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GEOGRAPHIC STRUCTURE OF POLLINATOR COMMUNITIES, REPRODUCTIVE ASSURANCE, AND THE EVOLUTION OF SELF-POLLINATION

David A. Moeller[1]

Department of Ecology and Evolutionary Biology, Cornell University, Ithaca, New York 14853 USA

Abstract. Reproductive assurance is often invoked as an explanation for the evolution of self-fertilization in plants. However, key aspects of this hypothesis have received little empirical support. In this study, I use geographic surveys of pollinator communities along with functional studies of floral trait variation to examine the role of pollination ecology in mating system differentiation among populations and subspecies of the annual plant *Clarkia xantiana*. A greenhouse experiment involving 30 populations from throughout the species' range indicated that variation in two floral traits, herkogamy and protandry, was closely related to levels of autofertility and that trait variation was partitioned mainly among populations. Emasculation experiments in the field showed that autonomous selfing confers reproductive assurance by elevating fruit and seed production. Surveys of pollinator communities across the geographic range of the species revealed that bee pollinator abundance and community composition differed dramatically between populations of the outcrossing subspecies *xantiana* and the selfing subspecies *parviflora* despite their close proximity. Specialist bee pollinators of *Clarkia* were absent from selfing populations, but they were the most frequent visitors to outcrossing populations. Moreover, within the outcrossing subspecies *xantiana*, there was a close correspondence between specialist abundance and population differentiation in herkogamy, a key mating system trait. This spatial covariation arose, in part, because geographically peripheral populations had reduced herkogamy, higher autofertility, and lower pollinator abundance compared to central populations of *xantiana*. Finally, I detected strong spatial structure to bee communities both across the range of the species and within the outcrossing subspecies. In both cases, spatial structure was stronger for specialist bees compared to generalist bees, and pollinator communities varied in parallel with population variation in herkogamy. These results provide evidence that mating system differentiation parallels spatial variation in pollinator abundance and community composition at both broad and more restricted spatial scales, consistent with the hypothesis that pollinator abundance and reproductive assurance are important drivers of plant mating system evolution.

Key words: autonomous self-fertilization; central and peripheral populations; Clarkia xantiana; *floral evolution; herkogamy; mating system; protandry; reproductive assurance; solitary bees; spatial variation; specialist pollinators.*

INTRODUCTION

The nature and intensity of species interactions often vary among populations (Thompson 1988); either because community composition varies locally or because the geographic ranges of interacting species do not completely overlap. Shifts in species interactions not only influence population dynamics in the short term, but also may lead to adaptive differentiation in traits that mediate species interactions (e.g., Carroll and Boyd 1992, Stachowicz and Hay 2000, Brodie et al. 2002). Integrating studies of species interactions and character variation in a spatial context has revealed new insights into the interplay of ecological and evolutionary processes (Travis 1996).

In flowering plants, the diversification of floral form has often been attributed to pollinator-mediated selection (e.g., Grant and Grant 1965, Stebbins 1970). From this perspective, adaptive evolution is expected to occur in response to changes in the abundance and/or identity of effective pollinators. One common observation is that plant species vary in floral traits across their geographic ranges (e.g., Johnson 1997, Jonas and Geber 1999, Mazer and Meade 2000). Although it is clear that pollinator preferences can maintain floral differences between closely related species (Fulton and Hodges 1999, Schemske and Bradshaw 1999), the role of pollinators in driving intraspecific differentiation in floral form remains less well understood. Evidence of spatial covariation between floral traits and pollinator identity comes mainly from species that exhibit variation in corolla size, color, or nectar spur length (Miller 1981,

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[1] Present address: Department of Plant Biology, University of Minnesota, Saint Paul, Minnesota 55108 USA. E-mail: moell021@umn.edu

Robertson and Wyatt 1990, Johnson 1997). These results suggest that geographic variation in key secondary sexual characters, which influence pollinator attraction or pollen transfer, can arise from differences in pollinator communities and pollinator-mediated selection.

Apart from floral evolution driven by different pollinator taxa or functional groups of pollinators, a major trend in flowering plant diversification is the evolution of self-fertilization or agamospermy, where plants no longer rely on vector-mediated pollen transfer (Jain 1976, Richards 1997, Holsinger 2000). In particular, the shift from outcrossing to autonomous self-pollination has received a great deal of attention from evolutionists because it has occurred repeatedly across many clades of flowering plants (Stebbins 1974) and has important consequences for population genetic structure (Hamrick and Godt 1996) and the adaptive potential of populations (reviewed in Takebayashi and Morrell 2001). Two major selective forces have been invoked to explain the evolution of self-fertilization: the transmission advantage of selfing over outcrossing genotypes and the reproductive assurance value of selfing in poor pollination environments (reviewed in Uyenoyama et al. 1993, Holsinger 2000, Goodwillie et al. 2005). This 50% transmission advantage of selfers depends on the assumption that pollinators are reliable such that selfing variants act as male parents for their own selfed seeds as well as the seeds of other plants; whereas reproductive assurance favors the evolution of selfing in regions where pollinator service is uncertain or chronically insufficient (Darwin 1859, Henslow 1879, Müller 1883). Therefore, the relative importance of these two forces in driving the evolution of selfing depends strongly on the pollination environment and leads to contrasting predictions about the relationship between population variation in mating system and pollinator abundance.

Theoretical models have demonstrated that pollen limitation due to poor or variable pollination environments can select for high selfing rates principally because the reproductive assurance value of selfing minimizes the ability of inbreeding depression, pollen discounting, and seed discounting to maintain outcrossing (Morgan and Wilson 2005, Porcher and Lande 2005). Field experiments have provided supporting evidence that self-fertilization often augments seed production (e.g., Goodwillie 2001, Herlihy and Eckert 2002, Elle and Carney 2003, Kalisz and Vogler 2003) and that selection strongly favors reduced mating system traits in environments where plant populations experience strong pollen limitation (Moeller and Geber 2005). It has recently been emphasized that assessing the role of ecological factors in mating system evolution may benefit from a geographical perspective (Barrett et al. 2001). For example, the reproductive assurance hypothesis predicts that population variation in mating system should covary with the long-term abundance of effective pollinators. In addition, observations that selfing populations frequently occur at the geographic periphery of the ranges of the species (e.g., Jain 1976, Wyatt 1988, Guerrant 1989) suggest that pollinator abundance may be lower in peripheral compared to central populations in parallel with mating system variation. Although there are many accounts of geographic variation in mating system traits, pollinator communities have rarely been characterized in sufficient detail over large spatial scales to evaluate these hypotheses rigorously. Here, I use a biogeographic approach involving pollinator surveys across the range of an annual plant, *Clarkia xantiana*, along with an experimental assessment of the functional significance of mating system trait variation to examine the role of pollination ecology in the evolution of self-pollination. Studies presented in this paper build on previous work on *C. xantiana* by Fausto et al. (2001), which suggested that pollinator abundance and reproductive assurance may play an important role in mating system differentiation.

