Vegetative sprouting as an additional pathway for a seed size-number trade-off: a field-parameterised simulation approach

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Abstract: Studies of perennial plants generally search for a seed size vs. seed number trade-off. Surprisingly, the fact that perennials may replace an investment in large seeds by the allocation to vegetative propagation has not yet been investigated as an additional pathway enabling species coexistence. We focused on the mechanisms of coexistence in *Carex elata* and *C. elongata*, two co-occurring clonal sedges dominant in European swamp alder forests. We asked the following questions: i) Is the number of germinated seeds a better predictor of species coexistence than the total number of seeds? ii) What recruitment conditions and competition rules determine vegetative sprouting to be an alternative to large, competitively superior seeds? We measured several species functional traits related to the colonisation and fitness of perennials. To examine the competitive hierarchy between species and microsite species preferences, we analysed the effects of environmental factors and plant densities on fitness-related traits using Structural Equation Modelling (SEM). Then, using a series of spatially explicit simulations partly parameterised based on the field measurement, we evaluated the importance of seed and ramet propagation and recruitment conditions for long-term species coexistence. SEM indicated a competitive hierarchy and a large overlap in microsite preferences between species. As a response to our initial questions we found that: i) Only differences in the numbers of germinated seeds, allowed the two species to coexist. If we consider only differences in the total number of seeds, the superior competitor (*Carex elata*) outcompeted the inferior competitor (*C. elongata*) in all scenarios. This is because the former produced about three-times as many seeds as the latter. ii) We show that vegetative sprouting represents an additional pathway for the seed size-number trade-off when the competitive superiority of species is attributed to vegetative propagation. This is another way that a species deals with the omnipresent seeds of other species. Taken together, our study demonstrates that differences in seed performance, coupled with differences in vegetative propagation related to competitive ability, are an additional mechanism allowing the coexistence of perennial plants.

Abbreviations: CFI – Comparative Fit Index; GLMM – Generalised Linear Mixed Model; MLM – Maximum Likelihood Method; RMSEA – Root Mean Square Error of Approximation Index; SEM – Structural Equation Modelling; SEVM – Spatial Eigenvalue Vector Mapping.

Introduction

Trade-offs between functional traits related to plant resource acquisition, competition and colonisation ability or stress tolerance are essential mechanisms supporting species coexistence (Tilman 1994, Muller-Landau 2010). A negative relationship between seed size and seed number was ascribed to a competition-colonisation trade-off in which the competitive benefit of large seeds is counterbalanced by the disadvantage of low seed production (Rees 1995, Geritz et al. 1999). According to this trade-off, small-seeded species possess a 'hit-and-run' strategy and invade new favourable sites due to their higher colonisation rates than those of large-seeded species, which profit from better competitive ability. Studies supporting plant coexistence based on trade-offs in reproductive traits have dealt primarily with annuals (e.g., Rees 1995, Ben-Hur et al. 2012), displaying only one reproductive mode, i.e., the allocation to generative reproduction. Surprisingly, studies on perennial plants have also generally focused on addressing the seed size vs. seed number trade-off (e.g.,

Eriksson and Jakobsson 1998, Guo et al. 2000, Leishman 2001, Eriksson 2005), but neglected the possibility that an investment in large seeds and successful seedling recruitment, may be replaced by vegetative propagation (Harper 1967, Abrahamson 1980, Cheplick 1995, Fischer and van Kleunen 2002).

Empirical data have shown that a trade-off between generative and vegetative reproduction commonly occurs in clonal plant species (Cheplick 1995, Thompson and Eckert 2004, van Drunen and Dorken 2012). The increase in seed size is accompanied by the decrease in vegetative propagation rate, and vice versa. Several authors stressed that a reproductive allocation trade-off will be apparent mainly in those environments where resources are limited or under stressful conditions (Weppler et al. 2006). A recent empirical study, compiling data from both databases and the field, supported that local processes such as specific environmental conditions and biotic interactions may affect the realisation of species' reproductive trade-offs (Herben et al. 2012). Seed recruitment of plants in perennial vegetation has often been shown to be

associated only with vegetation gaps (Bullock et al. 1995, Eriksson 2011). On the other hand, in dense vegetation, clonally growing ramets have a higher chance of establishment compared to seedlings (Williams et al. 1977). Grime (1998) postulated that founder effect may importantly influence the species composition in local space. The set of species that co-occur in a given area will depend upon early colonisation by appropriate dominants and subordinates (Egler 1954, Fukami 2004). A late arrival may be expected to delay an establishment of a species or may even exclude some species completely (Platt 1975, Grman and Suding 2010).

Studies addressing the seed size vs. seed number trade-off mostly used seed size or the total number of seeds as proxies of investment to generative reproduction (e.g., Guo et al. 2000, Leishman 2001). Nevertheless, clonal plants often reduce sexual fertility compared to non-clonal plants as a consequence of pollination failure (Charpentier et al. 2000), lack of mates or genetically conditioned sexual sterility (Eckert 2002). Hence, unviable seeds are highly probable to occur in clonal species. More suitable predictors of seed performance, e.g., seed germination, should be used in studies for an alternative estimation of generative reproduction output.

In this study, we developed a series of spatially explicit simulations to ask whether differences in reproductive strategies and competitive abilities between two perennial sedges dominant in European swamp broadleaf forests may be a sufficient mechanism for ensuring their local species coexistence. In the field study, we measured several life-history traits, which generally correlate with colonisation ability and fitness of perennials. To examine the competitive hierarchy between species, microsite species preferences and environmental niche overlap, we analysed the effects of environmental factors and plant densities on fitness-related traits using Structural Equation Modelling (SEM). Then, using a series of spatially explicit simulations partly parameterised by the field measurement, we evaluated the importance of seed and ramet propagation and recruitment conditions for long-term species coexistence. More specifically, we asked the following questions: i) Is the number of germinated seeds a better predictor of species coexistence than the total number of seeds? ii) What recruitment conditions and competition rules determine vegetative sprouting to be an alternative to large, competitively superior seeds?

