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Epizoochory via the hooves – the European bison (*Bison bonasus* L.) as a dispersal agent of seeds in an open-forest-mosaic

Epizoochore Samenausbreitung durch die Klauen des Wisents (*Bison bonasus* L.) in einem Wald-Offenland-Mosaik

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Abstract

Large herbivores are significant vectors for the long-distance dispersal of seeds in various habitats, both attached to animals (epizoochory) and via gut passage (endozoochory). The majority of studies on epizoochory have examined dispersal in the fur of domesticated ungulates. Studies on wild ungulates are important to understand dispersal processes in many habitats, but rare due to methodological constraints. We studied epizoochory of seeds by European bison in an open-forest-mosaic (nutrient-poor grassland and heathland, mixed forest) in NW Germany, where bison had been introduced for the purpose of nature conservation. At the study site it was possible to apply a method by which hoof material of free-ranging bison was non-invasively collected. We identified a total of 1082 seeds from 32 plant species in the hoof material. The three most abundant species were *Polygonum aviculare, Agrostis capillaris* and *Betula* spp. Seed species originated from various habitat types of the study area, while the majority of seeds derived from trampled areas. Compared to the non-dispersed plant species of the study area, dispersed plant species had a higher seed longevity index, suggesting that many seeds were picked up from the soil seed bank. Epizoochory ranking indices of dispersed seed species, classifying the importance of epizoochory, revealed that transport in the fur may be of minor importance for many species, i.e. epizoochory by the hooves turned out to be negatively correlated to epizoochory in the fur. We conclude that European bison disperses a considerable number of seed species through trampling. Further research should consider epizoochory via the hooves and include integrative approaches to understand the different dispersal mechanisms by ungulates and their long-term synergetic effect on plant communities.

Keywords: diaspore, external dispersal, large ungulate, secondary seed dispersal, trampling

Erweiterte deutsche Zusammenfassung am Ende des Manuskripts

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1. Introduction

Large herbivores are known to be important vectors for the long-distance dispersal of plants in various habitats, both via epizoochory (dispersal of seeds attached to animals) and endozoochory (dispersal of seeds via gut passage) (BONN & POSCHLOD 1998). Zoochorous dispersal of seeds by ungulates can thus maintain distribution and connectivity of plant species and populations (GERTSEEMA et al. 2002, COUVREUR et al. 2008). Epizoochory is especially understood as an important long-distance dispersal mode (MOUISSIE et al. 2005, MANZANO & MALO 2006), because in comparison to endozoochory it is not limited by physiological factors as gut passage (SORENSON 1986).

The majority of studies on epizoochory have examined dispersal of higher plants by brushing out the fur of domesticated live ungulates in various habitats (FISCHER et al. 1996, COUVREUR et al. 2004, WESSELS-DE WIT & SCHWABE 2010). Another approach is to experimentally test the adhesion and retention potential of seed species to a variety of fur types in standardized tests. The latter was done either with the help of shake tables (RÖMERMANN et al. 2005, TACKENBERG et al. 2006), or by using 'dummies' in the field (FISCHER et al. 1996, GRAAE 2002). Different to domesticated ungulates, the number of studies on epizoochorous dispersal by wild ungulates is low due to the methodological difficulty. Research on dead or immobilized deer (*Capreolus capreolus* L., *Cervus elaphus* L., *Dama dama* L.) and especially wild boar (*Sus scrofa* L.) has proved that these animal species transport various seed species both in their fur and hooves (HEINKEN & RAUDNITSCHKA 2002, SCHMIDT et al. 2004, PICARD & BALTZINGER 2012, DOVRAT et al. 2012). Investigations using free-ranging wild ungulates are to our knowledge missing until now.

Compared to laboratory experiments, studies on free-ranging ungulates give a more realistic picture of dispersal processes in real landscapes: Besides the diaspore (seed or fruit) traits, the quantity of seeds in the vegetation, the seed position and the animal's behaviour (roaming, feeding, habitat use) has to be taken into account (e.g. FISCHER et al. 1996). Hence, the epizoochorous dispersal process is taken as a complex interaction between plant, seed and ungulate characteristics. Moreover, ungulate trampling, defecation and selective foraging contribute to the spatial heterogeneity and floristic composition of landscapes (PASTOR et al. 2006, VAN WIEREN & BAKKER 2008), and, as ROSENTHAL et al. (2012) state, generate various disturbance patterns at different spatial scales resulting in diverse habitat structures. Trampling is a non-random disturbance determined through the attractiveness of patches within the habitat. So, highly trampled areas are more common close to water holes, resting places and attractive plants (JELTSCH et al. 1997). Besides direct effects due to defoliation and soil compaction trampling by ungulates also has less obvious effects in combining seed dispersal and microsite creation which can increase the probability of seeds to successfully germinate and establish (BAKKER et al. 2009, ROTUNDO & AGUIR 2004, ROSENTHAL et al. 2012).

