



## Density effects of two hemiparasitic *Melampyrum* species on grassland plant diversity

### Dichteeffekte von zwei hemiparasitischen *Melampyrum*-Arten auf die Pflanzenartenvielfalt im Grasland

Nico Heer<sup>1</sup>, Fabian Klimmek<sup>2</sup>, Yasemin Kurtogullari<sup>1</sup> , Daniel Prati<sup>3</sup>,  
Nora Simone Rieder<sup>4</sup> & Steffen Boch<sup>5,\*</sup> 

<sup>1</sup>Hintermann & Weber, Aarberggasse 61, 3011 Bern, Switzerland;

<sup>2</sup>Walter Zoo AG Gossau, Neuchlen 200, 9200 Gossau, Switzerland;

<sup>3</sup>Institute of Plant Sciences, University of Bern, Altenbergrain 21, 3013 Bern, Switzerland;

<sup>4</sup>IMPULS AG, Seestrasse 2, 3600 Thun, Switzerland;

<sup>5</sup>Swiss Federal Institute for Forest, Snow and Landscape Research WSL, Zürcherstrasse 111,  
8903 Birmensdorf, Switzerland

\*Corresponding author, e-mail: steffen.boch@wsl.ch

#### Abstract

Hemiparasitic plants can reduce interspecific competition by parasitizing competitive species or by reducing the density of dominant plants. However, effects of hemiparasites have been suggested to be density dependent and to follow an optimum curve, with the highest diversity values occurring at intermediate hemiparasite densities. As a proof of principle, we tested whether this holds true for the two hemiparasitic species *Melampyrum arvense* and *M. nemorosum*. We conducted vegetation surveys in thermophilous fringe communities on the island of Saaremaa (Estonia) and tested how the local species richness of all vascular plants, and forbs and graminoids separately, vary along density gradients of the two hemiparasitic species. In total, we investigated four spatially separated populations of *M. arvense*, and six separated populations of *M. nemorosum*. Within each population, we placed nine subplots along a *Melampyrum* density gradient. Using random slope, mixed-effects models, we found significant hump-shaped relationships of total vascular plant species richness with relative cover of *M. arvense* and *M. nemorosum*, with the highest richness values occurring at 13% and 40% cover, respectively. Starting at relatively low densities (> 29.7%) of *M. arvense* and relatively high densities (> 81.8%) of *M. nemorosum*, species richness was even lower than in plots without these hemiparasites. In contrast to results for total species richness, we found no significant effect of hemiparasite density when looking at graminoid and forb species separately. Our results help fill an important knowledge gap and underline the generality of density-dependent hemiparasite effects on local species richness. Our findings also clearly support the use of the density-gradient approach when the aim is to explore relationships between hemiparasites and species richness.

**Keywords:** alvar grassland, hemiparasite, *Melampyrum arvense*, *Melampyrum nemorosum*, Saaremaa, species richness

**Erweiterte deutsche Zusammenfassung am Ende des Artikels**

## 1. Introduction

Hemiparasitic plant species perform photosynthesis but form haustoria on the roots of other species and thereby extract water, nutrients, assimilates and secondary compounds from their hosts (ADLER 2000, CAMERON & SEEL 2007, TĚŠITEL et al. 2010, WESTWOOD et al. 2010). The genus *Melampyrum* is a member of the large parasitic *Orobanchaceae* family and comprises annual species that probably parasitize rather generally on various host species (MATTHIES 1996, KÄSERMANN 1999, HOLÁ et al. 2017, MATTHIES 2017). Like other hemiparasitic genera, such as *Rhinanthus*, *Melampyrum* is, however, considered to parasitize species with diffuse and wide-spreading root systems, such as grasses, more frequently than species with taproots (MATTHIES 1996, KÄSERMANN 1999, MUDRÁK et al. 2016).

Hemiparasitic plants can reduce interspecific competition by parasitizing competitive species or by reducing the density of dominant plants. In this way, they can facilitate coexistence and promote the stability of natural communities (PHOENIX & PRESS 2005). Annual hemiparasitic species can further promote the establishment of seedlings of other species by creating gaps after their senescence (JOSHI et al. 2000).

However, the effects of hemiparasitic species on local community diversity are still debated. The fact that positive, negative and neutral effects of hemiparasites on local diversity have all been found (GIBSON & WATKINSON 1992, AMELOOT et al. 2005, FIBICH et al. 2016) can be attributed to the most commonly used methodological approach, i.e. comparing sites with and without hemiparasites ('all or nothing' approach; e.g. GIBSON & WATKINSON 1992, FIBICH et al. 2016). In contrast to the intuitive expectation of a purely linear negative relationship between a dominant plant species and the species richness of a plant community, it has been shown that hemiparasite effects are most likely density dependent and follow an optimum curve with the highest diversity values occurring at intermediate hemiparasite densities (e.g. shown for *Rhinanthus alectorolophus* (Scop.) Pollich; HEER et al. 2018). This suggests that for studying hemiparasite effects on local species richness the 'all or nothing' approach can be misleading, as it does not cover hemiparasite density gradients. However, whether these relationships hold also true for other hemiparasites than *Rhinanthus* spp., such as *Melampyrum* spp., has remained largely unclear.

To help fill this knowledge gap and as a proof of principle regarding whether the findings of density-dependent effects of *Rhinanthus* on local community richness can be translated to other hemiparasites, we conducted vegetation surveys in ten sites distributed within two regions on the island of Saaremaa (Estonia). In particular, we tested how the local species richness of (1) all vascular plants and (2) forbs and graminoids separately vary along density gradients of the two hemiparasitic species *Melampyrum arvense* and *M. nemorosum*.

