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Original article

Regenerative role of seed banks following an intense soil disturbance

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Abstract

Our main aim was to determine the contribution of the seed bank to vegetation regeneration following a disturbance consisting in a deep ploughing and a thorough homogenisation of a perennial grassland. In the seed bank prior to disturbance, seed distribution through the vertical soil profile was evaluated to determine the initial seed species structure. Then, several characteristics of the shallow seed bank and the extant vegetation were evaluated prior and following field disturbance: seed species composition and abundance, and species composition of the aboveground vegetation. The contribution of seed rain versus seed bank was evaluated by means of the comparison of the vegetation developed in plots filled with sterilised soil (seed bank removal) and the vegetation developed in non-sterilised plots in the field. The distribution of seeds through the profile indicated a sharp decline in abundance with depth, and it was probably linked to propagule morphology, with small and rounded seeds proner to being buried deeper than larger seeds. In the grassland prior to disturbance, the aboveground vegetation and seed bank species composition showed very low similarity index, most likely because during the 5 years following field abandonment, sheep pressure had caused a faster change in aboveground vegetation species composition than in seed bank species composition. Ploughing and homogenisation of the grassland led to low seed abundance in the shallow soil layer caused by dilution of the seed bank. Regardless of impoverishment in seed abundance and species richness, comparison between sterilised and non-sterilised plots showed that the seed bank acted as an effective source of colonising species and determined the aboveground species composition. To summarise, this study outlines the importance of considering several characteristics of the seed bank, such as species composition and seed abundance, in the understanding of the function of seed bank and dynamics of the vegetation following a deep ploughing and homogenisation treatment.

Keywords: Grassland; Regeneration; Seed bank composition; Structure

1. Introduction

Seed banks are considered essential constituents of plant communities (Harper, 1977), since they contribute significantly to ecological processes. The recovery-ability of vegetation after disturbance is believed to lie mainly in the buried seed population (Uhl et al., 1981, 1982; Marks and Mohler, 1985; Lawton and Putz, 1988; Kalamees and Zobel, 2002). The understanding of the dynamics and function of seed banks has become a great challenge to plant community ecologists, as this understanding is necessary to enable determining the role of this community trait in ecosystem functioning and also to improve integrated management of ecosystems. Although a complete set of topics on seed banks such as the spatial distributions of seeds in soil (Thompson, 1986; Bigwood and Inouye, 1988; Lavorel et al., 1991), seed persistence in soil (Thompson et al., 1993; Hendry et al., 1995), effects of environmental factors on seed bank dynamics (Baskin and Baskin, 1989; Thompson, 1992; Kitajima and Tilman, 1996), and the resemblance of seed species

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composition on the aboveground vegetation (Bigwood and Inouye, 1988; Graham and Hutchings, 1988a, 1988b; Levassor et al., 1990; Lavorel et al., 1991; Lavorel and Lebreton, 1992) have been described, there are almost no studies about the regenerative potential of seed banks (but see Kalamees and Zobel, 2002; Middleton, 2003; Deiller et al., 2003).

The contribution of the seed bank to the regeneration process is mainly dependent on the existence, abundance and frequency of appearance of bare soil gaps varying in size and shape (Fenner, 1985; Rogers and Hartnett, 2001). It is widely accepted that the vegetation composition of scarcely disturbed communities, such as grasslands, is mainly affected by vegetative growth and only slightly influenced by the seed bank composition (Thompson and Grime, 1979; Van Andel et al., 1993; Eriksson, 1993; Kalamees and Zobel, 2002). Conversely, in frequently disturbed habitats, such as arable fields, vegetation composition is expected to be mainly determined by the seed bank composition (Marks and Mohler, 1985; Levassor et al., 1990; Lavorel et al., 1991; Warr et al., 1993). Our work evaluates to what extent seed bank characteristics such as composition, abundance and vertical distribution of seeds affect the development of the aboveground vegetation. Furthermore, this study brings a contribution not yet sufficiently analysed, through calibration of the regenerative potential of the seed bank versus the seed rain and comparing vegetation development in both situations. To our knowledge few studies have analysed the balance between the seed bank and the seed rain in the conformation of communities (see Kalamees and Zobel, 2002). We address the following specific questions: (1) Does the grassland prior to disturbance show any vertical distribution of seed species and abundance in seed bank? (2) Which seed bank features are affected after a deep ploughing and homogenisation treatment? (3) To which extent does regeneration following a disturbance depend on the seed bank or seed rain characteristics? (4) To which extent the seed bank composition after disturbance affects the resulting plant community composition?

2. Materials and methods

2.1. Study site

The study site was located 3 km east of Vitoria-Gasteiz (Basque Country) in Northern Spain (42° 51′N; 2° 37′W and 510°m in altitude), close to the boundary between the Eurosiberian and Mediterranean regions (Rivas-Martínez, 1987). Climatic conditions in this region show annual average rainfalls of 843 mm and mean average temperatures around 11.7 °C, ranging between 1.3 °C in the coldest and 26.1 °C in the warmest months. Rivas-Martínez and Loidi (1999) classified this climate as temperate submediterranean and subhumid. Rainfall is highly unpredictable in time and quantity and pronounced summer droughts are common.

