



## Microsite manipulation in lowland oak forest restoration results in indirect effects on acorn predation



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### ABSTRACT

Taking advantage of facilitation mechanisms to counteract harsh environmental conditions may prove to be a successful strategy in the restoration of degraded man-made ecosystems. Seed sowing is often a viable and less expensive technique to restore forest cover in environments subjected to heavy anthropic disturbance. In this context, nurse plants and shield objects can improve microsite conditions, enhancing the emergence and growth of seedlings. However, their presence can also create preferred microhabitats for seed predators, and as a result, may alter their movement decisions and foraging behavior. In this study, we investigated whether nurse shrubs and artificial shade structures, with and without previous soil amelioration, could facilitate acorn emergence. We also tested their effects on the predation of acorns by small mammals, within the framework of a larger project aimed at restoring a *Quercus robur* L. forest. The research was conducted in a former roadbuilder's yard located along the Mediterranean Corridor, in Northern Italy. The yard had been in use during the widening of a highway that runs parallel to a stretch of the Corridor. The area was restored in 2014. Mechanical preparation of the soil included two treatments, with and without the addition of compost and zeolite. Acorns were then sown in plots in different microsities, i.e., (1) close to a shrub (*Cytisus scoparius* L., Scotch broom), (2) to the north and (3) south of a shield object (a 30 cm × 30 cm wood particle board), and (4) without any protection element (control). We recorded acorn emergence and predation during the first growing season. At the end of the season, overall acorn emergence was observed to be low, and, surprisingly, was higher in the non-amended soil treatment. The amended soil had a higher temperature and a lower water volumetric content in the summer (JJA). No evidence of direct facilitation on emergence by shield objects or Scotch broom was found, but indirect effects were detected. Acorn predation was generally high (67.45%), but was negatively affected by the presence of *C. scoparius*. The distance of the plot from the rainwater drainage ditch surrounding the study site also influenced winter predation; a larger number of acorns were removed by rodents at longer distances from the drainage ditch. In order to restore degraded sites to lowland oak forests through direct seeding, it is necessary to deal with complex interactions between direct and indirect facilitation mechanisms, competition and predation. Predation was particularly important because of the high predation pressure on acorns.

### 1. Introduction

The ecological restoration of degraded man-made ecosystems usually requires plant establishment and survival in harsh or stressful environments, while limiting restoration costs and further interventions.

Seed sowing is often a viable and less expensive alternative to planting nursery-grown seedlings (Madsen and Löf, 2005), with seeds being easier to transport and manage than bareroot and container

seedlings. Direct seeding also allows the plant root system to develop naturally, thereby avoiding the root malformations that are generally associated with containers (Stanturf et al., 2000; Dey et al., 2008). Instead, the use of container stock can reduce the stress linked to nutrient or water limitation, the risk of poor survival, and number of operations necessary for site preparation and management, e.g. litter removal, mechanical scarification, repeated mowing to control competing ground vegetation (Smith et al., 1997; Dey et al., 2008). Moreover, after germination, seedlings can show slower development rates than planted

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trees (Nyland, 2007).

The early stages of seedling establishment are critical for the regeneration of plant populations. Several mortality factors, including predation, competition and abiotic stress can hamper seed regeneration (Nathan and Muller-Landau, 2000; Castro et al., 2004; Schurr et al., 2004; Davis et al., 2005; Riginos et al., 2005; Kipfer et al., 2009). In addition, excess light, high temperatures and summer drought, which are accentuated in degraded areas without forest cover, can severely limit the survival and growth of seedlings (Castro, 2006; Gómez-Aparicio et al., 2006). Site conditions and mechanisms that operate at a small scale may in particular limit the emergence and performance of seedlings (Collins and Good, 1987).

Taking advantage of intra- or interspecific facilitation mechanisms and the sheltering effects of abiotic elements in natural environments has proven to produce beneficial effects on the survival, initial growth and fitness of the neighbouring seedlings, particularly in climatically stressing sites (Callaway, 2007; Brooker et al., 2008; Marzano et al., 2013). Increasing evidence has pointed out the potential benefits of the application of this nursing effect to improve the success of restoration projects in degraded environments (Padilla and Pugnaire, 2006; Torroba-Balmori et al., 2015).

Nurse plants improve the emergence, survival and growth of seedlings (Castro et al., 2004; Gómez-Aparicio et al., 2004; Gómez-Aparicio et al., 2006; Torroba-Balmori et al., 2015) providing a favourable microhabitat in their surroundings. They offer shade, buffering against high radiation and temperatures, and can increase soil moisture and nutrient content (Callaway, 1995; Rey Benayas, 2005; Legras et al., 2010; Leiva et al., 2015).

In highly degraded systems, and in particular at the grassland stage, nurse objects, such as woody debris or artificial shade-structures can emulate the beneficial effects of nurse plants by reproducing the shaded environments that can be found under their canopies (Gómez-Aparicio et al., 2005; Rey Benayas et al., 2005; Badano et al., 2011).