## 2. Methods

### 2.1 Study system

Data were gathered as part of a student project during a plant ecological field course on the Baltic island of Saaremaa (57° 50' to 58° 40' N, 21° 45' to 23° 20' E) in July 2015. The bedrock of this island is formed by Silurian limestone, which is often covered by Weichselian moraines or Holocene marine sediments. The mean annual temperature on Saaremaa is about 6.0 °C, and the mean annual precipitation varies regionally between 540 mm and 675 mm (DENGLER & BOCH 2007). The island harbors a diverse set of vegetation types and is known for its unique flora. In particular, the alvar dry grasslands

growing on very shallow and humus-rich silt or clay soils over limestone bedrock have long attracted the interest of botanists (VILBERG 1927, ZOBEL & KONT 1992, PÄRTEL et al. 1999, BOCH 2005, BOCH & DENGLER 2006, DENGLER et al. 2006).

## 2.2 Study species

We chose two *Melampyrum* species that are common on Saaremaa and locally form dense populations: *M. arvense* L. is a generalist species, parasitizing on cereals and other grass species, but is also known to grow on many forb species (MATTHIES 1996, 2017). Its distribution ranges from Western Europe to the Ural Mountains and Turkey (EURO+MED 2006–2019). In Estonia the distribution center of *M. arvense* is on Saaremaa (KUUSK et al. 1996). Historically, the species was widely distributed amongst arable fields, but today it is restricted to various dry grassland communities, thermophilous fringes and semi-ruderal grasslands. The species is listed as Near Threatened in the Red Data Book of Estonia (NT; Estonian eBiodiversity database, <http://vana.elurikkus.ut.ee/prmt.php?lang=eng>, accessed 2021-03-03).

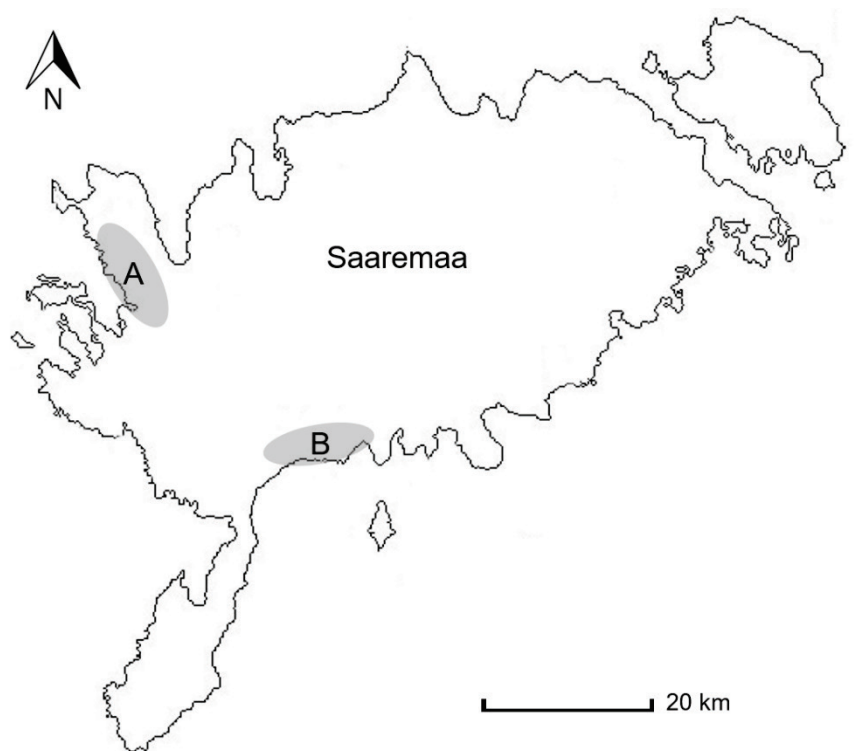
*Melampyrum nemorosum* L. is probably also a generalist species, parasitizing on grass species but also on various forb and woody species (KÄSERMANN 1999, HOLÁ et al. 2017). Its distribution also ranges from Western Europe to the Ural Mountains in the North (EURO+MED 2006–2019). In Estonia, the species is widely distributed (KUUSK et al. 1996; LC; Estonian eBiodiversity database, <http://elurikkus.ut.ee/>, accessed 2021-03-03).

## 2.3 Site and plot selection and vegetation data

On the island of Saaremaa, we selected two regions in which either *M. arvense* or *M. nemorosum* occurred frequently (Fig. 1). In the first region we detected four spatially separated populations of *M. arvense* in thermophilous fringes along agricultural fields that were likely mown, and in the other region we were able to find six separated populations of *M. nemorosum* in thermophilous fringe communities within an open *Quercus robur* L. forest area which was either unmanaged or grazed at very low intensity levels. Within each population, we placed nine circular 314 cm<sup>2</sup> (20 cm diameter) plots along a *Melampyrum* density gradient within a visually homogeneous area. We first visually subdivided each grassland into zones with a low, medium or high density of the particular *Melampyrum* species and then randomly placed three plots in each zone to maximize the range from a minimum (mostly equivalent to absence) to a maximum density within each meadow (up to 70% *M. arvense* and 100% *M. nemorosum* cover). We made sure that the plots were distributed across the whole population and that two plots of the same density class were not situated in the same *Melampyrum* patch. We chose small circular plots because parasitic effects on host plants are limited to the close neighborhood (approx. 10 cm around the parasite; GIBSON & WATKINSON 1992). Such small plot sizes are commonly used in hemiparasite studies (e.g. PYWELL et al. 2004, AMELOOT et al. 2006, MUDRÁK et al. 2016, HEER et al. 2018). In each of the 90 investigated plots, we estimated the cover of the particular *Melampyrum* species, counted the number of vascular plant species and also separately the species number of the two different functional groups graminoids and forbs.

