

Spicer & Morley:

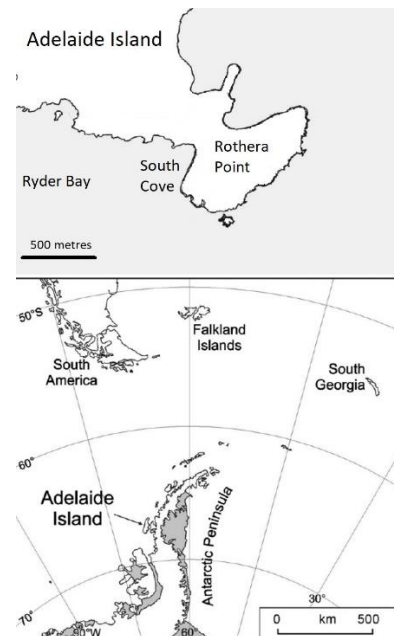
Will giant polar amphipods be first to fare badly in an oxygen-poor ocean? Testing hypotheses linking oxygen to body size.

Supplementary materials 1

Amphipod collection, species and habitat (with particular reference to oxygen) details

Collection Site

Collection of animal material was carried out on four separate days during the austral summer; 21st January and the 6th, 8th and 11th February, 2007. Amphipods used in the experiments reported here were collected by divers from beneath cobbles/boulders at a depth of 6 - 8 m from South Cove, Ryder Bay, Adelaide Island, Graham Land, Western Antarctic Peninsula (lat. 67°34'11"S, long. 68°08'89" W) (Fig. S1.1). The sea floor here is primarily natural bedrock and rubble (granite), with variable patches of fine biogenic and mineral debris (Fig. S1.2 below). The site has a score of 0 on the Wentworth [1] Classification Scale that quantifies rugosity of substrate.



It is a south-facing site which means that the main disturbance encountered is ice scouring and impact. The collection sites, which comprise IBIS sampling grids come under the Rothera Oceanographic and Biological Monitoring (RaTS) programme.

Figure 1.1 Location of the collection site (South Cove) and Rothera point on Adelaide Island (top panel) and Adelaide Island relative to the Western Antarctic Peninsula (bottom panel)

The sites are typically covered by winter fast ice for *circa* 3 months each year. Water temperatures range from -1.9 to +1.5 m at 15 m depth (RaTs), with the daily and weekly variability in December five times that in July.

Collection method

Amphipod collection was by means of the Souster and Yates Suction Sampler [2]. The bespoke device is driven by a submersible bilge pump (Xylem ISO 8849) which filters 31.5 L.min⁻¹ of sea water through a Professional hand held net Bag (EFE & GB nets, Totnes, 1 mm mesh size) in which the amphipods are retained. Divers would turn over larger boulders and vacuum the animals beneath them with the sampler. The Sampler was returned to the laboratory within 20 min of sampling, the net bag removed, and the amphipods transferred (using plastic tea strainers) to the aquarium facilities in the Bonner Laboratory, Rothera field station.

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Animal material collected

The four species presented in Fig S1.2 are those chosen for our study and were extremely common beneath the cobbles and the stones in South Cove (Fig. S1.3A), and are also common in similar habitats on the Antarctic Peninsula and beyond.



Figure S1.2 Species investigated in this study. (From top to bottom), Paraceradocus miersi, Prostebbingia brevicornis, Schraderia gracilis, Probolisca ovata

***Paraceradocus (Megamoera) miersi* (Pfeffer, 1888)**

(Family Maeridae [n.b. De Broyer et al.[3] places it in the family Melitidae]; Superfamily Hadzioidea; Suborder Senticaudata)

The individuals we used match very closely the pictures of this species presented in Souster [2], the identity of which was confirmed for Dr Souster by Dr. Anna Jazdzewska (p. 48)

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P. miersi is a detritivore/scavenger which lives under stones in burrows in sediment which they excavate ([4]; also see Fig. S1.3C below). It is a large species and occurs in east and west Antarctic Provinces and South Georgia district (0 - 344 m depth) [3].

