



MAY 19, 2023

## 7.2: Taxon Group: Crustacea - Peracarida

In 1 collection

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Darwin Tree of Life

OPEN ACCESS

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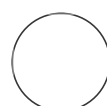
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**Protocol status:** Working  
This is a working protocol that may be subject to changes in the future.

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Inez Januszcak

### 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 superorder Peracarida is a large group of malacostracan crustaceans, having members in marine, freshwater, and terrestrial habitats. They are chiefly defined by the presence of a brood pouch, or marsupium, formed from thin flattened plates borne on the basalmost segments of the legs.

**Including:** Isopoda, Amphipoda, Cumacea, Tanaidacea, Mysidacea.

**Excluding:** Spelaeogriphacea and species/specimens under 5mm.

See the Guidelines for important details and checklists.

Photo guides available for specimens over 10mm and under 10mm.

**Keywords:** Peracarida, Peracarids, Crustacea, Darwin Tree of Life, Wellcome Sanger Institute, whole genome sequencing, DNA barcoding, SOP, Standard Operating Procedure

## GUIDELINES

### 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/rawl/grab.

#### Note

Use largest example of a species to maximise identification confidence and material available for sequencing.

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

If possible, extract specimens prior to sieving to avoid damage to specimens.

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

*This SOP can also be used for freshwater and terrestrial 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. Specimens can be anaesthetised by cooling on ice, in ice, in seawater slurry or in a domestic freezer, and sampled alive.

### 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**

**Pleopods** - If not enough material use pereopods (legs).

**Antenna 1** - For caprellid amphipods only. Take one and use half for DNA barcoding

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**

**Pleopods** - If not enough material use pereopods (legs) or whole organism.

**Antenna 1** - For caprellid amphipods only, use other half of antenna used for DNA barcoding. If not enough material use pereopods (legs) or whole organism.

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

Tissue should be frozen at at least -80°, for example in dry ice, in 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 vouchers and long term storage.

**Storage of voucher:**

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

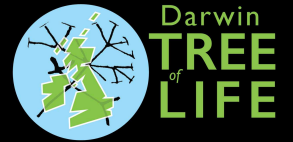
11. Vouchered tissue to be eventually preserved in 70-90% ethanol.

**Note**

If additional specimens (of the same species and from the same sampling event as the dissected specimen) are collected, please keep these intact and send to NHM in ethanol to act as corresponding voucher specimens.

**Photo guides below:**

# Darwin Tree of Life SOP: Peracarida (> 10mm)



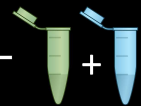
Samples for barcoding: in absolute ethanol



Samples for genome sequencing: red lentil sized pieces kept at or below -80° C.



Remainder  
of specimen



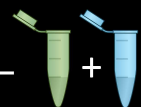
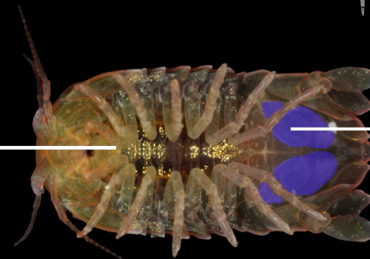
Antenna 1 (x1)

Remainder  
of specimen



Pleopods

Remainder  
of specimen



Pleopods



Voucher (remainder of specimen): in  
70-90% ethanol



Taxon habitus photograph © Chris Fletcher and the Trustees of the Natural History Museum, London. Pictorial dissection guide graphics: Olga Sivell (NHM)

Photo guide assembly: Chris Fletcher

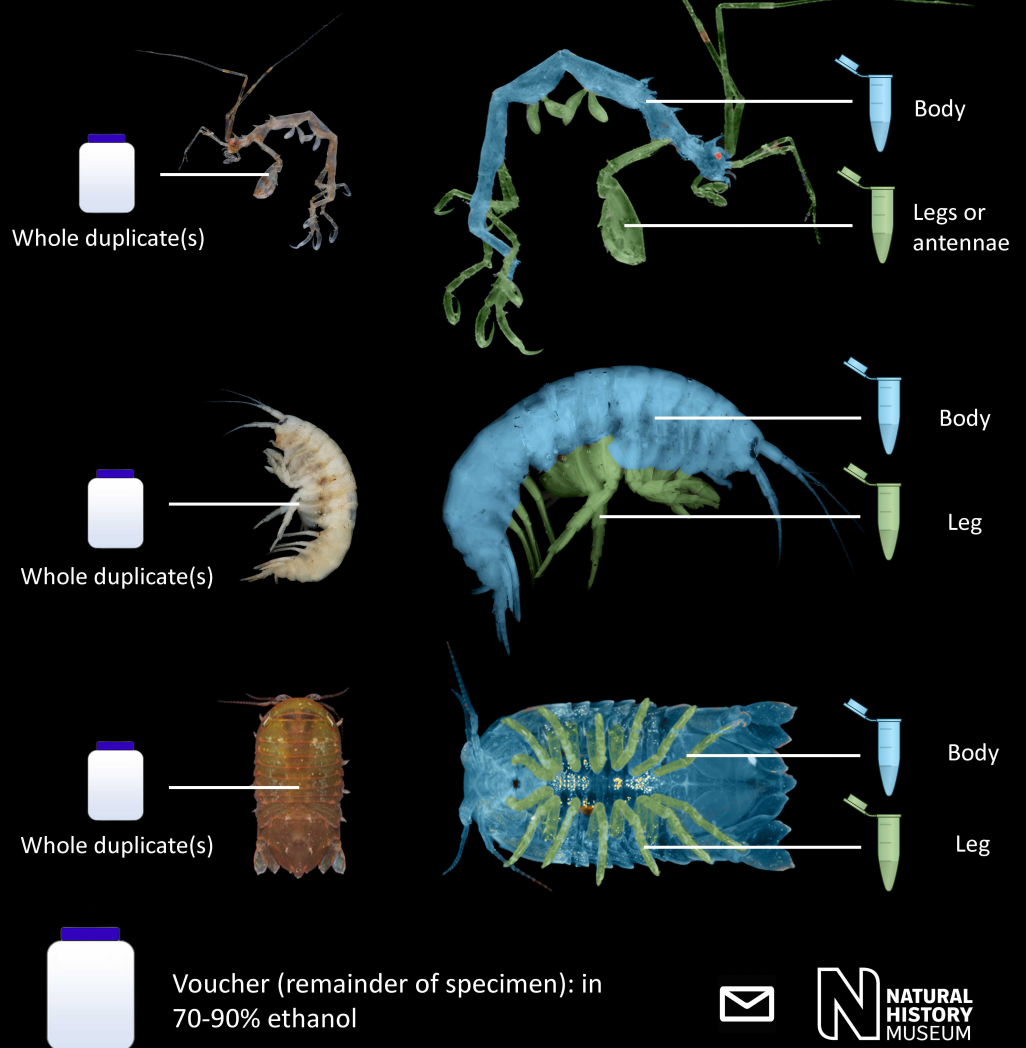
# Darwin Tree of Life SOP: Peracarida (< 10mm)



Samples for barcoding: in absolute ethanol



Samples for genome sequencing: red lentil sized pieces kept at or below -80° C.



Taxon habitus photograph © Chris Fletcher and the Trustees of the Natural History Museum, London. Pictorial dissection guide graphics: Olga Sivell (NHM)

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