Plant density and seed production of *Rhinanthus minor* under long-term Ca, N, P and K fertiliser application in the Rengen Grassland Experiment (Germany)

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Abstract

Rhinanthus minor L. is a summer annual and facultative hemiparasitic plant common in low-productivity grasslands. Survival of this species is strictly dependent on sufficient seed production in each vegetative season. We aimed to evaluate the effect of different fertiliser treatments on plant density per m², seed production per individual plant and seed production per m2. All data were collected in unfertilised control, Ca, CaN, CaNP, CaNPKCl and $CaNPK_2SO_4$ treatments of the Rengen Grassland Experiment in June 2010. Plant density per m²ranged from 5 to 745 in CaNPKCl and control treatments, seed production per individual plant from 24.9 to 65 seeds in control and $CaNPK_2SO_4$ treatments, and total seed production per m2 from 195 to 18142 seeds in CaNPKCl and control treatments, respectively. High density of small plants in low productivity control contrasted highlywith low density of tall plants in fully fertilised treatments. We concluded that for sufficient seed production per plot, high plant density of small *R. minor* plants is more relevant than low density of tall plants with high number of seeds.

Keywords: *Arrhenatherion*, *Polygono-Trisetion*, *Violion caninae*, yellow rattle

Introduction

Rhinanthus minor L. is a summer annual, facultative hemiparasitic therophyte species common in low productivity grasslands (Westbury, 2004). *R. minor* was formerly considered a weed due to low yield and forage quality, but today is highly valued for its ability to decrease competitive ability of highly productive forage species thereby facilitating restoration of species-rich grasslands. Survival of *R. minor* is strictly dependent on sufficient seed production in each vegetative season as it produces short-lived seeds. In monoculture, performance of individual *R. minor* plants is highest in substrates with high nutrient availability, but this can be *vice versa* when *R. minor* occurs in the real permanent grassland in mixture with potential host species. *R. minor* is highly sensitive to shade from tall canopy of potential host species especially at seedling stage. This might explain why *R. minor* is generally less common or absent in highly productive grasslands with aboveground annual biomass production > 5 t ha^{-1} (Hejcman *et al*., 2011). The aim of the research reported in this paper was to estimate the seed production of *R. minor* in different plant communities which developed over 65 years under different fertiliser treatments in the Rengen Grassland Experiment (RGE). We asked how is the plant density per m², seed production per individual plant and seed production per m² affected by fertiliser treatments in the RGE.

Materials and methods

The RGE was established in 1941 on low productive *Violion caninae* grassland in the Eifel Mts. (SW Germany, altitude: 475 m a. s. l.; precipitation: 811 mm; temperature: 6.9ºC) in a completely randomized block design with six fertiliser treatments (Control, Ca, CaN, CaNP, CaNPKCl and CaNPK2SO4, see Table 1) and five replicates (30 plots in total, individual plot size 5×3 m). Sixty-five years of fertilizer application resulted in development of different plant communities in close neighbourhood on a meadow that was cut twice per year in early July and in mid October. In each experimental plot, the number of *R. minor* plants was counted in m² monitoring plot in late June 2010. Ten randomly selected plants were then collected per plot and seed number per individual plant was determined in the laboratory. Seed production per m² was then calculated by multiplying plant density per m² with seed production per individual plant. One-way ANOVA followed by Tukey's *post-hoc* test were used to evaluate effect of treatment on obtained data.

Results and discussion

Density of *R. minor* plants was significantly affected by treatment and was highest in the unfertilized control where 745 plants per m² were recorded (Table 2). Very low density of plants, less than 3% relative to control, was recorded in all treatments with Ca, N and P application. In contrast to plant density, seed production per individual plant was significantly lowest in the control treatment where 24.9 seeds per individual plant were recorded. In $CaNPK_2SO_4$ treatment, seed production per individual plant was about 263% higher than in the control. Total seed production per m² was significantly affected by treatment and was highest in the control where 18142 seeds per m² were recorded. Total seed production per plot ranged from 1 to 3% relative to control in all treatments with Ca, N, P application.