In recent years, free-ranging ungulates have been introduced to semi-open landscapes within new management schemes, and also as means of conservation of the rare herbivores, e.g. European bison, *Bison bonasus* L. Free-ranging ungulates reduce encroachment of shrubs and trees, and hence maintain local biodiversity (BUNZEL-DRÜKE et al. 2009, STROH et al. 2012). During the Holocene European bisons were distributed throughout Central Europe and in the Western part of Russia. Since the 11th century extinction of these populations occurred, and in 1919 the last free roaming bisons were killed. In 1952 reintroductions into the reserve of the Białowieża national park and in other East European countries took place. Today, the only free roaming (wild) populations are in Poland, White Russia, Russia, Latvia, Ukraine, and Slovakia (KRASINSKA & KRASINKSI 2008). In Germany, bison herds are kept under semi-wild conditions in enclosures, except some wilderness projects in the Döberitzer Heide (Brandenburg) and the Rothaargebirge (North Rhine-Westphalia) where bison populations are controlled only by natural conditions, i.e. no supplemental feeding and free-ranging behaviour.

The European bison is one of the most effective wild ungulates with respect to endozoochorous dispersal (JAROSZEWICZ et al. 2008, 2009, JAROSZEWICZ 2013). However, there are no published studies about epizoochory by European bisons, neither for seeds transported in the hooves, nor in the fur, while CONSTIBLE et al. (2005) investigated the seed content in fur residuals of American bison (*Bison bison* L.) collected at resting places. For our study, we designed a method with which seeds contained in the hooves of moving European bison could be non-invasively collected. Aim of this study was to find out: 1) Which seed species are epizoochorously transported via the hooves of the European bison? 2) Which are the morphological and ecological traits of dispersed plant species and their dispersal units (i.e. diaspores) that are transported in the hooves by European bison?

2. Material and Methods

2.1 Study area

The study was carried out in an enclosure of 46 ha with five individuals of the European bison on a former military training area in the district of Cuxhaven, Lower Saxony, NW Germany (53° 49´ N, 8° 38´ E). Its geological substratum (acidic sand) derived from the Saale glacial during the Pleistocene. Climate is atlantic with an annual mean temperature of 8.9 °C and precipitation during the vegetation period of 417 mm (CORDES et al. 2006). The landscape mosaic contains open areas (15 ha) with plants typical for nutrient-poor habitats, e.g. *Agrostis capillaris, Betula pendula, Calluna vulgaris* and *Empetrum nigrum*. Due to its geomorphological structure, past military disturbance (including fire) and the presence of bisons, diverse habitat types occur here, among others nutrient-poor grassland, coastal heath, wet heath, trampled and ruderalised vegetation. Forest plantations with Norway spruce (*Picea abies)*, Japanese larch (*Larix kaempferi)* and Black cherry *(Prunus serotina)* cover 31 ha. A survey of all vascular plants was done in the enclosure before and during the sampling period. To produce a mean cover of the 95 plant species in the enclosure, we recorded plant abundance in the open areas and the forest patches as follows: very rare, 0–1% cover; rare, >1–5%; medium, >5–25%; or dominant, >25%.

2.2 Seed sampling

From May to August 2011, seeds were sampled from the hooves of a herd of five free-ranging European bison (4 cows, 1 bull) for 6 days every 3 weeks. The bisons were daily allured into a separated area, into which they entered through a gate; although the herd is semi-wild this was regularly practiced by the farmer, so that medical treatments still could be indirectly applied to the animals. Into this gate area, a row of four perforated hard-rubber mats (length 40 cm, width 60 cm, height 1.5 cm) were arranged on a green synthetic cover (length 1.65 m, width 0.68 m; Fig. 1). Before sampling, the synthetic ground cover and the mats were thoroughly washed, dried and brushed. Immediately after the bison herd had left, all accumulated material on the plastic ground and on the mats was collected into a plastic bag using a hand broom, plastic scraper and rubber glove. Moreover, the broom brush and the rubber gloves were searched for seeds.

The sampled material was spread out on a sieve (mesh size 1 mm), and stones, stems, leaves or roots were removed. The residual material was evenly distributed and aggregates were carefully crushed; it was left to dry at room temperature (19 °C) for 24 h to prevent the development of mold. In the laboratory the material was analysed with a dissecting microscope. Diaspores (in the following

