



Fingerprint of persistent organic pollutants in tissues of Antarctic notothenioid fish



Nerina B. Lana^a, Paula Berton^{a,b}, Adrian Covaci^c, Néstor F. Ciocco^{b,d}, Esteban Barrera-Oro^{e,f}, Adrián Atencio^g, Jorgelina C. Altamirano^{a,b,*}

^a Laboratorio de Química Ambiental, Instituto Argentino de Nivología, Glaciología y Ciencias Ambientales (IANIGLA)-CONICET, Mendoza, P.O. Box 131 ZC5500, Mendoza, Argentina

^b Facultad de Ciencias Exactas y Naturales, Universidad Nacional de Cuyo, Mendoza, Argentina

^c Toxicological Center, Department of Pharmaceutical Sciences, University of Antwerp, Universiteitsplein 1, 2610 Wilrijk, Belgium

^d Instituto Argentino de Investigación de Zonas Áridas (IADIZA)-CONICET, Mendoza, Argentina

^e Instituto Antártico Argentino (IAA), Buenos Aires, Argentina

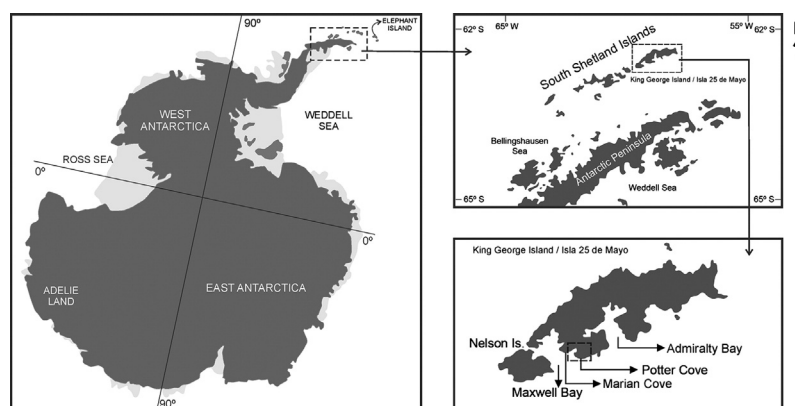
^f Museo Argentino de Ciencias Naturales Bernardino Rivadavia-CONICET, Buenos Aires, Argentina

^g Laboratorio de Estratigrafía Glaciar y Geoquímica del Agua y la Nieve (LEGAN)-IAA-CONICET, Mendoza, Argentina

HIGHLIGHTS

- POPs levels and tissue distribution in Antarctic notothenioid fish
- *Trematomus newnesi*, *Notothenia coriiceps* and *Notothenia rossii* analyzed for POPs
- This is the first report on POPs levels in the Antarctic notothenioid *Trematomus newnesi*.
- Gonads and gills of analyzed specimens presented the highest levels of studied POPs.

GRAPHICAL ABSTRACT



ARTICLE INFO

Article history:

Received 1 April 2014

Received in revised form 8 August 2014

Accepted 10 August 2014

Available online xxx

Editor: Eddy Y. Zeng

Keywords:

Antarctica

POPs

Tissue distribution

South Shetland Islands

Notothenioids

ABSTRACT

In the present work, persistent organic pollutants (POPs), such as polychlorinated biphenyls (PCBs), dichlorodiphenyltrichloroethane (DDT) and metabolites, polybrominated diphenyl ethers (PBDEs), and hexachlorocyclohexane (HCH) were analyzed in three Antarctic notothenioid fish species: *Trematomus newnesi* (TRN), *Notothenia coriiceps* (NOC) and *Notothenia rossii* (NOR). The contribution of each POP-family to the total load was as follows: Σ PCB (40%) > Σ DDT (27%) > Σ PBDEs (23%) > Σ HCH (10%). Among the 23 PCB congeners analyzed, penta-CBs homologues were the prevalent group, followed by hexa-CBs and hepta-CBs. DDT and its metabolites presented the following trend: p,p' -DDT > p,p' -DDE ~ p,p' -DDD. PBDE profile was dominated by BDE-47 and BDE-99 congeners, followed by BDE-100 > BDE-28 > BDE-154, BDE-153. Among HCHs, the γ -HCH isomer was detected in all samples, constituting 69% total HCH load, while α -HCH and β -HCH contributions were 15% and 16%, respectively.

The levels of POPs reported here suggest that NOR and NOC are more susceptible to accumulate the analyzed contaminants than TRN, a species not previously analyzed for POPs.

* Corresponding author.

E-mail address: jaltamirano@mendoza-conicet.gob.ar (J.C. Altamirano).

