High mercury levels in Antarctic toothfish *Dissostichus mawsoni* from the Southwest Pacific sector of the Southern Ocean

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Abstract

Mercury is a bioaccumulating toxic pollutant which can reach humans through the consumption of contaminated food (e.g. marine fish). Although the Southern Ocean is often portrayed as a pristine ecosystem, its fishery products are not immune to mercury contamination. We analysed mercury concentration (organic and inorganic forms – T-Hg) in the muscle of Antarctic toothfish, *Dissostichus mawsoni*, a long-lived top predator which supports a highly profitable fishery. Our samples were collected in three fishing areas (one seamount and two on the continental slope) in the Southwest Pacific Sector of the Southern Ocean during the 2016/2017 fishing season. Mercury levels and the size range of fish varied between fishing areas, with the highest levels (0.68 ± 0.45 mg kg^{-1} wwt) occurring on the Amundsen Sea seamount where catches were dominated by larger, older fish. The most parsimonious model of mercury concentration included both age and habitat (seamount versus continental slope) as explanatory variables. Mean mercury levels for each fishing area were higher than those in all previous studies of *D. mawsoni*, with mean values for the Amundsen Sea seamount exceeding the 0.5 mg kg^{-1} food safety threshold for the first time. It might therefore be appropriate to add *D. mawsoni* to the list of taxa, such as swordfish and sharks, which are known to exceed this threshold. This apparent increase in mercury levels suggests a recent contamination event which affected much of the Southwest Pacific sector, including both the Amundsen and Dumont D'Urville seas.

Keywords: Fisheries, Contaminants, Antarctic resources, Trace elements.

Funding

José Queirós was supported by FCT/MCTES through national funds (PIDDAC) and Portuguese Polar Program (PROPOLAR). He was also supported by a travel grant of SCAR Ant-ERA.

FCT-Portugal supported José Seco through a PhD grant (SRFH/PD/BD113487).

João Pedro Coelho is funded by CESAM (UID/AMB/50017/2019) and the Integrated Program of SR&TD 'Smart Valorization of Endogenous Marine Biological Resources Under a Changing Climate' (Centro-01-0145-FEDER-000018), co-funded by Centro 2020 program, Portugal 2020 and the European Regional Development Fund.

NHK Japan supported the cost onboard of José Queirós and material for the field work.

1 Introduction

Mercury is a natural pollutant that is released to the environment via processes such as hydrothermal venting and coastal erosion (Mason et al., 2012). However, its concentration in marine environments increased during the Industrial Era mostly due to the intensification of anthropogenic activities and climate change (Amos et al., 2013; Krabbenhoft and Sunderland, 2013; Stern et al., 2012; Streets et al., 2011; Sunderland and Mason, 2007). The Southern Ocean [here defined as the area managed by the Commission for the Conservation of Antarctic Marine Living Resources (CCAMLR), which covers c. 10% of the global marine area and has a northern boundary approximating the position of the Antarctic Polar Front; (CCAMLR, 2020)] is remote from anthropogenic sources and is often portrayed as a pristine ecosystem (Chown et al., 2017). Nonetheless observed concentrations can be as high as those elsewhere in the World Ocean because mercury is transported *via* oceanic and atmospheric currents (Cossa et al., 2011; Mason et al., 2012). As in other oceans, mercury concentrations vary depending on several factors, e.g. sea ice area and ocean bottom morphology (Aston and Fowler, 1985; Cossa et al., 2011).

Mercury accumulates along food-chains with top predators generally having the highest concentrations (Mason et al., 2012). Such values are dependent on the initial intake by primary producers (Mason et al., 2012; Mason et al., 1996), and increase at every trophic step (biomagnification), which suggests that top predators can be good indicators of ecosystem contamination (Becker et al., 2016; Blévin et al., 2013). High concentrations of mercury have been observed in top predators in the Southern Ocean, such as wandering albatrosses *Diomedea exulans* (up to 94 µg g^{-1} dw), reflecting high levels in the environment (Bustamante et al., 2016; Carravieri et al., 2016; Fontaine et al., 2015). Mercury is also a powerful toxin for humans, raising concerns about the health effects of consumption of contaminated animals, particularly marine fish (EPA, 2001; Hanchet et al., 2012; Karagas et al., 2012; Mason et al., 2012).

Antarctic toothfish *Dissostichus mawsoni* (family: Nototheniidae) is a top predator, feeding mainly on fish and squid (Fenaughty et al., 2003; Stevens et al., 2014), and is a valuable fisheries resource in the Southern Ocean (Hanchet et al., 2015). It is a benthopelagic species with a circumpolar distribution South of the Antarctic Polar Front (Duhamel et al., 2014), and a more southerly distribution than its congener *D. eleginoides.* It is found in depths < 2000m and is widely distributed on the continental shelf and slope around the Antarctic continent. It is also associated with offshore topographic features such as seamounts (Hanchet et al., 2015). *D. mawsoni* has a life-span of ~40 years (Brooks et al., 2011) and can reach lengths >1.5m and mass >100kg (Hanchet et al., 2015). These characteristics predispose *D. mawsoni* to accumulate mercury and persistent organic pollutants and suggest that the species is a potential indicator of mercury levels in the ecosystem (Corsolini et al., 2017; Sydeman et al., 2015). Current catches of *D. mawsoni* are ~4000 tonnes per year (CCAMLR, 2018), and its value varies between 25 and 50 US$ kg^{-1}, suggesting that the *D. mawsoni* fishery can generate revenues of more than 150 million US$ annually (Ainley et al., 2012; Baird, 2006; CCAMLR, 2016b; Grilly et al., 2015). This high value fishery is a potential source of mercury contamination for humans (Mason et al., 2012). Nevertheless, few research studies have addressed mercury levels in *D. mawsoni* (Hanchet et al., 2012; Son et al., 2014; Yoon et al., 2018).

These previous studies show that mercury levels in *D. mawsoni* can vary with biological, spatial and temporal factors, i.e. fish length, weight, maturation state, capture site, depth and year (Hanchet et al., 2012; Son et al., 2014; Yoon et al., 2018). The studies also suggest that average mercury values increased between 1998 and 2016 (~0.10 mg kg^{-1} in 1998, ~0.17 mg kg^{-1} in 2006 and 2012 and ~0.20 mg kg^{-1} in 2016) (Hanchet et al., 2012; Son et al., 2014; Yoon et al., 2018), but have remained below the upper food safety limit of 0.5 mg kg^{-1} for most marine species recognized by several agencies (EC, 2006; FSANZ, 2004). These previous studies provide limited information about how mercury levels vary between locations and how this interacts with the size and age of fish. Also, no recent (post 2006) data are available for the Amundsen Sea, which is an emerging fishery area (Brooks et al., 2016).

