

# Species introduction in restoration projects – Evaluation of different techniques for the establishment of semi-natural grasslands in Central and Northwestern Europe

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## Abstract

During recent decades, many studies have shown that the successful restoration of species-rich grasslands is often seed-limited because of depleted seed banks and limited seed dispersal in modern fragmented landscapes. In Europe, commercial seed mixtures, which are widely used for restoration measures, mostly consist of species and varieties of non-local provenance. The regional biodiversity of a given landscape, however, can be preserved only when seeds or plants of local provenance are used in restoration projects. Furthermore, the transfer of suitable target species of local provenance can strongly enhance restoration success.

We review and evaluate the success of currently used near-natural methods for the introduction of target plant species (e.g. seeding of site-specific seed mixtures, transfer of fresh seed-containing hay, vacuum harvesting, transfer of turves or seed-containing soil) on restoration sites, ranging from dry and mesic meadows to floodplain grasslands and fens. Own data combined with literature findings show species establishment rates during the initial phase as well as the persistence of target species during long-term vegetation development on restoration sites.

In conclusion, our review indicates that seed limitation can be overcome successfully by most of the reviewed measures for species introduction. The establishment of species-rich grasslands is most successful when seeds, seed-containing plant material or soil are spread on bare soil of ex-arable fields after tilling or topsoil removal, or on raw soils, e.g. in mined areas. In species-poor grasslands without soil disturbance and on older ex-arable fields with dense weed vegetation, final transfer rates were the lowest. For future restoration projects, suitable measures have to be chosen carefully from case to case as they differ considerably in costs and logistic effort. Long-term prospects for restored grassland are especially good when management can be incorporated in agricultural systems.

## Zusammenfassung

Während der letzten Jahrzehnte haben zahlreiche Untersuchungen gezeigt, dass die erfolgreiche Wiederherstellung artenreicher Graslandvegetation vielerorts durch die mangelnde Verfügbarkeit von Diasporen limitiert wird, da Zielarten in der

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Samenbank von Renaturierungsflächen fehlen und die Diasporenausbreitung in fragmentierten Landschaften erschwert ist. Handelsübliche Saatmischungen, die in Europa häufig im Rahmen von Begrünungsmaßnahmen verwendet werden, enthalten oftmals Arten oder Unterarten nicht-heimischer Herkunft. Die Biodiversität einer Region kann jedoch nur erhalten werden, wenn im Rahmen von Renaturierungsmaßnahmen gebietseigenes Saatgut oder Pflanzenmaterial verwendet wird. Durch die Einbringung standortangepasster Zielarten gebietseigener Herkunft kann der Renaturierungserfolg deutlich gesteigert werden.

Die vorliegende Veröffentlichung analysiert und bewertet unterschiedliche Verfahren zur Ansiedlung von Zielarten (z. B. Ansaat mit gebietsheimischem Saatgut, Übertragung von Mahd- oder Sauggut, Übertragung von Soden oder samenhaltigem Oberboden) im Rahmen der Wiederherstellung artenreicher Graslandvegetation (von Trocken- und Halbtrockenrasen bis hin zu Feuchtgrünland und Niedermoor). Eigene Daten in Kombination mit Literaturangaben dokumentieren sowohl die Artentransferraten in der Initialphase der durchgeführten Maßnahmen als auch die langfristigen Etablierungsraten der eingebrachten Zielarten. Zusammenfassend zeigen die Ergebnisse, dass die meisten der untersuchten Artentransfermaßnahmen das Problem der Diasporenlimitierung erfolgreich lösen. Die Etablierung artenreicher Graslandvegetation ist am erfolgreichsten, wenn Saatgut oder samenhaltiges Pflanzen- oder Bodenmaterial auf offenem Boden ehemaliger Äcker (nach Bodenbearbeitung oder Bodenabtrag) oder auf Rohböden (z.B. auf Abgrabungs- oder ehemaligen Bergbauflächen) ausgebracht werden. Bei der Ansiedlung in bestehenden artenarmen Grasländern oder auf älteren Ackerbrachen mit dichter Vegetation waren die Etablierungsraten der eingebrachten Arten am niedrigsten.

Für künftige Projekte zur Wiederherstellung artenreicher Graslandvegetation müssen die jeweils geeigneten Verfahren sorgfältig ausgewählt werden, da sie sich hinsichtlich des logistischen Aufwands und der Kosten deutlich unterscheiden. Die langfristigen Perspektiven für neu angelegte Grasländer sind besonders gut, wenn notwendige Managementmaßnahmen in landwirtschaftliche Systeme eingebunden werden können.

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## Introduction

Species-rich grasslands have been in constant decline in many parts of Europe during the past decades due to abandonment, afforestation, drainage, conversion into arable land and intensification of management (Poschlod & WallisDeVries 2002). This is particularly true in Northwestern and Central European countries with intensive agriculture such as Great Britain, The Netherlands, Belgium, France and Germany. Therefore, the restoration (i.e. the assistance of recovery; SER 2004) of species-rich semi-natural grasslands is a top priority of nature conservation activities in these countries since the 1970s (e.g. Bakker 1989, Walker et al. 2004). Early restoration efforts were often focused on the removal of nutrients, rewetting and management optimization. In many cases, such measures alone were unsuccessful and did not lead to the re-establishment of target communities although environmental conditions were favourably changed (Bakker & Berendse 1999; Walker et al. 2004). Numerous studies identified the lack of viable seeds in the soil seed bank and limited dispersal of target species as the main obstacles to restoration of species-rich grasslands (Bakker, Poschlod, Strykstra, Bekker, & Thompson 1996; Bossuyt & Honnay 2008; Bullock et al. 2002). Only few target species build up long-term persistent soil seed banks (Thompson, Bakker, & Bekker 1997; von Blanckenhagen & Poschlod 2005) and seed banks are usually depleted after periods of arable use or intensive grassland management (e.g. Bakker et al. 1996; Bekker et al. 1997; Bissels, Donath, Hölzel, & Otte 2005;

Hölzel & Otte 2004). Thus, species have to immigrate from source populations in the surroundings of restoration sites. Even in cases, where such remnant populations are present, the high degree of landscape fragmentation and lack of dispersal vectors such as flooding, wandering livestock and haymaking can hamper effective re-colonisation (Bakker et al. 1996; Poschlod & Bonn 1998). Furthermore, many grassland species show a low potential for long-distance dispersal (Bischoff 2002; Donath, Hölzel, & Otte 2003; Hutchings & Booth 1996).