Although high nutrient availability substantially increased the number of seeds per individual plant of *R. minor*, high number of seeds per individual plant was not able to overcome the effect of the low plant density on seed production per plot. Therefore the lowest seed production per plot was recorded in highly productive *Arrhenatherion* grassland in fully fertilized treatments where highest seed production per individual plant of *R. minor* was recorded. Low density of *R. minor* plants in fully fertilized plots with tall canopy of dominant grasses was given predominately by the competitive exclusion of seedlings, as they are known to be highly shade-intolerant (Keith, 2004) and by the lower ability of seedlings to attach to host plants. It is known that host plants develop different root architecture and exhibit higher meristematic activity under high compared to low phosphorus supply, and thus decreasing the probability of the parasitic plant becoming attached (Davies and Graves, 2000).

Conclusion

We concluded that plant density is more relevant for sufficient production of seeds per area than seed production per individual plant. This is supported by the fact that differences in plant density per plot can be substantially higher (almost 100 times in this study) than differences in seeds production per individual plant (almost 3 times in this study). Substantially lower differences in seed production per individual plant compared to differences in plant densities are given by the strict physiological limitation of seed numbers per individual plant. High density of small plants is therefore an efficient strategy for survival of *R. minor* in permanent low productivity grasslands.

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Table 1. Amounts of nutrients (kg ha^{-1}) supplied annually to the treatments since 1941 according to Schellberg *et al*. (1999), classification of plant communities which developed because of different fertilizer application into alliances according to Chytrý *et al*. (2009), total annual aboveground dry matter biomass production of vascular plants (BP, t ha^{-1}) according to Hejcman *et al*. (2010a) and standing dry matter biomass of bryophytes (SB, t ha^{-1}) in 2006 according to Hejcman *et al*. (2010b).

Treat. abbrev.	Applied nutrients	Alliance	BP	SB
A - Control	unfertilized control	*Violion caninae*	2.5	1.8
B - Ca	Ca = 715, Mg = 67	*Polygono-Trisetion*	2.9	0.46
C - CaN	Ca = 752, N = 100, Mg = 67	*Polygono-Trisetion*	4.9	0.02
D - CaNP	Ca = 936, N = 100, P = 35, Mg = 75	*Arrhenatherion*	6.5	0.25
E - CaNPKCl	Ca = 936, N = 100, P = 35, K = 133, Mg = 90	*Arrhenatherion*	8.9	0.15
F - $CaNPK_2SO_4$	Ca = 936, N = 100, P = 35, K = 133, Mg = 75	*Arrhenatherion*	9.6	0.09

Table 2. Plant density (number of plants) per m² (PD), plant density per m² relative to control (PDR), mean seed number per plant (SPP), seed number per plant relative to control (SPPR), seeds production per m² (SP) and seeds production per m² relative to control (SPR). ± values indicate SE. Calculated by one-way ANOVA followed by Tukey's post-hock test, treatments with the same letter are not significantly different. All ANOVA analyses were significant on 0.001 probability level.

Treat. abbrev.	PD	PDR	SPP	SPPR	SP	SPR
A - Control	$745^{a} \pm 113$	100%	$24.9^{a} \pm 2.1$	100%	$18142^{a} \pm 2608$	100%
B - Ca	$641^{a} \pm 71$	86%	$27.2^{a} \pm 2.2$	109%	$16560^{a} \pm 2064$	91%
C - CaN	$29^{b} \pm 22$	4%	$37.4^{ab} \pm 3.1$	150%	$1365^{b} \pm 1137$	8%
D - CaNP	$15^{b} \pm 6.4$	2%	$45.0^{b} \pm 3.4$	181%	$545^{b} \pm 192$	3%
E - CaNPKCl	$5^{b} \pm 5$	<1%	$48.4^{bc} \pm 6.9$	194%	$198^{b} \pm 198$	1%
F - $CaNPK_2SO_4$	$8^{b} \pm 5$	1%	$65.4^{c} \pm 9.3$	263%	$498^{b} \pm 330$	3%

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