In this study, mercury concentrations in *D. mawsoni* muscle from fish captured during the Austral summer of 2016/2017 in the Amundsen and Dumont D'Urville Seas were analysed to (1) determine and compare mercury levels in *D. mawsoni* captured in three different Antarctic fishing areas (comprising samples from the slope and seamount habitats, the main fishing grounds for Antarctic toothfish; Figure 1); (2) evaluate the relationship between mercury levels in *D. mawsoni* and biological (standard length, mass, age), and habitat variables (fishing area, seabed steepness, depth, geomorphology); and (3) assess changes in mercury levels in *D. mawsoni* in the last two decades by comparing our findings with previous studies.

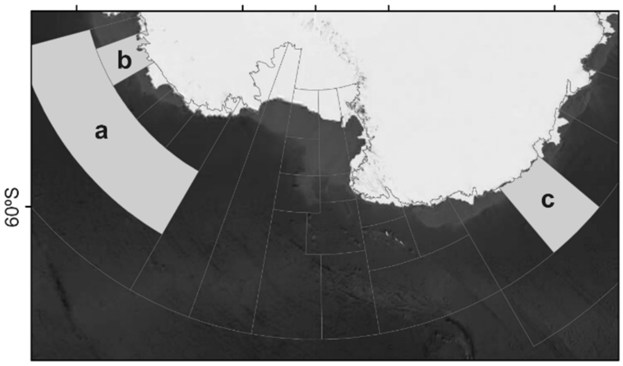
2 Materials and methods

2.1 Data Collection

Fieldwork was carried out between December 2016 and February 2017 on board a commercial fishing vessel licensed by CCAMLR (CCAMLR, 2017). Fish were captured using a bottom longline - autoline system (Fenaughty, 2008), baited with squid (Family: Ommastrephidae), in the Amundsen Sea seamount (n=39; CCAMLR 88.2H), Amundsen Sea slope (n=36; CCAMLR 88.2F) and Dumont D'Urville slope (n= 7; CCAMLR 58.4.1G) fishing areas (Figure 1). Specific coordinates of fishing locations are commercially sensitive and therefore not disclosed. However, these coordinates were included in relevant analyses.

Sagittal otoliths (hereafter otoliths) and muscle (fillet of < 30mm long and 10mm thick from the zone between the head and the first dorsal fin) were obtained from randomly selected fish heads (numbers given above) after the heads were removed by fishermen. Muscle samples were frozen and stored at -40ºC prior to analysis.

Figure 1. Sampling areas. a) Fishing area 88.2H – Amundsen Sea seamount; b) Fishing area 88.2F – Amundsen Sea slope; c) Fishing area 58.4.1G - Dumont D'Urville Sea slope.



2.2 Assessment of length, mass and weight of *D. mawsoni*

Otolith length (OL) and mass (OM) were measured using a digital calliper (±0.01mm) and a Mettler Toledo® UMX2 ultra-microbalance, respectively. Standard length (SL in mm), mass (M in g) and age (in years) were estimated using allometric equations from the literature

$$SL = 90.83046\ OL - 94.981$$ (Williams and McEldowney, 1990)

$$M = 2.17 \times 10^{-6}\ SL^{3.317}$$ (Williams and McEldowney, 1990)

$$Age = OM / 0.0128 + 0.0062$$ (Horn et al., 2003).

The OL and OM estimates used in the calculations were the averages of the left and right otolith values for each individual.

2.3 Total mercury (T-Hg) concentration in *D. mawsoni* muscle

Muscle samples were lyophilized and homogenized, and a microbalance was used to prepare a 35mg sample for mercury determination. Total mercury (hereafter T-Hg) was determined by atomic absorption spectrometry (AAS) using the Advanced Mercury Analyser (AMA) LECO® 254 (Costley et al., 2000), with a detection limit of 0.01ng of T-Hg, following Seco et al. (2019) at CESAM - Department of Chemistry, University of Aveiro (Portugal). Duplicates of each sample were performed to assess the precision of the analysis, with the coefficient of variation between replicates being 8.6 ± 8.9 %. Wet weight T-Hg was calculated using the water % in *D. mawsoni* muscle (% H_2O= ~26) calculated from the difference between the sample mass before and after lyophilization. To ensure the trueness of the results, an analytical quality control was performed using the certified reference material TORT-3 (lobster hepatopancreas T-Hg= 0.29 mg kg^{-1}) obtaining an extraction efficiency of 83 ± 5.3 %.

2.4 Relationship between T-Hg and habitat variables

Environmental T-Hg concentrations vary throughout the Southern Ocean, changing with topography, decreasing with depth (although methyl-mercury has similar concentrations throughout the water column) and distance from the continental coast and ice-pack (Cossa et al., 2011; Mason et al., 2012). *D. mawsoni* fishery occurs mainly on the slope, closer to the coast, and on offshore seamounts. CCAMLR manages the fishery using spatial units called Small Scale Research Units (CCAMLR, 2016a), which we refer to as fishing areas for brevity (Figure 1). Thus, to evaluate the influence of habitat on the T-Hg concentrations in *D. mawsoni* we considered the following candidate variables: seabed steepness, depth and geomorphology. Seabed steepness was calculated using the equation,

$$\% = (A\text{-}B) / \text{Line length (m)} \times 100$$

where A and B are the depth (in m) at each end of the longline. Depth at A and B and line length were obtained using vessel logbook data. Depth (D in m) was the average for the line, calculated using,

$$D = (A+B)/2$$

Geomorphology was a two-level categorical variable distinguishing the seamount area in the Amundsen Sea from the continental slope areas in the Amundsen and Dumont D´Urville seas. Depth and seabed steepness were continuous variables. Geomorphology was determined using the GPS coordinates in ArcGIS® ArcMap™ v10.2 with bathymetric basemap.

2.4 Statistical analyses

D. mawsoni biological variables (standard length (SL), mass and age) and T-Hg were tested for normality using the Shappiro-Wilk normality test. Differences between sampling areas in terms of *D. mawsoni* SL, mass, age and T-Hg were initially tested using the Kruskal-Wallis test, followed by a Dunn's multiple comparison test. A Mann-Whitney test was used to test for differences in T-Hg between samples from slope (Fishing area 88.2F and 58.4.1G) and seamount (Fishing area 58.4.1G) areas. These tests were executed in GraphPad Prism® v6.01.

Following these initial analyses, Generalized Linear Mixed Models (GLMMs) were used to provide a robust analysis of the relationship between T-Hg concentration and the candidate biological variables (SL, mass and age) and habitat variables (depth, geomorphology and steepness). Both T-Hg and SL were log transformed before the analysis as, for fish, the relationship between these variables approximates to linear following transformation (Gewurtz et al., 2011; Thomas et al., 2018).

