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Short communication

Effect of hydroseeding components on the germination of Mediterranean native plant species

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ABSTRACT

The use of native species by hydroseeding has been encouraged for the restoration of degraded Mediterranean areas, but their success is frequently low. Several biotic and abiotic constraints have been reported that partially explain such failure, but the effects of the slurry components on the performance of native species have not been evaluated so far. We used germination tests to assess the effects of four such components – bacteria-based fertilizing agent, biostimulant, dye and surfactant – on the germination of 12 native species used in hydroseeding of quarry slopes.

Hydroseeding components significantly affected germination only in four species. Bacteria-based fertilizing agent, biostimulant and a mixture of the four components increased the germination percentage of *Thymus mastichina* while bacteria-based fertilizing agent and biostimulant decreased the germination speed of *Cistus albidus*. Component- and species-specific effects on time to germination were observed in *Bituminaria bituminosa*, *C. albidus*, *Helichrysum stoechas* and *T. mastichina*.

The results showed that only a few of the native species studied were affected by a specific hydroseeding component. Therefore, poor establishment of native species observed in the field could not be wholly attributed to negative effects of those components on their germination. These might have contributed to failure in the field by further reducing the low germination speed of certain species. This study suggests that a better knowledge of species characteristics and species-specific responses to hydroseeding components may help to improve the success and the cost-effectiveness of this restoration procedure.

1. Introduction

Hydroseeding is frequently used in restoration of arid and semi-arid degraded areas such as mining wastes (Albaladejo-Montoro et al., 2000; Martínez-Ruiz et al., 2007) or road slopes (Tormo et al., 2006; Bochet et al., 2007; Matesanz and Valladares, 2007; García-Palacios et al., 2010), but also in degraded areas under sub-humid Mediterranean climate (Brofas et al., 2007; González-Alday et al., 2008; Alday et al. 2012; Oliveira et al., 2012). This technique involves sparring a homogeneous slurry of seeds, fertilizer, binder, mulch and other components (e.g. moisture retention agents, plant-growth promoters) over large and/or inaccessible areas. The use of native species instead of fast-growing commercial species has been proposed, because the latter strongly depend on irrigation, which is incompatible with sustainable water use in arid and semi-arid environments, and can outcompete native species and constrain vegetation dynamics in the long-term (Matesanz and Valladares, 2007; García-Palacios et al., 2010).

Species performance at the germination stage is expected to determine the success of hydroseeding. Bochet et al. (2007) demonstrated that species able to germinate earlier and at high germination rates colonise road slopes more successfully than species with low germination rates, especially during periods of low soil water availability. However, most studies that evaluate

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the success of native seed mixtures focus on plant establishment (Albaladejo-Montoro et al., 2000; Tormo et al., 2006; Brofas et al., 2007; Matesanz and Valladares, 2007; Martínez-Ruiz et al., 2007; González-Alday et al., 2008; García-Palacios et al., 2010) rather than on germination (Merlin et al., 1999; Bochet et al., 2007). Several studies reported failures in hydroseeding of Mediterranean native species, pointing out competition from fast-growing species, microsite suitability, and season of application as the main factors limiting their establishment (Matesanz and Valladares, 2007; García-Palacios et al., 2010; Oliveira et al., 2013).

Low establishment of native species introduced through hydroseeding was also recorded after a restoration project in a Mediterranean limestone quarry in southwestern Portugal (Oliveira et al., 2007). The germination performance of single species under standard conditions did not fully explain such a failure (Oliveira et al., 2012), and early seedling mortality, shortage of microsites suitable for seed germination (Tormo et al., 2006), and/or negative effects of substrate or slurry components on seed germination were considered the most probable causes for the low establishment of the native species in this case.

Hydroseeding slurries may include a number of components such as wood fibre mulches, organic amendments and fertilizers (Sheldon and Bradshaw, 1977; Albaladejo-Montoro et al., 2000; Brofas and Varelides, 2000; Martínez-Ruiz et al., 2007; Woods et al., 2012; Oliveira et al., 2013), tackifiers, stabilizers and surfactants (Merlin et al., 1999; Albaladejo-Montoro et al., 2000; Brofas et al., 2007; Martínez-Ruiz et al., 2007; Oliveira et al., 2013), dyes (Oliveira et al., 2013), natural biostimulants containing seaweed extracts (Oliveira et al., 2013), and plant growth-promoting bacteria (Brofas and Varelides, 2000; Martínez-Ruiz et al., 2007; Oliveira et al., 2013). Some studies provided evidences that mulches and organic amendments increase the efficiency of hydroseeding in the restoration of degraded areas, for example by improving soil properties, nutrient levels and water conditions, controlling soil erosion and increasing plant cover and biomass (Roberts and Bradshaw, 1985; Brofas and Varelides, 2000; Albaladejo-Montoro et al., 2000). In contrast, the effects of slurry components on seed germination are addressed only occasionally (Luken, 1990) and the results obtained indicate species, component and/or dosage specific-effects: stabilizers have either no effect on seed germination (Roberts and Bradshaw, 1985; Brofas et al., 2007) or species-specific inhibitory effects (Brofas and Varelides, 2000); mulches and organic amendments can promote seed burial and germination (Roberts and Bradshaw, 1985) while fertilizers may inhibit germination (Sheldon and Bradshaw, 1977; Roberts and Bradshaw, 1985); and biostimulants are known to enhance the germination of crop species only (Hernández-Rodríguez et al., 2010; Kumar and Sahoo, 2011). Interactions among slurry components are not taken into account when testing the effects of a specific component on seed germination with a single application. For instance, mulch can reduce the inhibitory effect of fertilizers on seed germination (Sheldon and Bradshaw, 1977) but such an effect varies with mulch type (Luken, 1990). Stabilizers can also reduce the positive effects of mulches (Sheldon and Bradshaw, 1977).

