

Pollen dispersal distance is determined by phenology and ancillary traits but not floral gender in an andromonoecious, fly-pollinated alpine herb

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Abstract

Pollen-mediated gene flow and spatial genetic structure have rarely been studied in alpine plants pollinated by Dipteran insects. Furthermore, it is not clear how different floral traits, such as floral gender, phenology, and ancillary traits, may affect pollen dispersal distance within a population. In this study, we conducted a paternity analysis to track pollen flow in a population of *Pulsatilla alpina*, an andromonoecious alpine herb producing male and bisexual flowers. We found that the pollen was dispersed over short distances (mean = 3.16 meters) with a dispersal kernel of Weibull distribution. Nonetheless, spatial genetic structure was weak in the population (Sp statistic = 0.013), pointing to effective seed dispersal. The pollen dispersal distance was independent of the gender of the flower of origin but depended positively on floral stalk height and negatively on flowering date and tepal length. Although male siring success did not correlate with pollen dispersal distance, selection may favor traits increasing pollen dispersal distance as a result of reduced bi-parental inbreeding. Our study has not only provided new insights into the nature of pollen dispersal, especially of alpine plants, but has also revealed the effects of floral traits on an important component of male reproductive success.

Introduction

Gene flow in sessile plants is largely mediated by dispersal of pollen and seeds, which together determine the spatial genetic structure within and among populations. The dispersal distance should depend on not only dispersal vectors but also on plant traits that attract and manipulate the position and behaviour of the pollinators. There has been substantial progress in investigating traits that affect seed dispersal (Vittoz and Engler 2007; Thomson et al. 2011; Côrtes and Uriarte 2013; Tamme et al. 2014), but we remain largely ignorant of how pollen dispersal distance varies among dispersal vectors and for different plant trait values, likely due to the difficulties of tracking the gene flow by pollen movements.

Around 90 percent of flowering plants rely on a diverse group of animals as their pollination vector (Ollerton et al. 2011). Nonetheless, our knowledge is still limited about how pollination vectors with different attributes, especially small insects, affect pollen dispersal (Ashley 2010). Although it is generally thought that animals with a larger body size lead to greater pollen dispersal distance and thus reduce genetic structures, such conclusions are predominantly drawn from comparisons between plants pollinated by birds and bees (especially hummingbirds and bumblebees) (Krauss et al. 2017; Wessinger 2021). In contrast, it remains unclear how small insects such as flies affect pollen dispersal distances, despite the fact that they are among the most important pollinators in alpine, arctic, and agricultural ecosystems (Inouye et al., 2015; see Table 1 for a summary table of studies on pollen dispersal distance in alpine plants).

A number of floral traits likely determine the foraging behavior of pollinators within a population (Waser 1983; Chittka and Thomson 2001; Ishii et al. 2008) and thus affect pollen dispersal distances. For example, in *Delphinium virescens*, bumblebee pollinators flew longer distances after visiting flowers with a low quantity of nectar, which would likely lead to longer pollen dispersal distances for the plant

(Waddington 1981). Furthermore, in hummingbird-pollinated *Ipomopsis aggregata*, mean pollen dispersal distance, estimated using pollen dyes, depended positively on the variance but negatively on the mean stamen length, whereas it was independent of flowering date, number of flowers, and corolla size (Campbell and Waser 1989). To our knowledge, few studies have evaluated the effect of floral traits on pollen dispersal at the individual level using paternity analyses to measure realized gene flow (e.g., Tomaszewski et al. 2018; Barbot et al. 2022), despite its importance as a component of male reproductive success and its potential implications on spatial genetic structure (but see studies focusing on density: DiLeo et al. 2018; Diaz-Martin et al. 2023).

It is generally assumed that individuals dispersing their pollen over greater distances should also enjoy higher male reproductive success in terms of the total numbers of progeny sired, largely because of a supposed increase in mate availability (Harder and Prusinkiewicz 2013) and/or because of a reduced chance of mating with relatives (Price and Waser 1979). As a result, traits that facilitate pollen dispersal should be favored by selection via male reproductive success (Harder and Prusinkiewicz 2013). In wind-pollinated *Mercurialis annua* (Tonnabel et al. 2019), for instance, taller plants had a higher male siring success because they dispersed their pollen further, and the pollen dispersal distance correlated positively with male siring success. Surprisingly, in insect-pollinated species, although mating between distant individuals usually led to increased performance in components of male reproductive success (Waser and Price 1991; Souto et al. 2002), few empirical studies have tested the positive correlation between pollen dispersal distance and male siring success.