Before GLMM, collinearity between candidate explanatory variables was tested using the "vif" function from the "car" package in R (Fox and Weisberg, 2019) to perform VIF (Variation Inflation Factor) analysis of both biological (SL, mass and Age) and habitat (Depth, Steepness and Geomorphology (slope vs seamount)) variables (Annex 1). Strong collinearity effects were found between SL and mass (biological variables) and Steepness and Geomorphology (habitat variables) suggesting that these pairs of variables should not be used together. We proceeded with SL because this variable has been used in previous fish studies (Gewurtz et al., 2011; Thomas et al., 2018) and with Geomorphology as this variablereflects the key geographical differences between sampling sites.

GLMMs were used to construct random intercepts models with longline as a grouping variable and the following candidate fixed effect variables – SL, age, geomorphology and depth as additive terms. The inclusion of longline as a grouping variable allows for the possibility that the baseline mercury level might vary between the specific locations where longlines are set. Model selection proceeded by discarding the variable with the highest *p*-value and concluded when all remaining variables were significant at $p<0.05$. AIC (Akaike Information Criterion) was used to confirm the improvement of the model throughout the selection process (Annex 2). GLMMs were performed using the function "lmer" from the "lme4" package (Bates et al., 2015) and R^2 was calculated using the function "r.squaredGLMM" from the "MuMIn" package (Barton, 2019) in the R environment (R core team, 2019). Figures and graphs were prepared using Adobe® Illustrator CC 2015.

Table 1: Fish biometrics and T-Hg concentrations in the three sampled areas. SSRU – small scale research unit; SL – standard length; M – mass; T-Hg – total mercury concentration. Values are mean ± standard deviation. Values with different superscript letters in the same column are significantly different (p <0.05).

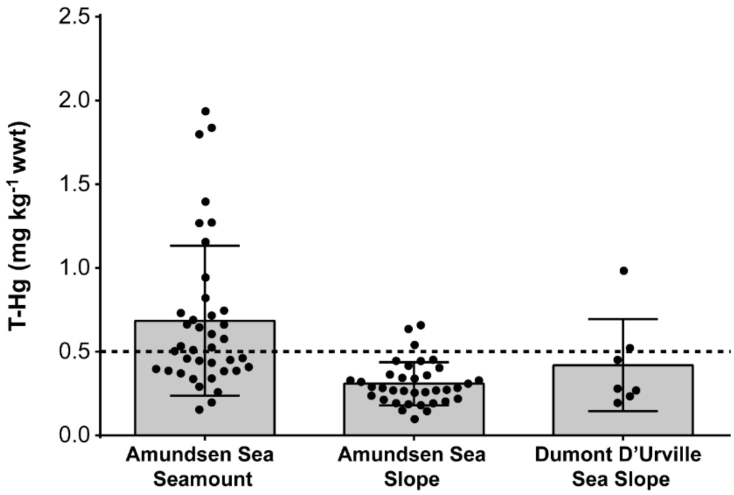
Fishing area	Fishing Area (SSRU)	n		Biological variables			Mercury
		longlines	fish	SL (mm)	M (Kg)	Age (years)	T-Hg (mg kg^{-1} wwt)
Amundsen Sea seamount	88.2 H	9	39	716 ± 66^{a}	6.6 ± 2.0^{a}	15 ± 2.7^{a}	0.68 ± 0.45^{a}
Amundsen Sea slope	88.2 F	6	37	572 ± 86^{b}	3.3 ± 1.9^{b}	6.6 ± 3.0^{b}	0.31 ± 0.13^{b}
Dumont D'Urville Sea slope	58.4.1 G	1	7	$678 \pm 87^{a,b}$	$5.7 \pm 2.5^{a,b}$	14 ± 3.8^{a}	$0.42 \pm 0.28^{a,b}$
Statistics (Kruskal-Wallis)				$p < 0.001$ U = 36.36	$p < 0.001$ U = 36.36	$p < 0.001$ U = 49.99	$p < 0.001$ U = 27.52

3 Results

3.1 Spatial differences in mercury levels

T-Hg values obtained in this study ranged from 0.10 to 1.94 mg kg^{-1} wwt, with a mean value (± SD) of 0.50 ± 0.37 mg kg^{-1} wwt and a median of 0.39 mg kg^{-1} wwt. Fish captured on the Amundsen Sea seamount (88.2H) had the highest T-Hg concentrations (0.15 to 1.94 mg kg^{-1} wwt, n = 39) and fish captured on the Amundsen Sea slope had the lowest concentrations (0.10 to 0.66 mg kg^{-1} wwt, n = 37) (Table 1). *D. mawsoni* captured in the Dumont D'Urville Sea had values ranging from 0.19 to 0.98 mg kg^{-1} wwt (n = 7) (Table 1, Figure 2). Dunn's multiple comparisons test identified significant differences in T-Hg values between the Amundsen Sea seamount (Fishing area 88.2H) and Amundsen Sea slope (Fishing area 88.2F) (Table 1). T-Hg values from the seamount area were typically higher than those from the slope areas (Mann-Whitney test: U = 282.0, p < 0.001) with a difference between means of ~0.36 mg kg^{-1}.

Figure 2. T-Hg concentrations in *Dissostichus mawsoni* in the three fishing areas. Bar ± Error Bar - Mean ± SD; Points - individual values; Dashed line - 0.5 mg kg^{-1}.



3.2 Relationship between mercury concentrations in *D. mawsoni* and biological and habitat variables.

Fish captured on the Amundsen Sea slope (Fishing area 88.2F) were, on average, the shortest, lightest and youngest fish, while those captured on the Amundsen Sea seamount (Fishing area 88.2H) were the longest, heaviest and oldest (Table 1). Dunn's multiple comparisons test identified significant differences between fishing areas for all biological variables (Table 1): Fish captured on the Amundsen Sea seamount had higher SL and mass than those captured on the Amundsen Sea slope, and fish captured on the Amundsen Sea slope were younger than those captured in the other fishing areas (Table 1).

The GLMM analysis identified one habitat variable, geomorphology (slope versus seamount), and one biological variable, age, as the best suite of predictors for mercury concentrations in *D. mawsoni* muscle (Table 2). The model was not improved by the inclusion of either depth ($p \approx 1.00$) or standard length ($p \approx 0.67$) (Annex 1). There was little difference between intercepts for different levels of the grouping variable, longline (variance = 0.0006), and the similar values for R^2m and R^2c confirm that the fixed effect variables explain almost all of the explained variability (Table 2).