Distribution of POPs among different tissues of the three species (muscle, liver, gonads, and gills) was also investigated. Considering lipid weight, the general pattern of POPs distribution in tissues indicated that while gonads showed higher levels of PCBs, DDTs and HCH, the most significant PBDE concentrations were recorded in gills. Also, a comparative analysis of POPs concentration in fish samples from Antarctic area was included.

© 2014 Elsevier B.V. All rights reserved.

1. Introduction

Although the human presence in Antarctica is low, with relatively little impact of wastewater and solid waste, the anthropogenic effect on the ecosystem has increased progressively, mainly through the commercial fishery of living resources such as finfish and krill (Ainley and Pauly, 2014; Kock, 1992). Contamination with persistent organic pollutants (POPs) was documented in the region since 1960s (Sladen et al., 1966; Tatton and Ruzicka, 1967). Due to their physicochemical properties and low decomposition rate, POPs like polychlorinated biphenyls (PCBs), polybrominated diphenyl ethers (PBDEs), hexachlorocyclohexane (HCH) and dichlorodiphenyltrichloroethane (DDT) are transported over long distances and/or widely dispersed into the environment after released. Transport of POPs can be accomplished through atmospheric and/or water flows as a gas phase and/or associated to particulate matter. Previous reports demonstrated that POPs associated with organic particles are transported by sedimentation from the pelagic zone to the sea-bed (Wania and Daly, 2002).

Cold condensation and global fractionation were proposed as the main mechanisms whereby POPs reach polar locations (Wania and Mackay, 1996). In this way, the more volatile POPs, such as HCH and low-PCBs reach polar regions in a higher rate than the less volatiles ones (highly halogenated PCBs, PBDEs and DDT) (Paasivirta et al., 1999). The combination of environmental conditions and physicochemical properties of POPs makes Antarctica to be a sink for such type of compounds (Wania and Mackay, 1996). Cold-adapted species present a slower metabolism, resulting in a slowdown of biological processes including growth and reproduction (Bargagli, 2005). This adaptation to cold environments can affect the fish's ability to detoxify or remove pollutants from its body which, added to the storage of lipids as an energy source, favor the bioaccumulation of hydrophobic chemicals during the Antarctic fish lifespan (Goutte et al., 2013). These factors also have significant influence on POPs biomagnification within the Antarctic food webs (Corsolini et al., 2006).

Antarctic fish constitute an important link of marine Antarctic food webs because they prey on a variety of benthic, epibenthic and planktonic organisms and are preyed by squids, other fish, penguins, flying sea birds, seals and whales (Barrera-Oro, 2002). Among fish, the suborder Notothenioidei is an endemic coastal demersal group, which includes six dominant families in terms of diversity (35%) and biomass (Kock and Kellermann, 1991). Based on their feeding strategies and their relevance to the marine environment under analysis (Barrera-Oro, 2003), three different species of the family Nototheniidae, *Trematomus newnesi* (TRN), *Notothenia coriiceps* (NOC) and *Notothenia rossii* (NOR), were selected for the present study. TRN is found in the permanent and seasonal packed-ice zones around Antarctica and adjacent islands. It is a benthos and plankton feeder, with benthic and benthopelagic habits (Eastman and Barrera Oro, 2010). NOC inhabits different areas of the same ichthyofaunistic subregion in the Atlantic Ocean sector, Southern Indian Ocean sector and High Antarctic Zone (Barrera-Oro, 2002). This fish species is euriphagous and changes its diet seasonally according to prey availability. It is a benthos feeder, with benthic and epibenthic habits. NOR inhabits the Scotia Arc, the western Antarctic Peninsula and circum-Antarctic waters of sub-Antarctic islands (Barrera-Oro, 2002). It is a benthos and plankton feeder and is characterized by offshore–inshore migrations in its life cycle. During its juvenile stage, NOR feeds on benthos, epibenthos, plankton and nekton (Casaux et al., 1990). It migrates then offshore to join the

adult population, and feeds primarily on krill (*Euphausia superba*) and fish (Barrera-Oro, 2002; Casaux et al., 1990).

It is known that POPs and metabolites present different degradation rates, as well as accumulation patterns among tissues, depending on their chemical structures and/or the metabolic system involved (Cipro et al., 2010; Ondarza et al., 2011; Tanabe et al., 1997). However, contaminants tissues distribution pattern not only are conditioned by the physicochemical properties of POPs and its major metabolites, but also by the biology and ecology of fish (Mormede and Davies, 2003; Storelli et al., 2009). Considering the feeding habits of the fish studied in this work, NOR and TRN species, both epibenthic and semipelagic water column feeders, it is possible to hypothesize that POPs and major metabolites should have a comparable pattern of accumulation in these two species. Furthermore, this accumulation pattern would likely be different in NOC, a mainly benthos feeder with a wide trophic spectrum. Although the number of NOC samples is low, our expected results could be taken as indicative.

