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# Improving the pasture quality through the annual legumes oversowing: previous results on the seed bank response

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**Summary** - The understanding of changes in botanical composition and seed bank dynamics was the aim of an experiment conducted on a natural pasture and oversown with annual ryegrass, subclover and burr medic. The site is located in western Sardinia and was grazed at 8 ewes/ha/year in April, September and December. Viable seed bank (VSB) composition was assessed by collecting intact soil cores at the end of summer 1998. VSB pool components were separated by 3 germination/dormancy breakdown treatments, thus estimating germinable (GSB) and dormant (DSB) seed bank. Autumn and spring overlying vegetation was estimated by non-destructive method using an HFRO sward stick, by recording 4 specific leaf contacts at the vertices of 1200 random quadrats (20 x 20 cm). Changes in overlying vegetation and VSB were analyzed after the durnig the first year after the oversowing: 15% of the VSB was represented by Trikkala subclover which contributed with almost all the seeds as readily germinable. ‘Anglona’ burr medic entrusted its persistence to the shedding of about 30% of its viable seeds as dormant. Among spontaneous species, more than the 50% of *Cistus* viable seeds was dormant, but many of these (about 40%) germinated when satisfied their cold requirements. The other components of the seed bank did not evidence significant chilling demand.

**Key-words:** improved pasture, annual legumes, persistence, seed bank

**Résumé** - La compréhension des changements dans la composition botanique et dans les dynamiques de la banque de graines était l'objectif d'une expérimentation conduite sur un pâturage naturel sursemé avec trèfle souterrain Trikkala et Clare, luzerne annuelle Anglona et ray-grass Nurra. Le site est localisé dans la Sardaigne Occidentale et il a été pâturé en Avril, Septembre et Décembre avec 8 brebis/ha/année. La composition de la banque de graines viables (VSB) a été étudiée en soumettant les échantillons de sols à trois traitements successifs alternant germination et dormance. Le taux de graines susceptibles de germer (GSB) et en dormance (DSB) a été estimé. Le trèfle souterrain Trikkala représente 15% des VSB alors qu'il constitue la presque totalité des GSB. La luzerne annuelle Anglona maintient 30% de ses VSB dormantes. Parmi le stock de graines d'espèces spontanées, 50% des VSB de ciste sont dormants mais presque 40% de ce stock est capable de germer dès que leur besoin en froid est satisfait. Les autres composants de la banque de graines n'ont montré aucun besoin de vernalisation.

**Mots-clés:** amélioration pastorale, Légumineuse annuelle, pérennité, banque de graines

## Introduction

Knowledge of the seed bank ecology can be very informative of the vegetation development and its persistence, providing substantial flexibility for potential community changes (Leck *et al.*, 1989).

In Mediterranean climates with severe summer drought, seed banks are mostly characterised by an abundant presence of “transient” species that contribute to the seed bank for a relative short period after shedding: winter annuals and large-seeded forbs. Other species, defined as “permanent”, show seasonal peaks of abundance, but are present throughout the year. The significance of persistent seed banks in grasslands is far from clear. However, the persistence of such seed banks seems dependent on a continuing lack of

disturbance (Thompson, 1987). Templeton and Levin (1979) stated that the seed bank keep a “memory” of the selective conditions that prevailed in the past as well as more recent conditions; animal predation, fire, overgrazing, tillage can affect the persistence of the seed bank, and, consequently, the regeneration of the vegetation (Clément and Touffet, 1990).

The existence of persistent seed bank species in the soil buffers both population density and botanical composition against environmental changes (Pagnotta *et al.*, 1997), in fact the soil seed bank composition is less influenced than the above-ground vegetation (Pywell *et al.*, 1997). Accordingly, Perez *et al.* (1998) found a lack of correspondence in the representation of species between the soil seed bank and the floristic composition of the vegetation. This reflected differences among species not only in seed production, but also in seed dormancy and survival.

Since persistence of seeds in the soil can have important implications for the restoration and the conservation of plant communities and species (Diemont, 1990), it is important to know how management and climate, as well as simply the passage of time, affect the size and composition of the soil seed bank. In this context, in 1997 an experimentation has begun on a Sardinian improved pasture with the aim to give a contribution for the knowledge of the mechanisms that regulate the equilibrium between the species within the seed bank and the overlying vegetation, in a condition of minimum disturbance. In this paper, the preliminary aspects of seed bank composition and vegetation will be focalised.

## Materials and methods

The study was conducted during 1997-99 at Usellus (Western Sardinia, Italy). The soil is clay-sandy sub-acid, with N content = 0.9 g kg<sup>-1</sup> (Kjeldhal) and P<sub>2</sub>O<sub>5</sub> content = 0.052 g kg<sup>-1</sup> (Bray and Kuntz). The climate of the area is Mediterranean semi-arid with annual rainfall of 510 mm.