Clarkia xantiana A. Gray (Onagraceae) is a winter annual endemic to southern California, USA, with the majority of populations occurring in the southern Sierra Nevada foothills. Populations exhibit substantial variation in floral morphology across a small geographic range (Moore and Lewis 1965, Eckhart and Geber 1999). The species is composed of two subspecies, *C. xantiana* ssp. *xantiana* and *C. xantiana* ssp. *parviflora* (Eastw.) Harlan Lewis (hereafter *xantiana* and *parviflora*), which have a parapatric distribution (Eckhart and Geber 1999). Subspecies *xantiana* is large flowered with herkogamy (spatial separation of anthers and stigma) and protandry (temporal precedence of anther dehiscence over stigma receptivity), whereas subspecies *parviflora* is small flowered with reduced herkogamy and protandry (Runions and Geber 2000). Mating system traits (herkogamy and protandry) are strongly correlated with outcrossing rates and levels of allozyme polymorphism in *Clarkia* species (Vasek 1965, Vasek and Harding 1976, Gottlieb 1984, Holtsford and Ellstrand 1992; M. A. Geber, *unpublished data*). Flower visitors to most *Clarkia* species are bees (Superfamily Apoidea: Series Apiformes [Goulet and Huber 1993]) including generalist foragers and at least 10 species of pollen specialists (MacSwain et al. 1973, Fausto et al. 2001). *Clarkia* specialist bees exclusively provision their nests with *Clarkia* pollen (MacSwain et al. 1973) and are effective in pollen removal and pollen transfer to stigmas (Eckhart et al. 2006; see Plate 1, Appendix F). Subspecies *xantiana* shares bee pollinators with other outcrossing *Clarkia* species while congeners are absent from most of *parviflora*'s range (Moeller and Geber 2005).

I used field and greenhouse studies to test predictions of the reproductive assurance hypothesis. First, I examined variation in mating system traits in 30 populations of *Clarkia xantiana* from across the geo-

PLATE 1. Many solitary bee pollinators of *Clarkia xantiana*, such as *Hesperapis regularis* (left), forage exclusively on species in the genus *Clarkia*. These bees have specialized scopae (middle), composed of sparse and unbranched hairs, which accommodate the large and often clumped pollen grains of *Clarkia* (right). A color version of Plate 1 is available in Appendix F. Photo credit: D. Moeller.

graphic range of the species and quantified the effect of morphological variation on rates of autonomous self-pollination in the absence of pollinators (autofertility). Second, I performed floral manipulations in highly selfing natural populations to test the hypothesis that self-fertilization provides reproductive assurance by elevating fruit and seed production. Third, I conducted surveys of bee pollinator assemblages in 15 populations spanning the species' range to (1) test for covariation between population differentiation in mating system traits and pollinator abundance and community composition, (2) test for spatial structure to specialist and generalist pollinator communities and its relationship to mating system variation, and (3) test the hypothesis that peripheral populations have reduced pollinator abundance and reduced mating system traits relative to central populations.

MATERIALS AND METHODS

Experiment 1: floral trait variation and autofertility

I used a greenhouse experiment to examine the distribution of floral trait variation within and among populations and its relation to levels of autofertility. The study included plants from 30 *Clarkia xantiana* populations distributed throughout the range of the species (15 populations of each subspecies). Five plants per population, each from a different maternal family, were grown in a completely randomized design under highly controlled conditions such that pollen transfer could not occur between flowers. Two floral traits, herkogamy (anther to stigma distance) and protandry (the temporal precedence of anther dehiscence over stigma receptivity), were measured for two flowers on each plant. The development of individual flowers was followed for one to seven days to measure protandry. Autofertility (rate of autonomous selfing in the absence of pollinators) was measured based on (1) the percentage of open-pollinated flowers on the whole plant that set fruit, and (2) the ratio of seed set from two open-pollinated flowers to the seed set from two hand-pollinated flowers. Hand pollinations were performed using large pollen loads (>500 pollen grains) from two unrelated pollen parents of the same subspecies. Treatments were randomly assigned to four flowers on each plant. I tested for differences in mating system traits and autonomous selfing rates between subspecies and among populations within subspecies using ANOVA. I examined the relationship between autofertility and quantitative variation in mating system traits using linear regressions both across and within subspecies.

Experiment 2: does autonomous selfing provide reproductive assurance?

I tested the hypothesis that self-pollination confers reproductive assurance using emasculation experiments in two populations of *parviflora* (Populations 11 and 12; Appendix A). The populations studied are the two westernmost *parviflora* populations shown in Fig. 1. I haphazardly selected plants distributed along four transects throughout each population ($N = 40$) and performed bud emasculations to excise the stamens from one flower prior to anthesis. A second flower bud on each plant was marked prior to anthesis but not manipulated. Flowers used for the two treatments were adjacent to one another on the main stem of the plant, and the position of these treatments on the stem was rotated among plants. For each population, I examined the effects of pollination treatments on fruit set (fruits per flower) using a χ^2 test and on seed set (seeds per flower) using a paired t test. Because one flower per plant was emasculated, reproduction in emasculated flowers may have been effected by two pollinator-mediated modes of reproduction, outcrossing or geitonogamous (between flower) selfing. For *parviflora* plants, however, the likelihood of geitonogamous selfing is low because floral displays typically consist of one or few flowers and pollinators infrequently forage on multiple flowers within an inflorescence (Moeller and Geber 2005). Floral displays tend to be small because flowers readily self-fertilize upon anthesis, which reduces floral longevity and the temporal overlap among flowers in an inflorescence.

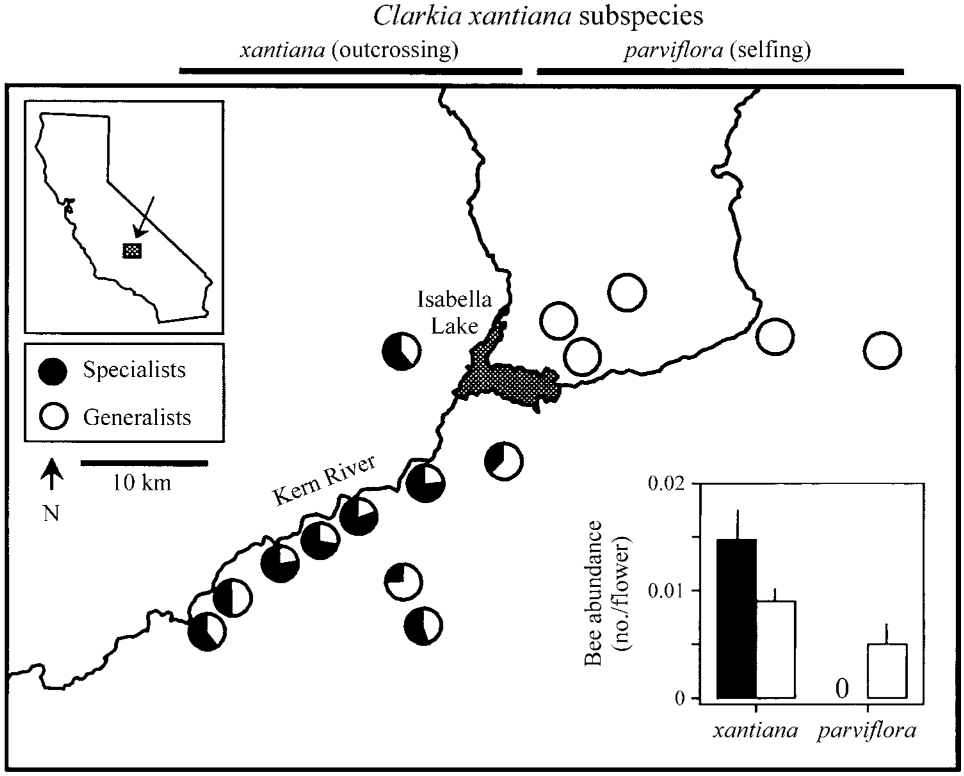

FIG. 1. Map of the distribution of *Clarkia xantiana* populations surveyed for pollinators in this study. Populations are indicated by pie diagrams showing the proportions of bees collected in each population that represent *Clarkia* specialist vs. generalist foragers. Populations to the west of Isabella Lake belong to subspecies *xantiana* (predominantly outcrossing), and those to the east belong to subspecies *parviflora* (predominantly selfing). The inset chart shows a comparison of specialist and generalist bee abundance (mean + SE) for populations of the two subspecies (no. bees collected per flower per 35-min observation period). For subspecies *xantiana*, geographically central populations are the five westernmost populations, and peripheral populations are the five easternmost populations.

