Procedures for sampling deep sea corals for DNA studies

I would like samples of deep sea coral species to contribute to ongoing studies of connectivity/larval dispersal in the southern ocean.

Importantly, this study relies on getting as many samples of corals from as many locations as possible, so that we can compare genetic variation;

a) between individuals from the same site
b) between populations from different depths/sites in the same location, and
c) between locations.

Species I am especially interested in…

Scleractinians (stony corals)

Solenosmilia variabilis (colonial, branching coral)
Madrepora oculata (colonial, branching coral)
Desmophyllum dianthus (solitary coral)
Endopachys greyi (solitary coral, often with polyp buds on the sides)
Stephanocyathus spp. (solitary coral)

Antipatharians (black corals)

Parantipathes spp. (bottle-brush black coral)
Stichopathes spp. (black coral – whip)

SEE ATTACHED PHOTOS FOR MORE DETAIL

What to do with the samples…

1. To ensure the DNA is kept in good condition, make sure catch samples are stored in sea-water on deck, out of the sun and kept cool. It often takes some time to process a catch, and the way samples are held during this period is critical for the quality of the DNA. If a cool room is available, all the better.

2. Identify if the coral has any live tissue….
   - For the **branching scleractinians** the best tissue will be associated with the tip of the branches where the polyps are located.
   - For **solitary corals**, the tissue usually recedes down into the centre of the polyp.
   - For **antipatharians**, the tissue is usually apparent as a white or opaque covering on the outside of the skeleton itself.

3. If there is tissue.…
   - MAKE SURE YOU HAVE CLEAN EQUIPMENT!! It is really important to avoid contamination– both between coral samples and from other species (including humans).
   - Wash all tissue sampling equipment that comes into contact with the coral with boiling hot water or bleach between specimens and dry them off with

clean paper towel. Use clean gloves for each sample if you have to physically handle the corals.

- For the **branching scleractinians**, cut off around 5-6 polyps with side-cutters or similar (this includes the tissue and the skeleton) and place in a 5ml+ sample vial with appropriate label inside the vial. Preserve with AT LEAST 75% ethanol. Make sure the remaining bits of the colony are also tagged with the same reference as that used for the vial, and preserved (in alcohol) for formal species identification.
  NOTE. If there are multiple pieces of a species of branching coral in a sample, then please take tissue samples from each piece (or as many as possible) for DNA analysis, and try to keep the main pieces labelled separately for ID purposes. It is possible that many of the pieces that come up from a site will be from the same colony, but to be safe, treat them as separate for the purpose of the collections.
- For the **solitary corals**, preserve the entire polyp in AT LEAST 75% ethanol. Its OK to put multiple polyps of a species from the same dredge/grab sample in a jar together. Remember to put a label inside the jar
- For **antipatharians**, snip off 4-5cm long pieces of the colony (ordinary scissors are usually strong enough for this) and place into sample vials (for the bottle brush corals, usually only 1 piece will fit in a 5ml vial; for the whip forms, collect 3 or four lengths from the colony to put in the vial). Preserve with AT LEAST 75% ethanol. Place label inside the vial. Make sure the remaining bits of the colony are also tagged with the same reference as that used for the vial, and preserved (preferably alcohol) for formal identification.
- When labelling, please make sure enough information is recorded to enable me to know the exact location the sample came from i.e. lat/long and depth as a minimum.

What to do if you're not sure…

- If you're not sure about the species ID, collect it anyway.
- If you're not sure if there is any live tissue, collect it anyway.
- If you're unsure about taking a genetic sample, at least make sure whatever sample is preserved goes in alcohol – that way I may still be able to use it.
- If you think there are too many bits of coral in a sample and that I won't need it all… COLLECT THEM ALL ANYWAY!!! I will definitely use them!
- If you have any questions …email me at karen.miller@utas.edu.au .

THANKS HEAPS!

Photos of target coral species to assist with identification…

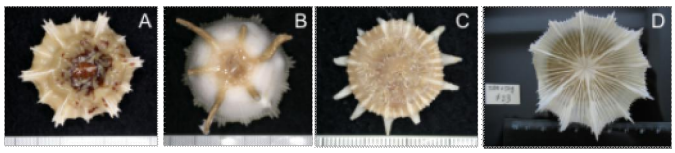
Stony Corals....

Solenosmilia variabilis
- branching coral
- forms large anastamosing colonies

Madrepora oculata
- branching, colonial coral
- distinguished by alternating polyps on branches

Desmophyllum dianthus
- solitary coral, attached to hard substrata
- up to 8cm diameter

Endopachys greyi
- solitary coral, often with polyps budding from the side
- free-living, small (<3cm diamter)



Stephanocyathus spp.
- solitary coral, free living
- often has "feet like" protrusions
- size up to ~ 8-10cm diameter

Black corals...

Parantipathes spp.
- bottle brush coral

Stichopathes spp
- whip coral.