In this study, we asked how floral traits affect pollen dispersal distance in a population of the andromonoecious alpine herb *Pulsatilla alpina*, which relies on dipteran insects as pollinators. To this end, we selected a population comprising mostly single-flowered individuals, i.e., with a single male or bisexual flower, so that we could explicitly evaluate the effect of traits at the flower level on dispersal distances. We exhaustively sampled all the flowering individuals in the study population and conducted a paternity analysis using microsatellite markers. We then addressed the following questions: 1) What is the pollen dispersal kernel of *P. alpina*, and does it differ between male and bisexual flowers? 2) What is the spatial genetic structure of the *P. alpina* population? 3) How does pollen dispersal distance depend on floral traits, specifically floral gender, phenology, tepal length, and stalk height? And 4) Does mating distance correlate positively with male siring success?

Materials And Methods

Study species and study sites

Pulsatilla alpina (L.) Delarbre (Ranunculaceae) is a perennial hemicryptophyte growing in sub-alpine to alpine habitats in central Europe (Lauber et al. 2018). Several vegetative and/or reproductive shoots emerge from a perennial underground rhizome soon after the snowmelt, from early May to July. Depending on their size and age, individuals produce between zero and ~ 20 flowers with around six white tepals (the sepals and petals are not distinguishable), each on its own reproductive shoot. The

species is andromonoecious: unisexual male flowers bear only stamens, whereas protogynous bisexual flowers bear stamens and one to a few hundred uni-ovulate pistils. The sex allocation of the species is size-dependent, with larger plants allocating absolutely and proportionally more resources to their female function (Chen and Pannell, 2023). Furthermore, small individuals may produce only a single male flower and thus function as pure males in the respective flowering season (Chen and Pannell, 2023). Both male and bisexual flowers are predominantly visited by flies, including houseflies and syrphid flies (Chen and Pannell, 2022). Ripe fruits (technically achenes) with elongated pappus hair are dispersed by wind in early autumn (Vittoz and Engler 2007). After fruit dispersal, above-ground vegetative parts senesce, but individuals persist underground over winter.

The study was conducted during the flowering season of 2022 in a single population of *P. alpina*, located at Solalex in the pre-Alps of Vaud canton, Switzerland ('Population S1+'; latitude: 46°17'42"N, longitude: 7°09'09"E; elevation: 1758 a.s.l.). The population was located on an open slope of sub-alpine grassland and covering an area with dimensions of about 20 m x 20 m and comprising about 150 mainly small and probably young individuals (following recent establishment after avalanche disturbances and/or herbivory by cattle). The individuals typically produced only a single male or bisexual flower during the season. We set up a 10 m x 15 m temporally fenced plot within the population, enclosing 135 flowering individuals, and removed all the floral buds (i.e., around 10 buds) outside the plot at the very beginning of the flowering season to prevent nearby individuals outside the plot from siring progeny in the plot (thereby improving our ability to assign paternity).

Flowering phenology and ancillary traits

We recorded the flowering state of all the individuals in the population every three to four days throughout the flowering season, from late May to late June 2022, noting the number of flowers, number of stalks, height of the tallest foliar stalk, gender, and position of the flowering individuals. On each census day, we recorded the sexual stage of all the flowers in terms of seven and five categories for bisexual and male flowers, respectively (see Chen and Pannell (2023b) for a detailed description of the categories). We also photographed each flower and later counted the number of stamens based on the photographs (see details below). The onset of the male stage (flowering date) was calculated as the first date on which anthers were seen to have dehisced (M_1 stage) for both male and bisexual flowers. The height of floral stalks and the length of tepals were measured at the end of the male stage (M_2 stage), following the methods used by Chen and Pannell (2022). Around three weeks after the end of the flowering season, all the flowers with developing fruits were enclosed in a paper bag until the end of the growing season (early August), at which point all the seeds were collected.

Paternity analysis and estimates of pollen dispersal

To estimate pollen dispersal distance, we used variation at ten microsatellite markers to assign paternity to mature seeds (for details, see Chen and Pannell, 2023b). Leaf samples of all flowering individuals were collected in July 2022 at the end of the flowering season and dried in silica gel before DNA

extraction. Up to ten mature seeds for all seed families were arbitrarily selected for each sampled flower for DNA extraction. Total DNA was extracted from the leaves and seed samples using the BioSprint 96 DNA Plant Kit (Qiagen, Germany).

PCR amplification was carried out in a final volume of 10 µl, including 5 µl of 2× Multiplex PCR Master Mix (Qiagen, Germany), 2 µl of diluted DNA, 1 µl of distilled water, and 2 µl of multiplex containing variable primer concentrations (Chen and Pannell 2023b). Thermal cycling was performed in a TProfessional Standard Thermocycler (Biometra GmbH, Göttingen, Germany) as follows: 95°C for 15 min; 36 and 41 cycles for leaf and seed samples, respectively, at a temperature of 94°C for 30 s, 60°C for 45 s, and 72°C for 45 s; and a final step at 72°C for 30 min before cooling down to 4°C. PCR products were analyzed by capillary electrophoresis on an ABI3100 Genetic Analyzer (Applied Biosystems).

