Universität für Bodenkultur Wien

Department für Nutzpflanzenwissenschaften

Institut für Pflanzenbau
Vorstand: Univ.-Prof. DI Dr. Hans-Peter Kaul
Betreuer: Univ.-Doz. DI Dr. Erich M. Pötsch

Qualitätskriterien geernteter Samenmischungen von ökologisch hochwertigen Grünlandflächen

Quality aspects of harversted seed material of semi-natural grassland

Dissertation
zur Erlangung des Doktorgrades
an der Universität für Bodenkultur Wien

eingereicht von
DI Petra Haslgrübler, BEd

Wien, August 2015

Die vorliegende Dissertation wurde vom österreichischen Bundesministerium für Land- und Forstwirtschaft, Umwelt und Wasserwirtschaft unterstützt und im Rahmen des INTERREG CENTRAL EUROPE Programms durch den europäischen Fonds für regionale Entwicklung (EFRE - Europäische territoriale Zusammenarbeit) ko-finanziert.

Danke …

... Herrn Dr. Erich M. Pötsch für die unermüdliche Betreuung, Korrektur- und Diskussionsbereitschaft und die Unterstützung bei der Entstehung der Publikationen und der Rahmenschrift. Besonders bedanke ich mich für die Motivation die nötige Energie zu finden, um die Arbeit fertig zu stellen.

... Herrn Dr. Bernhard Krautzer für die Überlassung des Themas und den großzügigen Freiraum in der Durchführung des Projektes. Besonderen Dank für die Balance zwischen Fordern und Fördern sowie dem offenen Ohr und die persönliche Unterstützung wenn es doch mal brannte.

... Herrn Dr. Wilhelm Graiss für die freundschaftliche und produktive Zusammenarbeit und die Hilfe bei der Beantwortung aller technischen Fragen.

... Herrn Dr. Albin Blaschka für die Hilfe bei den statistischen Auswertungen und den vielen gemeinsam getätigten Vegetationsaufnahmen. Ganz herzlichen Dank für die Geduld und die freundschaftliche Arbeitsweise.

... der Direktion der HBLFA Raumberg-Gumpenstein, dass ich die Möglichkeit bekam als Projektassistentin das SALVERE Projekt durchzuführen und für die Bereitstellung aller Betriebsmittel zur Erstellung der Dissertation.

... Frau Elisabeth Schwab für die Einführung in die vegetationskundlichen Aufnahmen und die Hilfe bei den zahlreichen Bonituren.

... dem gesamten Team der Abteilung Vegetationstechnik im Alpenraum, im speziellen Doris Holzinger, für die zahlreiche Unterstützung beim Auszählen und Zerlegen der unzähligen Proben, für die Anlage der Versuchsfläche und die Durchführung der Feldversuche. Dickes Dankeschön für die freundschaftliche Aufnahme in das Team und die unterhaltsame Arbeitsatmosphäre.

... Herrn Dr. Alan Hopkins für die rasche Bearbeitung der einzelnen Publikationen und der fachlichen Unterstützung beim Verfassen.

... Frau DI Johanna Huber für die vielen motivierenden Gespräche und die nötige Ausdauer beim Korrekturlesen.

... allen Personen, die in irgendeiner Form dazu beigetragen haben um diese Dissertation erfolgreich abzuschließen.

... meiner Familie, die mir einen langen Ausbildungsweg ermöglicht hat und mich in jeglicher Form gefördert, unterstützt und begleitet hat.

... meinem verständnisvollen Partner, der immer an mich geglaubt und mich in jeglicher Hinsicht unterstützt hat.

Inhaltsverzeichnis

Zusammenfassung

Heute zählen artenreiche Wiesen, welche einen hohen naturschutzfachlichen Wert haben, zu den am stärksten gefährdeten Biotoptypen der mitteleuropäischen Kulturlandschaft. Es ist wichtig, das Samenpotenzial noch vorhandener extensiv bewirtschafteter Wiesen als wesentliche Ressource für den Indikator High Nature Value Farmland zu erhalten und zu nutzen, um das gewonnene Samenmaterial bei regionalen Begrünungs- und Renaturierungsmaßnahmen einzusetzen. Aufgrund der spezifischen Anforderungen von Pflanzenarten an die jeweiligen Standortverhältnisse muss eine Spenderfläche mit vergleichbaren Standorteigenschaften ausgewählt werden, damit die Arten optimal an die Bedingungen der Empfängerfläche angepasst sind. Das benötigte Samenmaterial wird durch unterschiedliche Erntemethoden wie frischer Grünschnitt, Wiesendrusch und Ausbürsten geerntet. Um exakte Versuchsdaten und genaue Informationen zu den Ernte- und Etablierungsmethoden zu erarbeiten, wurde das INTERREG Projekt SALVERE (Semi Natural Grassland as a Source of Biodiversity Improvement) ins Leben gerufen.

Gegenstand der im Rahmen dieses Projektes entstandenen Dissertation war es, ein Verfahren zur Bestimmung wichtiger Qualitätsparameter wie Reinheit, Tausendkorngewicht und Keimfähigkeit von geerntetem Material von Spenderflächen zu entwickeln, das kostengünstig und praktikabel ist. Dafür wurde das von einer Glatthaferwiese geerntete Samenmaterial (Wiesendrusch und Ausbürstmaterial) luftgetrocknet und grob mit einem 6 mm Sieb gereinigt. Die größte Schwierigkeit war es, eine homogene Saatgutprobe zu ziehen. Dafür wurde aus vielen Einzelproben eine Mischprobe hergestellt, aus der letztlich Proben zu 5 g gezogen wurden, um die Reinheitsuntersuchungen und Tausendkorngewichtsbestimmungen durchzuführen, die in Anlehnung an die Vorgaben der „International Seed Testing Association“ erfolgten. Am Beginn wurde jede Probe sortiert, um die einzelnen Komponenten festzustellen.

Die Ergebnisse zeigten, dass mit beiden Erntemethoden (Wiesendrusch und Ausbürsten) ein Anteil von ca. 60 % reiner Samen erzielt wurde, allerdings war das Tausendkorngewicht unterschiedlich. Mit dem Ausbürstgerät wurden vor allem kleinere und leichtere Samen geerntet. Die Zerlegungsproben ergaben in beiden Mischungen einen 80 %igen Anteil an Gräsern. Das ist typisch für Glatthaferwiesen, die Anfang Juli geerntet werden. Bei einer Ernte im August oder September wäre der Anteil an Kräutern und Leguminosen wesentlich höher. Daher ist es ratsam, einen frühen und einen späten Erntetermin zu mischen, um das wesentliche Artenspektrum der Spenderfläche auf die Empfängerfläche übertragen zu können. Zur Bestimmung der Keimfähigkeit wurde auf Basis einer Literaturstudie eine geeignete Methode entwickelt. Dazu wurden im ersten Schritt drei Substrate – sterile Ansaaterde, Quarzsand und Filterpapier – getestet. Das homogenste und beste Ergebnis wurde mit steriler Ansaaterde erzielt, welche auch für die weiteren Untersuchungen herangezogen wurde. Weiters wurde getestet, ob Maßnahmen zum Abbau der Keimhemmung wie Vorkühlen, Zugabe von Kaliumnitrat (KNO_3) und Gibberellinsäure (GA_3) die Keimfähigkeit des Saatguts erhöhen. Es zeigte sich, dass durch diese Behandlungen die Keimfähigkeit nicht signifikant erhöht werden konnte, daher wurden sie in weiteren Tests nicht angewendet. Nach den Vortests im Keimschrank wurden die Haupttests im Gewächshaus durchgeführt. Nach der Reinigung wurde das Samenmaterial bei unterschiedlichen Temperaturen, nämlich bei Raumtemperatur (18-23°C), im Kühlhaus (2-5°C) und im Gefrierschrank (-18°C), gelagert. Die Keimfähigkeitstests wurden

für alle drei Lagerungsvarianten drei Jahre lang wiederholt. Es zeigte sich, dass das Saatgut unter kühlen Bedingungen (2-5°C) maximal zwei Jahre sehr gut lagerfähig ist. Der frische Grünschnitt wurde direkt nach der Ernte Anfang Juli auf der Empfängerfläche aufgebracht. Die Ansaat mit Wiesendrusch erfolgte Ende August mit einer Saatstärke von 3 g/m^2. Die Vegetation wurde zweimal jährlich bonitiert. Bereits nach vier Jahren konnte festgestellt werden, dass es keine signifikanten Unterschiede zwischen dem frischem Grünschnitt und der Wiesendruschvariante in der Übertragungsrate der Zielarten gab. Laut Saatgutgesetz (SaatG, 1997) ist das Inverkehrbringen von Wildpflanzensaatgut verboten, und laut österreichischer Naturschutzgesetzgebung ist es verboten, gezüchtetes Saatgut auf Naturschutzflächen auszubringen. Aus diesem Grund wurde im Jahr 2010 eine neue Verordnung (2010/60/EU) von der EU-Kommission herausgegeben, die den Handel und den Einsatz von Wildsamen erlaubt und regelt. Auf nationaler Ebene erschienen im Jahr 2014 in Österreich die ÖNORM L 1113 Begrünung mit Wildpflanzensaatgut sowie die Richtlinie ONR 121113, die nun diese ökologisch wertvolle Form der Begrünung ermöglichen und damit auch eine unmittelbare Umsetzung der in der vorliegenden Dissertation erarbeiteten Ergebnisse gewährleisten.

Abstract

In the last few decades there has been a significant decrease of biodiversity in Europe. To counter this trend, harvested seed mixtures from potential donor sites can be used to establish semi-natural grassland. Extensively managed semi-natural grassland which is the most important type of High Nature Value Farmland (HNVF) can be regarded as a seed source useful to establish new HNV areas. Indeed, they are normally rich in species of native provenance and for this reason they can be harvested to obtain valuable propagation material. Therefore a large number of different harvesting methods (green hay, on-site threshing and seed stripping) have been developed. It is important to guarantee a certain level of seed quality and successful germination as well as the regional provenance of the harvested seed mixtures. Seed material was harvested from an Arrhenatherion meadow. After the determination of purity and thousand seed weight, the pre-tests started in the climate chamber. The germination capacity of 400 randomly selected seeds was tested in two trials for four weeks to determine the substrate and dormancy breaking treatments. In the first trial, three substrates (organic growing media, quartz sand and filter paper) were tested. Germinated seedlings were counted as monocots and dicots. Organic growing media showed the highest germination rate. In the second trial, the germination capacity of the seed mixture was tested on organic growing media with and without pre-chilling after addition of potassium nitrate (KNO_3), addition of gibberellin acid (GA_3) and without addition of chemicals. In general, the germination capacity of the pre-chilled variants was lower. In the next steps the germination capacity tests (storage conditions and storage length) in the greenhouse started. The harvested seed material was stored under different conditions up to three years and tested once a year. A specific volume of seed material was sown on organic growing media. The results showed that storage under different conditions and the length of storage influenced the germination capacity significantly. Storage under cool and dry conditions revealed better results. There is a strong positive relationship (correlation) between the proportion of mature seed and germination percentage.

A receptor site was cultivated using green hay material from the donor site right after the harvest in July. The on-site threshing material was then sown at the end of August using 3 g/m². The vegetation of the receptor site was monitored twice a year. After already four years of observation no significant differences could be found between green hay and on-site threshing material concerning the transfer rate of target species. According to the seed material law (SaatG 1997) marketing of seeds of native plants is forbidden and due to the Austrian legislation of nature conservation, bred seed material must not be used on nature conservation areas. Therefore a new regulation (2010/60/EU) was established by the European Commission in 2010, which controls trade and use of seeds of native plants. On national level the Austrian norm "Revegetation with seeds of wild plants" (Begrünung mit Wildpflanzensaatgut, ÖNORM L 1113, ONR 121113) was launched in 2014 which now allows to use this ecologically valuable kind of re-cultivation and also ensures the direct implementation of the achieved results and findings of this thesis.

Einleitung

Aufbau der Dissertation und des Versuchsdesigns

Einleitung

Mehr als 40 % der Landfläche der Erde ist von Grünland bedeckt, und es kommt in fast allen Regionen der Erde vor (Pötsch et al., 2012). Der Mensch hat durch die unterschiedlichen Bewirtschaftungsweisen ein hohes Maß an Abwechslung und eine Vielfalt unterschiedlicher Grünland-Lebensräume geschaffen (Ellenberg und Leuschner, 2010). Bedingt durch intensive Bewirtschaftung und den Strukturwandel in der Land- und Forstwirtschaft hat die Biodiversität in den vergangenen Jahrzenten jedoch in dramatischem Ausmaß abgenommen (Hopkins et al., 1999). Die Erhaltung der seltenen Tier- und Pflanzenarten sowie die generelle Bewahrung der biologischen Vielfalt sind inzwischen zu einem besonderen Anliegen der Agrar- und Umweltpolitik geworden. Heute zählen artenreiche Wiesen, die einen hohen naturschutzfachlichen Wert haben, zu den am stärksten gefährdeten Biotoptypen der mitteleuropäischen Kulturlandschaft (Kirmer et al., 2012) und machen ca. 15-25 % der landwirtschaftlich genutzten Fläche in Europa aus (Pötsch et al., 2012). Extensiv bewirtschaftetes ökologisch hochwertiges Grünland stellt den wichtigsten Typ von High Nature Value Farmland (HNVF) dar, das als zielorientierter Basisindikator entsprechend der Evaluierung des Programms zur ländlichen Entwicklung LE 07-13 nominiert wurde (Bartel und Schwarzl, 2008).

Ein wesentlicher Teil des „High Nature Value Farmland" Konzeptes besagt, dass die Erhaltung der Biodiversität in Europa von der Aufrechterhaltung und Kontinuität extensiver landwirtschaftlicher Bewirtschaftungssysteme abhängt. Es ist wichtig, noch vorhandene extensiv bewirtschaftete Wiesen zu erhalten und nicht nur als Futterfläche sondern auch zur Gewinnung von wertvollem Samenmaterial als Spenderflächen zu nutzen. Das gewonnene Saatgut kann entweder direkt bei regionalen Begrünungs- und Rekultivierungsmaßnahmen eingesetzt werden oder als Ausgangsmaterial zur Saatgutproduktion dienen (Kirmer et al., 2012). Mit der Nutzung von Grünland zur Gewinnung von regionalem Saatgut wird zugleich einer Verbuschung und Verbrachung der Flächen entgegengewirkt.

Aktueller Wissensstand über die Nutzung von artenreichen, extensiven Wiesen zur Gewinnung von Saatgut

Noch ursprüngliche artenreiche Wiesen beheimaten die für die Region charakteristischen Unterarten und Ökotypen in regionaltypischer Artenzusammensetzung und sind deshalb für die Gewinnung von Saatgutmischungen, samenreicher Biomasse oder das Sammeln von Basissaatgut optimal geeignet (Krautzer und Klug, 2009). Die Nutzung dieses Potenzials für Renaturierungs- und Begrünungsmaßnahmen unterstützt den Erhalt dieser Lebensräume und könnte Landwirten ein zusätzliches Einkommen ermöglichen. Aufgrund der spezifischen Anpassungen der Pflanzenarten an die jeweiligen Standortverhältnisse, sollten die Standorteigenschaften der Spenderfläche mit jenen der Empfängerfläche übereinstimmen oder sich nur geringfügig davon unterscheiden (Kirmer und Tischew, 2006).

Besonders wichtig für die Etablierung standortgerechter Pflanzenbestände ist eine Bodenvorbereitung, die den Standortansprüchen des zu erzeugenden Vegetationstyps gerecht wird. Als Pflanzsubstrat sollte möglichst diasporenfreies, humusarmes Oberbodenmaterial („Zwischenboden") verwendet werden, welches aufgrund seines geringeren Nährstoffgehaltes und des geringen bis fehlenden Diasporengehaltes im Regelfall sehr gut für Begrünungen mit Saat- und Pflanzmaterial geeignet ist. Der Boden sollte gut abgesetzt sein. Die Samen sollten nur oberflächlich (max. 0,5 cm) abgelegt und danach gewalzt werden, damit ein guter Bodenschluss gewährleistet ist und eine schnelle Keimung erfolgen kann (Kirmer et al., 2012).

Die Saatgutmengen (bezogen auf die im Begrünungsmaterial vorhandenen reinen Samen) betragen zwischen 1-5 g/m². Bei extremen Standortbedingungen kann die empfohlene Aufwandsmenge bis zu 15 g/m² betragen. Höhere Aussaatmengen sind in keinem Fall notwendig (Krautzer und Hacker, 2006). Im Regelfall wird eine Spenderfläche einmal beerntet. Ein günstig gewählter Erntetermin führt zur erfolgreichen Übertragung einer breiten Palette von Arten, die sich bei günstigen Voraussetzungen auf der Empfängerfläche auch langfristig etablieren können. Der Erfolg der Ansaat (Begrünung) wird dabei entscheidend von den Bedingungen auf der Empfängerfläche beeinflusst. Einige Wochen nach Anlage der Grünlandfläche kann ein Reinigungsschnitt (Schnitthöhe mindestens 7-10 cm) erfolgen, ohne den Bestand negativ zu beeinflussen. Eine Düngung halbtrockener und feuchter Extensivwiesen soll im Regelfall nicht erfolgen. Ist eine gewisse Ertragsfähigkeit solcher Wiesen gewünscht, kann eine moderate Düngung mit Stallmist im Abstand von mehreren Jahren ausgeführt werden.

Die Zielvegetation muss zum einen an den Standort angepasst sein und zum anderen dem Begrünungsziel entsprechen. Prinzipiell sollten die Saatgutmischungen möglichst artenreich sein. Dadurch wird eine hohe Flexibilität gegenüber potenziellen Störungen erreicht (Risikostreuung), und durch die vielschichtige ober- und unterirdische Struktur der etablierten Vegetation kann der Standort optimal genutzt und damit auch gesichert werden (Kirmer et al., 2012).

Erntemethoden

Durch die Erntemethoden muss gewährleistet sein, dass das Samenmaterial einen hohen Anteil von unterschiedlichen Arten aufweist, was sich dann auch an der Zahl der erfolgreich auf die Empfängerfläche übertragenen Arten widerspiegelt. Um dies zu erreichen, muss der Erntezeitpunkt auf die Samenreife der Arten abgestimmt werden. Für die Einschätzung potenziell übertragbarer Arten auf der Spenderfläche ist kurz vor der Ernte eine Vegetationsaufnahme mit Erfassung des phänologischen Zustandes empfehlenswert. Die tatsächliche Anzahl keimfähiger Samen im geernteten Material ist von verschiedenen Faktoren wie z.B. Wiesentyp, Management (1./2. Schnitt), Tageszeit, Witterung und Erntezeitpunkt abhängig und damit schwierig zu quantifizieren. Der Gehalt an keimfähigen Samen kann durch Keimversuche ausgezählter oder abgewogener Proben ermittelt werden (Kirmer et al., 2012).

Frisches Mähgut (green hay)

Abbildung 1: Aufbringen des frischen Grünschnittes

Zur Gewinnung von frischem artenreichen Mähgut wird die Spenderfläche zu einem Zeitpunkt gemäht, an dem möglichst viele Arten die optimale Samenreife erreicht haben, wobei auch bei späteren Mähterminen noch Samen von bereits verblühten Arten im Mähgut enthalten sind (Donath et al., 2007, Hölzel und Otte, 2003). Am ertragsreichsten ist eine Mahd am frühen Morgen, wenn die Samen durch den Tau noch gut an den Pflanzen haften. Optimal ist eine sofortige Aufnahme des frischen Mähguts mit dem Ladewagen, da alle zusätzlichen Arbeitsgänge, z.B. Schwaden, Trocknen und Wenden zu Samenverlusten führen. Bei frischem Mähgut ist eine 100 %ige Samenausbeute möglich, und der Samenanteil im geernteten Material beträgt ca. 0,2-2 %, der Rest sind Blätter und Stängel. Es werden auch Kleintiere, vor allem Insekten und Spinnentiere, übertragen (Elias und Thiede, 2008, Kiehl und Wagner, 2006). Frisches Mähgut ist nicht lagerfähig und sollte deshalb unmittelbar nach der Ernte auf der Empfängerfläche ausgebracht werden. Es schützt die Empfängerfläche vor Erosion und Austrocknung. Um eine Selbstentzündung zu verhindern, sollte es nicht angehäufelt werden. Außerdem sollte auf möglichst kurze Transportwege geachtet werden (Kirmer et al., 2012).

Wiesendrusch (on-site threshing)

Abbildung 2: Parzellendrescher

Wiesendrusch wird mit einem Mähdrescher gemäht und im selben Arbeitsgang gedroschen. Die Schnitttiefe ist variabel einstellbar und bestimmt neben dem Erntezeitpunkt die Artenzusammensetzung und die Samenausbeute. Es sollten nur gut abgetrocknete Bestände gedroschen werden. Dichte Bestände können zuerst geschwadet und danach gedroschen werden, dies kann auch in feuchtem Zustand durchgeführt werden (Kirmer et al., 2012). Die Samenausbeute in Bezug zur Samenmenge auf der Erntefläche beträgt ca. 50-80 % (Scotton et al., 2012). Wird das Material eingelagert, muss es unter Luftzufuhr getrocknet und anschließend kühl und trocken gelagert werden. Es kann dann maximal 2 Jahre gelagert werden (Haslgrübler et al., 2014).

Ausgebürstete Samen (seed stripping)

Das Ausbürsten der Samen erfolgt im stehenden Bestand, dabei werden vorwiegend reife Samen abgestreift. Da die Vegetation nicht gemäht wird, kann dieselbe Fläche zu einem späteren Zeitpunkt erneut beerntet werden (Morgan und Collicutt, 1994). Für die Ernte durch Bürsten sind Spezialmaschinen erforderlich. Diese werden vor allem in den USA, Kanada und Großbritannien produziert. Dabei handelt es sich um einen gezogenen Anhänger, in dem sich eine rotierende Bürste befindet (Edwards et al., 2007, Morgan et al., 1995). Bei einer maschinellen Ernte mit rotierenden Bürsten ist die Samenausbeute stark von der Höhe und der Bewegungsrichtung der Bürsten abhängig. In hochwüchsigen Beständen liegt diese bei ca. 20-50 %, in niedrigwüchsigen Beständen bei ca. 50-75 %. Niedrige Bestände mit einer Bestandshöhe von 35-65 cm sind für die Beerntung durch Bürsten am besten geeignet (Scotton et al., 2012). Hohe Bestände weisen oft eine filzartige Struktur auf, wodurch sie schwieriger mittels Büsten zu beernten sind (Krautzer et al., 2013). Das geerntete Samenmaterial sollte luftgetrocknet werden und kann maximal zwei Jahre gelagert werden (Haslgrübler et al., 2014).

Abbildung 3: Seed stripper - Ausbürstgerät

Qualitätskriterien

Die Qualität von Samen und Samenmischungen, die bei Begrünungsmaßnahmen eingesetzt werden, ist für den Begrünungserfolg von großer Bedeutung. Dabei interessieren den Anwender vor allem die Zusammensetzung, die Keimfähigkeit, das Tausendkorngewicht und, daraus resultierend, die notwendige Ansaatmenge. Die Funktionsfähigkeit und die Kosteneffektivität der beschriebenen Methoden muss von Fall zu Fall separat bewertet werden. Die Erhebung der Qualitätsparameter einzelner Arten bzw. daraus hergestellter Mischungen mehrerer Arten richten sich prinzipiell nach den Methoden der "International Seed Testing Association" (ISTA 2011). Im Gegensatz dazu gibt es für die Qualitätsparameter von direkt geernteten Samenmischungen keine Richtlinien. In Anlehnung an bestehenden Methoden (Florineth, 2012, Heilinger und Florineth, 2003, Molder, 1995, 2008), bei denen mit derselben Wiesengesellschaft (Arrhenatherion) gearbeitet wurde, wurde eine Methode entwickelt um die Qualitätskriterien zu bestimmen.