The study was undertaken in a 3 ha perennial grassland (*Agropyro-Rumicion crispi*) that had been cultivated 5 years before, and developed under sheep grazing conditions. The dominant species included native perennials such as *Agropyron repens*, *Arrhenatherum bulbosum*, *Rumex crispus*, *Geranium dissectum*, and *Picris echioides*. Soils were vertisols with a slight hydromorphy.

2.2. Seed-bank sampling

In early February 1999, 30 soil cores were collected at random in the grassland prior to disturbance. Soil cores were 7 cm in diameter and 12 cm in height. In order to analyse the seed bank vertical distribution, each soil sample was divided into three fractions: the soil shallow layer (soil surface to 4 cm depth), the mid-layer (4–8 cm depth) and the deepest layer (8–12 cm depth). Each fraction (90) consisted of 140.45 cm^3. In late February 1999, the grassland suffered a disturbance consisting in a deeply ploughing (50 cm deep), followed by an intense tillering (30 cm) and hand-raking (5 cm) to obtain complete soil homogenisation and complete loss of the vertical distribution of the seed bank. Following the intensive soil management, another 40 soil cores (12 cm diameter and 3 cm height) were extracted at random in early March. The same soil volume per sample was collected in both seed bank types (140.45 cm^3) in order to make it possible to compare composition and abundance among soil cores before and after disturbance in the shallow layer. Given that the seed bank following disturbance is considered to be completely homogenised, and thus any previous spatial distribution lost, samples were collected only from the top 3 cm layer, so we did not analyse seed distribution with depth after field disturbance. Furthermore, it is widely accepted that seeds buried deeper than 3 cm have almost no contribution to the colonisation process mainly in old-fields (Fenner, 1985; Baskin and Baskin, 1998), and thus to the vegetation developed aboveground.

The direct germination method of Thompson and Grime (1979) was used to assess the readily germinable seed species composition (Gross, 1990; Rothrock et al., 1993). Each soil sample was merged with vermiculite (1:1), spread in a 12 × 12 × 2 cm tray and placed in a greenhouse. Soil samples were watered regularly and once seedlings emerged, they were identified and removed. Most studies in abandoned field communities show that provided suitable conditions, most seeds germinate within the first 2 months (Graber and Thompson, 1978; Lavorel et al., 1993). In this study, monitoring was undertaken weekly during the first month and fortnightly afterwards until day 108 (4 months) in the presence of the natural mixture of seed species.

2.3. Vegetation sampling

Vegetation prior to disturbance was measured as plant density and cover in 20 randomly placed quadrats (0.25 m^2) in July 1998. Vegetation following disturbance was measured at the maximum phenological peak of the community—in July 1999, 5 months after disturbance—at 40 random quadrats (0.25 m^2). Throughout each sampling period, each tiller of every species was counted and cover was estimated for those species with at least 1.5% of total cover. Nomenclature follows Aizpuru et al. (1999).

2.4. Soil sterilisation treatment: balance between seed bank and seed rain contribution to regeneration

In order to estimate the role of the seed bank for the establishment of vegetation after disturbance, a soil sterilisation experiment was undertaken. In early March 1999, immediately after the disturbance treatment, soil was sterilised in 40 quadrats (0.25 m^2) adjacent to those used for vegetation sampling. Soil sterilisation involved removing a 4 cm deep layer of soil from each 0.25 m^2 quadrat and subjecting it to steam heating at 120 °C for 70 min. Subsequently, the sterilised soil was replaced into the field quadrats. The comparison between plant development in plots with sterilised and non-sterilised soil showed us the relative contribution of seed bank and seed rain to vegetation development.

2.5. Numerical analyses

Data from the seed bank composition was highly skewed and although standard transformations (log and square root) were assayed to meet normality, they were insufficient to normalise the data in most cases (Reichman, 1984; Warr, 1991). Consequently, abundance and species richness was expressed as confidence intervals around means with a confidence coefficient of 0.95 (1000 randomisations), and non-parametric statistical methods were used for hypothesis testing. Dunn's multiple comparison test was used to test for multiple comparisons (Zar, 1999).

The seed bank species composition is a multidimensional data set (species × samples), so in order to search for global differences between depths it would be appropriate to undertake ordination constraining methods for hypothesis testing (McCune, 1997). Moreover, these techniques are free of the distribution restrictions characteristic of other analysis such as multivariate analysis of variance (Legendre, 1993). In order to select the appropriate ordination technique, the seed matrix was submitted to a detrended correspondence analysis (DCA), detrending by segments and with non-linear rescaling of axes, whereby the extracted gradient is scaled in units of average standard deviation (Gauch, 1982). The seed bank data set showed a relatively large gradient (4.35 S.D. units), so following the recommendations of Ter Braak (1986) and Legendre and Anderson (1999), a canonical correspondence analysis (hereafter CCA) was performed as long as it assumes unimodal responses. CCA ordination methods allow the constraining of CA axes to be linear functions of explanatory variables. Our null hypothesis (Ho) to test was: the effect of depth on the distribution of the seed bank prior to disturbance was not significantly different from random.

