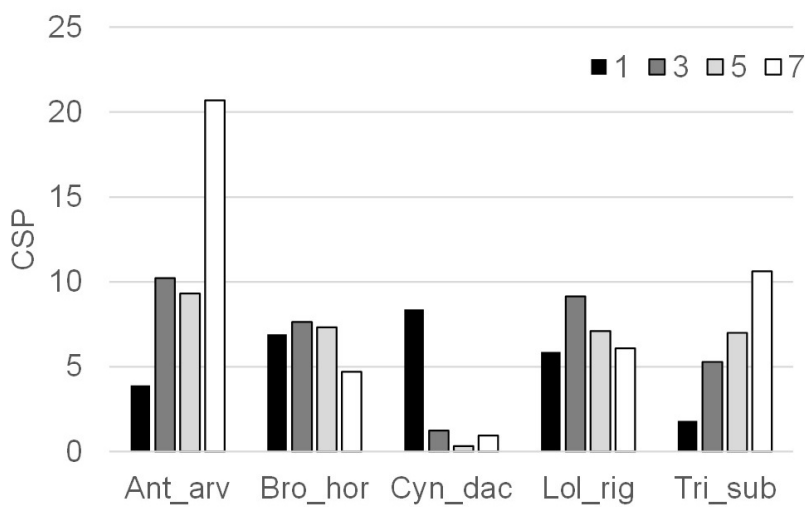
**Figure 3.** Two-dimensional nonmetric multidimensional scaling ordination (sMDS) of plant assemblage composition in the two seasons (S=spring; W=winter) and surveys (0-8).



**Figure 4.** Average abundance (CSP) of the four species contributing at least 5% to the dissimilarity the comparisons between plant assemblages in spring (S) and winter (W) according to SIMPER (Ant\_arv=*Anthemis arvensis*, Lol\_rig=*Lolium rigidum*, Fes\_lig=*Festuca ligustica*, Med\_pol=*Medicago polymorpha*).



**Figure 5.** Average abundance (CSP) of the three dominant families in spring (S) and winter (W).



**Figure 6.** Abundance (CSP) of the four species contributing at least 5% to the dissimilarity of the comparisons between plant assemblages in winter surveys (1, 3, 5, 7) according to SIMPER (Ant\_arv=*Anthemis arvensis*, Bro\_hor=*Bromus hordeaceus*, Cyn\_dac=*Cynodon dactylon*, Lol\_rig=*Lolium rigidum*, Tri\_sub=*Trifolium subterraneum*).



## Variations in rainfall and their effects

The rainfall pattern showed significant variability between years in all months of the year (Fig. 9).

$R^2$  between CSP of the taxa responsible for the inter-annual differences in the composition of plant assemblages in winter and the rainfall in the previous months were significant only for two species. We found a positive effect of August rainfall on *C. dactylon* ( $R^2=0.848$ ;  $y=-0.0695+9.072$ ) and a negative effect on *T. subterraneum* ( $R^2=0.9185$ ;  $y=-0.0749-0.3887$ ).

$R^2$  between CSP of the taxa responsible for the inter-annual differences in the composition of plant assemblages in spring and the rainfall in the previous months were significant only for two species. We found a positive effect of January rainfall on *L. rigidum* ( $R^2=0.706$ ;  $y=-0.0209+7.1764$ ) and a negative effect of May rainfall on *P. lanceolata* ( $R^2=0.713$ ;  $y=-0.0429+1.7018$ ).

## Discussion

The pastures in the study area exhibited typical Mediterranean pasture characteristics. They were indeed very species-rich pastures, as already observed in similar areas (Tárrega et al. 2009; Farris et al. 2010; Bagella et al. 2013), which is related to the traditional land use (Bagella et al. 2016). The dominance of Therophytes is another aspect typical of Mediterranean grasslands (Pitt and Heady 1978; Díaz-Villa et al. 2003; Tárrega et al. 2009; Fernández-Moya et al. 2011) because this life form is particularly well suited to disturbance tolerance (Grime 1977) and soil disturbance by trampling favoured seedlings recruitment (Noy-Meir et al. 1989). Fabaceae and Poaceae played a fundamental role in these grasslands. Fabaceae, including some excellent forage species such as *T. subterraneum*