I tested whether the bud emasculation procedure directly influenced reproductive success using a greenhouse experiment. One flower per plant was emasculated and subsequently hand pollinated and a second flower was hand pollinated but otherwise unmanipulated. Seed set (seeds per flower) did not differ significantly between the two treatments suggesting that the bud emasculation procedure does not directly reduce female fitness (paired t test: $N = 64$, $t = 0.21$, $P = 0.834$). In the field, it is possible that anther removal also has an indirect influence on reproduction by discouraging pollinator visitation. Although I have not tested for this possible effect, generalist and specialist bees frequently visit female-phase flowers that no longer contain pollen (*personal observation*).

Geographic surveys of bee visitor assemblages

I conducted standardized surveys of bee visitors to 10 populations of *xantiana* and five populations of *parviflora* distributed from the western to the eastern margin of the range of the species (Fig. 1; Appendix A). All populations were distinct colonies of potentially interbreeding plants and were medium to large in size (>1400 plants). Population surveys were conducted on 30 full days from 4 May to 7 July 2001. Within each population, collections were made according to a stratified random sampling protocol, with populations of *C. xantiana* divided into four sections of equal size and collections made along a 35 × 2 m belt transect spanning each section. I estimated plant and flower density by recording the number of plants and flowers observed in four plots (2 m^2) randomly placed along each of the transects. Bees were the only visitors observed in this study and in previous studies of *C. xantiana* (Fausto et al. 2001, Moeller 2004, Moeller and Geber 2005).

I sampled visitor communities at each site for two full days during the peak of flowering at a site; sampling days were four to six days apart. Collections were made during four sampling periods early in the day (0800–1100) and four sampling periods later in the same day (1100–1400), spanning the main period of bee visitation to *Clarkia* (MacSwain et al. 1973). During each 35-min sampling period, I repeatedly walked one transect and collected all flower visitors that made contact with *C. xantiana* flowers. Therefore, each transect was sampled early and late on each of two days. In total, collections were made during 16 sampling periods over nine hours and 20 minutes of collection time per site. Voucher specimens for bee species collected were deposited in the Cornell University Insect Collection (Voucher Lot no. 1244).

Pan trapping was employed as a second method to verify whether *Clarkia* specialist bees, the primary pollinators of *C. xantiana* (MacSwain et al. 1973), were present or absent from a site, independent of whether these specialists were found foraging on *C. xantiana*. I placed 10 pan traps (355-mL plastic bowls) of two colors that are effective at attracting bees (five each of blue and white) at 3-m intervals along a transect spanning each site. Pan trap colors were alternated along transects. Traps were filled with a dilute soap solution and placed into each site during the second day of collection.

Statistical analyses

Mating system trait variation and bee abundance.—The relationship between bee pollinator abundance and mating system variation was examined hierarchically by (1) testing for differences in pollinator abundance between populations of *xantiana* and *parviflora* and (2) testing for covariation between pollinator abundance and population variation in floral traits within subspecies. Analyses within subspecies were limited to the more variable *xantiana* because bee abundance was very low in all populations of *parviflora* (see *Results: Mating system variation and patterns of bee abundance*). Bee abundance was expressed as the number of bees collected per flower per observation period. All analyses were conducted on overall bee abundance as well as separately for specialist and generalist foragers. I tested for differences between subspecies using an ANCOVA that included two covariates, site elevation and mean plant density across each population. Within *xantiana*, I tested for a relationship between bee abundance and population variation in mating system traits using multiple regressions of bee abundance per flower on mean herkogamy, mean protandry, and the covariates elevation and plant density. Detailed data on mating system traits were available for eight of the 10 *xantiana* populations from a second greenhouse study (M. A. Geber and D. A. Moeller, *unpublished data*). For each population, 20–30 individuals, each from a different maternal family, were grown in a completely randomized design. Herkogamy and protandry were measured as described in Experiment 1. Finally, I tested the hypothesis that pollinator abundance was lower in geographically peripheral, compared to central, populations of *xantiana*. Peripheral populations are those at the eastern and northern edge of the range where *xantiana* overlaps in distribution with *parviflora* and where *xantiana* populations become increasingly sparse across the landscape.

Mating system variation and bee community composition.—I tested for a relationship between bee community composition and mating system variation using a series of multivariate analyses (ter Braak 1995). First, I conducted a detrended correspondence analysis (DCA) to identify the dominant axes of variability in community composition and subsequently to examine correlations between axis scores and the three predictor variables, mating system (*xantiana* vs. *parviflora*), elevation, and plant density. Second, I used canonical correspondence analysis (CCA) to test explicitly for a relationship between gradients in bee community composition and predictor variables. Conducting both analyses allows one to check that the major gradients in composition have been identified (DCA) and that the portion of the variability explained best by predictor variables has been extracted (CCA; ter Braak 1995). Third, to separate the effects of independent variables, I conducted three partial CCAs in which the significance of each predictor was tested while controlling for the remaining factors. For all CCAs, statistical significance was determined using Monte Carlo tests with 1000 permutations (CANOCO, Version 4; ter Braak and Smilauer 1998). Species abundance was $\log_{10}$-transformed and predictor variables were standardized to zero mean and unit variance.

Spatial structure and scale of bee community variation.—I tested for spatial structure to bee community variation by examining the relationship between bee community similarity and geographic distance for pairs of plant populations. Tests were conducted across both subspecies and separately for populations of *xantiana* alone. Pairwise geographic distance was calculated using latitude and longitude coordinates. Pairwise similarity between bee communities was calculated in EstimateS, version 6.0 (Colwell 1997), using the Sørenson qualitative and quantitative similarity indices. The Sørenson qualitative similarity index is based only upon the presence vs. absence of species. The quantitative version of the Sørenson index weights the presence of species by abundance. Both indices equal 1.0 when communities are identical. Comparison of a qualitative index to a quantitative index allows for a more detailed understanding of the nature of variation in community composition by separating correspondence between sites in species occurrence from the relative abundance of species. Tests of significance for relationships between community similarity and geographic distance matrices were conducted using Mantel tests with 1000 randomizations (R package, Legendre and Vaudor 2001). I tested for differences in spatial structure between specialist and generalist bee communities using ANCOVAs including pairwise community similarity as the dependent variable and geographic distance, specialist/generalist, and their interaction as independent variables. Geographic distance was natural log-transformed for analyses across both subspecies to satisfy ANOVA assumptions.

TABLE 1. ANOVAs testing for subspecific and population-level effects on pollinator abundance.

		Overall bee abundance		Specialist abundance		Generalist abundance	
Source of variation	df	*F*	*P*	*F*	*P*	*F*	*P*
A) Both subspecies							
Mating system	1,11	27.77	<0.001	17.24	0.002	6.16	0.030
Elevation	1,11	0.75	0.404	2.74	0.126	1.79	0.208
Plant density	1,11	2.31	0.157	0.59	0.460	2.14	0.171
B) Subspecies *xantiana*							
Herkogamy	1,3	33.03	0.010	16.45	0.027	0.01	0.921
Protandry	1,3	0.02	0.906	0.96	0.398	5.09	0.109
Elevation	1,3	0.03	0.875	0.64	0.483	4.02	0.139
Plant density	1,3	0.40	0.572	0.94	0.403	1.29	0.338
C) Subspecies *xantiana*							
Central vs. peripheral	1,6	8.93	0.024	3.35	0.117	1.26	0.305
Elevation	1,6	3.32	0.118	0.36	0.573	2.74	0.149
Plant density	1,6	0.56	0.482	0.33	0.584	0.00	0.977

Notes: (A) Differences in pollinator abundance between *Clarkia xantiana* subspecies *xantiana* and subspecies *parviflora*; (B) covariation between pollinator abundance and population variation in mating system traits, herkogamy and protandry, within subspecies *xantiana*; and (C) differences in pollinator abundance between central and peripheral populations of subspecies *xantiana*.