Sires (fathers) were assigned from among the 135 flowering individuals to all mature seeds for which more than five loci were genotyped with Cervus v 3.0.7, assuming a confidence level of 80% and an error rate of 0.018 (Kalinowski et al. 2007). Pollen dispersal distance was estimated by calculating the distance between the dam (mother) and the most likely sire for each of the successfully genotyped seeds. Although the species is self-compatible with an average selfing rate estimated to be 0.45, the selfed progeny ultimately contributes little to the next generation due to very high (0.93) inbreeding depression in the study population (unpublished data). Thus, in this study, we considered pollen dispersal only for outcrossing mating events. Male outcrossing sire success was calculated following Chen and Pannell (2023b).

Statistical analysis

We conducted the following analysis within the R statistical framework v 4.0.3 (R Core Team 2021). To quantify the pollen dispersal kernel of *P. alpina*, specifically, to compare that of bisexual and male flowers, we used the R package *dispgit* (Proença-Ferreira et al. 2023). We fitted the pollen dispersal distance with three separate models for all the flowering individuals, only individuals with a bisexual flower, and only individuals with a male flower, respectively. For each model, we used AIC values to compare the fits of Weibull, geometric, 2Dt, and exponential distributions (for the formula of each distribution, see Proença-Ferreira et al., 2023), i.e., four of the most common distributions for describing pollen dispersal (Austerlitz et al. 2004). The best-fitted distribution was then used in the following analysis to evaluate the effects of traits on pollen dispersal distance.

To assess the spatial genetic structure of the flowering individuals in the population, we used the software SPAGeDi version 1.5 (Hardy and Vekemans 2002), following the procedure described by Vekemans and Hardy (2004) based on pairwise kinship coefficients between individuals. We conducted Nason's estimator of kinship coefficient ($F_{(r)}$) (Loiselle et al. 1995). The average relationship coefficients of the ten microsatellite markers per distance class were estimated and their significance per class was tested with 1000 permutations. We used *Sp* to evaluate the extent of spatial genetic structure, which is

defined as: $S_P = -\beta / (1 - F_{(1)})$, where β is the regression slope of $F_{(r)}$ on $\ln(\text{spatial distance})$, and $F_{(1)}$ is the mean of $F_{(r)}$ among individuals for the first distance class (Vekemans and Hardy 2004).

To evaluate how floral gender, phenology, and ancillary traits affect pollen dispersal distances in *P. alpina*, we used a generalized additive model with a Weibull distribution (*gamlss* package; Stasinopoulos et al., 2018). The pollen dispersal distance of sires with a single bisexual or male flower was set as the response variable and floral gender (i.e., bisexual or male), flowering date, tepal length, and stalk height as explanatory variables. We included only the single-flowered sires with a complete set of measurements of the traits. We set the identity of the sires as a random variable because the mating events from the same flower share the same floral phenotype. We evaluated the residuals of the model using the *plot* function in the *gamlss* package (Figure S1; Stasinopoulos et al., 2018).

To investigate the relationship between pollen dispersal distance and floral male siring success, we used a generalized linear model (*glmer* function in the *lme4* package; Bates et al. 2015) with a Poisson distribution. We set the mean pollen dispersal distance of each single-flowered individual as an explanatory variable. We included an observation-level random variable to account for overdispersion (Harrison 2014). We evaluated the residuals of the model using the package *DHARMa* (Hartig 2019)

Results

Pollen dispersal kernel

We identified 513 outcross mating events for 854 genotyped seeds. Pollen dispersal distances for outcrossing were generally short, with an average of 3.16 m separating sire from the dam and 25%, 50%, and 75% of seeds sired by males < 1.0 m, < 2.15 m, and < 4.36 m away from the corresponding dam (Fig. 1A). Pollen dispersal of *P. alpina* was best fitted by a Weibull distribution for all three types of sires (Table S1). We found that the parameter b was close to 1 in all three cases, indicating a mostly fat-tailed distribution (Table 2; Fig. 1). The values of the parameters used in the Weibull distribution along with the mean dispersal distance, skewness, and kurtosis of the kernel can be found in Table 2.

Spatial genetic structure

Spatial genetic structure analysis among the 135 flowering individuals showed a significantly negative β value ($\beta = -0.013$, $P < 0.001$). The Sp statistic value of the flowering individuals was 0.013. Mean kinship coefficients ($\hat{F}_r$) across all distance classes was -0.0001 . Analysis of fine-scale genetic structure indicates a significant positive autocorrelation among individuals located up to around 3 meters apart (Fig. 1D).