Therefore, the active introduction of plant species of local provenance via propagule transfer is essential as a supplementary measure, when target species are absent on restoration sites or on neighbouring sites. During the past decades, various methods for propagule transfer have been tested over a broad range of habitats. Although such methods are increasingly used in restoration practice, it is difficult to evaluate the success of different techniques because information is scattered. The review of Walker et al. (2004) evaluated the effectiveness of measures for the restoration of species-rich lowland grassland on formerly agriculturally improved sites in the UK with a particular emphasis on abiotic constraints and management issues. In contrast, we will review the efficiency of various species transfer methods for the establishment of species-rich grasslands with habitat-specific species composition (i.e. a high proportion of target species) and summarise current knowledge from studies in Central and Northwestern Europe.

## Selection of studies and evaluation of restoration success

We reviewed the available literature on restoration of grasslands by measures of species introduction in Central and Northwestern Europe and complemented the information by own unpublished data and by additional unpublished data that were provided by authors of the included studies. For two techniques, seeding of site-specific seed mixtures and hay transfer (see below), it was possible to compile data in tables for systematic comparison. In general, publications documenting only a short time span of 1–2 years or focussing only on specific topics such as hay yield or soil parameters were excluded.

For seeding, only studies using seeds of local provenance with detailed information on composition of seed mixtures and establishment rates of sown species were selected (Table 1). For hay transfer, only studies that contained information about source communities, the amount of applied plant material and the number of transferred species, i.e. species from source communities with viable seeds in the hay, which were detected at least once during the evaluation period on receptor sites were included (Table 2).

In the literature, a broad variety of similarity measures and indices for the evaluation of restoration success complicates comparisons between studies. While some studies used species-pool data from the literature to calculate saturation indices for the evaluation of restoration success (e.g. Klimkowska, van Diggelen, Bakker, & Grootjans 2007; Wolters, Garbutt, & Bakker 2005), we decided to use vegetation data from source communities and – if available – from germination tests to calculate transfer rates of hay-transfer species. The absolute transfer rate is calculated as the percentage of transferred species in relation to the total number of species of the donor site. The relative transfer rate gives the percentage of transferred species in relation to the number of potentially transferable species (with viable seeds in the applied plant material). When data were available, transfer rates were calculated not only for all plant species of the donor sites but also for habitat-specific target species.

The available literature on other restoration techniques, such as vacuum harvesting, the transfer of seed-enriched chaff, threshing and brush harvesting, the transfer of raked material, turves and seed-containing soil, was more scattered and heterogeneous so that the data could not be compiled in tables. Nevertheless we give data on transfer and/or establishment rates of introduced plant species in the text to allow a comparison and evaluation of the different techniques. In our final evaluation we compared data on numbers of transferred/established plant species and on establishment rates of the introduced species, because these were the data given in most of the studies.

## Techniques for the establishment of plant communities

### Seeding of site-specific seed mixtures

Commercially produced mixtures comprising genetically uniform and optimized seeds for agriculture or gardening are commonly used for the restoration or re-creation of grasslands across Europe (Kirmer & Tischew 2006). As these seed mixtures are mostly propagated abroad (e.g. East Asia, the Balkans, New Zealand) they often contain foreign ecotypes, sub-species and even foreign species (Frank & John 2007; Marzini 2004), which may threaten local or regional genetic diversity. Several studies indicated that the introduction of foreign ecotypes can lead to higher failure rates in recruitment compared with seeds of local provenance (Bischoff & Müller-Schärer 2005; Hufford & Mazer 2003; Van der Mijnsbrugge, Bischoff, & Smith 2010). In addition, hybridisation between local and non-native genotypes may dilute native gene-pools and reduce the fitness of subsequent hybrid populations (Keller & Kollmann 2000; McKay, Christian, Harrison, & Rice 2005). Therefore, it is essential in ecological restoration to use local genotypes to preserve the genetic integrity of local populations (e.g. Sackville Hamilton 2001; Walker et al. 2004). One way to reach this goal is to define “seed zones” for collection and propagation based on geographical and climatic conditions (Hacker & Hiller 2003; Van der Mijnsbrugge et al. 2010). In practice, delineation of seed zones is quite difficult to achieve since the area should be small enough to maintain local ecotypes of very different species and large enough to allow economic seed propagation.

The composition of seed mixtures for restoration projects essentially depends on the local preconditions (e.g. climate, soil, water and nutrient availability). The mixtures tested in the reviewed studies contained between 15 and 51 species with grasses as matrix species and habitat-specific forbs (Table 1). Lepš et al. (2007) showed that mixtures of 15 species were more successful than mixtures of only 4 species. In Germany, the most common commercial seed mixtures contain only 4–6 grass species with a recommended seeding quantity of 20 g/m<sup>2</sup> corresponding to more than 20,000 grass seeds/m<sup>2</sup> (Stolle 2006a). This produces high competitive pressure regarding space, water and nutrients, especially on nutrient-limited sites. In contrast, Lindborg (2006) recommended a seeding density of 2100 and 4600 seeds/m<sup>2</sup>, respectively, in a study with hand-collected seeds of local provenance. In practical studies with site-specific seed mixtures, a seeding quantity of 1–5 g seeds/m<sup>2</sup> of site-adapted plant species was generally sufficient (Table 1).

In most studies 80–100% of sown species emerged on receptor sites, but the establishment rate at the end of the respective observation period (3–21 years) ranged between 32% and 96% (Table 1). Especially in species-poor grasslands, success is dependent on site preparation since less

**Table 1.** Evaluation of seeding of site-specific seed mixtures. Data on established species represent numbers or percentages of vascular plant species. The percentage of established species gives the proportions of species from the seed mixture that were detected at least once within the evaluation period on receptor sites. The final establishment rate is the percentage of successfully established species from the seed mixture at the end of the observation period. In addition, the percentage of established species from the seed mixture in relation to total cover is given for the end of the observation periods. g.: grasses, h.: herbs.

