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Brief Report

Is Shrub Encroachment Driving the Decline of Small Mammal Diversity in Pyrenean Grasslands? A Preliminary Study

Ignasi Torre [1,2,*] and Oriol Palau [2]

1 BiBio Research Group, Natural Sciences Museum of Granollers, c/Francesc Macià 51, E-08402 Granollers, Spain
2 Small Mammal Research Area, Natural Sciences Museum of Granollers, c/Francesc Macià 51, E-08402 Granollers, Spain
* Correspondence: itorre@mcng.cat

Abstract: The Pyrenean highlands hold the southernmost populations of some endemic and rare arvicoline species associated with grasslands. This area, as well as many other areas in the Mediterranean basin, has suffered from land abandonment due to socio-economic changes during the last decades. Those changes represented a reduction of the traditional livestock grazing by goats and sheep which naturally controlled the process of shrub encroachment. Today, maintenance of open habitats such as pasture grasslands needs to be performed by mechanical actions aimed at removing shrubs and woody vegetation. We document a case study on a plot sampled for five consecutive years (2017–2021) in which clear-cutting increased the cover of herbaceous vegetation by removing shrubs, potentially improving pasture quality. Also, changes in the small mammal community were detected, such as an increase in the species density and diversity (Shannon index) after clear-cutting, with the occurrence of typical grassland species (*Microtus arvalis*, *M. lavernedii*). Nonetheless, the effects were short-term (lasting two years), and then the community returned to the pre-treatment situation. Prior to intervention, the community was dominated by two generalist and widespread species (*A. sylvaticus* and *C. russula*), and the situation was similar two years after the intervention. Small increases in shrub cover produced relevant community changes by decreasing diversity and increasing dominance of the two most common species. These results suggested that shrub encroachment produced the banalization of small mammal communities dominated by generalist species.

Keywords: shrews; Rodents; clear-cutting; pasture; species diversity

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1. Introduction

The Mediterranean basin is considered a remarkable "hotspot" of biological diversity owing to the long history of assimilation between natural ecosystems and human interests [1,2]. In particular, macroclimate and biogeographical history regarding peninsular dynamics were considered key determinants of species richness and endemicity in the European continent [3]. Indeed, the Iberian Peninsula (as the other Mediterranean peninsulas) is considered an area of mammal endemism [4], and some of the mountain massifs of the NE, such as the Pyrenees, holding an astonishing mammal diversity [5]. Biogeographical studies concerning small mammal diversity pointed out the relevance of the axial Pyrenees as a physiographical region with its own chorotype of species sharing mid-European requirements [6]. In this area, some small mammals of northern-colder climates reach their southern distribution limits because their ranges are positively associated with moisture and negatively associated with temperature [5]. In particular, the Pyrenean highlands hold marginal populations of some arvicoline species associated to grasslands, such as the Pyrenean vole (*Microtus gerbi* Gerbe, 1879), the common vole (*Microtus arvalis* Pallas, 1778), and the montane vole (*Arvicola monticola* de Sélys-Longchamps, 1838). Furthermore, other grassland species of mid-European requirements but with higher tolerance to warm climates can be found in the area, such as the endemic Mediterranean field vole (*Microtus lavernedii* Crespon, 1844). Most of them are burrowing semi-fossorial small mammals feeding on grasses and playing a relevant role in the maintenance of grasslands [7]. Nonetheless, their fossorial activity (digging galleries, creating earth mounds) directly conflicts with human interests in montane grasslands such as agriculture and livestock production [7]. Although some of these species are widespread in Europe and considered agricultural pests (e.g., common vole), their populations in the Pyrenees are reduced and without evidence of typical population outbreaks frequently occurring in other areas of the Iberian Peninsula [8]. The degree of endemicity of some species, altogether with the overall rarity and future potential retraction of their ranges [9], makes this group a conservation concern.