Material and methods

Study site and species

The study was performed in an old-growth swamp broadleaf forest in the Vrbenské rybníky Nature Reserve located in southern Czech Republic (49° 00' N, 14° 25' E, 350 m a.s.l.). *Carex elata* All. and *C. elongata* L. (Cyperaceae) are the dominant species in the understorey vegetation (Douda et al. 2012). Both species are long-lived clonal perennials associated with alder carr throughout Europe, and frequently dominate the herbaceous layer (Douda et al. 2016). They produce densely tufted shoots, and can also produce short under-

ground rhizomes up to approximately 10 cm in length (Douda et al., personal observation). Both species also reproduce sexually by achenes. The achenes ripen at the beginning of June and germinate immediately after they are released from the mother plant or may be stored for several years in the soil seed bank (Schütz 2000).

Field measurements

Field measurements of species characteristics and environmental factors were made during the first week of June 2012 (see Appendix 1 for data). A 350-m-long transect in a south-north direction with a random starting point was established. Fifty-four plots of size 1.5 m × 1.5 m, each 5 m apart, were placed along the transect. Within each plot, all adult individuals of *Carex elata* and *C. elongata* were identified and counted. Seedlings, i.e., individuals with 3 or fewer leaves or less than 8 cm in height, were excluded from the data sampling because a determination of small individuals of both species may be misleading. In total, 78 and 390 individuals of *C. elata* and *C. elongata* were recorded, respectively. For fruiting plants (in total, 53 individuals of *C. elata* and 212 individuals of *C. elongata*), the mean number of germinated seeds per ramet, mean number of seeds per ramet, mean seed size (mg; calculated from the 50-seed mass of each fruiting individual which were germinated), the number of all seeds per individual, the number of germinated seeds per individual, the number of ramets and the total seed mass per individual (mg) were recorded. The dried seeds, including the perigynium, were weighed. The number of all seeds per individual was estimated as the ratio of total seed mass to mean seed size. The mean number of seeds per ramet was calculated as the ratio of the number of all seeds to the number of all ramets.

Seeds were stored in paper bags at room temperature for two weeks. Fifty seeds of each fruiting individual were germinated, if available. Seeds were placed on a single layer of filter paper in 90-mm-diameter Petri dish and moistened with water. Seeds were treated by cold-wet stratification in the dark at 4°C for 6 weeks to synchronise the timing of the germination. After that, they were incubated at 22°C for the 16-h light period and 10°C for the 8-h dark period each day (Schütz and Rave 2003). Germinated seeds were counted and removed at 2-day intervals for 45 days. The number of germinated seeds per individual was estimated as the ratio of total seed mass to mean seed size, multiplied by germination percentage. The mean number of germinated seeds per ramet was calculated as the ratio between number of germinated seeds and the number of ramets.

To examine microsite species preferences indicating the extent of their environmental niche overlap, we measured the soil conductivity, light availability, pH and water level on each plot (Douda 2010, Douda et al. 2012). The light conditions of each plot were examined using hemispherical photographs taken at the plot centres using a Sigma 8 mm fisheye. The percentage of total radiation transmitted by the forest canopy was the variable selected to describe light availability for the

understorey plants (Frazer et al. 1999, Douda et al. 2012). The depth of water level below the ground, pH and conductivity were measured in a soil pit as the mean value of 5 points within each plot. They had a regular spatial distribution, with one pit located in the centre and border pits in space between the centre and the plot corners. A field meter (pH/Cond 340i/SET) was used to estimate the pH and conductivity.

Additionally, to characterise an annual vegetative spread of both *Carex* species we used data from a common garden experiment that simulated the coexistence of four species in a swamp forest, including *C. elata* and *C. elongata*. The other species (i.e., *Calamagrostis canescens* (Weber) Roth and *Deschampsia cespitosa* (L.) P.B.) represent species that commonly co-occur with both sedges in a swamp forest (Douda et al. 2012). By measuring 260 individuals of each species, growing alongside the five individuals in 52 pots (60 × 60 × 36 cm) in the growing season of 2011, we counted the number of new vegetative sprouts per maternal plant having a rhizome of length greater than 1 cm (Appendix 1).

Data analyses

Differences between species in trait values, i.e., mean number of all seeds per ramet, mean number of germinated seeds per ramet, mean seed size, number of all seeds, number of germinated seeds and number of new vegetative sprouts were tested using Generalised Linear Mixed Models (GLMMs). A Gaussian distribution was implemented for mean seed size, mean number of seeds per ramet and mean number of germinated seeds per ramet, whereas a Poisson distribution was implemented for the number of seeds per plant, the number of germinated seeds per plant, and the number of new vegetative sprouts (i.e., the count data). GLMMs were performed using the package *lme4* (ver. 1.1-8) and function *glmer* in R software (R Core Team 2015).