Table 2: GLMM analysis final model. This is a random-intercept model with age and geomorphology as fixed-effect variables and fishing longlines as the random-intercept grouping variable. Significant explanatory variables for T-Hg in *Dissostichus mawsoni*. SE: Standard Error; df: degrees of freedom. R^2m: marginal pseudo-R^2 (variability explained by fixed effects); R^2c: conditional pseudo-R^2 (variability explained by fixed and random effects)

Variable	Estimate	SE	df	t-value	*p*-value	R^2m	R^2c
(intercept)	-0.46	0.11	34	-4.23	< 0.01		
Age	0.01	0.01	43	2.14	0.04	0.33	0.34
Geomorphology (slope)	-0.18	0.07	15	-2.57	0.02		

4 Discussion

4.1 Mercury concentrations in *D. mawsoni* from the Amundsen and Dumont D'Urville Seas

Mean mercury levels in *D. mawsoni* muscle recorded in the current study were higher than those reported in all previous studies (Hanchet et al., 2012; Son et al., 2014; Yoon et al., 2018). The highest single value in this study (1.94 mg kg^{-1} wwt) was almost 3 times higher than the previous maximum value [~0.70 mg kg^{-1} in *D. mawsoni* captured in 2006 (Hanchet et al., 2012)]. The mean value across all individuals in this study was 0.50 mg kg^{-1}, which is recognised as an upper limit for mercury levels in most marine species by various food safety agencies (EC, 2006; FSANZ, 2004). These agencies recognise a higher limit of 1 mg kg^{-1} for predatory marine species consumed by humans, including swordfish and sharks, known to have higher mercury concentrations. The current study results suggest that it might be appropriate to add *D. mawsoni* to these lists of species with higher mercury concentrations. Indeed, 33% of the studied individuals (including 56% of those from the Amundsen Sea seamount) had mercury levels >0.5 mg kg^{-1}.

The current study found a significant difference between individuals inhabiting slope and seamount sites. Individuals from the Amundsen Sea seamount had on average ~0.36 mg kg^{-1} wwt higher mercury levels than those from the neighbouring Amundsen Sea slope (Table 1). This result confirms that part of the *D. mawsoni* population, namely older individuals inhabiting seamount areas, has the capacity to accumulate high mercury levels. Similar spatial variability has been observed in the subantarctic congener *D. eleginoides* with contrasting results from Chile, the Prince Edwards Islands (Southern Indian Ocean) and South Georgia [Southern Atlantic Ocean (Guynn and Peterson, 2008)].

The analysis identified age as the best biological predictor of mercury levels in *D. mawsoni*, confirming that mercury bioaccumulates throughout the organism's life cycle (Jakimska et al., 2011). In a long-lived organism such as *D. mawsoni*, age may be better than size as an indicator of cumulative consumption of contaminated material. Previous studies of mercury in *Dissostichus* species have not examined relationships with age but have identified relationships with more easily acquired metrics of size (Guynn and Peterson, 2008; Hanchet et al., 2012; McArthur et al., 2003; Méndez et al., 2001; Son et al., 2014). Because of the considerable collinearity between age, length and mass (Annex

1) there is a high degree of comparability between studies. Furthermore, as age is related with an ontogenetic habitat change, thus different prey availability, it might reflect some biomagnification throughout the food-web.

D. mawsoni of all ages are found in shelf, slope and seamount areas (Hanchet et al., 2015). Nonetheless the predominant pattern is that juveniles mainly inhabit the continental shelf, moving to the slope and deeper waters as they grow. Consequently reproducing adults inhabit deeper areas further away from the continent (Hanchet et al., 2008). This leads to an expectation of increasing mercury concentration with depth based on age alone. However, the older mean age of fish in the Amundsen Sea seamount is not sufficient to explain the higher levels of mercury in this area.

Some of the observed spatial variability in mercury concentrations is related to habitat (seamount vs slope). The higher mercury levels in fish caught further away from the Antarctic continent contrasts with higher mercury levels in seawater and zooplankton (e.g. Antarctic krill *Euphausia superba*) closer to the continent (Cossa et al., 2011; Seco et al., 2019). The reasons for this contrast are currently unknown, but it does suggest differences in the factors affecting mercury concentration in the upper pelagic layers used by Antarctic krill and the seabed habitat used by *D. mawsoni*. Potential factors are discussed below.

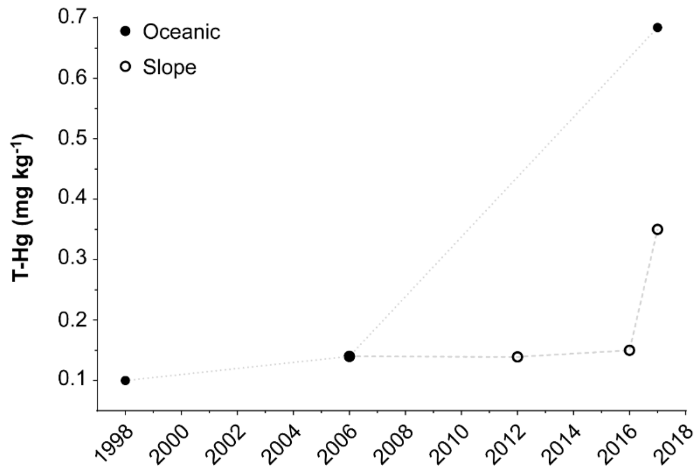
Mercury levels in Dumont D'Urville Sea slope were most similar to those from the Amundsen Sea slope. Adults from the Dumont D'Urville site were, on average, smaller and lighter than those from the Amundsen Sea seamount (Table 1). This might indicate that the Dumont D'Urville individuals belong to a group of fish that returned to the slope to improve their body condition after reproducing in oceanic waters, or that they have not yet migrated to northern waters (Hanchet et al., 2008).

4.2 Mercury in *D. mawsoni* over the last 2 decades and possible environmental implications

As a top predator with circumpolar distribution but relatively restricted postlarval ambit (Duhamel et al., 2014; Hanchet et al., 2015; Pinkerton et al., 2014), *D. mawsoni* is a potentially useful indicator of environmental mercury levels in the Southern Ocean. Conversely, mercury is an environmental marker which might provide insights into *D. mawsoni* ecology including migration and feeding relationships. Furthermore, regular commercial fishing in different Southern Ocean regions (CCAMLR, 2018) provides a way to monitor multiple areas. Although this fishery began in 1997, mercury levels have so far only been assessed for five seasons (1998, 2006, 2012, 2016 and 2017) (CCAMLR, 2018; Hanchet et al., 2012; Son et al., 2014; Yoon et al., 2018).

