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Trophic ecology drives trace element concentrations in the Antarctic octopod community

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Abstract

Despite the Antarctic Ocean being considered a pristine environment, elevated trace element concentrations have been reported in many marine organisms. The Antarctic Ocean is particularly vulnerable to climate change, which can also affect the bioaccumulation of trace element concentrations in biota. While Antarctic octopods are key components of the regional food webs as prey for a variety of predators (e.g., seals, fish, and seabirds), their contamination state by trace elements remains largely unknown. This study investigated the trace element concentrations in relation to the trophic ecology in Antarctic octopods. Stable isotope values (δ^{13}C and δ^{15}N) and trace element concentrations (Ag, As, Cd, Co, Cr, Cu, Fe, Hg, Mn, Ni, Pb, Se, V, and Zn) were measured in eight different species (*Adelieledone polymorpha*, *Pareledone aequipapillae, P. albimaculata, P. aurata, P. charcoti, P. cornuta, P. felix,* and *P. turqueti*) sampled near Elephant Island, close to the Antarctic Peninsula. Stable isotopes of δ^{15}N varied among species, with significant differences between *A. polymorpha* and *P. aurata* suggesting potential niche segregation. Trace element concentrations also differed among species and with sampling depth, which likely reflects their trophic ecology. The data presented in this study provides the first insight into the trace element concentrations for these endemic octopods in this vulnerable habitat and their stable isotope values.

Keywords: Cephalopods, Cd, Hg, Southern Ocean, Antarctica, Stable Isotopes

Introduction

Antarctica is recognized for its stable environment, sheltering numerous endemic species, and unique geography, which makes it especially susceptible to climate change as highlighted by increasing temperatures (Gille, 2002) and CO_2 saturation (Le Quéré et al., 2007). It is the southernmost continent and contains 90% of Earth's ice masses, which make up about 70% of Earth's freshwater reservoir (Bentley and Giovinetto, 1992). The surrounding Antarctic Ocean plays a crucial role in Earth's oceanic circulation by connecting three main oceans— the Atlantic, Indian, and Pacific Oceans—and contains up to 6% of the world's oceanic volume (Eakins and Sharman, 2010). In addition, the high levels of atmospheric transport and the connectivity of ocean currents make the Antarctic Ocean susceptible to inorganic pollutants (Potapowicz et al., 2019). Elevated concentrations of trace elements (in particular Cd and Hg) have been reported in Antarctic crustaceans (Keil et al., 2008; Petri and Zauke, 1993), fish (Bargagli et al., 2000; Bustamante et al., 2003; Goutte et al., 2015), seabirds (Blévin et al., 2013; Carravieri et al., 2013, 2014), and marine mammals (Andrade et al., 2007; Honda et al., 1987). For Subantarctic and Antarctic cephalopods, some studies demonstrated moderate Hg levels and very high Cd concentrations (Bustamante et al., 1998b; Matias et al., 2019; Trevizani et al., 2018; Seco et al., 2020). Global cephalopod studies have demonstrated their remarkable bioaccumulation potential for trace elements, such as Ag, Cd, and Hg (Bustamante et al., 2006; Martin and Flegal 1975; Miramand and Bentley 1992). Although the trace element concentrations in benthic octopods from temperate waters have been previously studied (e.g., Chouvelon et al., 2011; Raimundo et al., 2005; Rjeibi et al., 2014), comparative data from the Southern Ocean is lacking for most benthic octopods.

Cephalopods are key components of the Antarctic ecosystem (Collins and Rodhouse, 2006; Xavier et al., 2018). Benthic octopods have a role as both predator and prey, and are crucial in the transfer of energy and trace elements in the Antarctic food web, linking low trophic level consumers to high-level predators (Allcock,1997; 2005; Allcock et al., 2001; Daly, 1996; Piatkowski et al., 1998; 2003; Strugnell et al., 2017). A wide variety of predators feed on benthic octopods, for example: elephant seals (*Mirounga leonine*, Burdman et al., 2015; Daneri et al., 2000; Rodhouse et al., 1992), Weddell seals (*Leptonychotes weddellii*, Acevedo et al., 2015; Casaux et al., 1997; Lipinski and Woyciechowski, 1981; Negri et al., 2016), Patagonian toothfish (*Dissostichus eleginoides*, Xavier et al., 2002), and the black-browed albatross (*Thalassarche melanophris*, Xavier and Croxall, 2007). In turn, benthic octopods prey on crustaceans, polychaetes, bivalves, gastropods (Daly, 1996), and amphipods (Daneri et al., 2000; Piatkowski et al., 2003). Clear differences in the feeding ecology among octopod species were proposed based on the interspecific variation observed in beak morphology (Matias et al. 2019, Schwarz et al., 2019) and stomach content analyses (Büring, 2019; Daly, 1996; Piatkowski et al., 2003). These differences suggest that Antarctic benthic octopods occupy different trophic niches, despite their geographic co-occurrence.

The purpose of the present study is to investigate the role that the eight benthic octopod species, collected from near Elephant Island, might play in the transfer of trace elements in this pristine Antarctic ecosystem. We tested two hypotheses: 1) we expect to see differences between individuals of different sizes, due to ontogenetic shifts in feeding, as well as between the mantle and digestive gland tissues, the two main storage organs for trace elements in cephalopods. Furthermore, the relative trophic position of each octopod species (assessed through the δ^{15}N in mantle as a proxy) and their feeding habitat (δ^{13}C in mantle) should differ at the intraspecific and interspecific levels; and 2) we expect to see niche partitioning reflected by interspecific differences in the stable isotope values. The trophic ecology is supposed to mainly drive the trace element contamination differences recorded in individuals' tissues.

Material and Methods

Sample collection

Specimens were collected during a research cruise of the German research vessel '*Polarstern*' (ANT-XXVIII/4) in March/April 2012 near Elephant Island, South Shetland Islands (Lucassen, 2012). The sampling area ranged from 61°04' to 61°37′ S and 54°88 to 56°17′ W. Samples were caught in depths of 109–323 m by bottom otter trawling (OTB, Fig. 1; Table 1). A total of 60 individuals from eight different species (Table 1) were subsampled from approximately 800 collected specimens in total (Lucassen, 2012). For each specimen, the sampling depth (m), the mantle and total length (mm), and the sex (male, female, or sex unknown [indet./juvenile]) was noted. Specimens were stored at -40°C until they were analyzed for trace elements, stable isotopes, and genetics.

Species identification

Specimens were initially morphologically identified to species level on board of the research vessel, and identification were verified genetically for specimens that could not be confidently identified to species. When an identification was uncertain, a tissue snip was taken from the arm and stored frozen for later DNA analysis. DNA was extracted using EconoSpin columns (Epoch Life Science) with QIAGEN reagents, following protocols for the DNeasy Blood & Tissue Kit (QIAGEN). The mitochondrial gene cytochrome *c* oxidase subunit I (COI), was amplified using primers and protocols following Braid et al. (2014). The sequence reaction was performed using the primer HCO2198, which was the reverse primer used for the PCR

(Macrogen, Korea). Sequences were edited in CodonCode Aligner (CodonCode Corp., Dedham, MA, USA) and uploaded to the Barcode of Life Data System (Ratnasingham and Hebert, 2007) and subsequently submitted to GenBank. All sequences were screened for potential contamination using GenBank's Basic Local Alignment Search Tool (BLAST). Genetic identifications were made using the Full Database identification engine on BOLD.

Stable isotope analysis

Carbon and nitrogen stable isotopes were measured from freeze-dried tissue samples (0.2–0.4 mg) with a continuous flow mass spectrometer (Delta V Plus with a Conflo IV interface, Thermo Scientific, Bremen, Germany) coupled to an elemental analyser (Flash 2000, Thermo Scientific, Milan, Italy) following Lischka et al. (2018). Stable isotope values were calculated using the following formula: $\delta^{13}C$ or $\delta^{15}N = [(R_{sample}/R_{standard}) - 1] \times 10^3$, where R is $^{13}C/^{12}C$ or $^{15}N/^{14}N$; all results are expressed in the ‰ unit notation as a deviation of the standard (Vienna Pee Dee Belemnite for $\delta^{13}C$ and N_2 in air for $\delta^{15}N$). Internal laboratory standards (acetanilide and peptone) were used to assess the analytical precision, which was <0.10 ‰ for $\delta^{13}C$ and <0.15 ‰ for $\delta^{15}N$.

Trace element analysis

Digestive gland and mantle tissue samples were freeze-dried for 48–72 hours and ground into a homogenous powder. The water content in the octopod species (measured in *P. charcoti*, as an example) was 68±8% in the digestive gland, and 79±3% in the mantle tissue. Sample aliquots were prepared with ~200 mg dry weight (dw) of the tissue samples and digested overnight in a 3:1 mixture of 65% HNO_3 (Merck, suprapur quality) and 37% HCl (Merck, suprapur quality). The acidic digestion was followed by a mineralization, where samples were heated for 30 min in a Milestone microwave (maximum temperature of 105°C). Trace element concentrations (Ag, As, Cd, Co, Cr, Cu, Fe, Mn, Ni, Pb, Se, V, and Zn) were measured by inductively coupled plasma mass spectroscopy (ICP-MS)—with a Thermo Fisher Scientific X Series 2—and by optical emission spectroscopy (ICP-OES)—with a Varian Vista-Pro ICP—following Lucia et al. (2016). Dogfish liver (DOLT-4, National Research Council, Canada), lobster hepatopancreas (TORT-3, NRCC), and clam muscle tissue (IAEA-461, International Atomic Energy Agency, Austria) were used as certified reference materials (CRMs). CRMs, together with procedural blanks, were treated and analyzed in the same way as the samples. Recoveries of the elements ranged from 85–105% (*n*=9). The detection limits for Ag, Cd, Co, Cr, and Pb were 0.025 µg g^{-1}, Ni was 0.05 µg g^{-1}, Cu, Mn, and Se were 0.125 µg g^{-1}, As was 0.25 µg g^{-1}, V was 0.5 µg g^{-1}, and Fe and Zn were 5 µg g^{-1}, and based on 200 mg of sample material diluted in a volume of 50 ml. The Cd concentrations was only measured in the octopod digestive gland tissue and not in the mantle tissue due to a potential diffusion effect (Francesconi et al., 1993; Lischka et al., 2020a).

Sample aliquots (1–2 mg) of the dried homogenized digestive gland and mantle tissue were used to analyse Hg using an Advanced Mercury Analyser (ALTEC AMA 254, with a detection limit > 0.05 ng) as described in Bustamante et al. (2006). For every 10 samples, one standard sample of certified reference material DOLT 5 (Dogfish liver; NRCC) was analyzed with a recovery of 109 ± 2%. Results for all trace element concentrations are expressed in µg g^{-1} dw.

Statistical analyses

All statistical analyses were conducted with the statistical software R (R Core Team, 2017; Ihaka and Gentleman, 1996). The species *Pareledone felix* (*n*=2) and *P. turqueti* (*n*=1) were excluded from all statistical analysis and the interspecific comparisons due to their small sample sizes.

Analysis of covariance (ANCOVA) was performed on log-standardised δ^{15}N and δ^{13}C values to check if these were influenced by size (total length), sampling depth, sex (female, sex unknown [indet./ juvenile], male), and species.

Interspecific differences in stable isotope values were assessed with Tukey's post-hoc tests ('glht' function, R package 'multcomp', Hothorn et al., 2016). Adjustment of the *p*-values was conducted following Benjamini and Hochberg (1995).

A principal component analysis (PCA) was applied as an exploratory tool to examine differences in overall trace element concentrations among species for the digestive gland tissue. Prior to the PCA, data was normalised and z-transformed (auto-scaled, mean centred, and divided by the standard deviation).

To test whether tissue type, sampling depth, species, size (as total length), "sex", δ^{15}N and the residuals of δ^{13}C had an influence on trace element concentrations, generalised linear models (GLMs) with a negative binomial distribution and logit link function were applied (GLM, package 'MASS', Ripley et al., 2013). The fit of the models was confirmed by analysing the residuals. One model per trace element was fitted against non-transformed concentrations and the variables were added sequentially. Since the variables δ^{13}C and δ^{15}N were correlated (Pearson's r =0.71), only the residuals of the correlation between δ^{13}C and δ^{15}N were included in the models. Because size is affected by species, these variables were included as an interaction term in the models.

