

## ECOS Inquiry

**1. CONTRIBUTOR'S NAME: ALISON PERKINS**

**2. NAME OF INQUIRY: POLLEN GRAIN REFERENCE COLLECTION**

**3. GOALS AND OBJECTIVES:**

**a. Inquiry Questions:** How do you prepare dried plant specimens?

**b. Ecological Theme(s):** Identifying pollen requires visual recordings and descriptions of pollen from known sources. Because pollen grains are considered species specific, developing a reference collection with which unknown pollen can be compared will aid identification.

**c. General Goal:** To develop a pollen reference collection that can be used to determine pollen sources for food of Orchard Mason Bees and other pollinators.

**d. Specific Objectives:**

*Academic:* Development of a reference collection of plants to determine pollen sources

*Procedural/technical:* Learning basic lab techniques preparing specimens and using microscopes

*Social:* Working in groups

**e. Grade Level:** 9-12

**f. Duration/Time Required:** various

**4. ECOLOGICAL AND SCIENCE CONTEXT:**

**Background (for Teachers):**

Pollen grains are considered species specific and may be used to identify sources of food in Orchard Mason Bee nest cells. Because of variation within a taxon, however, a good pollen grain description or identification for a taxon should be based upon several pollen samples taken from voucher plants collected across a wide geographical range. Nevertheless, having a local specimen collection can be very useful, especially if plants are dried and preserved for as part of a school yard herbarium. In fact, a herbarium can be used in a number of ways if the information is collected correctly (see the ECOS Inquiry called Stories from the herbarium: Introduction to ethnobotany on the ECOS website - <http://www.bioed.org/ecos/Inquiries/inquiries.aspx>). Preserved specimens can then serve as “voucher” specimens.

**5. MOTIVATION AND INCENTIVE FOR LEARNING:**

Students get to go outside, learn about the plants and insects in their schoolyards, and observe pollination.

**6. VOCABULARY:**

**Herbarium** – a collection of preserved plant specimens

**Voucher specimen** – a specimen archived in a permanent collection that serves as physical evidence of occurrence at time and place.

**7. SAFETY INFORMATION:**

Some plants may have thorns or secrete irritants. Caution should be taken when handling these plants.

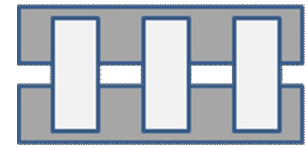
**8. MATERIALS LIST (including any handouts or transparency masters):**

- trowels
- work gloves
- labels
- permanent markers
- commercial plant press OR
  - blotting paper
  - newspaper
  - cardboard
  - scissors
  - old scrap wood (1 x 6' or 1 x 8")
  - old belts

## 9. METHODS/PROCEDURE FOR STUDENTS:

### a. Pre-investigation work:

Specimens should be pressed and dried in a plant press. An easy to make press includes some scrap wood 1 x 6s or 1 x 8s, cardboard pieces, newspaper, blotting paper, and some old belts. Cut the scrap wood to short pieces that can be laid down in a crossing pattern that will be about the size of the blotter paper (see picture to the right) and fasten them together with nails, brads, or staples. Make two. Arrange the cardboard so all the piping is in the same direction. Cut the cardboard pieces to the size of the blotting paper you'll be using. Take one of the wood pieces and lay it with the flattest side up. Layer cardboard-newspaper-blotter paper-plant-blotter paper-newspaper-cardboard until you've reached about 8 inches or so in depth. Place the other of the wood pieces with the flattest side down on top. Wrap the pile with a couple old belts and pull tightly. Place the press some place warm with a lot of air movement, preferably with the piping facing the air current (this improves air circulation around the plants and reduces the likelihood of mold and mildew). Once the voucher specimen is dried, identified, and a label placed with it, the specimen is ready to be placed in an herbarium. Laminating makes the specimens user friendly.



### b. Investigation work:

Students can collect voucher specimens throughout the spring, or any period when plants are flowering. For herbaceous plants, the entire plant is usually collected including roots, stems, leaves, flowers, and fruits, assuming that all of these parts are present. Remove the soil from the roots. If the plant is too large or unwieldy to fit in a plant press, collect and press representative parts, for example, the roots, part of the stem with characteristic leaves, and an inflorescence or part of it. Be sure to record the original plant height on the label. Be sure to collect reproductive parts.

For woody plants, representative parts are chosen as follows: tips of a terminal branch (about 30-35 cm, 12-14 in) showing leaf arrangement (leaves should be pressed open and flat); flowering and/or fruiting material; and other morphological structures unique to a species. As with herbaceous plants, extra reproductive parts are desirable.

Each specimen should have a proper label including:

- a. **Title**-identifying the political boundary within which the specimen was collected (e.g., Plants of Lewis and Clark Elementary, Missoula, MT)
- b. **Scientific name**- genus and specific epithet of the plant, including the author(s) who described it if possible (this information is usually included in most online field guides)
- c. **Collection site** - indicated by road intersections; latitude and longitude; coordinate system; township, range, and section; or any combination thereof

- d. As detailed a habitat description as possible, including soils, position on the landscape (e.g., east-facing slope)
- e. **Dominant plants** with which the voucher specimen was growing, if known
- f. Flower color, if applicable
- g. **Frequency** of the specimen (e.g., frequent, infrequent, rare)
- h. **Name of the collector(s)**
- i. **Collection number**
- j. **Date** the plant was collected (day, month, year).