In the present work the occurrence, distribution and isomeric profiles of target POPs, including PCBs, PBDEs, HCH, and DDT, and main metabolites, were investigated in three Antarctic notothenioid species: TRN, NOC and NOR. Additionally, distribution of the mentioned contaminants in tissues such as muscle, liver, gonads, and gills was evaluated to find target organs. This is the first report about these POPs groups and metabolites in TRN specimens.

2. Materials and methods

2.1. Reagents and materials

The following compounds were included in the analysis: 23 PCB congeners (*penta*-CB: 99, 101, 105, 118; *hexa*-CB: 128, 138, 146, 149, 151, 153, 156; *hepta*-CB: 170, 171, 174, 177, 180, 183, 187; *octa*-CB: 194, 195, 199; *nona*-CB: 206; *deca*-CB: 209), 7 PBDE congeners (nos: 28, 47, 99, 100, 153, 154, 183), HCH isomers (α -, β -, γ -), and DDT and metabolites (*p,p'*-DDE, *o,p'*-DDD, *p,p'*-DDD, *p,p'*-DDT). Abbreviations are expressed as follows: Σ PCB as the sum of the 23 congeners, Σ DDT as the sum of the 4 compounds, Σ PBDEs as the sum of the 7 congeners and Σ HCH as the sum of the 3 isomers.

Individual PCB, HCH and DDT standards were purchased from Dr. Ehrenstorfer Laboratories (Augsburg, Germany). PBDE standard mixtures were purchased from Wellington Laboratories (Guelph, Ontario, Canada). General reagents, such as acetone, n-hexane, dichloromethane (DCM), isooctane (all pesticide grade), and sulfuric acid (analytical grade) were purchased from Merck (Darmstadt, Germany). Silica gel 60 (63–230 mesh) and anhydrous Na₂SO₄ (Merck, Germany) were pre-washed with hexane aliquots and dried afterward. Before use, silica gel and Na₂SO₄ were heated at 150 °C for 24 h. Extraction thimbles were pre-extracted (1 h) with the solvent-extraction mixture used for the samples and dried at 100 °C for 1 h.

2.2. Collection, preservation of samples, and biometric determinations

Specimens of TRN (n = 21), NOC (n = 2), and NOR (n = 8) were collected during summer campaigns from year 2008 to 2011 at Potter Cove, King George Island/Isla 25 de Mayo, South Shetland Islands, close to the Scientific Station Carlini – formerly Jubany Station – (62° 14' S; 58° 40' W). Trammel nets (length 25, 35 and 50 m; width 1.5 m; inner mesh 2.5 cm; outer mesh 12 cm) were set for 6–96 h at

rocky, macroalgae beds at 5–50 m depths at three sites in the outer portion of the cove (Fig. 1). Each specimen was wrapped and kept in individual aluminum foil and taken to the laboratory where they were measured, weighed and stored at $-20\text{ }^{\circ}\text{C}$ until analysis. The length of the fish was measured from the front-tip of the mouth to the beginning of the caudal fin (standard length). Due to complexity in the logistic of sampling procedures, only in a few specimens it was possible to identify the sex on site before freezing. Details of sampling date and biometric data are presented in Table 1S.

2.3. Sample preparation

Specimens were defrosted and dissected before analysis. Muscle, liver, gonads and gills tissues were freeze-dried at $-55\text{ }^{\circ}\text{C}$ and 33 Pa until constant weight (ca. 72 h). The analytical methodology used is described in Covaci et al. (2006). Briefly, dried tissue aliquot of muscle ($\sim 2\text{ g}$), liver ($\sim 0.8\text{ g}$), gonads ($\sim 0.8\text{ g}$) or gills ($\sim 1\text{ g}$) was homogenized in an agate mortar, mixed with sodium sulfate, and spiked with internal standards (IS): 10 ng CB-143, 2 ng ϵ -HCH and 1 ng BDE-77. The homogenate was then Soxhlet-extracted with 100 mL n-hexane:acetone (3:1, v/v) for 2 h. An aliquot (ca. 1/10) of the resulting extract was used for the determination of lipid content by gravimetry (Roosens et al., 2008). The remaining extract was further cleaned up on $\sim 8\text{ g}$ acidified silica (H_2SO_4 44%, w/w) column; and analytes were eluted with 20 mL hexane and 15 mL DCM. The eluent was rotary evaporated to $\sim 2\text{ mL}$, further evaporated to incipient dryness under a gentle N_2 stream, and finally reconstituted with 150 μL isoctane.