Target community	Pre-restoration state of receptor site	Size of receptor site (m <sup>2</sup> )	Amendments or site preparation	Management on restoration sites	No. of species in seed mixture	No. of seeds in mixture (seeds/m <sup>2</sup> )	Seeding rate (g/m <sup>2</sup> )	Established species (%)	Final establishment rate (%)	% Cover of species from seed mixture	Study period (years)	References
<b>Ex-arable fields without topsoil removal</b>												
Flood meadow ( <i>Cnidion</i> )	Former cereal field	1,300,000	Ploughing, harrowing	Mowing once	11	–	–	82	82	–	3	Šeffer et al. (1999)
MG5a: <i>Centaureo–Cynosuretum</i>	Former cereal field	100	Deep ploughing (30–40 cm)	Mowing once, grazing	39	–	2.8	74	59	–	8	Pywell et al. (2002, unpubl.)
<i>Mesobromion</i>	Former cereal field	30,000	Ploughing, harrowing	Mowing once	27	–	2	96	96	82–85	5	Jongepierová et al. (2007, unpubl.)
CG3: <i>Bromus erectus</i> grassland	Former cereal field	50	Herbicide, rotovating	Mowing once	17	–	10.5	82	53	–	21	Pakeman, Pywell, and Wells (2002)
CG3b: <i>Bromus erectus</i> grassland	Former cereal field	100	Deep ploughing (30–40 cm)	Mowing once, grazing	41	–	2.7	75	–	–	4	Pywell et al. (2002, unpubl.)
Not specified	Ex-arable field (CZ)	100	Not specified	Mowing twice	15	2500 (g.) 1000 (h.)	–	93	80	c. 70	7	Lepš et al. (2007)
Not specified	Ex-arable field (NL)	100	Not specified	Mowing once	15	2500 (g.) 1000 (h.)	–	100	87	c. 85	7	Lepš et al. (2007)
Not specified	Ex-arable field (UK)	100	Not specified	Mowing once	15	2500 (g.) 1000 (h.)	–	100	100	c. 95	7	Lepš et al. (2007)
<b>Species-poor grassland with complete turf destruction</b>												
MG5: <i>Centaureo–Cynosuretum</i>	Species-poor grassland	700	5–10 cm deturfing	Mowing once	18	–	1	–	62	–	4	Pywell et al. (2007, unpubl.)
MG6 <i>Lolio–Cynosuretum</i>	Permanent pasture	1000	Herbicide	Four grazing/ mowing regimes	28	–	–	89	89	–	3	Coulson et al. (2001)
<b>Species-poor grassland with soil disturbance</b>												
MG5: <i>Centaureo–Cynosuretum</i>	Species-poor grassland	700	5–10 cm harrowing	Mowing once	18	–	1	–	32	–	4	Pywell et al. (2007, unpubl.)
<b>Mining areas and land fills</b>												
<i>Arrhenatheretum elatioris</i>	Disturbed loess (mined site)	3800	Fresh mulch (ca. 1 kg/m <sup>2</sup> )	Mown in 2007	21	496 (g.) 364 (h.)	2	100	81	96	7	Kirmer (2006, unpubl.)
<i>Arrhenatheretum elatioris</i>	Sandy loam (mined site)	1000	Fresh mulch (ca. 1 kg/m <sup>2</sup> )	No management	51	1130 (g.) 1320 (h.)	3.5	88	69	88	3	Stolle and Kirmer (2006, unpubl.)
<i>Dauco-Melilotion/Onopordion acanthii</i>	Loamy-clayey sand (mined site)	10,000	Hay mulch (ca. 1 kg/m <sup>2</sup> )	No management	51	3774 (g.) 914 (h.)	2	88	28	70	10	Stolle (2006b, unpubl.)
<i>Dauco-Melilotion/Onopordion acanthii</i>	Sandy silt (land fill)	10,000	Fresh mulch (ca. 1 kg/m <sup>2</sup> )	No management	23	586 (g.) 2499 (h.)	1	96	57	33	11	Stolle (2006c, unpubl.)
Not specified; probably calcareous grassland	Alkaline (waste heaps)	6 × 1 m <sup>2</sup>	–	Not specified, probably no management	36–41	10,000	–	100	50	–	6	Ash et al. (1994)
Not specified; probably calcareous grassland	Raw calcareous subsoil (mined site)	–	–	No management	26 herbs	260	–	–	50	–	5	Wathern and Gilbert (1978)

**Table 2.** Evaluation of species transfer by fresh seed-containing hay for different vegetation types on restoration sites with and without topsoil removal or soil disturbance. Data on transferred species represent numbers or percentages of vascular plant species (in parentheses: target species). Transferred species are those species with viable seeds in the hay, which were detected at least once within the evaluation period on receptor sites. The transfer rate was calculated as the percentage of transferred species in relation to the total number of species of the donor site. The relative transfer rate was calculated as the percentage of transferred species in relation to the number of potentially transferable species (with viable seeds in the applied plant material). The final establishment rate was calculated as the percentage of transferred species present at the end of the study period in relation to the total number of species of the donor site. For studies with different transfer rates due to variation between restoration fields, means ( $\bar{x}$ ) are given. Abbreviations: tsr: topsoil removal depth; f.wt.: fresh weight.