EXAMPLE:

PLANTS OF LEWIS AND CLARK ELEMENTARY, MISSOULA, MONTANA, U.S.A.  
 PORTULACACEAE  
*Lewisia rediviva* Pursh  
 3.4 mi NE on US 90 from its jct. with Van Buren Drive, N of Missoula. 46°52'38N  
 113°57'24W. Open, mesic with deep sandy soil. Elevation ca. 4589 feet.  
 Associates: *Quercus stellata*, *Ilex vomitoria*, *Schizachyrium scoparium*, *Sorghastrum nutans*, and  
*Cyperus retrorsus*.  
 Frequent  
 P. Alaback 1115 15 June 1994

Use a commercial or homemade plant press (see above) to press the plants. Place the press in a warm place with good air circulation. Each voucher specimen needs to be dried, identified, and labeled, and stored in a safe, dry place. Laminating makes the specimens user friendly.

**c. Extension:**

Students can prepare and examine pollen using standard microscopes. Below are instructions to prepare basic fuchsin gelatin and pollen slides from *Pollination Ecology: Field Studies of Insect Visitation and Pollen Transfer Rates*, by Judy Parrish. Pollen can be collected from flowers while the voucher specimen is being collected or after the specimen has been dried. Pollen can be collected in small envelopes until ready for preparation. The complete inquiry can be found on the Teaching Issues and Experiments in Ecology website at <http://tiee.ecoed.net/>.

A jelly containing stain to make a semi-permanent microscope slide of pollen, as suggested by Beattie, A. J. 1971. *Pan-Pacific Entomologist* 47: 82.

Use the following ingredients:

- \* distilled water, 175 ml,
- \* glycerin, 150 ml,
- \* gelatin, 50 g,
- \* crystalline basic fuchsin as desired - enough to make solution the “color of a fine claret,”
- \* crystalline phenol, 5 g - important in humid environments, may be left out if gelatin can be refrigerated. Don’t touch gelatin if phenol is used.

Procedure to Make the Fuchsin Gelatin:

1. add the gelatin to the distilled water in a beaker and heat until the gelatin dissolves,
2. add the glycerin,
3. add phenol, if desired,

4. add basic fuchsin crystals a few at a time until the solution is the color desired. Too light will not stain the pollen, but too dark may mask details of the pollen,
5. filter the solution through glass wool or cheesecloth,
6. pour into sterile containers such as petri plates that can be covered. If phenol is not used, refrigerate the plates and slides. They will keep about a month without refrigeration.

#### Preparation of Pollen Slides

1. keep the prepared slides, and the unused jelly, out of the sun, and cool enough not to melt,
2. with a dissecting needle, cut a small cube of the jelly out of the petri plate,
3. brush the cube of jelly against an anther containing pollen, or on the insect body part,
4. place the cube containing the pollen sample on a glass slide,
5. place a coverslip on top of the cube of jelly,
6. gently heat the slide over a candle flame until the jelly melts. Do not overheat, or scorch the slide. If it is warm and sunny, the jelly may be melted by placing the slide on a dark surface in the sun instead of using the candle flame. This will make a semi-permanent, stained specimen,
7. using a permanent marker, label the glass slide (date, species sample was collected on).

Below are some examples of plants and their pollen from the *Pollen as Indicators of Source Areas and Foraging Resources* website

[http://pollen.usda.gov/Reference\\_Collection/Reference\\_Collection.htm](http://pollen.usda.gov/Reference_Collection/Reference_Collection.htm):

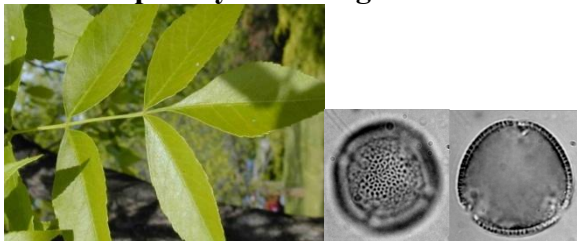
#### **Chenopodium album – lambsquarters**



#### **Melilotus officinalis - yellow sweetclover**



#### **Fraxinus pennsylvanica - green ash**



**10. ASSESSMENT:**

Older students can be required to create their own collection. The collection can be assessed based on proper care and labeling or completeness of a set of goals.

**11. EXTENSION IDEAS:**

Students can research the geographic range of plants, the history of the discovery of the plant, and the uses of plant through history.

**12. SCALABILITY:**

Students of all ages can collect plants with proper supervision.

**13. REFERENCES AND SOURCES FOR ADDITIONAL INFORMATION:**

Much of this information was taken from the *Pollen as Indicators of Source Areas and Foraging Resources* website

[http://pollen.usda.gov/Reference\\_Collection/Reference\\_Collection.htm](http://pollen.usda.gov/Reference_Collection/Reference_Collection.htm).

*Pollination Ecology: Field Studies of Insect Visitation and Pollen Transfer Rates*, by Judy Parrish (<http://tiee.ecoed.net/>).

**14. LIST OF EXPERTS AND CONSULTANTS**

Paul Alaback, an expert in local plants and phenology

**15. EVALUATION/REFLECTION BY FELLOWS AND TEACHERS OF HOW IT WENT**

This activity has not been tested.