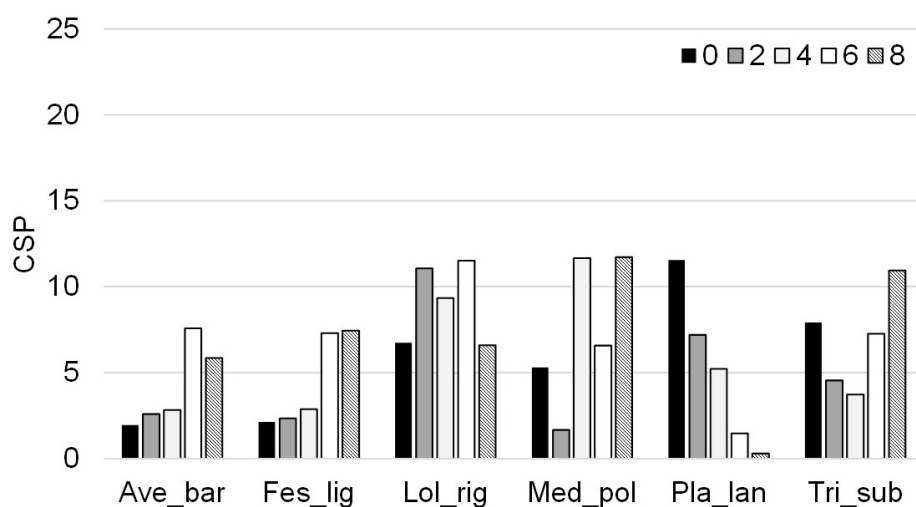
and *M. polymorpha*, ensure the supply of proteins to the grazing animal during the entire grazing season, improve soil fertility with natural N fixation (Lucas et al. 2010) and support C cycling and the productivity and persistence of these secondary grasslands (Bagella et al. 2020). Poaceae support the forage production during the coldest months, the excellent forage species *L. rigidum* with the contribution of *A. barbata* and *F. ligustica*. Moreover, they contribute to limiting soil erosion (Bagella et al. 2020).

As expected in the two seasons under comparison, the plant assemblage composition was different with a higher variability in winter than in spring.

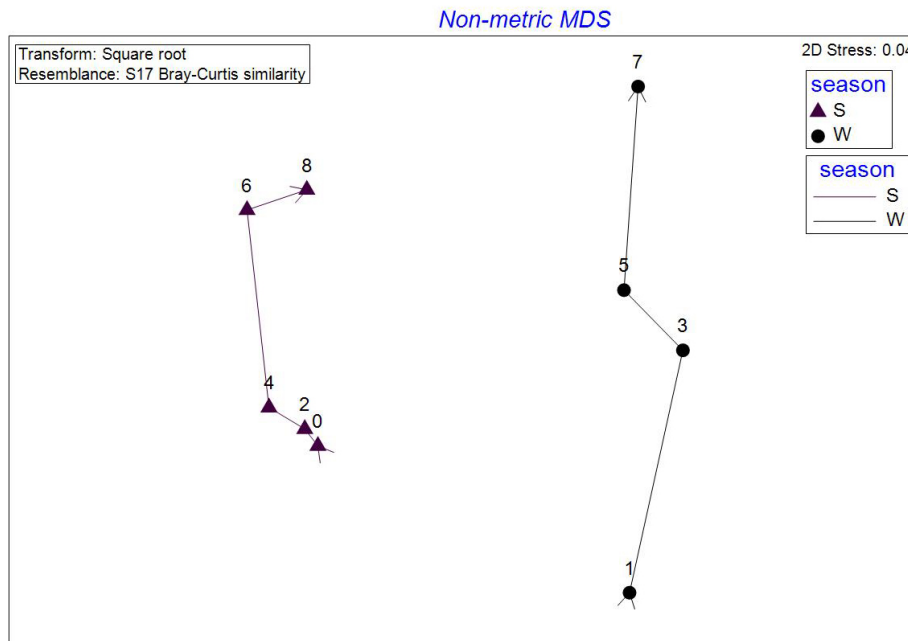
The spring vegetative cover was generally characterized by higher floral diversity and a more significant presence of forage grasses and legumes compared to winter. The bulk of primary production and biodiversity occurs in this season (Moreno and Pulido 2009), extending through late spring and early summer, during which plants undergo seed formation and eventual senescence. Specifically, during the spring season, *L. rigidum*, *M. polymorpha*, and *F. ligustica*, with an average coverage percentage close to 10% for *L. rigidum* and slightly lower for *M. polymorpha* and *F. ligustica*, represent a significant portion of the vegetation cover. *L. rigidum* and *M. polymorpha* are considered excellent and good forage plants, respectively, and *F. ligustica* is considered fair (Bagella et al. 2013). Therefore, the presence and substantial coverage of these species should constitute the primary support for pasture quality during the spring season.

On the other hand, the winter vegetation cover was dominated by *A. arvensis*, which is not consumed by grazing animals (Bagella et al. 2013). However, even in this season, though in limited quantities, other plant species are available.