To examine the competitive hierarchy between species and microsite species preferences, we analysed the effects of the environmental factors and plant densities of *Carex elata* and *C. elongata* on their fitness-related traits (i.e., the number of all seeds, number of germinated seeds, number of ramets, mean seed size and species density) measured in each plot. We used structural equation modelling (SEM), a multivariate technique that estimates the strength and sign of directional relationships for complex networks having multiple dependent variables (Bollen and Long 1993, Grace 2006). As a technique, SEM is more flexible than simple or multiple linear regression, because it deals with a system of regression equations. It represents a very useful statistical approach that can be used to unravel the linking structure of fitness-related traits that are correlated in a multivariate way (Shipley, 2004). The SEM diagram shows the direction of relationships and estimates the strength of those relationships (i.e., standardised path coefficients). A standardised path coefficient is the standardised slope of the regression of the dependent variable on the independent variable in the context of the other independent variables (Grace 2006). Medians of the fitness-related trait values of all individuals measured at each plot were used in the analyses because some plots had few plants

(particularly plots with a presence of *C. elata*). The median is a good measure of the centrality, particularly when data are overdispersed. Plots with absent species were not included in the analyses (i.e., 14 plots for *C. elata* and 6 plots for *C. elongata*).

In SEM, the environmental factors were considered exogenous variables in all equations, whereas fitness-related traits were endogenous variables. Densities of target species were considered to be exogenous variables in equations with fitness-related traits and endogenous variables in equations with environmental factors. We performed log-transformations, whenever it was necessary, to ameliorate the normality of variables. We used a maximum likelihood method (MLM) with robust standard errors. The most parsimonious models were selected using a combination of forward selection, backward elimination and Akaike Information Criteria. The Satorra-Bentler test statistic robust to non-normality with a rescaled version of the standard χ^2 was used to assess the overall fit of the final models. The quality of the models was also evaluated using the root mean square error of approximation index (RMSEA) and the comparative fit index (CFI). The following criteria were used to indicate SEM with a satisfactory fit: (1) *P*-values of chi-square tests > 0.05, (2) RMSEAs < 0.05, and (3) CFIs > 0.95 (Bollen and Long 1993). All analyses were performed using the function *cfa* and package *lavaan* (ver. 0.5-18) in the R software (R Core Team 2015).

Because the spatial autocorrelation of endogenous variables (i.e., fitness-related traits and density of target species) among plots, disrupts the assumptions of SEM and increases the risk of type-I errors, we accounted for spatial autocorrelation in all SEM. Moran's I was calculated to assess the spatially autocorrelated endogenous variables using the package *Ape* and function *Moran.I* (ver. 3.0-11) in the R software (R Core Team 2015). A distance matrix of plots and within-plot median values of particular endogenous variables were used in the Moran's I tests. After that, Spatial Eigenvalue Vector Mapping (SEVM) was adopted for endogenous variables with Moran's I *P* values < 0.05. The multivariate technique, SEVM, translates the spatial arrangement of data points into a set of eigenvectors, capturing the spatial effects (Dormann et al. 2007). We assessed eigenvectors using principal coordinate of neighbour matrices (PCNM). Only positive eigenvectors were considered to always represent significant autocorrelation. These eigenvectors were used as predictors in regression models with endogenous variables (i.e., fitness-related traits and density of target species) as dependent variables to determine spatially uncorrelated regression residuals (Dormann et al. 2007). The regression residuals were then input to SEM in place of the original (observed) endogenous variables. The SEVM was performed using SAM software (ver. 4.0) (Rangel et al. 2010).

To evaluate whether differences in seed performance, rate of ramet propagation and competitive ability support the species coexistence in clonal plant assemblages, we developed a series of spatially explicit models (Fig. 1, Table 1). Based on the results of the SEM, which showed negative density dependence of *Carex elongata* on *C. elata* and no effects of environmental conditions on local species distribution, we

Table 1. Parameters of spatially explicit models.

Model parameters	Description
Lattice	50 x 50 cells; cell states: empty, occupied by *Carex elata* OR occupied by *C. elongata*
Initial species distribution	at the start, each of the two species occupied 25% of randomly-selected cells; 50% of cells remained empty
Maturity	seeds and ramets became adults in the same time step and produce new seeds and new ramets in the next time step
Propagation via seeds	numbers of seeds randomly selected from a range of values based on field measurements made on 53 individuals of *Carex elata* and 212 individuals of *C. elongata*
Propagation via ramets	numbers of new vegetative sprouts randomly selected from a range of values based on measurements of 260 individuals of each species growing in a common garden experiment
Spread	each individual could spread to 8 neighbouring cells using seeds and ramets (propagation via seeds and ramets); seed and ramet dispersal to each cell was calculated as the number of seeds and ramets produced in the target cell and the 8 surrounding cells multiplied by the probabilities that all seeds and ramets reached the target cell
Mortality rates	percentages of randomly-selected plants that died in each step: 1%, 3%, 5%, 7% and 10%
Model time steps (t) and repetition (n)	t = 1,000, n = 10; if both species coexisted after 1,000 time steps, we considered their coexistence to be possible

modelled the coexistence of two species, including a superior competitor (i.e., *C. elata*) with the low number of germinated seeds, and an inferior one (i.e., *C. elongata*) with the large number of germinated seeds. We used a regular square lattice with 50 × 50 cells, where each cell had three states, i.e., emp-

Figure 1. Overview of spatially explicit simulations. Parameters included in the models are presented in italics.

ty, occupied by *C. elata* or occupied by *C. elongata*. Only one individual could occur in each cell. In each time step, an individual could spread to 8 neighbouring cells using seeds and ramets ensuring that there is a continuity at the edges (i.e., we consider a donut-shape) (Soetaert and Herman 2009). Each species initially occupied 25% of randomly-selected cells and 50% of cells remained empty. Seeds and ramets became adults in the same time step and produced new seeds and ramets in the next time step. We counted the numbers of cells occupied by the adults of each species after each time step. For each model, we repeated the simulation protocol 10 times and calculated the mean species densities for each time step. If both species coexisted after 1,000 time steps, we considered their coexistence to be possible.