Prostebbingia (Pontogeneilla) brevicornis (Chevreux 1906)

(Family Pontogeneiidae; Superfamily Calliopoidea; Suborder Senticaudata)

P. brevicornis occurs in west Antarctic province, South Georgia district and the sub Antarctic Islands province (0 -310 m depth) [3]. The individuals we used match very closely the pictures of this species presented in Souster [2], the identity of which was confirmed for Dr Souster by Dr. Anna Jazdzewska (p. 48). Interestingly she records the 'species' as '*P. brevilongicornis*'. In the manuscript we have used *P. brevicornis*, though it should be kept in mind that an expert in the systematics of the group opted for this dual 'identity'. *P. brevicornis*, together with *S. gracilis* (below) have been recorded previously from coarse sediments where macroalgae present, and are considered herbivores and detritivores [5].

Schraderia gracilis Pferrer, 1888

(Family Pontogeneiidae; Superfamily Calliopoidea; Suborder Senticaudata)

S. gracilis occurs in east west Antarctic province, South Georgia district and the sub Antarctic Islands province (0 - 338 m depth) [3]. The individuals we used match very closely the pictures of this species presented in Souster [2], the identity of which was confirmed for Dr Souster by Dr. Anna Jazdzewska (p. 48).

Probolisca (Metopa) ovata (Stebbing, 1888)

(Family Stenithoidea; Superfamily Amphilochoidea; Suborder Amphilochoidea)

The smallest species investigated is distantly-related to the other amphipods and occurs in west Antarctic province, South Georgia district and the sub Antarctic Islands province (0-570 m depth)) [3].

Environmental oxygen (O₂) status at the collection site

Unfortunately the RaTS monitoring scheme does not include dissolved O₂ as one of the parameters it measures (see the official website for what it does cover at <https://www.bas.ac.uk/project/rats/#about>).

The O₂ saturation of water overlying the boulder field and of water drawn from within presumed *Paraceradocus* burrows, excavated beneath boulders slightly embedded in the sea bottom (Fig. S1.3), was measured using a Presens system on water samples collected on the 6th and 8th February 2017 as follows.

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To obtain burrow water the following procedure was carried out. A team of divers carrying 4 x 20 mL thick plastic syringes each fitted with a 30 cm length of gas impermeable tubing (Tygon[®]) entered the water and descended to the boulder field. Firstly divers would locate possible burrows beneath large boulders either by making visual contact with *Paraceradocus* at the entrance to the burrow, or by coming to recognise the distinctive depression in the sediment below particular boulders (Fig. 1.3B). The tube attached to the syringe would then be carefully inserted into the potential burrow, to a depth of > 10 cm avoiding disturbance of any sediment. Slowly the syringe was filled with burrow water. Immediately the syringe was full of water, the tube was quickly extracted from the burrow and the end was sealed with a plastic clip to avoid leakage from the burrow water sample. The boulder was then overturned so that any amphipod species in the burrow or in other spaces beneath the rock could be checked visually and their presence and behaviours noted (Fig. 1.3C). Sealed water samples were returned to the surface, and analysed on the boat < 7 min after collection.

Water overlying the boulder field was sampled from the dive boat, from a depth of 30 cm below the surface, and the O₂ content determined < 30 sec after collection.