2.4. Instrumental analysis

Detection and quantification of analytes were carried out by using an Agilent 6890-5973 GC-MS instrument (Agilent, USA) equipped with an electron capture negative ionization (ECNI) source, and a $30\text{ m} \times 0.25\text{ mm} \times 0.25\text{ }\mu\text{m}$ DB-5 capillary column (J&W Scientific, Folsom, USA). Ion source, quadrupole, and interface temperatures were set at 170, 150, and $300\text{ }^{\circ}\text{C}$, respectively. Helium was used as carrier gas at constant flow (1.0 mL min^{-1}), with methane as moderating gas. The

electron multiplier voltage was set at 2200 V. An aliquot of the extract ($1\text{ }\mu\text{L}$) was injected in solvent vent mode (vent time 1.25 min, vent flow 54.2 mL min^{-1} , splitless time 1.50 min; initial injector temperature at $92\text{ }^{\circ}\text{C}$, maintained for 0.03 min, then heated at $700\text{ }^{\circ}\text{C min}^{-1}$ to $300\text{ }^{\circ}\text{C}$ and maintained for 30 min). Temperature of the DB-5 column was programmed from $90\text{ }^{\circ}\text{C}$ (1.25 min) to $310\text{ }^{\circ}\text{C}$ at a rate of $10\text{ }^{\circ}\text{C min}^{-1}$ holding for 6.75 min. Bromine isotope ions (m/z 79 and 81) were acquired in selected ion monitoring (SIM) mode for the whole run time. For PCBs and pesticides, two most intense characteristic ions were monitored in specific time segments according to elution characteristics (Ali et al., 2013; Jaspers et al., 2013). Typical dwell times were 20–25 ms.

2.5. Quality assurance

A procedural blank was analyzed every ten samples. This was carried out for each type of tissue analyzed. Procedural blanks were consistent among them (relative standard deviations – RSD < 30%). These values were used for correcting compounds concentrations by subtraction of the average blank value. Method limits of quantification (LOQ) were fixed at 3°SD of the procedural blanks. For compounds not detectable in blanks, LOQs were calculated from a signal to noise ratio of 10 (Ael et al., 2012). LOQs for the analyzed POPs and main metabolites ranged from 0.005 to 3.50 ng g^{-1} dry weight (d.w.) for muscle, from 0.01 to 5.50 ng g^{-1} d.w. for liver, from 0.02 to 11.50 ng g^{-1} d.w. for gonads and from 0.01 to 3.20 ng g^{-1} for gills. Recovery of internal standards was above 70%. A standard reference material SRM 1945 (PCBs, DDT, PBDEs and HCHs, in whale blubber) was used to test the accuracy of the method. The results demonstrated good repeatability for individual congeners (RSD between 1 and 20%) and good agreement with the certified values. Recovery values were above of 89%, except for the γ -HCH and CB-197 with recoveries of 83 and 72%, respectively. More information is included in the Supplemental material, Table 2S.

2.6. Analysis of reported POPs levels in Antarctic fish species

The bibliographic analysis of POPs levels reported in fish species from the Antarctic continent was based on relative mean concentration