Effects of floral traits on pollen dispersal distance

In total, 297 outcross mating events from 72 single-flowered sires were used for the analysis. Pollen dispersal distance depended negatively on flowering date (Fig. 2A; $P < 0.01$) and tepal length (Fig. 2B; P

< 0.001), positively with stalk height (Fig. 2C; $P < 0.05$), but was independent of floral gender (Fig. 1B and C; Table 3; $P > 0.05$).

Pollen dispersal distance and male siring success

Male outcross siring success did not depend on the mean pollen dispersal distance of the single-flowered individual ($N = 72$; Fig. 3; $P > 0.05$).

Discussion

Short pollen dispersal distance with weak spatial genetic structure

We found that pollen dispersal distance in the fly-pollinated alpine herb *P. alpina* was dominated by mating over short distances. Indeed, 75% of mating events were within 5 m. Although it has been suggested that pollen dispersal distance in herbaceous species is in general short (references in Tomaszewski et al., 2018; see also Ashley (2010) for a summary of trees and shrubs), the considerably short pollen dispersal distance in *P. alpina* is also likely a consequence of fly pollination and high population density (Levin and Kerster 1969a; van Rossum et al. 2011; Rader et al. 2011; Diaz-Martin et al. 2023). It is generally thought that plants pollinated by dipteran insects (e.g., house flies and syrphid flies) compared to hymenopteran insects (e.g., honey bees and bumble bees) have shorter pollen dispersal distances as a result of their different foraging behavior and flight distances (Inouye et al. 2015). Interestingly, in the congeneric but bee-pollinated *P. vulgaris*, the mean pollen dispersal distance ranged between 3 to 10 meters (DiLeo et al. 2018). Our results thus contribute to the general understanding of how pollination systems may shape within-population gene flow in flowering plants (Krauss et al. 2017).

Despite the short pollen dispersal distance and the mixed mating system and herbaceous growth form of *P. alpina*, we found the spatial genetic structure of the study population to be weak, likely as a result of efficient seed dispersal by wind. Spatial genetic structure in plants has been shown to depend on the mating system, pollination system, life form, and dispersal mechanism of the species (Vekemans and Hardy 2004), and efficient dispersal associated with any one of the factors should break down spatial patterns created by others (Meirmans et al. 2011). In general, high spatial genetic structure is expected for species pollinated by insects, especially those with predominant selfing, and those with a herbaceous growth form and seed dispersed by gravity (Vekemans and Hardy 2004). In contrast, seed dispersal by wind in *P. alpina* is likely to be efficient, especially as the species produces elongated floral stalks up to 70 centimeters above ground from which the bisexual flowers disperse their seeds (Chen and Pannell 2022), with a maximum dispersal distance of 80 meters (Vittoz and Engler 2007). Of course, we quantified only the pollen dispersal kernel in this study, so that estimates of both pollen and seed dispersal kernels together will be needed to provide a more complete picture of the mechanisms shaping the spatial genetic structure of *P. alpina* (e.g., García et al., 2007; Krauss et al., 2008).

Pollen dispersal distance is independent of floral gender

We did not find any difference in the pollen dispersal pattern between male and bisexual flowers in andromonoecious *P. alpina*, despite the fact that both types of flowers differed substantially in their sex allocation, morphology, and phenology (Chen and Pannell 2022, 2023b). First, we found the pollen dispersal kernels of male and bisexual flowers to be similar in terms of their shape, skewness, and kurtosis. Second, we detected no difference in mean dispersal distance, irrespective of morphology or phenology (a supplementary analysis using a univariate *gamlss* model with floral gender as an explanatory variable showed the same results; $P > 0.05$). These results conform to those of a previous study that found male and bisexual flowers to have similar male siring success (Chen and Pannell 2023b).

Direct comparisons between male and bisexual flowers in andromonoecious species have been made for various components of male fitness (Cuevas and Polito 2004; Schlessman et al. 2004; Dai and Galloway 2012), but not for pollen dispersal distance, as far as we are aware. The study on andromonoecious *Anticlea occidentalis* provided some indirect assessment of the effects of bisexual and male flowers on pollen dispersal distance by removing all the anthers from either bisexual or male flowers. That study showed that removing stamens of male flowers led to greater pollen dispersal distances compared to individuals with intact flowers or with their bisexual flowers emasculated, though the observed effects largely depended on both the paternal and maternal plants (Tomaszewski et al. 2018).