In each step, the number of germinated seeds and of all seeds for each of the individuals in the lattice matrix were randomly selected from a range of values based on field measurements made on 53 individuals of *Carex elata* and 212 individuals of *C. elongata*. Analogously, the numbers of new vegetative sprouts (ramets) were randomly selected from a range of values based on measurements of 260 individuals of each species growing in the common garden experiment. Seed and sprout dispersal to each cell, was calculated as the number of seeds and new vegetative sprouts produced in the target cell and in the eight surrounding cells, multiplied by the probability that all seeds and sprouts reached the target cell.

The following set of simulations was used to examine the plant traits and environmental limitations of species co-existence (see Tables 1-2 for description of all models and competitive ranking). We started with the simulation of both species propagating only via seeds to determine if differences in seed numbers accompanied with strong competitive asymmetry are sufficient for their coexistence (*Propagation via seeds* model). Then, we included propagation via ramets into the simulations to show the role of both seed and ramet propagation (*Propagation via seeds and ramets* model).
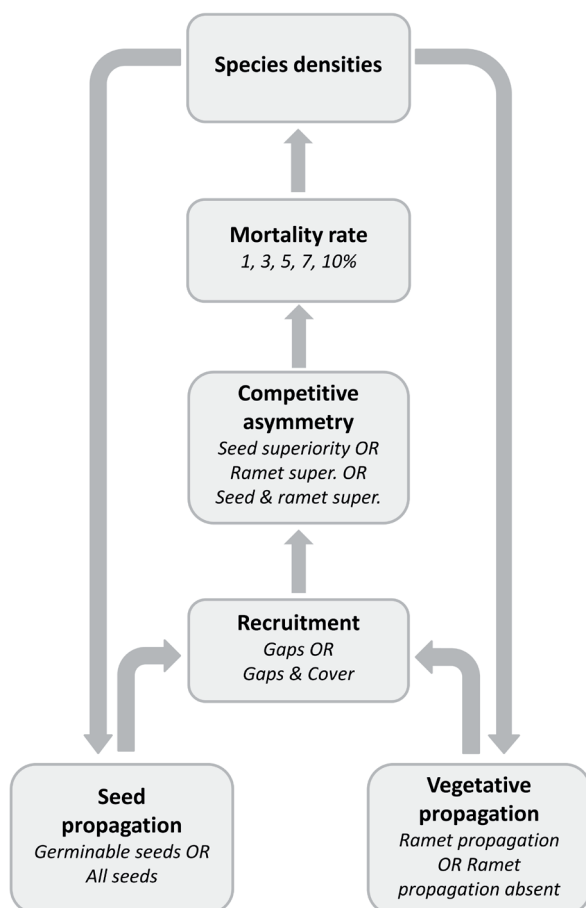
Table 2. Species competitive ranking (CR) related to the competitively superior species (i.e., *Carex elata*) used in different spatially explicit models. Competitive ranking was applied to all models but the *Equal competitive abilities* models. a, adult; r, ramet; s, seedlings (seeds); ELA, *C. elata*; ELO, *C. elongata*; for example, CR: *sELO* OR *aELO <- sELA* indicates that seedlings of *C. elata* have a competitive advantage over seedlings and adults of *C. elongata*, CR: *sELO* OR *rELO* OR *aELO == rELA* indicates that ramets of *C. elata* have an equal competitive ability to the seedlings, ramets and adults of *C. elongata*.

Propagation	Competitive asymmetry	Recruitment	Competitive ranking
Seeds		*Gaps*	*sELO <- sELA*
		Gaps & Cover	*sELO* OR *aELO <- sELA*
Seeds and ramets	Seed and ramet superiority	*Gaps*	*sELO* OR *rELO <- sELA* *sELO* OR *rELO <- rELA*
		Gaps & Cover	*sELO* OR *rELO* OR *aELO <- sELA* *sELO* OR *rELO* OR *aELO <- rELA*
	Seed superiority	*Gaps*	*sELO* OR *rELO <- sELA* *sELO* OR *rELO == rELA*
		Gaps & Cover	*sELO* OR *rELO* OR *aELO <- sELA* *sELO* OR *rELO* OR *aELO == rELA*
	Ramet superiority	*Gaps*	*sELO* OR *rELO == sELA* *sELO* OR *rELO <-* rELA
		Gaps & Cover	*sELO* OR *rELO == sELA* *sELO* OR *rELO* OR *aELO <- rELA*

Within the *Propagation via seeds and ramets* model, we attributed the competitive superiority of species to individual reproductive modes in three variants: i) the *Seed and ramet superiority* model attributing the competitive advantage of superior species to both modes of propagation; ii) the *Seed superiority* model assigning competitive advantage to seed recruitment of the superior competitor, whereas maintaining equal competitive ability for both species during propagation via ramets; and iii) the *Ramet superiority* model attributing competitive advantage to ramet propagation of the superior competitor, whereas maintaining equal competitive ability for both species during recruitment via seeds. In all models, the superior competitor replaced the inferior competitor whenever it was propagated (i.e., reached the cell) with the competitive advantage. In cases that both species had an equal competitive ability, we selected the one randomly attending to species proportion of seeds and new vegetative sprouts in the cell.