## 2.4 Statistical analysis

All statistical tests were performed in R version 3.5.1 (R CORE TEAM 2018). We used random slope, mixed-effects models (lme4 and lmerTest packages; BATES et al. 2015, KUZNETSOVA et al. 2017) to test the density effect of the two *Melampyrum* species separately on total vascular plant species richness and on the richness of forbs and graminoids. These models allowed both the mean and linear trend of *Melampyrum* density to vary randomly for each population in order to correct for population- and site-specific differences. *Melampyrum* was omitted from calculations on diversity measures. We also included *Melampyrum* cover as a quadratic term because we suspected non-linear relationships (HEER et al. 2018). For all mixed-effects models, we calculated R<sup>2</sup> as the marginal



**Fig. 1.** Location of the two study regions (in light grey) on the island of Saaremaa (Estonia). A: *Melampyrum arvense*, B: *Melampyrum nemorosum*.

**Abb. 1.** Lage der für beiden Untersuchungsgebiete auf der Insel Saaremaa (Estland). A: *Melampyrum arvense*, B: *Melampyrum nemorosum*.

coefficient (proportion of variance explained by the fixed factors alone) and as the conditional coefficient of determination (proportion of variance explained by both fixed and random factors; NAKAGAWA & SCHIELZETH 2013), using the MuMin package (BARTON 2018; Table 1).

### 3. Results

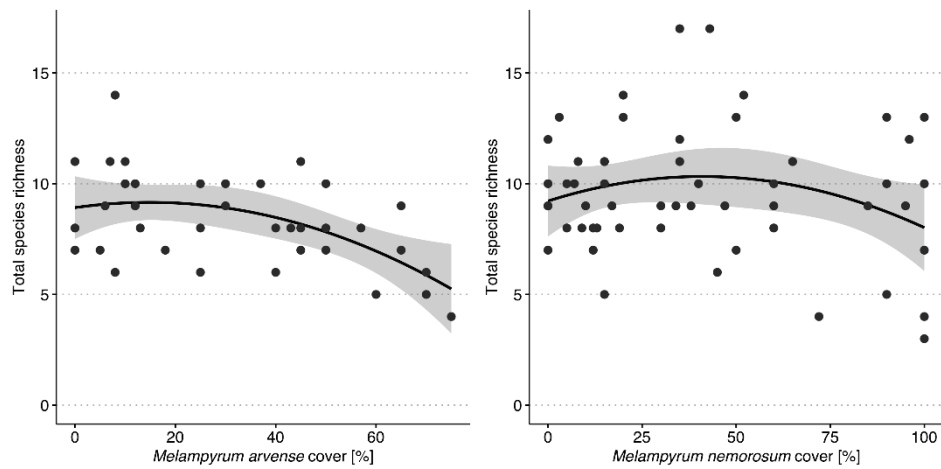
In *Melampyrum arvense* plots, the species richness was on average 8.3 per 314 cm<sup>2</sup> and ranged from 4 to 14 species. In *M. nemorosum* plots the species richness was on average 9.6 per 314 cm<sup>2</sup> and ranged from 3 to 17 species. We found significant hump-shaped relationships of total vascular plant species richness with relative cover of *M. arvense* and *M. nemorosum*, with the highest average richness values of 10.3 and 9.2 species occurring at 13% and 40% cover, respectively (Fig. 2, Table 1). The proportion of variance explained by the fixed factors alone (marginal coefficient) was intermediate for *M. arvense* (26%) and relatively low for *M. nemorosum* (7%). However, the proportion of variance explained by both the fixed factors and the random factors (conditional coefficient) was high (42% for *M. arvense* and 51% for *M. nemorosum*; Table 1). Compared with plots without these hemiparasites, total species richness increased by 2.5% in plots with *M. arvense* and by 10.7% in

**Table 1.** Summary of random-slope, mixed-effects models (position and linear trend of the curve varies randomly for each population), showing the effects of *Melampyrum arvense* and *M. nemorosum* density (including a quadratic term) on the total number of vascular plant species, as well as on the number of forb and graminoid species separately.  $R^2$  is given as the marginal coefficient (proportion of variance explained by the fixed factors alone) and the conditional coefficient (proportion of variance explained by both the fixed factors and the random factor) of determination for mixed-effects models. Significant differences are indicated by bold p-values ( $p < 0.05$ ).

**Table 1.** Zusammenfassung der «Random Slope Models» (gemischte lineare Modelle, bei denen Position und linearer Trend der Kurve zufällig für jede Population variieren), die die Dichteeffekte von *Melampyrum arvense* und *M. nemorosum* (einschließlich eines quadratischen Terms) auf die Gesamtzahl der Gefäßpflanzenarten sowie auf die Anzahl der Kräutern- und Gräserarten separat zeigen. Die Steigung gibt die Richtung des Effekts an. Signifikante Unterschiede sind durch fett gedruckte P-Werte ( $p < 0,05$ ) gekennzeichnet.