The main matrix consisted of seed species densities per soil sample and depth in the grassland previous to disturbance (47 species × 90 soil samples), whereas the constraining matrix was constructed with the three variables of depth defined as dummy variables (3 depths × 90 soil samples), thus, with only two degrees of freedom. To obtain the proportion of the total variation of the seed bank species composition explained by the constraining matrix—or the total variation explained (TVE)—a comparison between the inertia associated with the constrained analysis I(CCA) and the inertia of the unconstrained data set I(CA) was undertaken. A Monte Carlo permutation test was performed to determine the accuracy of the relationship (1000 randomisations) between the two data sets. The sum of all canonical eigenvalues—named trace—was used to build the F-ratio statistic (Ter Braak, 1990; Verdonschot and ter Braak, 1994; Legendre and Anderson, 1999). Only when $P < 0.05$ was the relationship between the two data sets considered significant.

If the CCA model was significant, a forward stepwise procedure was carried out to detect which constraining variables explained significant portions of the variability of the main matrix. Variables were ranged by means of their eigenvalues in a decreasing way and then, they were added one at a time to the model until the new variable included was not significant ($P > 0.05$). Improvement of the reduced model with each selected variable was determined by a Monte Carlo permutation test with 1000 randomisations. Ordination analyses were conducted with CANOCO for Windows v. 4.0 (Ter Braak and Smilauer, 1997).

The resemblance of the seed bank and the aboveground flora composition prior and following disturbance was estimated by means of Sorensen's similarity coefficients (Sorensen, 1948). This coefficient was chosen because it provides double weight to the presence of a species in both data sets, given that the presence of a species is more informative than its absence to understand function and structure of the plant community.

3. Results

3.1. Seed abundance and species richness in the seed bank prior and following disturbance

A gradual loss of seed abundance with depth was observed in pre-disturbed soil. Seed abundance was significantly higher in the shallow layer with no difference in densities between the deeper two fractions. Similarly, species richness declined with seed bank depth (Table 1), with each depth showing specific species composition (TVE = 5.13%; $P = 0.001$) (Table 2 and Fig. 1). The most abundant species detected in the shallow seed bank layer prior to disturbance were *Lolium perenne* (24.5% of the total seed abundance) and *Picris* spp. (25%) (Fig. 2). These species together with *Sinapis arvensis*: 3%, *Juncus bufonius*: 4.5% and *Verbena officinalis*, 3% represented ca. 60% of the total seed bank

Table 1
Seed bank and aboveground vegetation species richness and densities. Confidence interval of the mean is calculated by means of a bootstrapping procedure. Seed densities are expressed in individuals per dm^3. Seed bank species richness is measured for 140.45 cm^3. Vegetation density and species richness was expressed in 0.25 m^2. Significance was performed by means of a Dunn's Tukey-type multiple comparison test based on a Kruskal–Wallis analysis

	Density		Species richness	
	Significance	Confidence interval (5–95%)	Significance	Confidence interval (5–95%)
(a) Seed bank				
Pre-disturbance				
Shallow layer (0–4 cm)	a	133.85–180.42	a	6.7–8.6
Mid-layer (4–8 cm)	b	58.62–85.44	ab	4.83–6.3
Deepest layer (8–12 cm)	bc	41.73–60.15	bc	4–5.24
Post-disturbance (0–3 cm)	c	29.6–45.74	c	3.04–4.22
(b) Aboveground vegetation				
Sterilised soil	a	18.74–26.13	a	6.39–7.65
Pre-disturbance field	b	78.4–249.6	ab	10–14.2
Post-disturbance field	b	156.77–200.02	b	19.65–21.6

Table 2
Forward stepwise CCA for depth variables and the reduced model built with the two significant depths

Main matrix : pre-disturbance seed bank			
Steps	λ	Frat	*P*
Depth A	0.19	2.99	0.001
Depth B	0.10	1.58	0.028
Depth C	–	–	–
Reduced model			
TVE = 5.13	Frat = 2.293		
Drop TVE = 0	*P* = 0.001		

composition. In the mid and deepest layers, the most abundant seed species were *Lolium perenne* (20% and 19.2%, respectively), followed by *Sinapis arvensis* (11.5% and 11.3%, respectively). Species can be classified into three groups regarding the depth where they were mainly detected (Fig. 1 in the up right hand corner). Those more abundant in the shallow layer: *Lolium perenne*, *Picris* spp, *Juncus bufonius*, *Cerastium glomeratum*, *Rumex crispus*, etc., those occurring in the mid-layer: *Sinapis arvensis*, *Verbena officinalis*, *Sagina sabuletorum*, *Papaver rhoeas* and those mainly observed in the deepest layer: *Anagallis arvensis*, *Kickxia spuria*, *Arenaria serpyllifolia* and *Chenopodium album*.