To determine if the number of germinated seeds is a better predictor of species coexistence than the number of all seeds, we performed all simulations separately, once with germinated seeds (*Germinated seeds* model) and once with all seeds (*All seeds* model). Because the recruitment of plants in perennial vegetation is often associated only with vegetation gaps, we performed all simulations in two variants: first, with possible recruitment only in unoccupied cells (*Gaps* model) and second, with recruitment in all occupied and unoccupied cells (*Gaps & Cover* model). Previous spatial competition models have shown that species with different life-histories (i.e., one is a good coloniser/poor competitor, and another a poor coloniser/good competitor) coexisted depending on the disturbance frequency (Cadotte 2007). Therefore, we simulated different mortality rates (1%, 3%, 5%, 7% and 10%),

i.e., the percentages of plants that randomly died in each step, to capture the extent of mortality by disturbances under which coexistence is possible. Moreover, we simulated null scenarios with an equalised number of germinated seeds and ramets (*Equal number of seeds and ramets* model) and the competitive ability of the species (*Equal competitive abilities* model). The *Equal number of seeds and ramets* model was assessed with the number of all seeds and the number of germinated seeds and the number of new vegetative sprouts selected for each individual in the lattice in a species-nonspecific manner. In the *Equal competitive abilities* model we selected the one species to win in each cell, based on a probability corresponding to the species proportion of seeds and the number of new vegetative sprouts in the cell.

Results

Species differences in seed size, seed number, seed number per ramet and number of new sprouts

Carex elata had three times as many seeds (645.55 ± 113.89, mean $\pm$ se) as *C. elongata* (218.7 ± 17.26; $\chi^2 = 13972$, n = 265, $P < 0.001$; Fig. 2a). In contrast, *C. elongata* had 22.3 times as many germinated seeds (106.64 ± 11, mean germination percentage = $40.89 \pm 1.78\%$) as *C. elata* (4.79 ± 1.76, mean germination percentage = $0.72 \pm 0.24\%$; $\chi^2 = 4760.9$, n = 265, $P < 0.001$; Fig. 2a). No germinated seeds were recorded for 78% of the *C. elata* plants (n = 54). Similarly, the mean number of seeds per ramet was 2.6 times as many for *C. elata* (7.42 ± 1.03) as for *C. elongata* (2.83 ± 0.17; $\chi^2 = 33.798$, n = 265, $P < 0.001$; Fig. 2b), whereas the mean number of germinated seeds per ramet was 19 times as many for *C. elongata* (1.14 ± 0.08) as for *C. elata* (0.06 ± 0.02; $\chi^2 = $
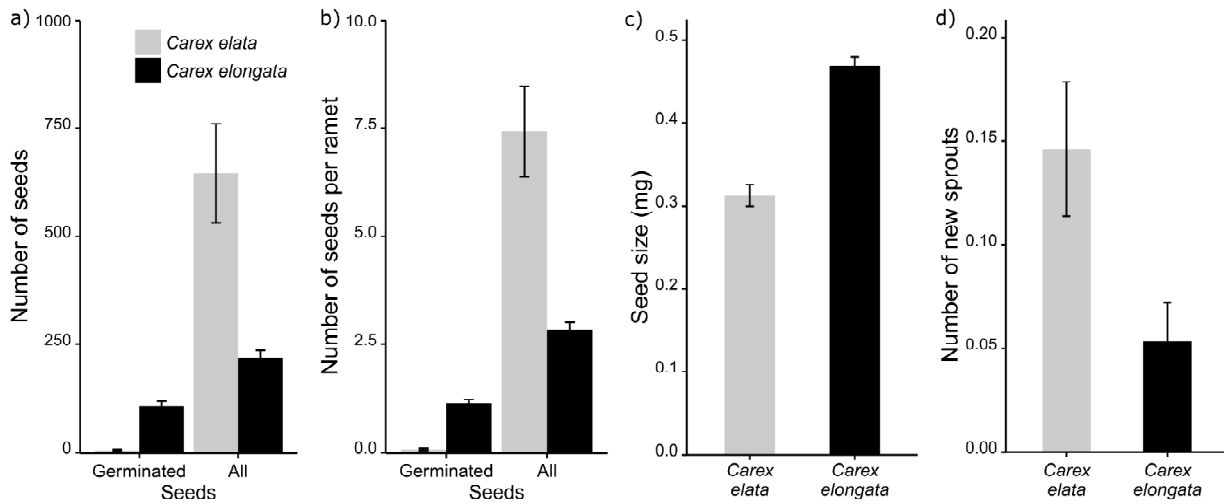
Figure 2. Differences in seed traits of *Carex elata* and *C. elongata*. Error bars are standard errors of the mean. All differences were significant (*P* < 0.05).