ANCOVAs were performed to see whether "sex", size, and/or stable isotopes had an influence on trace element concentrations for *P. charcoti,* because this species had a balanced sample size (11 females, 11 males). This model was chosen because the residuals of the *P. charcoti* dataset were normally distributed.

Results

Species identifications

Sixty specimens were morphologically identified to the species level (*Adelieledone polymorpha, Pareledone aequipapillae, P. albimaculata, P. aurata, P. charcoti, P. cornuta, Pareledone felix,* and *P. turqueti;* Table 1). Among them, the identification of 14 specimens were confirmed with DNA barcoding, confirming a total of eight species (99.83–100% match in the BOLD database). The morphological species identification (size variations, Fig. 2) suggests that *P. aurata* and *P. albimaculata* might include more species than confirmed with DNA barcoding.

Stable isotopes

The δ^{13}C and δ^{15}N values varied significantly among the species (ANCOVA, $p<0.001$; Table 2), indicating that the foraging habitats and diets differ among subantarctic octopods. On average, the highest δ^{13}C values were measured in *A. polymorpha*, *P. aequipapillae*, *P. felix* and *P. turqueti,* whereas *P. albimaculata, P. charcoti* and *P. cornuta* had similar (or overlapping) values, and *P. aurata* showed lowest δ^{13}C values (Fig. 3; Table S1). Statistically, only the δ^{13}C values of *A. polymorpha* and *P. aurata* differed significantly in the Tukey's post-hoc tests ($p<0.001$). In addition, sex had a significant effect on δ^{13}C (Table 2), with males and juveniles exhibiting the highest values ($p < 0.05$, Table 2). Regarding the δ^{15}N values, *A. polymorpha* exhibited the highest average value (Table S1), followed by *P. aequipapillae, P. albimaculata, P. felix,* and *P. turqueti* (Fig. 3). Lower average δ^{15}N values were measured in *P. cornuta,* but the lowest δ^{15}N values were recorded in 29 individuals of *P. aurata* and *P. charcoti* (which differed significantly from all other species; Tukey's post hoc test, $p<0.001$). Both δ^{13}C and δ^{15}N values are positively correlated (Pearson's $r=0.71$; $p < 0.05$) indicating that the highest δ^{13}C values were measured in specimens with the highest δ^{15}N values (Table 2). In addition, sampling depth had a significant effect on δ^{13}C and δ^{15}N values (Table 2), with the lowest δ^{13}C values and the highest δ^{15}N values measured in specimens sampled in deeper waters.

Trace elements

Average trace elements in the digestive gland were measured in decreasing order as follows: Fe> Cu> Zn> Cd> V> As> Ag> Se> Co> Mn> Ni> Pb> Cr> Hg (Table S1). In the mantle tissue, the average trace element concentrations decreased as follows: Zn> Cu> As> Fe> Se> V> Mn> Ni> Ag> Co> Hg> Cr> Pb (Table S1).

The PCA of the digestive gland tissue showed that Principal Component (PC) 1 explained 38.6% of the variance and was mainly driven by Ag, Cd, Co, Cu, Fe, Hg, and Pb, whereas PC2 explained 18.1% of the variance and was mainly driven by As, Cr, Mn, Ni, Se, and V (Fig. 4). The specimens of *P. charcoti* formed a distinct cluster along the PC1 influenced by highest Ag, Cd, Co, Cu, Pb, and Zn concentrations recorded in this species (see Table S1). The samples of *A. polymorpha*, *A. aequipapillae* and *A. albimaculata* formed a distinct group, characterised by higher As, Cr, and Ni concentrations. Overall, *P. albimaculata* displayed the highest concentrations of As and V (also supported by the GLMs, Table 3), *P. aequipapillae* the highest ones of Cr, Fe, Hg, and Mn (not confirmed by the GLMs), and *A. polymorpha* the highest Ni and Se concentrations (Table S1; supported by the GLMs, Table 3).

The GLMs analyses confirmed that the concentrations of As, Cd, Co, Fe, Ni, Pb, Se, V, and Zn significantly differed among species (Table 3) and that all concentrations, with the exception of As and Hg, are higher in the digestive gland compared to the mantle tissue. Arsenic tended to be more concentrated in the muscle than in the digestive gland in all species, whereas the Hg concentrations remained similar between the two tissues (Table S1). Sex had an impact on

Cd, Co, Cu, Fe, and Pb concentrations in both tissues, with juveniles (sex indet.) and males generally exhibiting higher concentrations relative to females (Table 3, Fig. S1). Finally, the concentrations of Cu (Fig. S2) and Zn decreased significantly with increasing size, while Se significantly increased with size (Table 3).

The stable isotope values of δ^{15}N had a significant positive effect on Ag, As, and Se concentrations and a significant negative effect on Cd concentrations (Table 3). None of the analyzed variables had an effect on Hg concentrations. The residuals of δ^{13}C did not show a significant effect on any of the analyzed trace elements.

Sampling depth had a significant effect on Co, Fe, Ni, Pb, Se, and V concentrations (Table 3), which were highest in specimens sampled in shallower waters. In contrast, As concentrations reached the highest concentrations in individuals collected in deeper waters (Table 3).

Pareledone charcoti

Focusing on the sample set of *P. charcoti* (*n*=11 males and 11 females), the concentrations of Ag, Co, Cr, Cu, Fe, Ni, Pb, Se, V, and Zn differed significantly between digestive gland and mantle tissue, with highest concentrations measured in the digestive gland (Table S2). Significantly higher concentrations of Cd, Co, Cr, Cu, Fe, Ni, Pb, Se, and Zn were also measured in males (Table S2; Fig. S2) compared to females. The concentrations of Hg and Zn increased with increasing δ^{15}N values ($p < 0.05$; Table S2).

Discussion

Although cephalopods, particularly benthic octopods, are a key component of the Antarctic food web, little is known about their trophic ecology in the Southern Ocean (Seco et al., 2020). In this study, we report the stable isotope values and the concentrations of 14 trace elements in eight different octopod species sampled near Elephant Island. For the first time, stable isotopes and trace elements are linked together to describe niche segregation by feeding and habitat tracers, in these endemic octopod species. Our study demonstrated that shallower living octopods are distinguished from deeper living species by their habitat and diet, and that reflected on their isotopic signature and trace elemental burden. This clear segregation may have ecological repercussions for predators that feed on different species or that hunt at different depths.

Trophic ecology

The δ^{15}N signature is a reliable indicator of the trophic position in specific food webs (Cherel et al., 2009; Richert et al., 2015). A relative comparison of this proxy among the species the Antarctic and Subantarctic octopus assemblages investigated here reveals that *Paraeledone aurata* and *P. charcoti* are distinguished from the other species by their low δ^{15}N values (Fig. 3), suggesting a lower trophic position. These results are consistent with those from a recent study on the feeding ecology of Antarctic octopods from the same area, which reported higher δ^{15}N values in *A. polymorpha* and *P. aequipapillae* relative to *P. charcoti* (Büring, 2019). These differences were linked to their stomach content analysis, which revealed that octopods living at greater depths (e.g. *A. polymorpha*) consumed prey at higher trophic levels, such as fish. In contrast, species living in shallower waters, such as *P. charcoti,* base their diet on crustaceans and amphipods (Piatkowski et al., 2003). In line with this observation, our specimens of *P. aurata* and *P. charcoti* were collected at depths ranging from 109 to 178 m, whereas individuals from the other analyzed species were sampled below 288 m depth. The significant effect of sampling depth found in the ANCOVAs (Table 2) may indicate a spatial segregation of the octopod species with depth, probably driving a contrasting feeding ecology between *A. polymorpha* and *P. aurata*, reflected by the significant difference in δ^{15}N values (Table S1).

The carbon stable isotope signature can be used to assess habitat differences between species, since δ^{13}C values exhibit a latitudinal gradient, with highest values measured inshore relative to offshore habitats, and higher values in benthic species relative to pelagic species (Cherel et al. 2009; Chouvelon et al., 2011). Most of the species analyzed herein (i.e., *P. aequipapillae, P. albimaculata, P. charcoti, P. charcoti, P. felix* and *P. turqueti*) displayed a narrow range in δ^{13}C values (from -25.42 to -24.09), suggesting that they share an overlapping feeding habitat. No significant effect of δ^{13}C values independently of the δ^{15}N values was measured in this study, likely due to the small δ^{13}C values variation, which never exceeded 2 ‰ (Table S1).

Arsenic

Most trace element concentrations in the digestive gland were generally higher than the mantle tissue, which is consistent with the detoxification and storage role of the digestive gland usually recognized in cephalopods, including octopods (e.g., Miramand and Bentley, 1992; Bustamante et al., 1998b; Miramand et al., 2006; Seixas et al., 2005). However, As concentrations were higher in the mantle tissue than in the digestive gland (Table S1). This could reflect the affinity of arsenobetaine, the major As species in cephalopods (Taylor et al., 2017) to the proteins in the muscular tissue (Shen et al., 2013). This has been already described in other cephalopods, such as nautilus, *Nautilus macromphalus* (Bustamante et al., 2000), and arrow squid, *Nototodarus sloanii* (Lischka et al., 2020a).

Some trace elements are not well documented in cephalopods (e.g., Ag, As, and V), which makes the interpretation for those elements challenging. Nevertheless, As concentration data is available for certain cephalopod species (e.g., *Todarodes filippovae*, Kojadinovic et al., 2011). The As concentrations measured in all octopods in this study were higher (digestive gland As concentration average = 37.48 µg g^{-1} dw) relative to pelagic squid species from previous studies (e.g., *Gonatus fabricii*, 10.18 µg g^{-1} dw, Lischka et al., 2020b; *Sthenoteuthis pteropus*, 18.33 µg g^{-1} dw, Lischka et al., 2018), but comparable to octopod species from temperate environments (e.g., *Octopus vulgaris*; ~40 µg g^{-1} dw, Raimundo et al., 2010). This is congruent with the well-supported hypothesis that the benthic *vs* pelagic habitat is the main driver of As accumulation since As tends to be trapped in sediments (Sanders, 1980). In the present study, the deeper living *P. albimaculata* and *P. polymorpha* had the highest concentrations of As among the eight studied octopod species (Table S1; Fig. 4). Cephalopods mainly accumulate As through their diet (Kojadinovic et al. 2011), and since information about the feeding ecology of this species is currently not available, conclusions between diet and As exposure cannot be drawn. However, our results show that sampling depth and $\delta^{15}N$ values both positively influenced As concentrations (Table 3) likely reflecting that feeding at higher trophic levels results in higher As contamination. This may suggest a higher As bioavailability in octopod prey with depth and/or the biomagnification of the organic As compounds between prey and octopods (Kubota et al., 2001; Neff, 1997; Tu et al., 2011). Further dietary studies are needed to confirm this hypothesis.

Cadmium

The variations in Cd concentrations have been described for cephalopod species from different oceanic origins (Table 4). It is noteworthy that the Antarctic octopods in the present study display similar Cd concentrations with temperate species, such as *Octopus vulgaris* from the Mediterranean Sea (Miramand and Guary, 1980) and *Eledone cirrhosa* from the English Channel (Miramand and Bentley, 1992). However, the latter species displayed a higher variation in Cd concentrations with respect to its location when compared to *E. cirrhosa* from the Bay of Biscay (Chouvelon et al., 2011) and from the Faroe Islands (Bustamante et al., 1998a), which displayed at least 50% lower and 1800% higher Cd concentrations, respectively, compared to the *A. polymorpha* samples (Table 4). In contrast, it could be hypothesized that the reason that benthic octopods show lower levels of Cd within the pan-Antarctic region is due to the more homogenous and stable environment that characterizes this area.