Because small mammal communities in montane grasslands are mostly composed of species with mid-European requirements, they can be expected to be severely affected by climate change, but also by landscape change (afforestation and shrub encroachment). The landscapes of the Pyrenees are subjected to the same process of change as observed in the whole Mediterranean basin [10]. This area suffered from land abandonment due to socio-economic changes during the last decades, reducing the traditional livestock grazing by goats/sheep, and increasing cows for beef, and horses [11]. Owing to food requirements by livestock [12], the decline of goats altogether with an increase of cows and horses contributed to shrub encroachment processes in grasslands. In this study, we presented preliminary results of the potential role of controlling shrub encroachment to improve pasture quality in the small mammal community of a montane grassland of the Eastern Pyrenees.

2. Materials and Methods

2.1. Study Area and Sampling Design

The study plot was situated on a subalpine meadow partially surrounded by forests (municipality of Vilallonga de Ter, Long: 2.269; Lat: 42.368; elevation 1554 m.a.s.l., Girona, NE Spain), with some terraces with stone walls, and stands of *Sarothamnus scoparius*, *Crataegus* sp., and *Corylus avellana*, mainly on the margins of the plot (Figure 1). Recently, some guidelines for the recovery of grasslands have been established in the Capçaleres del Ter i Freser Natural Park [13], and some ranchers followed recommendations to improve pasture quality. Clear-cutting was performed between autumn 2018 and summer 2019 with a manual brush cutter disc cutting at the ground level and the aerial biomass being removed [14,15]. Cattle grazing affects the study plot, with stocking densities from 0.3 cows/ha (90 cows/300 ha) in late spring to 1.1 cows/ha (90 cows/80 ha) in early winter.

The small mammal sampling followed the SEMICE monitoring scheme (www.semice.org, accessed on 1 September 2022), with a plot of 36 traps set on a grid (6 × 6, 15m distance between traps), alternating in position the same number of Longworth (Longworth Scientific Instrument Co., Oxford, UK) and Sherman traps (Sherman folding small animal trap; 23 cm × 7.5 cm × 9 cm; Sherman Co., USA), a combination that prevents size-specific biases in small mammal assessments in montane communities [16]. In the later years (2019–2021), Longworth traps were substituted by a very similar model, the Heslinga trap (http://www.heslingatraps.eu). Traps were provided with hydrophobic cotton for bedding [17] and baited with a piece of apple and a mixture of tuna and flour. We performed two sampling sessions—of three days each—each year, thus, totaling 10 sampling periods. We made the summer sessions between 29 June and 26 July, and the autumn sessions between 6 and 31 October. Traps were operated during three consecutive nights and revised during the early morning of the first, second, and third day. Rodents were marked with ear tags (Style 1005–1, National Band Co., Newport, KY, USA) and shrews were marked with fur clips, both were released at the point of capture [18]. Research on live animals followed ethical guidelines [19], the project was approved, and captures were performed under the special permission of the Catalan

Government (Generalitat de Catalunya). Small mammal nomenclature follows the Handbook of the Mammals of the World [20].

Figure 1. Map of the study plot, represented by the yellow square on the orthophoto map of the area.

Vegetation cover and height were assessed at the trap level by visual estimation (summers 2017, 2019, and 2021), considering the height of herbaceous, short and tall shrubs (in cm), and the cover of the same variables (in % area). These variables were estimated on a 5 m-radius centered on the trap following the SEMICE protocol (www.semice.org). Percentage cover was independent for the three vegetation profiles since the different strata can be superimposed. Distance to stone walls was assessed on a categorial basis (close: <1 m; near: 1–4 m, far: >4 m).