Probennahme

Bei der Probennahme soll ein ausreichend großes und repräsentatives Saatgutmuster aus der gesamten Erntemenge (Partie) gezogen werden, wobei jeder Bestandteil (reine Samen, Bruch, unerwünschte Arten, Spreu) im gleichen Verhältnis wie in der Partie vorhanden sein muss. Die Aussagekraft der Prüfungsergebnisse hängt entscheidend von der Sorgfalt der Probenahme ab. Die

Probengröße richtet sich nach der gesamten Erntemenge und ist für reine Samen laut ISTA (2011) definiert. Bei schwer fließendem Saatgut ist die Probennahme von Hand die geeignete Methode. Hierzu ist es erforderlich, eine größere Anzahl von Säcken zu öffnen und die Erstproben aus unterschiedlichen Tiefen mit der Hand zu entnehmen. Um Proben auch aus dem unteren Bereich zu erhalten, kann es erforderlich sein, eine bestimmte Anzahl von Säcken ganz oder teilweise zu entleeren. Die Behältnisse, aus denen die Erstproben entnommen werden, sind zufällig oder systematisch aus der ganzen Partie auszuwählen. Erstproben sind sowohl oben, in der Mitte, als auch unten aus den Behältnissen zu entnehmen. Erscheinen die Erstproben einheitlich, so werden sie in ein sauberes Gefäß geschüttet und nach Abschluss der Probenziehung durchmischt. Aus der Mischprobe werden die Teilproben durch wiederholtes Halbieren gewonnen. Es ist darauf zu achten, dass sich die Samenmischungen durch Rühren nicht entmischen (AGES, 2004, Hebeisen und Graff, 2008, ISTA, 2011).

Bestimmung von Reinheit und Tausendkorngewicht (TKG)

Die Zusammensetzung und Qualität des durch Mähen, Dreschen oder Bürsten gewonnenen Saatgutes kann von Jahr zu Jahr variieren. Die Anteile an Spreu und Verunreinigungen, wie z.B. Erde, können sehr hoch sein. Das trockene Material sollte deshalb vor der Lagerung und dem Ausbringen grob gereinigt werden. Dafür ist ein Sieb mit einer Lochgröße von 3-6 mm empfehlenswert. Sind größere und sperrigere Samen in der Samenmischung enthalten, richtet sich die Lochgröße des Siebes nach den größten Samen. Aus dem gereinigten Material wird eine homogene Probe gezogen. Im Labor werden Spreu und Samen getrennt und ihre Anteile bestimmt. Je nach Wiesentyp werden dafür 3-6 Arbeitstage benötigt. Für die Bestimmung des Tausendkorngewichtes werden achtmal 100 zufällig ausgewählte, volle Samen gezählt und gewogen (ISTA, 2011).

Keimfähigkeitsprüfung

Die Methode zur Ermittlung der Keimfähigkeit von Samenmischungen ermöglicht es, in einem überschaubaren Zeitraum mit begrenztem technischen und personellen Aufwand, Aussagen über das Samenpotenzial der direkt geernteten Samenmischungen zu treffen (Haslgrübler et al., 2013, 2014). Es wurden Vortests im Keimschrank durchgeführt, um ein geeignetes Substrat zu definieren und festzustellen, ob keimbrechende Maßnahmen notwendig sind. In weiteren Tests wurde die Dauer der Lagerfähigkeit und die richtige Lagerungstemperatur getestet.

Richtlinien und Zertifizierungssysteme

Insbesondere für große Begrünungsflächen und zur Sicherstellung einer saisonal nicht eingegrenzten Verfügbarkeit von regionalem Saatgut kommt der landwirtschaftlichen Vermehrung von Wildpflanzensaatgut eine große Bedeutung zu. Um sicherzustellen, dass dem ökologischen System kein Schaden zugefügt wird, ist nur Material (Wiesendrusch, Sammelgut, Vermehrungsgut) zuzulassen, welches für die Begrünung und Wiederherstellung in der gleichen Region verwendet wird. Die einzusetzenden Arten müssen im regionalen Bezug gewonnen sein und dürfen nur in festzulegenden zugehörigen Produktionsräumen vermehrt werden. Verwertung, Produktion und

Handel von regionalem Saatgut führen ohne gemeinsame Regeln (Zertifizierung, Etikettierung) für den Verbraucher zu einem unüberschaubaren Markt. Wildsamen aus unterschiedlichen Regionen konkurrieren mit gezüchteten Sorten der gleichen Pflanzenart. Im Sinne des Naturschutzes ist es notwendig, Regeln zu erstellen, um den europäischen Handel mit Wildsamen transparent gestalten zu können. Die wichtigste EU-Verordnung für Futterpflanzen (RICHTLINIE DES RATES vom 14. Juni 1966 über den Verkehr mit Futterpflanzensaatgut (66/401/EWG)) führte zu erheblichen Problemen zwischen den nationalen Naturschutzgesetzen und dem Schutz für Saatgutzüchtungen. Es durften von geschützten Sorten keine Wildformen ausgebracht werden und das Naturschutzrecht verbot die Ausbringung von Zuchtformen auf nicht prioritär landwirtschaftlich genutzten Flächen. Aus diesem Grund wurde im Jahr 2010 eine neue Verordnung von der EU-Kommission herausgegeben, die den Handel und den Einsatz von Wildsamen erlaubt und regelt (EU, 2010). Die Richtlinie 2010/60/EG über das Inverkehrbringen von Futterpflanzenmischungen zur Erhaltung der natürlichen Umwelt ist als Ausnahmeregelung vom Saatgutrecht für das Inverkehrbringen von Futterpflanzensaatgutmischungen zur Erhaltung der natürlichen Umwelt zu verstehen. Dadurch wurde ein Regelungsrahmen gesetzt, der die Gewinnung und Herstellung sowie den Einsatz von regionalen Wildpflanzenmischungen ermöglicht (Krautzer, 2015).
Zur Umsetzung in nationales österreichisches Recht wurde die „Durchführungsrichtlinie für die Zulassung von Saatgut von Futterpflanzenmischungen und das Inverkehrbringen von Saatgut dieser Mischungen“ erlassen. Diese Durchführungsrichtlinie dient der praktischen Umsetzung der Richtlinie 2010/60/EU in Zusammenhang mit § 4 „Erhaltungssorten“ der Saatgutverordnung BG Bl. II Nr. 417/2006 idgF. Die Bestimmungen gelten für sogenannte Erhaltungsmischungen (= vereinfacht eine Mischung zur Bewahrung der natürlichen Umwelt im Rahmen der Erhaltung pflanzengenetischer Ressourcen) und gelten für Mischungen, welche Saatgut von Futterpflanzen enthalten. Davon ausgenommen sind Mulch, Grünschnitt und diasporenhaltiger Boden. Das heißt, dass in der Praxis die meisten Wildpflanzensaatgutmischungen davon betroffen sind. Zur nationalen Umsetzung in Österreich existieren gegenwärtig zwei Zertifikate für Wildpflanzensaatgutproduzenten: das REWISA®-Zertifikat (www.rewisa.at – letzter Zugriff: 08.04.2015) und das Gumpensteiner Herkunftszertifikat (G-Zert) (http://www.raumberg-gumpenstein.at/cm4/de/aktuelles-lfz/forschung/4662-regionale-wildgraeser-und-wildkraeuter-nach-gumpensteiner-herkunftszertifikat-g-zert.html – letzter Zugriff: 08.04.2015). Auf nationaler Ebene wurden die Vorgaben der Richtlinie 2010/60/EU in die ÖNORM L1113 und L121113 (Begrünung mit Wildpflanzensaatgut) eingearbeitet, die 2014 neu erschienen ist (ÖNORM, 2014a, 2014b).
Zu dieser Thematik wurde im Rahmen des INTERREG CENTRAL EUROPE Programms (http://www.central2013.eu – letzter Zugriff: 07.08.2014) das SALVERE Projekt durchgeführt und durch den europäischen Fonds für regionale Entwicklung (EFRE - Europäische territoriale Zusammenarbeit) ko-finanziert. Das SALVERE Projekt („Semi Natural Grassland as a Source of Biodiversity Improvement”) soll dazu beitragen, ökologisch hochwertige landwirtschaftliche Flächen (HNVF) als eine wertvolle Ressource zu sehen. Damit soll eine nachhaltige Entwicklung des ländlichen Raumes und benachteiligter Gebiete gefördert werden. Das Projekt wurde von Januar 2009 bis Dezember 2011 durchgeführt. Es beteiligten sich acht Institutionen aus

insgesamt sechs Ländern (Österreich, Deutschland, Italien, Tschechien, Slowakei und Polen). Die wesentlichen Zielsetzungen dieses Projektes waren eine Analyse des Ist-Zustandes von hochwertigen landwirtschaftlichen Flächen (HNVF), unter Berücksichtigung von Agrarpolitik und Zukunftstrends, die Definition von Qualitätskriterien zur Keimfähigkeit sowie zu Ernte- und Lagermethoden des gewonnenen Samenmateriales, eine Verbesserung der Techniken zur Neuanlage von HNV-Flächen, sowie die Förderung der Verwendung von regionalem Saatgut zur Bewahrung der genetischen Eigenständigkeit des Landschaftsraumes. Die nachfolgenden Publikationen bauen im Wesentlichen auf Daten und Inhalte dieses Projektes auf.

Aufbau der Dissertation und des Versuchsdesigns

Die vorliegende, kumulative Dissertation gliedert sich thematisch in vier Kapitel, die im Folgenden näher erläutert werden.

In **Kapitel 1** geht es vor allem um die Erhaltung und Pflege der Artenvielfalt auf landwirtschaftlich genutzten Flächen. Dieses Thema ist ein besonderes Anliegen der Agrar- und Umweltpolitik und gewinnt in ganz Europa immer mehr an Bedeutung. Es wird eine Definition für High Nature Value Farmland (HNVF) vorgestellt und die unterschiedlichen HNVF-Typen werden erklärt. Es wird außerdem darauf hingewiesen, wie wichtig verbindliche europäische Richtlinien sind, um die Naturschutzziele in ganz Europa zu erfüllen und umzusetzen. Diese Publikation wurde 2011 im Rahmen des „16th Symposium of the European Grassland Federation" in Gumpenstein/Österreich, veröffentlicht.

Um die Qualität hinsichtlich Reinheit, Tausendkorngewicht, Keimfähigkeit und Lagerfähigkeit von geernteten Saatgutmischungen aus ökologisch hochwertigem Grünland zu bestimmen, wurde im Rahmen der vorliegenden Arbeit ein Verfahren entwickelt und Richtwerte erarbeitet, das kostengünstig und zugleich aussagekräftig ist. Dieses Verfahren wird in den **Kapiteln 2 und 3** im Detail erklärt. Zuerst wurden mit dem geernteten Saatgut Testreihen im Keimschrank durchgeführt, um ein geeignetes Substrat zu bestimmen und herauszufinden, ob keimbrechende Maßnahmen wie Vorkühlen, Kaliumnitrat (KNO_3) und Gibberellinsäure (GA_3) notwendig sind. In weiterer Folge wurden die Haupttests im Glashausversuch durchgeführt, und die Lagerfähigkeit des Saatgutes wurde getestet. Das Material dazu stammte von zwei unterschiedlichen Erntemethoden, nämlich von Wiesendrusch und von der Ausbürstmethode. Die Tausendkorngewichts- und die Reinheitsbestimmungen wurden in Anlehnung an die ISTA (ISTA, 2011) gemacht. Der Artikel in Kapitel 2 wurde in Grass and Forage Science 69 (3) (2013) publiziert und der Artikel in Kapitel 3 in Grass and Forage Science 70 (3) (2014) veröffentlicht.

Die Spenderfläche, von der das getestete Saatgutmaterial stammte, befand sich im nordwestlichen Teil von Österreich in der Nähe von Linz (48°18‘ N, 14°03‘ E; 310 m m.ü.A.). Es handelte sich um eine Glatthaferwiese beschrieben nach Koch (Koch, 1926). Die durchschnittliche Jahrestemperatur betrug 2009 9,6°C mit einem durchschnittlichen Jahresniederschlag von 1017 mm. Seit 1998 wurde die gesamte Fläche nicht mehr gedüngt, und einmal jährlich gemäht (Bogner, 1992). Beerntet wurde die Fläche am 1. Juli 2009. Insgesamt wurden drei verschiedene Erntemethoden angewendet: Frischer Grünschnitt, Wiesendrusch und Ausbürstmethode. Die Ernte des frischen Grünschnittes erfolgte am Morgen in feuchtem Zustand. Der Wiesendrusch wurde mit einem Parzellendrescher (Breite: 1,50 m) geerntet. Das Ausbürstmaterial wurde mit einem Seedstripper mit einer Breite von 1,20 m geerntet. Die Bürste rotierte aufwärts und wurde in einer Höhe von 15 cm fixiert. Der Grund für diese Höhe waren die niedrigsten reifen Samen. Die geernteten Saatgutmischungen wurden luftgetrocknet und vor der Lagerung mit einem 6 mm Sieb grob gereinigt.

Die Lagerung des Saatgutes erfolgte bei Raumtemperatur (18-23°C), im Kühlraum (2-5°C, mit 40 % relative Luftfeuchtigkeit) und im Gefrierschrank (-18°C). Nach der Ernte des Saatgutmaterials wurde versucht, eine geeignete Methode zur Bestimmung der Qualitätskriterien zu finden. Die erste große Herausforderung dabei war es, eine Probe des heterogenen Saatgutes zu ziehen. Es

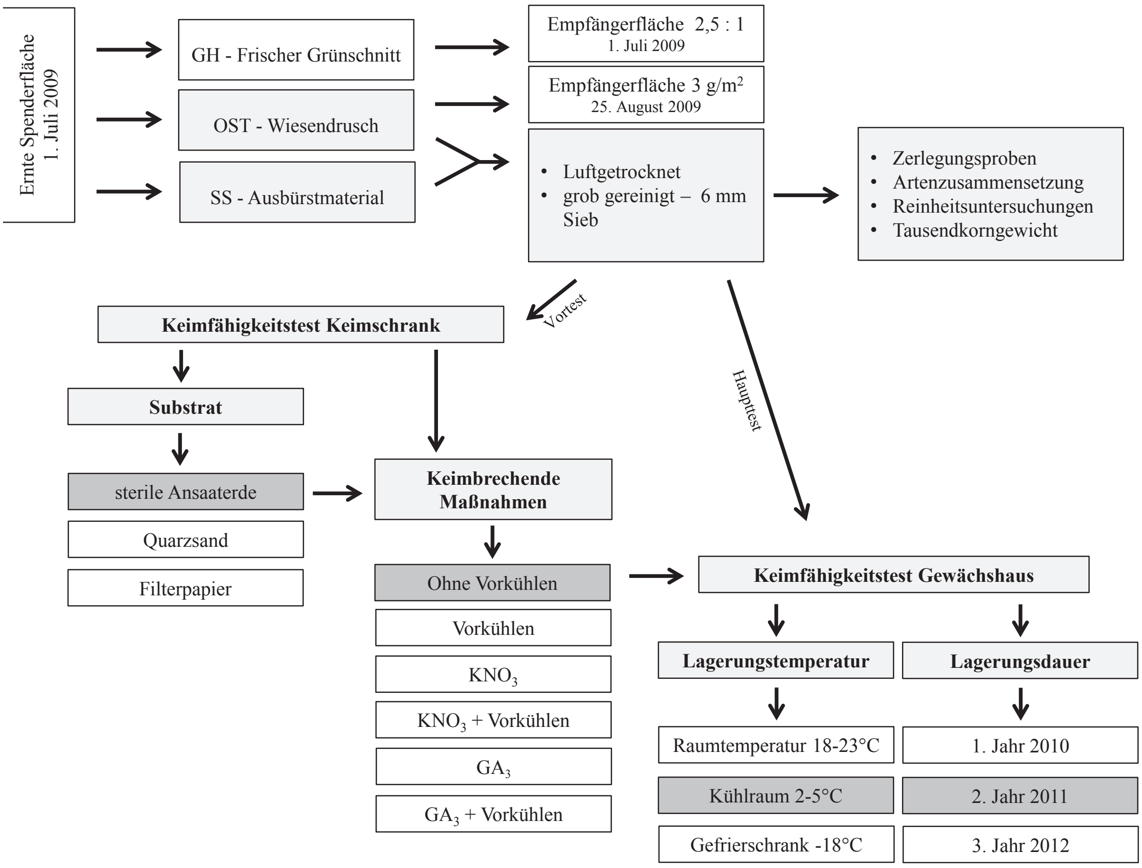

Abbildung 4: Versuchsdesign für die Bearbeitung der Kapitel 2 und 3. Bei den Keimfähigkeitstests sind jene Methoden, mit denen weitergearbeitet wurde dunkelgrau eingefärbt. Mit den hellgrau eingefärbten Erntemethoden wurden die Qualitätskriterien erarbeitet.

wurden Einzelproben aus unterschiedlichen Tiefen der Aufbewahrungssäcke gezogen und zu einer Mischprobe zusammengefasst. Um eine repräsentative Probe zu erhalten, wurden die Proben immer wieder geteilt und verkleinert. Aus der Mischprobe wurden 5 g Proben (n=25) gezogen, die im Labor in Spreu und Samen getrennt wurden. Daraufhin wurden deren jeweilige Anteile bestimmt. Für die Bestimmung des Tausendkorngewichtes wurden achtmal 100 zufällig ausgewählte, volle Samen gezählt und gewogen (ISTA, 2011).

Im ersten Jahr nach der Ernte wurden die Keimfähigkeitsversuche (Vortests) im Phytotron durchgeführt. Es wurden jeweils 4x100 Samen angekeimt. Insgesamt wurden zwei Erntemethoden (Wiesendrusch und Ausbürstmaterial) auf drei unterschiedlichen Substraten, nämlich sterile Ansaaterde, Quarzsand sowie Filterpapier, getestet. Im nächsten Schritt wurde festgestellt, ob keimbrechende Maßnahmen, wie Vorkühlen, Einsatz von Kaliumnitrat (KNO_3) oder Gibberellinsäure (GA_3) notwendig sind, um ein besseres Ergebnis bei der Keimfähigkeit zu erzielen. In der Literatur (Godefroid et al., 2010, Parsons et al., 2013, Shepley et al., 1972, Williams, 1983) wird darauf hingewiesen, dass solche Maßnahmen notwendig sein können, um die Keimhemmung aufzuheben. Die Ergebnisse der Tests zeigten, dass mit dem Substrat sterile Ansaaterde weitergearbeitet werden konnte und keine keimbrechenden Maßnahmen notwendig waren. Um zu testen, ob die Lagerung bei unterschiedlichen Temperaturen einen Einfluss auf die Keimfähigkeit hat, wurden

Wiesendrusch und Ausbürstmaterial drei Jahre lang bei unterschiedlichen Temperaturen, nämlich bei 18-23°C (Raumtemperatur), bei 2-5°C (Kühlraum, 40 % relative Luftfeuchtigkeit) und bei -18°C (Gefrierschrank) gelagert (Rao et al., 2006). Für die Haupttests wurde im Gewächshausversuch eine definierte Menge (4 x 1,25 g) der Samenmischung in Saatschalen (ein Äquivalent von ca. 3-5 g/m^2 reine Samen) auf steriler Ansaaterde in vierfacher Wiederholung ausgebracht. Einmal wöchentlich, über eine Zeitspanne von vier Wochen, wurden die gekeimten Pflanzen gezählt und aus den Schalen entfernt. Die ausgezählten Pflanzen wurden in Monocotyledone und Dicotyledone getrennt. Die meisten Samen keimten in den ersten zwei Wochen, daher reichte die Dauer von vier Wochen für die Auszählungen, um ein aussagekräftiges Ergebnis zu erzielen. Diese Tests wurden jährlich drei Jahre lang wiederholt, um die Lagerfähigkeit des Saatgutes zu prüfen.

In **Kapitel 4** wird gezeigt, wie eine ökologisch hochwertige Glatthaferwiese beerntet und auf einer Empfängerfläche neu etabliert werden kann. Glatthaferwiesen gelten als naturnahe Wiesen und zählen zu den artenreichsten Wiesen Mitteleuropas. Es wurden zwei unterschiedliche Ernte- bzw. Etablierungsmethoden, nämlich der Transfer von frischem Grünschnitt sowie von Wiesendrusch, getestet.

Die Empfängerfläche, auf der das getestete Saatgut ausgebracht wurde, befand sich im Ennstal in Gumpenstein (47°29‘N, 14°06‘ E; 740 m m.ü.A.). Vor der Ansaat wurde die Fläche tiefengepflügt, damit die Begrünung auf diasporenarmem Material erfolgte und der Konkurrenzdruck so gering wie möglich war. Der frische Grünschnitt wurde sofort nach der Ernte im Verhältnis 2,5 : 1 (Spenderfläche : Empfängerfläche) auf die Empfängerfläche ausgebracht. Der Wiesendrusch wurde am 25. August 2009 in einer Saatstärke von 3 g/m^2 angesät. Im Herbst 2009 erfolgte ein Reinigungsschnitt. Die pflanzensoziologischen Erhebungen erfolgten zweimal jährlich in den Jahren 2010, 2011, 2012 jeweils kurz vor der Mahd. Dazu wurde die Gesamtdeckung mit Vegetation auf den Versuchsparzellen geschätzt und aufgenommen. Der Anteil des offenen Bodens und der mit Steinen bedeckten Fläche summiert mit der Gesamtdeckung der Vegetation muss dabei die Zahl 100 ergeben. Es wurde keine Überschätzung oder Schätzung in unterschiedlichen Ebenen, wie es bei der Flächenprozentschätzung üblich ist (Schechtner, 1958, Weinzierl, 1917), durchgeführt. Es handelte sich also um eine Schätzung der projektiven Deckung, auch „sichtbare Deckung" genannt. Eine geschlossene Vegetationsdecke liegt nur dann vor, wenn sich der Wert 100 ausschließlich durch die Vegetation ergibt (Pötsch, 1997). Nach der Mahd ließ man das Schnittgut für ein bis zwei Tage auf der Fläche trocknen, bevor es von der Fläche entfernt wurde.

Die bisherigen Ergebnisse zeigen, dass sich bei der Wiesendruschvariante bereits nach drei Jahren 20 Zielarten und bei der Grünschnittvariante 16 Zielarten etabliert haben. Beide Methoden sind geeignet, um ökologisch hochwertige Glatthaferwiesen neu anzulegen. Der Beitrag, welcher Kapitel 4 behandelt, wurde 2013 beim „17th Symposium of the European Grassland Federation „The Role of Grasslands in a Green Future. Threats and Perspectives in Less Favoured Areas", in Akureyri/Island, publiziert.

Publikationen

Kapitel 1

“Establishment and Use of High Nature Value Farmland”

Zitat:

B. Krautzer, B., Bartel, A., Kirmer, A., Tischew, S., Feucht, B., Wieden, M., Haslgrübler, P., Rieger, E. and Pötsch, E.M. (2011): Establishment and use of high nature value farmland. Grassland Science in Europe, Vol. 16, “Grassland Farming and Land Management Systems in Mountainous Regions”, edited by E.M. Pötsch, A. Hopkins and B. Krautzer, Agricultural Research and Education Centre Raumberg-Gumpenstein, 457-469.