The Cd concentrations measured in this study were relatively high and similar to concentrations from urbanized coasts (Table 4). This could be because of an enrichment of Cd in the surface layers due to upwelling events (Bustamante et al., 1998b). It could also be due to the limited bioavailability of essential elements such as Cu and Zn, leading to an enhanced uptake of Cd (Gault-Ringold et al., 2012; Petri and Zauke, 1993). Therefore, organisms may have developed very efficient mechanisms of elemental uptake for Cu and Zn (Petri and Zauke, 1993). Because these mechanisms are probably not specific to Cu and Zn, Cd might be absorbed by the same physiological pathways (Penicaud et al., 2017). These high Cd concentrations might also be attributable to the 'Cd anomaly', which describes a latitudinal gradient of Cd concentration in marine invertebrates (including octopods), where higher Cd concentrations have been measured in marine organisms from subpolar areas. These descriptions include amphipods (Bargagli et al., 1996; Kahle and Zauke, 2002; Petri and Zauke, 1993), fish (Bustamante et al. 2003; Macdonald and Sprague 1988; Zauke et al. 1999), and cephalopods (Bustamante et al. 1998a, Cipro et al. 2017).

There is a general paucity of data concerning trace elements in Antarctic octopods, but the high Cd concentrations in the digestive gland in *Graneledone gonzalezi* and *Benthoctopus thielei* from the subantarctic Kerguelen Islands (Bustamante et al., 1998b) are within the same range of the present values, suggesting that the Cd anomaly may occur in the South Shetland Islands area.

This study further reported significant differences in Cd concentration data among different species (GLMs; Table 3), with *P. charcoti* and *P. aurata* displaying the highest Cd concentrations (see also Fig. 4). These results suggest that Cd concentration differences, consistent with the δ^{15}N signature (Table 3), can be used to highlight the diet partitioning of octopod species. For example, *P. charcoti* has shown high Cd levels and their diet includes Cd richer prey (amphipods, crustaceans; Büring, 2019), while *A. polymorpha* showed lower Cd levels and is known to consume prey of higher trophic levels (e.g., fish; Büring, 2019) with lower Cd concentrations (Storelli and Marcotrigiano, 2004). Our results suggest that Cd concentration data might be used to discriminate and characterise the ecological niche of octopod species.

Apart from interspecific differences, significant differences in Cd concentrations were observed between the sexes in the GLMs, with the highest concentrations measured in male *P. charcoti* specimens relative to females of the same species (Table 3, Fig. S1). *Paraledone charcoti* was the only species in the present study that had a balanced sample set (11 females and 11 males), which was the reason other species could not be evaluated this way. Since cephalopods mainly take up Cd though their diet (Bustamante et al., 2002; Koyama et al. 2000), this difference could be due to diet composition, highlighting a niche partitioning between sexes (Table 3). Such a difference in diet between sexes was previously reported for *O. mimus* from Northern Chilean waters, with females having a higher food intake relative to males (Cortez et al., 1995). Therefore, the amount of ingested prey but also their type should also influence Cd concentrations. In this respect, *P. charcoti* females might ingest more Cd-poor prey compared to males. To confirm sex differences in the diet of Antarctic benthic octopods, additional stomach content analysis, both morphologically and genetically, is necessary.

Mercury

The concentration of Hg measured in the mantle muscles of *P. turqueti* from the South Georgia coast was similar to the Hg concentrations measured in the present study (Matias el al., 2019; 2020). Worldwide, Hg concentrations fluctuated among octopod species and sampling locations, with the highest concentrations measured in *E. cirrhosa* from the Tyrrhenian Sea (Rossi et al. 1993; Barghigiani et al 2000). The Hg concentrations in the present study were similar to those reported for *O. vulgaris* from the Portuguese Coast (Seixas et al., 2005), and *E. cirrhosa,* from the Bay of Biscay (Chouvelon et al., 2011; Table 4). Benthic species in the Octopodidae usually display higher Hg concentrations compared to other cephalopod families (Penicaud et al., 2017). The specimens analyzed in the present study showed lower Hg concentrations relative to pelagic Antarctic squids; for example, Hg concentrations measured in the digestive gland of the squid *Kondakovia longimana* (0.045± 0.021 µg g^{-1} dw; Seco et al., 2020) were ten-fold lower when compared to *A. polymorpha* (Table 4). This highlights the interspecific as well as inter-location variability of Hg concentrations. Surprisingly, none of the proxies of trophic and habitat ecology have an effect on Hg concentrations among species, whereas Hg levels usually increases with benthic habitat and trophic position (Chouvelon et al., 2012). This result is likely related to the close ecological niches shared by Antarctic octopods and indicate that Hg would be a pertinent proxy for the trophic position and the habitat (benthic/pelagic) at the cephalopod assemblage scale (i.e., including benthic octopods and pelagic squids) in future comparative ecological studies (e.g., Seco et al., 2020).

Lead

Lead is one of the major pollutants in oceanic systems (Boyle et al., 2014). In the present study, Pb concentrations in the digestive gland varied among species, with *P. charcoti* exhibiting significantly higher concentrations compared to the other species analyzed (Fig. S1). This finding is consistent with a previous study on Antarctic benthic invertebrate communities, which also showed significant variations of Pb concentrations among species (Majer et al., 2014). This variation was attributed to the anthropogenic pollution in the Admiralty Bay at King George Island. However, because Elephant Island is a near-pristine ecosystem, it is unlikely that Pb has reached high levels due to anthropogenic causes. Instead, the shallow-living *P. charcoti* might be exposed to higher Pb concentrations due to diet, since it mainly feeds on invertebrates (Büring, 2019), which are known to bioaccumulate this element (Rainbow, 1997). Feeding at higher trophic levels should reduce the exposure to this element because it is bioreduced along the food webs (Michaels and Flegal, 1990). Furthermore, higher Pb concentrations are associated with near-surface water layers (Henderson and Maier-Reimer, 2002). The reasons for oceanic depth differences are not yet fully understood, and anthropogenic sources—e.g., aeolian dust from industrial applications—were described as a major contributor (Rosman et al., 1994; Sun and Xie, 2001). Although Pb concentrations in deeper waters of the Antarctic region of the Indian Ocean have increased over the last century, they are still lower when compared with oceanic regions that have higher regional anthropogenic emissions and slower vertical mixing rates (Echegoyen et al., 2014). Apart from the interspecific concentration differences found in the present study, Pb was generally higher in males and juveniles relative to females (Table 3, Fig. S1). A similar pattern has been observed in the squid *Gonatus fabricii* (from the Arctic ocean; Lischka et al., 2020b) and in *O. vulgaris* (from the Mediterranean Sea; Rjeibi et al., 2014), where Pb concentrations in the digestive gland were lowest in females and associated with feeding habits. An analysis of sex-specific dietary patterns is needed in order to make further conclusions about the effects of sex on Pb bioaccumulation.

Potential impacts on ecosystem

The shallower-living species *P. charcoti* had significantly higher concentrations of Ag, Cd, Co, Cu, Pb, and Zn, compared to all other species analyzed in the present study (Fig. 4). These elevated concentrations could be related to the diet of *P. charcoti*, which is known to consist mainly of crustaceans (particularly amphipods; Piatkowski et al., 2003; Büring, 2019). In contrast, the larger *A. polymorpha* has a diet that contains fish, which generally exhibit lower trace element concentrations (particularly, the non-essential elements Ag, Cd, and Pb) than crustaceans (Chouvelon et al., 2011; Pierce et al., 2008). Since diet can be considered a major source for trace element exposure (Bustamante et al., 1998a), the measured differences in trace element concentrations among species in this study are likely due to differences in feeding habits and rates. One of the main findings of this study is the significant difference in δ^{15}N values found in *P. charcoti* and *P. aurata* (relative to all other analyzed species, which is supported by ecological tracers (trace elements, isotopic signature). In addition, based on the ranges of the δ^{15}N values and the significant effect of depth on δ^{15}N values (Table 2), the analyzed Antarctic octopod community seems to be a widely unique group with two segregated species, *P. charcoti* and *P. aurata*. This might likely reflect distinct ecological niches, driven by depth and leading to different diets. However, considering the narrow range of δ^{13}C variation among all species, the feeding patterns of the octopods investigated in our study do not seem differently enough to emphasize contrasted foraging habitats.

The species assessed in this study are consumed by a variety of Antarctic top predators (see Introduction; e.g., Casaux et al., 1997; Daneri et al., 2000; Lipinski and Woyciechowski, 1981; Rodhouse et al., 1992; Xavier et al., 2002), which are exposed to trace elements, sometimes in elevated concentrations, when they feed on the species analyzed in the present study. In particular, the high Cd concentrations measured in the digestive gland samples might represent a significant point source for predators (Bustamante et al. 2008). High Cd concentrations were measured in Antarctic seals and explained by dietary exposure, which includes benthic octopods, particularly the shallower *P. charcoti*, which exhibits the highest Cd concentrations found in the present study (Casaux et al., 1997; Malcolm et al., 1994; Szefer et al., 1994). Our study highlights that incirrate octopods represent a vector for trace elements, including Cd and Hg, to top predators foraging near Antarctic Elephant Island. This has been previously shown for other oceanic areas (Bustamante et al., 1998a; Penicaud et al., 2017). Pinnipeds such as the Antarctic fur seal *Arctocephalus gazella* and the Southern elephant seal *Mirounga leonina* might be exposed to elevated trace element concentrations due to their octopod-rich diet (Burdman et al., 2015; Casaux et al., 1998). Consistently, elevated Cd concentrations have been measured in liver and kidney tissues of *A. gazella*, which were associated with hepatotoxicity and nephrotoxicity (De Moreno et al., 1997; Malcolm et al., 1994). Overall, the extent of the toxic effects of trace elements on both predators and prey still remains understudied. In this context, it is necessary to provide information to allow risk assessment evaluations.

Conclusion

In this study, stable isotopes (δ^{13}C and δ^{15}N) and trace elements (Ag, As, Cd, Co, Cr, Cu, Fe, Hg, Mn, Ni, Pb, Se, V, and Zn) were assessed in an octopod assemblage collected near Elephant Island in the Antarctic Ocean. Significant differences were measured between sexes and species. For example, *P. aurata* and *P. charcoti* exhibited significantly lower δ^{15}N values compared to *A. polymorpha*, which indicates that they feed at a lower trophic level. This is consistent with the sampling depths of *P. aurata* and *P. charcoti*, which occurred in shallower waters than *A. polymorpha*. This suggests. a spatial segregation of the octopod species with depth, which could drive a contrasting feeding ecology. Furthermore, most trace element concentrations (with the exception of Hg) varied between species, suggesting different bioaccumulation patterns, which likely reflect trophic habitat discrimination. Our results are consistent with the few previous dietary studies on these species and highlight the influence of benthic Antarctic octopods in the transfer of trace elements to their main predators, which is the first step in unravelling the complex interactions in this unique and irreplaceable environment.

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References

Acevedo, J., Carreno, E., Torres, D., Aguayo-Lobo, A., Letelier, S., 2015. Cephalopod remains in scats of Weddell seals (*Leptonychotes weddellii*) at Cape Shirreff, South Shetland Islands, Antarctica. Polar Biology, 38(9), 1559–1564.

Allcock, A.L., 1997. The genetics and taxonomy of Southern Ocean Octopodidae, with special reference to the genus *Pareledone* (PhD Thesis). University of Liverpool.

Allcock, A.L., 2005. On the confusion surrounding *Pareledone charcoti* (Joubin, 1905) (Cephalopoda: Octopodidae): endemic radiation in the Southern Ocean. Zoological Journal of the Linnean Society 143, 75–108.

Allcock, A.L., Piatkowski, U., Rodhouse, P.G.K., Thorpe, J.P., 2001. A study on octopodids from the eastern Weddell Sea, Antarctica. Polar Biology 24, 832-838.

Andrade, S., Carlini, A.R., Vodopivez, C., Poljak, S., 2007. Heavy metals in molted fur of the southern elephant seal *Mirounga leonina*. Marine Pollution Bulletin 54, 602–605.

Bargagli, R., Nelli, L., Ancora, S., Focardi, S., 1996. Elevated cadmium accumulation in marine organisms from Terra Nova Bay (Antarctica). Polar Biology 16, 513–520.

