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Suction samplers are a valuable tool to sample arthropod assemblages for conservation translocation

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Abstract

An important component of recent nature conservation is the ecological restoration of semi-natural grasslands. The aim of such projects is usually the restoration of typical plant communities; translocation of animals, by contrast, plays only a minor role. This is based on the assumption that a recovery of the flora will lead to recovered fauna; however, this is not always the case. Suction samplers with gauze collection bags are well suited to sample arthropods, and they may also be helpful for transferring animals. However, to date, the suitability of suction samplers as a translocation tool is unclear due to a lack of empirical data on the mortality rate of the sampled arthropod taxa. In this study, we sampled arthropods (leafhoppers, spiders, beetles, and true bugs) with a suction sampler on 21 calcareous grasslands. Immediately after sampling, animals were stored in collection bags and their mortality rate was determined. We compared storage periods (1, 2, and 3 h) and tested the suitability of a cool box to reduce mortality rates. Our study revealed that arthropod mortality was generally low (9% of all sampled individuals); however, the survival rate was affected by (1) storage time, (2) storage conditions, and (3) arthropod group. The mortality of beetles and true bugs was very low and not influenced by storage time or storage conditions. In contrast, leafhoppers and spiders had higher mortality, which increased with storage time and decreased by the use of a cool box. According to our results, suction samplers can be a valuable tool to sample arthropod assemblages for conservation translocation. In order to reduce mortality in sensitive groups such as leafhoppers and spiders, the storage process can be optimised. We thus recommend (1) using a cool box and (2) minimising the period until release of the collected arthropods at the restored site.

Introduction

Ecological restoration of semi-natural grasslands has become an increasingly important component of nature conservation throughout Europe (Bakker & Berendse, 1999; Strijker, 2005; Kiehl et al., 2010; Kollmann et al., 2019; Zerbe, 2019). This is the result of a considerable decline of species-rich grasslands, assumed to be primarily driven by land-use change (Sala et al., 2000; Foley et al., 2005; Hodgson et al., 2005; Stoate et al., 2009). For decades, the intensification of agricultural production, abandonment, afforestation, and urbanisation have been the

key drivers of the loss, fragmentation, and deterioration of semi-natural grasslands (Bakker & Berendse, 1999; WallisDeVries et al., 2002; Baur et al., 2006).

Restoration projects usually aim at reaching similar levels of (target) species diversity as observed on reference sites (Baur, 2014). However, in the course of the restoration process, including the evaluation of success, the focus is often on plant species and abiotic characteristics (Longcore, 2003; Ruiz-Jaen & Aide, 2005; Cristescu et al., 2013; Dietrich et al., 2013). This is based on the assumption that recovery of the flora will consequently lead to recovered fauna (Cristescu et al., 2013; Baur, 2014). Several studies have tackled this issue with contrasting results (Schultz et al., 2008; Cristescu et al., 2013; Baur, 2014). Whereas many highly mobile species are able to recolonise restored sites on their own, dispersal-limited animals often fail to

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do so (Zerbe & Wiegleb, 2009). Thus, Baur (2014) stressed the importance of integrating fauna-based criteria in the course of restoration projects. This is particularly important because high biodiversity, including both flora and fauna, increases the long-term resilience of an ecosystem (Fischer et al., 2006; Cardinale et al., 2012).

A common strategy to facilitate or accelerate restoration is to reintroduce plant species artificially (Kiehl et al., 2010; Godefroid et al., 2011). A variety of factors influences the establishment of the species. In addition to suitable abiotic and biotic conditions, these include the thoughtful selection of seeds, the use of seedlings, and the combination of plant material of various species-rich source populations (Kiehl et al., 2010; Godefroid et al., 2011; Prach et al., 2014). Reintroduction of animals is often restricted to single species (e.g., Thomas et al., 2009; Cristescu et al., 2013; Stringer et al., 2014; Carter et al., 2017) and there is often a disproportionate focus on mammals and birds (Fischer & Lindenmayer, 2000; Soorae, 2011, 2013, 2016, 2018; Bubac et al., 2019). However, it may be reasonable to reintroduce entire assemblages of dispersal-limited arthropod species as an accompanying measure to plant reintroductions (Baur, 2014). As with plants, species-rich source populations and suitable target patches must be available (Berger-Tal et al., 2019; Bubac et al., 2019). Additionally, large numbers of individuals for translocation are necessary (Fischer & Lindenmayer, 2000). To ensure a successful translocation, it is important that the largest possible number of animals survive when being collected and transported (JCCBI, 2010; IUCN/SSC, 2013).