RESULTS

Floral trait variation and autofertility

Herkogamy and protandry were significantly greater in populations of *xantiana* compared to *parviflora* (herkogamy, 3.29 ± 1.61 mm vs. 0.32 ± 0.41 mm [mean ± SE]; protandry, 3.93 ± 1.16 days vs. 0.19 ± 0.68 days), and both traits differed significantly among populations within subspecies (Appendix B). Among-population variation in herkogamy and protandry accounted for 79.5% and 88.0% of the total variation among individuals of both subspecies and accounted for 45.0% and 52.6% of variation within *xantiana* and 67.9% and 36.2% of variation within *parviflora*, respectively. In addition, geographically peripheral *xantiana* populations had significantly reduced herkogamy ($F_{1,13} = 22.13$, $P < 0.001$) but not protandry ($F_{1,13} = 1.01$, $P = 0.33$) compared to central populations (Appendix D).

Autofertility, as measured by percentage fruit set (fruits per flower), was considerably higher in *parviflora* (mean, 93.1%; range, 58.3–100%) than *xantiana* (mean, 9.7%; range, 0.0–40.0%) and differed significantly among populations within subspecies (Appendix B). There was strong evidence of covariation between autofertility and quantitative variation in herkogamy and protandry. For individuals combined across all 30 popuations, percentage fruit set declined with increasing herkogamy ($P < 0.0001$) and protandry ($P < 0.0001$). Within both subspecies, there was also evidence that percentage fruit set declined with increasing herkogamy (*xantiana*, $P = 0.016$; *parviflora*, $P = 0.009$) and protandry (*xantiana*, $P < 0.001$; *parviflora*, $P = 0.021$). Analyses based on the ratio of unmanipulated to hand-outcrossed seed set (seeds per flower) produced very similar results across subspecies and qualitatively similar, but marginally significant, results within subspecies. Whereas fruit set was recorded for all flowers on a plant (35.5 ± 1.6 flowers/plant [mean ± SE]), seed set was estimated for each treatment based on two flowers per plant resulting in more variability in the data set and lower statistical power, particularly for regressions within subspecies. The complete results of regression analyses are reported in Appendix C. Last, geographically peripheral populations of *xantiana* had significantly higher rates of autonomous selfing compared to central populations based on both fruit and seed set (fruit set, $F_{1,13} = 4.55$, $P = 0.05$; seed set, $F_{1,13} = 7.25$, $P = 0.02$; Appendix D).

Reproductive assurance

All unmanipulated flowers produced fruits, while significantly fewer emasculated flowers developed into fruits in both populations (Population 11, 57% fruit set for emasculated flowers, $\chi^2 = 19.5$, $P < 0.0001$; Population 12, 67% fruit set, $\chi^2 = 8.5$, $P < 0.005$). There was a greater difference between treatments in seed set (seeds per flower) where unmanipulated flowers produced 2.5 and 1.9 times as many seeds as emasculated flowers in Population 11 and 12, respectively (Population 11, $t_{32} = 7.9$, $P < 0.0001$; Population 12, $t_{30} = 4.8$, $P < 0.0001$). The greater difference between treatments in seed set vs. fruit set was most likely because many fully developed fruits from emasculated flowers contained few seeds (*personal observation*).

Mating system variation and patterns of bee abundance

The pollinator survey across 15 populations resulted in a collection of 1410 individuals from 57 bee species (Appendix E). All bees were native to western North America with the exception of four honey bees (*Apis mellifera*). The mean abundance of bees per population was 16.5 times higher in *xantiana*, and bee abundance (on a per-flower basis) was 4.4 times higher in *xantiana* than in *parviflora* populations (Fig. 1). Mating system (*xantiana* vs. *parviflora*) was the only significant predictor of overall bee abundance when accounting for covariates (Table 1). Specialists were represented by 834 individuals overall (59%) and were common in all 10 *xantiana* populations (23–80% of visitors), but specialists

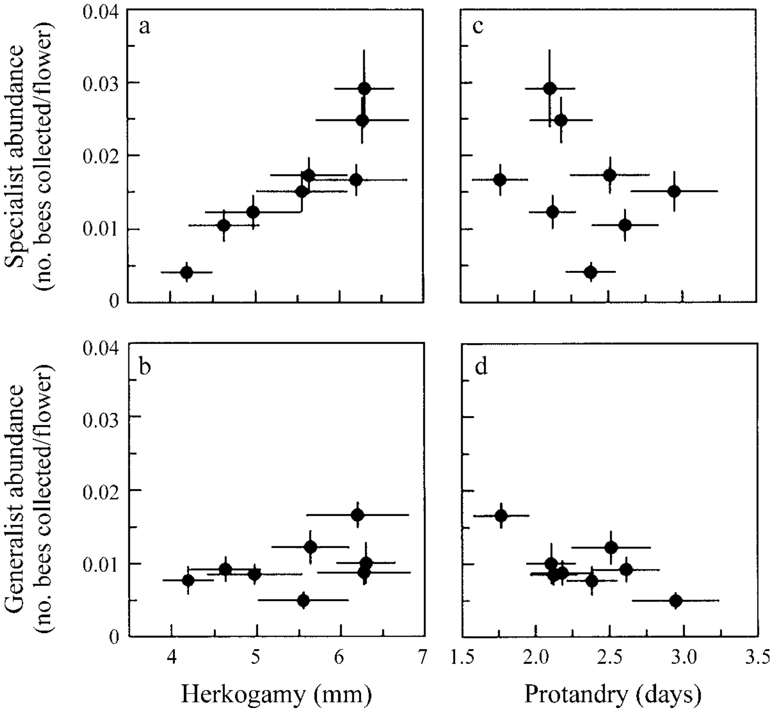

FIG. 2. The relationship between bee abundance (no. of bees collected per flower per 35-min observation period [mean ± SE], for specialists and generalists) and two floral traits, herkogamy and protandry (mean ± SE), for populations of subspecies *xantiana* (see *Materials and Methods: Statistical analyses: Mating system trait variation and bee abundance*).

were never collected in *parviflora* populations (Fig. 1, Table 1; Appendix E). Pan trap surveys confirmed that specialists were absent from *parviflora* populations, whereas specialists were abundant in pan traps from all 10 *xantiana* populations. The major specialist visitors to *xantiana* were *Hesperapis regularis* (Cresson) (see Plate 1, Appendix F), *Ceratina sequoiae* Michener, *Megachile gravita* Mitchell, and *Megachile pascoensis* Mitchell (Appendix E). The abundance of generalist bees was also significantly greater in *xantiana* populations, but to a lesser degree than specialists (Fig. 1, Table 1).

Within subspecies *xantiana*, there was a significant positive relationship between bee pollinator abundance and population variation in herkogamy, a key mating system trait (Table 1). This overall relationship was determined mainly by the abundance of specialists (Fig. 2, Table 1). I did not detect a significant relationship between generalist bee abundance and herkogamy. I also did not detect significant covariation between bee pollinator abundance (overall, specialist, or generalist) and population variation in protandry (Fig. 2, Table 1). Geographically peripheral populations of *xantiana* had significantly lower overall bee abundance compared to central populations, but specialist abundance did not differ significantly between regions (Table 1; Appendix D). The covariation between overall bee abundance and herkogamy as well as the difference in overall bee abundance between central and peripheral populations remain significant after using a Bonferroni correction to account for these two tests having been conducted on the same data set.