Seed abundance and species richness in the soil shallow layer decreased significantly following disturbance, being seed abundance reduced by 75% (Table 1). In the disturbed seed bank, six species represented more than 65% of the total seed bank composition: *Juncus bufonius* (20.3%), *Lolium perenne* (16.3%), *Verbena officinalis* (12.8%), *Sagina sabuletorum* (6%), *Mercurialis annua* (5.5%) and *Anagallis arvensis* (5%) (Fig. 2). Disturbance of the soil seed bank resulted in a decrease of the three most prominent species (*Lolium*, *Picris*, *Sinapis*), while it also resulted in an increase in the *Juncus* and *Verbena* species. Although the species composition of the shallow layer of both seed banks prior and following disturbance was very similar (Sorensen's coefficient = 0.79), new species were detected following disturbance: *Mercurialis annua*, *Trifolium repens* and *Pastinaca sativa*, while other species were not observed: *Crepis vesicaria*, *Lolium multiflorus*, *Galium aparine*, *Veronica persica* (Appendix A).

3.2. Similarity between seed bank and vegetation prior to disturbance

In the perennial grassland prior to disturbance, relatively low similarity between species composition in the vegetation and in the seed bank was detected (Sorensen's similarity index = 0.45). Considering the abundance of each species, vegetation was mainly dominated by perennial grasses whereas the seed bank was dominated by early succession forb species (Fig. 3a).

3.3. Contribution of the seed bank to the regeneration of vegetation following disturbance

Vegetation established 5 months following disturbance reflected seed bank species composition to some extent (Sorensen's similarity index: 0.61) (Fig. 3b). Aboveground vegetation and seed bank were mainly dominated by annual weeds such as, *Picris echioides*, *Sinapis arvensis*, *Medicago* spp. and *Anagallis arvensis*, but while perennial grasses greatly contributed to vegetation composition (30% of total density), no seeds of these species were detected in the seed bank. However, the relative abundance of each species in vegetation composition was not proportional to the abundance of each species in the seed bank.

Plant density was 12.2 times higher in plots not submitted to soil sterilisation treatment compared to sterilised plots (Fig. 4). The proportional contribution of each species to vegetation composition did not vary significantly whether the seed bank was present or not: *Lolium perenne*, *Anagallis arvensis*, *Picris echioides*, *Medicago* spp. and *Sinapis arvensis* were the most abundant annual species in both situations. Otherwise, perennial grass plants were completely absent from the sterilised plots.

4. Discussion

4.1. Seed abundance and species richness in the seed bank prior and following disturbance

The grassland seed bank mainly consisted of early succession species, e.g., *Picris echioides*, *Sinapis arvensis*, *Lolium*