Bargagli, R., Sanchez-Hernandez, J.C., Monaci, F., Focardi, S., 2000. Environmental factors promoting bioaccumulation of Hg and Cd in Antarctic marine and terrestrial organisms. Antarctic ecosystems: models for wider ecological understanding (ed. W. Davison, C. Howard-Williams, P. Broady) 308–314.

Barghigiani, C., Ristori, T., Biagi, F., De Ranieri, S., 2000. Size related mercury accumulations in edible marine species from an area of the Northern Tyrrhenian Sea. Water, Air, and Soil Pollution, 124(1-2), 169–176.

Benjamini, Y., Hochberg, Y., 1995. Controlling the false discovery rate: a practical and powerful approach to multiple testing. Journal of the Royal Statistical Society Series B, 57, 289–300.

Bentley, C.R., Giovinetto, M.B., 1992. Mass balance of Antarctica and sea level change. Wisconsin University, Madison Department of Geology, Geophysical and Polar Research Center, 8.

Blévin, P., Carravieri, A., Jaeger, A., Chastel, O., Bustamante, P., Cherel, Y., 2013. Wide range of mercury contamination in chicks of Southern Ocean seabirds. PLoS One 8, e54508.

Boyle, E.A., Lee, J.M., Echegoyen, Y., Noble, A., Moos, S., Carrasco, G., Zhao, N., Kayser, R., Zhang, J., Gamo, T., Obata, H., 2014. Anthropogenic lead emissions in the ocean: The evolving global experiment. Oceanography, 27(1), 69–75.

Braid, H.E., McBride, P.D., Bolstad, K.S., 2014. Molecular phylogenetic analysis of the squid family Mastigoteuthidae (Mollusca, Cephalopoda) based on three mitochondrial genes. Hydrobiologia, 725, 145–164.

Burdman, L., Daneri, G.A., Negrete, J., Mennucci, J.A., Marquez, M.E., 2015. Cephalopoda as prey of juvenile southern elephant seals at Isla 25 de Mayo/King George, South Shetland Islands. Iheringia. Série Zoologia, 105(1), 12–19.

Büring, T., 2019. Feeding ecology of Antarctic octopods (MSc thesis). University of Kiel.

Bustamante, P., Bocher, P., Cherel, Y., Miramand, P., Caurant, F., 2003. Distribution of trace elements in the tissues of benthic and pelagic fish from the Kerguelen Islands. Science of the Total Environment 313, 25–39.

Bustamante, P., Caurant, F., Fowler, S.W., Miramand, P., 1998a. Cephalopods as a vector for the transfer of cadmium to top marine predators in the north-east Atlantic Ocean. Science of the Total Environment 220, 71–80.

Bustamante, P., Cherel, Y., Caurant, F., Miramand, P., 1998b. Cadmium, copper and zinc in octopuses from Kerguelen Islands, Southern Indian Ocean. Polar Biology 19, 264–271.

Bustamante, P., Grigioni, S., Boucher-Rodoni, R., Caurant, F., Miramand, P., 2000. Bioaccumulation of 12 trace elements in the tissues of the nautilus *Nautilus macromphalus* from New Caledonia. Marine Pollution Bulletin, 40(8), 688-–96.

Bustamante, P., Lahaye, V., Durnez, C., Churlaud, C., Caurant, F., 2006. Total and organic Hg concentrations in cephalopods from the North Eastern Atlantic waters: influence of geographical origin and feeding ecology. Science of the Total environment 368, 585–596.

Bustamante P., González A.F., Rocha F., Miramand P., Guerra A. (2008) Metal and metalloid concentrations in the giant squid *Architeuthis dux* from Iberian waters. Marine Environmental Research, 66(2), 278–287.

Bustamante, P., Teyssié, J.-L., Fowler, S.W., Cotret, O., Danis, B., Miramand, P., Warnau, M., 2002. Biokinetics of zinc and cadmium accumulation and depuration at different stages in the life cycle of the cuttlefish *Sepia officinalis*. Marine Ecology Progress Series 231, 167–177.

Carravieri, A., Bustamante, P., Tartu, S., Meillère, A., Labadie, P., Budzinski, H., Peluhet, L., Barbraud, C., Weimerskirch, H., Chastel, O., Cherel, Y., 2014. Wandering albatrosses document latitudinal variations in the transfer of persistent organic pollutants and mercury to Southern Ocean predators. Environmental Science & Technology 48 (24): 14746–14755. dx.doi.org/10.1021/es504601m.

Casaux, R., Baroni, A., Carlini, A., 1997. The diet of the Weddell seal *Leptonychotes weddelli* at Harmony Point, South Shetland Islands. Polar Biology 18, 371–375.

Casaux, R., Baroni, A., Carlini, A., 1998. The diet of the Antarctic fur seal *Arctocephalus gazella* at Harmony Point, Nelson Island, South Shetland Islands. Polar Biology, 20(6), 424–428.

Cherel, Y., Ridoux, V., Spitz, J. and Richard, P., 2009. Stable isotopes document the trophic structure of a deep-sea cephalopod assemblage including giant octopod and giant squid. Biology Letters, 5(3), 364–367.

Chouvelon, T., Spitz, J., Cherel, Y., Caurant, F., Sirmel, R., Mèndez Fernandez, P., Bustamante, P., 2011. Species and ontogenic-related differences in δ^{13}C and δ^{15}N values and Hg and Cd concentrations of cephalopods. Marine Ecology Progress Series 433, 107–120.

Chouvelon, T., Spitz, J., Caurant, F., Mèndez-Fernandez, P., Autier, J., Lassus-Débat, A., Chappuis, A., Bustamante, P., 2012. Enhanced bioaccumulation of mercury in deep-sea fauna from the Bay of Biscay (north-east Atlantic) in relation to trophic positions identified by analysis of carbon and nitrogen stable isotopes. Deep Sea Research I 65, 113–124.

Cipro, C.V., Cherel, Y., Bocher, P., Caurant, F., Miramand, P., Bustamante, P., 2018. Trace elements in invertebrates and fish from Kerguelen waters, southern Indian Ocean. Polar Biology, 41(1), 175–191.

Collins, M.A., Rodhouse, P.G., 2006. Southern Ocean cephalopods. Advances in Marine Biology, 50, 191–265.

Cortez, T., Castro, B.G., Guerra, A., 1995. Feeding dynamics of *Octopus mimus* (Mollusca: Cephalopoda) in northern Chile waters. Marine Biology 123, 497–503.

Daly, H.I., 1996. Ecology of the Antarctic octopus *Pareledone* from the Scotia Sea (PhD Thesis). University of Aberdeen.

Daneri, G.A., Carlini, A.R., Rodhouse, P.G.K., 2000. Cephalopod diet of the southern elephant seal, *Mirounga leonina*, at King George Island, South Shetland Islands. Antarctic Science 12, 16–19.

De Moreno, J.E.A., Gerpe, M.S., Moreno, V.J., Vodopivez, C., 1997. Heavy metals in Antarctic organisms. Polar Biology, 17(2), 131–140.

Eakins, B.W., Sharman, G.F., 2010. Volumes of the World’s Oceans from ETOPO1. NOAA National Geophysical Data Center, Boulder, CO 7.

Echegoyen, Y., Boyle, E.A., Lee, J.M., Gamo, T., Obata, H., Norisuye, K., 2014. Recent distribution of lead in the Indian Ocean reflects the impact of regional emissions. Proceedings of the National Academy of Sciences, 111(43), 15328–15331.

Francesconi K.A., Moore, E.J., Joll, L.M., 1993. Cadmium in the saucer scallop, *Amusium balloti*, from western Australian waters: concentrations in adductor muscle and redistribution following frozen storage. Marine and Freshwater Research, 44(6), 787–797.

Gault-Ringold, M., Adu, T., Stirling, C.H., Frew, R.D., Hunter, K.A. (2012). Anomalous biogeochemical behavior of cadmium in subantarctic surface waters: mechanistic constraints from cadmium isotopes. Earth and Planetary Science Letters 341, 94–103.

Gille, S.T., 2002. Warming of the Southern Ocean since the 1950s. Science 295, 1275–1277.

Goutte, A., Cherel, Y., Churlaud, C., Ponthus, J.-P., Massé, G., Bustamante, P., 2015. Trace elements in Antarctic fish species and the influence of foraging habitats and dietary habits on mercury levels. Science of the Total Environment 538, 743–749.

Henderson, G.M. and Maier-Reimer, E., 2002. Advection and removal of ^{210}Pb and stable Pb isotopes in the oceans: a general circulation model study. Geochimica et Cosmochimica Acta, 66(2), 257–272.

Honda, K., Yamamoto, Y., Kato, H., Tatsukawa, R., 1987. Heavy metal accumulations and their recent changes in southern minke whales *Balaenoptera acutorostrata*. Archives of Environmental Contamination and Toxicology 16, 209–216.

Hothorn, T., Bretz, F., Westfall, P., Heiberger, R.M., Schuetzenmeister, A., Scheibe, S., Hothorn, M.T., 2016. Package ‘multcomp.’ Simultaneous inference in general parametric models. Project for Statistical Computing, Vienna, Austria.

Ihaka, R., Gentleman, R., 1996. R: A language for data analysis and graphics. Journal of Computational and Graphical Statistics, 5, 299–314.

Kahle, J., Zauke, G.-P., 2002. Bioaccumulation of trace metals in the copepod *Calanoides acutus* from the Weddell Sea (Antarctica): comparison of two-compartment and hyperbolic toxicokinetic models. Aquatic Toxicology 59, 115–135.

Keil, S., De Broyer, C., Zauke, G.-P., 2008. Significance and interspecific variability of accumulated trace metal concentrations in Antarctic benthic crustaceans. International Review of Hydrobiology 93, 106–126.

Koyama, J., Nanamori, N., Segawa, S., 2000. Bioaccumulation of waterborne and dietary cadmium by oval squid, *Sepioteuthis lessoniana*, and its distribution among organs. Marine Pollution Bulletin, 40, 961–967.

Kojadinovic, J., Jackson, C.H., Cherel, Y., Jackson, G.D., Bustamante, P., 2011. Multi-elemental concentrations in the tissues of the oceanic squid *Todarodes filippovae* from Tasmania and the southern Indian Ocean. Ecotoxicology and Environmental Safety, 74, 1238–1249. https://doi.org/10.1016/j.ecoenv.2011.03.015

Kubota, R., Kunito, T., Tanabe, S., 2001. Arsenic accumulation in the liver tissue of marine mammals. Environmental Pollution 115, 303–312.

Le Quéré, C., Rödenbeck, C., Buitenhuis, E.T., Conway, T.J., Langenfelds, R., Gomez, A., Labuschagne, C., Ramonet, M., Nakazawa, T., Metzl, N., 2007. Saturation of the Southern Ocean CO_2 sink due to recent climate change. Science 316, 1735–1738.

Lipinski, M., Woyciechowski, M., 1981. Cephalopods in the food of Weddell seals from the Admiralty Bay (King George Island, South Shetland Islands). Polish Polar Research, 2(3-4), 163-167.

Lischka, A., Lacoue-Labarthe, T., Hoving, H.J.T., JavidPour, J., Pannell, J.L., Merten, V., Churlaud, C., Bustamante, P., 2018. High cadmium and mercury concentrations in the tissues of the orange-back flying squid, *Sthenoteuthis pteropus*, from the tropical Eastern Atlantic. Ecotoxicology and Environmental Safety, 163, 323–330. https://doi.org/10.1016/j.ecoenv.2018.07.087

Lischka, A., Pook, C.J., Pannell, J.L., Braid, H.E., Gaw, S., Bolstad, K.S., 2020a. Distribution of trace elements in the tissues of arrow squid (*Nototodarus sloanii*) from the Chatham Rise, New Zealand: Human health implications. Fisheries Research, 221, 105383.

Lischka, A., Lacoue-Labarthe, T., Bustamante, P., Piatkowski, U. and Hoving, H.J.T., 2020b. Trace element analysis reveals bioaccumulation in the squid *Gonatus fabricii* from polar regions of the Atlantic Ocean. Environmental Pollution, 256, 113389.