Fish from each of our three study sites had higher mean mercury levels than those found in any previous study. Two of the sites (Amundsen Sea seamount and Dumont D'Urville Sea slope) also had higher maximum values than those found in any previous study. This suggests an increase in mean mercury levels for *D. mawsoni* in the southwest Pacific sector from 0.10 mg kg^{-1} to 0.50 mg kg^{-1} since 1998. After accounting for other influences, Hanchet et al. (2012) reported a 20% increase in mercury levels between 1998 and 2006 (c. 0.015 mg kg^{-1} yr^{-1}) in *D. mawsoni* flesh. Our results suggest a much greater increase between 2006 and 2017 (316%, ~0.045 mg kg^{-1} yr^{-1}) (Figure 3). Other studies conducted between these years are more consistent with earlier results, reporting mean mercury levels below 0.25 mg kg^{-1} in the southwest Pacific sector and elsewhere in the Southern Ocean (Son et al. 2014, Yoon et al. 2018). We calculated the size of fish using allometric equations, while previous studies used measurements of whole fish (Hanchet et al., 2012; Son et al., 2014; Yoon et al., 2018). This may have contributed to the lower mean size of fish in our study. However, the scale of the difference between studies suggests there has been a real and widespread increase in mercury levels.

Figure 3. Mean T-Hg concentrations in *D. mawsoni* over the last 2 decades in oceanic (solid line, include all offshore habitats but mainly seamounts) and slope (dashed line) environments. Values are from previous studies: 1998 and 2006 (Hanchet et al., 2012); 2012 (Son et al., 2014); 2016 (Yoon et al., 2018).



The contrast between our reults and those of previous studies suggests a recent contamination event which affected much of the southwest Pacific sector. Differences between study sites, which cannot be explained entirely by the demography of the *D mawsoni* population, indicate spatial differences in the level of contamination. The diet of *D. mawsoni* is reasonably consistent between years, consisting mainly of fish and cephalopods (Hanchet et al., 2015). This fact, coupled with the large spatial scale of the apparent recent increase in mercury levels, suggests that the change reflects an increase in mercury in the environment rather than a trophic change. However, a recent study of the Scotia Sea, on the opposite side of Antarctica, shows a decreasing trend in mercury levels in cephalopods over the last decade (Seco et al., 2020). This disparity indicates regional differences in mercury level trends across the Southern Ocean.

There are several processes which may contribute to such an increase: Firstly, mercury can be released into the ocean by melting ice (Gionfriddo et al., 2016). The Amundsen and Dumont D'Urville seas have experienced contrasting changes in seasonal sea ice over recent decades with the former losing and the latter gaining sea ice cover (Stammerjohn et al., 2012). These contrasts suggest that a widespread increase in mercury is not easily explained by changes in sea ice alone. Secondly, oceanic and atmospheric currents transport pollutants to the Southern Ocean, including from industrializing regions where increases in mercury emissions are ongoing (Krabbenhoft and Sunderland, 2013; Mason et al., 2012; Streets et al., 2019). Thirdly, mercury is released through geothermal activity, including hydrothermal vents (Mason et al., 2012). An intense contamination episode, such as a geothermal event, may be necessary to explain the sudden elevation of mercury levels relative to previous studies. Such events have been observed elsewhere in the Southern Ocean (Rogers et al., 2012), and a recent event on the Amundsen Sea seamount is plausible. Most likely, the increase in mercury levels in *D. mawsoni* observed over time results from a combination of these processes. Further monitoring in the region will be necessary to disentangle the possible effects of episodic contamination from long-term trends. Indeed, regular monitoring of mercury levels in *D. mawsoni* is recommended due to the potential effects of mercury on humans.

Our study adds to previous evidence from *D mawsoni* to suggest an increase in mercury levels in the Southern Ocean over recent decades. High mercury levels have already been found in other Southern Ocean fauna (Bustamante et al., 2016; Carravieri et al., 2016). Due to its toxicity (Wolfe et al., 1998), mercury is a potential stressor for these animals which could exacerbate the ecological impacts of climate change (Rintoul et al., 2018; Stern et al., 2012). Indeed, climate change might itself be a factor in the increase in mercury levels. This is further evidence of change in the Southern Ocean ecosystem and therefore the need for conservation of Antarctic fauna (Chown et al., 2012; Croxall, 1987; Croxall et al., 2002; Kerry and Riddle, 2009). Unfortunately, no environmental data was available to the sampling sites, but we advise that future studies try to incorporate more abiotic and biotic information on the models to unravel which factors might have higher influence in the mercury levels of *D. mawsoni* rather than just is location. Variation in *D. mawsoni* mercury levels between habitats and years suggest that this species might be a useful indicator of levels in the ecosystem, especially because this species is fished all around the continent under an observation program that covers 100% of the fleet, thus facilitating the collection of samples to perform such monitoring, and mercury might be a useful tracer of habitat use in *D. mawsoni*.

5 Conclusions

This study shows that mercury levels in the ecologically and economically important Antarctic toothfish, *D. mawsoni*, can exceed the 0.5 mg kg^{-1} food safety threshold. The results also confirm that mercury concentrations in *D. mawsoni* are spatially variable, with fish from the Amundsen Sea seamount having higher levels than those from Amundsen and Dumont D'Urville Seas slopes. They also showed that mercury levels in *D. mawsoni* have increased over recent decades with the greatest increase happening since 2006, in both oceanic and slope areas.

We suggest that mercury levels in *D. mawsoni* should be monitored regularly and that food safety agencies should recognize a higher limit of 1.0 mg kg^{-1} for *D. mawsoni*, as for swordfish, sharks, and other predatory species. As higher levels were observed in a specific area of the Southern Ocean, we advise continuous monitoring here to disentangle the effects of possible episodic contamination events from long-term changes. We also suggest that *D. mawsoni* might be a useful indicator of mercury levels in seabed habitats which are not well represented by samples from the upper pelagic layers.

6 Acknowledgements

The authors thank the crew of the fishing vessel for the help during field work. We would also like to thanks to NHK Japan by the financial support. JQ was supported by FCT/MCTES through national funds (PIDDAC) and Portuguese Polar Program (PROPOLAR). JQ was supported by a travel grant of SCAR Ant-ERA. We also acknowledge FCT-Portugal for supporting JS through a PhD grant (SRFH/PD/BD113487). JPC is funded by CESAM (UID/AMB/50017/2019) and the Integrated Program of SR&TD 'Smart Valorization of Endogenous Marine Biological Resources Under a Changing Climate' (Centro-01-0145-FEDER-000018), co-funded by Centro 2020 program, Portugal 2020 and the European Regional Development Fund. Authors would like to thanks to the reviewers which, with their great comments and advices, improved the quality of the manuscript

7 References

Ainley, D. G., Brooks, C. M., Eastman, J. T., Massaro, M., Unnatural selection of Antarctic toothfish in the Ross Sea, Antarctica. In: H. F., (Ed.), Protection of the Three Poles. Springer, Tokyo, 2012, pp. 53-75.

Amos, H. M., Jacob, D. J., Streets, D. G., Sunderland, E. M., 2013. Legacy impacts of all-time anthropogenic emissions on the global mercury cycle. Global Biogeochemical Cycles. 27**,** 410-421, 10.1002/gbc.20040.