	Total plant species richness			Forb species richness			Graminoid species richness								
	Estimate	SE	df	T	p	Estimate	SE	df	T	p	Estimate	SE	df	T	p
Intercept	8.95	0.56	6.98	15.99	<0.001	6.40	0.50	7.08	12.80	<0.001	2.50	0.40	4.76	6.27	<0.001
<i>M. arvense</i> density	-0.77	0.38	4.75	-2.02	0.102	-0.61	0.36	4.14	-1.67	0.168	-0.29	0.21	4.34	-1.42	0.223
<i>M. arvense</i> density <sup>2</sup>	-0.71	0.34	30.45	-2.08	<b>0.046</b>	-0.51	0.30	30.47	-1.68	0.103	-0.20	0.19	30.12	-1.07	0.294
$R^2$ marginal	0.26					0.20					0.10				
$R^2$ conditional	0.42					0.39					0.41				

	Total plant species richness			Forb species richness			Graminoid species richness								
	Estimate	SE	df	T	p	Estimate	SE	df	T	p	Estimate	SE	df	T	p
Intercept	10.32	0.88	7.13	11.78	<0.001	6.54	0.7	8.14	9.3	<0.001	2.96	0.5	7.01	5.88	<0.001
<i>M. nemorosum</i> density	0.01	0.50	7.97	0.02	0.982	-0.32	0.39	12.55	-0.81	0.433	0.10	0.25	8.37	0.39	0.707
<i>M. nemorosum</i> density <sup>2</sup>	-0.79	0.37	42.67	-2.16	<b>0.037</b>	-0.25	0.34	46.27	-0.73	0.470	-0.30	0.20	42.36	-1.45	0.154
$R^2$ marginal	0.07					0.04					0.02				
$R^2$ conditional	0.51					0.38					0.48				



**Fig. 2.** Relationships between the total species richness (*Melampyrum* excluded) and the percentage of *Melampyrum arvense* cover (left) and the percentage of *M. nemorosum* cover (right). The trend line indicates significant quadratic relationships, with the 95% confidence interval given in grey (untransformed raw data).

**Abb. 2.** Beziehungen zwischen dem Artenreichtum der Gefäßpflanzen (ohne *Melampyrum*) und der prozentualen Deckung von *Melampyrum arvense* (links) und der prozentualen Deckung von *M. nemorosum* (rechts). Die Trendlinie zeigt signifikante quadratische Beziehungen an, wobei das 95 %-Konfidenzintervall in Grau angegeben ist (untransformierte Werte).

plots with *M. nemorosum* at these cover values. Starting at relatively low densities (> 29.7% cover) of *M. arvense* and relatively high densities (> 81.8% cover) of *M. nemorosum*, species richness was even lower than the average values of 8.7 (*M. arvense*) or 9.3 (*M. nemorosum*) in plots without these hemiparasites (Fig. 2). In contrast to results for total species richness, we found no significant effect of hemiparasite density when looking at graminoid and forb species separately (Table 1).

#### 4. Discussion

Using a density-gradient approach, we found that the relationships between vascular plant species richness and *Melampyrum* density follow an optimum curve. This finding corresponds to that of HEER et al. (2018), who investigated effects of *Rinanthus alectorolophus* density on the species richness of meadows in Switzerland. Thus, our findings suggest that the density-dependent effects of hemiparasites on local species richness can probably be translated to further hemiparasitic species and therefore might be more widespread than originally assumed.

A common explanation for the positive effects of hemiparasites on plant diversity is the balancing of the competitive relationships within a community and the overall reduction of grassland productivity (DAVIES et al. 1997, BULLOCK & PYWELL 2005, MUDRÁK et al. 2016, HEER et al. 2018). However, in line with HEER et al. (2018), we found tipping points of particular hemiparasite densities, beyond which local species richness dropped below the average richness of plots without hemiparasites. This tipping point differed strongly between

the two investigated *Melampyrum* species, showing that optimum densities cannot be generalized among hemiparasitic species and that the density of individual species should therefore be considered in restoration measures. Whether these differences can be attributed to differing host selection of the two *Melampyrum* species needs to be further explored in future studies. Moreover, the densities of hemiparasites are considered to vary over time (AMELOOT et al. 2006, CAMERON et al. 2009) which might lead to cyclic effects of hemiparasites on plant diversity. Hemiparasites might promote diversity by suppressing dominant plant species and thereby promoting subordinate species until a certain density tipping point. After reaching this tipping point, subsequent invasion by competitive species becomes more likely because hemiparasites then might lose advantage when suitable hosts are no longer available and decline in abundance. The increase of competitive species then again might constitute resources and therefore can be again beneficial for hemiparasites. Thus, patches with high hemiparasite densities, so called “shifting clouds” can move through grasslands over time (CAMERON et al. 2009). The lack of information on *Melampyrum* densities in previous seasons is therefore a limitation of our study and all other previous observational studies investigating the effect of hemiparasites on diversity and composition in only one year. In case of low hemiparasite density plots, we therefore cannot distinguish between recently invaded plots and the ones where the parasite density had already declined because of the decreasing abundance of suitable hosts. However, spatially explicit monitoring over several years would be required to confirm whether hemiparasites exhibit such cycles of temporal changes in patch density. Until now, such temporal dynamics have not yet been monitored with permanent study plots and needs to be tested in future studies.

In addition, we found no significant effects of hemiparasite density on graminoid and forb species analyzed separately, indicating that the effect on total vascular plant species richness was not driven by gains or losses of species in only one of these two functional groups. This result also suggests that the two investigated *Melampyrum* species are generalist hemiparasites that parasitize many species rather than selectively attacking certain functional groups.

Our results further highlight that the density-gradient approach is more effective than the ‘all or nothing’ approach. The latter has been used in many previous studies, showing pure positive, neutral or pure negative hemiparasite effects on local plant species richness (e.g. GIBSON & WATKINSON 1992, JOSHI et al. 2000, AMELOOT et al. 2005, BARDGETT et al. 2006, FIBICH et al. 2016, CHAUDRON et al. 2021). Not accounting for changes along a hemiparasite-density gradient may even result in misleading management recommendations and provides no opportunity for compromise between the two contrasting opinions of hemiparasitic species as loved or hated. On the one hand, hemiparasites are recommended as a tool for grassland restoration (e.g. BODIS et al. 2021). On the other hand, from an agro-economic perspective hemiparasites are generally very unpopular because a reduction in yield is expected with their presence (e.g. MAGDA et al. 2004, AMELOOT et al. 2006, HEER et al. 2018). However, these polarized views cannot be applied to management decisions when it is understood that relationships between species richness and hemiparasite density follow an optimum curve.