Grassland type, target community	Pre-restoration state; tsr	Management after restoration	Size of receptor site (m <sup>2</sup> )	Dry weight of plant material (g/m <sup>2</sup> )	No. of viable diaspores (per m <sup>2</sup> )	No. of transferred species	Transfer rate (%)	Relative transfer rate (%)	Final establishment rate (%)	Study period (years)	References
<b>Ex-arable fields without topsoil removal</b>											
Floodplain meadows, <i>Cnidion</i>	Ex-arable fields	Mulching	1500–3500	750–1500	208–5175	15–64; $\bar{x}$ 39 (3–21; $\bar{x}$ 12)	21–78; $\bar{x}$ 53 (8–64; $\bar{x}$ 36)	18–91; $\bar{x}$ 66 (0–86; $\bar{x}$ 57)	9–67; $\bar{x}$ 48 (3–52; $\bar{x}$ 30)	4	Hölzel et al. (2006), Donath et al. (2007)
Mesic to wet meadows, <i>Arrhenatherion/Calthion</i>	Ex-arable fields	–	6 × 684	–	–	34	–	–	–	3	Manchester, et al. (1999)
Perennial field margins, <i>Trifolium medii</i>	Field margins	No management	3 × 15	860	5880	40 (33)	55 (68)	78	50 (61)	5	Anderlik-Wesinger (2002)*
Calcareous grassland, <i>Festuco Brometea</i>	Ex-arable fields	Mowing once or grazing	5000–6000	389–559, 1 application	1157–3023	57–59; $\bar{x}$ 58 (41–44; $\bar{x}$ 43)	67–69; $\bar{x}$ 68 (51–55; $\bar{x}$ 53)	88–90	66–68; $\bar{x}$ 67 (54–55; $\bar{x}$ 55)	13	Kiehl et al. (2006, unpubl.)
Calcareous grassland, <i>Festuco Brometea</i>	Ex-arable fields	Mowing once or grazing	10,000	805, 3 applications	3665	58 (44)	68 (55)	73	65 (55)	13	Kiehl et al. (2006, unpubl.)
<b>Species-poor grassland without topsoil removal or disturbance of the sward</b>											
Fen meadows, <i>Cirsietum rivularis/Molinietum</i>	Eutrophic fen meadow	Mowing once	114	600, 2 applications	2735–6121	6–8; $\bar{x}$ 7	–	14–27; $\bar{x}$ 21	14–27; $\bar{x}$ 21 (rel. transf. rate)	4–5	Biewer and Poschlod (1997), Poschlod and Biewer (2005)
Floodplain meadows, <i>Cnidion</i>	Species-poor grassland	Mowing once or twice	1000–3200	750–1500	435–3892	28–50, $\bar{x}$ 40 (0–11; $\bar{x}$ 6)	31–56; $\bar{x}$ 48 (0–64; $\bar{x}$ 32)	–	15–48; $\bar{x}$ 33 (0–41; $\bar{x}$ 21)	4	Hölzel et al. (2006), Donath et al. (2007)
<b>Species-poor grassland without topsoil removal but with soil disturbance</b>											
Fen meadows, <i>Cirsietum rivularis/Molinietum</i>	Eutrophic fen meadow, harrowing	Mowing once	114	600, 2 applications	2735–6121	11–16; $\bar{x}$ 14	–	36–37; $\bar{x}$ 37	35 (relative transfer rate)	4–5	Biewer and Poschlod (1997), Poschlod and Biewer (2005)
Floodplain meadows, <i>Cnidion</i>	Species-poor grassland, rotovation	Mowing once or twice	1000–3200	750–1500	435–3892	34–47, $\bar{x}$ 43 (3–10; $\bar{x}$ 7)	29–61; $\bar{x}$ 51 (16–55; $\bar{x}$ 36)	–	21–45; $\bar{x}$ 39 (16–37; $\bar{x}$ 25)	4	Hölzel et al. (2006); Donath et al. (2007)

**Table 2.** Table 2 (*Continued*)

Grassland type, target community	Pre-restoration state; tsr	Management after restoration	Size of receptor site (m <sup>2</sup> )	Dry weight of plant material (g/m <sup>2</sup> )	No. of viable diaspores (per m <sup>2</sup> )	No. of transferred species	Transfer rate (%)	Relative transfer rate (%)	Final establishment rate (%)	Study period (years)	References
<b>Topsoil-removal sites</b>											
Fen meadows, <i>Molinion</i>	Agricultural grassland; tsr: 0.4 m	Mowing once	9×30	180–450	4841–12,777	27 (23)	66 (89)	–	56 (65)	12	Patzelt (1998), Schächtele and Kiehl (2004, unpubl.). Rasran et al. (2006, 2007)
Fen meadows, <i>Scheuchzeria- Caricetea</i>	Eutrophic fen meadow; tsr: 0.4 m	Moderate grazing	10×25	350	2000	33	80	89	60	4	Hölzel and Otte (2003), Hölzel (unpubl.)
Floodplain meadows, <i>Cnidion/Molinion</i>	Ex-arable fields; tsr: 0.3–0.5 m	Mulching or mowing once	6×1000	900–1200	551–24,019	46–71; ø 58 (22–36; ø 26)	80–90; ø 85	90–95	80–90; ø 85	7	Kiehl et al. (2006), Hummitzsch, (unpubl.)
Calcareous grassland, <i>Festuco Brometea</i>	Ex-arable fields; tsr: 0.4 m	Mowing once or grazing	3000–5000	231–419	913–1068	57–61; ø 59 (50–55; ø 53)	67–72; ø 70 (63–69; ø 66)	94–96	67–72; ø 70 (63–69; ø 66)	13	Molder (1995)
<b>Mining areas and quarries</b>											
Mesic grassland <i>Arrhenatherion</i>	Former sand mine, soil application	Mowing twice	3×6	300	2110–3831	24–42; ø 33	54–79; ø 68	–	–	3	Kirmer (2004b, unpubl.)
Mesic hay meadow, <i>Arrhenatherion</i>	Lignite mining area	Mown in 2007	3000	c. 5 cm thick (f.wt.: 1 kg/m <sup>2</sup> )	–	78 (62)	80 (89)	–	67 (76)	9	Tränkle (1997, 1999)
Calcareous grassland, <i>Seslerio-Festucion pallentis</i>	Limestone quarry	No management	15×1	3–5 cm thick (c. 40 l/m <sup>2</sup> )	10,107	60 (40)	49 (54)	89 (89)	37 (46)	6	Kirmer (2004a, unpubl.)
Calcareous grassland, <i>Seslerio-Festucion pallentis</i>	Limestone quarry	No management	800+600	3–5 cm thick (c. 40 l/m <sup>2</sup> )	–	51	56	61	56	5	Tränkle (1997, 2002)
Semi-dry grassland <i>Festuco Brometea</i>	Lignite mining area	No management	1000	c. 5 cm thick (f.wt.: 1 kg/m <sup>2</sup> )	–	61 (44)	72 (72)	–	33 (43)	7	Kirmer (2004b, unpubl.)
Psammophytic grassland <i>Corynephorion</i>	Lignite mining area	No management	8×1	1200 (f.wt.: 2 kg/m <sup>2</sup> )	30,006 (17,010)	36–40; ø 38 (17–19; ø 18)	74–78; ø 76 (83–100; ø 92)	85–86; ø 86 (88–89; ø 89)	41–50; ø 46 (65–71; ø 68)	7	Kirmer (2004a, unpubl.)
Psammophytic grassland <i>Corynephorion</i>	Lignite mining area	No management	100	c. 5 cm thick (f.wt.: 1 kg/m <sup>2</sup> )	–	47 (17)	72 (89)	80 (94)	42 (83)	12	Kirmer (2004a, unpubl.)