In October 1997, 4.8 ha of low-quality pasture were improved oversowing subclover (15 kg ha<sup>-1</sup> of germinable seed composed by 65% of *Trifolium yannanicum* Katzn. and Morley ‘Trikkala’ and 35% *T. brachycalycinum* Katzn. and Morley ‘Clare’), burr medic (11 kg ha<sup>-1</sup> of *Medicago polymorpha* L. ‘Anglona’) and annual ryegrass (1.5 kg ha<sup>-1</sup> of *Lolium rigidum* Gaudin ‘Nurra’) and fertilising with 80 kg ha<sup>-1</sup> of P<sub>2</sub>O<sub>5</sub> and 20 kg ha<sup>-1</sup> of N. All legume seeds were previously scarified. Grazing took place in April (2 weeks), September (3weeks) and December (5 weeks) with an annual stoking rate of 8 ewes/ha.

The composition of the autumn and spring overlying vegetation was estimated using an HFRO sward stick, by recording 4 specific leaf contacts at the vertices of 1200 random quadrats (20 x 20 cm). On 21<sup>st</sup> September 1998, the viable seed bank (VSB) was estimated as follows: 144 intact soil cores were randomly collected, dry residues removed from the surface, driving an 8 cm diameter PVC cylinder at 5 cm of depth into the ground, digging a hole alongside, cutting off the base of the core and lifting it from the ground. Intact soil cores were watered regularly for a month in open-air conditions. The emerged seedlings were counted and classified (readily germinable seeds, RGS). Afterwards, each soil core was mixed with water and washed through two sieves (500 and 200 µm) for separating the mineral component. The organic material was poured into petri dishes on filter paper. The germinable (GSB) and the dormant (DSB) seed bank were estimated identifying and counting the seedlings emerged during the succession of 3 germination/dormancy breakdown treatments (Perez *et al.*, 1998):

- A) 21 day germination test, diurnal cycle of 8/16 darkness/light, T°C regimen of 20/30°C.
- B) 21 day dormancy assessment test, petri dishes at 3 to 5°C.

C) 35 days chemical breakdown test, petri dishes watered with a 600 mg/kg-solution of Gibberellic Acid, diurnal cycle of 8/16 darkness/light and a T°C regimen of 15/35°C. Finally, soil samples were watered for two weeks in controlled conditions to observe eventual further seedling emergences.

Estimate of GSB was obtained by summing RGS and the seedling counts from the A treatment. DSB was estimated by the seedling counts from C treatment. The sum of GSB and DSB represented the total VSB.

## Results and discussion

When collected, the intact soil cores surface showed a total lack of seedlings, demonstrating the absence of summer false breaks. The vegetation composition over the further months showed a good establishment of the introduced species (Fig. 1). In the

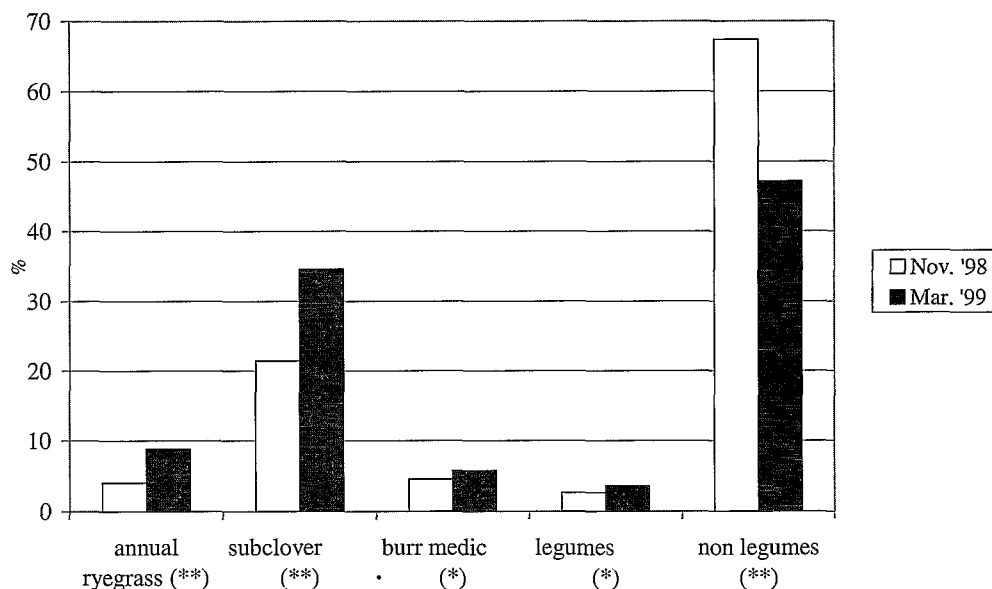


Figure 1 – Overlying vegetation composition. Results of  $\chi^2$  between component percentage contributions for Autumn and Spring are reported: \* = significantly different for  $P < 0.05$ ; \*\* = significantly different for  $P < 0.01$ .