Establishment and use of High Nature Value Farmland

Krautzer B.[1], Bartel A.[2], Kirmer A.[3], Tischew S.[3], Feucht B.[4], Wieden M.[5], Haslgrübler P.[1], Rieger E.[4] and Pötsch E.M.[1]

[1] *Federal Agricultural Research and Education Centre Raumberg-Gumpenstein, 8952 Irdning, Austria*

[2] *Umweltbundesamt, Spittelauer Lände 5, 1090 Wien, Austria*

[3] *Hochschule Anhalt, Bernburger Straße 55, 06366 Köthen, Germany*

[4] *Rieger-Hofmann GmbH, In den Wildblumen 7, 74572 Blaufelden-Raboldshausen, Germany*

[5] *Verband deutscher Wildsamen- und Wildpflanzenproduzenten e.V., Wetzlarer Straße 11, 35581 Wetzlar-Münchholzhausen, Germany*

Corresponding author: bernhard.krautzer@raumberg-gumpenstein.at

Abstract

The conservation and maintenance of biodiversity on agriculturally used areas has become a special concern of agrarian and environmental policy. Therefore, restoration projects with the objective of creating semi-natural grassland have become of increasing importance throughout Europe in recent years. Procedures that are as close to nature as possible have gained special significance. Species-rich semi-natural grassland is the only existing natural source that can provide the source material for restoration and reintroduction of High Nature Value Farmland (HNVF). In recent years, a large number of different harvesting methods and application techniques have been developed for exploitation and application of seed and plant material of regional semi-natural grasslands. In order to ensure and guarantee its use according to nature protection targets throughout Europe, binding European guidelines and an approved certification procedure for such material have to be developed.

Keywords: semi-natural grassland, ecological restoration, biodiversity

Introduction

All over Europe, agricultural intensification and, additionally, the abandonment of large areas, led to a strong decrease in biodiversity (Pötsch and Blaschka, 2003). The 1992 Rio de Janeiro Convention on Biological Diversity and recent EU regulations promote the protection of biodiversity and seek to address the strong reduction in biodiversity in Europe. To implement this aim, the availability of regional, native plant material is of extreme importance. This requirement is not sufficiently met in Europe, where seed of native ecotypes is still seldom available in large amounts. Extensively managed semi-natural grasslands, which are the most widespread type of High Nature Value Farmland (HNVF), can be regarded as the most important seed source. They are normally rich in species of native provenance and for this reason can be harvested to obtain valuable seeds useful for restoration and revegetation. The typical high diversity of HNV grasslands, in terms of species and site conditions, is their strong point but, at the same time, they pose the main challenge for an economically efficient harvesting system. Moreover, the normally used techniques to create forage meadows or to revegetate degraded areas with the help of commercial seed mixtures are not comparable to ecological restoration done with seed material from semi-natural grassland.

In 2009, an EC-funded Central-Europe project started in order to promote High Nature Value Farmland (HNVF) as a valuable resource to support sustainable rural development. As a main target, the project "Semi-natural Grassland as a Source of Biodiversity Improvement"

(SALVERE) intends to contribute to the practical realisation of EU regulations regarding biodiversity by utilising semi-natural grasslands as potential donor sites of seed to be used for the establishment of HNV areas (Scotton, 2009; SALVERE, 2011). Based on experiences made and information gained within this project, we present in this paper a short overview on the current situation of HNVF in Europe, the current state-of-the-art in the selection of donor sites, the exploitation of seed material, techniques and know-how for the establishment of semi-natural grassland, as well as the existing the still-necessary regulations.

Definition, relevance and state of development of High Nature Value Farmland (HNVF)

Since 2000 the agro-environmental indicator "High Nature Value Farmland" (HNVF) has been discussed and developed at the European scale, centred on the IRENA -Indicator No. 26 (EEA, 2006). Originally developed as an indicator referencing for the importance of certain farming practices for biodiversity in cultivated landscapes, it gained importance and relevance in 2005 as it was selected as an indicator for the Common Monitoring and Evaluation Framework (CMEF) of Rural Development Programmes (RDPs) according to Council Regulation (EC) No 1698/2005. Member states are obliged to report on the national area and maintenance of HNV farming and forestry for the mid-term evaluation in 2010 as well as to the *ex-post* evaluation of the Rural Development Programmes in 2015 (EC, 2006). According to the CMEF, HNVF is used as a "Baseline Indicator" for reference at the beginning of the RDPs, followed by an interpretation as "impact indicator" and as "result indicator".
Proposals for defining and mapping High Nature Value Farmland have been developed by the European Environment Agency (EEA) together with the Joint Research Centre (JRC) since 2003 (Andersen *et al*., 2004; EEA, 2005; JRC/EEA, 2006). In 2007 a report and separate guidance document to the Member States on the application of the HNVF indicator was published on behalf of the European Commission, DG Agriculture (IEEP, 2007; EC, 2009).
Following this document, the core of the HNVF concept is the link from management practices to biodiversity dependent on farmland habitats. Thus, the concept of HNVF can be seen as a two-fold approach: looking on the one hand to the state of the resource in terms of quantity and quality, and on the other hand to the driving forces, i.e. management practices that produce, influence and maintain the natural values.

The resource HNVF

From the ecological point of view, High Nature Value Farmland is a concept that may lead the focus on certain farmed areas, and which tend to be marginal in terms of their agronomic production capacity and to be outside of market-oriented policy interests. It raises the awareness to large areas of Europe used as extensive grassland, or in a diverse mosaic of small landscape elements and low intensity use. HNVF is defined as follows:
"High Nature Value farmland comprises those areas in Europe where agriculture is a major (usually the dominant) land use and

- where that agriculture supports or is associated with either a high species and habitat diversity, or
- the presence of species of European, and/or national, and/or regional conservation concern,
- or both."

Those areas have high overall biodiversity and landscape value, and are dependent on regular use, often in a traditional way. They have been seen as the ecological backbone of European cultural landscapes. Three types of HNVF are recognized (Andersen *et al*., 2004; IEEP, 2007; EC, 2009):

- Type 1 - Farmland with a high proportion of semi-natural vegetation.
- Type 2 - Farmland with a mosaic of low intensity agriculture and natural and structural elements, such as field margins, hedgerows, stone walls, patches of woodland or scrub, small rivers etc.
- Type 3 - Farmland supporting rare species or a high proportion of European or world populations.

However, because of their low agro-economic value, those farming systems are prone to abandonment or - whenever possible - intensification; for example, through irrigation and fertilizing. Both development paths would endanger the natural values. The concept of HNVF pulls those systems from behind the curtain and seeks to make them a topic in public discussion. The future agricultural policy is asked to pay attention to those extensive, large, and potentially threatened farming systems and areas. Policy should support agriculture in a way that those farming systems can be kept up and natural values can be maintained, even in a competitive agricultural surrounding.

The indicator HNVF

In the Evaluation Framework HNVF is seen as an indicator, against which the effectiveness and efficacy of the Rural Development Programmes should be tested. This requires a more operational definition of HNVF and a decision about what HNVF is and what it is not. Although theoretically well elaborated in different studies, this separation is not easy in practice, and may it have great implications on the resulting HNVF area.
Due to the diverse situation in member states regarding data quality and availability, and important differences in ecological conditions as well as in farming practices, a number of different approaches for the implementation of this indicator have evolved. Each state has reported its own baseline figure using different information sources and applying adapted criteria for the generation of the required area numbers. But those numbers are not really comparable throughout Europe because they are based on diverse methodologies. Some states apply a mapping concept, e.g. Germany, which tries to calculate the HNVF area through the monitoring of a number of stratified random sample plots. Others like France and Finland use a typology of their farms and evaluate the farming systems. The area calculation is largely influenced by statistical analysis of farm data and modelling of relationships. If land-use data are available in sufficient detail and completeness, the area can be calculated drawing on information systems like IACS (Integrated Administration and Control System) or LPIS (Land Parcel Information System), e.g. is done in Austria and Greece. Thus the required parcels can be selected through the application of criteria from land cover and management and summed up to the total area.

HNVF as a policy tool

At the policy level, HNVF has gained importance with its selection as an indicator for the evaluation of RDPs. The IRENA-process and studies done subsequently by the European Environment Agency (EEA) tried to determine the HNVF area for each member state. A map was produced showing the probability of HNVF throughout Europe. The intention of this map was to create an overview on the situation in Europe and more the kind of a target-identification for necessary policy support in those regions. When DG-Agriculture and regional development took over and defined the CMEF indicator in 2005, the understanding of its concept was still fuzzy and the method for implementation not well defined. Meanwhile it has developed towards a monitoring and evaluation approach, but there is still some obscurity on the target of the evaluation - farming practices at farm level, farming systems in terms of farm typologies,

agro-environmental measures and RDPs, or the biodiversity at the landscape level? Therefore, as mentioned above the implementation in member states shows great differences according to what the national emphasis is on. The use of this HNVF indicator as a trigger for European policy measures such as financing would need a lot of harmonization and coordination work. It does not seem feasible to reach a Europe-wide integrated CMEF indicator within the next few years. Nevertheless HNVF, and in particular the reported changes over the programme period, will definitely serve as a reference for the programme evaluation and thus influence the development of the next RDP periods.
However, the concept of HNVF has triggered a process in the political discussion. The values of certain low input/low output farming systems have moved into the public view and the concept of ecosystem services focuses on additional societal benefits of agriculture besides the agricultural production. In this context High Nature Value Farmland stands for valuable nature and bio-diversity.

Aims of semi-natural grassland restoration

Currently, 76% of grasslands of European interest are assessed as being in an unfavourable conservation status (EU2010 Biodiversity Baseline Report). Therefore, the protection of natural grasslands containing regional sub-species and ecotypes in region-specific compositions is of top priority in nature conservation. To reach this goal, not only the high ecological and aesthetic values of species-rich grasslands should be acknowledged but also their potential as donor sites for regional seed mixtures.
In general, restoration of species-rich grasslands is limited by several abiotic and biotic constraints. The success of restoration measures depends on abiotic factors such as nutrient status, pH-value of soil, and hydrology, as well as the availability of appropriate seed sources. Hence, restoration success is impeded by depleted seed banks of restoration sites, decrease or loss of target species in the surroundings and limited dispersal in fragmented landscapes. Early restoration efforts in the 1970s and 80s were mostly focused on the removal of nutrients, re-wetting and the re-introduction of an adequate management. In many cases such measures alone were frustratingly unsuccessful and did not lead to the re-establishment of target communities even after successful lowering of nutrient levels and productivity (Bakker and Berendse, 1999). Therefore, the introduction of target species is of decisive importance for restoration success. Seed mixtures directly harvested in genuine, natural grasslands can be used in ecological restoration, thereby contributing to the preservation and enhancement of regional biodiversity. Since the 1990s, different methods for ecological restoration have been used successfully by several working groups all over Europe (for reviews see Walker *et al*., 2004; Kirmer and Tischew, 2006; Klimkowska *et al*., 2007; Kiehl *et al*., 2010).

The most important grassland types and their suitability as donor-sites

Seed mixtures should be harvested in species-rich grasslands containing a species composition typical for the selected target community and for the region concerned. It is decisive to choose donor and receptor sites with similar site conditions (hydrology, substrate, nutrient status) to ensure that the plant species are optimally adapted to local climatic and edaphic conditions. Especially hydrology and nutrient status are decisive parameters to determine suitable donor communities:

- dry, nutrient-poor to mesotrophic sites: dry grasslands (Bromion)
- moist, mesotrophic sites: mesic grasslands (Arrhenatherion)
- wet, nutrient-rich sites: eutrophic floodplain grasslands (Deschampsion)
- wet, nutrient-poor sites: oligotrophic floodplain grasslands (Molinion) and fen grasslands

Criteria for the selection of donor sites

The main obstacle for the implementation of near-natural revegetation methods is the identification of suitable donor sites for seed harvesting. In Germany, donor-site registers have already been established in four federal states: Saxony-Anhalt (Hefter *et al*., 2010), Thuringia (Kirmer and Korsch, 2009), Schleswig-Holstein, and North Rhine-Westphalia.
For example, in 2003 the first donor-site register was installed in Saxony-Anhalt. The internet-based database comprises open habitats and grasslands with high nature conservation value, suitable for harvesting seeds and seed-rich plant material. The listing of an area as a donor site in the database does not include permission to harvest seeds. Any kind of harvesting (e.g. mowing, threshing, collecting seeds) requires a formal authorisation through the respective nature conservation authority and the approval of land owners and users. At the moment, the database contains almost 400 potential donor sites. It is embedded into an information system of nature-oriented greening measures (www.spenderflaechenkataster.de). This internet platform presents an overview of different restoration methods and gives information for their planning and implementation, as well as for the costs and the legal framework.
The internet-based database offers users multiple research functions for finding suitable donor sites, e.g. a general map and a search module. The donor site register allows a quick assessment of the suitability of potential donor sites according to nature conservation values and economic aspects. Registered donor sites must fulfil specific criterions:
- representative species composition (typical for the vegetation type and the region)
- low amount of problematic species (neophytes, strong competitors)
- not established or modified with standard seed mixtures containing cultivars from propagation
- ± regular management (preferably mowing)
- no change of use expected

Such a database enables an efficient inquiry about suitable donor sites and facilitates planning and realization of nature-oriented greening measures (e.g. harvesting of seeds via mowing, threshing, brushing, vacuuming). The use of species-rich donor sites in restoration or revegetation measures supports habitat protection, protects the biological diversity and preserves the floristic and genetic identity of the region.

Harvesting methods for site-specific seed and plant material

The selected grasslands may only contain a very low amount of problematic or neophytic species. The optimal harvesting time is when most target species have set seeds. In Arrhenatherion communities, a first harvest can be done between end of June and end of July. If the site was mown in early May, the harvesting cut can be delayed until the end of August. Bromion communities are harvested best between mid-July and beginning of September. Seed harvest in Molinion and Deschampsion communities should be done between end of August and end of September because of late-fruiting target species (e.g. *Cnidium dubium*). An additional harvesting cut in May is recommended to transfer early flowering species (e.g. *Cardamine* spp., *Ranunculus* spp.). In general, a later and/or second cut favours the transfer of herbaceous species whereas an early and/or first cut favours grasses. If harvesting time and method are different to the normal management regime, the site should not be harvested every year.
A lot of different harvesting techniques, partly well known for centuries and partly developed during the last decades, are used for the exploitation of regional plant and seed material (Krautzer *et al*., 2004; Kirmer and Tischew, 2006; Krautzer and Pötsch, 2009; Kiehl *et al*., 2010). The most common processes and methods are summarised below.

A widely used method is the mowing of suitable donor sites at the time when most of the desired species are at an optimum stage of seed maturity (June - August). To avoid excessive seed losses, the material is cut preferably early in the morning when it is moist with dew, and then immediately taken to the restoration area (receptor site) for distribution. Another possibility is to dry and store the mown material for later use. Nevertheless, this method requires increased manipulation and therefore higher costs. In addition, a large part of the seed material may be lost (ÖAG, 2000). The hay-flower sowing method uses seed-rich remains from the threshing floors of hay barns, which sometimes keep sufficient seed quantities and qualities.

With brushing and threshing methods (Edwards *et al*., 2007; Jongepierova *et al*., 2007; Scotton *et al*., 2009) site-specific seeds can be collected from suitable donor sites. To obtain the greatest possible number of mature seeds from the preferred species, particular attention has to be paid to the harvesting time. Seed mixtures with the highest species diversity are generally achieved by consecutive harvesting of donor sites according to species-specific seed maturation rates and schedules. In the Alps for example, seed yields are usually between 50-150 kg ha-1. The relationship of donor area to restoration area thus varies from approximately 1:1 to 1:4. If application of threshed seed material is not possible immediately after harvest, it must be dried and stored at a dry location.

A good method that is currently practised in several countries is the nursery or large-area production of seed of suitable species with agricultural and horticultural techniques. Above all, species that are used often and in large amounts can be produced at comparatively reasonable costs and implemented on appropriately large project areas. This method, for example, has been used successfully in Austria, Germany and Switzerland for restoration projects (Krautzer and Wittmann, 2006; Kirmer and Tischew, 2006; Rometsch, 2009). Similar approaches are now being implemented in the French Pyrenees (Malaval, 2006) in Iceland (Aradottir and Johannsson, 2006) and latterly in Norway (ECONADA, 2011).

In cases of land use change, the transfer of seed-rich top soil (mainly the first 5 cm, and at most the top 20 cm) from suitable donor sites is an occasionally used method, especially in cases of technical interventions (e.g. road construction, landscaping). Another possibility is the transplanting of turfs, in which soil-plant segments from donor sites to restoration sites are being transferred. Wherever possible the transplanting of turfs should take place as early as possible at the beginning of the vegetation period or after the start of the autumn vegetation pause, thus just after the melting of snow or directly before the onset of winter. With proper planning, grass turfs from building and construction sites can be directly transferred to restoration sites without intermediate storage (Krautzer and Klug, 2009).

Quality of native seed material

Exploitation, production and trade of regional seeds without any common rules lead to an unmanageable market for consumers. Wild forms compete against cultivars of the same plant species. Among declared “wild seed products” one will find a wide range of labels in terms certifications, assertions, documented provenances and qualities. On behalf of nature conservation, a system of rules is needed in order to support transparency of a European wild species seed market. On the other hand, seed consumers expect some minimum thresholds for quality aspects related to the composition of harvesting or propagation material, the concentration of pure seeds in harvesting materials and their germination capacity. Therefore, also a sufficient declaration on such quality aspects is important if native seed material is offered on the market.

Quality in terms of nature conservation

The idea of trading wild seeds is due to the consideration of a regional limitation of introducing wild plants as a crucial point of genetic adaptation. The commercial seed market offers several interesting species suitable for restoration, but they are generally to be described as being of non-local provenance. Through negative interaction with still-available local provenances their introduction may lead to undesired results such as hybridisation or displacement (Kirmer and Tischew, 2006). Only harvesting material and seeds that are collected, propagated and used in the same region will ensure ecosystem services, which will not be provided by cultivars and non-local propagation material (Blaschka *et al*. 2008). Therefore there is a need to define biogeographical regions to fulfil those benefits.
However, in Germany, Austria and Switzerland a sufficient definition of seed zones already exists (VWW, 2011; REWISA, 2011; CPS, 2009). One of the most important aspects is the non-conformance of those biogeographical boundaries with political ones! However, a well-defined national system of seed zones is inadequate when transnational trade occurs. Nowadays, the defined regions end at the borders of the member state, even though the physiographic province extends into the neighbouring country. A basis for a (still missing) international definition of European biogeographical regions could be the already existing system at the European Environment Agency (EEA, 2009). However, for a functioning European market-system with a regional supply of wild seed, transnational zones for production and use of native seed material have to be defined.

Quality in terms of consumers' expectations

Contractors are interested to get sufficient information about the quality of sowing material, especially in terms of seed proportion and germination capacity. Corresponding data are particularly in demand for large-scale restoration projects and trade. The viability or cost-effectiveness of the necessary assessments has to be proven from case to case.
The actual number of seeds in fresh green hay, hay mulch, stripped material or threshings, as well as the expenditures connected to the exploitation of the material, is dependent on various factors, such as the type of meadow, management, time of day, harvesting time in the course of the year, potential seed production and mechanisation (see Table 1).

Table 1. Share of grasses and herbs, amount of harvested seeds and expenditure of time for differing harvesting methods in Arrhenatherion and Molinion communities (expenditure for drying and cleaning is not included)

Harvesting method	Harvest time	Grasses:Herbs [%]	Pure seeds harvested [kg/ha]	Duration [h/ha]
Fresh green hay	End of June	80:20:00	100-200	1-2h*
Hay mulch	End of June	70:30:00	40	3-4h**
Threshings (plot thresher)	End of June	80:20:00	60-150	5-10***
Threshings (large thresher)	End of June	60:40:00	50-200	1,5-3*
Stripped seeds	End of June	80:20:00	20-60	1,5-3***

*depending on technical equipment; **including work processes for the drying of hay; ***depending on vegetation type

The species number and the composition of the harvested material are strongly dependent on the type of vegetation. Another influencing factor is the harvesting date. Later harvesting generally deceases the share of grasses in the mixture and thus fosters the establishment of herbaceous species (Hölzel and Otte, 2003). A harvesting date set too early hinders the full development of the seeds.

The assessment of purity, thousand-seed weight and germinating capacity of seed material harvested on donor sites is very complex and costly. Therefore, such information in practice will only be collected if the material is sold on the market or used at a large scale. However, determination of the purity of the harvested seed and plant materials is important to ascertain the volume of pure seeds that are contained in the material, which then defines the actual seed capacity of the entire material. The composition and quality of hay, hay mulch, stripped material or threshings differs greatly from year to year. The share of chaff and impurities, such as earth, can be very high.
Assessments on the germination capacity of harvested material are still in progress. First results from the SALVERE-project group indicate germination capacities between 40 and 70% from Arrhenatherion meadows. On meadows with a high share of species with seed dormancy (e.g. litter meadows), the actual germination capacity of harvested seed material can decrease notably (Haslgrübler, 2011).

Site preparation on receptor sites

A first step in grassland restoration and establishment and an important factor for restoration success is the site assessment and site preparation on receptor sites, thus creating optimal conditions for germination and establishment of introduced species. The special demands and threats of the habitat to be created, in terms of soil properties, nutrient supply, erosion tendency, competition phenomena with other plant species, sowing and planting time, availability of the seed and plant materials, etc., are to be determined as exactly as possible (ÖAG, 2000). Therefore, the choice of proper techniques for harvesting and application of species-rich grassland requires an assessment of the main factors of natural geographic region, climate, soil, erosion risk and other possible restoration targets (e.g. agricultural utilization, use as recreational area).

Site preparation in terms of regenerative measures

For successful species introduction into species-poor grassland, the sward has to be cut to a height of 3-5 cm, if necessary. Subsequently, the sward has to be opened. For large-area treatment, the use of curry comb, harrow, rotary hoe or flail chopper is recommended. During recent years, different specialised machinery for grassland regeneration has been developed and is available in grassland dominated areas. Several assessments showed that the stronger the intervention and disturbance of the sward, the higher the rate of successful species establishment (Walker *et al*., 2004, Hölzel *et al*., 2006).

Site preparation of arable land or ploughed grassland

The turning of the soil via ploughing or rotary hoeing is the standard method for the restoration of former intensively utilized grassland or arable land. Those soils are generally characterized by a high concentration of plant-available nutrients. One simple but time-consuming method to impoverish the soil is crop production period of 1-2 years without any fertilization.
Especially, restoration areas formerly used as arable land can potentially contain enormous amounts of weeds. Timely harrowing of soil under dry conditions fosters the accumulation of annual weeds which can then be combated mechanically by harrowing or grubbing several times before sowing. In humid regions, dry weather conditions are especially necessary for success when using these measures. In more continental regions with low precipitation, the germination of weeds from the soil seed bank may depend on moist conditions after grubbing. If those recommended methods of mechanical weed control are not applicable, the use of low persistence herbicides (e.g. glyphosate) could be considered (Pywell *et al*., 2007).

Sites with very nutrient-rich and weed-infested topsoil (particularly soil from arable land) can be very positively influenced by preliminary deep ploughing or topsoil inversion. To be used here could be a so-called trench excavator (deep plough), which requires a very powerful tractor. Thus the soil will be turned over to a depth of 40 cm to a maximum of 80 cm. Nutrient-rich and seed-rich layers are replaced and nutrient-poor substrate is turned up. The use of a trench excavator is not always permitted (e.g. Federal Soil Protection Act in Germany).

Site preparation after technical intervention

Many receptor sites requiring subsequent restoration are created through infrastructural interventions. Ground work (soil removal, intermediate storage and creation of an appropriate substrate layer) must only be carried out when the soil is suitably dried and during appropriate weather conditions. Soils with a clay content of over 30% are especially prone to soil compaction and are to be handled accordingly with care (BMLFUW, 2009). The general decision on the re-use of the topsoil-layer, with respect to its thickness, will depend on the content of nutrients and/or seeds of weeds and unwanted species. The extent of the applied soil layer, the space in which roots can penetrate, the water-storage capacity and the nutrient content of the substrates can be appropriately assessed during planning, and adjusted according to the desired type of vegetation (or vice versa).

