

Pollination of Pima Pineapple Cactus (*Coryphantha scheeri* var. *robustispina*): Does Pollen Flow Limit Abundance of This Endangered Species?

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Abstract—Pima pineapple cactus (PPC) (*Coryphantha scheeri* var. *robustispina*), a federally listed endangered species, occurs throughout southeastern Arizona and has relatively low population densities. To determine whether pollination limits reproduction of PPC we used florescent dye to quantify pollen flow between individuals in a PPC population. Preliminary results suggest *Diadasia* bees are capable of transferring pollen analogs nearly 1 km, but most transfers occur over a few hundred meters. PPC may not be pollen limited, at least when few competing cactus species are in bloom. Ongoing work will delineate the competition for pollinators between PPC and other cacti.

Introduction

Rare plants respond differently to a variety of ecological phenomena compared to their abundant counterparts. Rarity can be defined across gradients of geographic range, habitat specificity, and population size (Rabinowitz 1981). Rare plants can experience reproductive constraints through Allee effects, difficulties in the reproduction of organisms with small population sizes. These effects are caused by a variety of genetic, ecological, or demographic mechanisms associated with small population sizes and/or increased isolation, each of which can act alone or in combination (Forsyth 2003).

Pima pineapple cactus (PPC) (*Coryphantha scheeri* var. *robustispina*: Cactaceae) is a low-growing hemispherical succulent restricted to a relatively small range in the Altar and Santa Cruz Valleys in southeastern Arizona and Sonora, Mexico. It occurs on gently sloping bajadas and alluvial soils in the ecotone between Sonoran desert grassland and Sonoran desert scrub. PPC is federally endangered due to habitat loss, illegal collecting, habitat degradation and habitat alteration by non-native species, and range management practices (USDI 1993). The total number of PPC plants has yet to be determined, but approximately 3,000 have been located. The densities of populations vary between 1 plant per hectare to less than 1 plant per 10 hectares, and PPC occurs in a cactus community dominated by the genera *Opuntia*, and *Ferocactus*. PPC is primarily pollinated by the cactus specialist bee, *Diadasia rinconis*, and is also an obligate outcrosser (Roller 1996). Obligate outcrossers that require animal vectors for pollination are more likely to be affected by Allee effects (Huenneke 1991). Because PPC occurs at such low densities, lives in an ecotone, is pollinated by a specialist, and occurs

in a community where PPC competitors for pollinators are greatly more abundant, we believe that reproductive success potentially limits PPC abundance.

PPC aborts 18-59% of its flowers, but reasons for floral abortion are unknown (Roller 1996; Schmalzel 2000). There are two main explanations for floral abortion, proximate (ecological) and ultimate (evolutionary) mechanisms (Queller 1985). Of the proximate mechanisms, pollination failure, predation, and resource limitation are generally tested (Ackerman and Montalvo 1990; Parra-Tabla and Bullock 1998). An indirect pollen-flow experiment (i.e., delineation of the dispersal of pollen and pollen analogs) allows assessment of pollen limitation because individual pollen grains are counted rather than identifying seed paternity. An indirect pollen-flow experiment, coupled with selected crosses, would efficiently identify if pollination failure results in floral abortion. A pollen flow experiment would also illustrate how many individuals interact with each other in populations of rare organisms. These two benefits, identifying potential causes of floral abortion and estimating population interactions, are of prime importance in the reproductive ecology of PPC.

Reproduction is one of many areas of PPC ecology that has received little scientific attention. PPC flower buds begin to form in May and flowers bloom from June through August, although individual flowers bloom for only one day (Roller 1996). PPC flowers exhibit several synchronous flowering events a year and participation varies from only a few to nearly all individuals on a site (Roller 1996). Each flowering event occurs 5-7 days after a substantial (approximately 0.5 cm) "monsoon" rain. Female *Diadasia rinconis* are thought to be the primary pollinator of PPC; however, other bee species visit during flowering (Schmalzel 2000).