Lucassen, M., 2012. The expedition of the research vessel" Polarstern" to the Antarctic in 2012 (ANT-XXVIII/4). Berichte zur Polar-und Meeresforschung (Reports on Polar Research) 652.

Lucia, M., Strøm, H., Bustamante, P., Gabrielsen, G.W., 2016. Trace element concentrations in relation to the trophic behaviour of endangered Ivory Gulls (*Pagophila eburnea*) during their stay at a breeding site in Svalbard. Archives of Environmental Contamination and Toxicology 71, 518–529.

Macdonald, C.R., Sprague, J.B., 1988. Cadmium in marine invertebrates and arctic cod in the Canadian Arctic. Distribution and ecological implications. Marine Ecology Progress Series, 17–30.

Majer, A.P., Petti, M.A.V., Corbisier, T.N., Ribeiro, A.P., Theophilo, C.Y.S., de Lima Ferreira, P.A., Figueira, R.C.L., 2014. Bioaccumulation of potentially toxic trace elements in benthic organisms of Admiralty Bay (King George Island, Antarctica). Marine Pollution Bulletin 79, 321–325.

Malcolm, H.M., Boyd, I.L., Osborn, D., French, M.C., Freestone, P., 1994. Trace metals in Antarctic fur seal (*Arctocephalus gazella*) livers from Bird Island, South Georgia. Marine Pollution Bulletin 28, 375–380.

Michaels, A.F., Flegal, A.R., 1990. Lead in marine planktonic organisms and pelagic food webs. Limnology and Oceanography, 35(2), 287–295.

Matias, R., Seco, J., Gregory, S., Belchier, M., Pereira, M.E., Bustamante, P., Xavier, J.C. (2020) Antarctic octopod beaks as proxy for mercury concentrations in soft tissues. Marine Pollution Bulletin, 158, 111447.

Matias, R.S., Gregory, S., Ceia, F.R., Baeta, A., Seco, J., Rocha, M.S., Fernandes, E.M., Reis, R.L., Silva, T.H., Pereira, E., Piatkowski, U., Ramos, J.A., Xavier, J.C., 2019. Show your beaks and we tell you what you eat: Different ecology in sympatric Antarctic benthic octopods under a climate change context. Marine Environmental Research 150, 104757.

Miramand, P., Bentley, D., 1992. Concentration and distribution of heavy metals in tissues of two cephalopods, *Eledone cirrhosa* and *Sepia officinalis*, from the French coast of the English Channel. Marine Biology 114, 407–414.

Miramand, P., Guary, J.-C., 1980. High concentrations of some heavy metals in tissues of mediterranean octopus. Bulletin of Environmental Contamination and Toxicology 24, 783–788.

Neff, J.M., 1997. Ecotoxicology of arsenic in the marine environment. Environmental Toxicology and Chemistry 16, 917–927.

Negri, A., Gustavo A.D., Ceia, F., Vieira, R., Cherel, Y., Coria, N.R., Corbalán, A., Xavier, J.C. (2016). The cephalopod prey of the Weddell seal, *Leptonychotes weddellii*, a biological sampler of the Antarctic marine ecosystem. Polar Biology 39(3), 561–564.

Penicaud, V., Lacoue-Labarthe, T., Bustamante, P., 2017. Metal bioaccumulation and detoxification processes in cephalopods: A review. Environmental Research 155, 123–133. https://doi.org/10.1016/j.envres.2017.02.003

Petri, G., Zauke, G.-P., 1993. Trace metals in crustaceans in the Antarctic Ocean. Ambio-Journal of Human Environment Research and Management 22, 529–536.

Piatkowski, U., Allcock, A.L., Hevia, M., Steimer, S., Vecchione, M., 1998. Cephalopod ecology. In: Kattner, G. (ed.): The expedition ANTARKTIS XIV/2 of RV “Polarstern” in 1996/97. Berichte zur Polarforschung (Reports on Polar Research) 274, 41-49.

Piatkowski, U., Allcock, L., & Vecchione, M., 2003. Cephalopod diversity and ecology. In: Fütterer, D.K., Brandt, A., Poore, G.C.B. (eds.). The expedition ANTARKTIS-XIX/3-4 of the research vessel Polarstern in 2002. Berichte zur Polarforschung (Reports on Polar Research) 470, 32-38.

Pierce, G.J., Stowasser, G., Hastie, L.C., Bustamante, P., 2008. Geographic, seasonal and ontogenetic variation in cadmium and mercury concentrations in squid (Cephalopoda: Teuthoidea) from UK waters. Ecotoxicology and Environmental Safety 70, 422–432. https://doi.org/10.1016/j.ecoenv.2007.07.007

Potapowicz, J., Szumińska, D., Szopińska, M., Polkowska, Ż., 2019. The influence of global climate change on the environmental fate of anthropogenic pollution released from the permafrost: part I. Case study of Antarctica. Science of the Total Environment, 651, 1534–1548.

R Core Team (2017). R: A Language and Environment for Statistical Computing. R Found. Stat. Comput., Vienna, Austria. http://www.R-project.org

Raimundo, J., Costa, P.M., Vale, C., Costa, M.H., Moura, I. (2010). Metallothioneins and trace elements in digestive gland, gills, kidney and gonads of *Octopus vulgaris*. Comparative Biochemistry and Physiology Part C: Toxicology & Pharmacology 152(2), 139–146.

Rainbow, P.S., 1997. Trace metal accumulation in marine invertebrates: marine biology or marine chemistry? Journal of the Marine Biological Association of the United Kingdom, 77(1), 195–210.

Ratnasingham, S., Hebert, P.D., 2007. BOLD: The Barcode of Life Data System (http://www. barcodinglife. org). Molecular Ecology Notes, 7, 355–364.

Richert, J. E., Galván-Magaña, F., Klimley, A. P. (2015). Interpreting nitrogen stable isotopes in the study of migratory fishes in marine ecosystems. Marine Biology, 162(5), 1099–1110.

Rjeibi, M., Metian, M., Hajji, T., Guyot, T., Chaouacha-Chékir, R.B., Bustamante, P., 2014. Interspecific and geographical variations of trace metal concentrations in cephalopods from Tunisian waters. Environmental Monitoring and Assessment, 186, 3767–3783.

Rodhouse, P.G., Arnbom, T.R., Fedak, M.A., Yeatman, J., Murray, A.W.A., 1992. Cephalopod prey of the southern elephant seal, *Mirounga leonina* L. Canadian Journal of Zoology 70, 1007–1015.

Rosman, K.J.R., Chisholm, W., Boutron, C.F., Candelone, J.-P., Patterson, C.C., 1994. Anthropogenic lead isotopes in Antarctica. Geophysical Research Letters 21, 2669–2672.

Sanders, J.G., 1980. Arsenic cycling in marine systems. Marine Environmental Research, 3(4), 257–266.

Schwarz, R., Hoving, H.J.T, Noever, C., Piatkowski, U., 2019. Life histories of Antarctic incirrate octopods (Cephalopoda: Octopoda). PloS One, 14(7).

Seco, J., Xavier, J.C., Brierley, A.S., Bustamante, P., Coelho, J.P., Gregory, S., Fielding, S., Pardal, M.A., Pereira, B., Stowasser, G., Tarling, G.A., 2020. Mercury levels in Southern Ocean squid: Variability over the last decade. Chemosphere, 239, 124785.

Seixas, S., Bustamante, P., Pierce, G., 2005. Accumulation of mercury in the tissues of the common octopus *Octopus vulgaris* (L.) in two localities on the Portuguese coast. Science of The Total Environment 340, 113–122. https://doi.org/10.1016/j.scitotenv.2004.08.012

Shen, S., Li, X.F., Cullen, W.R., Weinfeld, M., Le, X.C., 2013. Arsenic binding to proteins. Chemical Reviews, 113(10), 7769–7792.

Storelli, M.M., Marcotrigiano, G.O., 2004. Content of mercury and cadmium in fish (*Thunnus alalunga*) and cephalopods (*Eledone moschata*) from the south-eastern Mediterranean Sea. Food Additives and Contaminants 21, 1051–1056. https://doi.org/10.1080/02652030400023127.

Strugnell, J.M., Allcock, A.L., Watts, P.C., 2017. Closely related octopus species show different spatial genetic structures in response to the Antarctic seascape. Ecology and evolution, 7(19), 8087–8099.

Sun, L., Xie, Z., 2001. Changes in lead concentration in Antarctic penguin droppings during the past 3,000 years. Environmental Geology 40, 1205–1208.

Szefer, P., Szefer, K., Pempkowiak, J., Skwarzec, B., Bojanowski, R., Holm, E., 1994. Distribution and coassociations of selected metals in seals of the Antarctic. Environmental Pollution 83, 341–349.

Taylor, V., Goodale, B., Raab, A., Schwerdtle, T., Reimer, K., Conklin, S., Karagas, M.R., Francesconi, K.A., 2017. Human exposure to organic arsenic species from seafood. Science of the Total Environment 580, 266–282.

Trevizani, T.H., Petti, M.A.V., Ribeiro, A.P., Corbisier, T.N., Figueira, R.C.L., 2018. Heavy metal concentrations in the benthic trophic web of Martel Inlet, Admiralty Bay (King George Island, Antarctica). Marine Pollution Bulletin 130, 198–205.

Tu, N.P.C., Agusa, T., Ha, N.N., Tuyen, B.C., Tanabe, S., Takeuchi, I., 2011. Stable isotope-guided analysis of biomagnification profiles of arsenic species in a tropical mangrove ecosystem. Marine Pollution Bulletin 63, 124–134.

Xavier, J., Rodhouse, P., Purves, M., Daw, T., Arata, J., Pilling, G., 2002. Distribution of cephalopods recorded in the diet of the Patagonian toothfish (*Dissostichus eleginoides*) around South Georgia. Polar Biology 25, 323–330.

Xavier, J.C., Croxall, J.P., 2007. Predator–prey interactions: why do larger albatrosses eat bigger squid? Journal of Zoology 271, 408–417.

Xavier, J.C., Cherel, Y., Allcock, L., Rosa, R., Sabirov, R.M., Blicher, M.E., Golikov, A.V., 2018. A review on the biodiversity, distribution and trophic role of cephalopods in the Arctic and Antarctic marine ecosystems under a changing ocean. Marine Biology, 165(5), 93.

Zauke, G.P., Savinov, V.M., Ritterhoff, J., Savinova, T., 1999. Heavy metals in fish from the Barents Sea (summer 1994). Science of the Total Environment, 227(2–3), 161–173.

Table 1. Specimen data for the eight octopod species included in this study. Acronyms are defined as follows: sample size (*n*), mean ± standard deviation (SD), mantle length (ML; mm), of the total length (TL; mm), and number of sex indetermined (Juvenile, J), female (♀) and male (♂) specimen.

Species	Sampling Depth (m)	*n*	ML Mean ± SD	ML min-max	TL Mean ± SD	TL min-max	J	♀	♂
Adelieledone polymorpha	293–323	15	75.1 ± 16.9	50–109	175.1 ± 43.9	110–265		7	8
Pareledone aequipapillae	295–320	4	38.5 ± 4.3	33–44	86.8 ± 19.1	67–110	2	2	
Pareledone albimaculata	295–320	5	45.4 ± 21.1	24–82	101.6 ± 21.7	70–128		4	1
Pareledone aurata	150–178	7	58.4 ± 26.2	28–100	142.4 ± 43.4	84–213		4	3
Pareledone charcoti	109–118	22	49.3 ± 7.7	35–63	112.7 ± 13.1	89–145		11	11
Pareledone cornuta	288–307	4	46.2 ± 6.3	38–54	104.0 ± 18.1	80–127	1		3
Pareledone felix	288–307	2	49.5 ± 17.9	34–65	125.0 ± 63.5	70–180	1	1	
Pareledone turqueti	288–307	1	46		112			1	

Table 2. Analysis of covariance (ANCOVA) for the linear models fitted to the δ^{13}C and δ^{15}N values of the mantle tissue from the different species used in this study. Abbreviations are defined as follows: TL = total length, Df = degrees of freedom, and 'depth' the sampling depth. Df – degrees of freedom. Asterisks indicate the level of significance: * = $p < 0.05$; ** = $p < 0.01$; and *** = $p < 0.001$.