Phenology and ancillary traits affect pollen dispersal distance

The impact of flowering phenology on different aspects of female reproductive success has been extensively studied for alpine plant populations (e.g., Kudo, 2006; Collin and Shykoff, 2010; Kameyama and Kudo, 2015; Preite et al., 2015), but we remain largely ignorant of how phenology affects the male components of reproductive success. Our present results for *P. alpina* indicate that pollen dispersal distance, a major component of male reproduction success, depended negatively on the flowering date (onset of the male function). Pollen dispersal distance in *Ipomopsis aggregata* was found to be independent of phenology, based on an investigation using pollen dyes (Campbell and Waser 1989). In contrast, Hirao et al. (2006) using a two-generation analysis (Smouse et al. 2001), found that the number of effective pollen donors was higher in the late than the early season in *Rhododendron aureum*. So far, we can only conclude that the effect of phenology on the pollen dispersal distance in alpine plants is species-specific, and any patterns that do exist will only emerge with the study of further species and populations.

The negative dependence of pollen dispersal distance on phenology in *P. alpina* is likely a result of an increase in flowering density of co-flowering species rather than a change in conspecific flowering density (Figure S2) or in potential mating distance throughout the flowering season (Figure S3). According to 'optimal foraging theory' (Pyke et al. 1977), a pollinator should tend to move shorter distances between flowers in more rewarding patches, leading to shorter pollen dispersal distances (Levin and Kerster 1969b; Diaz-Martin et al. 2023). In the study area, *P. alpina* is usually the sole flowering

species in its early flowering season, but it co-flowers with other fly-pollinated species (such as *Dryas octopetala* and *Ranunculus montanus*) in the later season (KC, personal observations), such that there is a rapid increase in the density of flowers in the community as the season progresses. If the dipteran insects, as generalist pollinators (Inouye et al. 2015), follow an optimal foraging strategy by assessing floral rewards at a community level, this may result in shorter pollen dispersal distance at the late flowering season.

In insect-pollinated *P. alpina*, pollen dispersal distances were positively dependent on floral stalk height. Although a positive correlation between stalk height and pollen dispersal distance has been generally expected and found in wind-pollinated species (Okubo and Levin 1989; Tonnabel et al. 2019; Zeng et al. 2023; but see Nakahara et al. 2018; Aljiboury and Friedman 2022), it has, to our knowledge, not been reported for species relying on animals as their pollen dispersal vector. Flowers presented on taller stalks likely attract more pollinators and their pollen may be dispersed further. For instance, syrphid flies, the major pollinators of *P. alpina* (Chen and Pannell 2022), were found to be more likely to visit taller plants within and among species in grassland habitats (Klecka et al. 2018a, b).

It is not clear why flowers with a larger tepal length had a shorter pollen dispersal distance in *P. alpina* than those with smaller tepals. Given that tepal length showed no correlation with stamen number (Chen and Pannell 2022), it is unlikely that the short dispersal distance is a result of pollinators staying longer in a flower for greater pollen rewards, as predicted by 'optimal foraging theory' (Pyke et al. 1977). Alternatively, dipteran pollinators may also visit the flowers for heat as a reward, which is common in the arctic, temperate, and alpine environments (Hocking and Sharplin 1965; Kudo 1995; Inouye et al. 2015). Indeed, it has been shown that the actinomorphic flowers of *P. alpina* could be around 10 Celsius degrees warmer than the air temperature (Dietrich and Körner 2014). If larger tepals lead to warmer temperatures in the flowers, pollinators may forage shorter distances around the patch and thus cause shorter pollen dispersal distances (Pyke et al. 1977). Although the actual mechanisms behind the observed patterns between pollen dispersal distance and different traits in *P. alpina* remain obscure, our results have nevertheless revealed how intra-specific variation in floral traits affects an important component of plant mating.

The effects of phenology and morphological traits on pollen dispersal distances in *P. alpina* imply that these traits may be under selection via male reproductive success, e.g., through enhancing the quality of offspring. It is worth noting that we detected no clear dependence between mean pollen dispersal distance and the number of seeds sired, in contrast with the common assumption of a positive correlation in theory (Okubo and Levin 1989; Fromhage and Kokko 2010) and the common observation of such a correlation for wind-pollinated species (Tonnabel et al. 2019; Zeng et al. 2023). Contrary to wind-pollinated species in which the pollination likely follows a mass-action mechanism, pollen flow between sires and dams in animal-pollinated plants may be much more complex (Harder 1990; Inouye et al. 2015; Krauss et al. 2017). It thus seems likely that traits that increase pollen dispersal distances in animal-pollinated species may not necessarily be related to increased male reproductive success in terms of numbers of progeny sired, as we have found here (see Chen and Pannell (2023b) for the

estimates of selection gradients via siring success). Nonetheless, traits facilitating pollen dispersal may still be favored as a result of enhanced *quality* of offspring, given the somewhat elevated relatedness of *P. alpina* individuals that are less than 3 m apart and the high level of inbreeding depression estimated for the species (unpublished data).

Declarations

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Author contribution

K.-H.C.: conceptualization, data curation, formal analysis, investigation, methodology, software, visualization, writing—original draft, writing—review and editing; J.R.P.: conceptualization, funding acquisition, project administration, resources, supervision, writing—original draft, writing—review and editing.

