Pollination Constancy Exercise

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The purpose of this exercise is to test whether or not certain pollinators visit flowers randomly, or specialize on certain flower species or types, a phenomenon known as "pollination constancy." Therefore, this exercise works best in a garden with a mixture of flower colors and shapes, so proper comparisons can be made. Before data collection can begin, students should familiarize themselves with the different flower species available on site, with the help of the teacher. Colors and shapes of each flower species should be defined before any data are taken. For example, if flowers have two colors, such as daisies, we should either decide what the dominant color is for that species, or if it seems appropriate, define it as its own color, white/yellow.

Usually though, flower colors will fall easily into the following categories: **white, yellow, orange, red, pink, purple, blue.**

Shape categories are either radially symmetric or bilaterally symmetric - **radial** or **bilateral** for short.

<u>Questions to ask before data collection:</u>

1. What might you predict about pollinator specialization? Do you think it would make sense for insects to specialize on flower types or not? Why? Might it be more advantageous for some species rather than others? (Think of advantages for either strategy, random or specialized.)

2. Based on what you know generally about insect vision and pollination, can you predict what flowers pollinators of different species might prefer? Might you expect different behavior from bees vs. butterflies? Why?

<u>Data collection</u>

Students work in pairs, with one observer and one data recorder. Each pair of students finds a pollinator (bee or butterfly, usually) and follows that individual exclusively for as long as possible. The observer keeps her eyes on the insect while giving the reporter data on flower species, shape, and color. Students can also observe how far pollinators travel between flowers. Do they go to the next immediate flower, or skip over some flowers? Some of these observations will be more qualitative than quantitative, but the observer should give as much information to the recorder as possible. To do the proper analysis, the minimum information recorded should be flower species, shape, and color, in the order visited.

After a set period of pollinator-watching, at least a half hour, we will reassemble as a class to analyze the data. We will do a separate analysis for each bee or butterfly, comparing the numbers of flowers visited of each color available, to the expected number if the insect were choosing flowers randomly. The expected numbers are just the total number of flowers visited, divided by the number of categories. For example:

One bee visited :
5 white
6 yellow
2 orange
0 red
0 purple
2 blue

Did the bee show pollination constancy or not?

To figure it out, we make a table comparing the actual numbers with the expected values, which are calculated based on the proportions of flower types in the patch:

Color	Observed	Expected
white	5	3
yellow	8	3
orange	3	3
red	0	3
purple	1	3
blue	1	3
total	18	18

You see the two totals in the "observed" and "expected" columns have to be the same. This is because our expected numbers are based on the same number of visits that we observed. Because we need to know the expected values of random visits based on the number of visits the bee actually made (=18 in this example), we divide that number of visits into the total number of categories (=18/6=3). (It's perfectly fine if this number is a decimal, even though it happened to come out evenly in this example.)

Depending on the level of your students, this can be taken a step further and analyzed with a chi-square test, available online here:

http://www.graphpad.com/quickcalcs/chisquared1.cfm

All you have to do is type in your category names (in this case, colors) and your observed and expected values in the table, and the page will calculate the statistical result for you. A statistically significant result means that these distributions can be stated to be different from each other, meaning the bee is not choosing flowers randomly. After you click the "calculate now" button, you will see the "P value" at the top of the page. In this case, $P=0.009$. The generally accepted cutoff for statistical significance is $P<0.05$, which means that we accept less than a 5% chance that the data we have came from a random distribution. Because $0.009<<0.05$, we are fairly confident that the bee is demonstrating a true preference for yellow.

But if you don't want to get into statistics with younger children, you can simply look at the values and point out the big differences - in this case, mainly white, yellow, red and purple. We would say that the bees appear to prefer white and yellow, and avoid red and purple. Thus the conclusion would be that this bee has some pollinator constancy for color.

In a given class you will do this test for several individual bees (or other pollinators). Look at all the results together and talk about whether it seems as though bees in general show flower constancy, or not.

The same test can be run on shapes to see if shape, independently of color, affects bee behavior.

Questions to ask after data are collected and "analyzed" :

1. If pollinators appear to show constancy, what might be an explanation for this? You can think about this in two ways. First, there may be an answer that has to do with immediate benefit for the insect. But also think about the long term, i.e. an evolutionary benefit. These two ways of thinking about behavior are known as "proximate" and "ultimate" causes, respectively.

Proximately, it may be more efficient for a pollinator to use a single search image when locating flowers. Insect brains, like human brains, are not good at looking for a lot of things at once.

Ultimately, flower constancy has a benefit for plants, because it ensures transfer of pollen to the right species. This benefits the bees too in the long term because pollination, remember, is a mutualism: if the bees do not properly pollinate the flowers, there will be fewer flowers around in the next generation, which means fewer resources for the descendants of these bees.

2. Do all bees prefer the same color, or do different individual bees favor different colors? Why or why not, do you think?

3. What data are we missing that might affect our conclusions? This is especially important to discuss if the data are not strong in showing a trend toward color preferences. That is, all analyses such as this rely on certain assumptions. What assumptions are we making in this observational study? Examples of these are assumptions about spatial distribution of flowers, relative numbers of flowers of each color available, relative flower sizes of the different colored flowers, different levels of rewards in different flower species, interactions with other pollinators, and any others you can think of.

4. Any other questions that you or your students come up with!