\*Data partly re-analysed.



disturbance (harrowing) led to a final establishment rate of 32% whereas deturfing almost doubled the number to 62% (Pywell et al. 2007). In addition, establishment rates were higher on restoration sites managed by mowing than on sites without management (Table 1). The effect of seed-ing persisted through the whole observation period and the sites showed mostly a high cover of sown species (e.g. Jongepierová, Mitchley, & Tzanopoulos, 2007; Kirmer 2006; Kirmer & Stolle 2006; Lepš et al. 2007; Šeffler, Stanová, & Mertanová 1999).

### Seed-enriched chaff, threshing and brush harvesting

One of the oldest methods for species introduction dating back to Roman times is the transfer of dried seed-enriched chaff, e.g. from hay-barn floors (“De re rustica” by Columella 50, cited in Lange 1976). It was used by farmers to create or re-seed grasslands until the 20th century (Duffey et al. 1971), but the seed content is hard to control. Therefore, agronomists in the 19th century discouraged its use to limit the spread of “weeds” (Stebler & Schröter 1892). Bosshard (1999) also pointed out that the use of seed-enriched chaff for restoration purposes will often result in vegetation composition dominated by unwanted species of low nature conservation value. On the other hand, a grassland restoration experiment in Norway showed that 16 target plant species could be established after application of seed-containing hay-barn chaff (Losvik & Austad 2002).

Threshing of seeds from species-rich grasslands has been established as a commercial method to restore target communities (Engelhardt 2000; Schwab, Engelhardt, & Bursch 2002). In southern Germany up to 92 species, including 69 target species, were transferred by threshing material onto industrially degraded dry slopes (Engelhardt 2006). Brush harvesting or seed stripping has been established as a commercial method to harvest seeds without cutting the vegetation. Brush harvesting is also suitable for grassland restoration, but seed content is lower and low-growing species are underrepresented in the seed material harvested in tall meadows compared with the transfer of freshly cut plant material (Edwards et al. 2007; Scotton, Piccinin, Dainese, & Sancin 2009). In shortgrass meadows, however, both techniques were equally efficient (Scotton et al. 2009).

### Transfer of fresh seed-containing hay

The collection of freshly cut seed-containing hay is easier than threshing or brush harvesting since it can be done with conventional cutters and forage trailers without special equipment. In this context it is important to distinguish between the use of freshly cut plant material – by others also referred to as plant clippings or green hay – and the use of dried plant material, i.e. hay *sensu strictu*. The latter was proposed by the pioneer of grassland restoration T.C.E. Wells (Wells, Frost,

& Bell 1986), but its application in restoration practice is limited because cutting, turning and hay swathing reduce the seed content considerably. In contrast, the transfer of freshly cut undried hay (hereafter referred to as hay transfer) has been increasingly documented over a wide range of habitat types (Table 2). Because of its potentially high seed content, the transfer of freshly cut undried hay is much more promising than the use of dried hay (e.g. Rasran, Vogt, & Jensen 2006).

Most data on hay transfer are available from small-scale experiments, but in some cases the method was applied on several hectares. In most of the studies, the plant material was cut at the time of maximal seed set and immediately transported to the receptor sites. The appropriate amount of applied hay depends on plant community type, productivity and site conditions (Table 2). At restoration sites with steep slopes and mobile substrate, e.g. in mined areas, the hay acts as a mulch layer and provides an effective erosion control (Kirmer 2004a; Kirmer & Mahn 2001). In addition, it can provide safe sites for germination and protection of seedlings against desiccation on bare soils but germination and seedling establishment of small-seeded light-demanding species may be hampered if the layer gets too thick (Donath, Hölzel, & Otte 2006; Eckstein & Donath 2005; Kirmer 2004a). For the restoration of low-productive calcareous grasslands, 300–600 g/m<sup>2</sup> of freshly cut hay with an area ratio of 2:1–3:1 between donor and receptor site were suitable (Kiehl, Thormann, & Pfadenhauer 2006). In spite of higher seed numbers, application of thick layers (800 g/m<sup>2</sup> hay from three cuttings) led to lower relative transfer rates in this study than a single transfer with thinner layers (Table 2). For the restoration of low-productive fen meadows, the application of freshly cut hay with an area ratio of 1:1 or 2:1 was sufficient (Klimkowska et al. 2010; Patzelt, Wild, & Pfadenhauer 2001; Rasran, Vogt, & Jensen 2007). During the restoration of mesic *Cynosurus*–*Centaurea* grasslands, however, application in very thin layers with a ratio of 1:3 (donor vs. receptor site) led to a lower number of established species in comparison with a ratio of 1:1 (Edwards et al. 2007). In mesotrophic–eutrophic grasslands with tall vegetation, such as floodplain meadows, target species established well when a 5–15 cm thick layer of freshly cut hay (dry weight 750–1500 g/m<sup>2</sup>) with an average ratio of 7:1 was applied (Donath, Bissels, Hölzel, & Otte 2007). This wide ratio was mainly caused by low biomass production during dry years. In the same region, the ratio was much closer (i.e. 3:1) in an earlier study that had been conducted during more productive years (Hölzel & Otte 2003). In the reviewed studies, the seed content of the transferred hay varied over a broad range. Provided that the proportion of target species was high and site conditions were suitable (see below), transfer of hay with several hundred viable seeds per m<sup>2</sup> was equally successful as the application of hay with several thousand or even more than ten thousand seeds per m<sup>2</sup> (Table 2).