Determining the amount and distance of pollen flow in the population will indicate how many individuals are interacting with each other in a specific area, thus enabling identification of neighborhoods of PPC plants (*sensu* Wright 1943). Our objective is to quantify the relationship between distance and pollen flow in a PPC population.

Methods

Methods for pollen flow experiments are adapted from Waser and Price (1982), Thompson et al. (1986), and Waser (1988). All known PPC plants in an approximately 40-ha area were surveyed and mapped with a global positioning system before they bloomed in the summer of 2003.

During the summer of 2003, flowering events took place on June 5, July 17, July 29, and August 20. The August event was later than indicated by previous research (Roller 1996). Using a toothpick, we placed small amounts of fluorescent powder, a mimic for pollen, on the anthers of PPC flowers. These flowers are the source flowers. The dye powder is transported on the body of the pollinator and is deposited on flowers subsequently visited by the pollinator. Four plants were chosen at random, and each plant received one color of dye for a total of four different colored source plants every flowering event. During the third flowering event (July 29) we dyed two plants with two colors, due to a small sample size ($n = 8$ plants). All source plants were separated by at least 50 m to minimize spatial autocorrelation.

The day after flowering the stigmas were removed from all PPC plants in the study area and individually placed in small glassine envelopes. The stigmas were then taken back to the laboratory where the number of dye grains was counted under a UV light through a 50x-dissecting microscope. Pollen grains were not counted because the proportion of self pollination associated with each stigma is unknown.

Future experiments will calibrate the relationship between pollen deposition and dye deposition (Waser 1988). Until this relationship is known we assume that the flow of dye grains is equivalent to pollen flow in the population. Previous studies report a 9% error rate when making this assumption (Waser 1988).

Data were log-transformed to fit simple linear regressions between the number of dye grains transported and the distance between source and recipient plants. We also analyzed mean transport distance. All analyses excluded source flowers as a recipient of dye because of the difficulty associated with detection of dye grains on these flowers.

Many flowers had few to none of the potential dye colors present. Reasons for the lack of deposition are unknown, so we defined the neighborhood size two different ways: considering only the flowers that transported dye or all potential recipient flowers in the population. In other words, each flower had the potential to receive four distinct colors, but many flowers did not receive all four colors, so we either considered the entire set with the missing colors (all potential crosses and colors) or we excluded the absence of dye and analyzed the data based only on the presence of the dye.

Results

The proportion of flowers that aborted increased over time (figure 1). However, the abortion rate data from the third flowering period was discarded due to small sample size ($n = 8$ plants) and asynchronous flowering. When analyzing all potential recipient flowers in the four flowering periods there was a significant decrease in the mean number of dye grains deposited through the summer (figure 2). When analyzing the flowers that received dye the mean number of dye grains transferred did not significantly differ between flowering events (ANOVA; mean = 1.50, $F = 1.15$, $df = 3, 696$, $p = 0.327$).

Flowering events 1, 2, and 4 had negative regression coefficients between the log-number of dye grains deposited and the log-distance dye grains traveled for all potential flower crosses and for flowers with dye present (all $p < 0.001$). Flowering event 3 did not have a significantly negative slope for flowers with dye present and for all potential flowers and thus was excluded from further regression analysis. The three remaining flowering events (1, 2, and 4) showed a consistent trend of decreased dye grain deposition with increased distance between the source and recipient flower.

The estimate of the x-intercept for the above regressions highlights the distance where the average dye deposition equals zero. When analyzing the x-intercept of only flowers with dye present no significant evidence of a relationship through time emerges (figure 3, dark bars). When expanding this x-intercept analysis to all potential recipients in the population, the x-intercept decreased throughout the season (figure 3, light bars). There is also evidence that the proportion of flowers that did not receive dye (i.e., “missed” crosses) increased as the season passed (regression coefficient = 5.492×10^{-3} , $t = 3.71$, $p = 0.066$).