## 5. Conclusions

In conclusion, our results that the relationship between species richness and *Melampyrum* density follows an optimum curve extend the findings of HEER et al. (2018) and show that hemiparasite effects on local species richness are likely density dependent in general. This clearly supports the use of the density-gradient approach instead of the ‘all or nothing’ approach in future studies, when the aim is to explore relationships between hemiparasites and species richness.

### Erweiterte deutsche Zusammenfassung

**Einleitung** – Hemiparasitische Pflanzenarten betreiben Photosynthese, bilden aber Haustorien an den Wurzeln anderer Arten und entziehen so ihren Wirten Wasser, Nährstoffe, Assimilate und sekundäre Verbindungen (ADLER 2000, CAMERON & SEEL 2007, TĚŠITEL et al. 2010, WESTWOOD et al. 2010). Sie können dadurch die interspezifische Konkurrenz in Pflanzengemeinschaften reduzieren, indem sie die Dichte dominanter Pflanzen verringern und generell die Produktivität verringern. Auf diese Weise können sie die Koexistenz erleichtern und die Stabilität von Pflanzengemeinschaften fördern (PHOENIX & PRESS 2005). Einjährige hemiparasitische Arten können zudem die Etablierung von Keimlingen anderer Arten fördern, indem sie nach dem Absterben Lücken schaffen (JOSHI et al. 2000).

Die Auswirkungen von hemiparasitischen Arten auf die Diversität von Pflanzengemeinschaften sind jedoch umstritten. Die Tatsache, dass sowohl positive, negative als auch neutrale Effekte von Hemiparasiten auf die lokale Diversität gefunden wurden (GIBSON & WATKINSON 1992, AMELOOT et al. 2005, FIBICH et al. 2016), kann auf den am häufigsten verwendeten methodischen Ansatz zurückgeführt werden, d. h. den Vergleich von Standorten mit und ohne Hemiparasiten ("Alles-oder-Nichts-Ansatz"; z. B. GIBSON & WATKINSON 1992, FIBICH et al. 2016). Entgegen der intuitiven Erwartung einer rein linearen negativen Beziehung zwischen einer dominanten Pflanzenart und dem Artenreichtum einer Pflanzengemeinschaft hat sich jedoch gezeigt, dass Effekte von Hemiparasiten höchstwahrscheinlich dichteabhängig sind und einer Optimumkurve folgen, wobei die höchste Diversität bei mittleren Hemiparasitendichten auftreten (z. B. gezeigt für *Rhinanthus alectorolophus* (Scop.) Pollich; HEER et al. 2018). Dies legt nahe, dass der "Alles-oder-Nichts"-Ansatz für die Untersuchung von Hemiparasiten-Effekten auf den Artenreichtum irreführend sein kann, da dieser den Dichtegradienten von Hemiparasiten nicht erfasst. Ob diese Zusammenhänge auch für andere Hemiparasiten als *Rhinanthus* spp. gelten, wie *Melampyrum* spp., wurde jedoch bislang noch nicht untersucht.

Um diese Wissenslücke zu schließen, untersuchten wir die Vegetation an zehn Standorten in zwei Regionen auf der Insel Saaremaa (Estland). Wir testeten, wie der lokale Artenreichtum von (1) allen Gefäßpflanzen und (2) Kräutern und Grasartigen separat, entlang von Dichtegradienten der beiden Hemiparasiten *Melampyrum arvense* und *M. nemorosum* variiert.

**Methoden** – Die Daten wurden im Rahmen eines pflanzenökologischen Feldkurses der Universität Bern auf der Ostseeinsel Saaremaa (57° 50' bis 58° 40' N, 21° 45' bis 23° 20' E) im Juli 2015 erhoben. Das Grundgestein dieser Insel besteht aus silurischem Kalkstein, der oft von weichselzeitlichen Moränen oder holozänen Meeressedimenten bedeckt ist. Saaremaa beherbergt eine Vielzahl von Vegetationstypen und eine einzigartige Flora. Insbesondere die Alvar-Trockenrasen, die auf sehr flachgründigen und humusreichen Schluff- oder Lehmböden über Kalkgestein wachsen, haben schon lange das Interesse von Botanikern auf sich gezogen (VILBERG 1927, ZOBEL & KONT 1992, PÄRTEL et al. 1999, BOCH 2005, BOCH & DENGLER 2006, DENGLER et al. 2006).

Für unsere Untersuchungen wählten wir zwei hemiparasitische Arten der Gattung *Melampyrum* (*Orobanchaceae*) aus, die auf Saaremaa häufig sind und lokal dichte Populationen bilden: *M. arvense* L. ist eine generalistische Art, die Getreide und andere Grasarten, aber auch viele krautige Arten parasitiert (MATTHIES 1996, MATTHIES 2017). Historisch war die Art in Ackerbaugebieten von Westeuropa bis zum Uralgebirge und der Türkei weit verbreitet. Heute ist sie auf verschiedene Trockensavannengemeinschaften, thermophile Säume und halbruderale Graslandschaften beschränkt (EURO+MED



2006–2019). *Melampyrum nemorosum* L. ist wahrscheinlich ebenfalls eine generalistische Art, die Gräser, aber auch verschiedene Kraut- und Gehölzarten parasitiert (KÄSERMANN 1999, HOLÁ et al. 2017). Ihr Verbreitungsgebiet reicht ebenfalls von Westeuropa bis zum Uralgebirge im Norden (EURO+MED 2006–2019).