2.2. Data Analysis

First, we analyzed the effect of vegetation treatment on vegetation profiles at the trap level, estimating four variables around each trap, and considering the three sampling periods (see below). Second, we analyzed whether vegetation treatment influenced the number of individuals captured at traps by species and session, considering the same three main sampling periods regarding vegetation treatment. Associations between response and environmental variables were established using generalized linear mixed models (glmer function in R) assuming Gaussian distribution of errors for mean abundance and vegetation data [21,22]. The selection of predictors incorporated into the models was assessed by paired correlations according to the strength of associations (Spearman correlation: $\rho \geq 0.8$) and testing the variation inflation factor (VIF) of the models [22] and selecting sets of independent variables with VIF < 3. The final set of suitable variables to model the small mammal abundance and diversity responses included the uncorrelated variables of vegetation cover (herbaceous, short and tall shrubs), and discarded redundant variables related to the height of vegetation [23,24]. We also included two categorical predictors: treatment, with three levels (pre-treatment, from summer 2017 to autumn 2018; early post-treatment, from summer 2019 to autumn 2020; late post-treatment, from summer 2021 to autumn 2021) and season (two levels: summer and autumn). Distance to stone walls was also included as a predictor. We included trap as a random factor to reduce the effects of non-independence of spatial and temporal data [25]. Prior to analyses, vegetation variables were standardized ($\bar{x}$ = 0, SD = 1).

3. Results

During the study period, we captured 97 individual small mammals of five species (132 including recaptures): the wood mouse (*Apodemus sylvaticus*) was dominant (69%), followed by the greater white-toothed shrew (*Crocidura russula*, 18.5%), the common vole (*Microtus arvalis*, 9.3%), the pygmy shrew (*Sorex minutus*, 2%), and the Mediterranean field vole (*Microtus lavernedii*, 1%). Common small mammals showed seasonal oscillations, with higher abundance in autumn than in summer, but this changed over the years (Figure 2). Species density/richness increased from 2017 (two species) to 2019 (three species) and 2020 (four species) and then dropped in 2021 (two species). During the study, we had only four casualties (4%).