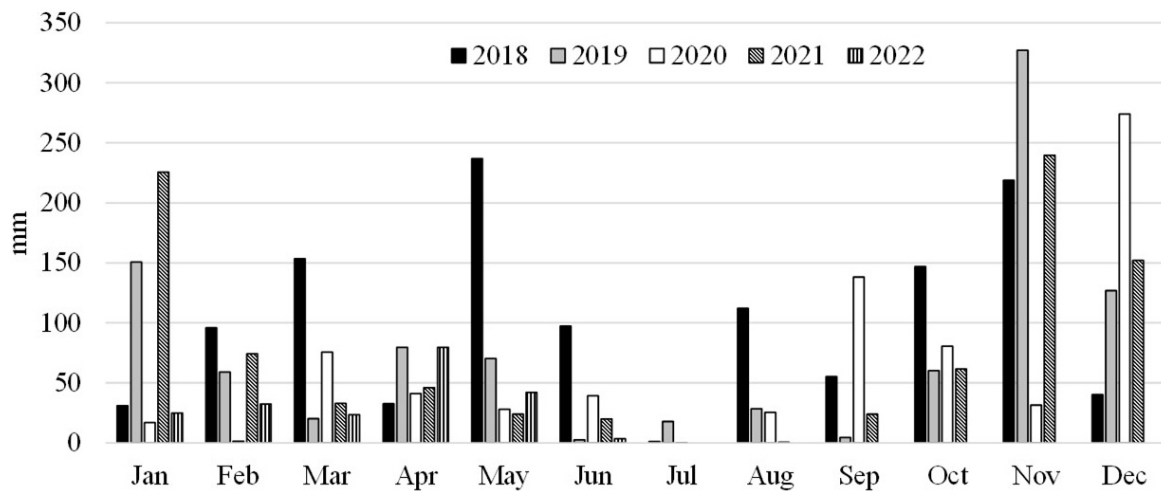
Plants grow in response to the initial autumn rain events (Pitt and Heady 1978). In the case of annual spe-



**Figure 7.** Abundance (CSP) of the six species contributing at least 5% to the dissimilarity of the comparisons between plant assemblages in spring surveys (0, 2, 4, 6, 8) according to SIMPER (Ave\_bar=*Avena barbata*, Fes\_lig=*Festuca ligustica*, Lol\_rig=*Lolium rigidum*, Med\_pol=*Medicago polymorpha*, Pla\_lan=*Plantago lanceolata*, Tri\_sub=*Trifolium subterraneum*).



**Figure 8.** Trajectories analysis relative to each season (S=spring; W=winter). Each point represents the average value of the three replicates.



**Figure 9.** Monthly rainfall amount (mm) during the survey period.

cies, the rainfall that induces seed germination is termed the break of season. If this event occurs too soon, it can induce germination of pasture species that do not survive and are termed false breaks (Chapman and Asseng 2001; Turner et al. 2001). In our study, we observed an effect of the August rainfall on the development of two species that most characterize the floristic composition during the winter season, which is more related to the rainfall in this month than in the following months. However, the two species on which these effects are most evident, *C. dactylon* and *T. subterraneum*, show an opposite response, positive in the former case and negative in the latter, which should be related to their different adaptation strategies.

*C. dactylon* is a perennial stolonifera C4 plant which grows mainly in summer under maximal light availability and high temperatures. The C4 strategy in the Mediterranean grassland species is unusual, typically dominated by annual C3 grasses and legumes that are active during the wet period of the year (Aires et al. 2008). Using this adaptation *C. dactylon* avoids competition for light, water and nutrients with other annual species whose seeds germinate later. The observed positive effect of August rainfall on the CSP of this species in the winter could be explained by the efficient use of water by a species with photosynthetic efficiency at high temperatures and radiation intensities (Galiano 1985). *T. subterraneum* is an annual ear-

ly-season species that starts growing slowly in autumn or winter if the weather is mild, grows rapidly in the spring and sets seed by the end of the spring (Papanastasis 1981). It was instead negatively affected by break season rainfall (Turner et al. 2001). The negative effect could probably be related to a false break due to a combination of a germination-inducing rainfall event followed by a period of drought determining a widespread death of seedlings, as already observed in Australian grasslands (Chappman and Asseng 2001). In the study area, competition for water with *C. dactylon*, which has higher evapotranspiration enhancing soil water depletion, exacerbates the situation (Aires et al. 2008).

Concerning the floristic composition in spring, we found a direct effect of the January rainfall on the cover of two species: *P. lanceolata* and *L. rigidum*, which CSP was positively and negatively affected, respectively. *P. lanceolata* is a perennial species which produces rosettes and reproduces by seed production and clonal propagation (Cavers et al. 1980). It consistently improved under high temperatures and drought conditions (Rasmussen et al. 2020). *L. rigidum* is an annual species whose seeds germinate during autumn and winter (Steadman et al. 2003). Seed dormancy prevents false breaks before this time due to sporadic summer rains but allows germination when rainfall is sufficient to sustain plant growth and development (Steadman et al. 2004). This should be why abundant rains in January favoured seed germination and seedling growth. The less demanding *P. lanceolata* was favoured in the year with less rainy January. These observations confirm that in an environment where water is scarce, the species abundance is regulated by competition-driven water depletion (Tsialtas et al. 2001).

Rainfall is a crucial determinant of production and composition in arid and semi-arid systems (Dudney et al. 2017), and extreme events, such as extended autumn droughts, are forecasted to increase in Mediterranean regions under climate change scenarios (Nogueira et al. 2017). Such changes, particularly those responsible for false breaks, may also adversely affect the soil seed bank, which is strategic for conserving and regenerating Mediterranean grasslands.

In this paper, we explored the inter-annual dynamics of floristic composition in the Mediterranean grasslands, considering the rain as a key factor for this dynamic. Other factors, such as temperature, remain to be investigated. Moreover, a more extended data series may allow us to strengthen our results.

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