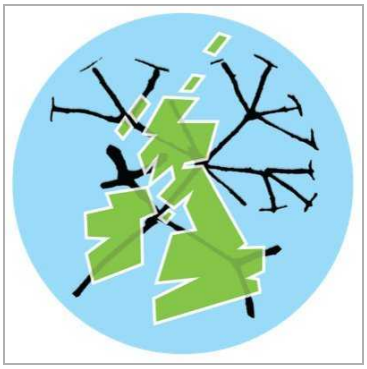

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**Protocol status:** Working
This is a working protocol that may be subject to changes in the future.

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# 7.1: Taxon Group: Crustacea - Decapoda

In 1 collection

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ABSTRACT

This is part of the collection "DToL Taxon-specific Standard Operating Procedures (SOPs) for Marine Metazoa", lead by the Other Metazoa Working Group. The SOP collection contains guidance on how to process the various marine Metazoa species within the scope of the Darwin Tree of Life project. The guidance specifically refers to the tissue samples needed for DNA barcoding (which takes place at the Natural History Museum (NHM) and at the Marine Biological Association (MBA)) and outlines the dissected tissues required for whole genome sequencing, which takes place at the Wellcome Sanger Institute. Every specimen is submitted for DNA barcoding first before potentially being sent to the Wellcome Sanger Institute.

In this instance, the subphylum Crustacea (SOP number 7) has been sub - divided into three particular groups; Decapoda (1), Peracarida (2) and Cirripedia (3). These other Crustacea SOPs can be found in the Marine Metazoa collection contents.

**Definition:** The Decapoda or decapods are an order of crustaceans within the class Malacostraca, including many familiar groups, such as crabs, lobsters, crayfish, shrimp, and prawns.

**Including:** Brachyura, Anomura, Caridea, Astacidea, Axiidea, Caridea, Gebiidea and Polychelida.

**Excluding:** Specimens smaller than 5mm.

See the Guidelines for important details and checklists.

GUIDELINES

**Keywords:** decapoda, decapods, crustacea, Darwin Tree of Life project, Wellcome Sanger Institute, Natural History Museum, whole genome sequencing, DNA barcoding

**Field sampling:**

**Guidance regarding regulatory compliance:** Prior to visiting a collection site , it must be determined whether the site is located within a Marine Protected Area and, if so, ensure that sampling permission has been obtained (on a site-by-site basis) from the relevant conservation body.

1. Environment to be sampled: Marine, brackish.

2. Trap/method of sampling: Individual collection by hand, intertidally or by diving. Possible incidental capture by dredge/trawl/grab.

After collection it is recommended to keep specimens kept in aerated or stirred cooled (preferable) or ambient seawater. Seawater can be replaced every few hours in place of aeration.

Specimens may require processing within a few hours to avoid deterioration.

*This SOP can also be used for freshwater decapod species if required.*

> Note
>
> Each specimen, regardless of species, must have its own relevant unique identifier (e.g. QR code) which will be attached to any subsequent tubes, genome or barcoding results.

**For genome sequencing:**

3. Under the Animal Welfare (Sentience) Act 2022, crabs and lobsters may not be sampled live. It is possible for other decapod groups to be sampled live.

All groups must be anaesthetised by cooling on ice, in ice and seawater slurry or in a domestic freezer. Once sampled, preserve the rest of the animal immediately.

Other approved methods such as Crustastun may be used if available.

For crabs and lobsters, nerve centers should be destroyed after anaesthetisation. Sample and preserve tissue samples immediately.

**Photography:**

4. Photograph dorsal, lateral and ventral views.

Not all views will be feasible for every species.

5. The image should be taken in the highest quality resolution -a macro lens is recommended.  The photos should be of high enough resolution to be diagnostic, when possible.

Photograph to include a unique identifier (e.g. QR code, specimen barcode) where possible; where no voucher specimen parts are retained the photograph will serve as voucher and should include identifying features.

**Dissection for DNA barcoding:**

6. The recommended tissue types for DNA barcoding are listed below:

Note

**Pleopod** - For groups with large pleopods, i.e., Axiidea, Caridea, Gebiidea and Polychelida.
**Pleonal (abdominal) tissue** - If not enough material is available, use non-diagnostic pereiopod (leg).
**Pereiopod (leg) tissue** - Dissect flesh or ensure appendage is free of epibiota.

Once the tissue for barcoding is removed, put the tissue in 100% ethanol. The rest of the frozen/live organism can then be dissected.

**Dissection for whole genome sequencing:**

7. The recommended tissue types for whole genome sequencing are listed below:

Note

**Pleopod** - For groups with large pleopods i.e., Axiidea, Caridea, Gebiidea and Polychelida.
**Pleonal (abdominal) tissue** - If not enough material is available, use non-diagnostic pereiopod (leg).
**Pereiopod (leg) tissue** - Dissect flesh or ensure appendage is free of epibiota.