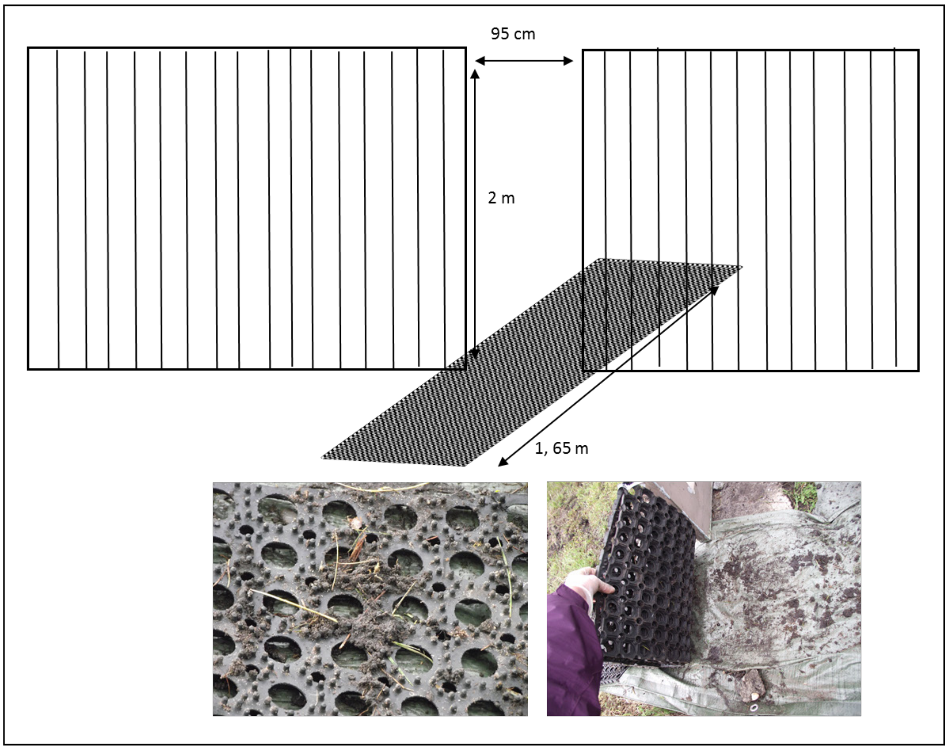

Fig. 1. Method for sampling material transported on hooves of free-moving European bison. The bison moved through a gate in a fence over a hard rubber path, on which material released from the hooves would accumulate to be later collected and analysed (Photo: K.A. Schulze).

Abb. 1. Methode zum Aufsammeln von Material aus Klauen von freilebenden Wisenten. Die Herde passierte einen mit perforierten Kunststoffläufern ausgelegten Durchgang eines Gatters, welcher mit einer Plane unterlegt war. Transportiertes Material aus den Klauen akkumulierte sich dort und wurde anschließend eingesammelt und im Labor untersucht (Foto: K.A. Schulze).

referred to as 'seeds') were sorted out and kept in small snap-cap glass vials. They were stored in a dark box at room temperature, and the seeds were only exposed to light during analysis. For seed identification length, width and weight (Sartorius CP324S) of each seed were measured. The plant species were identified using KIFFMANN (1960), BROUWER & STÄHLIN (1975), HANF (1990) and CAPPERS et al. (2006, also online version on www.plantatlas.eu/za.php, access: May–September 2011), and the Scottish Crop Research key (http://asis.scri.ac.uk/, access: May–September 2011). The nomenclature of plant species follows the taxonomic reference list for the German flora (German SL) by JANSEN & DENGLER (2008).

2.3 Data analysis

We measured and collected ecological and morphological properties of all dispersed and non-dispersed seed species in the study area mainly using the following databases: LEDA-traitbase (KLEYER et al. 2008, www.leda-traitbase.org, access May 2013: period of seed shedding, mean seed number per shoot, mean release height, longevity index), the Dispersal Diaspore Database (HINTZE et al. 2013, www.seed-dispersal.info, access February 2013: mean diaspore mass (in the following termed 'seed mass'), epizoochory ranking index: an index which classifies the importance of epizoochory in the fur for certain species), and Biolflor (KLOTZ et al. 2002: trampling index) in order to find out about differences between traits of dispersed and non-dispersed seed species (cf. STROH et al. 2012). We also classified species according to their preferred habitat type in the study area (see Appendix S1). With respect to mean seed mass we compared and checked-back our measured values to those of the Dispersal Diaspore Database.

In order to find out about similarities or significant differences between dispersed and non-dispersed seed species we compared certain plant and seed traits of the two groups. Due to non-normal distribution of the data (Kolmogorov Smirnov test, $p \leq 0.05$), the non-parametric Mann Whitney U-test was used for the analysis in SPSS Statistics 21.

3. Results

The material released from the bison hooves was a sandy-soil mixture with stones, leaves, stems, roots and seeds. We gained a total of 1,376 g dry weight of hoof material throughout the sampling period. In May, only the oldest bison cow walked through the narrow entrance. Therefore, little material (19 g) was collected in that month (June: 244 g, July: 667 g, August: 446 g).