Establishment of semi-natural grassland

Practically relevant restoration of semi-natural grassland has been successfully realised on the most differing sites for many years in different European countries (examples given in Kirmer and Tischew, 2006; Donath *et al*., 2007). The selection of a suitable method depends on the given aim (e.g. erosion prevention, development of extensive vegetation, compensation measures) and the site conditions of the receptor site. In general, the restoration method to be selected is that which enables the desired target community to can be developed with the least possible expenditure. Availability, practicability, costs, possible subsequent use and maintenance are to be taken into account. Fundamentally, the method should be adapted to the particular areas of origin to take into account climatic conditions and also the life cycle of insects, which are adapted to the regional blossoming period and special content material of plants local to an area.

A lot of successful techniques and strategies for the establishment of semi-natural grassland have been developed during the last years. Table 2 gives an overview of the most recommended techniques and materials depending on the most common initial situations. The use of seed-rich top soil or plant material from donor sites is, in practice, reduced to the rare situations where valuable donor sites are destroyed during construction work.

Under moist climate conditions, as well as in mountainous areas, restoration with seeds or seed mixtures should take place at the beginning of the vegetation period to make optimum use, on the one hand, of the winter moisture on drier sites, and on the other hand to guarantee development of the seedlings into plants that are capable of surviving the winter during the vegetation period. But in principle the application of extensive grassland areas throughout the entire vegetation period is possible, whereby persistent dry periods (e.g. in high summer) can lead to failures. In practice the time of restoration is generally in late summer to early autumn because, in that period, construction measures are to a great extent completed. According to the authors' experience, moist conditions and deep topsoil applications favour the development of grasses, whereas herbs have an advantage on nutrient-poor and dry sites.

Table 2. Strategies for the establishment of semi-natural grassland.

Initial situation	Materials	Recommended techniques
species-poor grassland	propagated regional seeds	overseeding device
	sieved threshings	band rotavator
ploughed grassland/arable land/ fallows	green hay	load wagon and manual or
	hay mulch	mechanical distribution
	threshings	rotavator
	hay flower	agricultural sowing and spreading devices
	propagated regional seeds	cover crop seeding
raw soils (e.g. road construction, landscaping, open cast mining areas)	green hay	load wagon and manual or
	hay mulch	mechanical distribution
	threshings	agricultural sowing and spreading devices
	hay flower	cover crop seeding
	propagated regional seeds	hydro-seeding
raw soils endangered by erosion	green hay	mechanical or manual distribution
	hay mulch	
	threshings	recommended seeding technique plus
	hay flower	additional protection by
	propagated regional seeds	a mulch layer or geotextiles

Many species of the extensive litter meadows (fen meadows, litter meadows, etc.) are so-called frost germinators. Therefore, with these types of vegetation sowing in winter has proved successful, whereby the seed must be sown from the middle of November to the beginning of December as long as the soil has no snow cover (Krautzer and Klug, 2010).

Restoration success

Semi-natural, species-rich grasslands are generally created over a very long period through extensive forms of use. Achieving the strived-for target state is therefore only possible through appropriately adapted utilization over a long period, sometimes after a decade or even longer. It is important that in the first year following the application as many grassland species as possible are regularly germinated and young plants are to be recognised. Some types of grassland species will appear only in the second or third year after the application or become visible even later, because their seeds have a distinct dormancy or the young plants are very difficult to find. But on no account should a high share of problematic species, such as common couch grass, creeping thistle, dock species or white clover be visible. The cover of grasses should generally be not too high, and before the first cut should not exceed 40-60%. The share of various functional groups should also be in a balanced ratio (grasses, legumes, other herbs). For most vegetation types, the vegetation cover should have achieved 40-60% after the first vegetation period, depending on vegetation type, to guarantee a receptor state. If this is not the case, subsequent sowing is required.

With increasing development time, the degree of cover derived from target species and the increasing similarity to the reference- or target state is decisive for success of the measures. The success of sowing (restoration) is decisively influenced by conditions on the receptor area. In the first year after the application, according to vegetation type (moist meadows, litter meadows, semi-dry grassland) the transfer rate is about 30-50%. On raw soil the transfer rates are generally higher and can reach 60% in the first year after the application. The transfer rate is dependent on differing factors, e.g. quality of the seed, soil preparation, site conditions, weather after the application, natural seed potential of the soil (weeds) and restoration method.

Regulations and implementation in Europe

To protect the market for licensed varieties, the important fodder plant directive was launched in 1966 (EEC, 1966). With some amendments it is today the main directive, which causes problems between many national nature conservation laws and those for seed breeding protection. In 2010 a new Commission directive was passed, which approves the trade of a small amount of 5% of „wild“ seeds among the cultivars. The European member states have to implement the directive by the end of November 2011 (EEC, 2010). This is the latest time for the start of a competition in trade between wild seeds and cultivars. There are only few points in the new directive supporting the use of wild forms but many formal conditions, like detailed registrations and declarations of every single mixture, which will hamper the development of a wild-seed market. To improve the situation of semi-natural grassland in all European member states, it is inescapable to start activities according to those directives, like careful implementation into national laws to protect the initiatives dealing with native seeds that are in the process of emerging. Member states should also start to influence the recently started process of a review of the European seed legislation. At a national level, only Germany adapted its nature protection law in view of the harvest, propagation and trade of native seeds (BNatSchG, 2010).

Prospects for the future

Semi-natural grasslands are the most important category of High Nature Value Farmland and provide a high level of biodiversity. Due to land abandonment and intensification this type of grassland is seriously endangered; the maintenance and development of semi-natural grassland has, therefore, become a special concern of agrarian and environmental policy. Semi-natural grassland can also be used as a natural source of biodiversity for different purposes and can itself contribute to the development and restoration of High Nature Value Farmland.
Ecological restoration of semi-natural grassland is a relatively new field of activity, and as a result there are still considerable gaps in our knowledge and know-how. Approaches to the technical aspects vary considerably, and the development of special restoration methods, especially for extreme site conditions, is partly far from sufficient. The legal standards and requirements also vary greatly from one country to another. What is commonly accepted or promoted in some countries is strictly forbidden in others. Above all, despite prohibitions and restrictions written into various nature-protection laws, the use of non-native plant species is often ignored or overlooked due to lack of the knowledge about alternatives or ability to properly identify plant material being offered for sale or used on site. There is also a considerable lack of information among the authorities concerning what became technically possible during the last years. According to the subject, the European environmental legislation seems to bet the right address to implement rules for seed supply intended for use in nature conservation. If there is no political majority for being incorporated into a European directive, there is at least practical use to launch just a recommendation for a regional wild seeds market at the European level.
However, the drawing up of binding European rules for the origin, quality, exploitation and establishment of semi-natural grassland as an essential part of the High Nature Value Farmland concept is urgently needed.

References

Andersen E., Baldock D., Bennett H., Beaufoy G., Bignal E., Brouwer F., Elbersen B., Eiden G., Godeschalk F., Jones G., McCracken D.I., Nieuwunhuizen W., van Eupen M., Hennekens S. and Zervas G. (2004) Developing a

high nature value farming area indicator. Internal report for the EEA. June. EEA, Copenhagen. Siehe auch http://eea.eionet.europa.eu/Public/irc/envirowindows/hnv/library
Aradottir A. and Johannsson M. (2006) Ecological restoration with native species in Iceland. Conference proceedings Soil Bioengineering: Restoration with native seed and plant material. HBLFA Raumberg-Gumpenstein, 175-179.
Bakker J.P. and Berendse F. (1999) Constraints in the restoration of ecological diversity in grassland and heathland communities. *Trends in Ecology and Evolution* 14, 63-68.
Blaschka A., Krautzer B. and Graiss W. (2008) Standortgerechte Begrünung im Landschaftsbau als Möglichkeit zur Lebensraumvernetzung - I. Was ist „standortgerecht"? Böschungen als Standort. Sauteria, Schriftenreihe für systematische Botanik, Floristik und Geobotanik, Universität Salzburg, Band 16, 50-55.
BMLFUW (2009) Rixchtlinie für die sachgerechte Bodenrekultivierung land- und forstwirtschaftlich genutzter Flächen. Bundesministerium für Land- und Forstwirtschaft, Umwelt und Wasserwirtschaft, Wien, 55 S.
BNatSchG (2011) Gesetz über Naturschutz und Landschaftspflege. www.bundesrecht.juris.de/bnatschg_2009.
CPS (2011) Swiss Commission for Wild Plant Conservation: www.cps-skew.ch (Accessed June 2011).
Donath T.-W., Bissels S., Hölzel N. and Otte A. (2007) Large scale application of diaspore transfer with plant material in restoration practice - Impact of seed and microsite limitation. *Biological Conservation* 138, 224-234.
ECONADA (2011) Ecologically sustainable implementation of the 'Nature Diversity Act' (Naturmangfoldloven) for restoration of disturbed landscapes in Norway. http://www.bioforsk.no/ikbViewer/page/tjenester/prosjekt?p_document_id=86296 (Accessed June 2011).
EEA - European Environment Agency (2005) Agriculture and the environment in EU-15 - the IRENA Indicator Report. EEA-Report No. 5. http://reports.eea.eu.int/eea_report_2005_6.
EEA - European Environment Agency (2006) The IRENA operations: http://www.eea.europa.eu/projects/irena. (Accessed June 2011).
EEA (2009) Biogeographical regions in Europe: www.eea.europa.eu/data-and-maps/figures/biogeographical-regions-in-europe (Accessed June 2011).
EEA - European Environment Agency (2010): Biodiversity Baseline Report. http://www.eea.europa.eu/publications/eu-2010-biodiversity-baseline (Accessed June 2011).
Edwards A.R., Mortimer S.R., Lawson C.S., Westbury D.B., Harris S.J., Woodcock B.A. and Brown V.K. (2007) Hay strewing, brush harvesting of seed and soil disturbance as tools for the enhancement of botanical diversity in grasslands. *Biological Conservation* 134, 372-382.
EEC (1966) Fodder plant directive: http://ec.europa.eu/food/plant/propagation/evaluation/ (Accessed June 2011).
EEC (2010) Preservation mixtures directive www.fera.defra.gov.uk/plants/consultations/documents/preservation-Directive.pdf
EC - European Commission (2006b) Rural Development 2007-2013. Handbook on Common Monitoring and Evaluation Framework, Guidance Document (Entwicklung des Ländlichen Raums 2007-2013. Handbuch für den gemeinsamen Begleitungs- und Bewertungsrahmen, Leitfaden). September 2006. Brussels. Generaldirektion Landwirtschaft.
EC - European Commission (2009) Guidance Document: The Application of the High Nature Value Impact Indicator. Programming Period 2007-2013. Report prepared for DG Agriculture. http://ec.europa.eu/agriculture/rurdev/eval/hnv/guidance_en.pdf
Haslgrübler P. (2011) Erste Ergebnisse zu Erträgen und Saatgutqualität von Spenderwiesen (mündliche Mitteilung)
Hefter I., Jünger G., Baasch A. and Tischew S. (2010) Gebietseigenes Wildpflanzensaatgut in Begrünungs- und Renaturierungsvorhaben fördern - Aufbau eines Spenderflächenkatasters und Informationssystems. *Naturschutz und Landschaftsplanung* 42 (11), 333-340.
Hölzel N. and Otte A. (2003) Restoration of a species-rich flood meadow by topsoil removal and diaspore transfer with plant material. *Applied Vegetation Science* 6, 131-140.
IEEP - Institute for European Environmental Policy (2007) Final Report for the study on HNV Indicators for Evaluation. Contract Notice 2006-G4-04. Report prepared for DG Agriculture. http://ec.europa.eu/agriculture/analysis/external/evaluation/report.pdf
Jongepierova I., Mitchley J. and Tzanopoulos J. (2007) A field experiment to recreate species rich hay meadows using regional seed mixtures. *Biological Conservation* 139, 297-305.
JRC/EEA - Joint Research Centre/European Environment Agency (2006) Background document on the methodology for mapping High Nature Value Farmland in EU27. M.L. Paracchini, J.M. Terres, J.E. Petersen, Y. Hoogeveen. October. http://eea.eionet.europa.eu/Public/irc/envirowindows/hnv/library
Kiehl K., Kirmer A., Donath T.W., Rasran L. and Hölzel N. (2010) Species introduction in restoration projects - evaluation of different techniques for the establishment of semi-natural grasslands in Central and Northwestern Europe. *Basic and Applied Ecology* 11, 285-299.
Kirmer A. and Korsch H. (2009) Spenderflächenkataster zur Gewinnung von autochthonem Grünland-Saatgut für Thüringen - Methodik, Stand und Perspektiven. Thüringer Landesanstalt für Umwelt und Geologie (Hrsg.)

Kirmer A. and Tischew S. (2006) Handbuch naturnahe Begrünung von Rohböden. Wiesbaden: Teubner B.G., 195 pp.
Klimkowska A., Van Diggelen R., Bakker J.P. and Grootjans A.P. (2007) Wet meadow restoration in Western Europe: a quantitative assessment of the effectiveness of several techniques. *Biological Conservation* 140, 318-328.
Krautzer B., Peratoner G. and Bozzo F. (2004) Site-Specific Grasses and Herbs. Seed production and use for restoration of mountain environments. Plant Production and Protection Series No. 32, Food and Agriculture Organisation of the United Nations, Rome, Italy, 112 pp.
Krautzer B. and Wittmann H. (2006) Restoration of alpine ecosystems. In: Restoration Ecology, the new frontier (edited by Jelte van Andel and James Aronson). Blackwell Publishing, pp. 208-220.
Krautzer B. and Klug B. (2009) Renaturierung von subalpinen und alpinen Ökosystemen. In: Zerbe S. and Wiegleb G. (Hrsg.), Renaturierung von Ökosystemen in Mitteleuropa. Heidelberg: Spektrum Akademischer Verlag, 209-234.
Krautzer B. and Pötsch E. (2009) The use of semi-natural grassland as donor sites for the restoration of high nature value areas. In: Proceedings 15th European Grassland Federation Symposium Brno, Czech Republic, 7-9 September 2009 (Cagas B., Radek M., Nedelnik J., editors). *Grassland Science in Europe* 14, 478-492.
Malaval S. (2006) Revegetation with native species in the French Pyrenees mountains. In Krautzer B. and Hacker E. (editors): Soil bioengineering-Ecological Restoration with Native Plant and Seed Material, Conference proceedings, 197-200.
ÖAG (2000) Richtlinien für standortgerechte Begrünungen. Österreichische Arbeitgemeinschaft für Grünland, LFZ Raumberg Gumpenstein, 54 S.
Pötsch E.M. and Blaschka A. (2003) Abschlussbericht über die Auswertung von MAB-Daten zur Evaluierung des ÖPUL hinsichtlich Kapitel VI.2.A ‘Artenvielfalt’. Gumpenstein, 37 pp.
Pywell R.F., Bullock J.M., Tallowin J.R.B., Walker K.J., Warman E.A. and Masters G. (2007) Enhancing diversity of species-poor grasslands: an experimental assessment of multiple constraints. *Journal of Applied Ecology* 44, 81-94.
REWISA (2011) Regionale Wildpflanzen und Samen: www.rewisa.at (Accessed June 2011).
Rometsch S. (2009) Recommendations for the production and use of wild flower seeds adapted to local ecological conditions in Switzerland. Proceedings of the International Workshop of the SALVERE Project, pp 19-20, http://www.salvereproject.eu/sites/default/files/SALVERE_Proceedings_May_2009.pdf (accessed July 2011).
SALVERE (2011) Semi-natural Grassland as a Source of Biodiversity Improvement: www.salvereproject.eu. Last visit June 2011
Scotton M. (2009) Semi-natural grassland as a source of biodiversity improvement – SALVERE. Proceedings of the international Workshop of the SALVERE-Project 2009, Agricultural Research and Education Centre Raumberg-Gumpenstein, pp. 2-5 http://www.salvereproject.eu/sites/default/files/SALVERE_Proceedings_May_2009.pdf (accessed July 2011).
Scotton M., Piccinin L., Dainese M. and Sancin F. (2009) Seed harvesting for ecological restoration: Efficiency of haymaking and seed-stripping on different grassland types in the eastern Italian Alps. *Ecological Restoration* 27, 66-75.
VWW (2011) Verband der deutschen Wildpflanzen- und Wildsamenproduzenten: www.natur-im-vww.de (Accessed June 2011).
Walker K.J., Stevens P.A., Stevens D.P., Mountford J.O., Manchester S.J. and Pywell R.F. (2004) The restoration and re-creation of species-rich lowland grassland on land formerly managed for intensive agriculture in the UK. *Biological Conservation* 119, 1-18.

Kapitel 2

“Quality and germination capacity of seed material harvested from an Arrhenatherion meadow”

Zitat:

Haslgrübler, P., Krautzer, B., Blaschka, A., Graiss, W. and Pötsch, E.M. (2013): Quality and germination capacity of seed material harvested from an Arrhenatherion meadow. Grass and Forage Science 69 (3), 454-461.

Grass and Forage Science
The Journal of the British Grassland Society | The Official Journal of the European Grassland Federation

Quality and germination capacity of seed material harvested from an Arrhenatherion meadow

P. Haslgrübler, B. Krautzer, A. Blaschka, W. Graiss and E. M. Pötsch

Agricultural Research and Education Centre Raumberg-Gumpenstein, Institute for Plant Production and Cultural Landscape, Irdning, Austria

Abstract

Arrhenatherion meadows are an endangered type of semi-natural grassland in Europe, and their conservation and restoration is an important policy objective. Recent research has led to development of techniques for harvesting seed material from regional donor sites and strategies for re-establishment of species-rich grassland, but their practical application requires that consumers are guaranteed sufficient seed quality of directly harvested material. Methods for the evaluation of purity, thousand seed weight (TSW) and germination capacity were developed and tested, a pre-condition to define the optimal seeding rate for this vegetation type (2000–3000 seeds m^{-2}). The assessment was based on seed obtained by two harvesting techniques: on-site threshing (OST) and seed stripping (SS). Materials from both methods obtained 63% pure seeds. The TSW differed significantly between harvesting methods: the OST provided greater seed weight (1·057 g) than the SS (0·84 g). Two trials were implemented to define criteria for the germination capacity test. Organic growing media obtained the best and most homogenous results in a first comparative germination test. In the second trial, different dormancy-breaking treatments were compared, each with and without pre-chilling, namely: addition of potassium nitrate (KNO_3), addition of gibberellic acid (GA3) and without addition of chemicals (WA). The germination capacity of the treated variants KNO_3, GA3 and pre-chilling was lower than that for WA. The harvesting method significantly influenced quality and quantity of the seed material, whereas the substrate and dormancy-breaking treatments had no effect.

Correspondence to: P. Haslgrübler, Agricultural Research and Education Centre Raumberg-Gumpenstein, Institute for Plant Production and Cultural Landscape, Raumberg 38, 8952 Irdning, Austria.
E-mail: petrahaslgruebler@gmx.at

Received 30 August 2012; revised 23 January 2013

Keywords: semi-natural grassland, donor sites, seed quality, seed stripping, on-site threshing, pre-chilling

Introduction

Recent changes in land use, characterized by agricultural intensification or land abandonment, have caused a dramatic decline in the species richness of semi-natural grasslands in Europe and have led to a strong decrease in biodiversity (Poschlod and Wallis-DeVries, 2002; Duprè *et al.*, 2010; Farruggia *et al.*, 2011). Until the nineteenth century, farmland in Europe included vast areas of semi-natural grassland, but in more recent times, most of these areas have become increasingly fragmented as meadows within the landscape (Hedberg and Kotowski, 2010). In addition to its role in providing forage for livestock production, semi-natural grassland is the only source of appropriate seed materials for ecological restoration of valuable native grassland types, either as source material for propagation of seed of native provenance or as directly harvested seed material (Poschlod and Wallis-DeVries, 2002; Kiehl *et al.*, 2010; Scotton *et al.*, 2012).

The conservation and restoration of semi-natural grasslands and their biodiversity value has been an important priority of European agri-environmental policy since the 1990s (Hopkins *et al.*, 1999). The availability of site-specific native seed material and also the knowledge on how to use it are among the greatest challenges for restoration of the rare grassland habitats that have been lost in Central Europe. Within the project SALVERE (semi-natural grassland as a source of biodiversity improvement), one of the aims was to tackle this challenge by finding and testing methods and procedures to use species-rich meadows as donor sites and to process the harvested material (Haslgrübler *et al.*, 2011).

There are many recent examples of successful ecological restoration of grasslands throughout Europe (Hopkins *et al.*, 1999; Kirmer and Tischew, 2006; Krautzer and Hacker, 2006; Jongepierová *et al.*, 2007;

doi: 10.1111/gfs.12063

Krautzer and Klug, 2009; Hedberg and Kotowski, 2010; Kirmer *et al.*, 2012; Scotton *et al.*, 2012). Harvesting of seed material that originates from within the same biogeographical region as the restoration projects where it is used is a means of ensuring that any restoration and ecosystem services will not be provided by inappropriate use of cultivars or propagation material of non-local provenance (Keller and Kollmann, 1999; Donath and Eckstein, 2008; Kiehl *et al.*, 2010). The opportunities for seed harvesting also represent a potential additional economic benefit and can be viewed as a special ecosystem service of species-rich meadows.

There are no existing regulations for this kind of harvested material for use in ecological restoration, although published literature on the topic dates from more than 30 years ago (Grime *et al.*, 1981; Williams, 1983). One important challenge with seed material directly harvested from a donor site is the changing diversity and heterogeneity in the seed composition, varying between harvests, even within the same site of origin. Seed mass and seed size also influence the germination capacity and the amount of pure seed in the harvesting material. Under a range of different environmental conditions, the germination and establishment success increase with increasing seed mass (Kahmen and Poschlod, 2008). In this article, we address this topic and give information about methods for testing the quantity and quality criteria for on-site threshing (OST) and seed-stripping (SS) material harvested from an Arrhenatherion meadow.

The core questions we consider are as follows: (i) Which factors influence germination capacity? (ii) Are there any differences between the harvesting methods applied? (iii) Is it possible to give common recommendations concerning quality and quantity criteria of harvested seed materials?

Material and methods

Study site

The experimental site was at Welser Heide, approximately 40 km west of Linz, Austria (48°18N, 14°03E; altitude 310 m a.s.l.). The site can be characterized as an Arrhenatherion meadow (Koch, 1926) as described by Mucina *et al.* (1993). The mean annual air temperature in 2009 (year of harvest) was 9·6°C with monthly values ranging from −3·2°C in January to 19·5°C in August. The annual precipitation in 2009 was 1017 mm. Since 1998, the total area of about 200 ha had not received fertilizers, and it was mown only once a year, at the end of June, followed by removal of the biomass. Under this management regime, the site changed from a nutrient-rich and species-poor meadow to a nutrient-poor and species-rich Arrhenatherion meadow (Bogner, 1992). The site at Welser Heide had never been resown with commercial seed material, and therefore all the species present are local ecotypes. The most dominant species were *Arrhenatherum elatius, Avenula pubescens, Festuca rubra, Festuca rupicula, Poa pratensis, Dianthus carthusianorum, Galium album, Salvia pratensis, Securigera varia* and *Thymus praecox* (nomenclature follows Adler *et al.*, 2008).

Seed harvesting methods

The tested material was harvested on 1 July 2009 at a time of optimum seed maturity according to Meier (2001). Two different harvesting techniques were applied: (i) a plot combine thresher (OST) with a cutting width of 150 cm and (ii) a pull-type seed stripper (SS) with a width of 1·2 m drawn at a speed of 3 km h^{-1}. The seed stripper is a machine with a rotating drum or brush that combs the seed off the plants and throws it into a hopper (Morgan and Collicutt, 1994). Due to the advanced phenological stage of grasses, the brush axle was fixed at 15 cm height above the ground, rotating upwards to collect also seeds of low-growing forbs. Mature seeds, and also stalks and leaves from grasses and forbs, are brushed from the plants into a container (Scotton *et al.*, 2009). The harvested material was air-dried, and a sieve with a mesh size of 6 mm was used to clean the seed from the stalks and leaves.

