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Establishing a semi-natural grassland: Effects of harvesting time and sowing density on species composition and structure of a restored *Arrhenatherum elatius* meadow

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ABSTRACT

Species-rich semi-natural grasslands play an essential role in the conservation of European biodiversity. To restore them, existing grasslands may be used as a seed source. A key factor for successful restoration is the transfer of all species of the plant community to the site being restored. This approach, however, often poses practical problems due to variations in species phenology, so that only time-staggered harvesting allows the seeds of many species to be collected. Poor harvesting methods may reduce the number of species transferred from the donor to recipient site. The effects of harvesting at various time points by various methods were evaluated here in a restoration experiment on an ex-arable field in Northern Italy. Propagation materials from the first or second regrowth (or both) of a meadow dominated by *Arrhenatherum elatius* were collected by four harvesting methods. The materials were spread at the sowing density between 830 and 14360 seeds m^{-2}. Species composition and structure of the vegetation were examined during 6 years. Untransferred species were almost exclusively those not present at harvesting as mature seed. Compared with materials from one regrowth period, materials from both regrowth periods significantly increased the number of transferred species per plot (26.5 vs. 28.5, respectively) and the absolute transfer rate (64% vs. 75%). Higher sowing density yielded a greater number of positive than negative effects. It favored stable establishment of donor site species and significantly reduced the presence of weeds. Nonetheless, due to the initial dominance of species with high sowing density, evenness of the restored plots was lower than that at the donor site but increased with time, at least under low- and medium-sowing-density conditions. Multiple harvesting at time points when several species with mature seed are present increases the species transfer rate. Medium sowing density positively affects restoration development because it promotes the establishment of donor site species with lower sowing density, prevents species with the highest density from dominating the vegetation during the first few years, and reduces weed cover.

1. Introduction

Species-rich semi-natural grasslands of agricultural origin are necessary for conservation of European biodiversity (EEA, 2004). Their reduction and deterioration due to abandonment or agricultural intensification are among the recent negative environmental changes in Central Europe. Therefore, in addition to protection of still existing grasslands, restoration of degraded grasslands has become increasingly important for conservation of biodiversity (Carter and Blair, 2012).

Spontaneous establishment of grassland species is hindered by environmental and anthropogenic factors. High rates of species immigration to degraded sites through natural seed dispersal are favored only in regions with high species richness (MacArthur and Wilson, 1967). Species that exist in highly diverse grasslands are poorly represented in the seed bank of soils that have been cultivated for long periods (Stevenson et al., 1995).

As an alternative to seed immigration from nearby vegetation, addition of seeds produced at agricultural cultivation sites or harvested from grasslands has been used successfully to restore species-rich grasslands (e.g. Graiss et al., 2013; Hölzel and Otte, 2003; Kiehl and Wagner, 2006; Prach et al., 2014). In this regard, the important ecological and technical problems involve species richness, sowing density (SD), and the proportions of species in the propagation material. Species richness plays an important role in

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restoration of a grassland with increased ecosystem services, but establishment of all target species has never been observed (Camill et al., 2004; Kiehl et al., 2010). Predictions regarding the effects of SD on species richness in grasslands can be contradictory (Münzbergová, 2012). At less productive sites, higher SD may enhance the establishment of all added species, but at more productive sites, it can also reduce species richness by giving an advantage to more competitive species. These predictions turned out to be true in restoration experiments with tallgrass prairies in North America (e.g. Dickson and Busby, 2009), but were not always confirmed in European grasslands (Münzbergová, 2012; Stevenson et al., 1995). At least during the first few years, SD and species proportions in the seed mixture were found to affect relative abundance levels of species in the restored grasslands in the Czech Republic (Münzbergová, 2012).

Added seeds can be collected from single-species cultivation sites (Steinauer, 2003) or from species-rich grasslands for restoration purposes. The second approach is easy when sufficient areas of species-rich grasslands are present in the region in question. Seeds of some species from agricultural cultivation sites are not available in all biogeographical regions (areas of relatively homogeneous ecological conditions with common characteristics: Directive 2007/2/EC) (Kiehl et al., 2010). Moreover, they are often used outside their provenance region; consequently, these species can now be found in areas where they have never been found previously (Rometsch, 2009).

Arrhenatherum elatius hay meadows represent important grassland vegetation in Central Europe, where they have been extensively mown as a key source of herbage with considerable species richness (Rodwell et al., 2007). Due to agriculture intensification, they are now much less common than they had been until the 1970s, to the extent that they are currently regarded as plant communities to be protected and restored (Dietl, 1995). Typically, *A. elatius* meadows are mown two to three times a year. Plant phenology must therefore match the sequences of light and shade generated (Ellenberg, 1978). Most grasses of these meadows flower in late spring, from May to June (Schneider, 1954). Legumes show the greatest growth and flowering during the second regrowth. Some herbs flower only during the second regrowth (Ellenberg, 1978).

