Echinacea Project Summer Research Proposal
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The efficiency of *Echinacea angustifolia* pollinators and the relationship of pollinator and distance between the parental plants.

Introduction:

Echinacea angustifolia is a native plant common to the tall grass prairie the central United States. These native grasslands have been constantly shrinking and are now less then 0.1 percent of the*ir* original size. (Sampson and Knopf, 1994). *Echinacea angustifolia* is a good model plant for studying the effects of the prairie fragmentation due to its longevity and self-incompatibility, characteristic of many plants native to this habitat (Wagenius et al. 2007).

E. angustifolia lacks an efficient technique for pollen distribution manually, so the species relies on 26 native species of bees. This means the movement of the pollen is dependent on the movement of these pollinators. The challenge of the pollinators of *E. angustifolia* is two-fold; pollinators must get pollen to the styles of *E. angustifolia*, as well as have pollen that is compatible.

The self-incompatibility in the *E. angustifolia* is based on the S allele. If the either of the two S alleles of the pollen are the same as the style's alleles, the pollen is rejected and the achene does not get fertilized. This process acts as a good measure against inbreeding, which has shown to greatly decrease the fitness of *E. angustifolia* (Wagenius et al. 2010). However in the fragmented prairies that the *E. angustifolia* this self-incompatibility trait reduces the number of possible mating partners of each plant.

The main barrier that *E. angustifolia* faces in producing viable seeds is pollen limitation, rather than pollinator limitation (Wagenius et al. 2007). This means the most important factor in the pollinator efficiency is where the pollen comes from. *E. angustifolia* has been shown to have a correlation between population size of the population and rate of pollen compatibility, where the smaller populations have a lower compatibility rate then larger populations (Wagenius et al. 2007). If the pollinator travels further between plants, the pollinator would have a higher chance of holding compatible pollen.

Previous research on pollinator efficiency has been done by Katie Koch, and Andrew Kaul. Their data shows that *Melissodes* females are the most efficient, followed by *Agopostemon, Lasioglossum, Halictus, Auglochlorella*, and finally *Melissodes* male. I would like to increase the data set of this experiment so that solid conclusions can be drawn. Up until now there are 78 total data points, my goal is to add 50 additional data points, and *have* each group of pollinators represented.

Questions:

Which *Echinacea angustifolia* pollinator causes the highest proportion of the styles to shrivel?

- Is there a certain functional group of bee that has higher efficiency?
- Does amount of pollen carried by the pollinators follow the trend of pollenating efficiency?

Is there a relationship between the distance between the parental plants and the pollinator?

- Do pollinators that travel further have a higher efficiency than pollinators that travel less?

Question One Methods:
This observational data will take place in the common garden two, or Jennifer's common garden. The benefit of changing locations will be additional information at a different location, but also the ability to find the paternity using microsatellites that Jennifer discovered for each plant in the garden. I will be using the same methods as Andrew Kaul used in 2012. A set number of flowering plants will be selected randomly to be a part of the study, as well as a set number of times (three). The virgin inflorescence of each selected plant will be covered with the pollinator exclusion bag to prevent insect damage or contamination. Two rows of untouched styles will mature before the plant is exposed to pollinators. Where the styles are on the head (base or top) will be recorded. The bracts of these mature styles will be painted to distinguish between other styles, the pattern being: color #1, no color, Color #2. A digital camera will be used to record the pollinator visit to help with the classification of the pollinator, and a stopwatch to measure the duration of the visit. Approximately 24 hours after the visit I will return and assess the status of the styles recording how many styles are shriveled. The shriveled styles will have their bracts painted third distinct color. Those achenes will be collected to answer the second question.

Materials:
Pollinator exclusion bags, clipboard, measuring tapes, flags, stopwatch, acrylic paint, video camera (2?), tripod (2?), pollinator identification guide

Question Two Methods:
The pollenated styles from the above methods should develop into achenes. The achenes will be harvested when they fully developed. The achenes will be separated by flower and by pollinator (If the head is used more than once). In a greenhouse the achenes will be germinated and be grown until the first true leaf emerges. The leaf will be harvested and have the DNA extracted. The DNA of each achene will be run in a PCR with each of the primers from each of the parental plants. The products of the PCR will be run through a sequencing gel. With the gel results the parents plants of each achenes will be able to be determined.

Literature Cited

Samson and Knopf. 1994. Prairie Conservation In North America. Bioscience.

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