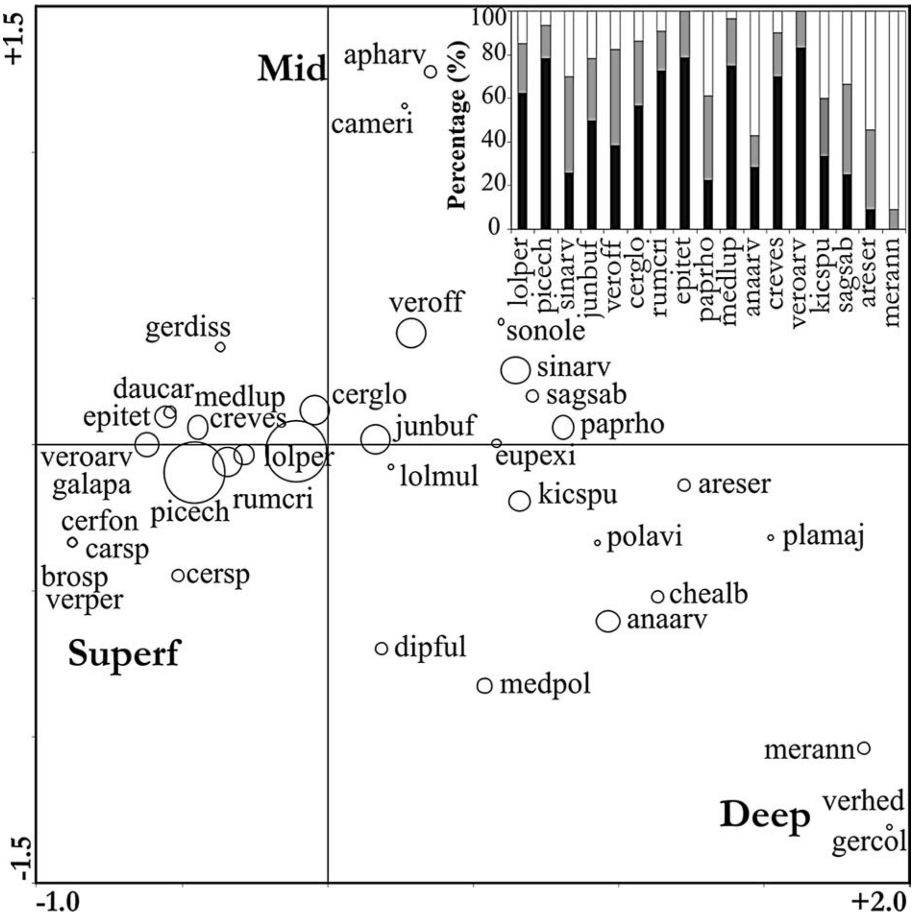

Fig. 1. CCA biplot (axes 1 and 2) representing the seed bank composition previous to disturbance in relation to depth. Dummy depth variables were introduced as centroids. “Deepest” layer (8–12 cm deep) was included as a passive variable. This dummy variable was not considered in the analyses to avoid multicollinearity problems. “Shallow”: shallow layer and “Mid”: mid-layer. Circle sizes represent species abundance per soil sample. In the top right corner: species abundance percentages with depths are displayed. Black bars: shallow layer; grey bars: mid-layer; white bars: deepest layer. Species abbreviations are indicated in Appendix A.

perenne, etc. This result is similar to other studies carried out in recently disturbed habitats (Moore, 1980; Cavers and Benoit, 1989; Milberg and Hansson, 1993). The relatively high seed densities found in this perennial grassland are more similar to arable lands (Brenchley and Warington, 1930, 1933; Kropac, 1966; Jensen, 1969) than to genuine grasslands, which usually show relatively smaller seed banks (Chippindale and Milton, 1934; Champness and Morris, 1948; Major and Pyott, 1966; Bakker, 1989). Large seed banks beneath grasslands have only been found where previous arable cultivation has occurred (Schenkeveld and Verkaar, 1984; Lavorel et al., 1993). Indeed, the agricultural history of this grassland indicates that it was cultivated only 5 years prior to the present study.

Surprisingly, the seed bank had developed a marked vertical distribution in only 5 years. Seed abundance and species richness showed a decreasing trend with depth, being significantly higher in the shallow layer than in the mid and deepest layers. Many other studies have reported similar trends of decreasing densities with depth (Putz and Appanah, 1987; Kramer and Johnson, 1987; Kitajima and Tilman, 1996). More interestingly, species composition was related to the vertical distribution. Thus, some species were more abundant near the surface, whereas others were mainly detected in the mid-layer and a third group of species that mainly appeared in the deepest layer. Patterns of species distribution with depth have been detected in other studies (Kellman, 1978; Young, 1985; Young et al., 1987). Most probably, the vertical structure may lie in the different ability of seeds to penetrate the soil (Thompson et al., 1993), together with the fact that viability is retained for longer when seeds are buried in the soil (Rampton and Ching, 1970).

The ability of seeds to penetrate into the soil may lie in the relationship between the morphological features of the dispersal structures (size and shape) and its tendency to get buried (Thompson and Grime, 1979; Thompson, 1987; Leck, 1989; Thompson et al., 1993). Dispersal structures that are large (*Rumex crispus*), elongated (*Lolium perenne*) and/or have a rough surface (*Picris* spp.) showed a strong tendency to occur preferably in the most shallow layer, due to greater soil resistance, conversely, dispersal structures that are small (*Papaver rhoeas*), compact (*Anagallis arvensis*) and/or with a smooth surface (*Sinapis arvensis*) are more prone to getting buried. The relationship of seed size and shape with seed

