Collecting, Drying and Storing Specimens for DNA Analysis

DNA sampling consumes both time and money so efforts to increase the probability of success are encouraged. The following notes outline our ideal requirements and we ask that submitters of material aim to follow as much of this guidance as possible. This document should be read in conjunction with the DNA submission form (here).

Types of Fungi

Although most of our work so far has focussed on analysing fruiting bodies with gills, we are willing to consider other basidiomycetes including brackets and resupinates. We will also consider ascomycetes (jellies, cups, morels, truffles etc) but please note that we have had limited experience to date with this group.
At present, we are unlikely to accept plant parasites ('dots & spots'):

- Mildews (powdery and downy)
- Slime moulds (Myxomycetes)
- Moulds (Hyphomycetes)
- Water moulds (Oomycetes)
- Rusts (Pucciniales)
- Smuts (Ustilaginales)

Materials Required (but not essential):

- Sharp, sterile knife
- Food Dehydrator
- Lidded containers capable of being frozen
- Glassine envelopes
- Voucher Packets

Collecting

Efforts to avoid contamination from other fungal bodies should be made by collecting specimens in separate containers. Geoffrey Kibby suggests that specimens should be individually wrapped in kitchen foil but we appreciate that not all mycologists carry such material with them in the field.

Drying & Freezing

Thorough and prompt drying of a specimen is a key component to a successful outcome. It is essential to prevent mould growing on the specimen, therefore specimens should be dried as soon as possible after collection.

Drying methods

For larger fungi, the drying process can be accelerated by cutting the sample into smaller pieces with a dry, sharp, sterile knife which has been wiped clean with isopropyl alcohol or bleach. A shortened drying time reduces the possibility of rotting.
The best DNA results come from using a piece taken from the hymenium (spore-producing surface). Whilst only a small sample is required for extraction the piece received by us needs to be large enough to be easily visible and capable of manipulation. A larger piece should be preserved for subsequent microscopic examination.

Whilst traditional wisdom has been that drying should ideally be done at no warmer than 45°C, recent research indicates that fungal DNA is actually far more resilient than previously thought and concludes that higher drying temperatures up to 70°C should provide excellent quality of materials for DNA extraction:

Drying method 1

For smaller fungi or slivers of larger ones, place the sample on a piece of paper on top of a warm radiator, or in a warm conservatory or airing cupboard for several days. Unfortunately, this method can increase the risk of airborne cross contamination.

Drying method 2

Alternatively for smaller fungi or slivers of larger ones, use silica gel in a small container at room temperature for 1-1.5 days.Silica gel is available online, and can be dried and reused.

Drying method 3

For larger fungi, use a food dryer (dehydrator). Basic ones are inexpensive (internet search 'food dehydrator' e.g. Oypia Food Dehydrator for about £35) but we could consider grant-support for those who would benefit from their use. These provide effective drying by exposing the sample to moving air at a fairly low temperature. The aim is to achieve a balance between the effective extraction of water and preventing contamination by other organisms.

Drying method 4

Also for larger fungi, oven dry at 70°C for 3-4 hours (or 60°C for 7-8 hours).

Freezing post drying

Whichever method you use to dry the specimen, the dried specimens should then be placed in a lidded container and frozen for 2 days (in a domestic freezer at -18°C) to kill any bugs. The frozen specimen should then be thawed at room temperature, using absorbent kitchen paper (ideally in conjunction with silica gel to avoid condensation issues).

Storage

After drying, freezing and thawing, specimens should be placed into a labelled envelope (ideally a semi-transparent glassine type), together with notes identifying the sample. The envelope or tube should then be placed inside a folded paper voucher packet on which all the relevant details have been written. The voucher packet is available for download here and should be printed on A4 paper (skip page 2 to save ink if required). Voucher packets can be stored effectively in sealed polythene bags with silica gel packets in air-tight food storage boxes.

Provision of supporting data

Essential:

- Date of collection, site name and grid reference, name(s) of person submitting the specimen, finder, and identifier, together with substrate and/or association if known
- Likely species (or genus) and any reasons for initial ID
- Names of any books, papers and/or other mycologists consulted
- Description of method used to preserve the specimen
- Reasons why you think the specimen may be a good candidate for DNA sequencing (e.g. potentially new to Norfolk, new to GB, very few records on the national database etc)

Additional:

- Description of specimen, including details of size, shape, smell, habitat, spore size, colour, ornamentation etc.
- Images of the specimen in situ and/or at home, in particular any those showing features that support your provisional ID
- Notes and images of spores and/or spore print
- Images of microscopic features that support your provisional ID

We know from experience that DNA work is a very steep learning curve and appreciate that the above requirements may seem very demanding; however, each of the above steps are likely to increase the successful sequencing of your specimen and provide essential data and evidence for future use of the results. Please don't be put off putting forward interesting specimens and if you have any questions feel free to contact any of the DNA Team for help and advice.

NFSG DNA Team October 2023