Figure 3. Structural equation modelling (SEM) representing the effects of environmental variables and plant densities on fitness-related traits of *Carex elongata* (a) and *C. elata* (b). Unidirectional arrows represent standardised path coefficients, with the line thickness being proportional to the coefficient strength. Bidirectional arrows show the correlations between exogenous variables. R^2 indicates the amount of variation of the target variable explained by the model. res. indicates endogenous factors, for which spatial autocorrelation was detected and the residuals of regression models were inputted into SEM instead of the originally observed variables. *** *P* < 0.001; ** *P* < 0.01; * *P* < 0.05.
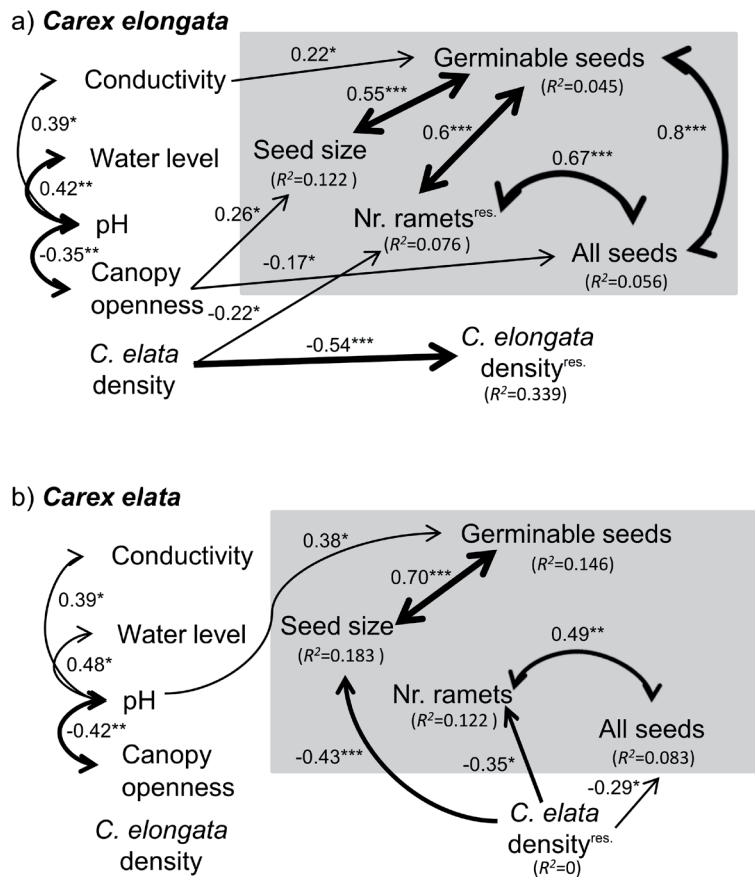
70.342, n = 265, *P* < 0.001; Fig. 2b). The mean seed size was 1.48 times as many for *C. elongata* (0.46 ± 0.01 mg) as for *C. elata* (0.31 ± 0.01 mg; χ^2 = 42.803, n = 265, *P* < 0.001; Fig. 2c). Only low number of plants (i.e., 1.25% and 0.38% for *C. elata* and *C. elongata*, respectively) propagated via vegetative sprouts more than 1 cm in length. The number of new vegetative sprouts was 2.73 times greater for *C. elata* (0.15 ± 0.03) than for *C. elongata* (0.05 ± 0.02; χ^2 = 149.58, n = 520, *P* < 0.001; Fig. 2d).

Factors influencing fitness-related traits: plot scale

The final SEM fitted the data structure well for both *Carex elata* (χ^2 = 14.742, n = 30, *P* = 0.791; RMSEA = 0; CFI = 1) and *C. elongata* (χ^2 = 18.843, n = 48, *P* = 0.338; RMSEA = 0.048; CFI = 0.988). For *C. elongata*, the density and number of ramets decreased with the density of *C. elata* (Fig. 3a). The number of all seeds decreased with increasing canopy openness (Fig. 3a). Seed size increased with canopy

openness, and the number of germinated seeds increased with increasing soil conductivity (Fig. 3a). The number of germinated seeds was positively correlated with the seed size, number of ramets and number of all seeds. The number of ramets was positively correlated with the number of all seeds. For *C. elata*, the number of ramets, seed size and number of all seeds significantly decreased with increasing density of *C. elata* (Fig. 3b). The number of germinated seeds of *C. elata* increased with seed size and soil pH (Fig. 3b). The soil conductivity and water level were positively correlated with pH, and canopy openness was negatively correlated with pH.

Spatially explicit models of species coexistence

After 1,000 time steps, species coexisted at all mortality rates in all competitive scenarios when recruited in gaps via germinated seeds or germinated seeds and ramets except in the *Ramet superiority* scenario (Fig. 4). In the *Ramet superiority* scenario, species coexisted at 5 and 7% of the mortality rate when recruited in gaps and under cover. Species also coexisted at 1, 3, 5 and 7% of the mortality rate under the null model with an equalised number of seeds when recruited in gaps, and competitive superiority was attributed to the ramet propagation. In all other simulations, one of two species survived and the other became extinct. The inferior competitor (i.e., *Carex elongata*) won in all simulations when both species had equal competitive abilities or under the *Ramet superiority* scenario when it recruited via germinated seeds and ramets both in gaps (at all mortality rates) and in gaps and cover (at a 10% mortality rate).

The germination, competition and gap recruitments had the following effects on species coexistence: (1) If germination differences between both species were not considered the species that produced more seeds won (*All seeds* model). (2) If we simulated equal competition ability between species, the species with better establishment won (the one with more germinable seeds). (3) The coexistence of species was strongly dependent on recruiting in gaps. The results for the *Ramet superiority* scenario in gaps were more dependent on the absence of seed superiority than on the ramet superiority itself (when seeds were competitively equal, the species with highest germination won). Indeed, vegetative spread was very low (ca 1%), so it is not an estrange result at all. The results for the *Ramet superiority* scenario in a gap and under a canopy (cover) show that vegetative spread is the alternative pathway when species must deal with the omnipresent seeds of other species. However, this is only true if vegetative recruitment is not limited to the presence of vegetation gaps.

Discussion

Seed traits as estimates of competitive ability and recruitment potential in clonal plants

Species differed in the traits closely related to seed recruitment potential, specifically in the number of all seeds and of germinated seeds. However, only the number of germi-

nated seeds was shown to explain species coexistence when it was implemented into the spatially explicit simulations. If we considered only differences in the number of all seeds, the superior competitor (i.e., *C. elata*) outcompeted the inferior competitor in all scenarios because the former had three times as many seeds as the latter (Fig. 4). Although studies on perennial plants generally focused on addressing the seed size vs. seed number trade-off (e.g., Leishman 2001, Lönnber and Eriksson 2012), our study indicates that neither seed size nor the seed number provide good estimates of the competitive ability and recruitment potential of perennial plants. In our study, the superior competitor had a considerably higher number of seeds but a lower seed size than the inferior competitor. Perennials may allocate resources to clonal growth rather than to the seeds (Cheplick 1995, van Drunen and Dorken 2012). The low germination of *C. elata* was linked to the lower ratio of the number of germinated seeds to the number of ramets. On the other hand, we recorded many germinated seeds and the higher ratio of the seed number to the ramet number for *C. elongata*. We can speculate that an investment of *C. elata* into vegetative biomass resulted in the allocation of limited resources to achene development (Lee 1988, Fang et al. 2006). In contrast, a higher number of germinated seeds in *C. elongata* may correspond to a plant strategy that maximises seed reproduction success (Weiner et al. 2009).