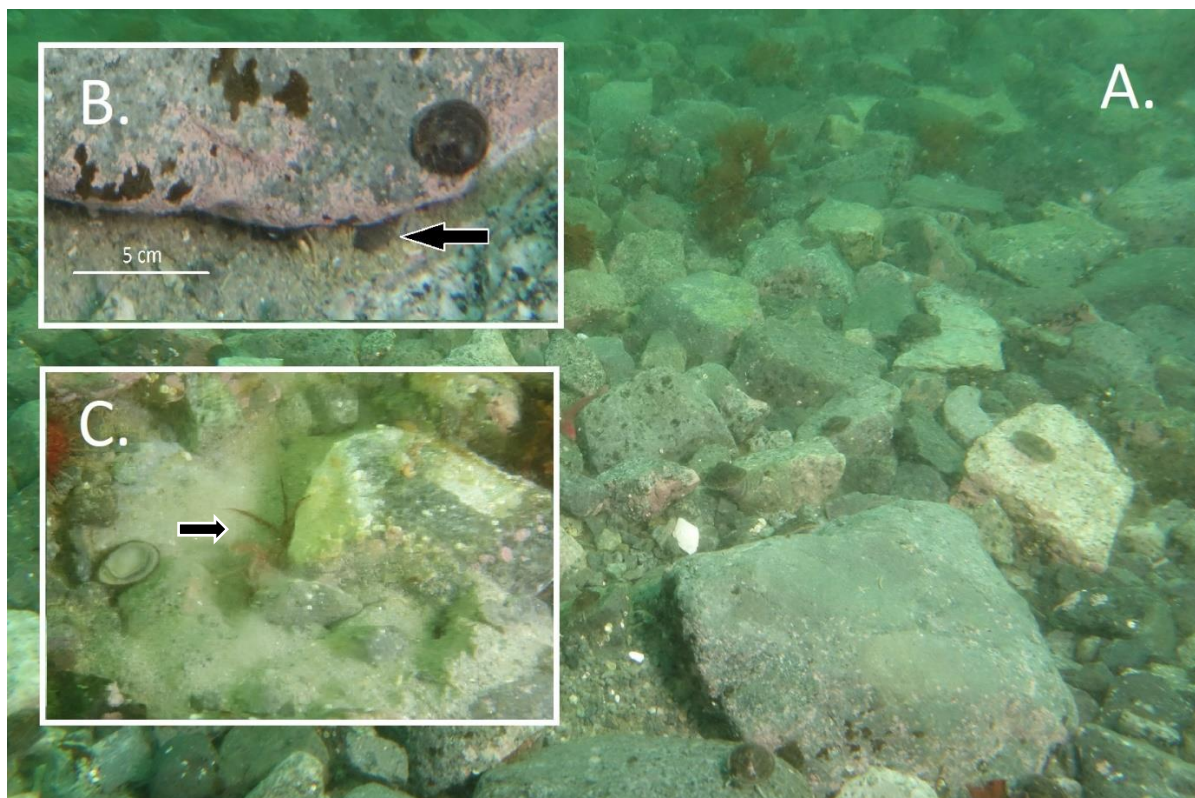


Figure S1.3 The sampling site comprised cobbles and boulders in relatively shallow water around 4-6 m depth (A). *Paraceradocus* burrows could be located either by the presence of the animal at the mouth of the burrow or by the depression in the biogenic and mineral sediment beneath, and to the side of, the boulders (B – arrow marks opening to burrow). When the boulder was removed, it was often possible to see the *Paraceradocus* in its typically inverted position, still lying within its burrow (C – arrow marks animal in its burrow).

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Presented in Figure S1.4 is the O₂ saturation (% a.s. or air saturation) of sea water collected from different conditions in South Cove. During the sampling period the overlying sea water was hypersaturated with O₂. The O₂ concentration of burrow water from confirmed *Paraceradocus* burrows was substantially lower than overlying sea water but was still around 100 % a.s. saturation. The O₂ content of water filled spaces beneath boulders lying on substrate with a visibly high biogenic content were substantially lower than that in the burrow water.

In summary while the overlying water was oversaturated with O₂, O₂ conditions beneath the boulders where the amphipods were collected ranged from mildly hypoxic (beneath boulders) to normoxic (in burrows).



Figure S1.4 O₂ saturation (as % air saturation) of sea water from different conditions in South Cove (8/2/17). Each point is one measurement.

Conclusion: While *Paraceradocus* inhabiting their burrows experienced normoxic conditions smaller individuals or other smaller species living under boulders may have experienced exposure to mildly hypoxic conditions.

References

- [1] Wentworth CR 1922. A scale of grade and class terms for clastic sediments. *J. Geol.* **30**, 377-392.
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- [5] Jazdzewska AM, Siciński J. 2017. Assemblages and habitat preferences of soft bottom Antarctic Amphipoda: Admiralty Bay case study. *Polar Biol.* **40**, 1845-1869.