The seeds contained in the hoof material occurred in various states. Some seed species, e.g. *Erica tetralix*, *Juncus tenuis* and *Stellaria media,* were represented both by seeds and capsules. Among the grasses, seeds were either released as fruits with glumae, as florets, or as inflorescenses (multiple spikelets), e.g. *Holcus lanatus*. Especially, seeds from *Agrostis capillaris* were found in different constitutions, i.e. as whole inflorescenses with fertile and sterile spikelets or solely as florets.

Among the four sampling dates a total number of 1,082 seeds of 32 plant species were collected. Seed number and species diversity was the highest in July ($n_{species}$ = 23, n_{seeds} = 425); August (19, 349); June (12, 244) and May (8, 64). Over the whole sampling period the most abundant seed species were *Polygonum aviculare* (56% of the total number of seeds), *Agrostis capillaris* (17%), *Betula pendula* and *B. pubescens* (8%) and *Juncus tenuis* (4%) (Fig. 2, Appendix S1). Among all 32 plant species recorded, four woody and one herbaceous species were found in all months; these were *Betula* spp., *Larix kaempferi*, *Picea abies* and *Polygonum aviculare,* and >60% of the dispersed species were from July to August.

Altogether 34% (32 of the 95) of the seed plant species recorded in the study area were epizoochorically dispersed by bisons (Appendix S1). Six dispersed species (*Crataegus monogyna*, *E. tetralix*, *Larix kaempferi*, *Molinia caerulea*, *Persicaria minor* and *Picea abies*) were most likely not fruiting during the study period, and seeds of *Betula* spp. were already found months before seed shedding. On the other hand only four of the 63 non-dispersed species were not fruiting during the study period. Seeds of plant species occurring very rare or rare in the study area were also frequently recorded (Fig. 2). The seeds encountered in the hoof material derived from plant species of nearly all habitat types within the enclosure, but mainly seed species common for woodlands, trampled sites and nutrient-poor grassland areas were recorded, and the minority of species derived from wet areas (Fig. 3). While plant species from trampled areas, trees and shrubs were overrepresented, species from wet and coastal heathlands and forest understory were poorly represented.

Comparing the mean release heights of all herbaceous plant species at the study site we found that a large amount of seeds (63%) derived from the three smallest species, i.e. *Juncus tenuis*, *Poa annua* and *Polygonum aviculare*. These species were also characterized by the highest trampling resistances (Appendix S1), while also many tall plants with low trampling resistance, including trees, were transported.

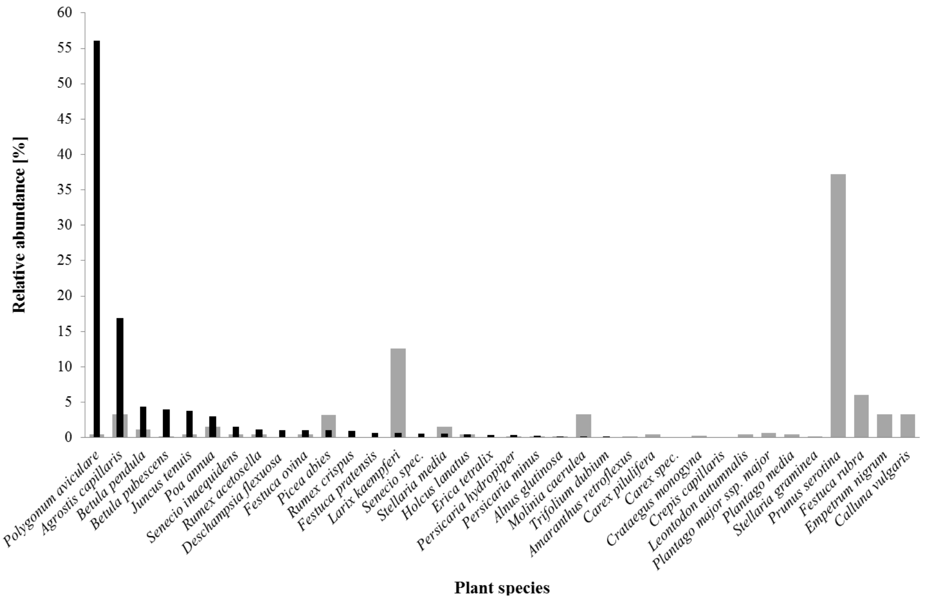

Fig. 2. Relative abundance of seed species in the hooves (black, $n_{species}$ = 32) of the European bison compared with their relative abundance (grey) in the vegetation of the study area ($n_{species}$ = 95). The four most abundant plant species which were non-dispersed are also shown.

Abb. 2. Relative Abundanz der ausgebreiteten Samenarten in den Klauen des Wisents (schwarz, n_{Arten} = 32) verglichen mit ihrer relativen Abundanz (grau) in der Vegetation (n_{Arten} = 95). Zusätzlich gezeigt werden die vier häufigsten Pflanzenarten im Gebiet, deren Samen nicht in den Wisentklauen gefunden wurden.