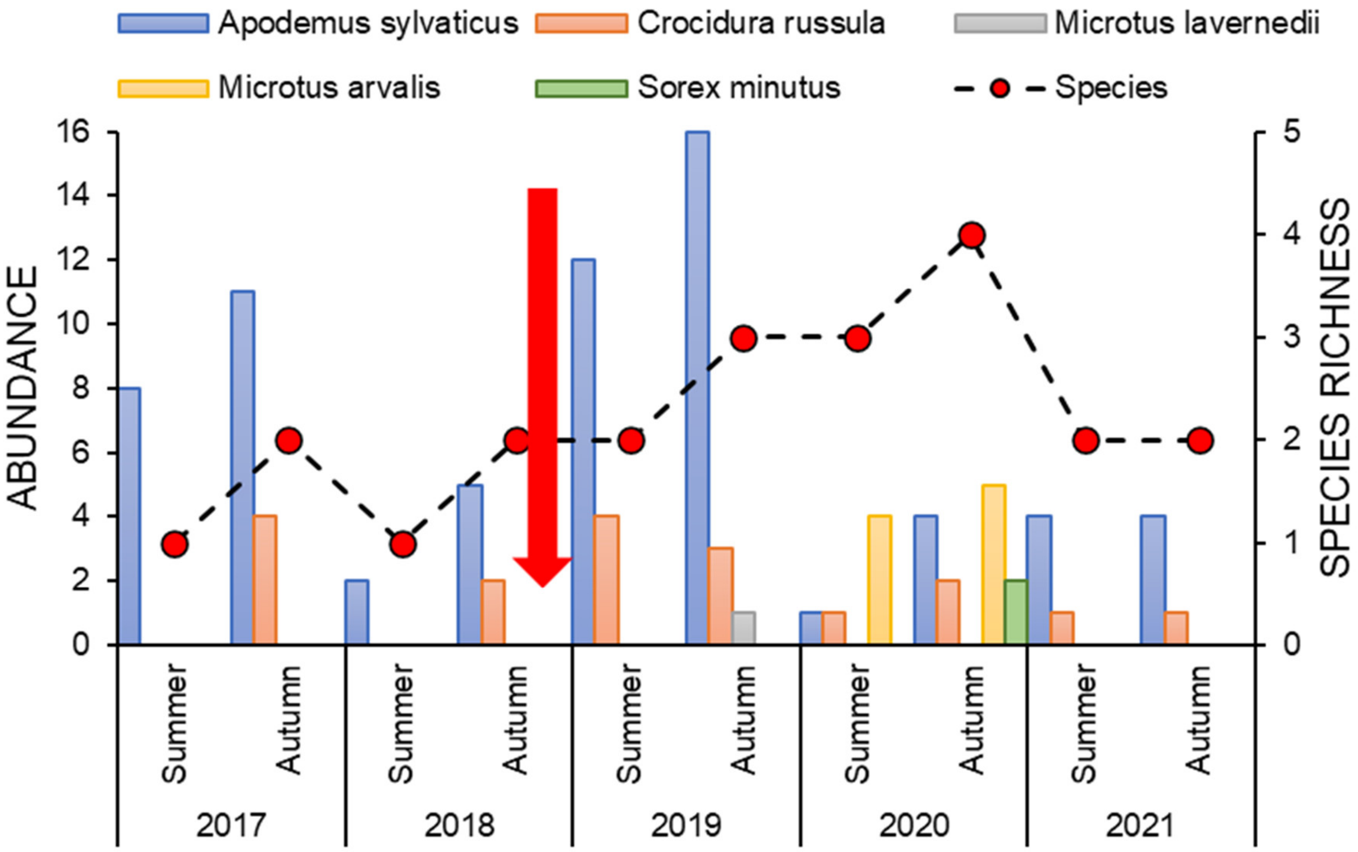

Figure 2. Number of small mammal individuals captured (left axis) and species density (right axis) along the study period (summer 2017–autumn 2021). The red arrow indicates the intervention to control for shrub encroachment.

Cover of herbaceous and short shrub vegetation were affected by treatment, while tall shrub cover was barely affected (Table 1). After treatment (Post-1, summer 2019), herbaceous cover increased (+16%) and short shrub cover decreased (−12%). Two years after treatment (Post-2, summer 2021), the cover of short shrubs increased to the values of the pre-treatment period (+15%), tall shrub cover slightly increased (+3%), but the herbaceous cover was higher than in the pre-treatment period (+17%) (Figure 3). Furthermore, effects of treatment affected the small mammal community, with the appearance of the field vole during the autumn of the first year after treatment, the common vole during the summer of the second year, and the pygmy shrew during the autumn of the second year (autumn 2020). The community returned to pre-treatment values during 2021, with only captures for the two more common species (wood mice and white-toothed shrews).

Table 1. Results of the GLMMs were performed with the three variables related to vegetation cover as a response, the treatment (three levels: Previous, Post-1, and Post-2) as a fixed factor, and the trap as a random factor. AICc values, and variance explained by fixed (marginal) and fixed + random (conditional) factors are also shown. Pre-treatment (previous) was the reference for statistical comparisons.

		Herbaceous Cover (%)		**Shrub Cover (<1.5 m, %)**		**Shrub Cover (>1.5 m, %)**	
		Estimate	SE	Estimate	SE	Estimate	SE
Treatment	(Intercept)	−0.67 ***	0.13	0.19	0.13	−0.12	0.14
	Post-1	0.97 ***	0.10	−0.71 ***	0.13	0.05	0.13
	Post-2	1.02 ***	0.10	0.14	0.13	0.31 *	0.13
	AICc	496.73		565.69		566.18	
	R^2 marginal	0.22		0.14		0.02	
	R^2 conditional	0.61		0.40		0.44	

Signif. codes: *** 0.001; ** 0.01; * 0.05.