Mating system variation and patterns of bee community composition

The multivariate analyses detected strong gradients in bee community composition and identified mating system as the major factor explaining variability in the distribution of bee species among populations. High eigenvalues obtained from the unconstrained ordination (DCA) showed that important gradients in community

TABLE 2. Multivariate analyses of bee community composition in relation to mating system variation in *Clarkia xantiana*.

Analysis		Predictor variable	Covariable	λ_1	λ_2	λ_3	P
1	DCA	···	···	0.79	0.28	0.15	···
2	CCA	M, E, D	···	0.67	0.24	0.14	0.005
3	CCA	M	E, D	0.64	0.56	0.46	0.001
4	CCA	E	M, D	0.24	0.56	0.46	0.349
5	CCA	D	M, E	0.14	0.56	0.46	0.589

Notes: We used detrended correspondence analysis (DCA), canonical correspondence analysis (CCA), and partial CCA in which the significance of individual predictor variables was tested while controlling for other covariables. The three predictor variables in the analyses are (M) mating system (*xantiana* vs. *parviflora*), (E) elevation, and (D) plant density (plants/m^2). For each analysis, the first three eigenvalues (λ) are shown along with P values from Monte Carlo tests of the significance of the first ordination axis (from CCA).

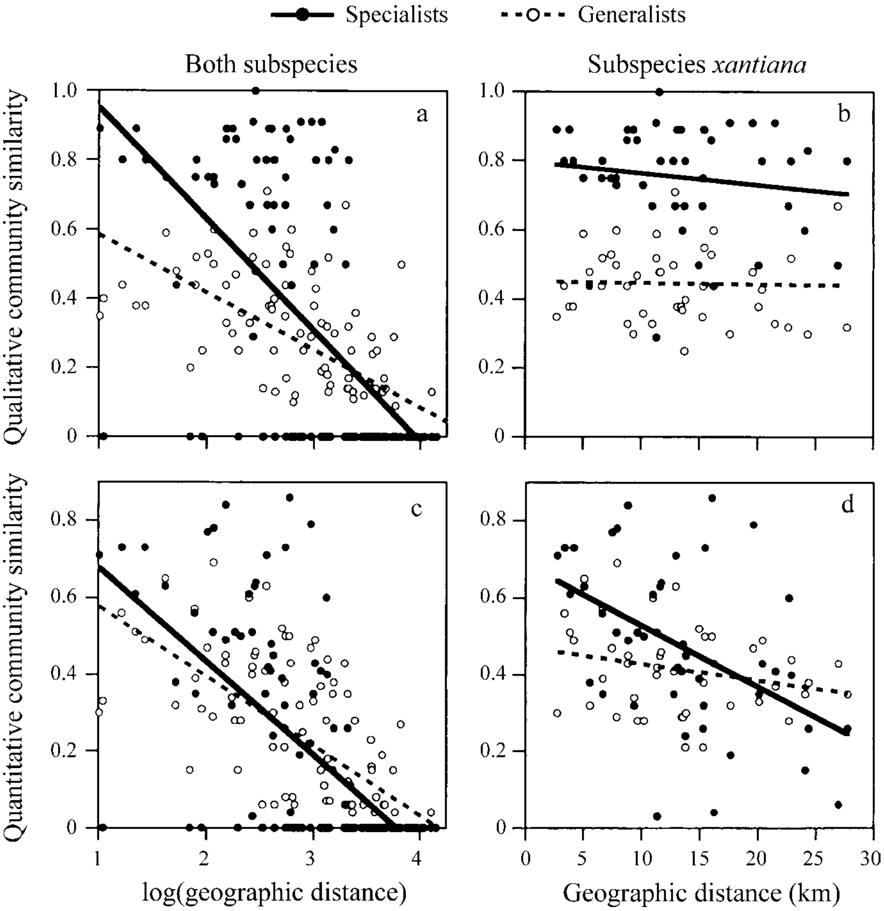

FIG. 3. Spatial structure of bee community composition exhibited as the relationship between bee community similarity and geographic distance for pairs of plant populations (a and c) across the range of the species and (b and d) within the outcrossing subspecies *xantiana*. Similarity was measured using both qualitative (species presence vs. absence; a and b) and quantitative (species presence weighted by abundance; c and d) indices.

composition were extracted from the data set (Table 2). The first axis was strongly correlated with mating system ($r = -0.92$), but only weakly correlated with elevation and plant density ($r = 0.16$ and -0.31, respectively). The null hypothesis that bee community composition is independent of predictor variables was rejected using a Monte Carlo permutation test of the first canonical axis ($P = 0.005$) and all canonical axes ($P = 0.03$). The CCA with all three predictor variables (Analysis 2 on Table 2) revealed that the first axis was determined principally by mating system (canonical coefficient = –0.80), and partial CCAs (Analyses 3–5 on Table 2) confirmed that mating system was the only significant predictor of bee community composition. The second axis was correlated with elevation (canonical coefficient = 0.50), but the partial CCA indicated that elevation was not significantly associated with community composition (Table 2).

Spatial structure and scale of bee community variation

There was pronounced spatial structure to bee community composition across the geographic transect spanning the range of *C. xantiana*. Overall bee community similarity between populations decreased strongly with geographic distance using either qualitative or quantitative similarity indices, indicating a high rate of species turnover across a range of 63.6 km (Mantel tests: qualitative, $r = -0.67$, $P < 0.001$; quantitative, $r = -0.70$, $P < 0.001$). Spatial structure was stronger for specialist compared to generalist foragers using the qualitative index (Fig. 3a; specialist/generalist × ln distance, $F_{1,206} = 10.75$, $P = 0.001$). A similar pattern was found using the quantitative index, but the difference between slopes was only marginally significant (Fig. 3c; specialist/generalist × ln distance, $F_{1,206} = 3.12$, $P = 0.079$). These results indicate that, over a broad spatial scale, bee communities (particularly specialist communities) vary substantially in composition and in concert with mating system differentiation.

Within the range of *xantiana*, I did not detect a significant decline in similarity with distance based on the qualitative index; the pattern of bee species occurrence was largely constant across the range of

27.7 km (Fig. 3b; Mantel $r = -0.13$, $P = 0.217$). When species occurrence was weighted by abundance, however, there was evidence of spatial structure to bee community composition (Mantel $r = -0.43$, $P = 0.011$). As with analyses across both subspecies, spatial structure in relative abundance for *xantiana* populations was stronger for specialist compared to generalist foragers (Fig. 3d; specialist/generalist × distance, $F_{1,86} = 5.07$, $P = 0.027$). Finally, there was concordance between spatial structure in bee communities and in one mating system trait, herkogamy. Bee community similarity between populations declined as differences in herkogamy between populations increased (Mantel $r = -0.38$, $P = 0.044$). Community similarity was not significantly correlated with population differences in protandry (Mantel $r = 0.21$, $P = 0.218$). These results suggest that there is fine-scale parallel variation between bee communities and mating system.

Discussion

Recent theoretical and empirical studies have provided increasing support for the long-standing hypothesis that reproductive assurance often drives mating system evolution (Fishman and Wyatt 1999, Fausto et al. 2001, Kalisz et al. 2004, Moeller and Geber 2005). Traits that promote self-pollination may be favored under strong pollen limitation of reproduction because inbreeding depression becomes a less potent force opposing the selection of selfing (Morgan and Wilson 2005) and because the reproductive assurance value of selfing can have important demographic consequences for plant populations (Cheptou 2004, Morgan et al. 2005). The apparent importance of pollination ecology in the evolutionary dynamics of mating systems has led to the hypothesis that spatial variation in plant–pollinator interactions may explain broad-scale geographic patterns of mating system variation. Observations in some systems are consistent with this idea (e.g., Barrett et al. 1989, Inoue et al. 1996), but detailed tests of the hypothesis have not found evidence for it (Schmidt-Adam et al. 2000, Herrera et al. 2001, Eckert 2002, Molina-Freaner et al. 2003). In *Clarkia xantiana*, there is strong spatial covariation between population differentiation in mating system traits and the abundance and composition of pollinator communities at both broad and more restricted spatial scales. These results combined with evidence from emasculation experiments that selfing elevates female fitness are consistent with the hypothesis that reproductive assurance is an important force directing mating system evolution.