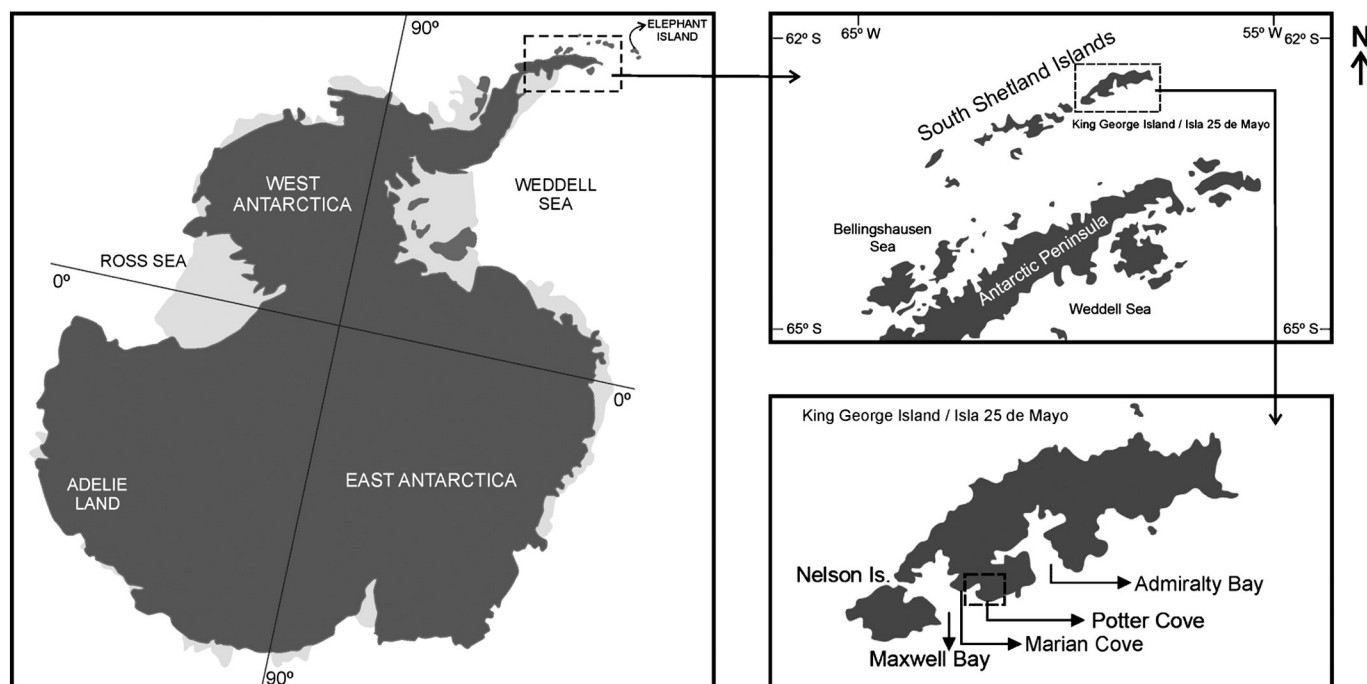


Fig. 1. The studied area in King George Island/Isla 25 de Mayo, South Shetland Islands, with indication of the sampling site, Potter Cove ($62^{\circ}14'\text{ S}$; $58^{\circ}40'\text{ W}$).

for a specific tissue type, a POP family and/or metabolite, and a fish species. It was calculated as follows: $RMC_s^{POP} = MC_s^{POP} / MC_{TRN}^{POP}$, where RMC is the relative mean concentration, MC is the mean concentration of the target POP family in a specific tissue of the fish species (represented as 's' in the equation). Reported data below LOD of the technique was not included in the analysis. TRN was arbitrarily chosen as referent (TRN) since this is the first time that this species is analyzed and reported in literature for POPs study. Additionally, the largest number of specimens analyzed in this work belonged to this species.

2.7. Statistical analyses

Statistical analyses were performed using the software SPSS 20.0 for Windows (SPSS Inc., Chicago, IL). Level of statistical significance was defined at $p < 0.05$. For calculations of sums and means, a value of $f * LOQ$ was assigned to concentrations of compounds $< LOQ$, where 'f' is the proportion of determinations with concentrations above the LOQ (or the detection frequency) (Covaci et al., 2008). Non-parametric statistics were used since parametric requirements, including data normality (Shapiro–Wilk), variances homogeneity (Levene test), and data transformations (\log_{10} , square root, etc.), were not satisfied. Comparison of POP concentrations among species and tissues were made using Kruskal–Wallis H test (KW) and Wilcoxon rank sum tests with post-hoc correction (Wxon).

3. Results and discussion

All analyzed samples contained detectable levels of the target POPs. POP concentrations were normalized against lipid content of the analyzed tissue. Results are summarized in Table 1.

The contribution of targets family to the total load was leaded by PCBs. Average concentration of ΣPCB was about 40% of the total load, followed by ΣDDT and $\Sigma PBDEs$ with 27% and 23%, respectively. ΣHCH represented only 10% of the total load (Fig. 1S, Supplementary material).