Discussion

In one model of a perfectly pollinated system each flower would receive pollen from each source plant at a quantity inversely proportional to the distance from the source plant. This model population exists when three assumptions are met: (1) there are no barriers to pollen dispersal (i.e., no isolation), (2) each flower has an equal chance of being pollinated, through space and time, and (3) pollinators are present and their activity is constant. If these assumptions were met every stigma we observed should have received all four dye colors. Most stigmas did not have this expected pattern, especially after the first flowering event. Thus, we would like to determine which assumptions were violated and led to the resulting lack of deposition in the three flowering events.

Three non-mutually exclusive hypotheses may explain why the reproduction of PPC was relatively low. The lack of reproductive success may have resulted from a paucity of pollinators available to deposit sufficient quantities of pollen either through (1) the pollinator being absent or (2) through PPC competitors securing the pollinator’s attention. Or, (3) there could have been a lack of plant-available resources, which would constrain PPC fruit development regardless of the success of pollination. This latter hypothesis is supported

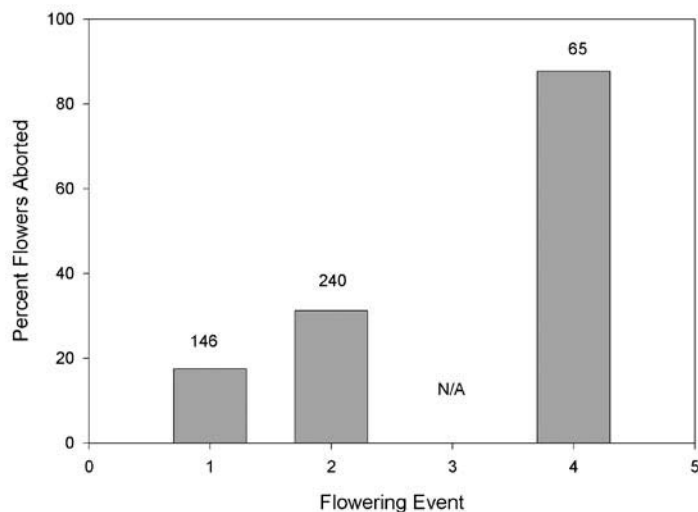


Figure 1—Percent of flowers aborting during each flowering event; numbers indicate sample size.

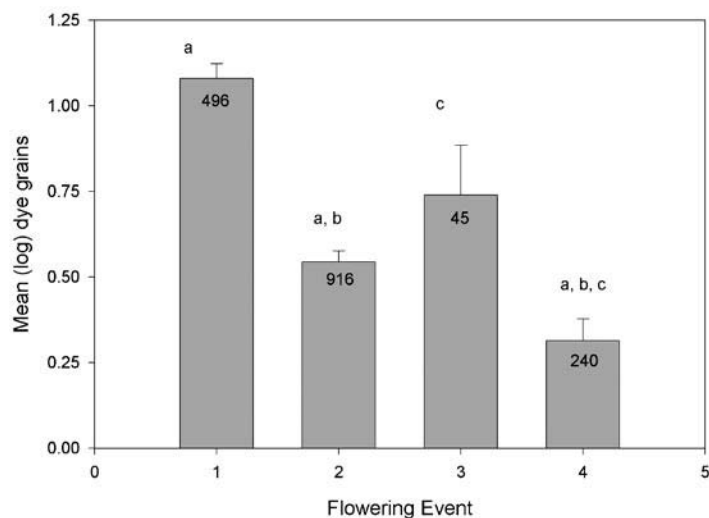


Figure 2—Mean number of dye grains deposited for all potential crosses; a, b, c denotes a significant difference ($p < 0.01$) between means. Numbers indicate sample size.

by the increase in proportion of flowers aborted throughout the season and cannot be excluded at this time. Nevertheless this mechanism does not account for the observed differences in dye dispersal, which suggests that pollinator presence and/or abundance explains the decrease in pollination success.

Of our initial three assumptions, (1) no isolation, (2) all flowers have an equal chance of pollination, and (3) pollinators are present and their activity is constant, we believe that a lack of pollinator activity provides the best explanation for our results. *Diadasia* bees are capable of moving pollen analogs nearly 1 km, and we suspect they have the capacity to move dye farther. This suggests that all plants on our site had the potential to receive dye from all of the four source plants, thereby eliminating assumption number 1. The observed lack of transferred dye may indicate an Allee effect in which PPC is constrained in reproduction by its small population size.