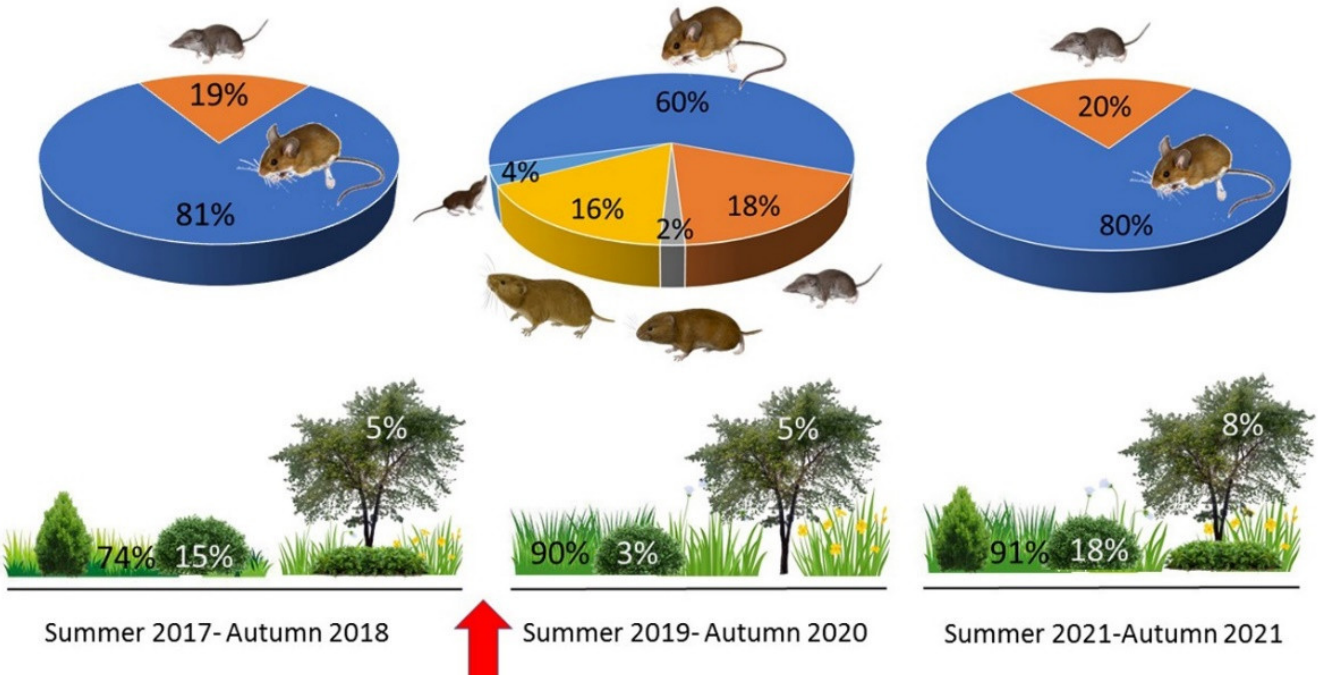

Figure 3. Graphical representation of changes that occurred in the sampling plot along the study period (summer 2017–autumn 2021), regarding the intervention to control for shrub encroachment (red arrow), and their effects on vegetation cover profiles (herbs, short and tall shrubs) and the small mammal community (frequencies of occurrence).

At the trap level, wood mice showed a negative response to herbaceous cover and a positive response to the cover of tall shrubs, showing marginal seasonality (more abundance in autumn). White-toothed shrews were positively affected by the three vegetation profiles, showing a negative effect on the distance to stone walls. Also, they showed marginal seasonality similar to wood mice (more abundance in autumn). The common vole was the only species showing a positive effect of the early post-treatment (Post-1). Species density (the mean number of species captured per trap) increased during the early post-treatment period, being similar during the pre-treatment and late post-treatment periods (Table 2).

Table 2. Results of the GLMMs performed with the three small mammal species, the total number of individuals, and species density/richness, as response variables, treatment and season as fixed factors, vegetation cover (herbaceous, short and tall shrubs) and distance to stone walls as covariates, and trap as a random factor. AICc values, and variance explained by fixed (marginal) and fixed + random (conditional) factors are shown. For the treatment factor, the pre-treatment (previous) was the reference for statistical comparisons.