3.1. PCBs

PCB was the predominant group along the target POPs, with levels ranging from 11.1 to 99.0 $ng\ g^{-1}$ lipid weight (l.w.) (Table 1). Six of the analyzed 23 PCB congeners (CB-101, 105, 99, 118, 194, and 199), were detected in more than 50% of the samples, whereas CB-209 was detected in less than 20%. Our results showed that analyzed PCBs congeners had no particular accumulation pattern, which would be conditioned by biotic (age, gender, body condition, reproductive status, and diet), ecological (migratory habits, areas of occurrence, and depth) and environmental factors (temperature, contamination load) (Mormede and Davies, 2003; Storelli et al., 2009). There is evidence that PCB accumulation can be influenced by trophic level position of the species in high latitude environments. In this sense, higher chlorinated PCB (*penta* to *hepta*-CBs) are found relatively abundant in fish and seabird eggs (Corsolini et al., 2003; Goutte et al., 2013), while low-chlorinated PCBs are mainly reported in the lower trophic levels (Corsolini et al., 2003; Goutte et al., 2013). *Tri*- and *tetra*-CB congeners could not be analyzed due to poor sensitivity on the GC–ECNI/MS instrument and therefore, it is not possible to make any assumptions regarding these congeners. However, we are aware that *tri*- and *tetra*-CB congeners were reported to be present in biota, including fish specimens from Antarctic continent (Cipro et al., 2013).

The contribution of each PCB congeners family to the total load was as follows: *penta*-CBs presented the highest levels, followed by *hexa*-CBs and *hepta*-CBs (mean: 5.4 $ng\ g^{-1}$ l.w., 1.9 $ng\ g^{-1}$ l.w. and 0.6 $ng\ g^{-1}$ l.w., respectively; all species and tissues combined) (Fig. 2S, Supplementary material). Highly chlorinated PCBs (*octa*- to *deca*-CBs) presented the lowest levels in all tissues. The abundance of *penta*- and *hexa*-CBs was already reported in fish worldwide (Covaci

et al., 2006; Ondarza et al., 2011; Storelli et al., 2009). These two homologue PCB groups predominate in technical mixtures (UNEP, 2013) and present distinctive physicochemical properties, including low vapor pressure, high stability, and lipophilic character. Therefore, these two families of congeners are easily transported and accumulated in the environment, as well as in living organisms (Borghesi et al., 2008). Regarding *hepta*-CB homologues, those with chlorine atoms substituted in the 2,4,5 position (e.g. CB-170, 180, and 187) are the most resistant to fish metabolic degradation; and in turn are sparsely dispersed in long-distance atmospheric processes (Corsolini et al., 2003). This could explain the relatively high levels of these heavy congeners. Additionally, the presence of *hepta*- to *deca*-CBs was previously associated with local sources, like waste burning and dumping sites in the King George Island/Isla 25 de Mayo (Montone et al., 2003).

Among the studied species, NOR and NOC presented the highest $\Sigma PCBs$ levels ($\Sigma PCBs$ 239 and 183 $ng\ g^{-1}$ l.w., respectively; all tissues combined), whereas TRN presented values considerably lower ($\Sigma PCBs$ 122 $ng\ g^{-1}$ l.w.). These differences could be related to the diet of the species. TRN does not vary substantially its diet along the year, preying on some epibenthic organisms, but also on plankton components in the water column (Casaux et al., 1990). On the other hand, NOR and NOC prey mainly on benthic organisms and have a wider trophic spectrum (10 taxa) than TRN. Sediments found in stomach contents studies of NOR and NOC specimens from the same study area confirm their benthic habits (Casaux et al., 1990). This suggests that part of the PCBs intake by these fish species is through their food, which is then readily available and can accumulate in it.

Despite differences in total concentration, distribution of PCB congener among the three fish species was similar to the previously described pattern (*penta*-CBs > *hexa*-CBs > *hepta*-CBs; Fig. 2). Furthermore, no significant differences were found when homologue groups profile was compared among the three species (KW *penta*-CBs: $X^2 = 5.69$, $p = 0.05$; *hexa*-CBs: $X^2 = 5.01$, $p = 0.08$; *octa*-CBs: $X^2 = 4.69$, $p = 0.09$; *nona*-CBs: $X^2 = 4.96$, $p = 0.08$; *deca*-CBs: $X^2 = 0.43$, $p = 0.80$), except for *hepta*-CBs (KW $X^2 = 6.59$, $p = 0.03$). Concentrations differences were found between NOR and TRN (Wxon $W = 2573.5$, $p = 0.01$), with slightly higher concentration in the former species. Even though it is difficult to explain this difference, Storelli et al. (2009) suggested that the species have a selective metabolism for individual congeners and/or some congeners have higher biomagnification potentials, leading to selective enrichments in higher organisms of trophic web (Storelli et al., 2009).

Gonads presented the highest levels of $\Sigma PCBs$ (216 $ng\ g^{-1}$ l.w., all species combined), while gills, muscle, and liver tissue had similar levels (123, 97 and 108 $ng\ g^{-1}$ l.w., respectively; all species combined). Previous reports indicated that PCBs distributions among tissues may depend on differences of physiological characteristics among organisms (age, sex and dietary habits), as well as lipid content and composition (different ratios of triacylglycerols, phospholipids and cholesterol) in analyzed tissues. Lipids provide energy for swimming during migration and are transferred from muscle to gonads during reproduction. This remobilization may be the main cause of the high PCB levels in gonads (Corsolini et al., 2005).