Germination experiments

Existing literature relating to successful germination treatments was studied, but existing reports were found to be available mainly for single species (Williams, 1983; Flüeler, 1992; Bell *et al.*, 1995; Godefroid *et al.*, 2010; ISTA, 2011). A combination of different treatments was therefore used to test the germination capacity of the harvested material (Molder, 1995; Heilinger and Florineth, 2003).

Germination trials were implemented one year after harvest, and the harvested material was dried, cleaned and then stored at room temperature (15–20°C) according to the common project protocol. The first trial was conducted to determine the most suitable substrate for the germination test with samples of material obtained using both harvesting methods (OST and SS). After these preliminary tests, the second trial was implemented to test the influence of different dormancy-breaking treatments.

Before the germination trial started, a homogeneous sample was taken and the purity assessments and the thousand seed weight (TSW) were determined based on the guidelines issued by the International Seed Testing

Association (ISTA, 2011). For determination of the TSW, eight batches of 100 randomly chosen full seeds were counted and weighed. For the purity assessments, the percentage composition by weight of seeds and chaff of twenty-four subsamples from both harvesting methods was determined, but the various species of seeds and inert particles were not classified. The species obtained by both harvesting methods (OST and SS) were identified through seed separation, and the seeds of all species were weighed. The duration of the germination trials was 4 weeks, and all experiments were carried out with four replicates per variant. In the first germination trial, three different types of substrate with material from both harvesting methods (OST and SS) were tested: (i) quartz sand, (ii) organic growing media (OGM) and (iii) filter paper (Don, 2009; ISTA, 2011). The quartz sand (0·5–2·0 mm grain size) was washed, hydro-classified, sieved and made free of carbon and organic waste, and had a pH value of 7·6. The organic growing medium was a mixture of organic components and mineral particles; they were nutrient rich for the first weeks and free of small and large particles. The substrate was also free of seeds, fungi, bacteria or toxic substances and had a pH value of 5·6. The filter paper was classified according to ISTA (2011), which prescribe that particular substrate must contain sufficient moisture at all times to meet the requirements for germination (Don, 2009). Four batches of 100 randomly selected seeds were sown in white pots (15·5 cm × 11 cm), after adding the respective substrates. The seeds were placed on the substrate, covered with a fine layer of the appropriate substrate in the variants quartz sand and OGM and watered with distilled water.

In the second germination trial, different dormancy-breaking treatments were as follows: (i) potassium nitrate (KNO_3 0·2%); (ii) gibberellic acid (GA3 0·05%); and (iii) without additives (WA) each in combination with and without pre-chilling. At the beginning of the experiment, 80 mL of each treatment solution was used (Bell *et al.*, 1995; Don, 2009; ISTA, 2011). Afterwards, only distilled water was added twice a week, in amounts that totalled 400 mL per pot during 4 weeks. The seeds were placed on the substrate and covered with a thin layer of the same material. The pre-chilled variants were additionally covered with a transparent lid and stored for 1 week in a cooling chamber under controlled temperature of 2–5°C. Both germination trials were conducted in a climate chamber (KBWF 720, Binder) under controlled conditions (Table 1). The germinated seeds were counted once a week, separated into monocotyledons and dicotyledons and then removed. The germination capacity indicates the proportion of seeds that produced seedlings (ISTA, 2011).

Statistical analysis

The statistical analyses were carried out using R 2.15.1 (R Core Team, 2012). To test for normality of the data (results for purity, TSW and germination capacity), the Shapiro–Wilk normality test was used. As a consequence, for the analysis of the germination trials ‘substrate’ and ‘dormancy-breaking treatment’, the data were log-transformed to provide normally distributed data for further analysis. In all other cases, the data were normally distributed. The purity and the TSW were tested with a one-sample *t*-test to determine the 95% confidence interval. A Welch two-sample *t*-test was used to test the significance of difference of the TSW between the harvesting methods. A two-way ANOVA was used to define the substrate best suited for germination, to identify differences between the harvesting methods and to determine the influence of pre-chilling and the addition of chemicals on the germination capacity. Finally, the Bonferroni post hoc test was used to prove the significance of differences between variants where necessary.

Results

Purity assessments, thousand seed weight and seed separation

The OST material showed an average purity of 63·05% with an average TSW of 1·0573 g. In the SS material, 63·11% of pure seeds with a mean TSW of 0·84 g were assessed. The calculated confidence interval of the TSW and the purity for materials obtained by both harvesting methods were highly significant ($P < 0{\cdot}001$). The difference in the TSW between the two harvesting methods was also significant ($P < 0{\cdot}001$), but there were no significant differences concerning the proportion of harvested pure seeds. The results of the seed separation showed that thirty three species were harvested with the on-site threshing method, of which there were nine grasses that comprised 80% by weight and four legumes and twenty forb species that together comprised the other 20% by weight. Twenty-eight different species were harvested with the seed-stripping technique; nine different grasses comprised 79% by weight, and five legumes and fourteen forbs represented the remaining 21% by weight.

Germination capacity in the trial ‘substrate’

The primary aim of the germination capacity trial ‘substrate’ was to determine a suitable substrate for the subsequent experiments (Table 2). The two-way

Table 1 Germination cabinet conditions for the assessment of the germination capacity.

First germination trial: 'substrate'	Second germination trial: 'dormancy-breaking treatment'
Daylight (14·200 lux)/Night: 8 h/16 h	Daylight (14·200 lux)/Night: 12 h/12 h
Humidity: 85% ± ≤ 2·5	Humidity: 85% ± ≤ 2·5
Temperature: 20°C/30°C ± 0·1–0·5°C	Temperature: 20°C/30°C ± 0·1–0·5°C
Duration: 29 March 2010–26 April 2010	Duration: 19 August 2010–16 September 2010
	Pre-chilling: 12 August 2010–19 August 2010 (3–4°C)

Table 2 Results of the thousand seed weight (TSW) and the purity of the material harvested by on-site threshing (OST) and seed stripping (SS).

Harvesting material	Mean-value	95% Confidence interval	*P*-value
OST – TSW	1·0573 g	0·9955–1·1191	<0·001***
SS – TSW	0·84 g	0·8108–0·8702	<0·001***
OST – pure seeds	63·05%	59·24–66·85	<0·001***
SS – pure seeds	63·11%	58·93–67·29	<0·001***

Significance level *, $P < 0·05$; **, $P < 0·01$; ***, $P < 0·001$.

anova (Table 3) showed significant differences between the harvesting methods, whereas the substrate did not influence the germination capacity significantly. There was a significant interaction between the harvesting method and the substrate ($P = 0·028$, Eta Sq = 0·33) caused by a higher germination capacity of the OST material on organic growing media.

The viability of seeds that had not germinated was not tested further, because after the end of the germination trial, the seeds could not be isolated from the quartz sand substrate and the OGM, and in the variant 'filter paper', all of the non-germinated seeds were additionally infected with fungi. The crucial difference between the substrates was the water-holding capacity. Over the total observation period, the variant 'filter paper' (120 mL) needed less water compared with the OGM (400 mL) and quartz sand (500 mL). The outcomes showed that the OGM had a higher water-absorbing capacity than quartz sand. In the variant 'filter paper', an infection with fungi was observed; this required the paper to be changed after 2 weeks, which is impractical. As a consequence, the 'filter paper' variant was not used in the subsequent experiments. The OST material revealed the highest and most homogeneous germination rate on the substrate OGM, shown also by a low standard deviation between the replicates (Table 4). Therefore, the substrate OGM and the harvesting material OST were selected for the following trial 'dormancy-breaking treatment'.

Germination capacity in the trial 'dormancy-breaking treatment'

The two-way anova showed a significant difference between the treatments WA, KNO_3 and GA3 and the pre-chill variants (Table 5). The interaction between pre-chill and treatment was not significant ($P = 0·678$, Eta Sq = 0·41). The mean total values showed that the different variants reached a germination capacity of between 41% and 56% (Table 6). The highest germination rates were obtained for the variants without any dormancy-breaking treatment (WA), which indicates that pre-chilling had a decreasing effect on the germination capacity.

The results of the specific separation of the germinated seeds (Figure 1) demonstrate a well-balanced proportion of monocotyledonous and dicotyledonous seedlings. Despite a lower proportion of forbs and legumes (20%, compared with 80% grasses) in the harvested material, they were much more competitive concerning the total germination capacity.

Discussion

For the successful ecological restoration of semi-natural grassland, both seed quantity and seed quality are of great importance. Therefore, this article has

Table 3 Results of the two-way anova of the germination trial 'substrate'.

Variant	Df	Sum Sq	Mean Sq	*F*-value	*P*-value	Partial Eta Sq
Harvesting method	1	0·096	0·096	13·275	0·002**	0·42
Substrate	2	0·015	0·008	1·043	0·373	0·10
Harvesting method × Substrate	2	0·064	0·032	4·395	0·028*	0·33
Residuals	18	0·130	0·007			

Significance level: *, $P < 0·05$; **, $P < 0·01$; ***, $P < 0·001$. R^2: 0·57; adjusted R^2: 0·45.

Table 4 Average germination capacity (mean value) in percentage of on-site threshing and seed-stripping material from an Arrhenatherion meadow dependent on different substrates.

Harvesting method	Quartz sand	Organic growing media	Filter paper
Mean OST	59·5	68·3	58·0
s.d.	±10·08	±3·59	±2·58
Mean SS	73·0	67·0	69·8
s.d.	±4·69	±2·71	±4·03

addressed the specific parameters of seed purity, TSW, relative proportions by weight of monocotyledonous and dicotyledonous seeds in the mixture and the spontaneous germination capacity of harvested seed material, specifically in the context of an Arrhenatherion meadow. The focus of this study was to test and characterize the germinability of harvested seed material from a donor site, to provide practical and useful information for practitioners concerned with planning of ecological restoration projects. Parameters studied were different harvesting techniques (on-site threshing and seed stripping) and factors influencing the germination capacity ('substrate' and 'dormancy-breaking treatment').

The main challenge is presented by the diversity and variability of the tested material. Many factors influence seed production of plants and the growth of semi-natural grassland during the year (Scotton *et al.*, 2012). The composition and quality of seed material harvested by stripping or by on-site threshing differ greatly from year to year (Morgan and Collicutt, 1994). Thus, harvesting time and harvested volume of the seed material are dependent on the weather conditions during the respective vegetation period (Scotton *et al.*, 2012). Our results showed considerable differences in the number of harvested species obtained between the two methods (on-site threshing and seed stripping) assessed during seed separation, but both mixtures showed the same proportion of grass seeds, which was 80% by weight. This is typical for Arrhenatherion meadows, as early harvesting in June means a higher proportion of grasses, whereas harvesting carried out in July or August increases the proportion of forbs (Hölzel and Otte, 2003; Scotton *et al.*, 2012). In general, it is desirable to mix an early and a late harvest to provide the greatest possible spectrum of species. A harvesting date set either too early or too late can lead to the disappearance of several species (Kirmer and Tischew, 2006; Krautzer *et al.*, 2011; Scotton *et al.*, 2012). Species number and the composition of the harvested material are strongly dependent on the type of meadow (Kiehl *et al.*, 2010).

The results presented in this article confirm that on-site threshing and seed stripping of potential donor sites provide effective methods for harvesting seed material with a satisfactory germination capacity for successful use in the restoration of semi-natural grasslands (Scotton *et al.*, 2012). Determination of the purity of the harvested seed material is one important part of the information needed to define the seeding rate of the entire material. After the cleaning process with a sieve of a mesh size of 6 mm, both harvesting techniques showed comparable proportions of pure seeds. The results of the TSW differed significantly.

In the germination trials, most of the seeds germinated within the first 2 weeks, allowing the conclusion that an observation period of 4 weeks is sufficient for a proper testing of such heterogeneous material. The germination trial 'substrate' did not show any significant difference between the different substrates tested, but between the two harvesting methods highly significant results were found, and this can be explained by the harvest of only late mature seeds with the seed-stripping technique (Scotton *et al.*, 2009). Another explanation could be the varying seed size and mass (Kahmen and Poschlod, 2008) obtained by different harvesting methods. Flüeler (1992) reports that the germination capacity of heavy seeds is higher than that of seeds of lighter weight. Milberg *et al.* (2000) found that the response to light has an influence on the germination capacity in relation to seed size: light requirement for germination is more likely

Table 5 Results of the two-way ANOVA of the germination trial 'dormancy-breaking treatment'. Treatment clusters WA, KNO_3 and GA3.

Variant	Df	Sum Sq	Mean Sq	*F*-value	*P*-value	Partial Eta Sq
Treatment	2	0·147	0·074	4·688	0·023*	0·34
Pre-chill	1	0·082	0·082	5·199	0·035*	0·22
Treatment × Pre-chill	2	0·012	0·006	0·383	0·687	0·41
Residuals	18	0·283	0·016			

Significance level: *, $P < 0·05$; **, $P < 0·01$; ***, $P < 0·001$. R^2:0·46; adjusted R^2: 0·31.

Table 6 Germination capacity (mean value) as percentage, of on-site threshing material from an Arrhenatherion meadow with different dormancy-breaking treatments (KNO_3: potassium nitrate; GA3: gibberellic acid) and without chemical treatment (WA), with/without pre-chilling in each case.

Treatment	Without pre-chilling	With pre-chilling	Means
Mean WA	56·25	50·0	53·1[a]
SD	±3·10	±3·92	
Mean KNO_3	55·0	51·0	53·0[ab]
s.d.	±11·69	±5·89	
Mean GA3	48·75	41·0	44·9[b]
s.d.	±5·25	±2·55	
Means	53·3[a]	47·3[b]	

a, b: Different letters indicate significant differences.

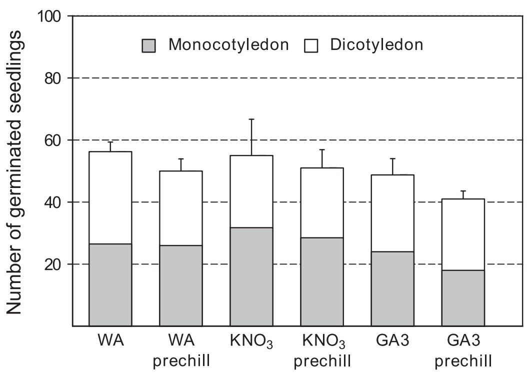

Figure 1 Proportion of germinated seeds (monocotyledons and dicotyledons) of all variants in the dormancy-breaking experiment. WA: without dormancy-breaking treatments; KNO_3: potassium nitrate addition; GA3: gibberellic acid addition; all with and without pre-chilling.

in small- than in large-seeded species and that its likely ecological role is a response to depth of burial.

The results of TSW assessment show that smaller seeds are harvested with the seed stripper, probably explaining the higher germination capacity assessed. A recommendation can be given for using the substrate OGM, as this provided the most homogeneous results, and it has a high water-holding capacity and is easily available. In the germination trial 'substrate', all replicates reached a germination capacity of over 50%, which can be considered high when compared with the results of Heilinger and Florineth (2003), where the same type of meadow was tested for its germination capacity.

The germination trial 'dormancy-breaking treatment' was carried out to clarify whether harvested seeds need to be stimulated with either potassium nitrate or gibberellic acid or by pre-chilling to obtain a higher germination capacity. Seeds from different populations seem to respond differently to the stratification treatments (Grime *et al.*, 1981; Milberg and Andersson, 1998). Our results showed that pre-chilling had a decreasing effect on the germination capacity. Shepley *et al.* (1972) reported that a 0·1% solution of GA3 led to full germination within 2 d. In contrast, Vandvik and Vange (2003) reported that addition of gibberellic acid to stratified seeds did not increase germination. Our variants treated with GA3 showed in total that there was a decrease in germination capacity below 50%. The proportion of dicotyledonous seedlings was higher regarding the dormancy-breaking effect on forbs and legumes (Williams, 1983; Flüeler, 1992), but stimulation effects of GA3 are not universal (Bell *et al.*, 1995). Potassium nitrate has been used in dormancy-breaking treatments of monocotyledons (Williams, 1983; ISTA, 2011), thus resulting in higher number of monocotyledon seedlings in KNO_3 treatments. The average germination capacity in the variant KNO_3 was not significantly different to the variant without additives, although this effect may have been different with a lower or higher nitrate concentration. Williams (1983) found that seeds were strongly influenced by temperature regime, potassium nitrate and gibberellic acid. In fact, the heterogeneity of the harvested material can also be an advantage for restoration projects, as the probability for a successful restoration increases with a greater flexibility and adaptability of different ecotypes to the various site conditions on receptor sites.

Conclusion

The results obtained in this study enable us to provide recommendations concerning quantity and quality criteria of harvested seed material from semi-natural grasslands, specifically from Arrhenatherion meadows, for practical users. A practical method is demonstrated to test the germination capacity with the use of organic growing media and without the use of dormancy-breaking treatments (such as pre-chilling, GA3 and KNO_3). Based on the presented methods for the assessment of TSW, purity and germination capacity, it is possible to define the required seeding rate of 2000–3000 seeds m^{-2} (Kirmer *et al.*, 2012) of the harvested material of an Arrhenatherion meadow. This is an essential pre-condition to guarantee successful ecological restoration of receptor sites.

Acknowledgments

This study was supported by the Austrian Ministry for Agriculture, Forestry, Environment and Water

Management and co-financed within the framework of the CENTRAL EUROPE Programme by the European Regional Development Funds (ERDF). We would like to thank all persons who contributed to this research work.

References

Adler W., Oswald K. and Fischer M.A. (2008) *Exkursionsflora für Österreich, Liechtenstein und Südtirol. (Botanical Fieldguide for Austria, Lichtenstein and South Tyrol)*. Austria: Ulmer Verlag.

Bell D.T., Rokich D.P., McChesney C.J. and Plummer J.A. (1995) Effects of temperature, light and gibberellic acid on the germination of seeds of 43 species native to Western Australia. *Journal of Vegetation Science*, **6**, 797–806.

Bogner D. (1992) Entwicklung der Landwirtschaft auf der Welser Heide und ihr Einfluß auf die Landwirtschaft seit der Frühzeit. (Agricultural development on the Welser Heide and the influence of the agriculture since prehistoric times). *Kataloge des oberösterreichischen Landesmuseums*, **54**, 401–408.

Don R. (2009) *Ista handbook on seedling evaluation third edition with amendments 2009*. Bassersdorf, Switzerland: The International Seed testing Association (ISTA).

Donath W.T. and Eckstein R.L. (2008) Bedutung genetischer Faktoren für die Wiederansiedlung seltener Pflanzengemeinschaften – genetische Diversität in der Praxis des Naturschutzes. (Importance of genetic factors of the reestablishment of rare plant communities – genetic diversity for applied nature conservation). *Naturschutz und Landschaftsplanung*, **40**, 21–25.

Duprè C., Stevens C.J., Ranke T., Bleeker A., Peppler-Lisbach C., Gowing D.J.G., Dise N.B., Dorland E.D.U., Bobbink R. and Diekmann M. (2010) Changes in species richness and composition in European acidic grasslands over the past 70 years: the contribution of cumulative atmospheric nitrogen deposition. *Global Change Biology*, **16**, 344–357.

Farruggia A., Dumont B., Scohier A., Leroy T., Pradel P. and Garel J.-P. (2011) An alternative stocking management designed to favour butterflies in permanent grasslands. *Grass and Forage Science*, **67**, 136–149.

Flüeler R.P. (1992) *Experimentelle Untersuchungen über Keimung und Etablierung von alpinen Leguminosen. (Experimental analysis of germinaton and establishemnt of alpine legumes)*. Zürich, Switzerland: Geobotanisches Institut der ETH Zürich.

Godefroid S., van de Vyver A. and Vanderborght T. (2010) Germination capacity and viability of threatened species collections in seed banks. *Biodiversity and Conservation*, **19**, 1365–1383.

Grime J.P., Mason G., Curtis A.V., Rodman J. and Band S.R. (1981) A comparative study of germination characteristics in a local flora. *Journal of Ecology*, **69**, 1017–1059.

Haslgrübler P., Krautzer B., Tamegger C., Ševčíková M., Tischew S., Rieger E., Kizekova M., Golinski P. and Scotton M. (2011) SALVERE – Semi natural grassland as a source of biodiversity improvement – a Central Europe Project. *Grassland Science in Europe*, **16**, 526–528.

Hedberg P. and Kotowski W. (2010) New nature by sowing? The current state of species introduction in grassland restoration, and the road ahead. *Journal for Nature Conservation*, **18**, 304–308.

Heilinger K. and Florineth F. (2003) Qualitatsprüfung von nicht gezüchtetem Saatgut. (Quality aspects of uncultivated seed mixtures). *Rasen Turf Gazon*, **3**, 56–66.

Hölzel N. and Otte A. (2003) Restoration of a species-rich flood meadow by topsoil removal and diaspore transfer with plant material. *Applied Vegetation Science*, **6**, 131–140.

Hopkins A., Pywell R.F., Peel S., Johnson R.H. and Bowling P.J. (1999) Enhancement of botanical diversity of permanent grassland and impact on hay production in environmentally sensitive areas in the UK. *Grass and Forage Science*, **54**, 163–173.

ISTA (2011) *International rules for seed testing edition 2011*. Bassersdorf, Switzerland: The International Seed Testing Association (ISTA).

Jongepierová I., Mitchley J. and Tzanopoulos J. (2007) A field experiment to recreate species rich hay meadows using regional seed mixtures. *Biological Conservation*, **139**, 297–305.

Kahmen S. and Poschlod P. (2008) Does germination success differ with respect to seed mass and germination season? Experimental testing of plant functional trait responses to grassland management. *Annals of Botany*, **101**, 541–548.

Keller M. and Kollmann J. (1999) Effects of seed provenance on germination of herbs for agricultural compensation sites. *Agriculture, Ecosystems and Environment*, **72**, 87–99.

Kiehl K., Kirmer A., Donath T.W., Rasran L. and Hölzel N. (2010) Species introduction in restoration projects – Evaluation of different techniques for the establishment of semi-natural grasslands in Central and Northwestern Europe. *Basic and Applied Ecology*, **11**, 285–299.

Kirmer A. and Tischew S. (2006) *Handbuch naturnahe Begrünung von Rohböden (Handbook near-natural restoration of raw soil.)* Wiesbaden, Germany: Vieweg+Teubner Verlag.

Kirmer A., Krautzer B., Scotton M. and Tischew S. (2012) *Praxishandbuch zur Samengewinnung und Renaturierung von artenreichem Grünland. (Practical handbook for seed harvest and ecological restoration of species-rich grasslands)*. Anhalt, Germany: Hochschule Anhalt; Irdning, Austria: Lehr- und Forschungszentrum Raumberg-Gumpenstein.

Koch W. (1926) Die Vegetationseinheiten der Linthebene. (Vegetation units of the Linth plain). *Jahrbuch St. Gallischen naturwissenschaftlichen Gesellschaft. St. Gallen*, **61**, 1–146.

Krautzer B. and Hacker E. (2006) *Soil bioengineering: ecological restoration with native plant and seed material*. Irdning, Austria: Research and Education Centre for Agriculture Raumberg-Gumpenstein; Achen, Germany: Federation for Soil-Bioengineering.

Krautzer B. and Klug B. (2009) Renaturierung von subalpinen und alpinen Ökosystemen. (Restoration of subalpine and alpine ecosystems). In: Zerbe S. and Wiegleb G. (eds) *Renaturierung von Ökosystemen in Mitteleuropa (Restoration of ecosystems in central Europe)*, pp. 209–234. Heidelberg, Germany: Spektrum Akademischer Verlag.