Suction samplers are a valuable tool to obtain seeds for translocation in restoration projects (Zerbe & Wiegleb, 2009). Moreover, the use of a suction sampler with a finegauze collection bag on the inside of the inlet nozzle is well suited to sample arthropods (e.g., Eschen et al., 2012; Trivellone et al., 2012; Helbing et al., 2017). This sampling technique has the advantage that it enables the collection of large numbers of individuals without much effort (Standen, 2000; Stewart, 2002; Brook et al., 2008). However, it is to be expected that some individuals die inside the collection bags. Death may occur due to damage coming from sucked-in debris (Stewart, 2002; Ramires et al., 2007). Moreover, the high density of individuals and missing refuges may cause an enhanced level of stress and an increased risk of being caught by predators. In scientific studies using suction samplers, the collected animals are usually killed immediately to allow identification in the laboratory. There is no experience with the transfer of vital arthropod assemblages. Thus, empirical data on the mortality of various taxa of arthropods are lacking, but these are fundamental to assess the use of suction samplers for

conservation translocation. We therefore conducted a study on calcareous grasslands and determined the mortality rate of individuals of four arthropod taxa – leafhoppers (Hemiptera: Auchenorrhyncha), spiders (Araneae), beetles (Coleoptera), and true bugs (Hemiptera: Heteroptera) – caused by suction sampling and subsequent storing in gauze bags. We compared various storage periods and tested the use of a cool box. The rationale behind this was to reduce the physical activity of arthropods due to decreased temperatures and darkness. We hypothesize that this lowers activity-dependent lethal effects such as predation or density stress and thus reduces mortality.

Materials and methods

Study area

The study was conducted in the Diemel Valley, Germany, along the border between North Rhine-Westphalia and Hesse (51°23′N, 8°39′E and 51°36′N, 9°24′E). The valley stretches 70 km from east to west with an elevational gradient from 100 to 500 m a.s.l. The climate is suboceanic (Müller-Wille, 1981) and varies with elevation (mean annual temperature: 6.5–9 °C, mean annual precipitation: 600–1 000 mm; MURL NRW, 1989). The Diemel Valley contains the largest area of semi-dry calcareous grasslands (ca. 750 ha) in the northern half of Germany (Fartmann, 2004).

Sampling design

We sampled a total of 21 calcareous grassland patches in August and September, 2018, using a G-Vac suction sampler (Stihl SH 56, Waiblingen, Germany; 12.5 cm diameter suction tube; 710 m³ h⁻¹ air flow rate) with a fine-gauze collection bag (300 µm mesh size; ca. 6 l volume) on the inside of the inlet nozzle (Figure 1A and B). We took 50 suction samples (= a total sampled area of 0.6 m²) per collection bag and filled four collection bags in each study patch. The samples were taken randomly within each patch and only under dry and sunny weather conditions. After sampling, we removed the collection bags from the suction sampler and sealed them. We placed three of the four bags in the shade of adjacent shrubs for periods of 1, 2, and 3 h, respectively. The fourth bag was placed inside a passive cool box with two freezer packs and stored for 3 h (Figure 1C). In order to simulate vibrations caused by transport, all collection bags were turned every 15 min.

After the storage periods, each sample was poured into a bucket. All individuals that moved very actively and tried to escape were collected using a suction exhauster (Figure 1C), killed, and counted in the laboratory; similar to the methods applied by Bucher et al. (2016) and Helbing et al. (2017). The remaining individuals were counted on-







Figure 1 (A) Suction sampler. (B) Fine gauze collection bag (300 μm mesh size; ca. 6 l volume) on the suction tube. (C) Passive cool box and suction exhauster. [Colour figure can be viewed at wile yonlinelibrary.com]

site and released afterwards. All individuals classified as alive after storing had no visible damage; hence, time-de-layed mortality due to the sampling and storage process is very unlikely. By sampling leafhoppers, spiders, beetles, and true bugs, our samples consisted of the majority of transferable arthropods when using a suction sampler (Brook et al., 2008; Sanders & Entling, 2011).

Throughout the duration of sample storage, we measured temperatures adjacent to the collection bags outside and inside the cool box using data loggers (iButton; Maxim Integrated, San José, CA, USA) with a measurement resolution of 1 min and an accuracy of 0.5 °C. To prevent the logger outside the cool box from being affected by direct sunlight, it was attached to a radiation shield at a height of 28 cm above the ground.