oversowing year particularly, the subclover showed a better establishment capability than burr medic, with respectively about 20 and 10 % of the total composition. The scarce contribution of annual ryegrass was justified by the low sowing rate. The overlying vegetation composition, after the first self-reseeding, evidenced a progressive increase in the presence of the introduced species, moreover of subclover and annual ryegrass, while a reduction in the non-legumes species contribution occurred, allowing to the improvement of the forage quality. Seed bank showed different behaviours of the main components (Fig. 2).

Within each component, the contribution to the GSB ranged from 50% in *Cistus* to near 100% in subclover, demonstrating a ready germination capability in good moist and temperature conditions.

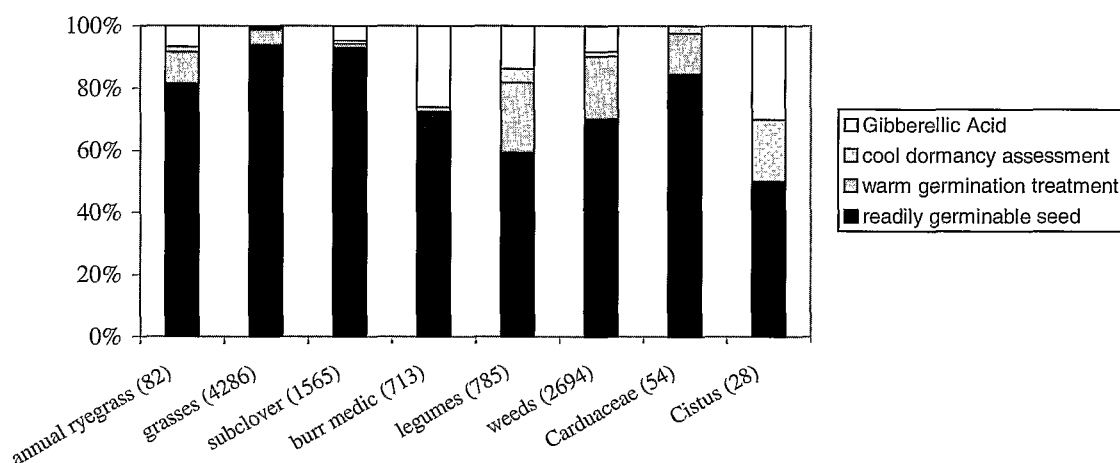


Figure 2 - Viable seed bank: germination/dormancy breakdown treatment. Between parenthesis, the total number of seedlings emerged for each component after the three treatments is reported.

About the 20% of *Cistus* viable seeds germinated after the dormancy assessment treatment (B). Burr medic and *Cistus* seemed to contribute largely to the DSB: more than the 25% of their seeds germinated only after the C treatment. After the dormancy breakdown treatments, only 12 seedlings emerged later in the watered soil samples: 6 of them were burr medics, the other spontaneous legumes. Several vegetation components produced a limited amount of dormant seeds, mainly spontaneous grasses (1%), subclover (5%) and annual ryegrass (7%).

## Conclusions

The viable seed bank density of the 0-5 cm layer was 10,200 seeds  $m^{-2}$ . 'Trikkala' subclover has shown a good establishment response, with an increasing colonisation capability, due to an important contribution to the viable seed bank (15%) with almost all the seeds as readily germinable. 'Anglona' burr medic seemed to establish more hardly but shedding, at the first self-reseeding, about 200 dormant seeds  $m^{-2}$ , a good seed reserve for achieving the species survival. Interesting informations were collected about some weeds persistence capability: more than the 50% of *Cistus* viable seeds is dormant, but many of these (about 40%) germinated when satisfied their cold requirements. The other components of the seed bank did not evidence significant chilling demand.

Further investigations will concern the analysis of the ungerminated seeds, to check the effectiveness of the methodology and to assess the total seed bank, by counting the nonviable seeds and, as a consequence, estimating the potential seed bank contribution of each component. Aimed at studying the mechanisms that regulate the seed bank dynamic, all the estimates will be repeated over years, correlating them with meteorological and topographical data inside the experimental area.

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