In this study, we evaluated the germination performance of Mediterranean species in response to components of the hydroseeding slurry using germination tests in the laboratory. The main goal was to determine if those components, currently used to hydroseed quarry slopes, affected the germination of 12 native species used in experimental hydroseedings made at that same quarry. Improved knowledge on this subject is expected to support both the selection of hydroseeding components and of native species in future restoration projects.

2. Materials and methods

The twelve native species used in this study had been previously used in experimental hydroseedings at SECIL limestone quarry in SW Portugal (38.49615° N, −8.95097°W) (Oliveira et al., 2012; unpublished data): *Anthyllis vulneraria* L. (Fabaceae), *Bituminaria bituminosa* (L.) C.H.Striton (Fabaceae), *Cistus albidus* L. (Cistaceae), *Coronilla glauca* L. (Fabaceae), *Dactylis glomerata* L. (Poaceae), *Helichrysum stoechas* (L.) Moench (Asteraceae), *Ononis natrix* L. (Fabaceae), *Origanum vulgare* L. (Lamiaceae), *Sanguisorba minor* Scop. (Rosaceae), *Sedum sediforme* (Jacq.) Pau (Crassulaceae), *Thymbra capitata* (L.) Cav (Lamiaceae) and *Thymus mastichina* L. (Lamiaceae). Seeds were obtained from two commercial seed suppliers (Semillas Silvestres and Corporación Zulueta), except for *H. stoechas*. Seeds of this species were provided by Seed Bank A.L. Belo Correia (Jardim Botânico, Museu Nacional de História Natural e da Ciência, Lisbon). All seeds were stored for two months in the Seed Bank drying room (16 °C, 30% RH) until used.

Four hydroseeding liquid components were tested, in the same concentrations as commonly used at the quarry: a bacteria-based fertilizing agent (Finn MB, FINN ™, 0.2642 ml/L), a seaweed extract and amino acid-based biostimulant (Pronto, Priya Chemicals™, 0.3816 ml/L), a green dye (Regreen, BoskFarma™, 0.8807 ml/L) and a surfactant (Agri2, Bosk™, 0.0396 ml/L).

The effects of hydroseeding components on germination percentage, germination speed, and time to first germination of the selected species were evaluated using standard germination tests in incubators. The hydroseeding components were applied in six treatments. Four treatments consisted of a solution of each component in the same concentrations as used at the quarry: Bacteria-based fertilizing agent (Bac), Biostimulant (Bio), Dye (Dye), Surfactant (Sur). An additional treatment consisted of a mixture of those four components (Mix), each at the same concentration as in the single-component treatment. Distilled water served as control (Con).

Each treatment/species was replicated six times, each replicate consisting of 30 seeds placed in a 9 cm diameter Petri dish on a filter paper moistened with 3 ml of the correspondent component solution at the beginning of the experiment. Only plump seeds of each species were used for germination tests, and those from species exhibiting physical seed dormancy (*C. glauca*, *B. bituminosa*, *O. natrix* and *C. albidus*) were previously scarified using sand paper.

Petri dishes were placed in a temperature-controlled incubator (Fitoclima S600, Aralab, Lisbon) with 12 h light at 18 °C and 12 h dark at 9 °C. Incubation temperatures correspond to mean maximum and mean minimum temperatures for November, March and April, the months when hydroseeding is usually performed at the quarry (SECIL's local meteorological data averaged over the 2000–2007 period). Germination was daily recorded during the first week, and every two or three days afterwards, over 35 days. Radicle emergence (approximately 1 mm) was the criterion used for scoring a seed as germinated. Germination percentage was calculated as the number of germinated seeds at the end of the test/(number of initial seeds-number of empty seeds)×100. Germination speed (T_{50}) was calculated as the number of days to achieve 50% of germination percentage, and time to germination as the number of days until the first germination occurred.

The effects of treatments on germination percentage, T_{50}, and time to germination were analysed with one way ANOVA using treatments as a fixed factor. Data were transformed whenever assumptions of homogeneity of variance and normality were not met (germination percentages were arcsine transformed, and T_{50} and time to germination were log transformed). When assumptions were not met after data transformation, a Kruskal–Wallis test was performed. When a significant effect of treatments was found,