All authors gave final approval for publication and agreed to be held accountable for the work performed therein.

Data availability

The datasets generated and analysed during the current study will be available in the online repository Zenodo after acceptance.

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<https://doi.org/10.1111/JSE.12960>

Tables

Table 1

Summary of studies reporting direct estimates of pollen dispersal distances in alpine plants. References:

a. Petré et al. (2021) b. Buehler et al. (2012) c. Scheepens et al. (2012) d. gisdóttir et al. (2009) e. Thomson and Thomson (1989) f. Pluess and Stöcklin (2004) g. Campbell and Waser (1989) h. Chen and Pannell (2022) i. This study j. Matter, Kettle, Ghazoul and Pluess (2013) k. Matter, Kettle, Ghazoul, Hahn, et al. (2013)

Species	Pollination system	Mean pollen dispersal distance (m)	Methods used to estimate dispersal distance	Traits studied for within-population variation
<i>Arabis alpina</i>	Generalized (fly, bee) ^a	< 50 ^b	Paternity analysis ^b	
<i>Campanula thyrsoidea</i>	Bumblebee ^c	17.4 ^c 34-62.1 ^c 4.85 ^d	Paternity analysis ^c , Pollen dyes ^{c,d}	Density ^c
<i>Erythronium grandiflorum</i>	Bumblebee ^e	0.68-16.44 ^e	Pollen dyes ^e	
<i>Geum reptans</i>	Fly ^f	< 1 ^f	Pollen dyes ^f	
<i>Ipomopsis aggregata</i>	Hummingbird ^g	1.41-2.63 ^g	Pollen dyes ^f	Phenology, floral morphology ^g
<i>Pulsatilla alpina</i>	Fly ^h	3.17 ⁱ	Paternity analysis ⁱ	Floral gender, phenology, floral morphology ⁱ
<i>Ranunculus bulbosus</i>	Generalized (fly, beetle, bee) ^j	109.5-296.5 ^{j,*} 15.9 ^k	Inferred from fitted dispersal kernel ^j , Paternity analysis ^k	
<i>Trifolium montanum</i>	Bees ^j	3490.6 ^{j,*} 10.3 ^k	Inferred from fitted dispersal kernel ^j , Paternity analysis ^k	

Note: * The extremely long pollen dispersal distance reported is likely due to a non-exhausted sampling of the sires.

Table 2

Estimates of parameters for the Weibull distribution pollen dispersal kernel for all the sires (A), single-flowered hermaphrodites (B), and single-flowered males (C).

	a (upper CI, lower CI)	b (upper CI, lower CI)	Mean (SD) (m)	Skewness	Kurtosis
A. All sires	3.26 (2.99, 3.54)	1.07 (1, 1.15)	3.17 (2.95)	1.8	7.73
B. Sires with a bisexual flower	3.33 (3.00, 3.70)	1.09 (1, 1.19)	3.22 (2.96)	1.75	7.47
C. Sires with a male flower	3.28 (2.58, 4.14)	0.92 (0.78, 1.08)	3.41 (3.69)	2.26	10.85

Table 3

Partial effects of floral gender and floral traits on pollen dispersal distances in single-flowered individuals (N = 297 mating events from 72 sires). Only outcrossing mating events were included in the analysis.

	Estimate	Standard error	t-value	P-value
(Intercept)	1.15	0.06	20.12	***
Floral gender	-0.08	0.14	-0.58	n.s.
Petal length	-0.19	0.05	-4.05	***
Stalk height	0.13	0.06	2	*
Flowering date	-0.21	0.07	-3.2	**