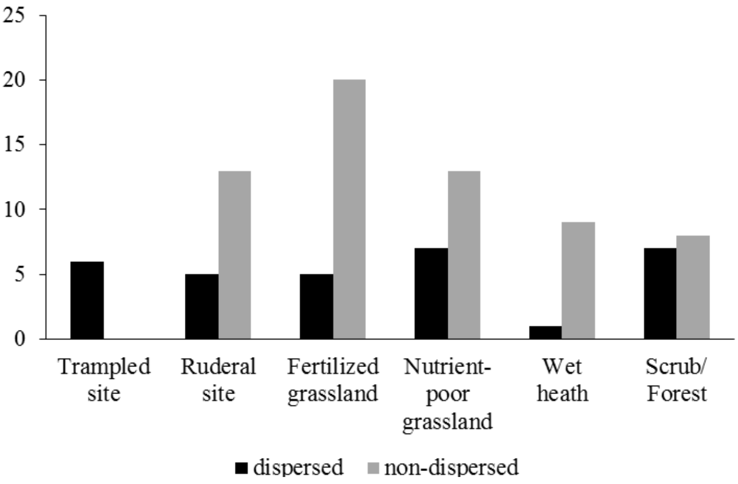

Fig. 3. Total number of dispersed (black, $n_{species}$ = 32) and non-dispersed plant species (grey, $n_{species}$ = 63) compared with their preferred habitat type.

Abb. 3. Absolute Anzahl ausgebreiteter (schwarz, n_{Arten} = 32) und nicht-ausgebreiteter Pflanzenarten (grau, n_{Arten} = 63) in Bezug auf ihren bevorzugten Habitattyp (von links nach rechts: Trittrasen, Ruderalflur, Wirtschaftsgrünland, Magerrasen/Küstenheide, Feuchtheide, Gebüsch und Wald).

Table 1. Results of analysis of traits of dispersed and non-dispersed seed species, using the Mann Whitney U-test. The data source is shown for each trait: BiolFlor = KLOTZ et al. (2002); D3 = Dispersal Diaspore Database; flora = Floraweb, ld = LEDA-traitbase; pers = personal measurements.

Tabelle 1. Ergebnisse des statistischen Vergleichs (Mann-Whitney-U-Test) bestimmter Pflanzen- und Sameneigenschaften zwischen ausgebreiteten und nicht-ausgebreiteten Arten. Verwendete Datenbanken und ihre Abkürzungen s. o.

Trait	N species dispersed/ non-dispersed	Median dispersed species	Median non-dispersed species	*U*	*p*
Mean seed mass [mg] ^{pers,D3}	29/60	0.39	0.54	846	0.834
Seeds per shoot^{ld}	24/52	1060	568	617	0.993
Seed longevity index^{ld}	25/60	0.48	0.25	483	0.010*
Mean release height^{flora,ld}	29/61	0.43	0.48	837	0.685
Trampling index^{BiolFlor}	22/39	4.50	5.00	377	0.430
Epizochory ranking index^{D3}	22/53	0.30	0.63	418	0.056(*)

We did not find significant differences in seed mass, seed number per shoot, seed release height and trampling index between dispersed and non-dispersed seed species. However, the seed longevity in the soil of dispersed species was significantly higher than that of seeds from non-dispersed plant species (Table 1). A large amount of dispersed seeds has the potential for long-term persistence in the soil seed bank, i.e. *Juncus tenuis, Poa annua* and *Rumex acetosella.* With respect to the relevance of epizoochory in the fur as a dispersal mode, the ranking indices revealed that dispersal ranks from seed species detected in the hoof material were (marginally significantly) lower than from the non-dispersed species from the vegetation in the enclosure (Table 1). Accordingly, for <10% of the seed species dispersed by bison in our study, epizoochory has been classified as an important dispersal mode, whereas the majority of non-dispersed plant species were represented by high indices.

4. Discussion

4.1 Methodological aspects

Studying epizoochorous dispersal by wild and live ungulates represents a methodological challenge because it is not possible to get near to these animals, and any direct interference may lead to irritation and changes in the behaviour of the animals. Thus, until now research on epizoochory by wild ungulates is mainly restricted to dead or immobilized animals (HEINKEN & RAUDNITSCHKA 2002, SCHMIDT et al. 2004, PICARD & BALTZINGER 2012, DOVRAT et al. 2012), or epizoochory is inferred indirectly via fur residuals (CONSTIBLE et al. 2005) or the seed bank at resting or rubbing places (HEINKEN et al. 2006).