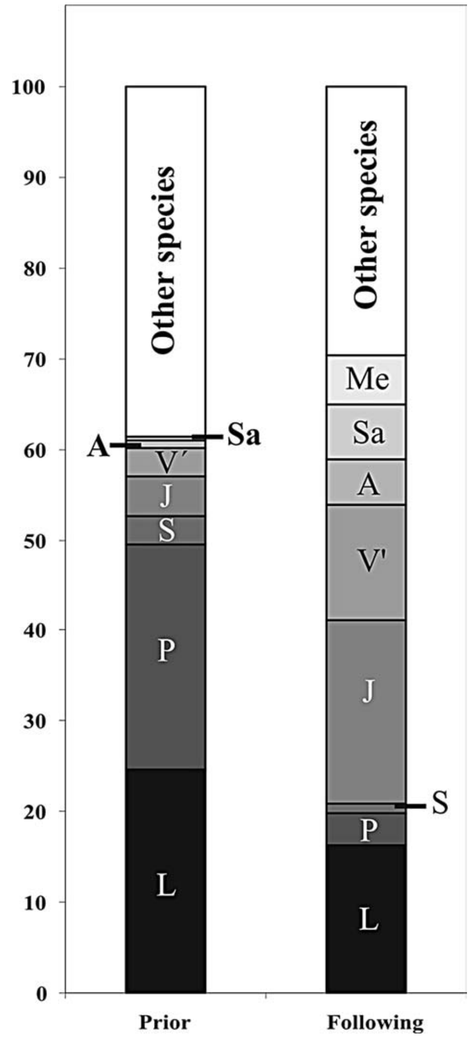

Fig. 2. Percentage density contribution of the eight more common species to the soil shallow seed bank layers prior and following disturbance. A: *Anagallis arvensis*, J: *Juncus bufonius*, L: *Lolium perenne*, Me: *Mercurialis annua*, P: *Picris echioides*, S: *Sinapis arvensis*, Sa: *Sagina sabuletorum*, V′: *Verbena officinalis*.

bank persistence (following Thompson et al., 1993) seems to fulfil in temperate climates (Thompson and Grime, 1979; Thompson et al., 1993), on the contrary, in Mediterranean old fields, no clear relationship has been found (see Lavorel et al., 1993). In our study, some species did not respond as predicted. *Lolium perenne* is considered to have transient seed banks (Thompson et al., 1993), however, this study demonstrated important stocks (37%) of seeds of this species buried below 4 cm depth. The historic management of this field may have led to burying episodes of *Lolium perenne* seeds to deeper layers and once buried, seeds may have obtained physiological dormancy (Rampton and Ching, 1970).

Not surprisingly, following disturbance, seed abundance and species richness decreased in the shallow layer, mainly as a result of dilution of the seed bank (Colbach et al., 2000; Rokich et al., 2000). Indeed, low seed densities have been found in other recently ploughed fields, which gradually became enriched with species over time (D'Angela et al., 1988; Levassor et al., 1990; Lavorel et al., 1991).

4.2. Similarity between seed bank and vegetation prior to disturbance

In the perennial grassland prior to disturbance, the seed bank was dominated by early successional species while conversely, the aboveground vegetation was mainly dominated by perennial grasses. Seed banks are accumulated over time from the vegetation previously established in a site (Brown and Oosterhuis, 1981; Hill and Stevens, 1981), thus, it was evident that the seed bank composition reflected the historical heritage of the grassland. Sorensen's similarity index (0.45) between the seed bank and the vegetation composition was not as low as those found in seed banks of other typical grasslands (Thompson and Grime, 1979; Thompson, 1986; D'Angela et al., 1988; Bigwood and Inouye, 1988; Kitajima and Tilman, 1996), but was quite inferior to that found by Lavorel and Lebreton (1992) for their 7- and 15-year-old-fields (0.70). In the study by the latter authors, annual species became dominant in mid-succession communities, while in this study, a community dominated by perennial grasses rapidly became dominant, probably as a consequence of efficient vegetative growth (*Arrhenatherum bulbosum* and *Agropyron repens*). High sheep loads together with climatic conditions favourable for perennial grasses establishment, would probably have shaped our grassland community, turning it into creeping perennial grasses with their main reproductive investment directed towards vegetative spreading. Most likely, while aboveground vegetation composition and structure were being shaped by sheep grazing, the persistent seed bank, comprised of long-lived seeds, did not undergo changes in species composition. Milberg and Hansson (1993) also found slight effects of vegetation changes on seed bank species composition in grazed and non-grazed limestone grasslands, although, Milberg (1992) reported converse results in a study of a tall grassland prairie.

Prior to disturbance, no seeds of *Arrhenatherum bulbosum* and *Agropyron repens* were found in the seed bank, despite both species dominating the aboveground vegetation. The germination biology of this species may prevent seed accumulation in the soil (Type I transient seed banks; following the classification of Thompson and Grime, 1979). The absence of a seed bank for species relying on vegetative reproduction seems to be a common trait (Whitney, 1986; Viragh and Gerencser, 1988; Graham and Hutchings, 1988a, 1988b; D'Angela et al., 1988; Rice, 1989; Houle, 1991; Milberg, 1992). Conversely, some species with large contributions to seed bank composition were not observed in the extant vegetation (i.e. *Juncus bufonius*, *Papaver rhoeas*, *Rumex crispus*, *Anagallis arvensis*). This may be due to the interaction of two factors: firstly, these species are provided with long-lived seeds able to form persistent seed banks (Milberg and Hansson, 1993) and secondly, they are only able to germinate and establish when disturbance events happen (Thompson and Grime, 1979).