Studies dealing with trade-offs between competition/utilisation and colonisation in clonal plants primarily noted differences in clonal growth forms. A trade-off between allocation to long vs. short spacers has been described (Cheplick 1997), and the possible coexistence of species with such differences in life histories was confirmed (Benot et al. 2013). In our study, albeit as tussock plants, both sedges were able to propagate via underground rhizomes (longer than 1 cm from a maternal plant); *Carex elata* propagated via rhizomes with the frequency three times that of *C. elongata*.

Species competitive ranking and microsite preferences

The results of structural equation modelling indicated a competitive hierarchy and large overlap in microsite preferences between the species (Fig. 3). Both competitive asymmetry and overlapping environmental niches between species are an essential assumption underlying the coexistence of species with different colonisation abilities (Geritz et al. 1999). We recorded a negative effect of *C. elata* plant density on *C. elongata* plant density and the number of ramets. In contrast, *C. elongata* density did not negatively affect the reproductive traits of *C. elata* nor its density. Van de Koppel and Crain (2006) noted that competition among *Carex* plants inhibits their growth and recruitment, due to a deep wrack layer produced by tussocks. In our study, an analogous mechanism may be an important trigger of interspecific competition because *C. elata* produces a large number of leaves every year. These leaves decompose only slowly (Douda et al., personal observation).

Environmental conditions (i.e., canopy openness, conductivity and pH of the soil) influenced plant seed traits but did not affect the local distribution of plants or the number
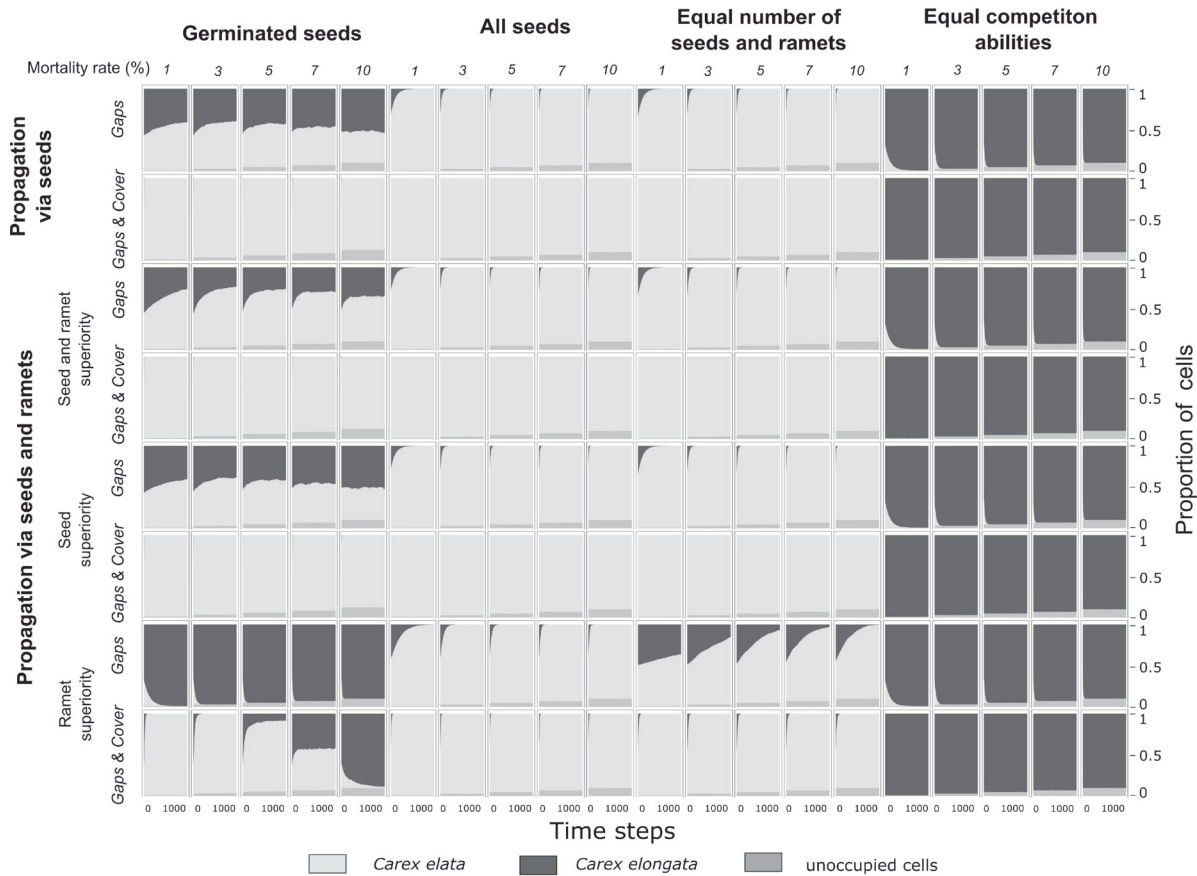
Figure 4. Spatially explicit simulations of the coexistence of species differing in the number of germinated seeds, rate of vegetative propagation and competitive ability (see Table 1 and 2 for description of the parameters of particular models). The mean proportions of *Carex elata* (pale grey), *C. elongata* (dark grey) and unoccupied cells (grey) during the 1,000 steps of simulations are given (n = 10). Each model was simulated for 1%, 3%, 5%, 7% and 10% mortality rate.

of ramets. Increasing light and nutrient supply in a forest ecosystem generally increases seed production and viability (Meekins and McCarthy 2000, Baskin and Baskin 2014). Previous studies have shown that the distribution of herbaceous plants in temperate forests is driven by different species preferences for microtopography (Beatty 1984, Douda et al. 2012), overstory composition and structure (Beatty 1984, Scheller and Mladenoff 2002) or soil nutrients (Bruelheide and Udelhoven 2005). However, the occurrence and abundance of clonal plants, which are dominant components of temperate forests, may only poorly reflect environmental heterogeneity (Gilbert and Lechowicz 2004, Douda et al. 2012). Douda et al. (2012) found that the distributions of *C. elata* and *C. elongata* are most strongly influenced by dispersal limitation rather than by environmental conditions.