	Df	Sum of squares	Mean square	F value	Significance
δ^{13}C					
Depth	1	18.22	18.22	61.55	***
Species	8	42.52	5.31	20.72	***
Size (TL)	1	0.16	0.16	0.63	
Sex	2	1.89	0.94	3.68	*
δ^{15}N	1	2.39	2.38	9.30	**
Residuals	47	12.05	0.26		
δ^{15}N					
Depth	1	45.33	45.33	372	***
Species	8	52.02	6.50	53.72	***
Size (TL)	1	0.11	0.11	0.93	
Sex	2	0.02	0.01	0.09	
δ^{13}C	1	1.16	1.16	9.56	**
Residuals	47	5.69	0.12		

Table 3. Output of the generalised linear models (GLMs) for variables that significantly influence the trace element concentrations of the analysed octopod species. Species included were: *A. polymorpha*, *P. aequipapillae*, *P. albimaculata*, *P. aurata*, *P. charcoti*, and *P. cornuta*. The *p*-values of the variables are shown according to likelihood ratio tests (*** 0.001, ** 0.01, * 0.05). Negative (↓) and positive (↑) effects for the continuous variable size (total length [TL]) are indicated with arrows (↑↑↑ = 0.001, ↑↑ = 0.01, ↑ = 0.05). For Cd, tissue type was not included in the GLM because it was not analysed (NA). Models for each element included δ^{15}N and the residuals (Resid) of δ^{13}C.

	Ag	As	Cd	Co	Cr	Cu	Fe	Hg	Ni	Pb	Se	V	Zn
Tissue type	***	***	NA	***	*	***	***		***	***	***	***	***
Species		***	***	**			*		***		**	***	***
Depth		***		***			***		***	**	***	*	
Size (TL)						↓↓↓					↑		↓
Sex			***	***		**	*			*			
δ^{15}N	↑	↑↑↑	↓								↑		
Resid ($\delta^{13}C$)													
Species x TL		**		*									

Table 4. Concentrations of Cd in the digestive gland and Hg (mean ± standard deviation [SD]) in muscular tissues of octopods from temperate and (sub-)Antarctic areas. All concentrations are presented in $\mu g\ g^{-1}$ dw. Abbreviations as followed: (^{A}) = arm muscles and (^{M}) = mantle muscles. Converted values from wet weight are indicated with *. ‘Year’ indicates the tissue collection date.

Species	Cd in the digestive gland Mean ± SD	Hg in the muscle Mean ± SD	Location	Year	Reference
Antarctic species					
Adelieledone					
A. polymorpha	112 ± 44.4	0.56 ± 0.13^{M}	Antarctic Peninsula	2012	This study
A. polymorpha		0.322 ± 0.088^{M}	South Georgia coast	2004	Matias el al., 2019
A. polymorpha		0.126 ± 0.032^{M}	South Georgia region	2004	Matias et al., 2020
Benthoctopus					
B. thielei	215		Kerguelen Islands	1995	Bustamante et al., 1998b
Graneledone					
G. gonzalezi	369		Kerguelen Islands	1995	Bustamante et al., 1998b
Pareledone					
P. aequipapillae	80.92 ± 23.02	0.20 ± 0.03^{M}	Antarctic Peninsula	2012	This study
P. albimaculata	85.89 ± 43.27	0.15 ± 0.02^{M}	Antarctic Peninsula	2012	This study
P. annata	175± 65.22	0.13 ± 0.03^{M}	Antarctic Peninsula	2012	This study
P. charcoti	152± 87.33	0.41 ± 0.22^{M}	Antarctic Peninsula	2012	This study
P. cornuta	112 ± 11.98	0.12 ± 0.01^{M}	Antarctic Peninsula	2012	This study
P. felix	76.4 ± 1.76	0.39 ± 0.03^{M}	Antarctic Peninsula	2012	This study
P. subtilis	164 ± 140	0.19 ± 0.04^{M}	Antarctic Peninsula	2012	This study
P. turqueti	204	0.28^{M}	Antarctic Peninsula	2012	This study
P. turqueti		0.434 ± 0.128^{M}	South Georgia Coast	2004	Matias el al., 2019

P. turqueti		0.196 ± 0.083^{M}	South Georgia region	2004	Matias et al., 2020
Temperate species					
Eledone					
E. cirrhosa	79.87 ± 4.38*		Cotentin Coast, English Channel	1987	Miramand and Bentley, 1992
E. cirrhosa	16.3 ± 9.6	0.34 ± 0.072^{M}	Bay of Biscay	2005-2008	Chouvelon et al., 2011
E. cirrhosa		6.06^{M}	Tyrrhenian Sea	1999	Barghigiani et al., 2000
E. cirrhosa	2333 ± 1000*	0.144± 0.071^{M}	Faroe Islands	1997	Bustamante et al. 1998a, 2006
Octopus					
O. vulgaris	50 ± 10		Monaco, Mediterranean Sea	1980	Miramand and Guary, 1980
O. vulgaris	94–185		Matosinhos, Portugal	2002	Raimundo et al., 2008
O. vulgaris	136–269		Northwest Coast, Portugal	2001	Raimundo et al., 2005
O. vulgaris	20–122		South Coast, Portugal	2001	Raimundo et al., 2005
O. vulgaris	65.8± 24.2	0.13± 0.02^{M}	Sfax, Tunisia	2010	Rjeibi et al. 2014
	42.5± 20.3	0.18± 0.06^{M}	Bizerte, Tunisia		
	31.0± 9.2	0.13±0.06^{M}	Monastir, Tunisia		
O. vulgaris		0.43 ± 0.13^{A}	Cascais, Portugal	2002-2003	Seixas et al., 2005
O. vulgaris		0.213 ± 0.02*^{M}	Azores	1990-1991	Monteiro et al., 1992

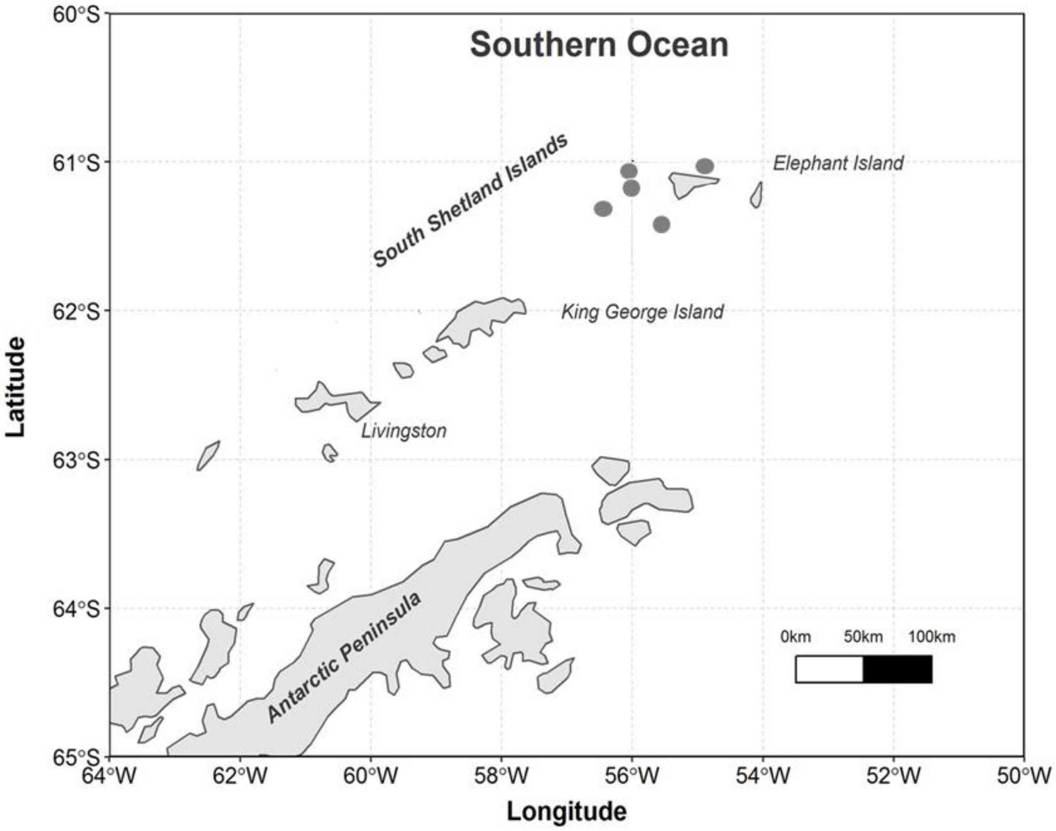

Figure 1. Sampling locality of the specimen used in this study (grey circles), adapted from Schwarz et al., 2019.

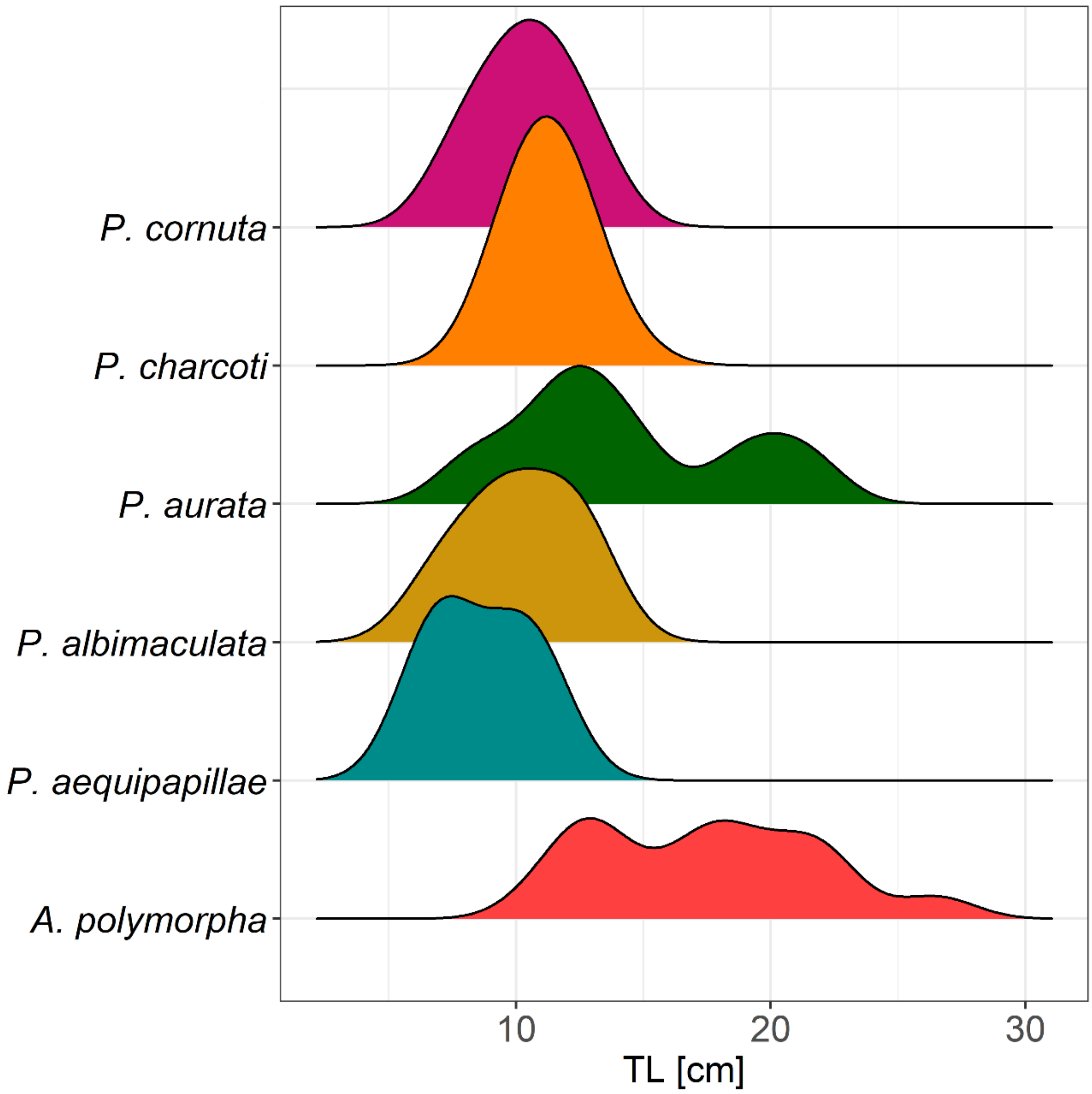

Figure 2. Frequencies of sizes for the analysed octopod specimens analysed in our study (only including species with *n*>3) from near Elephant Island, Antarctic Ocean.