Our method to non-invasively gain and analyse the seed content of hoof material from free-ranging wild European bison worked very well. Previous studies (FISCHER et al. 1995, STENDER et al. 1997, HEINKEN & RAUDNITSCHKA 2002, PAULIUK et al. 2011, PICARD & BALTZINGER 2012) have shown that dispersal via the hooves accounts for a considerable proportion of epizoochory by ungulates. Our results indicate which seed species are frequently dispersed by bison and which temporal changes occur. Quantity of seed and species richness were clearly related to the weather conditions (K.A. Schulze, unpubl. data); at times of high precipitation prior or during the sampling week (mainly in summer) more material was transported (see also CLIFFORD 1956 in BONN & POSCHLOD 1998).

Some methodological problems were identified and need to be optimized in future studies. We cannot exclude that some seeds arrived on the rubber mats by wind, but due to the short exposition time this should be a minor fraction. The high abundance of *Polygonum aviculare* seeds can be explained by its local distribution. Although it only occurred rarely within the enclosure, it mainly grew on the trampled path between gate and trough, and consequently the European bison repetitively passed over these plants. Similarly, PAULIUK et al. (2011) found certain moss species with high abundance in sheep fur that were dominant at resting places of the animals. So, our study proved at least short-distance dispersal of *Polygonum aviculare* via bison hooves.

4.2 Epizoochory by European bison

To our knowledge this study is the first showing that the European bison functions as an epizoochorous dispersal agent in its habitat. It has already been proven that this ungulate species is one of the most important endozoochorous long-distance seed dispersers in European natural temperate forest landscapes (JAROSZEWICZ et al. 2009, JAROSZEWICZ 2013). Our study suggests that seed dispersal by European bison is even more significant than highlighted in these investigations because a certain amount of seeds is transported by the hooves, and internally and externally transported plant species complement each other: Only six of the 32 dispersed plant species were among the 30 species dispersed endozoochorously by the five individuals of European bison during our study period (K.A. Schulze, unpubl. data, see Appendix S1; cf. COUVREUR et al. 2005 for donkeys).

Dispersed seeds originated from various habitats, mainly from the open parts of the study area, but also from the forest patches. This is in accordance with the endozoochory results of JAROSZEWICZ et al. (2009) and JAROSZEWICZ (2013), and also those from other wild ungulates (HEINKEN et al. 2001, SCHMIDT et al. 2004, VON OHEIMB et al. 2005, JAROSZEWICZ et al. 2013). However, the number of internally dispersed seeds and plant species can be much higher: JAROSZEWICZ et al. (2009, 2013) detected a total number of 4,177 seedlings from 109 plant species emerging from dung samples of wild and semi-wild European bison in a study period of 12 months. Also for other wild ungulates high numbers have been reported, especially for red deer (*Cervus elaphus*) (VON OHEIMB et al. 2005, JAROSZEWICZ et al. 2013).

More seed species were dispersed epizoochorously in the hooves by the five studied individuals of European bison compared to other wild ungulates. HEINKEN & RAUDNITSCHKA (2002) discovered a total of 15 and eight plant species in hoof material of shot roe deer and wild boar in a forest habitat. Also, PICARD & BALTZINGER (2012) detected seeds of only ten different plant species in the hooves of three shot wild ungulate species (*C. elaphus, C. capreolus, Sus scrofa*) in an open-forest-mosaic. Our study integrated over a longer period of time, but the higher number of species may be also caused by the larger hooves of European bison. Grasses (e.g. *Agrostis capillaris*) and *Betula* spp. were also recorded as highly abundant in the hooves of the wild ungulates in these studies.

As expected, a recognizable amount of dispersed plant species derived from disturbed, trampled areas. Their high tolerance to trampling is partially due to their small size which should further favour attachment of seeds to hooves. Assuming tolerance for trampling is also expressed by the ability to spread and colonize such habitats due to successful dispersal by the hooves it might be a good strategy for these species to reach new disturbed areas.

Contrarily, no plant species from the forest understory and only one species from the wet heath were found in the soil material collected from bison hooves. Habitat use of the bison herd may be the main reason for this pattern. For example the understory vegetation was not really used for feeding since food plant availability during the vegetation period was higher in the open area of the enclosure (K.A. Schulze, unpubl. data). The forest area was mainly used for resting, and bison roamed on more or less fixed trails. As bison prefer to wallow on the forest ground at their resting places and also to rub their body against tree trunks, forest plant species might thus rather be dispersed via the matted-dense fur of bison. Because of our relatively short study period also some plant species with late seed maturation (which occur especially in the heathlands and wet areas, see Appendix S1) may have been under-represented in the samples, but altogether the number of undispersed species which set seeds later than August was low.