Dissect up to ten, lentil-sized pieces in separate tubes.

Tissue should be frozen at at least -80°, for example in dry ice, a liquid nitrogen charged dry shipper or in a -80° freezer.

**Storage of frozen tissue:**

8. If barcoded tissue passes the DNA barcoding stage, subsequent frozen tissue of specimen to be sent to Wellcome Sanger Institute.

> Note
>
> Please refer to DNA barcoding SOP v2.1.

9. Leftover tissue from specimens must be sent to NHM for vouchering and long term storage.

**Storage of voucher:**

10. Vouchers to be sent to/kept at NHM.

11. Vouchered tissue to be eventually preserved in 70-90% ethanol.

**Photo guide below:**

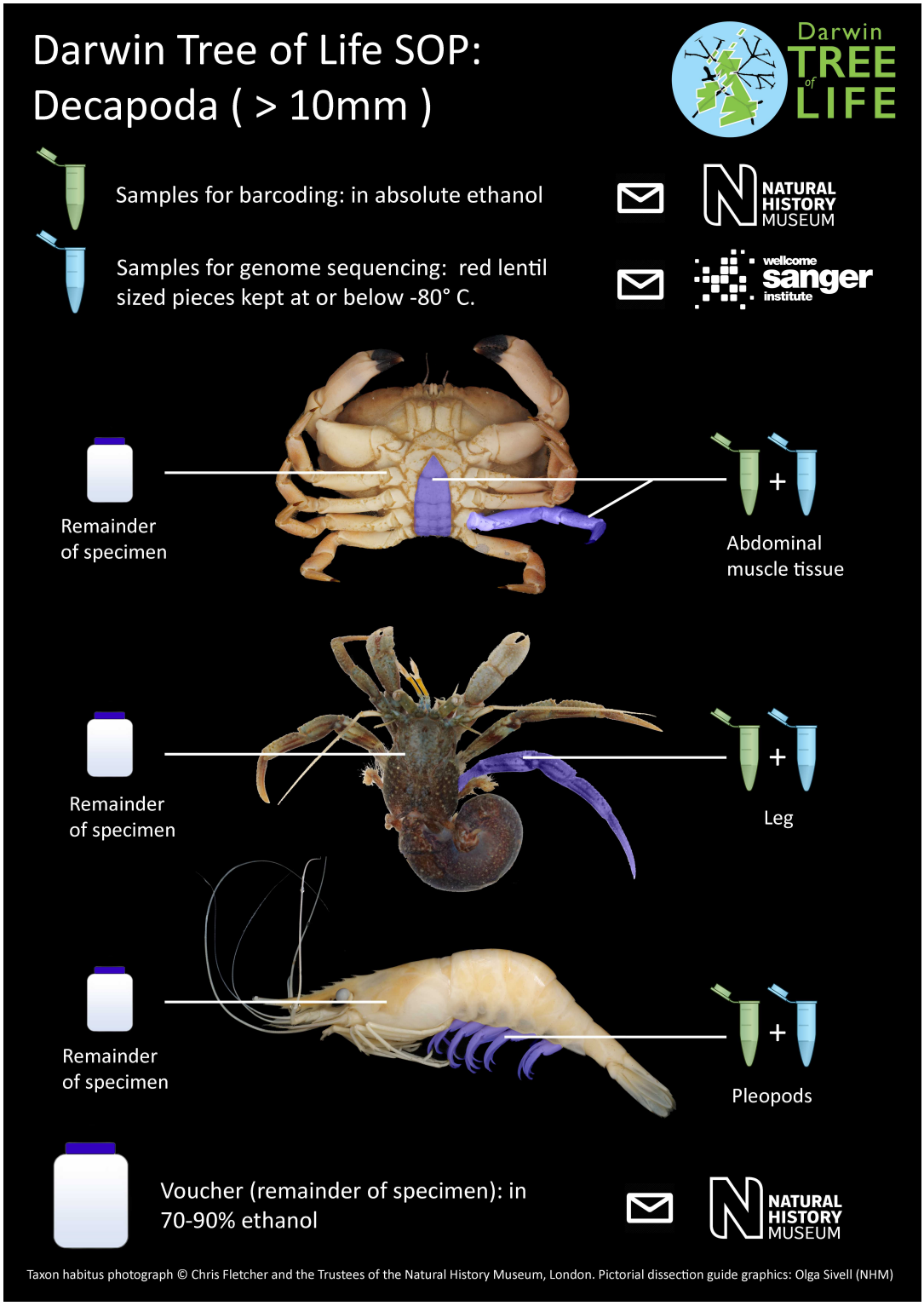


Photo guide assembly: Chris Fletcher