Except for gonads (KW $X^2 = 2.49$, $p = 0.287$), median concentrations of PCB homologue groups in tissues significantly differed among species (KW muscle: $X^2 = 21.60$, $p = 0.00$; liver: $X^2 = 9.24$, $p = 0.01$; gills: $X^2 = 8.86$, $p = 0.01$; Fig. 2). Main differences were found between NOR and TRN tissues (Wxon muscle: $W = 153,171.5$, $p < 0.00$; liver: $W = 86,369$, $p < 0.00$; gills: $W = 73,648.5$, $p < 0.00$). Additionally, PCB levels were 1.7 times higher in NOR than in TRN (Table 1). CB-101 and -99 were the most abundant PCB congeners in gonads (19 and 8.1 $ng\ g^{-1}$ l.w., respectively) and gills (14 and 6.7 $ng\ g^{-1}$ l.w., respectively) of TRN. Lower concentrations of these congeners were found in muscle and liver. The highest levels of CB-153 and -118 were found in liver and gills of TRN (Table 1). Highly chlorinated PCBs (*hepta*- to *deca*-CBs) were mainly found in muscle tissue of

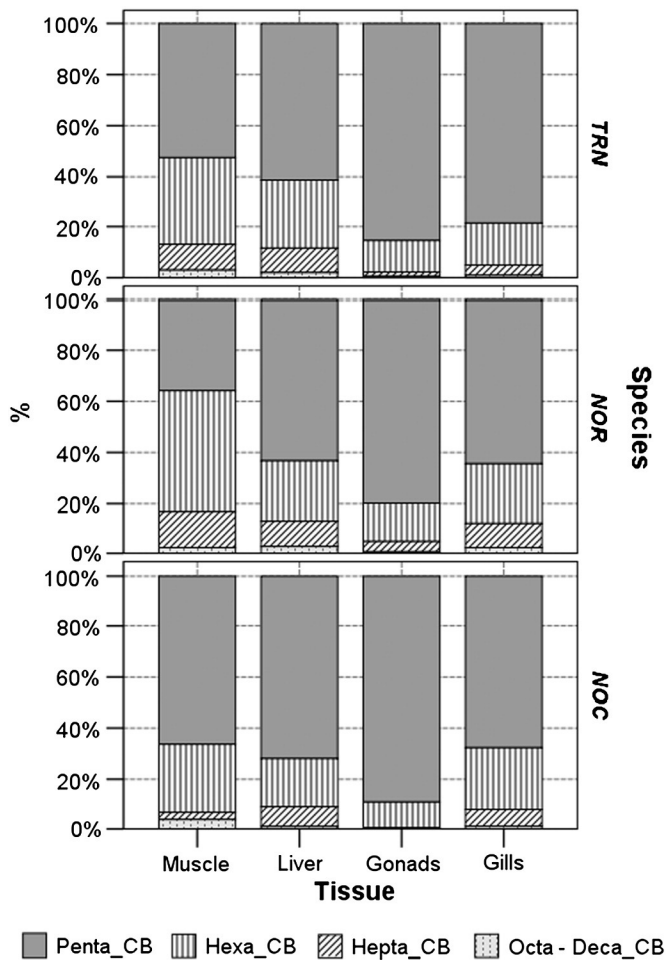


Fig. 2. Congeners composition (percentage) of PCBs in tissues in analyzed notothenioid species.

TRN, while these congeners were below to LOQ in liver, gonads and gills. Similar to TRN, NOR also showed CB-101 and -99 as the most abundant congeners, with levels in gonads 2.5 times higher than in gills, muscle and liver. Congeners among CB-187 to CB-209 were below to the LOQ in gonads. Remaining PCB congeners showed accumulation in muscle tissue, mainly; with CB-153 and -138 prevailing (Table 1). NOC tissues showed a similar trend to NOR and TRN, where CB-101 and -99 prevailed in gonads and gills. NOC muscle tissues presented the lowest levels of PCBs (Table 1). In muscle, lighter PCBs congeners (*penta-* to *hepta-*CBs) had higher concentrations than *octa-* to *deca-*CBs. The relative high concentration of low-chlorinated PCBs in gills and gonads tissues indicates that PCBs uptake is mainly from the water column (Ondarza et al., 2011) due to the lower hydrophobicity of these congeners ($\log Kow < 6.2$). Biomagnification process through the food chain may explain the high concentration of high-chlorinated PCBs in liver and muscle tissue (Corsolini et al., 2006).