Krautzer B., Graiss W., Peratoner G., Partl C., Venerus S. and Klug B. (2011) The influence of recultivation technique and seed mixture on erosion stability after restoration in mountain environment. *Natural Hazards*, **56**, 547–557.

Meier U. (2001) *Entwicklungsstadien mono- und dikotyler Pflanzen - BBCH Monografie. (Development stages of monocotyledon and dicotyledon species)*. Berlin, Germany: Biologische Bundesanstalt für Land- und Forstwirtschaft.

Milberg P. and Andersson L. (1998) Does cold stratification level out differences in seed germinability between populations? *Plant Ecology*, **134**, 225–234.

Milberg P., Andersson L. and Thompson K. (2000) Large-seeded spices are less dependent on light for germination than small-seeded ones. *Seed Science Research*, **10**, 99–104.

Molder F. (1995) *Vergleichende Untersuchungen mit Verfahren der oberbodenlosen Begrünung unter besonderer Berücksichtigung areal- und standortbezogener Ökotypen, (Comparative study of revegetation methods without topsoil regarding site specific ecotypes)*. Gießen, Germany: Justus-Liebig-Universität Gießen.

Morgan J.P. and Collicutt D.R. (1994) Seed stripper harvesters. *Restoration and Management Notes*, **12**, 51–54.

Mucina L., Grabherr G. and Ellmauer T. (Hrsg.) (1993) *Die Pflanzengesellschaften Österreichs. Teil I: Anthropogene Vegetation. (Plant communities of Austria. Part I: Anthropogenic Vegetation)*. Jena, Germany: Fischer Verlag.

Poschlod P. and WallisDeVries M.F. (2002) The historical and socioeconomic perspective of calcareous grasslands—lessons from the distant and recent past. *Biological Conservation*, **104**, 361–376.

R Core Team (2012) *R: a language and environment for statistical computing*. Vienna, Austria: R Foundation for Statistical Computing.

Scotton M., Piccinin L., Dainese M. and Sancin F. (2009) Seed harvesting for ecological restoration: efficiency of haymaking and seed-stripping on different grassland types in the Eastern Italian alps. *Ecological Restoration*, **27**, 66–75.

Scotton M., Kirmer A. and Krautzer B. (2012) *Practical handbook for seed harvest and ecological restoration of species-rich grasslands*. Padova, Italy: Padova Cleup.

Shepley S.C., Chang J.L.L. and Chang C. (1972) Does gibberellic acid stimulate seed germination via amylase synthesis? *Plant Physiology*, **49**, 441–442.

Vandvik V. and Vange V. (2003) Germination ecology of the clonal herb *Knautia arvensis*: regeneration strategy and geographic variation. *Journal of Vegetation Science*, **14**, 591–600.

Williams E.D. (1983) Effects of temperature, light, nitrate and pre-chilling on seed germination of grassland plants. *Annals of Applied Biology*, **103**, 161–172.

Kapitel 3

“Influence of different storage condidions on quality characteristics of seed material from semi-natural grassland“

Zitat:

Haslgrübler, P., Krautzer, B., Blaschka, A., Graiss, W. and Pötsch, E.M. (2014): Influence of different storage conditions on quality characteristics of seed material from semi-natural grassland. Grass and Forage Science 70 (3), 549-556.

Grass and Forage Science
The Journal of the British Grassland Society | The Official Journal of the European Grassland Federation

Influence of different storage conditions on quality characteristics of seed material from semi-natural grassland

P. Haslgrübler, B. Krautzer, A. Blaschka, W. Graiss and E. M. Pötsch
Agricultural Research and Education Centre Raumberg-Gumpenstein, Irdning, Austria

Abstract

Neither methodology nor guidelines are available for defining quality characteristics and storage conditions of seed material harvested from semi-natural grassland. Seeds from an *Arrhenatherion* meadow were harvested via on-site threshing and seed stripping. After determination of purity, thousand seed weight and pre-tests in a phytotron, germination-capacity trials were carried out in a greenhouse. The harvested seed material was stored for up to 3 years under different conditions: (i) room temperature 18–23°C, (ii) cooling chamber (2–5°C with 40–50% humidity) and (iii) freezer (−18°C). There was a significant impact of the tested harvesting methods on seed separation, thousand seed weight and purity, but not on the germination capacity. Different storage conditions and storage length significantly influenced the germination capacity. There were also generally higher germination values for the seed-stripping material than the on-site threshing material (ca. 70 and 60%, respectively, in the first year). Germination capacity decreased significantly with time and was <15% after 3 years. We conclude that harvested seed material from semi-natural grassland should preferably be stored under cool conditions and used within 2 years.

Keywords: *Arrhenatherion* meadow, germination capacity, grassland restoration, seed stripping, seed storage, on-site threshing

Correspondence to: P. Haslgrübler, Agricultural Research and Education Centre Raumberg-Gumpenstein, Institute of Plant Production and Cultural Landscape, Raumberg 38, 8952 Irdning, Austria.
E-mail: petrahaslgruebler@gmx.at

Received 20 December 2013; revised 15 May 2014

Introduction

Throughout most of the twentieth century, there was a tremendous decline in the area of semi-natural grassland in Europe and a huge loss of biodiversity (Hoekstra *et al.*, 2005; Wellstein *et al.*, 2007; Duprè *et al.*, 2010; Wesche *et al.*, 2012 Krause and Culmsee, 2013). To counter this trend, various agri-environmental policies and habitat-protection strategies (e.g. NATURA 2000) and new conservation management practices have been introduced (Stevenson *et al.*, 1997; Lencová and Prach, 2011; Schmiede *et al.*, 2012; Kirkham *et al.*, 2013). During the past 10–15 years, there has been a considerable development in ecological restoration, and nowadays this plays an important role in contributing to the protection of biodiversity (Society for Ecological Restoration International Science & Policy Working Group, 2004; Shackelford *et al.*, 2013). There are numerous examples of successful ecological restoration measures throughout Europe (Rydgren *et al.*, 2010; Lencová and Prach, 2011; Kirmer *et al.*, 2012a; Mitchley *et al.*, 2012), and effective methods for harvesting seed and plant material have been developed (Morgan *et al.*, 1995; Pywell *et al.*, 2002; Kiehl *et al.*, 2010; Rydgren *et al.*, 2010; Scotton *et al.*, 2012). The usual procedure is the transfer of propagules from a donor community to a receptor site, with the species composition as complete as possible, and this requires on-site harvesting of material. Contractors and others involved in restoration require information about the quality of the sowing material, especially in terms of seed proportion and its germination capacity (Kirmer *et al.*, 2012b). There are, however, no prescribed methods for the determination of quality aspects such as total number of species, moisture content, purity, thousand seed weight (TSW) and germination capacity of harvested seed mixtures from donor sites, although for several cultivated species there are strict procedural requirements imposed by Certifying Authorities (ISTA, 2011).

Seed production of plants and the biomass yield of grasslands are dependent on precipitation and temperature during the year and therefore they differ from year to year (Kirmer *et al.*, 2012b). Seed mass and size influence the TSW and the germination capacity of the harvested seed material as well as the seeding density. For successful restoration of semi-natural grassland, an amount of 2000–5000 seeds m^{-2}, depending on vegetation type, is seen as the optimal seeding density (Krautzer and Klug, 2009; Kirmer *et al.*, 2012b). Seed size, lifespan and germination date may therefore be regarded as co-adapted features of plant life history (Silvertown, 1981). The main challenge presented by harvested seed material is its heterogeneity, caused by the high diversity of species, different seed sizes and various germination requirements. We developed a method, based on literature studies (Grime *et al.*, 1981; Williams, 1983; Molder, 1995; Heilinger and Florineth, 2003), to gain valid information about the seed potential of a harvested donor site within a defined period of time, and requiring as little technical and personnel expenditure as possible.

The main questions of our research were: (i) are there any differences in seed quality characteristics of the seed material according to the harvesting methods and (ii) is there a change in the germination capacity of the seed material concerning storage length, storage conditions and harvesting methods? The results will provide relevant quality characteristics of seed material from semi-natural grassland that are essential requirements for generating specific methodology and guidelines.

Materials and methods

Study site

The donor site is an *Arrhenatherion* meadow classified by Koch (1926, as described by Mucina *et al.*, 1993) situated in the north-western part of Austria near Linz (48°18′ N, 14°03′ E; 310 m a.s.l.). The soil is characterized by a high pH-value and a low concentration of phosphorus and potassium (Table 1), which are unfavourable soil conditions for intensive grassland farming. This nutrient status reflects the low-input management at this semi-natural grassland site over a long period. The extremely high magnesium content and high total carbonate concentration are of geogenic origin. The soil is rich in humus and total nitrogen, resulting from the site remaining undisturbed for long periods of time, which has led to an accumulation of organic matter in the upper soil layer (BMLFUW, 2006). Since 1998, the total area of 200 ha has remained unfertilized and mown only once a year, at the end of June, followed by removal of mown biomass. The average long-term (1971–2000) air temperature was 8·8°C with a maximum of 13·5°C and a minimum of 4·9°C, and the average annual precipitation was 754 mm. In the year of harvest (2009), the mean annual air temperature at the donor site was 9·6°C and annual precipitation was 1017 mm.

Table 1 Soil parameters of the study site at Welser Heide in Upper Austria (average of three randomly taken bulk samples, 0–10 cm soil depth).

Soil parameter	Method*/ extractant	Values	Classification†
pH-value	$CaCl_2$	7·2	Alkaline, high
Phosphorus (mg kg^{-1} soil)	Calcium acetate lactate	15·8	Very low
Potassium (mg kg^{-1} soil)	Calcium acetate lactate	73·9	Low
Magnesium (mg kg^{-1} soil)	By Schachtschabel (1954)	450·4	Very high
Total carbonate (mass%)	Scheibler method‡	13·3	High
Total nitrogen (mass%)	Dry combustion	0·5	–
Humus (mass%)	Dry combustion	10·4	Very high

*According to the Austrian Standard System (Ö-NORM).
†According to BMLFUW (2006).
‡Described in Blum *et al.*(1996).

Field harvesting methods

The seed-rich material was harvested via (i) on-site threshing (OST) and (ii) seed stripping (SS) on 1 July 2009. For each harvesting method, a total area of 270 m^2 (three replicates with 3 × 30 m with a distance of 2·8 m between the plots) was selected. The optimal time for harvesting was determined through phenological surveys of the main characteristic species *Arrhenatherum elatius, Festuca rubra, Festuca rupicola, Poa pratensis, Dianthus carthusianorum, Galium album, Salvia pratensis, Securigera varia* and *Thymus praecox* (nomenclature after Adler *et al.* (2008) according to the BBCH scale (Meier, 2001)). For on-site threshing, a plot combines thresher with a cutting width of 150 cm, and for seed stripping, a pull-type seed stripper (120 cm width) was used and both drawn at 3 km h^{-1} (Lochner, 1997). Seed stripping is a method for harvesting seed from donor sites without cutting the vegetation (Morgan *et al.*, 1995). The method uses a rotating brush, so that the mature seeds, stems and leaves are brushed from the plants into a container (Morgan and Collicutt, 1994; Edwards *et al.*, 2007;

Scotton *et al.*, 2009). As the main species at the study site were at an advanced stage of seed ripening, the brush axis of the seed stripper was fixed at a height of 15 cm above the ground surface. The weather conditions on the day of harvesting were hot and cloudy with 85% humidity and low wind speed. After harvesting, the material was air dried for 3 d at room temperature and partially cleaned with a 6-mm sieve. The air-dried material was then stored under a range of conditions before investigations of germination under greenhouse conditions.

Experimental design

The experimental design for the greenhouse test was defined following the results of pre-tests in a phytotron (Haslgrübler *et al.*, 2014). The material was stored over 3 years (2009–2012) under different storage conditions: (i) room temperature, 18–23°C (room), (ii) cooling chamber, 2–5°C with 40–50% humidity (cool) and (iii) freezer, −18°C (freeze). The different storage temperatures were based on the results of previous experiments conducted by Welty *et al.* (1987), Budelsky and Galatowitsch (1999) and Turner *et al.* (2013). Before the harvested material was stored, seed samples (of each 5 g per harvesting method and replication) were separated in the laboratory under a microscope. All species in the harvested seed material were identified, and their seeds were counted and weighed separately. The seed moisture content was measured after half a year of storage under the three different conditions listed above. For determination of the TSW, twenty samples of 100 randomly available full seeds were counted and weighed. For the purity assessments, twenty samples were divided into chaff and full seeds, but the various species of seeds and inert matter were not classified. The seed material of each storage variant was mixed thoroughly and four samples of 100 g each were selected (Stevenson *et al.*, 1997; Scotton *et al.*, 2012). Additionally, four subsamples of 1·25 g were taken of each 100 g sample and divided into monocotyledons, dicotyledons and chaff. The separated seeds were counted and weighed for both harvesting methods. Afterwards, the subsamples of 1·25 g were mixed again and surface-sown in bulb trays (40 cm × 60 cm × 6 cm) on organic growing media (OGM), which performed best in the pre-tests (Haslgrübler *et al.*, 2014). In total, twenty-four trays (two harvesting methods × three storage variants × four replicates) were placed in the greenhouse and watered at least once a day to keep the OGM on a constant level of soil moisture. The germination capacity (GC) test in the greenhouse was carried out every year during the storage period and lasted for 4 weeks each (July 2010, June 2011 and June 2012). The air temperature in the greenhouse was measured and controlled during the testing period. The average day/night temperatures were 24·0/18·6°C in 2010, 26·4/18·6°C in 2011 and 24·9/17·2°C in 2012. The germination capacity indicates the proportion of seeds which have produced seedlings (ISTA, 2011). Germinated seeds were counted, removed weekly and divided into monocotyledons and dicotyledons.

Statistical analysis

For the statistical analyses, we used R 2.15.1 (R Core Team, 2012). The normal distribution of the residuals was tested and confirmed by a Shapiro–Wilk test, and homogeneity of variances was assessed visually (Crawley, 2007). The Wilcoxon rank sum test with continuity correction (Mann–Whitney *U*-test) was used to test whether significant differences in the number of species exist. The Welch's two-sample *t*-test was applied to find differences in TSW and purity between the two harvesting methods. A three-way ANOVA model was used to test the impact of storage conditions, storage length and harvesting method on germination capacity. Finally, the Tukey HSD post hoc test was used to indicate significant differences between variants.

Results

Seed quality characteristics

The total number of species in the seed material was significantly different between the tested harvesting methods. The results of the seed separation showed an average number of twenty-four different species for OST material. These included eight grasses, which in total comprised 78·3% by weight, two legumes which comprised 1·0% by weight and thirteen non-legume forbs with 20·8% by weight. The SS material contained twenty-one different species of which nine grasses contributed 76·4% by weight, two legumes with 0·2% by weight and nine non-legume forbs contributed 23·4% by weight. There were no significant differences in the numbers of harvested grasses and legumes, whereas the number of non-legume forbs was significantly higher in the OST material (Table 2).

The moisture content of the harvested seed mixtures for both OST and SS was between 7·5 and 10·7%, depending on the storage conditions (room, cool, freeze). The TSW of the harvested OST material was 1·027 g (±0·095), while that of the SS material reached 0·841 g (±0·006), with a significant impact of the harvesting methods ($P < 0·001$). There was no sig-

Table 2 Number of harvested species (means and standard deviation) for samples obtained by on-site threshing (OST) and seed stripping (SS), divided into grasses, forbs and legumes.

	OST			SS		
	Grasses	**Forbs**	**Legumes**	**Grasses**	**Forbs**	**Legumes**
Mean number	8·3	13·3	2·0	9·3	9·0	2·3
s.d.	±0·5	±1·0	±0·8	±1·0	±2·5	±1·3
Share of seed (% by weight)	78·3	20·7	1·0	76·4	23·4	0·2
s.d.	±9·1	±10·0	±1·1	±10·8	±10·9	±1·1

nificant difference ($P = 0{\cdot}98$) in the share of pure seeds in the harvested material between OST with 63·05% (±9·92) and SS with 63·11% (±9·00).

Germination capacity of the harvested material

The germination capacity was significantly influenced by the factors storage length and storage conditions but not by the tested harvesting methods, with significant two- and three-way interactions for all tested main factors (adjusted $R^2 = 0{\cdot}91$; Table 3). Over time, the three storage conditions (SL*SC; $P < 0{\cdot}001$) as well as the two harvesting methods (SL*HM; $P = 0{\cdot}034$) developed differently. The germination capacity of the OST material significantly declined with storage length, independent of storage conditions (Figure 1). After 1 year, the germination capacity reached an average value of 54% but dropped to 44% in the second year for samples stored under room temperature, whereas samples stored under cool and freeze conditions still retained high values, ranging between 56 and 60%. After 3 years of the experiment, all three storage variants showed a strong and significant reduction of the germination capacity to below 20%. In general, OST samples stored at room temperature had the lowest germination capacity for all years.

The germination capacity of the SS material decreased significantly with the length of storage period for all storage treatments, with a significant interaction between these factors (Figure 1). In the first year, the seed stored at room temperature obtained the highest germination rate (78%), with the strongest variation, in contrast to the samples stored under cool conditions (48%). This material achieved higher germination in the second year (58%). In the third year of testing, the germination capacity strongly decreased to <15%, indicating that seeds harvested from semi-natural grassland have limited storage ability and should therefore be used within 2 years as far as possible.

Table 3 Three-way ANOVA for germination capacity of seed material harvested with on-site threshing (OST) and seeds stripping (SS), stored under different conditions (room, cool, freeze) over 3 years (2010, 2011, 2012).

	Df	Sum Sq	Mean Sq	*F*-value	*P*-value
Storage length (SL)	2	31651	15825·5	330·20	<0·001
Storage conditions (SC)	2	415	207·4	4·33	0·018
Harvesting methods (HM)	1	44	43·7	0·91	0·344
SL*SC	4	1664	416·0	8·68	<0·001
SL*HM	2	345	172·7	3·60	0·0339
SC*HM	2	815	407·7	8·51	<0·001
SL*SC*HM	4	641	160·3	3·34	0·016
Residuals	54	2588	47·9		

Discussion

Number of species and thousand seed weight

The harvesting method influences the plant species richness, the total number of species and the biodiversity of the seed material, as shown by Morgan *et al.* (1995) and Edwards *et al.* (2007). In the present study, the tested seed material was rich in terms of plant-species diversity, and in the mixtures obtained by both harvesting methods, the proportion of harvested grasses was higher than that of other species. This species balance can be explained by the harvesting date, at the beginning of July. Especially in the case of *Arrhenatherion* meadows, early harvesting time (June and July) results in a higher proportion of seeds of grasses under the conditions prevailing in Central Europe, whereas late harvesting (August and September) increases the proportion of seeds of forbs and legumes (Hölzel and Otte, 2003; Edwards *et al.*, 2007; Kirmer *et al.*, 2012b). The assessment of the TSW shows

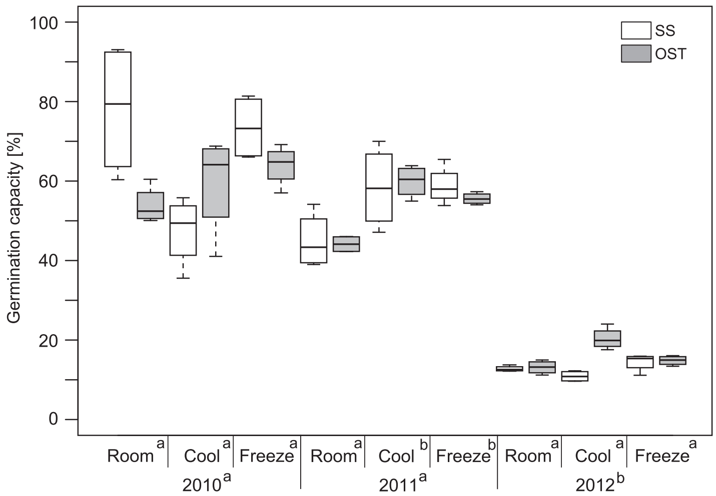

Figure 1 Germination capacity of seeds harvested by on-site threshing (OST) and seed stripping (SS) from an *Arrhenatherion* meadow stored under different conditions over 3 years. a, b – different letters indicate significant differences, based on a Tukey HSD *post hoc* test.

significantly higher values for the seed material harvested by on-site threshing. It can be assumed that the rotating brush of the seed stripper technique favours the harvesting of grasses with small and light seeds.

Germination capacity and storage conditions

Seed moisture content is one of the most important factors influencing seed quality and storability. Generally, if seed moisture content increases, storability decreases but moisture content can be higher at lower storage temperature (Taylor, 1987; ISTA, 2011). The results of the seed moisture content are below the predetermined critical value. The Austrian seed law and also Taylor (1987) prescribe a maximum moisture content for grass seeds of <14%, and for small-seeded legumes, it should be <12%.

In the present study, and independent of the storage conditions and harvesting methods, the germination rates for the seed mixtures in the first year can be considered high, compared, for instance, with the results of Heilinger and Florineth (2003) and Oliveira *et al.* (2012). In the first 2 years, the germination capacity of the SS material was, in general, higher than that of the OST material. This is in contrast to the findings of Eriksson (1999), Münzbergová and Plačková (2010) and Lönnberg and Eriksson (2013) who report that larger-seeded species have a better germination capacity than smaller-seeded species. Our results may be explained by the fact that the seed-stripping technique favours the harvesting of small-sized mature grass seeds with high germination capacity.

In the first year of this study, only the SS material stored in the cooling chamber obtained seed of a lower germination capacity. More detailed analysis showed that in these subsamples, the proportion of forbs was the lowest but, on the other hand, the purity assessment gave a similar result for both harvesting methods. Obtaining homogenous samples from such diverse seed material still remains a challenge. This problem can be reduced by taking a representative sample and an increased number of five to six subsamples. Seed material stored under cool conditions reached higher germination capacities compared with samples stored at room temperature, which is also confirmed by the findings of Welty *et al.* (1987) and Haslgrübler *et al.* (2013).

We observed a rapid reduction of the germination capacity to <15% after 3 years of storage, which indicates that the material should be used within 2 years after harvest. This has also been confirmed by other authors (Grime *et al.*, 1981; Welty *et al.*, 1987; Bonner, 1990). The longer the samples are stored, the lower the variation of the germination capacity becomes, which could be explained by different germination strategies of the various species. After storage over time, some species lose vitality earlier than others (Grime *et al.*, 1981) and some even break dormancy (Jorritsma-Wienk *et al.*, 2007). Dormancy is an innate seed property that reflects the environmental conditions in which the seed is able to germinate (Dyer, 2004; Finch-Savage and Leubner-Metzger, 2006). This should guarantee that germination occurs when conditions for establishing a new plant generation are likely to be suitable.

Seed material harvested from suitable semi-natural grasslands can be seen as a useful natural source for ecological restoration. It is important to translate research findings into practice, based on the best knowledge available and implemented by managers with up-to-date paradigms (Hobbs and Harris, 2001). To be an effective and successful measure for ecological restoration, numerous preconditions have to be fulfilled, for example site suitability and availability, harvesting time and technique as well as species richness and optimal storage conditions of the harvested material. All these characteristics show a clear influence on species composition as well as on seed quality.

Conclusions

The presented methodology provides a practicable way to determine the quality of seed mixtures harvested from an *Arrhenatherion* meadow without any expensive laboratory equipment that would be required for testing single cultivated species. The species composition of the material harvested either with a plot combine thresher or with a pull-type seed stripper is strongly influenced by the harvesting time. Knowledge about the proportion of mature seeds in the harvested seed material and its germination capacity is essential for calculation of the necessary transfer rate from donor site to receptor site. Furthermore, the quality assessment of the harvested material (e.g. presence of invasive species or undesirable contents) as well as the thousand seed weight, purity and moisture content provides essential information to determine the optimal use of the material. We conclude that storage under cool conditions contributes to the best germination results, but our results show that seed material should not be stored for more than 2 years. The findings and concluded recommendations will be integrated into specific guidelines for relevant stakeholders.