However, we were unable to determine if our second assumption, that flowers have unequal pollination success, was violated and could have confounded our results.

The pollination success of PPC in our study site decreased during the summer. Previous studies did not look at pollination success for this length of time (Thompson et al. 1986; Waser 1988). The decrease in pollination success could be explained equally well by decreases in pollinator abundance and activity. However, female *Diadasia* bees decline only slightly between June and July, whereas we found that abortion rates nearly doubled during the same period (Ordway 1987). Evidence of partial bivoltinism occurring in *Diadasia rinconis* suggests that the second generation of bees would emerge to pollinate *Ferocactus wislizenii* (Neff and Simpson 1992). It is unknown whether these lines of evidence support the prediction that there were enough *Diadasia* present to successfully pollinate PPC.

PPC generally produces flowers between the flowering period of its main competitors *Opuntia*, and *Ferocactus*. In southern Arizona *Opuntia* cacti typically bloom coincident with the spring rains, and *Ferocactus* plants begin to bloom a few weeks after the beginning of the “monsoons” which typically start in July (Turner et al. 1995). Future experiments will determine if plant-available resources are limiting PPC reproduction during the flowering season, but we believe that the decline in reproductive success can be attributed to a lack of pollinator resources.

PPC occurs in relatively low densities, is pollinated by a cactus specialist bee, and probably cannot out compete its competitors for this resource. PPC produces flowers temporarily between its major competitors, likely because this period is a stable point in time during which PPC can maximize its chance of successful pollination. Our data reveal that the proportion of flowers missed by pollinators increased when PPC was blooming at the same time as its competitors. This could account for the decrease in pollination success through the summer. When *Ferocactus* begins to bloom the potential exists that *Diadasia* preferentially pollinate *Ferocactus*, which is a more stable resource compared to the sporadic resources PPC provides. However since a nucleus of dye transfer exists during all flowering periods a significant number of bees may have discovered this sporadic reward during the later part of the summer.

An estimate of neighborhood size can be developed from the greatest distance at which the average plant receives pollen. This point is modeled by the x-intercept in figure 3. In a highly fragmented population with widely spaced individuals the x-intercept represents the maximum distance PPC plants could be separated and still receive significant pollen. In our un-fragmented study population this x-intercept determines the number of potential sources of pollen a PPC plant could receive. Future research will determine the distance correlated with the x-intercept. This distance would be critical for reserve design in areas of fragmented PPC habitat.

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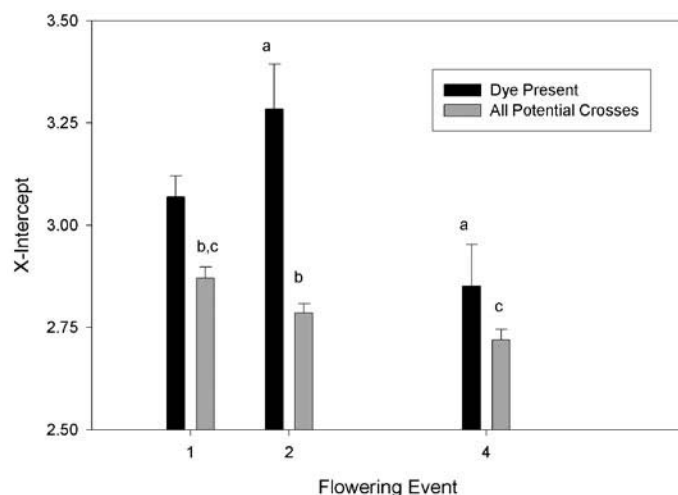


Figure 3—X-Intercepts of the regression of log dye grains deposited versus log distance for flowers with dye present and for all potential crosses; a, b, c indicates significant difference ($p < 0.05$) between pairs.

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