Auf der Insel Saaremaa wählten wir zwei Regionen aus, in denen entweder *M. arvense* oder *M. nemorosum* häufig vorkam (Abb. 1). In der ersten Region entdeckten wir vier räumlich getrennte Populationen von *M. arvense* in thermophilen Saumgesellschaften entlang landwirtschaftlicher Felder. In der zweiten Region konnten wir sechs getrennte Populationen von *M. nemorosum* in thermophilen Saumgesellschaften innerhalb eines offenen *Quercus robur*-Waldgebietes finden. Innerhalb jeder Population platzierten wir neun kreisförmige 314 cm<sup>2</sup> (20 cm Durchmesser) Untersuchungsflächen entlang eines *Melampyrum*-Dichtegradienten innerhalb eines visuell homogenen Gebietes. Wir unterteilten jede Wiese visuell in Zonen mit geringer, mittlerer oder hoher Dichte der jeweiligen *Melampyrum*-Art und platzierten dann zufällig drei Untersuchungsflächen in jeder Zone, um den Bereich von einer geringen bis zu einer maximalen Dichte innerhalb jeder Wiese zu erfassen (bis zu 70 % *M. arvense* und 100 % *M. nemorosum*-Deckung). Wir wählten kleine kreisförmige Parzellen, da parasitäre Effekte auf Wirtspflanzen auf die unmittelbare Nachbarschaft beschränkt sind (ca. 10 cm um den Parasiten herum; GIBSON & WATKINSON 1992). In jeder der 90 Untersuchungsflächen schätzten wir die Deckung der jeweiligen *Melampyrum*-Art, zählten die Anzahl der Gefäßpflanzenarten und auch separat die Artenzahl von Grasartigen und Kräutern.

Wir verwendeten «Random Slope Models» eine Art von gemischten linearen Modellen, um den Dichteeffekt der beiden *Melampyrum*-Arten auf den Artenreichtum der Gefäßpflanzen zu testen. Wir haben zudem die *Melampyrum*-Deckung als quadratischen Term einbezogen, da wir nicht-lineare Beziehungen vermuteten (HEER et al. 2018).

**Ergebnisse** – Die Beziehungen zwischen dem Artenreichtum der Gefäßpflanzen und der Deckung von *Melampyrum arvense* und *M. nemorosum* folgte einer Optimumskurve, wobei die höchsten durchschnittlichen Artenzahlen von 10,3 und 9,2 Arten bei 13 % bzw. 40 % Bedeckung auftraten (Abb. 2, Tab. 1). Im Vergleich zu Untersuchungsflächen ohne Hemiparasiten stieg der Gesamtartenreichtum bei diesen Deckungswerten in Untersuchungsflächen mit *M. arvense* um 2,5 % und in Parzellen mit *M. nemorosum* um 10,7 %. Ab relativ niedrigen Dichten (> 29,7 % Deckung) von *M. arvense* und relativ hohen Dichten (> 81,8 % Deckung) von *M. nemorosum* lag der Artenreichtum sogar unter den Durchschnittswerten von 8,7 (*M. arvense*) bzw. 9,3 (*M. nemorosum*) in Untersuchungsflächen ohne diese Hemiparasiten (Abb. 2). Wir fanden jedoch keinen signifikanten Effekt der Hemiparasitendichte, wenn wir die Grasartigen und Kräuter getrennt betrachteten (Tab. 1).

**Diskussion** – Unser Ergebnis, dass die Beziehungen zwischen Gefäßpflanzenartenreichtum und *Melampyrum*-Dichte einer Optimumskurve folgen, entspricht jenem von HEER et al. (2018), die den Artenreichtum von Wiesen in der Schweiz entlang von *Rinanthus alectorolophus*-Dichtegradienten untersuchten. Unsere Ergebnisse deuten somit darauf hin, dass die dichteabhängigen Effekte von Hemiparasiten auf den lokalen Artenreichtum wahrscheinlich auf weitere hemiparasitische Arten übertragen werden können und weiter verbreitet sind als ursprünglich angenommen.

Eine gängige Erklärung für die positiven Effekte von Hemiparasiten auf die Pflanzendiversität ist die Reduktion der interspezifischen Konkurrenz in Pflanzengemeinschaften, durch die Verringerung der Dichte dominanter Pflanzen sowie der Produktivität (DAVIES et al. 1997, BULLOCK & PYWELL 2005, MUDRÁK et al. 2016, HEER et al. 2018). Ebenso wie HEER et al. (2018) fanden wir ab einer bestimmten Hemiparasitendichte einen Umkehrpunkt, ab dem die lokale Artenvielfalt unter den Wert ohne Hemiparasiten fiel. Dieser Umkehrpunkt unterschied sich stark zwischen den beiden untersuchten *Melampyrum*-Arten, was zeigt, dass optimale Dichten von Hemiparasiten nicht verallgemeinert werden können. Ob diese Unterschiede auf eine unterschiedliche Wirtsauswahl der beiden *Melampyrum*-Arten zurückgeführt werden können, muss in zukünftigen Studien untersucht werden.

Zudem fanden wir keine signifikanten Effekte der Hemiparasitendichte auf Grasartige und Kräuter, wenn wir diese separat analysierten. Dies zeigt, dass der Effekt auf den gesamten Gefäßpflanzen-Artenreichtum nicht von Gewinnen oder Verlusten von Arten in nur einer dieser beiden funktionalen

Gruppen zurückzuführen ist. Dieses Ergebnis legt zudem nahe, dass die beiden untersuchten *Melampyrum*-Arten eher generalistische Hemiparasiten sind, die verschiedene Pflanzenarten parasitieren und nicht selektiv bestimmte funktionelle Gruppen angreifen.