A greenhouse experiment revealed that two key mating system traits, herkogamy and protandry, differed considerably between *C. xantiana* subspecies and variation was partitioned largely among populations within subspecies. These patterns are consistent with previous field and greenhouse studies of floral morphology in *C. xantiana* (Eckhart and Geber 1999, Runions and Geber 2000) and argue for using a geographic perspective to evaluate the role of plant–pollinator interactions in mating system evolution. The strong covariation between mating system traits and the likelihood of fruit and seed set in a pollinator-free environment (autofertility) indicates that reduced herkogamy and protandry strongly promote autonomous selfing. Notably, mean autofertility was 9.5% for *xantiana* plants and 91.8% for *parviflora* plants (based on fruit set). Evidence that autofertility declines with herkogamy and protandry was also apparent within each of the subspecies, where the extent of phenotypic variation was reduced. Although realized selfing rates were not measured for natural populations, data from this and previous studies on *C. xantiana* suggest that rates of selfing in nature are influenced by variation in floral morphology and that self-pollination is largely autonomous. First, self-pollination is effected mainly at the onset of anthesis or shortly thereafter in *C. xantiana* (prior or competing selfing) rather than following opportunities for outcrossing (delayed self-pollination; Runions and Geber 2000). Second, previous work has shown that within-inflorescence bee movement is uncommon for most bee pollinators and therefore that rates of geitonogamous selfing are likely to be low (Moeller and Geber 2005). Third, herkogamy and protandry are closely related to outcrossing rates and allelic polymorphism among natural populations of *C. xantiana* and other *Clarkia* species (Vasek and Harding 1976, Gottlieb 1984, Holtsford and Ellstrand 1992) suggesting that these floral traits serve as suitable proxies for mating system variation in this system.

The importance of ecological context to the evolution of mating systems depends in part on the extent to which selfing provides reproductive assurance. Floral manipulations performed in two *parviflora* populations suggest that autonomous selfing strongly elevates female reproductive success. Emasculated flowers, which could set seed only via pollinator-mediated pollen transfer (outcrossing or geitonogamous selfing), produced considerably fewer fruits and seeds compared to unmanipulated flowers suggesting that seed production is limited by pollen receipt. In contrast to *parviflora*, previous work in natural populations of *xantiana* has shown that when pollinators are uncommon, plants often suffer from strong pollen limitation of reproduction (Moeller 2004). Moreover, the very low fruit and seed set of *xantiana* flowers in a pollinator-free greenhouse suggests that the capacity to self-fertilize is limited in most populations. Therefore, the effect of the pollination environment on reproductive success in *C. xantiana* depends upon the orientation and development of floral organs. That is, seed production in *parviflora* is buffered from poor or unpredictable pollination environments whereas seed production in *xantiana* varies with spatial and temporal variation in the abundance and identity of pollinators.

To evaluate explicitly the correspondence between pollinator abundance and population variation in mating system in *C. xantiana*, I conducted pollinator surveys in 15 plant populations spanning the geographic range of the species. Bee pollinator abundance was markedly lower in populations of the predominantly selfing *parviflora* compared to populations of the predominantly outcrossing *xantiana*. Populations of the two subspecies also differed in the composition of bee communities despite the small distance separating populations of contrasting mating system (<10 km). Especially notable is the absence of *Clarkia* pollen specialists from the range of selfing (*parviflora*) populations. These results are consistent with the finding of Fausto et al. (2001) of lower visitation rates in populations of subspecies *parviflora* and also agree with their preliminary characterization of bee communities. In addition, I have conducted less exhaustive surveys of bee visitors in two other years (1999, 2000) and failed to find specialists in selfing populations. Therefore, it appears that the results of this study are not a temporal anomaly; there is a consistent pattern of low bee pollinator abundance and an absence of *Clarkia* pollen specialists from these *parviflora* populations.

Fine-scale population sampling also allowed for tests of covariation between mating system and pollinator abundance within *xantiana*. I found a close correspondence between specialist bee abundance and population variation in herkogamy. Generalist abundance, however, exhibited little association with either mating system trait. Bee community composition was geographically structured over tens of kilometers and community composition varied in parallel with herkogamy. Spatial structure was caused mainly by changes in the composition of specialist communities across the geographic transect. Although individual specialist species were broadly distributed across the range of *xantiana*, specialist species varied considerably in relative abundance (significant spatial structure based on the quantitative similarity index). It is particularly interesting that measures of pollinator abundance and community composition were strongly associated with herkogamy but not protandry despite its potential influence on the likelihood of autonomous selfing. Consistent with results from this study, a field experiment showed that the strength of selection on herkogamy in experimental populations was significantly related to pollinator abundance, whereas this relationship was less apparent and not significant for protandry (Moeller and Geber 2005). The mechanisms underlying these different patterns for herkogamy and protandry are not clear, however, and require further study of floral development and its ecological consequences.

Spatial variation in plant–pollinator interactions has been found in a diverse array of systems (e.g., Herrera 1988, Horvitz and Schemske 1990, Fenster and Dudash 2001), but little is understood about the spatial structure and scale of variation. In addition, limited information on pollinator movement and pollinator perception of resources at broad spatial scales impedes our ability to identify the causes of spatial variation in plant–pollinator interactions (Bronstein 1995). Aside from their ecological implications, landscape-level studies may provide novel insights into the role of pollinator communities in generating intraspecific diversity in floral traits. In this study, the evidence for a relationship between community similarity and geographic distance at both a broad spatial scale (across the range of the species) and at a more limited spatial scale (within *xantiana*) suggests that plant–pollinator interactions exhibit clinal variation across *C. xantiana*'s populations rather than pronounced heterogeneity across the landscape. Highly structured variation in plant–pollinator interactions may facilitate adaptive divergence in ecologically important floral traits by virtue of limited gene flow among populations. Results from this study further imply that external ecological factors that vary in a heterogeneous manner over small spatial scales in this region (e.g., dominant vegetation and substrate) seem unlikely to explain the spatial gradient in bee pollinator abundance and community composition. Rather, pollinator population dynamics may respond to ecological factors varying over large spatial scales such as climate or the distribution of plant populations. Although it is currently only possible to speculate, preliminary surveys suggest that the frequency of *Clarkia* populations across the landscape (both *C. xantiana* and other pollinator-sharing *Clarkia* species) and the predictability of floral resources across years are lower in *parviflora*'s range, which may affect the persistence of *Clarkia* specialist pollinators. The population viability of less mobile pollinators, such as *Clarkia* specialist solitary bees, may be sensitive to the spatial and temporal dynamics of plant populations because their small foraging ranges and restricted dispersal may limit resource acquisition and colonization potential (Gathmann and Tscharntke 2002). Further detailed work on pollinator biology in this system and others is required to understand better the ecological factors governing pollinator population dynamics and distribution.