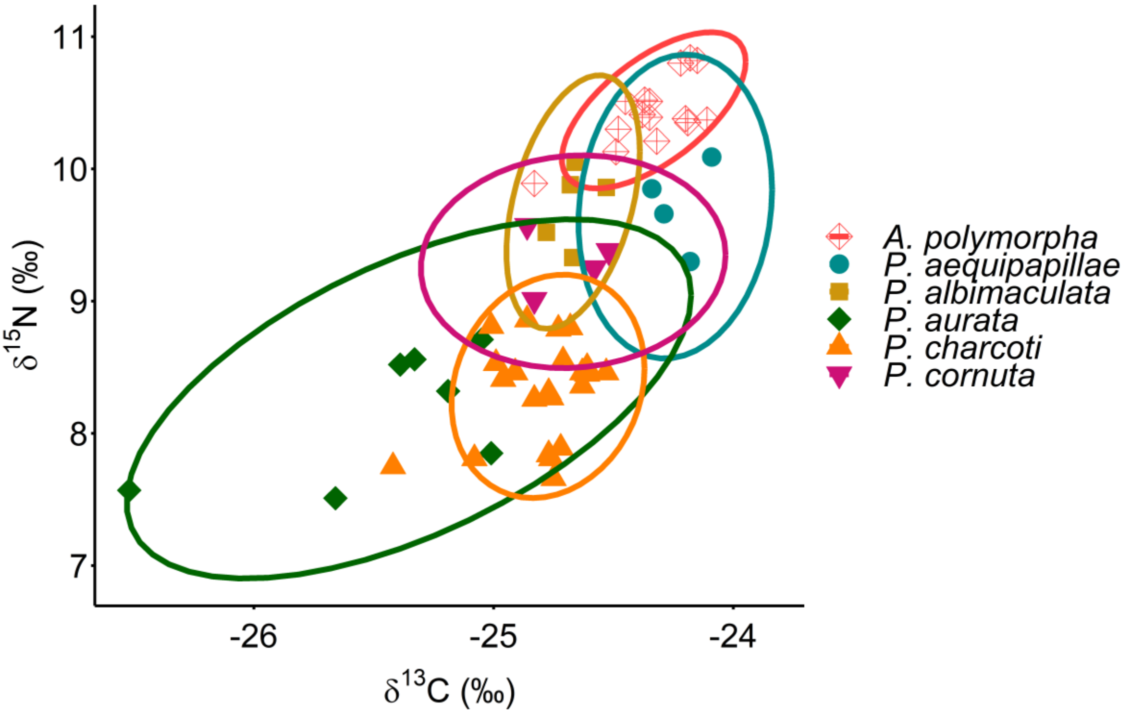

Figure 3. Carbon (δ^{13}C) and nitrogen (δ^{15}N) stable isotope values (‰) in eight octopods species from the genera *Adelieledone* and *Pareledone* collected from near Elephant Island, Antarctic Ocean. Ellipses indicate the 95% confidence interval around the species groupings.

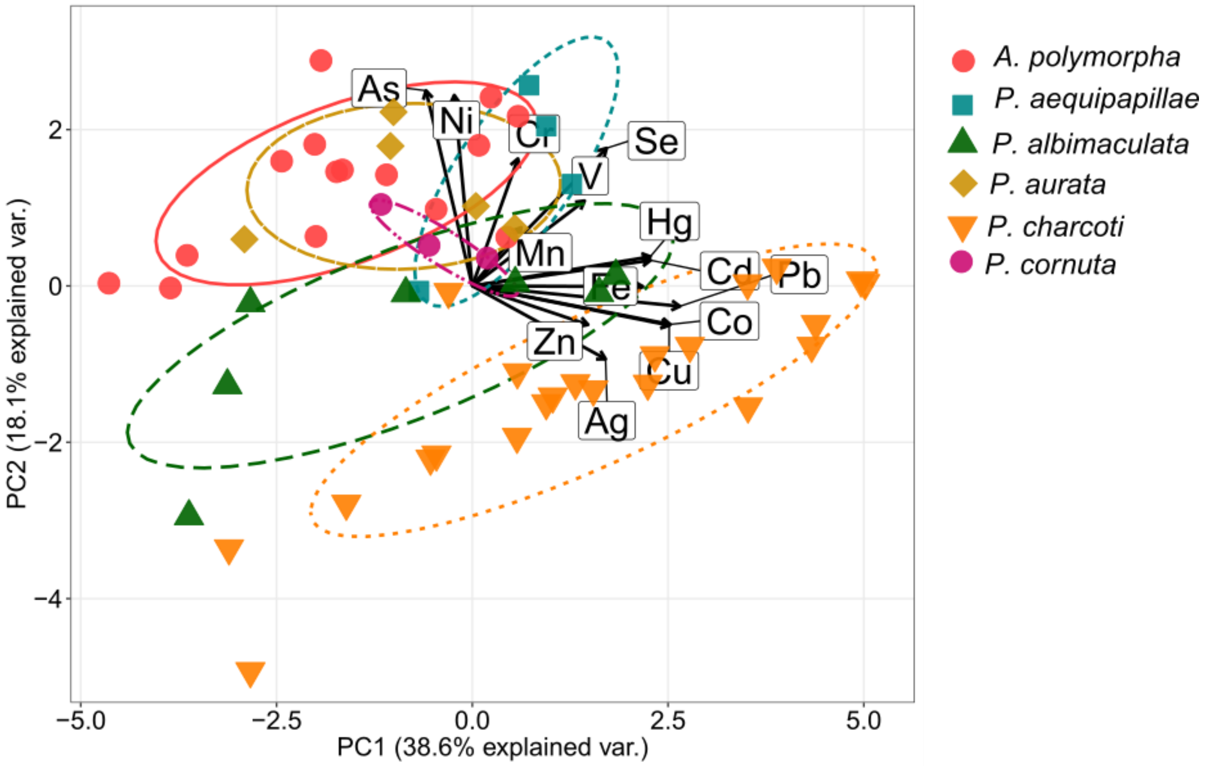

Figure 4. Principal component analysis (PCA) of the digestive gland trace element concentrations of six octopods species from the genera *Adelieledone* and *Pareledone* collected near Elephant Island, Antarctic Ocean. Due to the low sampling number, *P. felix* and *P. turqueti* were excluded from the PCA.

Supplementary Material

Table S1. Trace-element concentrations and stable isotope values in the digestive gland and mantle tissue (µg g^{-1} dw) of the eight octopod species analysed in this study, collected from near Elephant Island, Antarctic Ocean. Data is provided for the mean, standard deviation (SD), and minimum (min) and maximum (max) concentrations. NA= data not analysed.

Digestive Gland								
	A. polymorpha n=15		*P. aequipaillae n*=4		*P. albimaculata n*=5		*P. aurata n*=7	
	mean ± SD	min-max	mean ± SD	min-max	mean ± SD	min-max	mean ± SD	min-max
Ag	18.9 ± 20.8	3.56–68.5	9.25 ± 6.13	2.56–14.7	10.4 ± 10.0	2.87–26.5	11.5 ± 10.4	2.36–28.3
As	50.4 ± 7.79	36.4–60.9	43.3 ± 4.54	38.3–47.8	60.1 ± 10.8	51.5–77.4	27.3 ± 9.11	11.2–34.3
Cd	112 ± 44.4	50.9–193	80.9 ± 23.0	55.1–104	85.9 ± 43.3	54.8–153	169 ± 106	63.5–370
Co	5.46 ± 6.54	0.83–24.1	13.4 ± 11.4	2.48–29.5	4.64 ± 3.65	1.88–10.8	8.60 ± 6.48	2.11–18.0
Cr	0.44 ± 0.36	0.14–1.59	1.21 ± 0.88	0.28–2.37	0.63 ± 0.51	0.25–1.48	0.21 ± 0.07	0.16–0.37
Cu	369 ± 192	124.5–760	874 ± 129	757–1058	668 ± 339	332–1133	669 ± 392	300–1320
Fe	270 ± 188	53.1–689	2050 ± 786	1328–3104	1110 ± 656	370–2057	699 ± 485	169–1347
Hg	0.38 ± 0.05	0.28–0.48	0.6 ± 0.21	0.42–0.9	0.34 ± 0.10	0.23–0.48	0.41 ± 0.21	0.18–0.65
Mn	3.99 ± 1.39	2.63–8.05	16.7 ± 18.3	3.24–42.1	11.7 ± 10.5	3.68–28.4	5.94 ± 2.68	3.69–11.4
Ni	8.50 ± 3.97	2.81–17.6	5.84 ± 2.71	3.69–9.79	5.05 ± 1.05	3.72–6.25	4.13 ± 1.61	1.48–6.04
Pb	0.46 ± 0.35	0.10–1.19	1.03 ± 0.15	0.94–1.26	1.11 ± 0.51	0.48–1.75	0.92 ± 0.69	0.17–1.93
Se	14.6 ± 3.8	8.33–20.9	13.1 ± 1.44	12.0–15.2	11.7 ± 3.29	8.02–16.1	10.8 ± 4.52	4.87–16.9

V	35.2 ± 22.1	8.63–94.3	24.7 ± 4.35	19.2–29.8	56.7 ± 53.4	14.0–131	14.3 ± 12.3	1.63–32.0
Zn	251 ± 69.2	153–404	189 ± 44.8	141–247	187 ± 28.3	146–219	175 ± 64.8	103–299

	P. charcoti *n*=22		*P. cornuta* *n*=4		*P. felix* *n*=2	*P. turqueti* *n*=1
	mean ± SD	min-max	mean ± SD	min-max	min-max	concentration
Ag	40.4 ± 20.5	6.98–87.5	25.4 ± 30.5	7.9–71.1	5.98–8.52	4.90
As	20.8 ± 4.96	11.0–32.0	48.1 ± 8.3	41.0–58.5	29.7–55.9	77.0
Cd	152 ± 87.3	52.3–327	112 ± 12.0	97.0–126	75.2–77.6	204
Co	17.1 ± 9.84	2.45–35.1	4.09 ± 0.45	3.59–4.62	2.37–3.07	10.1
Cr	0.42 ± 0.43	0.10–1.99	0.48 ± 0.16	0.31–0.67	0.18–0.20	0.55
Cu	1166 ± 680	339–2553	933 ± 233	614–1111	229–399	823
Fe	1115 ± 609	349–2834	1359 ± 384	980–1864	545–820	1600
Hg	0.51 ± 0.20	0.18–0.92	0.30 ± 0.03	0.26–0.34	0.37–0.69	0.82
Mn	5.55 ± 1.61	3.08–8.35	11.3 ± 6.18	3.64–16.8	3.55–5.54	10.6
Ni	2.55 ± 1.19	1.07–5.81	5.19 ± 0.59	4.71–6.03	1.50–3.91	6.49
Pb	1.87 ± 1.30	0.50–4.46	1.28 ± 0.26	0.91–1.50	0.51–0.52	1.13
Se	12.3 ± 4.46	2.99–21.9	9.6 ± 2.12	7.42–12.5	7.58–12.1	18.8
V	53.5 ± 36.7	1.81–152	21.3 ± 11.5	9.16–36.4	9.91–43.7	103
Zn	322 ± 89.5	172–557	176 ± 26.9	149–203	237–304	262