Acknowledgments

This study was supported by the Austrian Ministry for Agriculture, Forestry, Environment and Water Management, and co-financed within the frame of the CENTRAL EUROPE Program by the European Regional Development Funds (ERDF). We would like to thank all persons who contributed to this research work.

References

Adler W., Oswald K. and Fischer M. A. (2008) *Exkursionsflora für Österreich, Liechtenstein und Südtirol (Botanical field guide for Austria, Leichtenstein and South Tyrol)*. Linz, Austria: Ulmer Verlag.

Blum W.E.H., Spiegel H. and Wenzel W.W. (1996) *Bodenzustandsinventur – Konzeption, Durchführung und Bewertung; Empfehlungen zur Vereinheitlichung der Vorgangsweise in Österreich (2. Auflage) (Soil conditions inventory – design, implementation and evaluation; Recommendations for the standardization of the procedure in Austria)*. Wien, Austria: Bundesministerium für Land- und Forstwirtschaft, Bundesministerium für Wissenschaft, Verkehr und Kunst.

BMLFUW (2006) *Richtlinien für sachgerechte Düngung, Anleitung zur Interpretation von Bodenuntersuchungsergebnissen in der Landwirtschaft (Guidelines for fertilization, instructions for the interpretation of soil survey results in agriculture)*. Wien, Austria: Fachbeirat für Bodenfruchtbarkeit und Bodenschutz des Bundesministeriums für Land-, Forstwirtschaft, Umwelt und Wasserwirtschaft.

Bonner F.T. (1990) Storage of seeds: potential and limitations for germplasm conservation. *Forest Ecology and Management*, **35**, 35–43.

Budelsky R.A. and Galatowitsch S.M. (1999) Effects of moisture, temperature, and time on seed germination of five wetland Carices: implications for restoration. *Restoration Ecology*, **7**, 86–97.

Core Team R. (2012) *R: a language and environment for statistical computing*. Vienna, Austria: R Foundation for Statistical Computing.

Crawley M.J. (2007) *The R book*. Chichester, UK: John Wiley Sons.

Duprè C., Stevens C.J., Ranke T., Bleeker A., Peppler-Lisbach C., Gowing D.J.G., Dise N.B., Dorland E.D.U., Bobbink R. and Diekmann M. (2010) Changes in species richness and composition in European acidic grasslands over the past 70 years: the contribution of cumulative atmospheric nitrogen deposition. *Global Change Biology*, **16**, 344–357.

Dyer A. (2004) Maternal and sibling factors induce dormancy in dimorphic seed pairs of *Aegilops triuncialis*. *Plant Ecology*, **172**, 211–218.

Edwards A.R., Mortimer S.R., Lawson C.S., Westbury D.B., Harris S.J., Woodcock B.A. and Brown V.K. (2007) Hay strewing, brush harvesting of seed and soil disturbance as tools for the enhancement of botanical diversity in grasslands. *Biological Conservation*, **134**, 372–382.

Eriksson O. (1999) Seed size variation and its effect on germination and seedling performance in the clonal herb *Convallaria majalis*. *Acta Oecologica*, **20**, 61–66.

Finch-Savage W.E. and Leubner-Metzger G. (2006) Seed dormancy and the control of germination. *New Phytologist*, **171**, 501–523.

Grime J.P., Mason G., Curtis A.V., Rodman J. and Band S.R. (1981) A comparative study of germination characteristics in a local flora. *Journal of Ecology*, **69**, 1017–1059.

Haslgrübler P., Krautzer B., Graiss W., Blaschka A. and Pötsch E. (2013) Quality characteristics of seed material from selected species or a nutrient poor *Arrhenatherion* community. *Grassland Science in Europe*, **18**, 442–444.

Haslgrübler P., Krautzer B., Blaschka A., Graiss W. and Pötsch E.M. (2014) Quality and germination capacity of seed material harvested from an *Arrhenatherion* meadow. *Grass and Forage Science*, **69**, 454–461.

HEILINGER K. and FLORINETH F. (2003) Qualitatsprüfung von nicht gezüchtetem Saatgut (Quality aspects of uncultivated seed mixtures). *Rasen-Turf-Gazon*, **3**, 56–66.

HOBBS R.J. and HARRIS J.A. (2001) Restoration ecology: repairing the earth's ecosystems in the new millennium. *Restoration Ecology*, **9**, 239–246.

HOEKSTRA J.M., BOUCHER T.M., RICKETTS T.H. and ROBERTS C. (2005) Confronting a biome crisis: global disparities of habitat loss and protection. *Ecology Letters*, **8**, 23–29.

HÖLZEL N. and OTTE A. (2003) Restoration of a species-rich flood meadow by topsoil removal and diaspore transfer with plant material. *Applied Vegetation Science*, **6**, 131–140.

ISTA (2011) *International rules for seed testing, edition 2011*. Bassersdorf, Switzerland: The International Seed Testing Association (ISTA).

JORRITSMA-WIENK L.D., AMELOOT E., LENSSEN J.P.M. and DE KROON H. (2007) Differential responses of germination and seedling establishment in populations of *Tragopogon pratensis* (Asteraceae). *Plant Biology*, **9**, 109–115.

KIEHL K., KIRMER A., DONATH T.W., RASRAN L. and HÖLZEL N. (2010) Species introduction in restoration projects – evaluation of different techniques for the establishment of semi-natural grasslands in Central and Northwestern Europe. *Basic and Applied Ecology*, **11**, 285–299.

KIRKHAM F.W., BHOGAL A., CHAMBERS B.J., DUNN R.M. and TALLOWIN J.R.B. (2013) Effects of spreading species-rich green hay on the botanical composition of an agriculturally improved hay meadow in northern England. *Grass and Forage Science*, **68**, 260–270.

KIRMER A., BAASCH A. and TISCHEW S. (2012a) Sowing of low and high diversity seed mixtures in ecological restoration of surface mined-land. *Applied Vegetation Science*, **15**, 198–207.

KIRMER A., KRAUTZER B., SCOTTON M. and TISCHEW S. (2012b) *Praxishandbuch zur Samengewinnung und Renaturierung von artenreichem Grünland (Practical handbook for seed harvest and ecological restoration of species-rich grasslands)*. Anhalt, Germay: Hochschule Anhalt, Irdning, Austria: Lehr- und Forschungszentrum Raumberg-Gumpenstein.

KOCH W. (1926) Die Vegetationseinheiten der Linthebene (Vegetation units of the Linth plain). *Jahrbuch der St. Gallischen Naturwissenschaftlichen Gesellschaft für das Vereinsjahr*, **61**, 1–146.

KRAUSE B. and CULMSEE H. (2013) The significance of habitat continuity and current management on the compositional and functional diversity of grasslands in the uplands of Lower Saxony, Germany. *Flora – Morphology, Distribution, Functional Ecology of Plants*, **208**, 299–311.

KRAUTZER B. and KLUG B. (2009) Renaturierung von subalpinen und alpinen Ökosystemen (Restoration of subalpine and alpine ecosystems). In: Zerbe S. and Wiegleb G. (eds) *Renaturierung von Ökosystemen in Mitteleuropa (Restoration of ecosystems in central Europe)*, pp. 209–234. Heidelberg, Germany: Spektrum Akademischer Verlag.

LENCOVÁ K. and PRACH K. (2011) Restoration of hay meadows on ex-arable land: commercial seed mixtures vs. spontaneous succession. *Grass and Forage Science*, **66**, 265–271.

LOCHNER D. (1997) Restoration and reclamation review. *Student On-Line Journal*, **2**, 1–6.

LÖNNBERG K. and ERIKSSON O. (2013) Rules of the seed size game: contests between large-seeded and small-seeded species. *Oikos*, **122**, 1080–1084.

MEIER U. (2001) *Entwicklungsstadien mono- und dikotyler Pflanzen – BBCH Monografie (Development stages of monocotyledon and dicotyledon species)*. Berlin, Germany: Biologische Bundesanstalt für Land- und Forstwirtschaft.

MITCHLEY J., JONGEPIEROVÁ I. and FAJMON K. (2012) Regional seed mixtures for the re-creation of species-rich meadows in the White Carpathian Mountains: results of a 10-yr experiment. *Applied Vegetation Science*, **15**, 253–263.

MOLDER F. (1995) *Vergleichende Untersuchungen mit Verfahren der oberbodenlosen Begrünung unter besonderer Berücksichtigung areal- und standortbezogener Ökotypen (Comparative study of revegetation methods without topsoil regarding site specific ecotypes)*. Gießen, Germany: Justus-Liebig-Universität Gießen.

MORGAN J.P. and COLLICUTT D.R. (1994) Seed stripper harvesters. *Restoration and Management Notes*, **12**, 51–54.

MORGAN J.P., COLLICUTT D.R. and THOMAS J.D. (1995) *Restoring Canada's native prairies, a practical manual*. Winnipeg, MB, Canada: Prairie Habitats.

MUCINA L., GRABHERR G. and ELLMAUER T. (1993) *Die Pflanzengesellschaften Osterreichs. Teil I: Anthropogene Vegetation (Plant communities of Austria. Part I: anthropogenic vegetation)*. Jena, Germany: Fischer Verlag.

MÜNZBERGOVÁ Z. and PLAČKOVÁ I. (2010) Seed mass and population characteristics interact to determine performance of *Scorzonera hispanica* under common garden conditions. *Flora – Morphology, Distribution, Functional Ecology of Plants*, **205**, 552–559.

OLIVEIRA G., NUNES A., CLEMENTE A. and CORREIA O. (2012) Testing germination of species for hydroseeding degraded Mediterranean areas. *Restoration Ecology*, **20**, 623–630.

PYWELL R.F., BULLOCK J.M., HOPKINS A., WALKER K.J., SPARKS T.H., BURKE M.J.W. and PEEL S. (2002) Restoration of species-rich grassland on arable land: assessing the limiting processes using a multi-site experiment. *Journal of Applied Ecology*, **39**, 294–309.

RYDGREN K., NORDBAKKEN J.-F., AUSTAD I., AUESTAD I. and HEEGAARD E. (2010) Recreating semi-natural grasslands: a comparison of four methods. *Ecological Engineering*, **36**, 1672–1679.

SCHACHTSCHABEL P. (1954) Das pflanzenverfügbare Magnesium des Boden und seine Bestimmung (Plant-available magnesium of the soil and its determination).

Zeitschrift für Pflanzenernährung, Düngung, Bodenkunde, **67**, 9–23.

Schmiede R., Otte A. and Donath T.W. (2012) Enhancing plant biodiversity in species-poor grassland through plant material transfer – the impact of sward disturbance. *Applied Vegetation Science*, **15**, 290–298.

Scotton M., Piccinin L., Dainese M. and Sancin F. (2009) Seed harvesting for ecological restoration: efficiency of haymaking and seed-stripping on different grassland types in the eastern Italian Alps. *Ecological Restoration*, **27**, 66–75.

Scotton M., Kirmer A. and Krautzer B. (2012) *Practical handbook for seed harvest and ecological restoration of species-rich grasslands*, p. 116. Padova, Italy: Padova Cleup.

Shackelford N., Hobbs R.J., Burgar J.M., Erickson T.E., Fontaine J.B., Laliberté E., Ramalho C.E., Perring M.P. and Standish R.J. (2013) Primed for change: developing ecological restoration for the 21st century. *Restoration Ecology*, **21**, 297–304.

Silvertown J.W. (1981) Seed size, life span, and germination date as coadapted features of plant life history. *The American Naturalist*, **118**, 860–864.

Society for Ecological Restoration International Science & Policy Working Group (2004) *SER international primer on ecological restoration*. Tucson, AZ: Society for Ecological Restoration International. Available at: http://www.ser.org/resources/resources-detail-view/guidelines-for-developing-and-managing-ecological-restoration-projects (accessed 16 May 2014).

Stevenson M.J., Ward L.K. and Pywell R.F. (1997) Re-creating semi-natural communities: vacuum harvesting and hand collection of seed on calcareous grassland. *Restoration Ecology*, **5**, 66–76.

Taylor G. (1987) Determination of seed moisture content in small-seeded pasture legumes. *Australian Journal of Experimental Agriculture*, **27**, 377–387.

Turner S.R., Steadman K.J., Vlahos S., Koch J.M. and Dixon K.W. (2013) Seed treatment optimizes benefits of seed bank storage for restoration-ready seeds: the feasibility of prestorage dormancy alleviation for mine-site revegetation. *Restoration Ecology*, **21**, 186–192.

Wellstein C., Otte A. and Waldhardt R. (2007) Impact of site and management on the diversity of central European mesic grassland. *Agriculture, Ecosystems and Environment*, **122**, 203–210.

Welty R.E., Azevedo M.D. and Cooper T.M. (1987) Influence of moisture content, temperature, and length of storage on seed germination and survival of endophytic fungi in seeds of tall fescue and perennial ryegrass. *Phytopathology*, **77**, 893–900.

Wesche K., Krause B., Culmsee H. and Leuschner C. (2012) Fifty years of change in Central European grassland vegetation: large losses in species richness and animal-pollinated plants. *Biological Conservation*, **150**, 76–85.

Williams E.D. (1983) Germinability and enforced dormancy in seeds of species of indigenous grassland. *The Annals of Applied Biology*, **102**, 557–566.

Kapitel 4

“Establishment of an Arrhenatherion meadow through on-site threshing material and green hay transfer”

Zitat:

Graiss, W., Haslgrübler, P., Blaschka, A., Pötsch, E.M. und Krautzer, B. (2013): Establishment of an Arrhenatherion meadow through on-site threshing material and green hay transfer. Grassland Science in Europe, Vol. 18th „The Role of Grasslands in a Green Future. Threats and Perspectives in Less Favoured Areas”, edited by Á. Helgadóttir and A. Hopkins, Akureyri, 341-343.

Establishment of an *Arrhenatherion* meadow through on-site threshing material and green hay transfer

Graiss W., Haslgrübler P., Blaschka A., Pötsch E.M. and Krautzer B.
Agricultural Research and Education Centre (AREC) Raumberg-Gumpenstein, Raumberg 38, A-8952 Irdning, Austria
Corresponding author: Bernhard.Krautzer@raumberg-gumpenstein.at

Abstract

Arrhenatherion meadows are highly abundant in semi-natural grasslands of Central Europe and are regarded as an important resource for the ecological restoration of species-rich grassland. The effectiveness of different restoration methods of former intensively used arable land was tested by means of a field experiment. A donor site was harvested via i) on-site threshing (OST) and ii) fresh hay cutting - green hay (GH). The harvested material was transferred to a receptor site, which was deep ploughed before applying the seed and plant material. Three years after the establishment 20 target species with a transfer rate of 61% were observed for the OST method, resulting in a vegetation cover of target species of 72%. In the same year, the GH method achieved a number of 16 target species at the transfer rate of 52% and a share of target species at the vegetation cover of 74%. The number of target species and also the transfer rate increased over the years of observation. Both establishment methods are, in combination with an adequate site preparation, effective methods for the ecological restoration of *Arrhenatherion* meadows on former arable land. A PCoA analysis showed a trend of increasing similarity of donor site and receptor site over time, but up to now without any statistical significance.

Keywords: receptor site, target species, ecological restoration, transfer rate

Introduction

Semi-natural grasslands are the result of long and sustainable utilization of meadows and/or pastures by humans in Central Europe. *Arrhenatherion* meadows are one of the most common plant communities of these semi-natural grasslands and also an important resource for the ecological restoration of species-rich grassland. Practically relevant restoration of semi-natural grassland has been realized successfully on differing sites for many years in several European countries (Prach *et al.*, 2012; Scotton *et al.*, 2012). Restoration success can be defined as the development towards a natural, self-maintaining habitat with the physical and biological characteristics defined in the restoration project (Scotton *et al.*, 2012). The aim of this research work was to study the transfer rate of target species and the development of the vegetation cover on the receptor site established with on-site threshing material and green hay.

Materials and methods

The donor site (nutrient-poor *Arrhenatherion* meadow) is located in Upper Austria, at 48°18'N, 14°03'E; 310 m a.s.l. The receptor site was an area of plain arable land, located at 47°29'N, 14°06'E, in the Enns valley, at an altitude of 700 m a.s.l. Before establishment, site preparation was done by deep ploughing (80 cm), not only to reduce the nutrient level but also to deplete the soil seed bank of unwanted weeds (Scotton *et al.*, 2012). The soil properties of the donor site and the receptor site are presented in Table 1. The on-site threshing (OST) material was harvested with a plot combine thresher at a cutting width of 1.5 m on 1 July

2009. The harvested material was air-dried and roughly cleaned with a 6 mm sieve. After determination of purity and thousand seed weight of the OST material (described in Haslgrübler *et al*., 2011) the receptor site was sown with a seed density of 3 g m^{-2} on 25 August 2009. The green hay (GH) was harvested on 1 July 2009 with a hand-operated motor mower early in the morning, because the morning dew enables the seed to stick to the plant. The fresh material was immediately brought to the receptor site to avoid decay and seed losses, as recommended by Scotton *et al*. (2011). The green hay application was implemented immediately after cutting with 3.5 kg m^{-2} on the receptor site, representing a biomass ratio between donor: receptor site of 2:1. In the year of establishment the GH treatment received a cleaning cut after two months in order to control weeds. The experimental trials were established as a complete block design with 3 replicates of 174 m^2 each. The projective cover of the different species on the donor site was surveyed according to Schechtner (1958) in the harvesting year 2009 and on the receptor site twice a year between 2010 and 2012. The development of the different species was assessed over a period of three years under a 2-cut regime. The count of target species was done over all replicates; therefore no statistical tests were possible.

Table 1. Soil properties of the donor and restoration site.

	Gravel %	N % mass	pH $CaCl_2$	P (CAL) mg kg^{-1}	K (CAL) mg kg^{-1}
Donor site (0–20 cm)	44.14	0.52	7.2	15.8	73.9
Receptor site (0–20 cm)	47.83	0.15	6.2	56.0	91.9

A MANOVA and Principal Coordinates Analysis (PCoA) was used for data analysis, done with the statistics language R, Version 2.15.2 with the package vegan, Version 2.0-5 (Oksanen *et al*., 2012). The analysis was based on a distance matrix focusing on beta diversity calculated with the metric Jaccard index as implemented in vegan (functions 'vegdist' and 'betadisper' in vegan). The grouping variable was year, respectively age of the plot.

Results and discussion

Restoration success depends not only on the number of different species at the donor site and on the transfer rate, but also on site conditions of the receptor area. The OST method showed the highest number of species in 2010, including annual weeds like *Capsella bursa-pastoris* and *Viola arvensis*. The number of target species and also the transfer rate increased over the years. In 2012, twenty target species were observed over all replicates in the OST treatment, and the transfer rate was 60.9% of the total species count. The proportion of target species of the total projective vegetation cover increased over the years and reached 72.2% in 2012. The total number of species in the GH treatment remained stable over the years. The number of target species over all replicates increased slightly between 2010 and 2012. In 2012, the GH treatment reached a total number of 16 target species resulting in a transfer rate of 51.6% of the total species count. The mean relative cover of target species found over all replicates in the vegetation cover was 74.3%. The high amount of biomass/mulch from the GH method in the first year after establishment prevented the germination of forbs and therefore more grasses than forbs were transferred.

In 2012, three years after establishment, more target species were found in the plots of the OST treatments than in the GH plots (Table 2).

The results of the MANOVA (function 'adonis' in vegan) showed no significant differences between the two restoration methods (P = 0.883, R^2 = 0.00754) and years (P = 0.919, R^2 = 0.04828) concerning group (OST vs. GH) variance based on a dissimilarity matrix. The result of the following PCoA for donor and receptor site shows the development of the two methods OST and GH together over three years. The variance (represented by the mean distance from the centroid) within the replicates was, independent of the method in the first two years (2010 to 2011), higher compared to the donor site, and lower in 2012, though without any statistical significance. The receptor site was still rather far from the donor site in terms of its species composition; however, the development of the vegetation seems to be heading in the anticipated direction. Part of the difference might possibly be explained by the distinction of climatic conditions and soil properties of the donor and the receptor site; e.g. the pH value on the receptor site is lower and the soil P content much higher than at the donor site (Table 1).

Table 2. Species number, vegetation cover and transfer rate of target species from on-site threshing (OST) and green hay (GH).

Treatments	Donor site	Receptor site					
Parameter		OST			GH		
Date of survey	6/2009	6/2010	6/2011	6/2012	6/2010	6/2011	6/2012
Number of species	67	38	27	28	30	30	30
Numb. of target species (over all replicates)	48	16	19	20	14	15	16
Transfer rate of target species (% species)	-	50.7	60	60.9	47.7	48.5	51.6
Total cover (%)	97.6	58.7	91.0	85.6	67.0	73.3	86.0
Proportion of target species (% total cover)	83.9	70.1	55.5	72.2	79.0	73.6	74.3

Conclusion

On-site threshing and green hay are suitable harvesting methods and an effective way for an ecological restoration of *Arrhenatherion* meadows on arable land in combination with adequate site preparation, as shown by the results of our project. Four years after establishment of the experimental site there were no significant differences found between on-site threshing and green hay, and therefore it can be stated that the choice of restoration method for establishing an *Arrhenatherion* meadow depends more on the availability of equipment and other practical conditions, such as the accessibility of the sites.

References

Haslgrübler P., Krautzer B. and Graiss W. (2011) Germination capacity of threshed material from an *Arrhenatherion* meadow. *Grassland Science in Europe* 16, 523-525.

Oksanen J., Guillaume Blanchet F., Kindt R., Legendre P., Minchin P.R., O'Hara R.B., Simpson G.L., Solymos P., Stevens M.H.H. and Wagner H. (2012) Vegan: Community Ecology Package. R package version 2.0-5. http://CRAN.R-project.org/package=vegan.

Prach K., Jongepierová I. and Řehounková K. (2012) Large-scale restoration of dry grasslands on ex-arable land using a regional seed mixture: establishment of target species. *Restoration Ecology* 21, 33-39.

Schechtner G. (1958) Grünlandsoziologische Bestandsaufnahme mittels,Flächenprozentschätzung. *Zeitschrift für Acker- und Pflanzenbau* 105, 33-43.

Scotton M., Kirmer A. and Krautzer B. (2012) *Practical Handbook for Seed Harvest and Ecological Restoration of Species-Rich Grasslands*, CLEUP, Padova, Italy, 116 pp.

Diskussion und Ausblick

Dem Verlust der Biodiversität entgegenzuwirken und einen nachhaltigen, schonenden Umgang mit den immer knapper werdenden Ressourcen zu pflegen, ist zu einer wesentlichen gesellschaftlichen Zielsetzung geworden. Auf internationaler sowie auf nationaler Ebene wird ständig an der Weiterentwicklung der Ernte- und Etablierungsmethoden gearbeitet, um naturnahe Grünlandgesellschaften als Quelle zur Steigerung der Biodiversität zu nutzen und um die ökologische und nachhaltige Sanierung geeigneter Begrünungsflächen zu gewährleisten. Die Gewinnung und Verwendung regionaler Wildpflanzen bei Begrünungs- und Rekultivierungsmaßnahmen ist ein wesentlicher Beitrag dazu. Gerade im Naturschutz bietet sich gemeinsam mit der Landwirtschaft großflächig die Möglichkeit, selten gewordene Grünlandgesellschaften der frischen und halbtrockenen Standorte durch Kombination mit passendem Begrünungsmaterial wieder in der Landschaft zu etablieren (Kirmer et al., 2012). Aber auch im Zuge landschaftsbaulicher Maßnahmen sowie im besiedelten Bereich können optisch attraktive Samenmischungen von ökologisch wertvollem Grünland (High Nature Value Farmland) beispielsweise auf öffentlichen Grünflächen (z.B. Straßenböschungen, Hochwasserschutzdämme, Verkehrsinseln etc.), bei der Begrünung von Gewerbeflächen, auf Flachdächern aber auch im privaten Garten verwendet werden (ÖAG, 2000, ÖNORM, 2014a, 2014b).