Figures

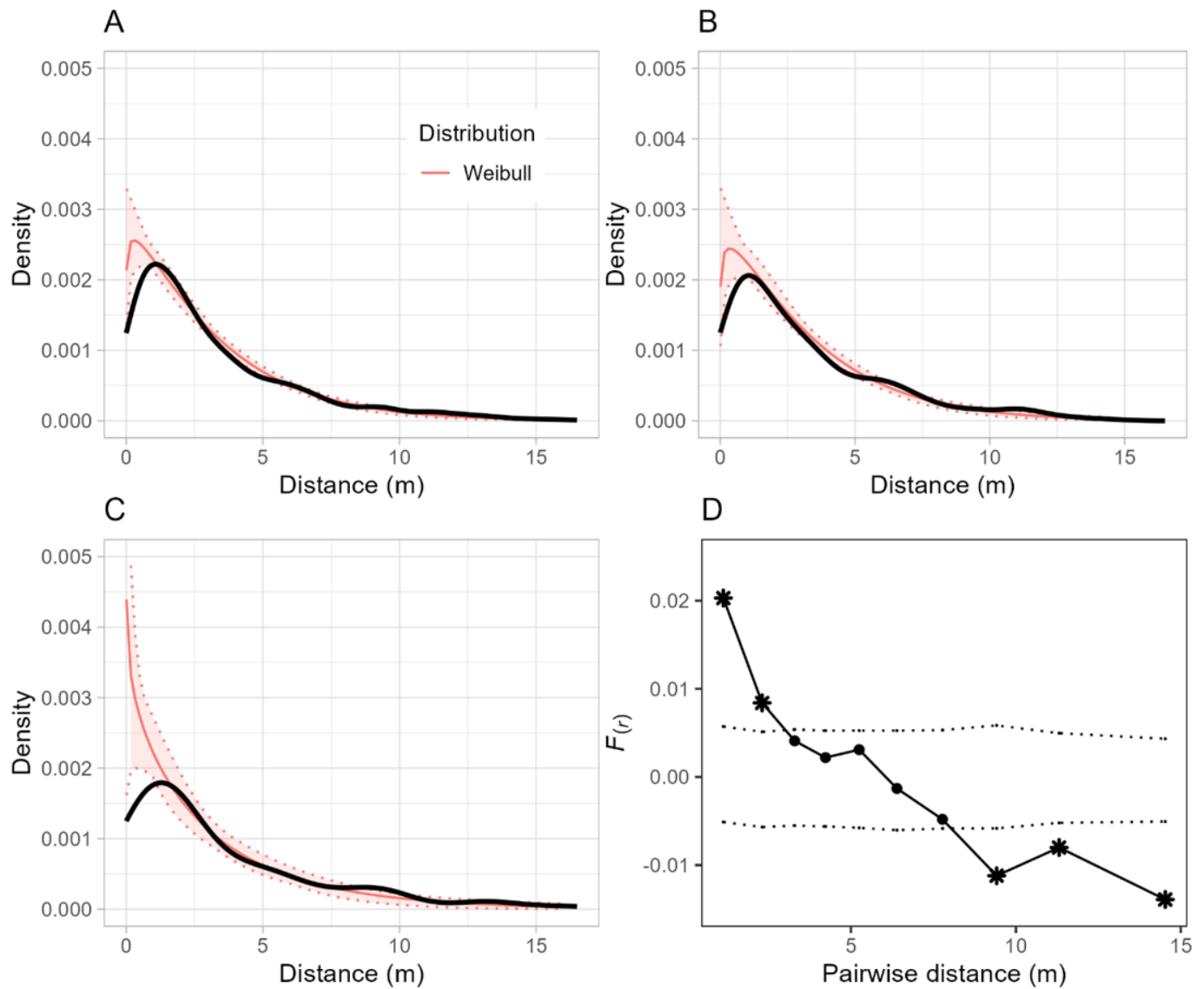


Figure 1

Pollen dispersal kernels for all sires (A), for sires with a single bisexual flower (B), and for sires with a single male flower (C). Black lines represent the density distribution of raw data. Red lines represent the best-fitted kernel with a Weibull distribution and dotted red lines show the 95% confidence intervals of the distribution. Only outcross mating events were included in the analysis. (D) Results of a spatial autocorrelation analysis based on all flowering individuals ($N = 138$), and on the use of Nason's kinship coefficients ($F_{(r)}$). The average kinship coefficients (marked as dots) are positioned along the x-axis at the mean pairwise distance within each of the ten distance classes. The average $F_{(r)}$ values deviating significantly from expected for a random distribution of genotypes are marked with an asterisk. The dotted lines represent 95% confidence intervals.