In all reviewed studies, species richness and the number of target species were significantly higher on plots that received seed-containing hay than on control plots. The number of

transferred species depends strongly on species richness of the donor sites. An evaluation of eight newly created calcareous grasslands north of Munich showed that in total 102 plant species from a species-rich nature reserve (including 73 target species specific for calcareous grasslands and 16 red-list species) had established successfully 13–15 years after hay transfer (Kiehl 2009). In restored floodplain meadows on topsoil removal sites, absolute transfer rates per restoration field reached 80–90% (Table 2). In total, up to 120 plant species with 57 target species (mostly *Cnidion* and *Molinion* species) that included 33 red-list species were transferred (Hölzel & Otte 2003, and unpublished data). In several studies, the final transfer rate of all species tended to decrease in comparison with the absolute transfer rate (Table 2). This was partly due to loss of non-target species but also due to loss of rare species that had established only with few individuals on small experimental plots or due to missing management, e.g. in former mining areas.

### Transfer of raked material

Raked material can be easily gathered since the removal of litter and moss layers in grasslands by raking is often recommended as a management measure in grasslands of high conservation value in order to create gaps for species recruitment. Stroh, Storm, Zehm, and Schwabe-Kratochwil (2002) and Stroh, Storm, and Schwabe (2007) transferred 78% of the plant species of a xeric *Allio-Stipetum* on calcareous sand dunes successfully onto restoration plots. Not only low-growing vascular plant species but also bryophytes and lichens of low-productive grasslands can be successfully transferred with raked material. Jeschke (2008) transferred 19 bryophyte and lichen species, including six red-list species successfully to topsoil removal sites; without topsoil removal this number decreased to only 12 species and one red-list species.

### Vacuum harvesting

Unlike brush harvesting, vacuum harvesting can be applied long after seed shed and propagules of low growing plant species are more likely to be transferred. Seeds, litter and also invertebrates are collected by pushed or handheld machines, which were originally constructed for leaf vacuuming or blowing. Stevenson, Ward, and Pywell (1997) transferred 32 out of 66 calcareous grassland species with four harvestings between July and October whereas Thormann, Kiehl, and Pfadenhauer (2003) collected viable seeds of 53 plant species, including 33 target species at one harvest in a calcareous grassland in July. Similar to other species-transfer methods, the number and the proportion of species in the collected seed material strongly depend on the species composition of the donor vegetation. Although Riley, Craft, Rimmer, and Smith (2004) gathered seeds from 19 out of 30 species, 74% of the germinated seedlings came from two species only.

Despite the advantage of collecting shed seeds and seeds of small plant species, vacuum harvesting is a time consuming method for seed collection best applied at sites that cannot be harvested with mowing machinery. It should be carried out on alternating donor sites in order to allow for natural seedling recruitment, which is especially important for short-lived plants.

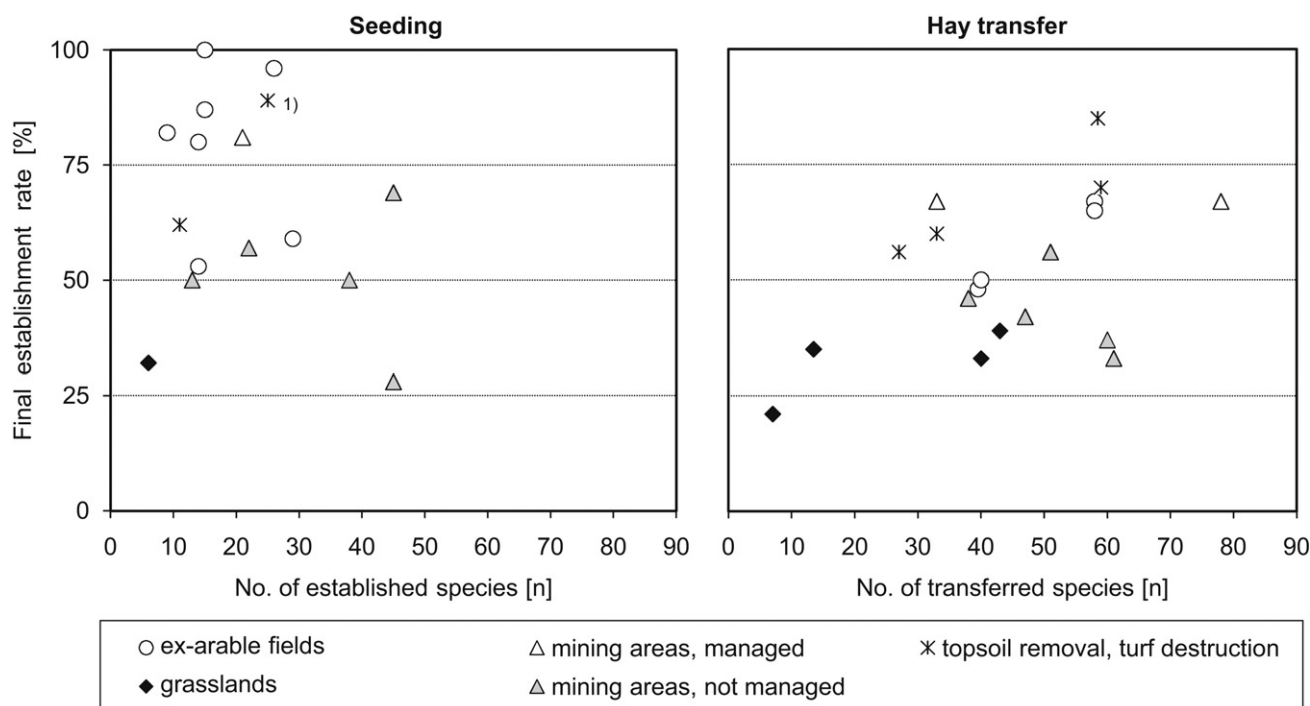
### Transfer of turves and seed-containing soil

The translocation of turves and seed-containing soil can be accomplished by different means (Bullock 1998): (1) hand turving involving cutting and lifting of small turves using spades, (2) machine turving with earth-moving machinery, (3) macroturving with specially designed equipment to cut and lift very large turves and (4) spreading of excavated soil and vegetation (=sod dumping). In Great Britain, there is a long tradition of habitat transfer covering a wide range of plant communities (Byrne 1990; Bullock 1998). Schwickert (1992) gives an overview about various transplantation projects in Germany and Switzerland.