Interestingly, not only low-growing species were dispersed, i.e. the ones which can directly come into contact with the hooves. This is because plant species from grazed areas may attach to the hooves after they have been trampled down, as indicated by the fact that seed release height and trampling index are no predictors for seed species selection. Our results also suggest an additional dispersal mechanism: Seeds of many herbaceous and woody plant species contained in the soil are secondarily dispersed through the hooves. The winged, light fruits of *Betula* spp. for example are present on herbaceous vegetation and the soil surface in large amounts (HEINKEN & RAUDNITSCHKA 2002). Secondly, the higher seed longevity index of dispersed plant species compared to non-dispersed species and that a considerable amount of species was dispersed out of their seed shedding period indicate that many species included in the soil seed bank were secondarily dispersed, enhancing the dispersal distance of plant species (SORENSON 1986).

We suppose that trampling actually has a combined effect on the soil seed bank and plant community dynamics: First, it is an effective dispersal mechanism for many plant species because establishment of seeds might be enhanced when within a "package of mud" (BONN & POSCHLOD 1998). In more loamy soils than in our study area even more material may stick to the hooves and thus favour epizoochory. Secondly, seeds embedded in deeper soil layers might benefit from the loosened soil in times of high precipitation. As a consequence post-dispersal ungulate trampling may enhance plant establishment (FAUST et al. 2011, ROSENTHAL et al. 2012). Similarly, JAROSZEWICZ (2013) encountered that through endozoochorous dispersal by bison, dispersed seeds do not only establish on, but also move into the soil seed bank from dung pats.

4.3 Three complementary zoochorous dispersal modes?

It has been assumed that dispersal via ungulate hooves is coincidental and 'by-chance-dispersal', although a number of species have frequently been recorded in epizoochory studies on ungulates (BONN & POSCHLOD 1998). This is because of a large heterogeneity in plant growth height and seed morphology of species dispersed in the hooves of sheep (FISCHER et al. 1995), cattle (STENDER et al. 1997), roe deer and wild boar (HEINKEN & RAUDNITSCHKA 2002). Still, we suppose that external transport by the hooves rather can be seen as a complementary dispersal mode not only to endozoochory (see above), but also to epizoochory in the fur: Analyzing epizoochory ranking indices of dispersed seed species revealed that traits which favour dispersal in the fur of animals (i.e. hooks and other appendages) are apparently of minor importance for dispersal by hooves. Similarly, plant species with a considerable height of seed presentation, i.e. in the height of the body of the disperser (FISCHER et al.

1996), will also not be preferred (cf. STENDER et al. 1997). Instead, our study indicates that multiple factors like seed longevity and habitat type facilitate seed dispersal by ungulate hooves.

4.4 Implications for future research

Our study illustrates that dispersal via hooves should be incorporated in future research on zoochory because many plant species are transported through this dispersal mode. To overcome the methodological problem of over-representation of seeds from trampled sites near the gate (see Section 4.1) we propose to put perforated hard-rubber on synthetic cover as used in our study on frequently used animal trails, preferably combined with controls beyond the paths when investigating epizoochory via hooves.

As for endozoochory and epizoochory in fur, the quality and quantity of seed species transported via the hooves will be strongly influenced by the behaviour of the ungulates. Further research would be necessary to confirm whether (a) the variety of dispersed seed species is dependent on the ungulate species, and (b) epizoochory in hooves is complementary to both endozoochory and epizoochory in the fur. Such integrated approaches should include comparisons within the same habitat (a) of epizoochory by the hooves for more ungulate species (for endozoochory in the Białowieża forest see JAROSZEWICZ et al. 2013), and (b) of all three dispersal mechanisms on one (or more) ungulate species. Modelling approaches which include a variety of abiotic and biotic factors as conducted by COUVREUR et al. (2008) and D´HONDT et al. (2012) for epi- and endozoochorous dispersal by domestic ungulates, as well as telemetry data could help to understand how the complex ungulate-plant-interactions and the complementary dispersal modes affect the plant communities at the landscape-scale in the long run. Also in the scope of nature restoration, results about how different ungulate species may contribute to the maintenance of plant species and their spatial connectivity through (epi)-zoochorous dispersal and behavioral habits would be important.

Erweiterte deutsche Zusammenfassung

Einleitung – Die Ausbreitung von Diasporen ist ein essentieller ökologischer Prozess zur Erhaltung der Phytodiversität in einer Landschaft. Es ist bekannt, dass Großherbivoren in ihrem Habitat als passive (epizoochor, externe Samenanheftung) und aktive (endozoochor, Nahrungsaufnahme) Ausbreitungsvektoren über große Distanzen wirken. Die Mehrzahl der relevanten Untersuchungen (u. a. FISCHER et al. 1996, COUVREUR et al. 2004, COSYNS et al. 2005) wurde mit domestizierten Huftieren durchgeführt. Dagegen ist weniger bekannt über die Rolle wildlebender Huftiere als Ausbreitungsvektoren, weil derartige Studien überwiegend an geschossenen oder anderweitig immobilisierten Huftieren durchgeführt worden sind.

