Abstract
Within the genus Clarkia, individual species have evolved floral traits that enable self-fertilization (selfing), as opposed to pollinator-dependent mating between genetically distinct individuals (outcrossing). Insufficient pollen deposition, which may reduce individual seed production, is hypothesized to have contributed to the evolution of selfing. If seed set is limited by pollen deposition (i.e., pollen limited), then natural selection may favor the evolution of traits associated with selfing. Our experiment confirmed that pollen limitation occurs in the self-compatible wildflower, *Clarkia unguiculata*, a predominantly outcrossing species that often occurs in sympathy with its siring sister species *Clarkia exilis*. In Spring 2010, we selected 600 plants at three different sites in the southern Sierra Nevada. We subjected two flowers per plant to one of the following experimental treatments: (1) naturally pollinated or (2) hand-pollinated with supplemental pollen from neighboring plants. We alternated treatments with respect to flower position on the plant to control for proximity to light. We compared seed production between fruits derived from flowers that were pollinated naturally and those that were pollen supplemented. During Spring of 2010, we performed this treatment at three different sites in the southern Sierra Nevada Mountains near Lake Isabella, California. At each site 200 *C. unguiculata* plants were chosen, 100 early in the season, and 100 late in the season. Approximately four weeks after pollination, fruits were glued to prevent seed dispersal and allowed to mature until we returned to harvest them. Thereafter I dissected them in a lab and quantified seed set.

Methods
In order to quantify pollen limitation in *C. unguiculata* we subjected two flowers per plant to one of the following experimental treatments: (1) naturally pollinated or (2) hand-pollinated with supplemental pollen from neighboring plants. We alternated treatments with respect to flower position on the plant to control for proximity to light. We compared seed production between fruits derived from flowers that were pollinated naturally and those that were pollen supplemented.

Results
Pollen limitation occurs in *Clarkia unguiculata*
We detected pollen limitation in *C. unguiculata* (pollination treatment effect p=0.0473) in both early and late in the flowering season. Pollen limitation occurred at a lesser degree during the late season. The seed set was significantly higher in the supplemented fruits than the naturally pollinated. An analysis of the effect of site by ANOVA showed geographic location had no effect on the data set and the data was consistent throughout our sites.

Abortion rates are influenced by timing
Our analysis of the data showed the abortion rates to be significantly higher in the late season than the early season (p=0.0001). Abortion rates were nearly tripled in the late season for both treatment groups.

Discussion
After sampling at our locations and performing statistical analyses of the data, we confirmed that pollen limitation does occur in *Clarkia unguiculata*. While some of our sampling was victim to herbivory and/or fungal pathogens, 706 of the fruits were successfully dissected and analyzed at the Mazer Lab, located at University of California, Santa Barbara.

While pollen limitation was detected early and late in the flowering season, the degree to which it occurred was more pronounced in the early than the late season. High abortion rates in the late season may have masked the degree to which pollen limitation occurred, since a plant may allocate its resources to a fixed number of seeds regardless of the amount of pollen it receives. This is consistent with the dryer and hotter conditions present in the late season.

The data is consistent with the fact that selfers have evolved to flower in the early flowering season and outcrossers in the later. Because pollen limitation occurs early in the season, the Mechanism of selfing is advantageous. However in the late season, the advantage of developing selfing mechanisms may be nullified if high abortion rates cause the plants yield a fixed number of seeds.

Habitat fragmentation and encroachment are known to disrupt plant-pollinator interactions, inevitably causing poor pollinator service to occur. While selfing may evolve as a mechanism to cope with these perturbations, the potential emergence of a drier climate in the Sierra Nevada may increase abortion rates and interfere with the ability of plant species to adapt to environmental change.

References


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