Aston, S. R., Fowler, S. W., 1985. Mercury in the open Mediterranean: evidence of contamination? Science of the Total Environment. 43**,** 13-26, 10.1016/0048-9697(85)90028-2.

Baird, R. J., 2006. Aspects of Illegal, Unreported and Unregulated Fishing in the Southern Ocean. Springer, Dordrecht, Netherlands.

Barton, K., 2019. MuMIn: Multi-model inference. R package version 1.43.15.

Bates, D., Maechler, M., Bolker, B., Walker, S., 2015. Fitting linear mixed-effects models using lme4. Journal of Statistical Software. 67**,** 1-48, 10.18637/jss.v067.i01.

Becker, P. H., Goutner, V., Ryan, P. G., González-Solis, J., 2016. Feather mercury concentrations in Southern Ocean seabirds: Variation by species, site and time. Environmental Pollution. 216**,** 253-263, 10.1016/j.envpol.2016.05.061.

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, 10.1371/journal.pone.0054508.t001.

Brooks, C. M., Andrews, A. H., Ashford, J. R., Ramanna, N., Jones, C. D., Lundstrom, C. C., Cailliet, G. M., 2011. Age estimation and lead–radium dating of Antarctic toothfish (*Dissostichus mawsoni*) in the Ross Sea. Polar Biology. 34**,** 329-338, 10.1007/s00300-010-0883-z.

Brooks, C. M., Crowder, L. B., Curran, L. M., Dunbar, R. B., Ainley, D. G., Dodds, K. J., Gjerde, K. M., Sumaila, U. R., 2016. Science-based management in decline in the Southern Ocean. Science. 354, 185-187, 10.1126/science.aah4119.

Bustamante, P., Carravieri, A., Goutte, A., Barbraud, C., Delord, K., Chastel, O., Weimerskirch, H., Cherel, Y., 2016. High feather mercury concentrations in the wandering albatross are related to sex, breeding status and trophic ecology with no demographic consequences. Environmental research. 144**,** 1-10, 10.1016/j.envres.2015.10.024.

Carravieri, A., Cherel, Y., Jaeger, A., Churlaud, C., Bustamante, P., 2016. Penguins as bioindicators of mercury contamination in the southern Indian Ocean: geographical and temporal trends. Environmental Pollution. 213**,** 195-205, 10.1016/j.envpol.2016.02.010.

CCAMLR, 2016a. General measures for exploratory fisheries for *Dissostichus* spp. in the Convention Area in the 2016/17 season. Conservation Measure 41-01.

CCAMLR, Non-Contracting Party IUU Vessel List. Vol. 2018, 2016b.

CCAMLR, List of authorised vessels. Vol. 2018. CCAMLR, 2017.

CCAMLR, Toothfish fisheries. Vol. 2018, 2018.

CCAMLR, Convention Area. In: CCAMLR, (Ed.), Vol. 2020. CCAMLR, 2020.

Chown, S. L., Brooks, C. M., Terauds, A., Le Bohec, C., van Klaveren-Impagliazzo, C., Whittington, J. D., Butchart, S. H., Coetzee, B. W., Collen, B., Convey, P., Gaston, K. J., Gilbert, N., Gill, M., Hoft, R., Johnston, S., Kennicutt, M. C., 2nd, Kriesell, H. J., Le Maho, Y., Lynch, H. J., Palomares, M., Puig-Marco, R., Stoett,

P., McGeoch, M. A., 2017. Antarctica and the strategic plan for biodiversity. PLoS Biology. 15, e2001656, 10.1371/journal.pbio.2001656.
Chown, S. L., Lee, J. E., Hughes, K. A., Barnes, J., Barrett, P. J., Bergsrom, D. M., Convey, P., Cowan, D. A., Crosbie, K., Dyer, G., Frenot, Y., Grant, S. M., Herr, D., Kennicutt, M. C., Lamers, M., Murray, A., Possingham, H. P., Reid, K., Riddle, M. J., Ryan, P. G., Sanson, L., Shaw, J. D., Sparrow, M. D., Summerhayes, C., Terauds, A., Wall, D. H., 2012. Challenges to the future conservation of the Antarctic. Science. 337, 158-159, 10.1126/science.1222821
Corsolini, S., Ademollo, N., Martellini, T., Randazzo, D., Vacchi, M., Cincinelli, A., 2017. Legacy persistent organic pollutants including PBDEs in the trophic web of the Ross Sea (Antarctica). Chemosphere. 185, 699-708, 10.1016/j.chemosphere.2017.07.054.
Cossa, D., Heimbürger, L. E., Lannuzel, D., Rintoul, S. R., Butler, E. C. V., Bowie, A. R., Averty, B., Watson, R. J., Remenyi, T., 2011. Mercury in the Southern Ocean. Geochimica et Cosmochimica Acta. 75, 4037-4052, 10.1016/j.gca.2011.05.001.
Costley, C. T., Mossop, K. F., Dean, J. R., Garden, L. M., Marshall, J., Carroll, J., 2000. Determination of mercury in environmental and biological samples using pyrolysis atomic absorption spectrometry with gold amalgamation. Analytica Chimica Acta. 405, 179-183, 10.1016/S0003-2670(99)00742-4.
Croxall, J. P., 1987. The status and conservation of Antarctic seals and seabirds: a review. Environment International. 13, 55-70, 10.1016/0160-4120(87)90044-4.
Croxall, J. P., Trathan, P. N., Murphy, E. J., 2002. Environmental change and Antarctic seabird populations. Science. 297, 1510-1514, 10.1126/science.1071987.
Duhamel, G., Hulley, P. A., Causse, R., Koubbi, P., Vacchi, M., Pruvost, P., Vigetta, S., Irisson, J., Mormede, S., Belchier, M., Dettai, A., Detrich, H. W., Gutt, J., Jones, C. D., Kock, K. H., Lopez Abellan, L. J., Van de Putte, A. P., Biogeographic patterns of fish. In: C. De Broyer, et al., Eds.), Biogeographc atlas of the Southern Ocean. The Scientific Committee on Antarctic Research, Cambridge, UK, 2014, pp. 328-498.
EC, E. C., Commission regulamentation (EC) No 1881/2006. Setting maximum levels for certain contaminants in foodstuffs. 2006.
EPA, Mercury update: impact on fish advisories. In: O. o. Water, (Ed.). Environmental Protection Agency, Washington, United States, 2001.
Fenaughty, J. M., 2008. The autoline system - an updated descriptive review of the method with recommendations to clarify CCAMLR conservation measures regulating longline fisheries within the convention area. Document WG-FSA-08/60. CCAMLR, Hobart, Australia. 27,
Fenaughty, J. M., Stevens, D. W., Hanchet, S. M., 2003. Diet of Antarctic toothfish (*Dissostichus mawsoni*) from the Ross Sea, Antarctica (subarea 88.1). CCAMLR Science. 10, 113-123,
Fontaine, M., Carravieri, A., Simon-Bouhet, B., Bustamante, P., Gasco, N., Bailleul, F., Guinet, C., Cherel, Y., 2015. Ecological tracers and at-sea observations document the foraging ecology of southern long-finned pilot whales (*Globicephala melas edwardii*) in Kerguelen waters. Marine Biology. 162, 207-219, 10.1007/s00227-014-2587-3.
Fox, J., Weisberg, S., 2019. An R Companion to Applied Regression. Sage, Thousand Oaks, California.
FSANZ, F. S. A. a. N. Z., Mercury in fish. 2004.

