Collecting quillworts to preserve DNA

Materials (links are just examples, can be substituted)

- Coin envelopes <u>https://www.amazon.com/Quality-Park-Envelopes-Brown-</u> Kraft/dp/B000Y50ZD8/ref=dp prsubs 2?pd rd i=B000Y50ZD8&psc=1
- Plastic bags <u>https://www.amazon.com/dp/B0781BDHZJ?psc=1&ref=ppx_yo2_dt_b_product_details</u>
- Silica gel
 <u>https://www.amazon.com/gp/product/B071S914ZP/ref=ppx_yo_dt_b_asin_title_o07_s</u>

 <u>01?ie=UTF8&th=1</u>
- Alcohol wipes <u>https://www.amazon.com/Sanitizing-Alcohol-Fragrance-Individually-</u> <u>Portable/dp/B014EVXZ9S/ref=sr 1 20?keywords=alcohol+wipes&qid=1638887236&sr=</u> <u>8-20</u>
- Ziploc bags
- Paper towels and water (purified or distilled if possible) for cleaning leaves
- Pen/marker

For systematic work, I try to sample 5 individuals selected pseudo-randomly from each population. For populations genetics, sampling 25-30 individuals is standard, but quillwort populations are often smaller than this. Sampled individuals can be flagged and labeled for further study (to collect material to determine ploidy, spores, etc.).

Procedure

- Select sporophylls (leaves) that are young and healthy looking. These are towards the center of the plant, generally a bright green color. Avoid older, damaged, or sick-looking leaves.
- Cut a minimum of ~3 in. (7.5 cm) of leaf (combined length if individual leaves smaller). If possible, collect up to 12 in. (30 cm) per plant to have some backup material.

- 3. Remove all soil, algae, or other contaminants from leaf and wipe with alcohol wipe until clean.
- Cut leaf into ~1 in. (2.5 cm) sections and put in coin envelope. Seal when finished. Label envelope with sample ID, collection site info, date, etc. Put envelope in plastic bag with silica gel.
- Keep samples away from heat and direct sun, especially during initial drying. Room temperature (~70-75 F) is OK.
- 6. After 1-2 days, check that silica gel isn't saturated (color changes from orange to dark blue-green). If all silica gel in a bag has changed color, replace it. Silica gel can be dried and reused by warming in an oven at 200 F for 30-60 minutes. Sealed plastic bags still leak air over time, so for long term storage put sample bags in a larger Ziploc bag or an air-tight container. Samples can also be stored in a freezer for an additional layer of preservation.