Statistical analysis

Prior to analyses, we visually checked histograms for normal distribution and homoscedasticity (Quinn & Keough, 2002). Normally distributed data or data that could be normalized by transformation were analysed via parametric tests, otherwise we applied non-parametric approaches. Differences between the four taxonomic groups (leafhoppers, spiders, beetles, and true bugs) and between the three storage periods (1, 2, and 3 h) were compared using repeated measures ANOVA (parametric: rANOVA; non-parametric: Friedman test). As post-hoc test, we applied the Holm-Sidak test and Dunn's test, respectively. Storage times of 3 h outside the cool box and 3 h inside the cool box were compared with a paired t-test (parametric) or paired Wilcoxon-test (non-parametric). We used the paired t-test to test for significant differences in the

temperatures outside and inside the cool box. All tests were conducted with SigmaPlot v.14 (Systat Software, San José, CA, USA).

Results

We counted in total 26 357 individuals in the suction samples of the 21 study patches. The four taxonomic groups were found in different abundance (Friedman test: $\chi^2 = 53.97$, d.f. = 3, P<0.001). Leafhoppers (12 782 specimens) and spiders (10 551) were most abundant, beetles (2 223) and true bugs (801) were also sampled in all patches, but in much lower abundance (Figure 2).

Temperature was significantly lower inside that outside the cool box (Figure 3); however, the cooling capacity of the box was poor, as the median temperature inside the box over a period of 3 h was 22 °C. This is also reflected by the minimum values, which ranged between 13 and 26 °C (median 21 °C).

Mortality rate was generally low with an overall number of 2 437 dead individuals (9% of all sampled individuals). After a storage period of 1 h without cooling, about 7% of the leafhoppers had died (Figure 4A). Extending the storage time increased mortality (maximum of 19% after 3 h; rANOVA: $F_{2,40} = 23.87$, P<0.001). Comparing the storage time of 3 h outside vs. inside the cool box also revealed a significant difference: using the cool box reduced the mortality to 14%. Overall, the mortality of spiders was lower than that of leafhoppers but the pattern was similar: storage period affected mortality, with lowest values after 1 h of storage and highest values after 3 h ($F_{2,40} = 3.38$, P<0.05; Figure 4B). Mortality after 2 h of storage was not

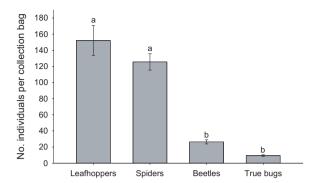


Figure 2 Mean (\pm SE; n = 21 plots) number of individuals per collection bag (i.e., 50 suction samples per bag) of leafhoppers, spiders, beetles, and true bugs. Means capped with different letters are significantly different (Dunn's test: P<0.05).

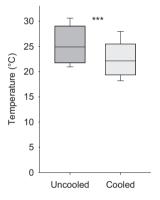


Figure 3 Temperature outside and inside the cool box (based on mean values generated from measurements recorded every minute for 3 h). Box plots show 10th and 90th percentiles (whiskers), 25th and 75th percentiles (top and bottom of the box), and median (line within the box). The asterisks indicate a significant difference between treatments (paired t-test: P<0.001).

significantly different from the two other storage periods. Similar to leafhoppers, the use of the cool box for 3 h significantly reduced the mortality rate of spiders compared to storage for 3 h outside. In beetles and true bugs, the mortality was generally low and did not differ among the storage periods or the cool box (Friedman test, beetles: $\chi^2 = 0.22$, P = 0.90; true bugs: $\chi^2 = 0.62$, P = 0.74, both d.f. = 2; Figure 4C and D).

Discussion

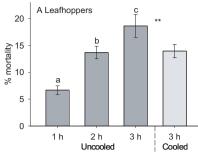
Our study revealed that arthropod mortality after suction sampling was generally low (9% of all sampled individuals). Overall, the survival rate was affected by (1) storage time, (2) storage conditions, and (3) arthropod group. The mortality of beetles and true bugs was low and not influenced by storage time or storage conditions. In contrast, leafhoppers and spiders had a higher mortality; mortality increased with storage time and decreased by the use of a cool box.

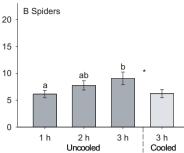
Arthropod mortality in our study may be caused by effects of the suction or the storage. Suction sampling may lead to lethal damage through collision of the individuals with the walls of the suction sampler or with swirling debris. In addition, storing arthropods within collection bags may result in death due to predation, lethal damages through jumping, or suffocation and dehydration of individuals covered by debris inside the collection bags (Stewart, 2002; Ramires et al., 2007). Predation rates and jumping behaviour are strongly dependent on the activity of the individuals (Speight et al., 2008). Hence, both should be higher under the warmer and lighter conditions in storage bags outside the cool box than in those inside the cool box.