Thus, the species composition and the proportions of seeds in propagation material cannot be changed easily when an *A. elatius* meadow is used as the seed source for grassland restoration. To some extent, however, these factors can be adjusted via decisions regarding:

+ regrowth and the time point within this period when the seeds are harvested: harvesting in more than one regrowth period allows more species to be transferred; changing the relative amounts of propagation materials from different regrowth periods also allows for modification of single-species percentages in the seed mixture;

+ the amounts of propagation materials: changing them alters the SD, and if propagation materials are harvested at different time points, also alters the species percentages in the seed mixture;

+ the harvesting method: because available protocols vary in efficiency in terms of both species amounts and proportions of harvested seeds (Scotton et al., 2009a), the method used may change both SD and species percentages.

Here, by harvesting of seeds from a species-rich grassland and using them directly for restoration, we conducted an experiment to optimize restoration of an *A. elatius* hay meadow. The specific questions addressed were as follows:

+ What are the effects of spreading the propagation materials from two harvesting regrowth periods as opposed to the materials from only one regrowth period?

+ How can SD influence the species richness, composition, and structure of the restored vegetation?

+ Do harvesting methods affect restoration results?

2. Materials and methods

2.1. The restoration trial and field surveys

Donor and recipient sites (Vicenza, Northeast Italy) were 11 km apart from each other. The mean annual rainfall (1177 and 1266 mm year^{-1} at the recipient and donor site, respectively) was well distributed across the four seasons. The mean annual temperature was 13.3 °C at the recipient site (79 m above the sea level [a.s.l.]) and 11.1 °C at the donor site (435 m a.s.l.). At both sites, soils derive from a calcareous substratum and have sub-alkaline pH. The recipient site (Maragnole, high plain, zero slope) was cultivated with maize before 2008 (the first year of the experiment). It had a clay loam soil (to the depth of 50 cm) containing 2.23% of organic matter, 27.3 mg kg^{-1} Olsen P, and 257 mg kg^{-1} exchangeable K. The main weeds that were present at the beginning of the experiment were *Cynodon dactylon*, *Sonchus asper* and *Capsella bursa-pastoris*. The main agricultural practice in the area was dairy farming. Semi-natural hay meadows have traditionally performed an important function in the conservation of biodiversity, but they currently occur only as intensively used, species-poor grasslands. Nevertheless, species-rich meadows still existed in the nearby calcareous Prealps, where the donor site (Pianari) was located. The 20% sloping meadow was mown two to three times per year and was not fertilized much. The soil was 31 cm deep and composed of loamy fine sand. It contained 8.1% of organic matter and 8.9 and 133 mg kg^{-1} Olsen P and exchangeable K, respectively. The vegetation was a poor dry-soil form of *A. elatius* meadows (*Arrhenatherion elatioris* Koch 1926), with 63 species found within the area of 1800 m^2. During the first regrowth, the grasses, particularly *A. elatius* and *Trisetum flavescens*, were dominant (relative cover of 64%). Legumes (especially *Trifolium pratense*, *Onobrychis viciifolia*, and *Lotus corniculatus*) constituted 16% of the total cover, and forbs (mainly *Rhinanthus freynii* and *Galium album*) the residual 20%.

At the recipient site, 3000 m^2 were ploughed in November 2008 and rotary-hoed in March 2009. The surface was then subdivided into three blocks of eight plots 10 × 10 m each. The weeds that grew after the hoeing were chemically controlled with two herbicide treatments (May and July 2009: glyphosate, application rate 3 kg ha^{-1}).

At the donor site, three blocks of four plots (10 × 10 m each) were marked in March 2009. In June (the end of the first regrowth), seeds were harvested by the following methods:

+ on-site threshing (with a wheat thresher);

+ seed-stripping (by means of a pull-type stripper with downward brush rotation covering the full vegetation height);

+ harvesting as green hay (mowing and immediate manual collection);

+ harvesting as dry hay (mowing, one manual turning on the following morning, and baling in the afternoon on the same day).

The resulting propagation materials were hay-flower from threshing (OST) or seed stripping (SS), green hay (GH), and dry hay (DH). GH and DH harvesting was repeated in July 2009 (the end of the second regrowth). OST, SS, and DH were dried and stored until September. For each plot, two samples of propagation material were analysed for seed content in the following months by manual separation of mature seeds to the species level. The amounts of each sample corresponded to a donor site area of 1 m^2, i.e. 10.8, 15.2, 644, 361, 971, and 322 g for OST1, SS1, GH1, DH1, GH2, and DH2, respectively.