Aufgrund der spezifischen Anpassungen der Pflanzenarten an die jeweiligen Standortverhältnisse muss jedoch eine Spenderfläche mit vergleichbaren Standorteigenschaften zur Empfängerfläche ausgewählt werden, damit die Arten optimal an die Bedingungen der Empfängerfläche angepasst sind. Um einen möglichst hohen Anteil an Zielarten zu erreichen, muss der Erntezeitpunkt auf die Samenreife der Zielarten abgestimmt werden (Donath et al., 2007, Hölzel und Otte, 2003). Sind von den Zielarten nicht ausreichend Samen im Erntegut vorhanden oder fehlen wichtige Arten, so ist eine mehrfache Beerntung des Bestandes oder eine Zusaat von regionalem Wildpflanzensaatgut in Erwägung zu ziehen (Kirmer et al., 2012, Scotton et al., 2012). Für die Einschätzung potenziell übertragbarer Arten auf der Spenderfläche ist kurz vor der Ernte eine Vegetationsaufnahme mit Erfassung des phänologischen Zustandes empfehlenswert (Bohner et al., 2011, Meier, 2001). In den letzten Jahrzehnten haben sich unterschiedliche Erntemethoden als praktikabel erwiesen, und diese werden ständig verbessert (Baasch et al., 2012, Jongepierová et al., 2007, Mitchley et al., 2012, Prach et al., 2012). Die gängigsten Methoden zur Ernte von ökologisch wertvollen Samenmischungen sind die Ernte von frischem Grünschnitt, Wiesendrusch, Ausbürstmaterial sowie der Einsatz von regionalem Wiesensaatgut aus landwirtschaftlicher Produktion (Kapitel 1). Die Samen- und Biomasseproduktion eines Wiesenbestandes sind abhängig vom Niederschlags- und Temperaturverlauf im Jahr, das heißt, der Erntezeitpunkt und die Erntemenge sind vom Witterungsverlauf der jeweiligen Vegetationsperiode abhängig.

Die Zerlegungsproben der beiden Erntemethoden ergaben, dass die Anzahl der Zielarten unterschiedlich war, jedoch die prozentuelle Verteilung von Gräsern, Kräutern und Leguminosen gleich war. Der Anteil an Gräsern lag bei 80 %, was für Glatthaferwiesen, die Anfang Juli beerntet werden, typisch ist. Speziell bei trockenen bis halbtrockenen Wiesentypen ist bei einer frühen Ernte im Juni der prozentuelle Anteil der Gräser höher, während eine Ernte im Juli oder August den Anteil der Kräuter fördert (Edwards et al., 2007, Hölzel und Otte, 2003). Generell wäre es

daher wünschenswert, eine frühere und eine spätere Ernte zu mischen, um ein möglichst hohes Artenspektrum zu gewinnen (Kirmer et al., 2012). Nach der Ernte wurden der Wiesendrusch und das Ausbürstmaterial luftgetrocknet und mit einem 6 mm Sieb grob gereinigt, danach wurde der Anteil an reinen Samen festgestellt. Es zeigte sich, dass der Anteil an reinen Samen bei beiden Erntemethoden (Wiesendrusch und Ausbürstmaterial) gleich war. Die Untersuchungen des Tausendkorngewichtes ergaben dagegen unterschiedliche Ergebnisse. Im Wiesendrusch waren größere und schwerere Samen. Das lässt den Schluss zu, dass bei der Ausbürstmethode eher kleinere und leichtere Samen geerntet werden. Um möglichst alle reifen Samen (große und kleine) zu ernten sollten im Idealfall beide Erntemethoden zum Einsatz kommen.
Zur Bestimmung der Keimfähigkeit wurde eine Methode angewendet, bei der nach dem Ausschlussverfahren gearbeitet wurde (Haslgrübler et al., 2013, 2014) (Kapitel 2,3). Bei den Vortests im Keimschrank wurde ein Substrat definiert und festgestellt, ob Maßnahmen zum Abbau der Keimhemmung notwendig sind. Bei den Haupttests im Gewächshaus wurden die Lagertemperatur sowie die Dauer der Lagerung getestet. Bei den ersten Tests im Keimschrank wurde die Keimfähigkeit auf drei verschiedenen Substraten (sterile Ansaaterde, Quarzsand und Filterpapier) getestet. Sterile Ansaaterde erzielte die besten und homogensten Ergebnisse. Dieses Substrat ist sehr gut verfügbar und hat außerdem die höchste Wasserspeicherkapazität. In weitere Folge wurde getestet, ob der Keimerfolg durch Maßnahmen wie Vorkühlen, Einsatz von Kaliumnitrat (KNO_3) oder Gibberellinsäure (GA_3) erhöht werden konnte. Diese keimbrechenden Maßnahmen werden in der Literatur (Godefroid et al., 2010, Grime et al., 1981, ISTA, 2011, Williams, 1983) für Einzelarten empfohlen, die auch in der verwendeten Samenmischung enthalten waren. Der Test dieser Maßnahmen im Rahmen der vorliegenden Arbeit zeigte jedoch negative Wirkungen auf die Keimfähigkeit, daher wurden für die weiteren Tests keine Maßnahmen zum Abbau der Keimhemmung angewendet (Kapitel 2). Der Effekt keimbrechender Maßnahmen muss in Abhängigkeit vom Vegetationstyp differenziert betrachtet werden. Das heißt für Glatthaferwiesen zeigt sich ein negativer Effekt, hingegen bei Pfeifengraswiesen ist dieser deutlich positiv (Graiss et al., 2009, Krautzer et al., 2013). Nach Abschluss der Vortests, wurden die Haupttests im Gewächshaus durchgeführt (Kapitel 3). Nach der Trocknung und Reinigung wurde das Samenmaterial bei unterschiedlichen Temperaturen (Raumtemperatur 18-23°C, Kühlraum 2-5°C, Gefrierschrank -18°C) gelagert. Die Keimfähigkeitstests wurden für alle Lagertemperaturen über drei Jahre wiederholt.
Die Ergebnisse im ersten Jahr zeigten sehr hohe Keimfähigkeitswerte verglichen mit denen von Heilinger und Florineth (2003), sowie von Oliveira (2012). Im dritten Jahr sank die Keimfähigkeit rapide unter 15 %. Diese Beobachtung wurde auch schon von anderen Autoren (Bonner, 1990, Grime et al., 1981, Welty et al., 1987) bestätigt. Je länger die Samen gelagert werden, desto geringer wird die Keimfähigkeit der einzelnen Arten und somit sinkt auch die Keimfähigkeit der gesamten Mischung (Grime et al., 1981). Es zeigte sich, dass Wiesendrusch oder Ausbürstmaterial maximal für zwei Jahre unter kühlen Bedingungen (Kühlraum 2-5°C) gelagert werden sollte. Die Keimfähigkeitsprüfung im Gewächshaus erwies sich als praktikabel und kostengünstig im Gegensatz zu Keimfähigkeitsprüfungen von Einzelarten in einem zertifizierten Labor. Aus den Ergebnissen zu Reinheit, Tausendkorngewicht und Keimfähigkeit wird bestimmt, wie viele Gramm

des grob gereinigten Materials ausgesät werden muss, um pro Quadratmeter Begrünungsfläche ca. 2000 bis maximal 5000 Samen aufzubringen (Kirmer et al., 2012). Das Samenmaterial sollte eine Mindestkeimfähigkeit von 50 % erreichen, damit es verwendet werden kann. Um einen abnahmefähigen Zustand der mit solchen Materialien begrünten Empfängerfläche zu gewährleisten, sollte nach einer Vegetationsperiode eine Vegetationsdeckung von mindestens 60 % erreicht werden (Kirmer et al., 2012, Scotton et al., 2012). Ist das nicht der Fall, kann eine Nachsaat erforderlich sein. Vor Anlage der Empfängerfläche wurde diese rigolt (tiefengepflügt), um den Konkurrenzdruck durch unerwünschte Arten so gering wie möglich zu halten. Weiters wurde im Herbst 2009 ein Reinigungsschnitt durchgeführt, um sicher zu gehen, dass sich die Zielvegetation etablieren konnte (Kapitel 4). Auf der Empfängerfläche wurden frischer Grünschnitt und Wiesendrusch getestet. Frischer Grünschnitt kann nicht gelagert werden, und somit ist für die Anlage einer Fläche nur eine Ernte möglich, allerdings wird die Empfängerfläche gut gegen Erosion und Austrocknung geschützt (Kirmer et al., 2012). Im gegenständlichen Fall war das Übertragungsverhältnis von Spenderfläche zu Empfängerfläche 2,5 : 1. Dieses Verhältnis wird in der Literatur (Kirmer et al., 2012, Scotton et al., 2012) empfohlen, allerdings konnte festgestellt werden, dass die Mulchschicht teilweise zu dick war und somit die aufkeimenden Pflanzen erstickt wurden. Es ist daher empfehlenswert, vor der Ernte eine Schnittprobe zu entnehmen und diese zu analysieren, um das genaue Übertragungsverhältnis zu bestimmen. Für ein optimales Wachstum darf die Dicke der Mulchschicht nie mehr als 3-4 cm betragen, das Material muss locker aufgebracht und noch ausreichend lichtdurchlässig sein (Kirmer et al., 2012, Scotton et al., 2012). Die Vegetationsentwicklung auf der untersuchten Empfängerfläche zeigte, dass es nach vier Jahren keine signifikanten Unterschiede hinsichtlich der Anzahl der übertragenen Zielarten zwischen den beiden Erntemethoden (Grünschnitt bzw. Wiesendrusch) mehr gibt. Ausschlaggebend für den Begrünungserfolg ist eine gute Flächenvorbereitung. Die untersuchten Erntemethoden eignen sich sehr gut für Rekultivierungsprojekte (Scotton et al., 2012).
Bei der Verwendung von standortgerechten Saatgutmischungen ist zu beachten, dass auch dem Saatgutgesetz unterliegende Arten, wie beispielsweise Glatthafer, Wiesenfuchsschwanz, Rotschwingel oder Hornklee enthalten sein können. Hier sind die Vorgaben der Europäischen Richtlinie 2010/60/EU zu beachten, welche das Inverkehrbringen von Futterpflanzen-Saatgutmischungen zur Erhaltung der natürlichen Umwelt (sogenannte Erhaltungsmischungen) regelt. Außerdem gibt diese Richtlinie vor, dass die Keimraten der Bestandteile in der Erhaltungsmischung ausreichend sein müssen, um die Art des Lebensraumes am Entnahmeort wieder herzustellen. Entsprechende Prüfungen sind laut Richtlinie nach den international gebräuchlichen Methoden bzw., sofern diese nicht existieren, nach anderweitigen geeigneten Methoden durchzuführen. Das heißt, dass die in dieser Arbeit vorgestellten Methoden zur Prüfung der Reinheit, des Tausendkorngewichtes und der Keimfähigkeit hier Anwendung finden können. Um den Einsatz solcher Saatgutmischungen auch national umzusetzen, erschien in Österreich im Jahr 2014 die ÖNORM L1113 Begrünung mit Wildpflanzensaatgut und ONR 12113 Begrünung mit Wildpflanzensaatgut – Lebensraumtypen und Saatgutmischungen (ÖNORM, 2014a, 2014b). Die Anwendung von Wildpflanzensaatgut im Landschaftsbau bedingt das Einhalten von Standards, die zum Teil im Widerspruch zu bestehenden normativen Vorgaben stehen, worauf bereits bei der Formulierung von Ausschreibungen geachtet werden muss. Die Funktionsfähigkeit und die Kosteneffektivität der unterschiedlichen Methoden muss von Fall zu Fall separat bestimmt und ermittelt werden.

Literatur zur Rahmenschrift

AGES (2004): Normen und Verfahren der repräsentativen Probenahme einschließlich Kontrolle der Kennzeichnung, Verpackung und Verschließung, Österreich, 40 S.

Baasch, A., Kirmer, A. und Tischew, S. (2012): Nine years of vegetation development in a postmining site: effects of spontaneous and assisted site recovery. Journal of Applied Ecology 49 (1), 251-260.

Bartel, A. und Schwarzl, B. (2008): Agrar-Umweltindikator ‚High Nature Value Farmland' – Verifizierung der Gebietskulisse für Österreich, Studie des Umweltbundesamtes im Auftrag des BMLFUW, Wien, 118 S.

Bogner, D. (1992): Entwicklung der Landwirtschaft auf der Welser Heide und ihr Einfluß auf die Landwirtschaft seit der Frühzeit. Kataloge des OÖ. Landesmuseums N.F. 54, 401-408.

Bohner, A., Krautzer, B., Starz, W., Graiss, W. und Haslgrübler, P. (2011): Extensive Wiesen - Bedeutung, Nutzung und Pflege, Sonderbeilage 03/2011. ÖAG - Österreichische Arbeitsgemeinschaft für Grünland, Irdning.

Bonner, F.T. (1990): Storage of seeds: Potential and limitations for germplasm conservation. Forest Ecology and Management 35 (1–2), 35-43.

Donath, T.W., Bissels, S., Hölzel, N. und Otte, A. (2007): Large scale application of diaspore transfer with plant material in restoration practice - Impact of seed and microsite limitation. Biological Conservation 138 (1-2), 224-234.

Edwards, A.R., Mortimer, S.R., Lawson, C.S., Westbury, D.B., Harris, S.J., Woodcock, B.A. und Brown, V.K. (2007): Hay strewing, brush harvesting of seed and soil disturbance as tools for the enhancement of botanical diversity in grasslands. Biological Conservation 134 (3), 372-382.

Elias, D. und Thiede, S. (2008): Verfrachtung von Heuchschrecken (Insecta: Ensifera et Caelifera) mit frischem Mähgut im Wulfener Bruch (Sachsen-Anhalt). Hercynia N.F. 41, 253-262.

Ellenberg, H. und Leuschner, C. (2010): Vegetation Mitteleuropas mit den Alpen: In ökologischer, dynamischer und historischer Sicht, Ulmer Eugen Verlag. 1357 S.

EU (2010): RICHTLINIE 2010/60/EU Der Kommission - mit Ausnahmeregelungen für das Inverkehrbringen von Futterpflanzensaatgutmischungen zur Erhaltung der natürlichen Umwelt. (07.08.2014).

Florineth, F. (2012): Pflanze statt Beton: Sichern und Gestalten mit Pflanzen, Patzer Verlag, Berlin-Hannover. 272 S.

Godefroid, S., Van de Vyver, A. und Vanderborght, T. (2010): Germination capacity and viability of threatened species collections in seed banks. Biodiversity and Conservation 19 (5), 1365-1383.

Graiss, W., Krautzer, B. und Blaschka, A. (2009): Anlage und Pflege naturschutzfachlich wertvoller Streuwiesen im mittleren Ennstal, Lehr- und Forschungszentrum für Landwirtschaft Raumberg-Gumpenstein, Irdning, 20 S.

Grime, J.P., Mason, G., Curtis, A.V., Rodman, J. und Band, S.R. (1981): A Comparative Study of Germination Characteristics in a Local Flora. Journal of Ecology 69 (3), 1017-1059.

Haslgrübler, P., Krautzer, B., Blaschka, A., Graiss, W. und Pötsch, E.M. (2013): Quality and germination capacity of seed material harvested from an Arrhenatherion meadow. Grass and Forage Science 69 (3), 454-461.

Haslgrübler, P., Krautzer, B., Blaschka, A., Graiss, W. und Pötsch, E.M. (2014): Influence of different storage conditions on quality characteristics of seed material from semi-natural grassland. Grass and Forage Science 70 (3), 549-556

Hebeisen, T. und Graff, L. (2008): Richtlinie für die Probenahme, Kennzeichnung und Verschließung von Saatgut, Schweizerische Eidgenossenschaft, Zürich, 23 S.

Heilinger, K. und Florineth, F. (2003): Qualitatsprüfung von nicht gezüchtetem Saatgut. Rasen Turf Gazon 3, 56-66.

Hölzel, N. und Otte, A. (2003): Restoration of a species-rich flood meadow by topsoil removal and diaspore transfer with plant material. Applied Vegetation Science 6 (2), 131-140.

Hopkins, Pywell, Peel, Johnson und Bowling (1999): Enhancement of botanical diversity of permanent grassland and impact on hay production in Environmentaly Sensitive Areas in the UK. Grass and Forage Science 54 (2), 163-173.

ISTA (2011): International Rules for Seed Testing, Edition 2011, The International Seed Testing Association (ISTA), Bassersdorf.

Jongepierová, I., Mitchley, J. und Tzanopoulos, J. (2007): A field experiment to recreate species rich hay meadows using regional seed mixtures. Biological Conservation 139 (3-4), 297-305.

Kiehl, K. und Wagner, C. (2006): Effects of hay transfer on long-term establishment of vegetation and grasshoppes on former arable land. Restoration Ecology 14, 157-166.

Kirmer, A. und Tischew, S. (2006): Handbuch naturnahe Begrünung von Rohböden, Strategien und Beispiele für eine nachhaltige und ökologische Renaturierung, Vieweg Teubner Verlag, Wiesbaden. 184 S.

Kirmer, A., Krautzer, B., Scotton, M. und Tischew, S., Eds. (2012): Praxishandbuch zur Samengewinnung und Renaturierung von artenreichem Grünland, Hochschule Anhalt, Irdning, Lehr- und Forschungszentrum Raumberg-Gumpenstein, 221 S.

Koch, W. (1926): Die Vegetationseinheiten der Linthebene, unter Berücksichtigung der Verhältnisse in der Nordostschweiz, systematisch-kritische Studie. Doktorarbeit der Eidgenössischen technischen Hochschule in Zürich, St. Gallen, 134 S.

Krautzer, B. (2015): Rahmenbedingungen für Produktion und Anwendung von Wildpflanzensaatgut. Begrünung mit Wildpflanzensaatgut, HBLFA Raumberg-Gumpenstein. Irdning, 1-6.

Krautzer, B. und Hacker, E., Eds. (2006): Ecological Restoration with Native Plant and Seed Material. Soil Bioengineering, HBLFA Raumberg-Gumpenstein, Irdning, German Federation for Soil-Bioengineering, Achen, 291 S.

Krautzer, B., Haslgrübler, P., Blaschka, A., Graiss, W. und Pötsch, E. (2013): Quality characteristics of seed material harvested from Molinion litter meadows. Grassland Science in Europe 17th Symposium of the European Grassland Federation „The Role of Grasslands in a Green Future. Threats and Perspectives in Less Favoured Areas, Akureyri, 487-489 S.

Krautzer, B. und Klug, B. (2009): Renaturierung von subalpinen und alpinen Ökosystemen. In ZERBE, S. and WIEGLEB, G.: Renaturierung von Ökosystemen in Mitteleuropa, Spektrum Akademischer Verlag, Heidelberg, 209-234 S.

Meier, U. (2001): Entwicklungsstadien mono- und dikotyler Pflanzen - BBCH Monografie, Biologische Bundesanstalt für Land- und Forstwirtschaft, Berlin, 165 S.

Mitchley, J., Jongepierová, I. und Fajmon, K. (2012): Regional seed mixtures for the re-creation of species-rich meadows in the White Carpathian Mountains: results of a 10-yr experiment. Applied Vegetation Science 15 (2), 253-263.

Molder, F. (1995): Vergleichende Untersuchungen mit Verfahren der oberbodenlosen Begrünung unter besonderer Berücksichtigung areal- und standortbezogener Ökotypen. Institut für Bodenkunde und Bodenerhaltung, Gießen, 175 S.

Molder, F. (2008): Keimfähigkeitsprüfung von inhomogenen Diasporengemischen. Die neue Landschaft 8 54-57.

Morgan, J.P. und Collicutt, D.R. (1994): Seed stripper harvesters. Restoration & Management Notes 12, 51-54.

Morgan, J.P., Collicutt, D.R. und Thomas, J.D. (1995): Restoring Canada's native Prairies, A pracital Manaual, Prairie Habitas, Manitoba, Canada. Prairie Habitats.

ÖAG (2000): Richtlinie für standortgerechte Begrünungen; Ein Regelwerk im Interesse der Natur, Österreichische Arbeitsgemeinschaft für Grünland und Futterbau (ÖAG), HBLFA Raumberg-Gumpenstein.

Oliveira, G., Nunes, A. Clemente, A. und Correia, O. (2012): Testing Germination of Species for Hydroseeding Degraded Mediterranean Areas. Restoration Ecology 20 (5), 623-630.

ÖNORM (2014a): Bergrünung mit Wildpflanzensaatgut L 1113. Österreichisches Normungsinstitut, Wien.

ÖNORM (2014b): Begrünung mit Wildpflanzensaatgut - Lebensraumtypen und Saatgutmischungen L121113. Österreichisches Normungsinstitut, Wien.

Parsons, A.J., Rasmussen, S., Liu, Q., Xue, H., Ball, C. und Shaw, C. (2013): Plant growth – resource or strategy limited: insights from responses to gibberellin. Grass and Forage Science 68 (4), 577-588.

Pötsch, E.M. (1997): Auswirkungen langjähriger Wirtschafts- und Mineraldüngeranwendung auf Pflanzensoziologie, Ertrag, Futterinhaltsstoffe und Bodenkennwerte von Dauergrünland. Institut für Pflanzenbau, Wien, 116 S.

Pötsch, E.M., Krautzer, B. und Buchgraber, K. (2012): Status quo und Entwicklung des Extensivgrünlandes im Alpenraum. Lehr- und Forschungszentrum Raumberg-Gumpenstein, 17. Alpenländisches Expertenforum, Irdning. 1-8.

Prach, K., Jongepierová, I. und Řehounková, K. (2012): Large-Scale Restoration of Dry Grasslands on Ex-Arable Land Using a Regional Seed Mixture: Establishment of Target Species. Restoration Ecology, 33-39.

Rao, N., Hanson, J., Dulloo, M., Ghosh, K., Nowell, D. und Larinde, M. (2006): Manual of seed handling in genebanks. Handbooks for Genebanks, Bioversity International, Rome, Italy. 147 S.

SaatG 1997 (1997): Bundesgesetz über die Saatgutanerkennung, die Saatgutzulassung und das Inverkehrbringen von Saatgut sowie die Sortenzulassung (Saatgutgesetz 1997 - SaatG 1997), BGBl. I Nr. 72/1997 idF BGBl. I Nr. 39/2000, BGBl. I Nr. 108/2001, BGBl. I Nr. 109/2001, BGBl. I Nr. 110/2002 und BGBl I Nr. 83/2004.

Schechtner, G. (1958): Grünlandsoziologische Bestandsaufnahme mittels „Flächenprotzentschätzung". Sonderdruck aus „Zeitschrift für Acker- und Pflazenbau" 105 (1), 33-43.

Scotton, M., Kirmer, A. und Krautzer, B., Eds. (2012): Practical handbook for seed harvest and ecological restoration of species-rich grasslands, Cleup, Padova 116 S.

Shepley, S.C., Chang, J.L.L. und Chang, C. (1972): Does Gibberellic Acid Stimulate Seed Germination via Amylase Synthesis? Plant Physiology 49 (3), 441-442.

Weinzierl, v.T.R. (1917): Das k. k. Kraglgut eine Weide- und Versuchswirtschaft in Österreich, M. & H. Schaper Verlag Hannover.

Welty, R.E., Azevedo, M.D. und Cooper, T.M. (1987): Influence of moisture content, temperature, and length of storage on seed germination and survival of endophytic fungi in seeds of tall fescue and perennial ryegrass. Phytopathology 77 (6), 893-900.

Williams, E.D. (1983): Effects of temperature, light, nitrate and pre chilling on seed germination of grassland plants. Annals of Applied Biology 103 (1), 161-172.