Mantle

Mantle	*A. polymorpha n*=15		*P. aequipaillae* *n*=4		*P. albimaculata* *n*=5	
	mean ±sd	min-max	mean ±sd	min-max	mean ±sd	min-max

δ^{13}C	-24.34 ± 0.18	-24.83 – (-24.11)	-24.23 ± 0.11	-24.34-(-24.09)	-24.66 ±0.09	-24.78-(-24.53)
δ^{15}N	10.34 ± 0.26	9.89-10.84	9.72 ±0.33	9.30 -10.09	9.73± 0.29	9.33-10.05
Ag	NA	NA	3.04 ±0.90	2.04-4.15	1.76 ±0.92	0.75-2.95
As	65.4 ±10.9	47.2-89.7	52.9 ±4.34	47.9-58.1	72.3 ±10.7	58.4-84.2
Cd	NA	NA	NA	NA	NA	NA
Co	0.90 ±0.48	0.31-2.23	1.58 ±0.93	1.07-2.97	0.54 ±0.12	0.44-0.72
Cr	0.20 ±0.07	0.11-0.36	0.50 ±0.52	0.13-1.26	0.16 ±0.03	0.13-0.21
Cu	107 ±95.4	40.5-403	129 ±23.5	109-161	77.4 ±44.7	33.5-152.3
Fe	79.6 ±91.4	20.86-362	27.1 ±10.6	16.0-37.3	22.7 ±22.8	5.63-61.5
Hg	0.56 ±0.13	0.40-0.76	0.20 ±0.03	0.17-0.24	0.15 ±0.02	0.13-0.19
Mn	3.27 ±1.35	2.13-7.82	2.06 ±0.34	1.63-2.39	2.14 ±0.52	1.5-2.76
Ni	4.06 ±1.44	1.75-7.21	2.05 ±0.74	1.20-2.76	1.41 ±0.33	1.02-1.81
Pb	0.09 ±0.04	0.05-0.17	0.07 ±0.01	0.06-0.08	0.12 ±0.04	0.09-0.18
Se	7.09 ±2.02	3.54-11.2	5.45 ±0.97	4.13-6.42	4.45 ±0.06	4.36-4.51
V	7.65 ±4.31	3.45-18.7	3.73 ±2.67	1.40-7.45	5.90 ±6.39	1.28-16.4
Zn	151 ±41.2	103-244	89.5 ±7.34	84.0-99.7	91.2 ±12.4	78.7-111
	P. charcoti n=22		*P. cornuta n*=4		*P. felix n*=2	*P. turqueti n*=1
	mean ±sd	min-max	mean ±sd	min-max	min-max	concentration
δ^{13}C	-24.81±0.20	-25.42-(-24.53)	-24.70±0.17	-24.86-(-24.52)	-24.89-(-23.97)	-24.11
δ^{15}N	8.32±0.37	7.66-8.86	9.3±0.24	9.01-9.57	9.78-10.03	9.87
Ag	0.29 ±0.42	0.04-1.66	1.09 ±0.64	0.51-1.78	2.2-6.29	2.19
As	22.1 ±4.64	15.4-30.9	66.4 ±8.23	60.5-78.6	51.0-55.3	97.9
Cd	NA.	NA.	NA.	NA.	NA.	NA.
Co	0.27 ±0.25	0.05-1.15	0.37 ±0.09	0.27-0.49	1.43-1.64	1.66
Cr	0.24 ±0.23	0.11-1.15	0.14 ±0.04	0.11-0.19	0.12-0.22	0.15
Cu	33.5 ±21.2	15.9-98.7	53.1 ±22.5	34.7-82.9	73.7-202	134
Fe	9.36 ±6.79	5.51-39.0	29.6 ±20.2	7.80-52.9	26.8-131	30.6
Hg	0.41 ±0.22	0.15-1.13	0.12 ±0.01	0.12-0.13	0.37-0.41	0.28
Mn	3.63 ±1.83	1.43-7.48	2.67 ±0.88	1.98-3.95	1.66-2.52	3.29
Ni	0.52 ±0.14	0.30-0.76	0.86 ±0.12	0.78-1.04	0.97-2.26	2.63
Pb	0.05 ±0.03	0.03-0.15	0.10 ±0.01	0.09-0.11	0.06-0.13	0.15
Se	2.34 ±0.40	1.84-3.42	2.90 ±0.54	2.57-3.70	5.97-7.64	6.61
V	1.66 ±0.53	0.91-2.77	1.43 ±0.37	1.12-1.95	6.95-16.0	21.2
Zn	99.9 ±16.8	72.1-136	96.2 ±16.5	76.1-112.1	125-154.7	106

Table S2. Analysis of covariance (ANCOVA) for the linear models fitted to the trace element concentrations of *Pareledone charcoti.* Asterisks indicate the level of significance: * = p < 0.05; ** = p < 0.01; and *** = p < 0.001. NA= not analysed. The following variables were analysed: size (total length; TL); depth (sampling depth), δ^{15}N (nitrogen stable isotope ratio); and Resd *(*δ^{13}C) (residuals of δ^{13}C). Negative (↓) and positive (↑) effects for the continuous variable size (total length [TL]) are indicated with arrows (↑↑↑ = 0.001, ↑↑ = 0.01, ↑ = 0.05).

	Ag	As	Cd	Co	Cr	Cu	Fe	Hg	Ni	Pb	Se	V	Zn
Tissue	***		NA	***		***	***		***	***	***	***	***
Sex			***	***		**	**		***	***	**		***
Size													
δ^{15}N								↑			↑		↑
Resd(δ^{13}C)													

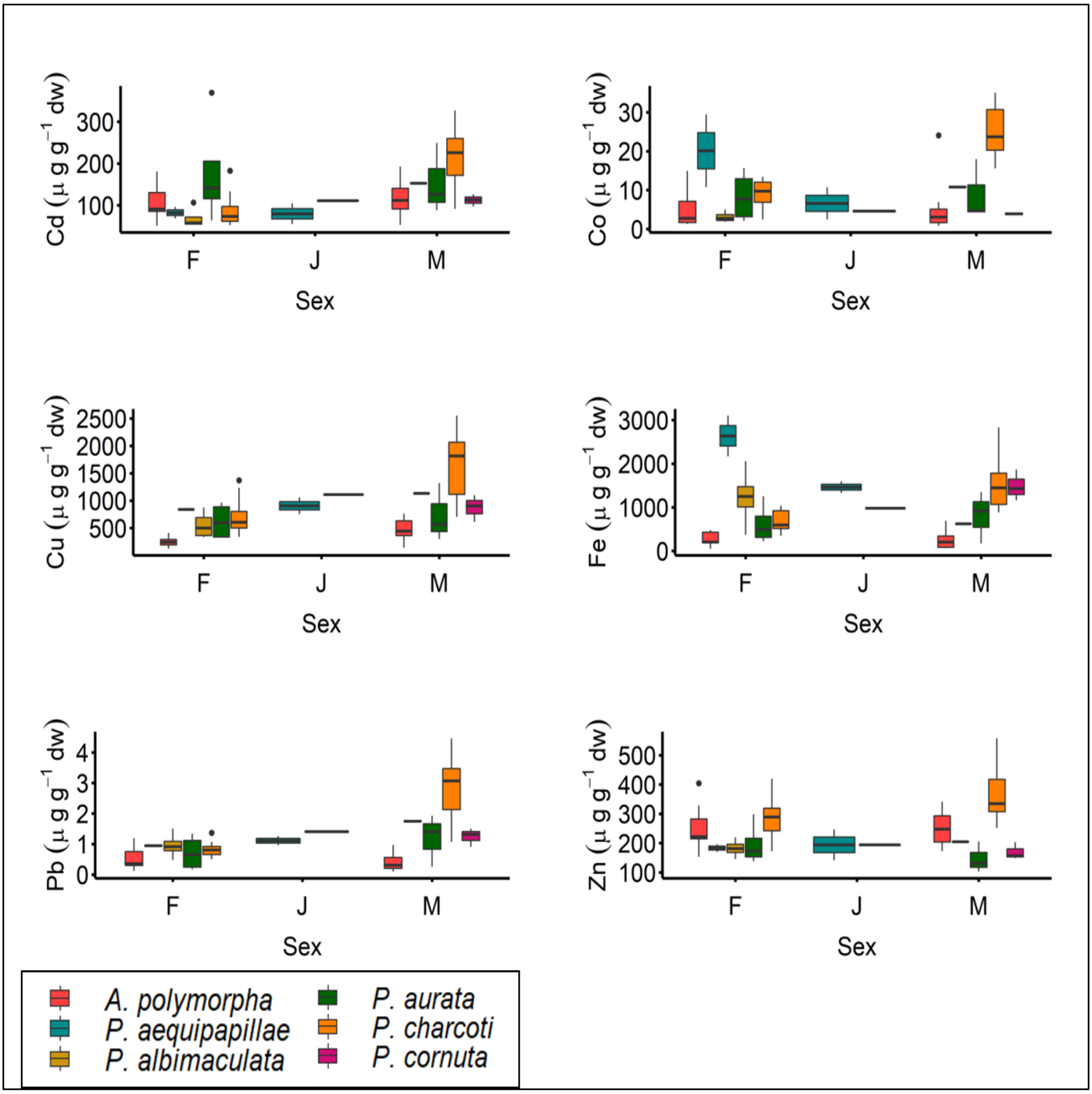

Figure S1. Digestive gland concentrations of trace elements (µg g^{-1} dw) between female (F), sex indet./juvenile (J) and male (M) specimens of six octopod species in the genera *Adelieledone* and *Pareledone* from the Antarctic Ocean. Elements that had significant differences in the GLMs are included (Cd, Co, Cu, Fe, Pb, and Zn). *Pareledone felix* and *P.turqueti* were excluded from the GLMs due to their low sampling size.

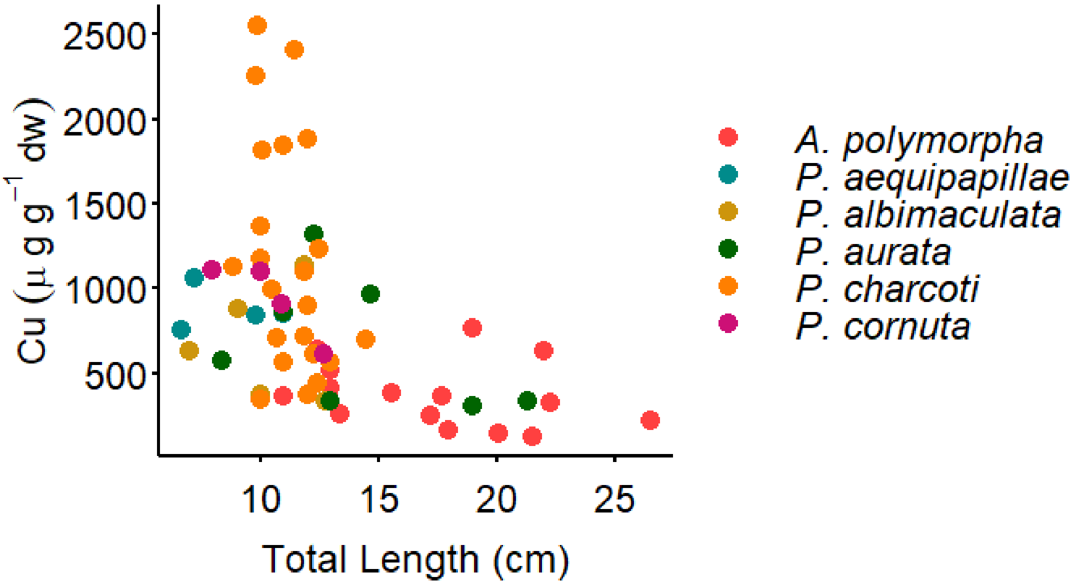

Figure S2. Digestive gland Cu concentrations (µg g^{-1} dw) of six octopod species in the genera *Adelieledone* and *Pareledone*, from the Antarctic Ocean in relation to size (total length; TL). *Pareledone felix* and *P. turqueti* were excluded from the GLMs due to their low sampling size.