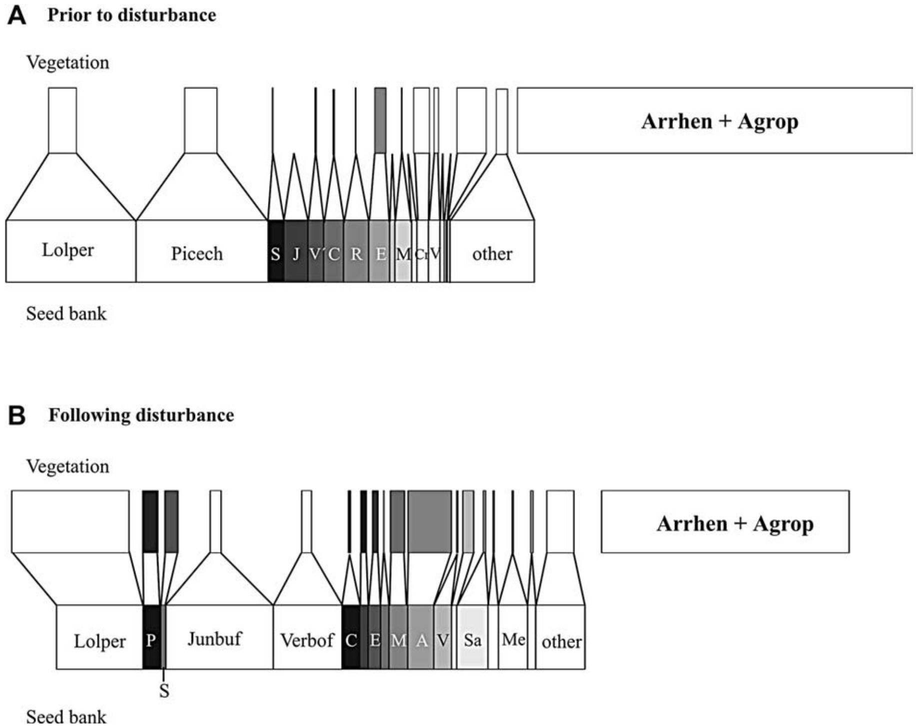

Fig. 3. Proportional contribution of each species to the composition of the aboveground vegetation and the seed bank (a) prior to disturbance and (b) following disturbance. A: *Anagallis arvensis*, C: *Cerastium* spp., Cr: *Crepis vesicaria*, E: *Epilobium tetragonum*, J: *Juncus bufonius*, M: *Medicago* spp., Me: *Mercurialis annua*, P: *Picris echioides*, R: *Rumex crispus*, S: *Sinapis arvensis*, Sa: *Sagina sabuletorum*, V: *Veronica arvensis*, V′: *Verbena officinalis*. Rest of the species abbreviations in Appendix A.

4.3. Contribution of the seed bank to the regeneration of vegetation

The early successional plant community developed following disturbance was mainly composed of annual weed species similar to those already present in the soil seed bank. Many other studies have also reported high similarities between aboveground vegetation and seed bank in habitats that suffer periodical disturbances (Jensen, 1969; Wilson et al., 1985; Levassor et al., 1990; Lavorel et al., 1991). These results support Moore's (1980) hypothesis that vegetation and seed bank composition should be similar in frequently disturbed communities dominated by early successional annuals with persistent seed banks. Similarities between both community attributes may lie in the opportunistic behaviour of species that comprise the seed bank. As soon as any disturbance occurs, these species become dominant in the aboveground vegetation so that the vegetation composition in the first stages of succession reflects the seed bank composition.

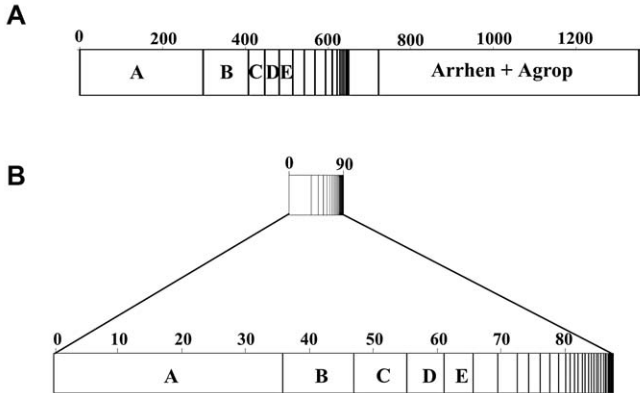

Fig. 4. Aboveground vegetation composition following disturbance (a) in quadrats with the intact seed bank and (b) in sterilised plots. Units are individual/m^2. A: *Lolium perenne*, B: *Anagallis arvensis*. C: *Picris echioides*, D: *Medicago* spp., E: *Sinapis arvensis*, Arrhen: *Arrhenatherum bulbossum* and Agrop: *Agropyrum repens*.

Seeds and propagules stored in the soil played a crucial role in colonisation of disturbed habitats, while seed rain had a discrete contribution. This was evidenced by the low vegetation densities and slow colonisation rate (personal observation) in sterilised plots, thus, vegetation that developed initially in the early secondary succession following disturbance, depended to a great extent on the seed bank and vegetative propagule bank composition. Despite plant density being 12.2-fold lower in sterilised plots, the proportional contribution of each species to the aboveground vegetation composition was very similar to plots whose seed bank was left untouched (see Fig. 4). This similarity is the result of secondary seed dispersion from the surrounding bare soil, because primary seed dispersal from the extant vegetation did not occur until more than a month after this study had finished.