Although authors at least as early as Darwin recognized the potential importance of plant–pollinator interactions to the selection of selfing (Darwin 1859, Henslow 1879, Müller 1883), recent work has reinvigorated consideration of ecological context and reproductive assurance (e.g., Wyatt 1983, Fishman and Wyatt 1999, Herlihy and Eckert 2002, Kalisz et al. 2004, Moeller and Geber 2005). Data from this study demonstrate tight linkages between ecological factors and mating system over even small spatial scales. One often-cited hypothesis is that selfing taxa evolve in peripheral populations at the range margin of sister outcrossing taxa where pollinator and mate availability are predicted to be low (e.g., Jain 1976, Solbrig and Rollins 1977, Wyatt 1988, Herlihy and Eckert 2005). Consistent with this idea, peripheral populations of *xantiana* had reduced herkogamy, higher rates of autonomous selfing, and lower pollinator abundance compared to central populations. Together, the results of this study suggest that floral variation within *C. xantiana* represents evolutionary responses to variation in pollinator abundance and the reproductive assurance value of selfing. Reciprocal transplant studies between *xantiana*'s and *parviflora*'s range have also shown that selection by pollinators and mating system differences are important to adaptive differentiation (Geber and Eckhart 2005). I cannot exclude the possibility that variation in other selective factors (e.g., inbreeding depression) has resulted in population differences in mating system across *C. xantiana*'s range and in turn influenced the distribution and abundance of bee pollinator species. In this second hypothesis, increased reliance on selfing in plant populations may select for reduced investment in male function and secondary sexual characters (e.g., nectar production), which could affect pollinator resource acquisition and therefore the performance of pollinator populations. In either case, these results suggest an intimate interplay between ecological and evolutionary processes and illustrate the utility of examining species interactions in a geographic context.

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APPENDIX A

Latitude, longitude, and elevation of *Clarkia xantiana* populations used for the geographic survey of pollinator communities (*Ecological Archives* E087-090-A1).

APPENDIX B

ANOVAs testing for differences between subspecies and among populations within subspecies for mating system traits and autofertility (*Ecological Archives* E087-090-A2).

APPENDIX C

Results of linear regressions of fruit or seed set on individual variation in herkogamy and protandry across subspecies and within each subspecies (*Ecological Archives* E087-090-A3).

APPENDIX D

A figure comparing mating system traits, autofertility, and pollinator abundance between geographically central vs. peripheral populations of *Clarkia xantiana* ssp. *xantiana* (*Ecological Archives* E087-090-A4).

APPENDIX E

A complete list of the bee species collected on flowers of *Clarkia xantiana* along with their overall abundance and patterns of site occupancy (*Ecological Archives* E087-090-A5).

APPENDIX F

A color version of Plate 1 (*Ecological Archives* E087-090-A6).

Ecological Archives E087-090-A1

David A. Moeller. 2006. Geographic structure of pollinator communities, reproductive assurance, and the evolution of self-pollination. *Ecology* 87:1510–1522.

Appendix A (Table A1). Latitude, longitude, and elevation of *Clarkia xantiana* populations used for the geographic survey of pollinator communities.

Population	Location	Subspecies	Latitude	Longitude	Elevation (m)
1	Kern River Canyon	*xantiana*	35° 28.82N	118° 42.62W	457.2
2	Kern River Canyon	*xantiana*	35° 30.36N	118° 41.42W	550.7
3	Kern River Canyon	*xantiana*	35° 31.77N	118° 39.26W	673.6
4	Kern River Canyon	*xantiana*	35° 32.44N	118° 36.85W	725.4
5	Kern River Canyon	*xantiana*	35° 33.93N	118° 34.03W	740.6
6	Kern River Canyon	*xantiana*	35° 35.94N	118° 30.38W	710.1
7	Breckenridge Mountain, East face	*xantiana*	35° 28.87N	118° 32.45W	1210.0
8	Breckenridge Mountain, East face	*xantiana*	35° 29.68N	118° 33.96W	1828.7
9	Erskine Creek Canyon	*xantiana*	35° 35.48N	118° 26.73W	899.1
10	Greenhorn Mountains	*xantiana*	35° 41.23N	118° 32.31W	1573.6
11	NE of Isabella Lake	*parviflora*	35° 40.58N	118° 21.33W	838.2
12	NE of Isabella Lake	*parviflora*	35° 41.99N	118° 22.06W	863.4
13	Fay Creek Canyon	*parviflora*	35° 43.48N	118° 18.25W	1094.2
14	South Fork Valley, E of Onyx	*parviflora*	35° 43.03N	118° 10.06W	834.8
15	NE base of Pinyon Peak	*parviflora*	35° 41.90N	118° 03.60W	1331.9

Ecological Archives E087-090-A2

David A. Moeller. 2006. Geographic structure of pollinator communities, reproductive assurance, and the evolution of self-pollination. *Ecology* 87:1510–1522.

Appendix B (Table B1). ANOVAs testing for differences between subspecies and among populations within subspecies in (A) two mating system traits (herkogamy and protandry) and in (B) levels of autofertility (fruit set per flower or seed set per fruit; see *Methods*).

		Herkogamy		Protandry	
A.	df	*F*	*P*	*F*	*P*
Subspecies	1, 119	356.7	<0.0001	1305.3	<0.0001
Population (subsp)	29, 119	3.6	<0.0001	5.0	<0.0001
		Fruit set		Seed set	
B.	df	*F*	*P*	*F*	*P*
Subspecies	1, 116	2837.1	<0.0001	842.9	<0.0001
Population (subsp)	29, 116	1.7	0.024	1.1	0.323

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Ecological Archives E087-090-A3

David A. Moeller. 2006. Geographic structure of pollinator communities, reproductive assurance, and the evolution of self-pollination. *Ecology* 87:1510–1522.

Appendix C. The relationship between autofertility (measured by fruit and seed set in a pollinator-free greenhouse) and two mating system traits, herkogamy and protandry. Simple linear regressions were conducted on plants of (A) both subspecies (df = 1, 143), (B) subspecies *xantiana* alone (df = 1, 70), and (C) subspecies *parviflora* alone (df = 1, 70).

	Autofertility							
	Fruit set				Seed set			
	slope	intercept	*F*	*P*	slope	intercept	*F*	*P*
A. Both subspecies								
herkogamy	-0.179	0.831	238.5	< 0.001	-0.180	0.840	179.9	< 0.001
protandry	-0.189	0.897	821.7	< 0.001	-0.188	0.904	450.5	< 0.001
B. Within *xantiana*								
herkogamy	-0.016	0.147	6.0	0.016	-0.018	0.155	2.1	0.153
protandry	-0.039	0.250	23.8	< 0.001	-0.024	0.191	2.4	0.128
C. Within *parviflora*								
herkogamy	-0.068	0.959	7.2	0.009	-0.075	0.961	2.8	0.099
protandry	-0.074	0.946	5.6	0.021	-0.010	0.951	3.7	0.057

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David A. Moeller. 2006. Geographic structure of pollinator communities, reproductive assurance, and the evolution of self-pollination. *Ecology* 87:1510–1522.

Appendix D. A figure comparing mating system traits (herkogamy and protandry), autofertility (fruit and seed set in a pollinator-free greenhouse), and pollinator abundance (overall and specialist abundance) between geographically central vs. peripheral populations of *Clarkia xantiana* ssp. *xantiana*.