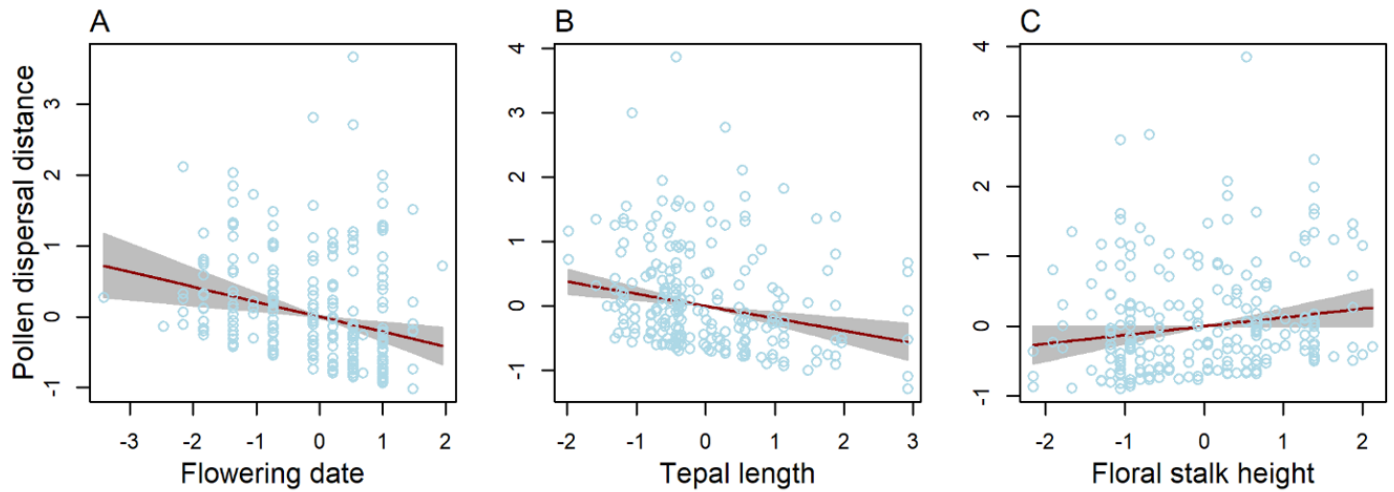


Figure 2

Partial effects of floral traits on pollen dispersal distances of single-flowered individuals ($N = 297$ outcross mating events from 72 sires). Floral traits are all standardized. The mating events are represented by blue dots. The 95% confidence interval of the estimates is shown around the regression lines.

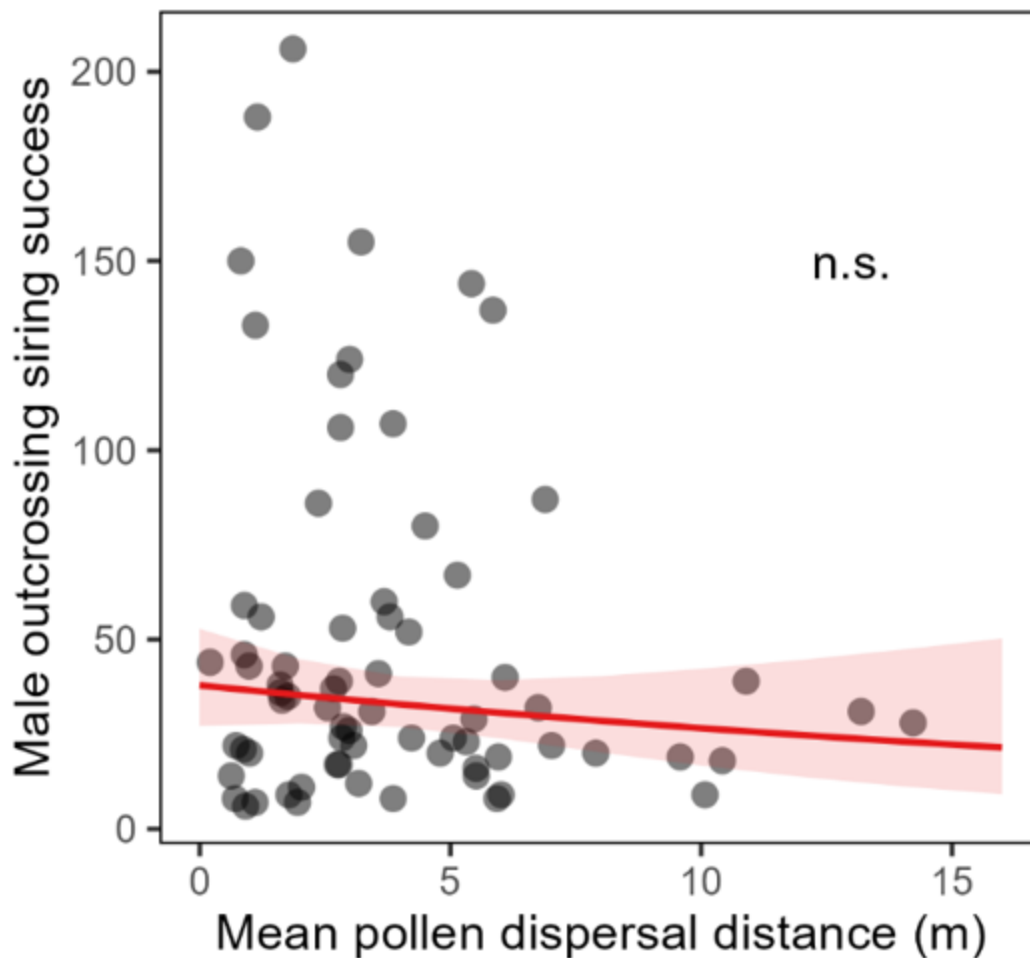


Figure 3

Absence of a significant relationship between mean pollen dispersal distances and male outcross siring success of individuals evaluated using a generalized linear model ($N= 72$; $P > 0.05$). Each point represents one individual. The 95% confidence interval of the estimates is shown around the regression line.

Supplementary Files

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