5. Conclusion

In the early successional grassland, the seed bank vertical distribution was developed in only 5 years following cultivation. This was probably due to a set of jointly acting processes, such as, the morphological features of seeds and the historical management of the field. Following the deep ploughing and the subsequent soil homogenisation treatment, the seed abundance in the shallow layer was drastically reduced due to dilution. Given that the regeneration process in early secondary succession depends to a great extent on the presence of the soil seed bank rather than on seed rain, and given that seeds below a few cm depth are not able to emerge, the decrease in seed bank densities in the shallow layer most likely has implications in the plant community regeneration process. Indeed, the aboveground species composition of the recently disturbed field reflected a higher proportion of the soil seed bank species composition. All these changes may be responsible for the subsequent vegetation composition and community-structure.

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Appendix A. Mean seed densities (number dm^{-3}) and mean plant covers (percentages) previous and after the ploughing treatment

Species	Abbr.	SB previous to disturbance						SB_post		VEG_pre		VEG_pos	
		Depth A		Depth B		Depth C		Mean	S.D.	Mean % cover	S.D.	Mean % cover	S.D.
		Mean	S.D.	Mean	S.D.	Mean	S.D.						
Picris spp.	picsp	39.4	39.4	7.6	14.8	3.4	5.9	+		0.50	0.8	13.5	11.4
Lolium perenne	lolper	38.7	29.7	14.0	10.3	9.6	14.7	5.9	6	*		*	
Cerastium spp.	cersp	7.8	13	3.1	9.8	+		+		*		*	
Rumex crispus	rumcri	7.6	17.6	+		+		+				*	
Juncus bufonius	junbuf	7.1	10.2	4.0	6.9	3.2	6.5	7.3	11.6			*	
Epilobium tetragonum	epitet	6.2	10	+				+		1	0.8	*	
Medicago lupulina	medlup	4.9	18.4	+				+				9.4	7.9
Sinapis arvensis	sinarv	4.7	11.1	8.1	12.5	5.6	6.7	+		*		8.3	8.6
Verbena officinalis	verboff	4.7	8.4	5.5	11.3	+	5.1	4.6	9.2	*		*	
Veronica arvenis	verarv	3.6	6.7	+				+		*		*	
Crepis vesicaria	creves	3.3	9.3	+		+				5.21	2.5	3.1	2.7
Anagallis arvensis	anaarv	1.4	3.9	0.7	2.2	3	4.5	1.8	3.1			*	
Sagina sabuletorum	sagsab	0.7	2.2	1.2	3.9	1	4.1	2.1	4.6			*	
Papaver rhoeas	paprho	+		+		2.9	7.3	+				*	
Daucus carota	daucar	+		+				+		*		*	
Kickxia spuria	kicspu	+		+		+		+		*		+	
Dipsacus fullonum	dipful	+				+		+				5.3	2.6
Medicago polymorpha	medpol	+				+		+				11.9	10.2
Lolium multiflorus	lolmul	+		+		+						*	
Poa trivialis	poatri	+											
Geranium sp	gersp	+		+		+		+		4.40	2.7	4.8	4.6
Bromus sp	brosp	+						+				*	
Grass (*Agrop+Arrhen*)	grass									75	22.5	30.8	26.5
Chenopodium album	chealb	+		+		+		+				*	
Poligonum aviculare	polavi	+		+		+		+				*	
Galium aparine	galapa	+										*	
Sonchus oleraceus	sonole	+		+		+						*	
Euphorbia exigua	eupexi	+		+		+							
Veronica persica	verper	+										*	
Aphanes arvensis	apharv	+		+				+				*	
Arenaria serpyllifolia	areser	+		+		+		+		*		*	
Campanula erinus	cameri	+		+				+				*	

Cardamine hirsuta	carhir	+						+					
Conyza sumatraensis	consum	+											
Nigella gallica	niggal	+											
Cirsium arvense	cirarv			+								6.7	5.7
Vicia sativa	vicsat			0.2	1.3					0.25	0.6		
Lathyrus sp	latsp											*	
Potentilla reptans	potrep											*	
Convolvulus arvensis	conarv											*	
Polygonum convolvulus	polcon											*	
Mercurialis annua	merann			+		2.5	5.8	1.9	3.9			*	
Lathyrus aphaca	lataph											*	
Trifolium repens	trirep							+		*		*	
Sherardia arvensis	shearv											*	
Pulicaria dysenterica	puldys											*	
Trifolium pratense	tripra									0.25	0.6	*	
Taraxacum officinale	taroff									*			
Pastinaca sativa	passat							+		0.25	0.6		
Solanum nigrum	solnig							+					

Abbr., abbreviation; SB_post, seed bank after perturbation; VEG_pre: vegetation previous to disturbance; VEG_post, vegetation after disturbance; +, number of seeds in the experiment below 3; *, presence of the species cover below 1%.

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