In most experiments, large turves were transferred (more than 0.5 m × 0.5 m, minimum depth 0.3–0.5 m: Bank, Bemmerlein-Lux, & Böhmer 2002; Bruelheide & Flintrop 1999; Cullen & Wheeler 1993; Klötzli 1980; Müller 1990; Park 1989; Trueman, Mitchell, & Besenyei 2007; Worthington & Helliwell 1987). They were laid out area-wide with only small gaps. Only a few studies used smaller turves (minimum 10 cm<sup>2</sup>) and planted them with low intensities (Kirmer 2004a; Šeffer et al. 1999; Wathern & Gilbert 1978). In many sod dumping experiments, topsoil with vegetation was spread on receptor sites with an area ratio of 1:1 and a depth of up to 50 cm (e.g. Bank et al. 2002; Good, Wallace, Stevens, & Radford 1999; Müller 1990; Vécirín & Muller 2003; Worthington & Helliwell 1987). Experiments with only 10 cm depth were documented by Bank et al. (2002) and Kirmer (2004a).

In most experiments, the transfer rate of target species was rather high. Trueman et al. (2007) reported that the transfer rate after habitat transfer of species-rich mesotrophic grassland remains 100% even after 5 years. Bullock (1998) analysed 18 individual translocations in Great Britain showing transfer rates between 54% and 91% after 3–7 years. He stated that an area-wide transplantation was more successful than sod dumping. On the other hand, Good et al. (1999) demonstrated that sod dumping was very cost-saving (up to 4-fold) and still satisfactory in the transfer of target species. On dry sites, they found a transfer rate of 49% for turf transplantation and 54% for sod dumping after 4 years. On wetter sites, the transfer rates reached 71% for turf transplantation and 76% for sod dumping. Kirmer (2004a) and Vécirín and Muller (2003) found transfer rates of target species of 69% (psammophytic grassland, 6 years) and 71% (alluvial meadow, 2 years) after sod dumping. Hence, the sod dumping technique can be used as a method for introduction of soil





**Fig. 1.** Efficiency of species introductions by seeding (left) and transfer of fresh seed-containing hay (right). For seeding, the number of established species from the seed mixture is given and for hay transfer it is the number of species from the donor site transferred successfully to restoration sites. The final establishment rate is the percentage of successfully established species from the seed mixture or from the donor sites at the end of the observation period. The figure summarizes data from Tables 1 and 2. For studies reporting two or more transfer rates due to variation between restoration fields, mean values are shown. <sup>1)</sup>Turf destruction by herbicides.

and propagules of suitable plant species in order to initiate the development of a similar plant community on the restoration site.

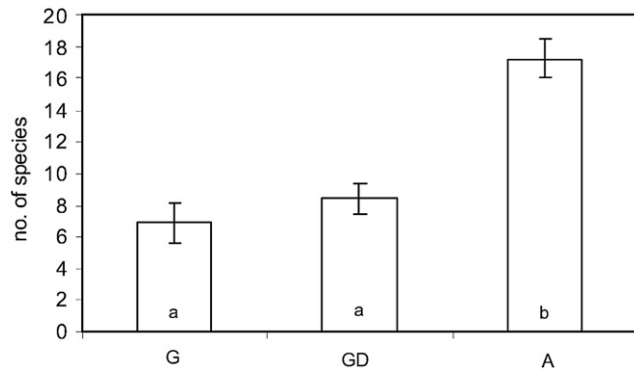
In all soil transfer projects the main problem was eutrophication due to enhanced nutrient mineralisation in the applied soil. Especially the use of large amounts of topsoil can lead to ruderalisation and requires the control of ruderal species by management measures (e.g. Cullen & Wheater 1993; Klötzli 1980; Park 1989; Worthington & Helliwell 1987). Therefore, most authors recommend topsoil removal at the receptor site (at least 20–30 cm, e.g. Vécir & Muller 2003) and the use of only 1–10 cm topsoil from donor sites (e.g. Bank et al. 2002; Kirmer 2004a).

### Preconditions for the successful establishment of target communities

The success of grassland restoration depends not only on species richness, species composition and seed content of the introduced seed mixtures, plant material or soil but also on the availability of suitable microsites for seedling recruitment (Pywell et al. 2002; Walker et al. 2004). Our review indicates that the establishment of species-rich grasslands with habitat-specific species composition is most successful, when seeds, seed-containing plant material or soil are

spread on bare soil of ex-arable fields after tilling or topsoil removal, or on raw soils, e.g. in mined areas (Fig. 1, Tables 1 and 2). Topsoil removal enhances species establishment not only by lowering the nutrient status to levels of the donor sites but also by reducing the seed bank of resident and unwanted species (Marrs 2002; Verhagen, Klooker, Bakker, & van Diggelen 2001). These are the main reasons for its positive effect on establishment and persistence rates of introduced target species in calcareous grasslands, floodplain meadows and fens (Edwards et al. 2007; Hölzel & Otte 2003; Kiehl & Pfadenhauer 2007; Klimkowska et al. 2007; Rasran et al. 2007). The generally high species transfer rates on ex-arable fields without topsoil removal (Fig. 1), however, indicate that the availability of bare soil is of even higher importance than low nutrient availability. The latter seems especially true in cases when nutrient availability is reduced due to periodic drought periods (Donath et al. 2007; Kiehl 2009).

The final establishment rates were low (<40%), when restoration sites were dominated by grasslands with dense swards (Fig. 1). Donath et al. (2007) showed that establishment of transferred species was generally lower in existing species poor grasslands than on ex-arable fields (Fig. 2, Table 2). Analogously, on ex-arable fields dominance of competitive remnant species, i.e. *Elymus repens*, reduced the final transfer rate to only 21% (Donath et al. 2007). Competition can be lowered by herbicide treatment (e.g. Coulson, Bullock,



**Fig. 2.** Number of successfully introduced species detected on restoration sites in the fourth year after transfer of fresh seed-containing hay. Data show means ( $\pm 1$ SE) for restoration sites grouped by their former use (G=grassland,  $N=19$ ; A=arable field,  $N=13$ ) and supplementary measure (GD=grassland disturbed by rotoavation,  $N=19$ ). Different letters at the bottom of the bars indicate significant differences revealed by multiple *U*-tests ( $\alpha_{\text{Bonferroni corrected}} < 0.017$ ). Data modified from Donath et al. (2007).