Spatially explicit simulation of species coexistence of clonal plants

The results of spatially explicit simulations answered whether the difference of propagation importance between seed and ramet in long-term species coexistence depended on recruitment conditions and whether competition advantage was attributed to seed and/or ramet propagation. Both species coexisted when they propagated only via seeds and recruited in gaps. The results did not change when we included ramet propagation into the simulations because the rate of ramet propagation was too low to achieve the effect of seed recruitment. These results show that tussock plants that differ only in the number of seeds and competitive abilities may coexist based on seed number and competition trade-offs, but only if seedling recruitment is limited to empty sites. In this case, the low availability of sites, where species may recruit via seeds, limits the competitively superior species with a low fecundity, which leads to species coexistence.

On the other hand, both species coexisted when we attributed the competitive superiority only to ramet propagation of *C. elata*, although in limited range of mortality rates (i.e., 5–7%). In this case, the low propagation rate of ramets of the superior competitor ensured the coexistence of the inferior competitor with the omnipresent seeds. Species coexisted only if recruitment both in gap and under cover was allowed because if recruited only in gaps, the rate of ramet propagation was not sufficient to achieve the recruitment of seeds of *C. elongata*. This is because the restriction of seed and ramet recruitment to gaps imposes a very strong founder effect of seeds of inferior competitor (Platt 1975, Grime 1998).

In accordance with simulation studies on the competition-colonisation trade-off, higher rates of mortality generally increased the success of the species producing more germinating seeds, whereas lower plant mortality supported the superior competitor (Cadotte 2007). Disturbance usually decreases the competitive advantage of a superior competitor by releasing resources to the inferior competitor, thus promoting coexistence (e.g. Elias and Dias 2009). The effect of mortality rate was mostly weak except for the model with competitive advantage attributed to ramet propagation, where coexistence was possible only at an intermediate level of disturbance (Fig. 4). It has been shown that the relative risk of extinction depends on the balance between disturbance mortality rate and population growth (Shaffer 1981, Lande 1993). Increasing mortality rate interrupts competitive exclusion by reducing the densities of all species (Huston 1979, Chesson and Huntly 1997) but also makes species more prone to extinction (Fox 2013). Mortality had an effect by modulating the availability of gaps as there were no differences in mortality between species.

Recently, Herben et al. (2012) supported the existence of a seed reproduction-vegetative propagation trade-off for a large dataset of plant species growing in a botanical garden. They have shown that this trade-off may become evident only when species are influenced by site conditions or biotic interactions. For example, at sites with high productivity, local seedling establishment is limited due to high above-ground competition. Thus, allocation to clonal propagation increases the chances of plant recruitment in dense vegetation due to resource integration between mother and offspring ramets (Williams et al. 1977, Abrahamson 1980).

One characteristic of our models should be discussed. We considered the same spatial range of seed dispersal and vegetative propagation for both species. Thus, our models more strongly corresponded to studies that considered the effect of seed number and population growth than to those that considered differences in species dispersal abilities (Eriksson 2005, Ben-Hur et al. 2012). According to our results, *Carex elongata* maximised the number of seeds that may germinate and thereby gained a fecundity advantage for rapidly colonising new sites, whereas *C. elata* recruiting via a lower number of germinated seeds or via ramets have a competitive advantage and utilise the local space.

Conclusions

Based on a combination of field trait measurement, germination trial and spatial explicit simulations, we supported that species coexistence depends on seed germination and vegetative spread. Specifically, the germinated seed number is better than the seed number to account for the colonisation of empty space; vegetative spread is a tool for dealing with founder effect and account for species recruitment under the vegetation cover; and mortality creates gaps and allows the survivability of species which are better colonising empty space than occupied space. As a response to our initial questions we found that: i) Only differences in the numbers of germinated seeds has been shown to allow species coexistence

in our simulations. If we consider only differences in the total number of seeds, the superior competitor outcompeted the inferior competitor in all scenarios because the former produced about three-times as many seeds as the latter. ii) We show that vegetative sprouting represents an additional pathway for the seed size-number trade-off when the competitive superiority of species is attributed to ramet propagation. This is another way in which species deal with the omnipresent seeds of other species. Taken together, our study demonstrates that differences in seed performance and vegetative propagation related to competitive ability, may be an additional mechanism allowing perennial plants to coexist. This may be advantageous in the conditions of the forest understory, where vegetative propagation of plant species appears to be omnipresent (Bierzychudek 1982). Several studies noted difficulties in attempts to explain the local coexistence of clonal plants based on only the concept of environmental niche segregation (Wijesinghe et al. 2005, Reynolds et al. 2007) or the seed size vs. seed number trade-off (Eriksson 2005). Future research in this area presents a challenge because, at present, knowledge is still insufficient to determine the role of the generative and vegetative propagation in the coexistence of clonal plants and the diversity of clonal plant assemblages.

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Electronic Appendix

Fitness-related data measured in field study. The file may be downloaded from www.akademiai.com.