Unsere Ergebnisse unterstreichen außerdem, dass der „Dichtegradienten“-Ansatz effektiver ist als der „Alles-oder-Nichts“-Ansatz. Letzterer wurde in vielen früheren Studien verwendet, die rein positive, neutrale oder rein negative Effekte von Hemiparasiten auf den lokalen Pflanzenartenreichtum zeigten (z. B. GIBSON & WATKINSON 1992, JOSHI et al. 2000, AMELOOT et al. 2005, BARDGETT et al. 2006, FIBICH et al. 2016) und berücksichtigt nicht die Veränderungen entlang von Hemiparasiten-Dichtegradienten. Da Hemiparasiten einerseits als Mittel zur Wiederherstellung der Artenvielfalt im Grünland empfohlen werden, andererseits aber aus agrarökonomischer Sicht generell sehr unbeliebt sind, da mit ihrer Anwesenheit eine Ertragsminderung erwartet wird (MAGDA et al. 2004, AMELOOT et al. 2006, HEER et al. 2018), könnte dieser einseitige Ansatz sogar zu irreführenden Managementempfehlungen führen und bietet keine Möglichkeit für einen Kompromiss zwischen den beiden gegensätzlichen Auffassungen. Unsere Ergebnisse sprechen somit eindeutig für die Verwendung des Dichtegradienten-Ansatzes anstelle des "Alles-oder-Nichts"-Ansatzes in zukünftigen Studien, wenn es darum geht, Beziehungen zwischen Hemiparasiten und dem lokalen Artenreichtum zu untersuchen.


## Acknowledgements


We thank Melissa Dawes for help editing our manuscript, the editor Balázs Deak for the handling of the manuscript and the two anonymous reviewers as well as Jakub Těšitel for the constructive comments.

## Author contributions

All authors conceived and designed the project. NH, FK, YK and NR conducted the field survey. DP, NH and SB analyzed the data. SB wrote the first draft of the paper. All authors contributed to the final manuscript.

## ORCID iDs

Steffen Boch  <https://orcid.org/0000-0003-2814-5343>

Yasemin Kurtogullari  <https://orcid.org/0000-0002-9928-137X>

## References

- ADLER, L.S. (2000): Alkaloid uptake increases fitness in a hemiparasitic plant via reduced herbivory and increased pollination. – *Am. Nat.* 156: 92–99.
- AMELOOT, E., VERHEYEN, K., BAKKER, J.P., DE VRIES, Y. & HERMY, M. (2006): Long-term dynamics of the hemiparasite *Rhinanthus angustifolius* and its relationship with vegetation structure. – *J. Veg. Sci.* 17: 637–646.
- AMELOOT, E., VERHEYEN, K. & HERMY, M. (2005): Meta-analysis of standing crop reduction by *Rhinanthus* spp. and its effect on vegetation structure. – *Folia Geobot.* 40: 289–310.
- BARDGETT, R.D., SMITH, R.S., SHIEL, R.S., PEACOCK, S., SIMKIN, J.M., QUIRK, H. & HOBBS, P.J. (2006): Parasitic plants indirectly regulate below-ground properties in grassland ecosystems. – *Nature* 439: 969–972.
- BARTON, K. (2018): MuMIn: multi-model inference. R package version 1.42.1. – URL: <https://CRAN.R-project.org/package=MuMIn>.
- BATES, D., MÄCHLER, M., BOLKER, B. & WALKER, S. (2015) Fitting linear mixed-effects models using lme4. – *J. Stat. Software* 67: 1–48.