		A. sylvaticus		***C. russula***		***M. arvalis***		**Total**		**Species**	
		Estimate	**SE**	**Estimate**	**SE**	**Estimate**	**SE**	**Estimate**	**SE**	**Estimate**	**SE**
	(Intercept)	0.95 ***	0.19	−0.05	0.08	−0.08	0.08	0.85 ***	0.22	0.43 **	0.15
Treatment	Post-1	0.11	0.08	0.02	0.04	0.07 *	0.03	0.23 *	0.09	0.15 *	0.06
	Post-2	−0.04	0.08	−0.05	0.04	−0.01	0.03	−0.10	0.09	−0.08	0.06
Vegetation	Herb	−0.08 **	0.00	0.00	0.00	0.00	0.00	−0.01	0.00	0.00	0.00
	Shrub (<1.5 m)	0.00	0.00	0.00 **	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	Shrub (>1.5 m)	0.00 ***	0.00	0.00 **	0.00	0.00	0.00	0.01 ***	0.00	0.01 ***	0.00
	Stone wall	−0.01	0.01	−0.01 *	0.00	0.01	0.00	−0.01	0.01	−0.01	0.01
Season	Summer	−0.10	0.05	−0.04	0.02	−0.02	0.02	−0.18 **	0.06	−0.10 *	0.04
	AICc	285.92		−37.15		−95.42		354.97		184.03	
	R2 marginal	0.22		0.08		0.07		0.21		0.22	
	R2 conditional	0.30		0.10		0.12		0.28		0.32	

Signif. codes: *** 0.001; ** 0.01; * 0.05.

Community diversity (Shannon index) reached its highest during 2020 (H′ = 1.09), with an increase in species richness altogether with a decrease in abundance of the two common species, thus increasing equitability. During the pre-treatment period community diversity showed the minimum (H′ = 0.29), and during the late post-treatment dropped to similar values (H′ = 0.5).

4. Discussion

Our preliminary results confirmed that clear-cutting increased the cover of herbaceous vegetation by simply removing the cover of competing shrubs, potentially improving pasture quality [15]. These results agree with other studies conducted in similar mountain grasslands, clear-cutting increasing the herbaceous cover to 90%, with cover remaining almost invariable some years before treatment [14]. Even though clear-cutting has been proposed as a management practice to control shrub encroachment, this measure alone is not enough to counteract natural plant succession [26]. Indeed, in our study plot, the cover of short shrubs returned to a pre-treatment situation two years after the intervention. This means that other active management practices, such as extensive livestock grazing, need to be implemented to maintain grasslands [27]. Moderate cattle stockings can prevent shrub encroachment in Pyrenean grasslands [27] owing to the increase of cows in the area during the last decades [11]. Nonetheless, goats are the only cattle significantly feeding on woody plants [12], having the possibility to efficiently control shrub encroachment, but that kind of cattle decreased by 57% in the area during the last decades [11]. In fact, shrub encroachment and afforestation increased in the Natural Park, and especially in the study area (municipality of Vilallonga de Ter), following a consistent pattern of landscape change during the last decades in the Pyrenees [10]. Increasing cattle diversity can be an option for managing grasslands because the potential role of cows alone in controlling shrub encroachment can be questioned. Indeed, combining the effects of cows, affecting vegetation seedlings and resprouting by trampling [27], with goats, removing woody aerial parts by grazing [12], will be a more efficient way to prevent grassland regression and to preserve their plant diversity [28].