Gewurtz, S. B., Bhavsar, S. P., Fletcher, R., 2011. Influence of fish size and sex on mercury/PCB concentration: importance for fish consumption advisories. Environment International. 37**,** 425-434, 10.1016/j.envint.2010.11.005.

Gionfriddo, C. M., Tate, M. T., Wick, R. R., Schultz, M. B., Zemla, A., Thelen, M. P., Schofield, R., Krabbenhoft, D. P., Holt, K. E., Moreau, J. W., 2016. Microbial mercury methylation in Antarctic sea ice. Nature Microbiology. 1**,** 16127, 10.1038/nmicrobiol.2016.127.

Grilly, E., Reid, K., Lenel, S., Jabour, J., 2015. The price of fish: A global trade analysis of Patagonian (*Dissostichus eleginoides*) and Antarctic toothfish (*Dissostichus mawsoni*). Marine Policy. 60**,** 186-196, 10.1016/j.marpol.2015.06.006.

Guynn, K. D., Peterson, M. S., 2008. Mercury concentrations in the Patagonian toothfish, *Dissostichus eleginoides* Smitt 1898, among three distinct stocks. Polar Biology. 31**,** 269-274, 10.1007/s00300-007-0354-3.

Hanchet, S., Dunn, A., Parker, S., Horn, P., Stevens, D., Mormede, S., 2015. The Antarctic toothfish (*Dissostichus mawsoni*): biology, ecology, and life history in the Ross Sea region. Hydrobiologia. 761**,** 397-414, 10.1007/s10750-015-2435-6.

Hanchet, S. M., Rickard, G. J., Fenaughty, J. M., Dunn, A., Williams, M. J. H., 2008. A hypothetical life cycle for Antarctic toothfish (*Dissostichus mawsoni*) in the Ross Sea region. CCAMLR Science. 15**,** 35-53,

Hanchet, S. M., Tracey, D., Dunn, A., Horn, P., Smith, N., 2012. Mercury concentrations of two toothfish and three of its prey species from the Pacific sector of the Antarctic. Antarctic Science. 24**,** 34-42, 10.1017/s0954102011000654.

Horn, P. L., Sutton, C. P., DeVries, A. L., 2003. Evidence to support the annual formation of growth zones in otoliths of Antarctic toothfish (*Dissostichus mawsoni*). CCAMLR Science. 10**,** 125-138,

Jakimska, A., Konieczka, P., Skóra, K., Namiesnik, J., 2011. Bioaccumulation of metals in tissues of marine animals, part I: the role and impact of heavy metals on organisms. Polish Journal of Environmental Studies. 20**,** 1117-1125,

Karagas, M. R., Choi, A. L., Oken, E., Horvat, M., Schoeny, R., Kamai, E., Cowell, W., Grandjean, P., Korrick, S., 2012. Evidence on the human health effects of low-level methylmercury exposure. Environmental Health Perspectives. 120**,** 799-806, 10.1289/ehp.1104494.

Kerry, K. R., Riddle, M., 2009. Health of Antarctic Wildlife. Springer-Verlag Berlin Heidelberg.

Krabbenhoft, D. P., Sunderland, E. M., 2013. Global change and mercury. Science. 341**,** 1457-1458, 10.1126/science.1242838.

Mason, R. P., Choi, A. L., Fitzgerald, W. F., Hammerschmidt, C. R., Lamborg, C. H., Soerensen, A. L., Sunderland, E. M., 2012. Mercury biogeochemical cycling in the ocean and policy implications. Environmental Research. 119**,** 101-17, 10.1016/j.envres.2012.03.013.

Mason, R. P., Reinfelder, J. R., Morel, F. M. M., 1996. Uptake, Toxicity, and Trophic Transfer of Mercury in a Coastal Diatom. Environmental Science & Technology. 30**,** 1835-1845, 10.1021/es950373d.

McArthur, T., Butler, E. C. V., Jackson, G. D., 2003. Mercury in the marine food chain in the Southern Ocean at Macquarie Island: an analysis of a top predator, Patagonian toothfish (*Dissostichus eleginoides*) and a mid-trophic species, the warty squid (*Moroteuthis ingens*). Polar Biology. 27**,** 1-5, 10.1007/s00300-003-0560-6.

Méndez, E., Giudice, H., Pereira, A., Inocente, G., Medina, D., 2001. Preliminary Report on the Total Mercury Content of Patagonian Toothfish (*Dissostichos eleginoides*). Journal of Food Composition and Analysis. 14, 547-549, 10.1006/jfca.2001.1006.

Pinkerton, M. H., Bury, S. J., Brown, J. C. S., Forman, J., Kilimnik, A., 2014. Stable isotope analysis of tissue samples to investigate trophic linkages of Antarctic toothfish (*Dissostichus mawsoni*) in the Ross and Amundsen Sea regions. document WG-EMM-14/50. CCAMLR, Hobart, Australia. 30p,

R core team, R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria, 2019.

Rintoul, S. R., Chown, S. L., DeConto, R. M., England, M. H., Fricker, H. A., Masson-Delmotte, V., Naish, T. R., Siegert, M. J., Xavier, J. C., 2018. Choosing the future of Antarctica. Nature. 558, 233-241, 10.1038/s41586-018-0173-4.

Rogers, A. D., Tyler, P. A., Connelly, D. P., Copley, J. T., James, R., Larter, R. D., Linse, K., Mills, R. A., Garabato, A. N., Pancost, R. D., Pearce, D. A., Polunin, N. V., German, C. R., Shank, T., Boersch-Supan, P. H., Alker, B. J., Aquilina, A., Bennett, S. A., Clarke, A., Dinley, R. J., Graham, A. G., Green, D. R., Hawkes, J. A., Hepburn, L., Hilario, A., Huvenne, V. A., Marsh, L., Ramirez-Llodra, E., Reid, W. D., Roterman, C. N., Sweeting, C. J., Thatje, S., Zwirglmaier, K., 2012. The discovery of new deep-sea hydrothermal vent communities in the southern ocean and implications for biogeography. PLoS Biology. 10, e1001234, 10.1371/journal.pbio.1001234.