Material und Methoden – Wir haben in einem 46 ha großen Wald-Offenland-Mosaik (Magerrasen, Heide, Mischwald) eines küstennahen Naturschutzgebietes in Nordwestdeutschland (ehemaliger Truppenübungsplatz Altenwalde, Landkreis Cuxhaven) die Rolle des Wisents als epizoochoren Ausbreitungsvektor untersucht. Die Wisente werden zur Offenhaltung von Zwergstrauch- und Gras-geprägten Lebensräumen eingesetzt. Aufgrund der spezifischen Gegebenheiten im Gehege war es uns möglich eine Methode zu entwickeln, durch welche wir auf indirekte Weise Material sammeln konnten, das durch die Wisent-Klauen transportiert wird (s. Abb. 1). Die gefundenen Diasporen (Samen und Früchte) wurden bestimmt. Die ausgebreiteten Arten wurden, auch in Bezug auf verschiedene Pflanzenmerkmale, mit den nicht ausgebreiteten Arten im Gehege verglichen.

Ergebnisse – Insgesamt identifizierten wir 1.082 Samen von 32 Pflanzenarten (79 Samen pro 100 g Trockenmasse) in den Klauen-Proben; das waren 34 % der insgesamt im Gehege gefundenen Pflanzenarten. Am stärksten vertreten waren *Polygonum aviculare, Agrostis capillaris* und *Betula* spp. (Abb. 2, Anhang S1). Die Samen- und Pflanzeneigenschaften der ausgebreiteten Arten waren heterogen. Die ausgebreiten Samen gehörten Pflanzenarten diverser Biotoptypen an, jedoch ist die Mehrzahl der Samen den Trittgesellschaften zuzuordnen (Abb. 3). Im Vergleich zwischen ausgebreiteten und nicht-ausgebreiten Samenarten stellten wir einen signifikanten Unterschied bezüglich ihrer Überlebensdauer in der Samenbank fest (Tab. 1): Ausgebreitete Pflanzenarten haben ein höheres Potential zur Überdauerung, sind also eher als langlebig einzustufen. Darüber hinaus fanden wir einen niedrigeren „Epizoochory Ranking Index" (Skalierung der Bedeutung von Epizoochorie einer Pflanzenart für eine Ausbreitung im Fell) von ausgebreiteten im Vergleich zu nicht-ausgebreiteten Arten (Tab. 1).

Diskussion und Schlussfolgerung – Unsere Methode, auf Gummimatten durch die Klauen transportiertes Material mit Samen zu gewinnen, war geeignet, um die Ausbreitungsmechanismen von Pflanzen auf diesem Wege zu demonstrieren und wird für weitere Studien zur Ausbreitung durch wildlebende Huftiere empfohlen. Der Wisent stellt nicht nur einen hocheffizienten endozoochoren Ausbreitungsvektor dar (JAROSZEWICZ et al. 2009, JAROSZEWICZ 2013), sondern breitet darüber hinaus mit Hilfe seiner Klauen eine große Anzahl Samen verschiedener Pflanzenarten aus. Dies scheint insbesondere für Arten von Tritthabitaten ein wichtiger Modus zu sein. Der Befund, dass ausgebreitete Arten nicht unbedingt niedrigwüchsig sind, häufig eine langlebige Samenbank haben und oft vor ihrer Fruchtzeit ausgebreitet wurden, lässt darauf schließen, dass eine Vielzahl von Samen von Pflanzenarten unterschiedlichster Habitate durch Viehtritt aus der Samenbank herausgelöst wird (sekundäre Diasporenausbreitung). Die geringe Bedeutung der Epizoochorie vieler ausgebreiteter Arten nach dem ‚Epizoochory Ranking Index' kann als ein Hinweis dafür interpretiert werden, dass es sich bei der Ausbreitung durch Klauen um einen nicht nur zur Endozoochorie, sondern auch zur Epizoochorie im Fell ergänzenden Ausbreitungsmechanismus handeln könnte. Für zukünftige Studien über die Rolle von wildlebenden Huftieren als Vektoren sollte der Transport von Samen in den Hufen berücksichtigt werden. Dabei ist ein integrierender Untersuchungsansatz empfehlenswert, um sowohl die Komplementarität der drei Ausbreitungsmodi wie auch ihre Langzeit-Auswirkung auf die Pflanzengesellschaften zu untersuchen.

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Supplements and Appendices

Additional supporting information may be found in the online version of this article.

Zusätzliche unterstützende Information ist in der Online-Version dieses Artikels zu finden.

Appendix S1. Summary of the results of seed species which were recorded in the hooves of European bison, and of the non-dispersed species in the enclosure.

Anhang S1. Zusammenfassung der Ergebnisse der Samenarten, die in den Klauen-Proben festgestellt wurden, und der nicht ausgebreiteten Arten im Gehege.

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