Interestingly, clear-cutting changes on vegetation had a significant effect on the composition of the small mammal community inhabiting the study plot. During the two years after the intervention (summer 2019–autumn 2020), we detected a significant increase in the species density [29] and diversity (Shannon index), with the occurrence of typical grassland species such as common and field voles. *Microtus arvalis* is considered a widespread species with the potential as a pest for agroecosystems [30], but in the Pyrenees, it can be considered a rare species associated with highlands [16], and with populations lacking apparent cyclicity. Common voles showed a significant response to clear-cutting and colonized the plot for the first time during the summer of the second post-treatment year. Small increases in shrub cover produced relevant community changes, by decreasing diversity and increasing dominance of the two most common species. Indeed, *A. sylvaticus* and *C. russula* could be favored by shrub encroachment owing to the increased abundance of shrubby habitats in Mediterranean landscapes [31].

The axial Pyrenees are home to a very diverse small mammal community [6], but biodiversity associated with grasslands in the Mediterranean Region is under the threat of climate and landscape change (e.g., shrub encroachment and afforestation, [32]). We are aware that the small mammal community of grasslands is constituted by fossorial (hypogeal) species not (or barely) detected by the sampling method used in this study (e.g., moles *Talpa aquitania*). Indeed, the low abundance of some hypogeal arvicolines in our study plot—and elsewhere in the Pyrenees (www.semice.org)—could be a reflection of sampling issues. Nonetheless, other authors captured those species with the same epigeal devices used in this study (e.g., Sherman traps [33]; Longworth traps [34]). Our perception is that specialist small mammal species associated with grasslands are becoming scarce and hence, its conservation concern increased. Some evidence is the lack of recent detections for endemic species (e.g., *Microtus gerbi*), the low abundance and apparent lack of cyclicity of populations for common European species such as *M. arvalis* [35], and the new specific status reducing the geographical range of the once widespread field vole (e.g., *M. lavernedii*). Nonetheless, the three species detected just after treatment represented a very reduced fraction of the whole community (<20%). In fact, the grassland community was dominated—throughout the seasons and years—by two widespread and generalist small mammal species: *A. sylvaticus* and *C. russula*. Grassland regression produced a banalization of communities by the combined effects of increased dominance of generalist species and the scarcity of specialist species, as was found for other taxa [32]. Despite that *C. russula* can be negatively affected by climate and landscape change (afforestation) in the Mediterranean lowlands [36], this shrew showed potential for expanding its range both northwards and upwards [37], as was yet observed in the axial Pyrenees (authors unpub.). This species showed a positive association with shrub cover, which provides protection against predators and harsh climate [37], thus, being favored by shrub encroachment. Interestingly, *C. russula* abundance was also associated with stone walls, which may provide a habitat niche in the absence of natural landscape traits [38]. The conservation of stone walls, despite having lost the purpose for which they were built (e.g., crops), can be relevant for epigeal small mammals in areas with low woody vegetation cover such as grasslands. *A. sylvaticus* can be the dominant small mammal species in the Mediterranean and montane communities of the NE Iberian Peninsula [16,39], and this was the most common species in the grassland studied. This species behaves as ecotonic [40], taking advantage of forest edges to quickly colonize the grassland during favorable periods. Despite being widespread and without conservation concern, both are key species playing relevant ecological roles in the Mediterranean as seed dispersers [41], pest predators [42], and as prey producing demographic responses of raptors and mesocarnivores [35,39,43].

Summarizing, our preliminary study suggests that clear-cutting reverted shrub encroachment and increased the diversity of the small mammal community in a Pyrenean grassland. However, the effects were short-term—lasting about two years—and then the community returned to the pre-treatment situation. Shrub encroachment represents a significant conservation problem, and it is difficult to fight against it under the process of landscape change in the Mediterranean basin. The role of cattle grazing in controlling shrub encroachment can be of limited importance, owing to the need for a reduction of cow stockings to a minimum number per ha, and herds of goats are declining. Nonetheless, cattle grazing in montane grasslands produced negative effects on small mammal communities through grazing-induced changes in vegetation height and volume and in soil structure [24]. Thus, a compromise between increasing cattle stockings to effectively control shrub encroachment and their negative effects on small mammal communities need to be counter-balanced to preserve the small mammal diversity of montane grasslands.

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