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., Pereira, E., 2020. Mercury levels in Southern Ocean squid: Variability over the last decade. Chemosphere. 239, 124785, 10.1016/j.chemosphere.2019.124785.

Seco, J., Xavier, J. C., Coelho, J. P., Pereira, B., Tarling, G., Pardal, M. A., Bustamante, P., Stowasser, G., Brierley, A. S., Pereira, M. E., 2019. Spatial variability in total and organic mercury levels in Antarctic krill *Euphausia superba* across the Scotia Sea. Environmental Pollution. 247, 332-339, 10.1016/j.envpol.2019.01.031.

Son, K. T., Kwon, J. Y., Jo, M. R., Yoon, M., Song, K. C., Choi, W. S., Yeon, I. J., Kim, J. H., Lee, T. S., 2014. Total Mercury Contents of Antarctic Toothfish *Dissostichus mawsoni* Caught in the Antarctic Sea. Fisheries and Aquatic Sciences. 17, 427-431, 10.5657/fas.2014.0427.

Stammerjohn, S., Massom, R., Rind, D., Martinson, D., 2012. Regions of rapid sea ice change: An inter-hemispheric seasonal comparison. Geophysical Research Letters. 39, L06501, 10.1029/2012gl050874.

Stern, G. A., Macdonald, R. W., Outridge, P. M., Wilson, S., Chételat, J., Cole, A., Hintelmann, H., Loseto, L. L., Steffen, A., Wang, F., Zdanowicz, C., 2012. How does climate change influence Arctic mercury? Science of the Total Environment. 414, 22-42, 10.1016/j.scitotenv.2011.10.039.

Stevens, D. W., Dunn, M. R., Pinkerton, M. H., Forman, J. S., 2014. Diet of Antarctic toothfish (*Dissostichus mawsoni*) from the continental slope and oceanic features of the Ross Sea region, Antarctica. Antarctic Science. 26, 502-512, 10.1017/s095410201300093x.

Streets, D. G., Devane, M. K., Lu, Z., Bond, T. C., Sunderland, E. M., Jacob, D. J., 2011. All-time releases of mercury to the atmosphere from human activities. Environmental Science & Technology. 45, 10485-10491, 10.1021/es202765m.

Streets, D. G., Horowitz, H. M., Lu, Z., Levin, L., Thackray, C. P., Sunderland, E. M., 2019. Global and regional trends in mercury emissions and concentrations, 2010–

2015. Atmospheric Environment. 201, 417-427, 10.1016/j.atmosenv.2018.12.031.

Sunderland, E. M., Mason, R. P., 2007. Human impacts on open ocean mercury concentrations. Global Biogeochemical Cycles. 21, 10.1029/2006gb002876.

Sydeman, W. J., Poloczanska, E., Reed, T. E., Thompson, S. A., 2015. Climate change and marine vertebrates. Science. 350, 772,

Thomas, S. M., Melles, S., Bhavsar, S. P., 2018. Spatiotemporal variations in mercury bioaccumulation at fine and broad scales for two freshwater sport fishes. Water. 10, 1625, 10.3390/w10111625.

Williams, R., McEldowney, A., 1990. A guide to the fish otoliths from waters off the Australian Antarctic Territory, Heard and Macquarie Islands. Australian Antarctic Division, Australia.

Wolfe, M. E., Schwarzbach, S., Sulaiman, R. A., 1998. Effects of mercury on wildlife: a comprehensive review. Environmental Toxicology and Chemistry. 17, 146-160, 10.1002/etc.5620170203.

Yoon, M., Jo, M. R., Kim, P. H., Choi, W. S., Kang, S. I., Choi, S. G., Lee, J. H., Lee, H. C., Son, K. T., Mok, J. S., 2018. Total and Methyl Mercury Concentrations in Antarctic Toothfish (*Dissostichus mawsoni*): Health Risk Assessment. Bulletin of Environmental Contamination and Toxicology. 100, 748-753, 10.1007/s00128-018-2326-4.

Figures

Figure 1. Sampling areas. a) Fishing area 88.2H – Amundsen Sea seamount; b) Fishing area 88.2F – Amundsen Sea slope; c) Fishing area 58.4.1G - Dumont D'Urville Sea slope.

Figure 2. T-Hg concentrations in *Dissostichus mawsoni* in the three fishing areas. Bar ± Error Bar - Mean ± SD; Points - individual values; Dashed line - 0.5 mg kg^{-1}.

Figure 3. Mean T-Hg concentrations in *D. mawsoni* over the last 2 decades in oceanic (solid line, include all offshore habitats but mainly seamounts) and slope (dashed line) environments. Values are from previous studies: 1998 and 2006 (Hanchet et al., 2012); 2012 (Son et al., 2014); 2016 (Yoon et al., 2018).

Tables

Table 1: Fish biometrics and T-Hg concentrations in the three sampled areas. SSRU – small scale research unit; SL – standard length; M – mass; T-Hg – total mercury concentration. Values are mean ± standard deviation. Values with different superscript letters in the same column are significantly different (p <0.05).

Table 2: GLMM analysis final model. This is a random-intercept model with age and geomorphology as fixed-effect variables and fishing longlines as the random-intercept grouping variable. Significant explanatory variables for T-Hg in *Dissostichus mawsoni*. SE: Standard Error; df: degrees of freedom. R^2m: marginal pseudo-R^2 (variability explained by fixed effects); R^2c: conditional pseudo-R^2 (variability explained by fixed and random effects)

Supplementary Material

Annex 1. Collinearity analysis of both biological and habitat candidate variables using VIF (Variation Inflexion Factor) analysis. From the 1^{st} to the 2^{nd} column, mass (biological variable) and steepness (habitat variable) were deleted. The last column (grey shading) shows the results of post hoc VIF analysis applied to the final model, indicating that moderate correlation between the two explanatory variables does not adversely affect model results.

Annex 2. GLMMs performed, p-value and AIC. Grey shadow shows the final model.

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	1st	2nd	Final Model
Biological Variables			
SL	18.0	3.6	
Mass	14.4		
Age	3.6	3.6	2.0
Habitat variables			
Steepness	6.8		
Depth	3.3	1.1	
Geomorph	6.6	1.1	2.0

Annex 2. GLMMs performed, *p*-value and AIC. Grey shadow shows the final model.

Generalized Linear Mixed Models (GLMM)		
Models	***p*-value**	**AIC**
T-Hg ~ SL + age + depth + geomorphology + (1|longline)	intercept = 0.90 SL = 0.70 age = 0.12 depth = 1.00 geomor = 0.02	26.19
T-Hg ~ SL + age + geomorphology + (1|longline)	intercept = 0.87 SL = 0.67 age = 0.08 geomor = 0.02	9.48
T-Hg ~ age + geomorphology + (1|longline)	intercept < 0.01 age = 0.04 geomor = 0.02	8.64