Stevenson, & Pywell 2001; Tallowin & Smith 2001), but the use of herbicides has low acceptance in nature conservation in many European countries. Although sward disturbance, e.g. by harrowing or rotoavation, considerably increases the establishment rates of target species (Tables 1 and 2; Fig. 2) establishment success fails to reach levels found on tilled ex-arable fields without established vegetation and on sites prepared by deep ploughing, turf stripping or topsoil removal (Donath et al. 2007; Edwards et al. 2007; Walker et al. 2004).

In the long run, nutrient impoverishment may become decisive for the persistence of target communities because tall species of productive grasslands can be favoured by high nutrient availability and out-compete low growing target species (Jongepierová et al. 2007; Tränkle 2002). Pywell et al. (2002, 2007) also found a higher similarity to target communities when the nutrient level was decreased by deturfing or deep ploughing.

Especially on raw soils with low water-holding capacity (e.g. in mined areas or on topsoil-removal sites), restoration success can be increased by creating safe sites for germination and establishment, which prevent desiccation of seedlings. The transfer of seed-containing plant material (e.g. fresh hay or raked material) or application of an additional mulch layer after seeding of site-specific seed mixtures creates safe sites and provides in addition an effective erosion control (e.g. Kirmer 2004a).

In fens and wet meadows, the presence of undisturbed hydrological conditions or careful rewetting of drained sites is a prerequisite for the successful restoration of target communities (Pfadenhauer & Grootjans 1999). Topsoil removal, which leads to lowering of the soil surface and reduces the distance from the groundwater, can be used to provide hydrological conditions, which are adequate for target species (Rasran et al. 2007; Schächtele & Kiehl 2004).

In addition to the optimization of abiotic conditions, the long-term management of newly created grasslands has to be assured in order to suppress the spread of ruderals and woody species. The reviewed studies indicate that mowing once a year or moderate grazing are both suitable management regimes (Tables 1 and 2). Low final establishment rates on restoration sites without management in mining areas and quarries (Fig. 1) can be explained by a loss of low-growing target species due to increasing competition.

## Comparison of different methods for species introduction – final evaluation and conclusions

Walker et al. (2004) concluded that seed limitation can be successfully overcome by species introduction and presented data on the diversification and re-creation of grasslands by seeding or plug-planting. Our review indicates that rules for seeding of site-specific seed mixtures are necessary (e.g. definition of seed zones, see above) and that other near-natural species-introduction measures using locally available seed-containing plant material have several advantages.

In contrast with commercial seed mixtures, freshly cut hay and plant material collected by threshing or raking often contains seeds of rare species, which are not commercially available. Therefore, the number of successfully transferred species is often higher in grasslands restored by hay transfer (>40 species in 11 out of 20 studies) than in grasslands restored by seeding (<30 species in 15 out of 17 studies; Tables 1 and 2). High final establishment rates of species introduced by seeding are mostly due to the introduction of low numbers of rather common species (Fig. 1, Tables 1 and 2).

Generally, the use of fresh hay or raked material is cheaper than the use of site-specific seed mixtures because many wild plants are difficult to cultivate and the propagation of seeds may take several years (Kirmer & Tischew 2006). Nevertheless, the propagation of seeds and creation of species-rich site-specific seed mixtures will be necessary in areas where donor communities of sufficient size for hay transfer have disappeared due to land use change. Planting of propagated pluglings (Walker et al. 2004), however, is even more expensive than sowing (Röder & Kiehl 2007). The transfer of hay or raked plant material favours not only the transfer of vascular plants but also the introduction of mosses and lichens (Jeschke 2008; Jeschke & Kiehl 2006; Poschlod & Biewer 2005). Hay threshing and brush harvesting are more expensive than simple hay transfer due to the use of specialised equipment. Nevertheless, the use of threshed or brush harvested, dried and volume-reduced plant material is recommendable when harvesting of seeds and sowing cannot take place at the same time, e.g. in the case of compensatory measures or when high transport costs from remote donor sites make bulk reduction necessary.

The transfer of raked material or vacuum harvesting are suitable species-transfer measures, when diaspore-rich plant material cannot be harvested by mowing due to inaccessibility for mowing machinery or low standing crop of target communities, e.g. in psammophytic grasslands. Due to the high proportion of hand work, however, these methods are applicable only at smaller scales. The translocation of turves and seed-containing soil demands high logistic efforts. Because of the destruction of the donor community it should be restricted to donor sites, which are sure to be destroyed by infrastructural interventions (e.g. road and railway construction).

In summary, our review shows that the use of near-natural restoration methods can lead to high establishment rates of target species, especially when species establishment is favoured by the availability of bare soil, e.g. on tilled ex-arable fields, raw soils of mining areas or topsoil removal sites. In general, most of the receptor sites develop towards the intended target communities provided that an appropriate management (grazing or cutting) can be assured in the long run. Due to the different successional states, ancient and newly established grasslands can differ in dominance of specific species groups (e.g. legumes), but differences decrease over time in most cases if the management is appropriate (e.g. Kiehl & Pfadenhauer 2007). Although some differences to century-old ancient grasslands may persist – even more pronounced in the seed bank than in the above ground vegetation (Schmiede, Donath, & Otte 2009) – many restoration sites can reach a high nature conservation value within a few years after species introduction (e.g. Donath et al. 2007; Hölzel & Otte 2003; Kiehl & Wagner 2006; Kirmer 2004a). Long-term prospects for restored grasslands with high species diversity, habitat-specific species composition and vegetation structure are especially good if management can be incorporated in agricultural systems. The assessment of fodder quality showed that many semi-natural grasslands are suitable for farming systems (Bullock, Pywell, & Walker 2007; Donath, Hölzel, Bissels, & Otte 2004; Tallwin & Jefferson 1999). Newly restored low-productive grasslands can also be successfully used for recreation and education purposes as well as for landscape design (Gnädinger & Haase 2003; Joas, Gnädinger, Wiesinger, Haase, & Kiehl 2010; Simmons, Venhaus, & Windhager 2007).

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