A key point that influences the recruitment process success is then the interaction among environmental factors that affect regeneration and the behavior of seed dispersal and predation agents (Crawley and Long, 1995; Sunyer et al., 2015). This interaction can actually alter the probability of seed survival and seedling establishment, with the spatial pattern of seed removal usually resulting from predator preferences for certain microhabitats (Clark et al., 1999; Jordano and Schupp, 2000; Pérez-Ramos and Marañón, 2008). Thus, even though nurse plants and shield objects could improve microhabitat conditions and promote seed germination and growth, they could also influence animal behavior, driving seed and seedling predator movements. Their presence, which in particular affects the foraging behavior of animals that usually feed on seeds, could result in increasing predation.

It is well known that wild ungulates and granivorous rodents in Europe can predate a large number of seeds and browse saplings, thus reducing the success of direct seeding in restoration projects (Birkedal et al., 2009; Jinks et al., 2012; van Ginkel et al., 2013). The seed predation rate is strongly affected by habitat complexity, which in turn can influence the abundance of ungulates and rodents. For instance, wild ungulates tend to avoid habitats with complex structure that could hamper their movements, while small mammals prefer to forage in habitats with shrubs and rocks, where they perceive a minor predation risk (Gómez et al., 2003; Orrock et al., 2004; Fedriani and Manzaneda, 2005; Pérez-Ramos et al., 2008; Leverkus et al., 2013). The presence of coarse woody debris in forests usually intensifies the removal of seeds by rodents, and the presence of shrubs and shelters lengthens the time that the rodent spends handling and choosing viable seeds (Perea et al., 2011; van Ginkel et al., 2013).

In the context of a large lowland oak forest restoration project, we have investigated whether the presence of shelter elements can improve acorn emergence and seedling survival of pedunculate oak (*Quercus robur* L.) or whether it can support the movement decisions and foraging behavior of small rodents, by analyzing the main factors that affect

spatio-temporal variations in seed predation.

Since acorn germination in *Quercus* species is conditioned by soil moisture (Pérez-Ramos et al., 2013) and can present delayed emergence (González-Rodríguez et al., 2012), using shrubs (Gómez-Aparicio et al., 2004) and artificial shield objects on planting sites could ameliorate microclimatic conditions, and thus enhance emergence rates (Smit et al., 2008). The large size of oak cotyledons can increase the odds of both regeneration success and seed predation. The latter, which is mainly by small mammals (Gómez et al., 2003; Smit et al., 2008), is usually more intense in the post-dispersal phase and is one of the main factors that endangers oak recruitment. The regeneration performance of oak is related to several variables (Annighöfer et al., 2015). Acorn predation before germination largely determines the success of regeneration (Harmer, 1994), with competition, pests, water supply, light availability and browsing being other variables leading to recruitment failure (Nilsson et al., 1996).

We have hypothesized that improving the physical and chemical conditions of the soil and facilitating seed germination with nurse shrubs and shield objects could influence oak establishment but also the removal of acorns by small mammals. Our main aims were thus to (1) assess the impact of soil amelioration on acorn emergence and predation rate; (2) evaluate the role of enhanced seeding microsites (close to nurse shrubs and shield objects) on the regeneration performance and acorn predation; (3) determine whether the relative position of sowing sites can modify the predation pattern.

## 2. Materials and methods

### 2.1. Study site

The experiment was conducted in a degraded area in northwestern Italy (45°11'38.60" N, 7°50'38.04" E, ca. 190 m a.s.l.). The site is located along the Rail Freight Corridor 6 (Mediterranean Corridor) (European Commission, 2018), linking the southwestern Mediterranean region of Spain and the Hungarian border with Ukraine. The area, which is bordered by a highway, was formerly a roadbuilder's yard. In November 2014, after the road works ended, the area was restored through both seed sowing and seedling planting (2 year old nursery plants), followed by hand sowing with a grassland species mixture. The tree and shrub species used in the experiment were those that usually grow in the lowland oak-hornbeam Mesophytic deciduous forest, i.e., the natural late seral forest ecosystem of the Po Plain, which is dominated by *Q. robur* and *Carpinus betulus* L.

No trees were present when the experiment was started. The soil texture was sandy loam. The pH ranged from 7.41 to 8.08 and soil organic matter content ranged from 1.5% to 3%. The cation exchange capacity (CEC) was lower than 10 meq/100 g.

The climate is temperate, with an annual mean temperature of 11.4 °C and average annual precipitation of 806.2 mm (ARPA Piemonte-Verolengo meteorological station 10 km from the study site, period 1988–2010). The rainfall is not homogeneously distributed during the year, with spring and autumn being the wettest seasons.

A rainwater drainage ditch surrounds the area. At the beginning of the experiment, the area was fenced off against the introduced eastern cottontail (*Sylvilagus floridanus*) and wild ungulates to avoid browsing damage to seedlings and seed predation.

The experimental design (Fig. 1) included two soil treatments (non-amended and amended). In November 2014, the treatments were applied in rows after a mechanical site preparation. Using a tractor the rows were ripped (to a 70 cm depth) and ploughed (to a 40 cm depth). The distance between rows was approximately 2.5 m. Furrows were then formed along the southern edge of the rows. Half of the rows were just ripped and ploughed (non-amended). Since we wanted to improve the organic matter content up to 3% and raise the CEC to 15 meq/100 g, the other rows were ripped, ploughed and amended using 4.2 Mg/ha of compost and 43 Mg/ha of zeolite, with a diameter of 3–8 mm. The

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