- BOCH, S. (2005): Phytodiversität, Charakterisierung und Syntaxonomie der Trockenrasen auf Saaremaa (Estland). – Diploma thesis, depon. Library, Univ. Lüneburg.
- BOCH, S. & DENGLER, J. (2006): Floristische und ökologische Charakterisierung sowie Phytodiversität der Trockenrasen auf der Insel Saaremaa (Estland). – Arb. Inst. Landschaftsökol. Münster 15: 55–71.
- BÓDIS, J., FÜLÖP, B., LÁBADI, V., MÉSZÁROS, A., PACSAI, B., SVAJDA, P., VALKÓ, O. & KELEMEN, A. (2021): One year of conservation management is not sufficient for increasing the conservation value of abandoned fen meadows. – *Tuexenia* 41: 381–394.
- BULLOCK, J.M. & PYWELL, R.F. (2005): *Rhinanthus*: A tool for restoring diverse grassland? – *Folia Geobot.* 40: 273–288.
- CAMERON, D.D. & SEEL, W.E. (2007): Functional anatomy of haustoria formed by *Rhinanthus minor*: linking evidence from histology and isotope tracing. – *New Phytol.* 174: 412–419.
- CAMERON, D.D., WHITE, A. & ANTONOVICS, J. (2009): Parasite-grass-forb interactions and rock-paper-scissor dynamics: predicting the effects of the parasitic plant *Rhinanthus minor* on host plant communities. – *J. Ecol.* 97: 1311–1319.
- CHAUDRON, C., MAZALOVÁ, M., KURAS, T., MALENOVSKÝ, I. & MLÁDEK, J. (2021): Introducing ecosystem engineers for grassland biodiversity conservation: A review of the effects of hemiparasitic *Rhinanthus* species on plant and animal communities at multiple trophic levels. – *Perspect. Plant Ecol. Evol. Syst.* 52: 125633.
- DAVIES, D.M., GRAVES, J.D., ELIAS, C.O. & WILLIAMS, P.J. (1997): The impact of *Rhinanthus* spp. on sward productivity and composition: implication for the restoration of species-rich grasslands. – *Biol. Conserv.* 82: 87–93.
- DENGLER, J. & BOCH, S. (2007): Taxonomy, ecology, and distribution of six remarkable plant taxa (Spermatophyta, Bryophyta, and Lichenes) of the Estonian dry grassland flora. – *Phyton* 47: 47–71.
- DENGLER, J., LÖBEL, S. & BOCH, S. (2006): Dry grassland communities of shallow, skeletal soils (*Sedo-Scleranthenea*) in northern Europe. – *Tuexenia* 26: 159–190.
- EURO+MED (2006–2019): Euro+Med PlantBase – the information resource for Euro-Mediterranean plant diversity. <http://ww2.bgbm.org/EuroPlusMed/> [accessed 2021-03-03].
- FIBICH, P., LEPŠ, J., CHYTRÝ, M. & TĚŠITEL, J. (2016): Root hemiparasitic plants are associated with high diversity in temperate grasslands. – *J. Veg. Sci.* 28: 184–191.
- GIBSON, C.C. & WATKINSON, A.R. (1992): The role of the hemiparasitic annual *Rhinanthus minor* in determining grassland community structure. – *Oecologia* 89: 62–68.
- HEER, N., KLIMMEK, F., ZWAHLEN, C., FISCHER, M., HÖLZEL, N., KLAUS, V.H., KLEINEBECKER, T., PRATI, D. & BOCH, S. (2018): Hemiparasite-density effects on grassland plant diversity, composition and biomass. – *Perspect. Plant Ecol. Evol. Syst.* 32: 23–29.
- HOLÁ, E., KOCKOVÁ, J. & TĚŠITEL, J. (2017): DNA barcoding as a tool for identification of host association of root-hemiparasitic plants. – *Folia Geobot.* 52: 227–238.
- JOSHI, J., MATTHIES, D. & SCHMID, B. (2000): Root hemiparasites and plant diversity in experimental grassland communities. – *J. Ecol.* 88: 634–644.
- KÄSERMANN, C. (1999): *Melampyrum nemorosum*. – In: KÄSERMANN, C. & MOSER, D.M.: Merkbücher Artenschutz – Blütenpflanzen und Farne: 198–199. Bundesamt für Umwelt, Wald und Landschaft, Bern.
- KUUSK, V., TABAKA, L. & JANKEVICIENE, R. (Eds.) (1996): Flora of the Baltic Countries. Compendium of Vascular Plants, Vol. 2. – Eesti Loodusfoto AS, Tartu: 372 pp.
- KUZNETSOVA, A., BROCKHOFF, P.B. & CHRISTENSEN, R.H.B. (2017): lmerTest package: tests in linear mixed effects models. – *J. Stat. Softw.* 82: 1–26.
- MAGDA, D., DURU, M. & THEAU, J.-P. (2004): Defining management rules for grasslands using weed demographic characteristics. – *Weed Sci.* 52: 339–345.
- MATTHIES, D. (1996): Interactions between the root hemiparasite *Melampyrum arvense* and mixtures of host plants: Heterotrophic benefit and parasite-mediated competition. – *Oikos* 75: 118–124.
- MATTHIES, D. (2017): Interactions between a root hemiparasite and 27 different hosts: growth, biomass allocation and plant architecture. – *Perspect. Plant Ecol. Evol. Syst.* 24: 118–137.
- MUDRÁK, O., DE BELLO, F., DOLEŽAL, J. & LEPŠ, J. (2016): Changes in the functional trait composition and diversity of meadow communities induced by *Rhinanthus minor* L. – *Folia Geobot.* 51: 1–11.
- NAKAGAWA, S. & SCHIELZETH, H. (2013): A general and simple method for obtaining  $R^2$  from generalized linear mixed-effects models. – *Methods Ecol. Evol.* 4: 133–142.

- PÄRTEL, M., KALAMEES, R., ZOBEL, M. & ROSÉN, E. (1999): Alvar grasslands in Estonia: variation in species composition and community structure. – *J. Veg. Sci.* 10: 561–568.
- PHOENIX, G.K. & PRESS, M.C. (2005): Linking physiological traits to impacts on community structure and function: the role of root hemiparasitic *Orobanchaceae* (ex-*Scrophulariaceae*). – *J. Ecol.* 93: 67–78.
- PYWELL, R.F., BULLOCK, J.M., WALKER, K.J., COULSON, S.J., GREGORY, S.J. & STEVENSON, M.J. (2004): Facilitating grassland diversification using the hemiparasitic plant *Rhinanthus minor*. – *J. Appl. Ecol.* 41: 880–887.
- R DEVELOPMENT CORE TEAM (2018): R: A language and environment for statistical computing. – R Foundation for Statistical Computing, Vienna. – URL: <http://www.r-project.org/>.
- TĚŠITEL, J., PLAVCOVÁ, L. & CAMERON, D.D. (2010): Heterotrophic carbon gain by the root hemiparasites, *Rhinanthus minor* and *Euphrasia rostkoviana* (*Orobanchaceae*). – *Planta* 231: 1137–1144.
- VILBERG, G. (1927): Die Alvare und die Alvarvegetation in Ost-Harrien [in Estonian, with German summary]. – *Tartu Ülikooli Juures Oleva Loodus, Seltsi Aruanded* 34: 11–139.
- WESTWOOD, J.H., YODER, J.I., TIMKO, M.P. & DE PAMPILIS, C.W. (2010): The evolution of parasitism in plants. – *Trends Plant Sci.* 15: 227–235.
- ZOBEL, M. & KONT, A. (1992): Formation and succession of alvar communities in the Baltic land uplift area. – *Nord. J. Bot.* 12: 249–256.