Bee Sampling Protocol –June 2018

Effects of grazing versus fire for prairie management.

Materials:

GPS unit
Batteries
Stop watch
3.25oz plastic bowls; yellow, white, and blue
Binder clips
Wooden dowels or lengths of bamboo, cut to approximately 0.5m
Dawn dish soap
Gallon jug of water
Aerial insect nets
Ethyl acetate
Eyedroppers
Ethyl acetate kill jars
50mL polypropylene centrifuge tubes
cotton balls
Glassine envelopes
Field notebook
Pencil
Heavy paper
Whirl-Pak® bags
Brine shrimp net
70% ethanol
Cooler
Laser printer and acid-free cardstock for locality labels
Mason jar and metal ring of lid
Fine mesh
Scissors to cut mesh to size
Timer
Hair dryer
Petri dish (and a dry place to store them)
Pins
Foam block

Bee protocols:

We surveyed bees using two methods - passive trapping and active netting. Bowl traps are a highly replicable means of collecting bees over a period of time, with little-to-no observer bias. However, not all bees are attracted to standard bowl traps. Therefore, directed netting at a site by an observer can serve to round-out species lists and provide a more accurate view of species richness when combined with bowl data.

Deploying bowl traps:

Obtain 30 standard 3.25 oz plastic bowls in three colors (white, yellow, and blue). At a randomly selected end of a predetermined transect, deploy bowls in a ladder-like pattern. Place a wooden dowel or length of bamboo approximately 0.5m long into the ground. Use a binder clip to hold the lip of the bowl to the elevated end of the stake. Fill the bowl with a mixture of water and Dawn dish soap. Using this same method, place a second and third bowl set five meters away from the transect in opposite directions, in effect creating a 10m long perpendicular transect. Use one bowl of each color in this set. Pace out twenty meters along the original transect, towards the other end. Place another set of three bowls, using all three colors. Repeat this until 30 bowls are in place. (Note: This modification of a standard bee-bowl transect, in which bowls are placed every five meters, served to create 20-meter gaps through which cattle could pass without encountering bowls.)

Netting bees:

Next, collect bees using an aerial net during a meandering walk of the site. The duration of this walk can vary depending on site size, but should always be recorded so that the collection effort can be calculated later. Wander through the site, looking for flowers. Net bees that are observed on flowers. (Note: We only collected bees when they were on flowers, in an attempt to minimize detectability biases.) Once captured in a net, bees tend to fly upwards. Hold the end of the net up so that bees corral themselves into the end of the net. Insert a 15mL polypropylene centrifuge tube containing an ethyl-acetate soaked wad of cotton into the net and maneuver it underneath a single bee. With some manipulation of the net and the tube, the bee will fall into the tube. Cap the tube while it is still in the net to prevent escape. Using a pencil, record the species of flower the bee was found on, the site name, date and time on a glassine envelope. Place the now quiet bee into this labeled envelope and put the envelope into a charged ethyl-acetate kill jar. Repeat until all bees are removed from the net. Continue the meandering walk, repeating the above procedures when bees are encountered on flowers.

Collecting bowl traps:

Approximately 24 hours later, collect bowls. Unclip bowls from their stakes, and dump the contents of each bowl through a brine shrimp net. (Note: We removed moths and butterflies from our samples, with butterflies set aside and moths discarded.) Collect stakes, bowls, and clips. Once all bowls are removed, or periodically if necessary, transfer the contents of the shrimp net to a single Whirl-Pak®. Label the bag with a permanent marker, noting the site, date, number of bowls collected, the time at which the last bowl was deployed and at which the last bowl was collected. Replicate this information in pencil on a sturdy piece of paper and insert into the bag. Fill bags with sufficient 70% ethanol to fully cover all insects. Seal the Whirl-Pak® by squeezing out as much air as possible, rolling the top of the bag down to the level of the ethanol, bending the wires together and twisting them together. Place samples in the freezer until they can be washed, dried, and pinned.

Processing bowl samples:

Once in a laboratory, bowl traps can be processed. Remove a single sample from the freezer and empty into a Petri dish. Split the sample into multiple dishes if it is too large. Add liquid so that insects can be moved around and slightly disentangled. Using a stereo-microscope and forceps,

separate bees from non-bees, placing non-bees into a separate Petri dish. Pour the bees into a mason jar. Add warm water and a small amount of Dawn dish soap. Place a piece of fine mesh over the mouth of the jar and attach the metal ring component of the jar lid. Cover the mesh with a gloved hand and shake the jar vigorously for one minute. Rinse the soap out through the mesh. This will take multiple rinse cycles. Once insects are no longer soapy, blow a hairdryer through the mesh, rotating the jar until no insects stick to the sides of the jar. Adding small pieces of balled-up paper towel can help fluff bees and remove some excess moisture. Remove metal ring and mesh. Pour all insects from jar into a covered petri dish. Copy collection information from Whirl-Pak® bag onto a slip of paper and include it in the Petri dish. The sample may then be refrozen to be pinned at a later date, or pinned immediately.