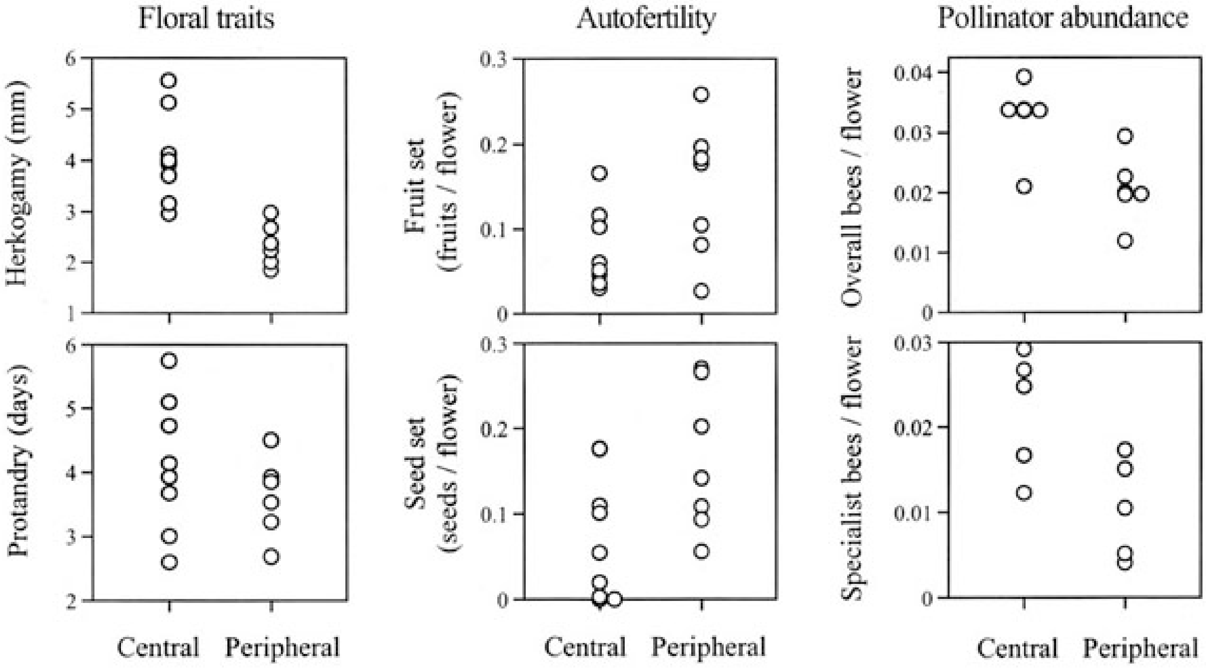

FIG. D1. A figure comparing mating system traits (herkogamy and protandry), autofertility (fruit and seed set in a pollinator-free greenhouse), and pollinator abundance (overall and specialist abundance) between geographically central vs. peripheral populations of *Clarkia xantiana* ssp. *xantiana*. Floral traits and autofertility were measured for eight central and seven peripheral populations. Pollinator abundance was estimated for 5 central and 5 peripheral populations.

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Ecological Archives E087-090-A5

David A. Moeller. 2006. Geographic structure of pollinator communities, reproductive assurance, and the evolution of self-pollination. *Ecology* 87:1510–1522.

Appendix E (Table E1). Bee species collected on flowers of *Clarkia xantiana*. Species are ranked by overall abundance. The subgenus for each species is shown in parentheses, when possible. Taxonomic designations follow Michener (2000)[†]. Pollen specialists are denoted by "S" and were designated according to MacSwain et al.'s (1973) monograph on bee visitors to *Clarkia* species. The number of sites occupied by each species is shown separately for subspecies *xantiana* (10 sites) and *parviflora* (5 sites).

			No. sites occupied	
Species		Abundance	*xantiana*	*parviflora*
Hesperapis (*Panurgomia*) *regularis* (Cresson)	S	258	10	0
Lasioglossum (*Evylaeus*) *pullilabre* (Vachal)	S[‡]	250	9	0
Ceratina (*Zadontomerus*) *sequoiae* Michener	S	168	9	0
Anthophora (*Mystacanthophora*) *urbana* Cresson		144	10	2
Megachile (*Megachiloides*) *gravita* Mitchell + *Megachile* (*Megachiloides*) *pascoensis* Mitchell [§]	S	125	8	0
Lasioglossum (*Dialictus*) sp.		96	7	0
Bombus (*Pyrobombus*) *vandykei* (Frison)		55	6	0
Hoplitis albifrons maura Kirby		48	8	0
Agapostemon (*Agapostemon*) *texanus* Cresson		46	3	1
Bombus (*Pyrobombus*) *vosnesenskii* Radoszkowski		28	10	0
Megachile sp. 1		18	2	0
Melissodes sp. 2		16	5	0
Andrena (*Diandrena*) *lewisorum* Thorp	S	15	2	0
Xylocopa (*Notoxylocopa*) *tabaniformis* Smith		10	5	2
Eucera (*Synhalonia*) *venusta* ssp. *carinata* (Timberlake)	S	9	1	0
Coelioxys sp.		9	4	0
Diadasia angusticeps Timberlake	S	9	2	0
Halictus (*Halictus*) *farinosus* Smith		8	4	0
Protosmia (*Chelostomopsis*) *rubifloris* (Cockerell)		8	2	0
Osmia (*Melanosmia*) sp.1		8	1	2
Bombus (*Crotchiibombus*) *crotchii* Cresson		7	3	0

Ceratina sp. 2	6	0	2
Apis mellifera L.	4	3	0
Osmia (*Melanosmia*) sp. 2	4	3	0
Ashmeadiella sp. 1	4	4	0
Melissodes communis Cresson	4	3	0
Melissodes sp. 1	3	1	0
Halictus (*Seladonia*) *tripartitus* Cockerell	3	1	1
Andrena (*Melandrena*) *cerasifolii* Cockerell	3	3	0
Megachile sp. 2	3	2	0
Melissodes lupina Cresson	3	3	0
Habropoda tristissima Cockerell	3	2	0
Synhalonia stretchii Cresson	3	1	0
Augochlorella pomoniella (Cockerell)	2	1	1
Hoplitis sp. 1	2	0	2
Hoplitis (*Monumetha*) *fulgida* (Cockerell)	2	1	0
Megachile sp. 3	2	1	0
Ashmeadiella sp. 2	2	0	2
Ceratina sp. 3	2	1	1
Ceratina sp. 1	2	2	0
Anthophora (*Paramegilla*) *centriformis* Cresson	2	0	2
Diadasia bituberculata (Cresson)	2	2	0
Heriades sp.	1	1	0
Hylaeus sp.	1	1	0
Hoplitis sp. 2	1	1	0
Andrena sp. 1	1	1	0
Andrena sp. 2	1	1	0
Chelostoma sp.	1	1	0
Trachusa (*Heteranthidium*) *timberlakei* Schwartz	1	1	0
Anthophora (*Melea*) *bomboides* Kirby	1	1	0
Melissodes nigracauda LaBerge	1	1	0
Bombus (*Pyrobombus*) *mixtus* Cresson	1	1	0
Anthophora sp.	1	0	1
Anthidium sp.	1	0	1
Dianthidium sp.	1	0	1
Centris (*Xerocentris*) sp.	1	0	1

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† Michener, C. D. The Bees of the World. The Johns Hopkins University Press, Baltimore, Maryland, USA.

‡ *Lasioglossum pullilabre* is considered a putative specialist but this has not been confirmed.

§ *Megachile gravita* and *Megachile pascoensis* were combined because females of the two species could not be distinguished. Males are readily distinguished and their distribution among sites indicated that the two species frequently co-occur.

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Ecological Archives E087-090-A6

David A. Moeller. 2006. Geographic structure of pollinator communities, reproductive assurance, and the evolution of self-pollination. *Ecology* 87:1510–1522.

Appendix F. A color version of Plate 1.

A color version of Plate 1. Many solitary bee pollinators of *Clarkia xantiana*, such as (left) *Hesperapis regularis*, forage exclusively on species in the genus *Clarkia*. These bees have specialized scopae, (middle) composed of sparse and unbranched hairs, which accommodate the (right) large and often clumped pollen grains of *Clarkia*. Photo credit: D. Moeller. Click here for a large image of the plate for closer viewing.

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