Yellow or Red? Study Design for a Question on the Effect of Flower Color on Pollination of *Potentilla Glandulosa* in Freeman Meadow

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Potentilla glandulosa

Abstract

We wanted to choose a study design to test the effect of color on insect pollination of *Potentilla glandulosa* in Freeman Meadow, a high Sierra meadow in California. This plant, a member of the family Rosaceae, has yellow flowers. Our hypothesis was that if some of the flowers were colored red, the pollination rate would decrease. Test data were collected and analyzed using Analysis of Variance and showed that a within subjects study would be the most efficient way to proceed.

Introduction

Animal pollinators are very important to the success of many plant species. Pollination is required for sexual reproduction, which promotes out-crossing and maintenance of genetic variation. This variability is important for an organism's ability to adapt to changes in its environment. Plants rely on mechanisms such as animals and wind to move pollen, the genetic material, from one plant to another. Pollinators can recognize factors such as flower color, shape, markings, or scent as attractants.

One particular method that plants use to control pollinators is flower color. Pollinators vary in their ability to perceive different colors, or wavelengths of light. For example, insects tend to have a difficult time recognizing the color red, while this color is well within the range of wavelengths recognized by birds. We decided to test the relationship between flower color and insect pollinator activity on *Potentilla glandulosa* in a high mountain meadow. Our hypothesis is that insect pollinators are more attracted to flowers with yellow petals than with petals of another color. If some flowers on a plant were left yellow, and others changed to another color such as red, we predicted that the amount of pollinator traffic would vary. As a first step, we examined the volume of pollinator traffic on each of two halves of plants containing unaltered (yellow) flowers.

Methods

Study Area

Freeman meadow is a high elevation mountain meadow to the northwest of Yuba Pass, in the Tahoe National Forest. We observed pollinators of *Potentilla glandulosa*, which were found growing in a dry perennial streambed in a sunny part of the meadow.

Freeman Meadow

Data Collection

We mapped out a transect that included most of the *Potentilla glandulosa* plants and randomly assigned coordinates within our transect to five pairs of observers. Each pair located the plant closest to their assigned coordinates, randomly divided the plant in half (tossing a stick to determine the angle at which to divide the plant) and noted the number of flowers in each half (A and B). Each pair then situated themselves to observe the plant from different angles and record visitation by pollinators. Each pollinator was only counted once, on its initial choice of sides. We found that it was necessary to have two observers because not all flowers in a cluster faced in the same direction. We collected data for three ten-minute intervals to test different observation times.

Walking Transects

Counting pollinators

Results

We determined the sample size needed for a within-subjects design experiment to detect a 20% decrease in pollinator visitation with $\alpha = 0.05$ and 80% power by comparing each half of the

plant. We used the same total plant data to determine sample size needed for a between-subjects design to detect a 20% decrease in pollinator visitation also with $\alpha = 0.05$ and with 80% power by using the total for each plant and comparing it to the other plants in the group. The raw data we collected is expressed in the following table:

Table 1. Raw data on pollinator visitation.

	A half				B half			
Plant #	0-10 min	10-20 min	20-30 min	Number of flowers	0-10 min	10-20 min	20-30 min	Number of flowers
1	7	8	23	25	9	10	19	26
2	10	9	6	25	10	7	8	25
3	9	5	8	9	3	5	6	10
4	3	2	3	10	4	2	0	10
5	3	1	4	8	3	2	4	9

This chart describes the number of distinct pollinators that landed on any flower on each half of the five plants over three contiguous 10-minute intervals. It also notes the number of flowers on each side of the five plants.

Table 2. Summary of total pollinator visits/flower for whole plant and for each plant half.

Sample Site	Total Number Visits per flower	Difference in Number of Visits per Flower between A half and B half
1	1.49	0.06
2	1.00	0.00
3	1.89	1.04
4	0.70	0.20
5	1.00	0.00

Using the observed average number of pollinator visits per whole plant (mean = 1.217, sd = 0.473), we calculated a 20% reduction in the mean visitation rate to be a reduction of 0.36425 visits/plant.

Using the program PS (Dupont and Plummer 1997), we found that to achieve power of 80% in a between-subjects design, the sample size, *n*, would need to be 22 plants (Figure 1). Similarly, to achieve a power of 80% in a within-subjects design would require a sample size of 11 (Figure 2).

Figure 1. Power curve for between-subjects design. A sample size of 22 plants is required to have 80% power, $\alpha = 0.05$, and an effect size equal to a 20% reduction in pollinator visitation.

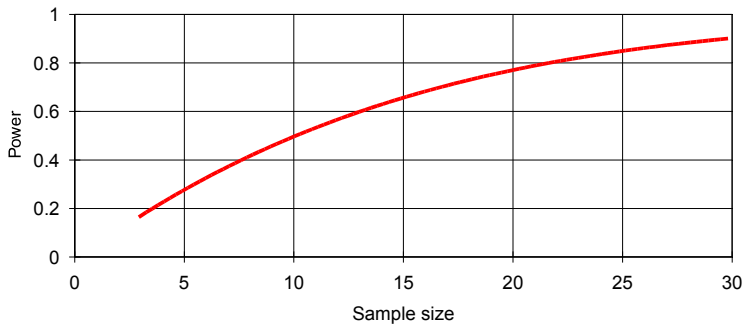
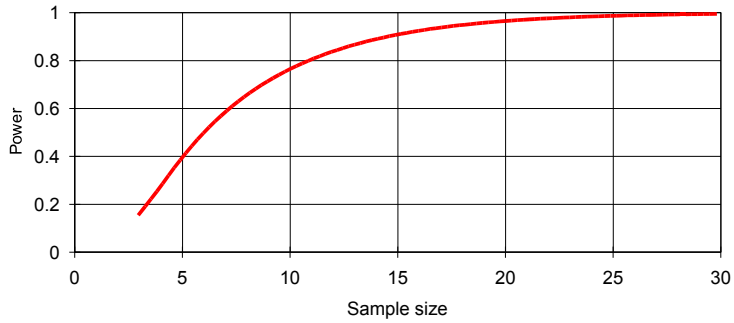


Figure 2. Power curve for within-subjects design. A sample size of 11 plants is required to have 80% power, $\alpha = 0.05$ and an effect size equal to a 20% reduction in pollinator visitation.



Discussion

We wanted to test a basic scientific hypothesis about pollinator response to different flower colors, specifically yellow and red. We determined that a 20% reduction in visitation rate would be biologically interesting, and an experimental design with a 5% Type I error rate and 80% power to be sound. Our field measurements show that a within-subjects experimental design is likely to be more efficient than a between-subjects design, based on sample size.

The sample size for the between-subjects design is 22, with a 50% decrease in sample size for a within-subjects design. This result is mainly due to the highly correlated measurements between the untreated halves of the plant. A next step is to measure the response to the red color treatment. We could also examine the temporal data to determine the minimum observation time needed to detect a significant difference in pollination rate.

We performed uncontrolled observations to ask a general question about pollinator attraction, then performed more controlled observations that allowed us to construct sound methods for a future mensurative experiment, described above. A further step would be to perform strict manipulative experiments with pollinators and color in the lab in order to more reliably determine causal relationships. Although the results of these experiments would be limited to our population of inference, the population of *Potentilla glandulosa* plants in Freeman meadow, the results would add evidence to general theory about pollinator attraction.

Literature Cited

Dupont, W.D. and W.D. Plummer. 1997. PS – power and sample size program available free on the internet. Controlled Clinical Trials 18 : 274.