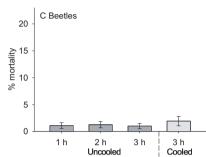
The two arthropod groups with on average larger specimens – beetles and true bugs – were relatively resistant to the effects of suction and storing as the low mortality rates in our study indicate. Beetles are characterised by a heavily chitinized body with hard-shelled forewings (Dettner & Peters, 2003) and thus seem to be well protected against mechanical damage and against most predators in the bags. True bugs suffered higher mortality rates than beetles. Nevertheless, they were also robust and even soft-bodied species such as plant bugs (Miridae) mostly survived the storage.

Leafhoppers are small, hardly chitinized, and actively jumping arthropods (Dettner & Peters, 2003; Speight et al., 2008). These characteristics make them more sensitive to mechanical damage and arthropod predation (Stewart, 2002; Ramires et al., 2007). Additionally, when buried by debris inside the collection bags they may be too weak to escape, risking suffocation and dehydration (cf., Ramires et al., 2007). Spiders are also known to be sensitive to mechanical damage (Parry & Brown, 1959; Wilson, 1970; Anderson & Prestwich, 1975; Ramires et al., 2007; Kropf, 2013) and especially the smaller ones should also suffer from predation by other arthropods in the collection bags. In line with this, we regularly observed spiders and some predatory true bugs preying on other spiders and, more frequently, leafhoppers.

Storage in the cool box reduced the mortality of both leafhoppers and spiders. The lower temperature may have lowered the activity of the individuals and, hence, activity-dependent mortality may have been lower. Despite a significant cooling effect, the conditions inside the cool box were still quite warm (median temperature ca. 22 °C). Hence, we speculate that the darkness inside the box was







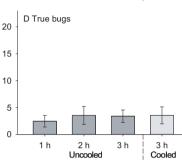


Figure 4 Mean (\pm SE; n = 21 plots) mortality (%) of (A) leafhoppers ($n_{total} = 12 782$), (B) spiders ($n_{total} = 10 551$), (C) beetles ($n_{total} = 2 223$), and (D) true bugs ($n_{total} = 801$) after storage for 1-3 h outside the cool box or 3 h inside the cool box. Means within a panel capped with different letters are significantly different (Holm-Sidak test: P<0.05). The asterisks indicate a difference between cooled and uncooled (3 h storage) samples (A, C, D: non-parametric test for paired samples; B: paired t-test; *0.01<P<0.05, **P<0.01).

also important for a reduced activity and higher survival rates. However, a device with a higher cooling capacity may lead to a lower mortality than the passive cool box we used.

Implications for conservation translocations using a suction sampler

Suction samplers are well suited to sample arthropods (Stewart, 2002) and to translocate plant seeds in restoration projects (Zerbe & Wiegleb, 2009; Zerbe, 2019). According to our results, they are also a valuable tool to sample arthropod assemblages for conservation translocation. A large proportion of transported individuals survived and the mortality of the more sensitive taxa (leafhoppers and spiders) could be reduced by decreasing storage time and by using a cool box. Low loss rates are mandatory in translocation projects for reasons of ethics, economics, and ecological sustainability. The current guidelines for translocations underline that it is important to preserve the source populations and to minimise stress or suffering of translocated individuals (JCCBI, 2010; IUCN/SSC, 2013).

Translocation projects are often expensive and time consuming (Fischer & Lindenmayer, 2000; Carter et al., 2017; Berger-Tal et al., 2019). An advantage of using a suction sampler for translocation is the little material requirements – no heavy machinery is needed for transportation and handling can be done by a single operator.

Assessing the scale of expected benefits of a conservation translocation is part of its planning stage (IUCN/SSC, 2013). The dispersal abilities of arthropods vary greatly among species (Carter et al., 2017). Some are able to move

long distances (Kisimoto & Rosenberg, 1994; Reynolds et al., 2017) or can be transported passively by hay transfer (Kiehl & Wagner, 2006), sheep (Fischer et al., 1996) or vacuum-harvested seeds (Kiehl et al., 2010). For such good dispersers, active translocations are of subordinate importance because they are able to recolonise restored sites on their own (Carter et al., 2017). Furthermore, the capture efficiency of a suction sampler differs between taxa. For example, it is low for soil-dwelling beetles or hidden nocturnal beetles and spiders, whereas vegetation-dwelling species are caught easily (Sanders & Entling, 2011). But if source populations of catchable, rare and dispersal-limited species exist, a suction sampler should work well for conservation translocation (Baur, 2014; Carter et al., 2017) and has the potential to complement the reintroduction of plant species.

Suction samplers can be a valuable tool to translocate species assemblages. In order to reduce mortality in sensitive groups such as leafhoppers and spiders, only the storage process can be influenced. We thus recommend (1) using a cool box and (2) minimising the period until release at the restored site.

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