Collecting, Drying, and Storing Chestnut Anthers Version 2.0

Paul Sisco
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Following is a step-by-step guide to collecting, drying, and storing chestnut anthers. This closely follows the method used at the Meadowview Research Farms in 2008.

Most important points:

1. Male catkins are best collected when in **prime** condition – in full bloom with **yellowish-white** cast and a strong, pungent aroma. If they are turning brown, they are past prime.
2. The last male catkins to bloom are the ones on the ends of bisexual flowers – those having a female flower at the base. This helps extend the male catkin-collection season.
3. The anthers must be dried overnight in **open** vials in a **sealed** can with **desiccant** in a refrigerator. **This drying is a critical step.**
4. After drying, the vials need to be sealed and kept in a cool, dry place until use.

Equipment needed:

1. Pill vials. These are available in many sizes from your local pharmacy. Be sure to get a **solid (no holes)**, **screw-on** cap if possible. Photo below is of a solid, pop-on cap type. I like a style of cap that is easy to pour the anthers on for pollination and then easy to get the anthers back into the vial.

2. Desiccant capsules help keep the pollen dry. At TedPella.com I order product # 19956, $35.14 for a box of 100 capsules.
   http://www.tedpella.com/desiccat_html/descant.htm#anchor736580

3. Glass plate or other smooth surface for collecting the anthers. I got a large, thick glass plate at a local glass supply outlet. At Meadowview, they use a smooth table-top that can be easily wiped down between each pollen sample.

4. Ethanol (grain alcohol) for wiping down the glass plate and for flaming the tools that come in contact with the anthers. The local liquor store sells 95% ethanol under brand names such as Everclear®

5. Single-edged razor blades for moving anthers around on the smooth surface. One source is the paint supply department at Loew's.

6. New, never-used gallon paint can for storing vials in the refrigerator. These are also found in the paint departments of home supply stores.

7. DampRid® or similar product for removing moisture from the air. It is available in quart or half-gallon milk-carton-style packaging at our local home improvement store (Lowe's, Home Depot, hardware store).

8. Parafilm® is helpful for spreading over the top of open vials during the pollen-drying process. It is easy to stretch over the tops of the vials, and holes are easily punched in it for air exchange. It is available from scientific supply houses. At Fisher Scientific on-line, the order number is 13-374-16 for a 2" x 250' roll, costing $28.67 plus shipping.

9. A small sifter is useful for separating the anthers from trash. I found a good one at the local Wal-Mart store

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10. A trigger-type lighter (seen in the photo above) is helpful for flaming. These are found near charcoal, lighter fluid, and other supplies for outdoor cooking in grocery stores and hardware stores.

11. Labeling tape and fine-point, water-proof marker for labeling the vials.

12. Splatter screens for removing anthers from catkins. These are available at Bed, Bath, and Beyond. $9.99 for a set of three.

Step-by-step Directions:

1. Collect fresh branches of male catkins, preferably in the morning. Choose ones that are in full-bloom, yellowish-white, and with a strong aroma. If you will be transporting the catkins for several hours or shipping them overnight to someone, clip off the individual catkins and stuff them into a bag labeled with the tree name. If catkins need to be stored overnight, keep them in the refrigerator. (Alternative used by Hill Craddock is to leave the catkins out overnight on the glass plate, so that new anthers will extrude in the morning.)

2. Wipe down the glass plate or other clean surface thoroughly with 70% ethanol.

3. Make sure the plate is dry before putting the catkins on it.

To dilute the 95% to 70% ethanol, use the formula:

$$\frac{\text{Desired total volume} \times 0.70}{0.95} = \text{volume of 95\% alcohol to add to water}$$

Example: To make 100 mls of 70% ethanol using 95% ethanol:

(100*0.7)/0.95 = 73.7 mls 95%; then bring up to 100 mls with water

In other words, add 73.7 mls 95% to 26.3 mls water = 100mls 70%

4. When you get ready to collect anthers, lay the catkins out on the glass plate or other smooth surface. Be sure to retain the labeled bag for identification of the pollen.

5. Strip the catkins off by rubbing the catkins against the splatter screen over the glass plate, making sure that you have **washed your hands thoroughly** before handling a new pollen source.

6. Collect the anthers together with a clean single-edged razor blade or other sharp instrument and either directly lift them into the sifter using the flat edge of the blade or scrape them onto a new paper lunch bag, creased down the middle, and then pour the mixture of anthers, filaments, and other catkin parts into the sifter. Sift the anthers onto a clean area of the plate and discard any trash remaining in the sifter.

7. Be sure to flame-sterilize the sifter and razor blade between each pollen source.

8. Collect the pollen into a fresh vial and label it with the pollen source and date of collection. Be sure to label the vial as well as the cap, in case the cap gets separated from the proper vial.



9. **Very important step:** the pollen must be dried overnight in a sealed, refrigerated can with **fresh** DampRid® or other moisture-absorbing compound. More expensive versions of drying compounds such as Drierite® can be purchased from Scientific Supply houses. These often contain colored indicator dyes to let one know when the drying compound has absorbed all the moisture it can. The color turns from blue to pink when the drying compound becomes saturated with water.

   Put a 1 – 2" layer of fresh, active drying compound into the bottom of the clean paint can, place the open vials into the compound so they stand up straight, and carefully seal the can by tapping on the lid. It helps to prevent accidental spilling of the anthers if one can stretch Parafilm® over the top of the open vials with punched holes in it. Use a wide needle or other punching device to punch lots of holes for air transfer. Put the sealed can into a refrigerator overnight.

10. The next morning, open the can, put a fresh, clean drying capsule in each vial, quickly replace all the proper caps on the vials, and seal the caps with Parafilm® stretched and sealed around each one. If you have labeled the caps, make sure you get the proper cap to the proper vial. **Don't mix up your pollens!** Then reseal the paint can and put it back into the refrigerator.



11. Replace the drying compound in the bottom of the can as needed.

12. When taking pollen vials to the field for use, put them in a small cooler with an ice or freezer pack until you are ready to use the pollen.