3.2. DDT and metabolites

Within the term “total DDT” (ΣDDT), we included DDT (p,p' -DDT) and its metabolites (p,p' -DDE, o,p' -DDD, p,p' -DDD). This group was the second most predominant of the studied contaminants, with levels within the range 9.9–199 ng g⁻¹ l.w. (Table 1). The contribution of each compound to the total DDT load was as follows: p,p' -DDT > p,p' -DDE ~ p,p' -DDD (Fig. 3S, Supplementary material), whereas o,p' -DDD was not detected in any sample (Table 1). Fish biotransform p,p' -DDT into its most stable metabolite p,p' -DDE (Vives et al., 2005). According

to literature, p,p' -DDE represents 90–99.7% of ΣDDT s in biological samples, while o,p' -DDE is usually below LOQ (Storelli et al., 2009; Weijs et al., 2010). Consequently, the authors selected p,p' -DDE as the most representative metabolite to identify the pollution produced by DDTs congeners.

Among the studied species, the highest level of total DDT was found in NOR (254 ng g⁻¹ l.w.), while comparable DDT levels were found in NOC and TRN (60 and 54 ng g⁻¹ l.w., respectively). A comparison of median levels of DDT and DDT-metabolites among the studied species revealed no significant differences (KW o,p' -DDD: $X^2 = 0.00$, $p = 1.0$; p,p' -DDD: $X^2 = 3.52$, $p = 0.17$; p,p' -DDT: $X^2 = 3.71$, $p = 0.15$), except for p,p' -DDE (KW $X^2 = 17.02$, $p < 0.01$). The lowest p,p' -DDE level was found in TRN in relation to NOC (Wxon W = 2067, $p = 0.01$) and NOR (Wxon W = 2416, $p < 0.00$).

Gonads presented the highest ΣDDT levels (230 ng g⁻¹ l.w.); while concentrations in liver, gills and muscle tissues were comparable (ΣDDT 63, 41 and 34 ng g⁻¹ l.w., respectively). Median levels of ΣDDT in tissues (all species combined) were not significantly different (KW muscle: $X^2 = 0.89$, $p = 0.63$; gonads: $X^2 = 1.64$, $p = 0.44$; gills: $X^2 = 0.57$, $p = 0.74$), except for liver (KW $X^2 = 12.63$, $p = 0.002$). Differences on liver concentrations were observed between TRN and NOR (Wxon W = 1302.5, $p < 0.00$), with the lowest values found in TRN specimens.

The distribution of the DDT and metabolites among tissues slightly differed with the contribution fraction of each compound to the total load (previously discussed). The observed order was: p,p' -DDE > p,p' -DDT > p,p' -DDD, except in gonads and liver of NOR species, and in liver and muscle of TRN species, where p,p' -DDT concentrations were the highest (Fig. 3). Since fish can biotransform p,p' -DDT into p,p' -DDE (Vives et al., 2005), the p,p' -DDE/DDT ratio is often used as an indicator of the DDT input time. A high p,p' -DDE/DDT ratio (>0.6) indicates older input, while low values point toward fresh DDT input (Yogui et al., 2003). Furthermore, van den Brink et al. (2011) confirmed that p,p' -DDE levels, as well as PCBs, are decreasing in Antarctic pelagic organisms, while increasing in benthic organisms (van den Brink et al., 2011). Consequently, concentrations of hydrophobic organic contaminants in pelagic biota may indicate fresh input of contaminants in the environment, while the concentrations in benthic organisms are more related to the total (background) environmental burden in Antarctica. In the present study, p,p' -DDE/DDT ratios in different tissues were >0.6, except for NOR gonads (Fig. 3). These results suggest that DDT residues in Antarctic organisms derive from old DDT input. Additionally, the high values of the ratio here reported for demersal fish could be due to (secondary) exposure to high concentrations of p,p' -DDE congener (Barrera-Oro, 2002).

3.3. PBDEs

The third group of POPs is constituted by PBDEs. The $\Sigma PBDE$ levels ranged between 1.2 and 114 ng g⁻¹ l.w. (Table 1). Among the PBDE congeners, BDE-47, -99, -28, -154 and -100 were the most predominant in more than 70% the analyzed samples. These results are consistent with previous studies in Polar Regions (Corsolini et al., 2006; Haglund et al., 1997; Ikononou et al., 2002a; Wolkers et al., 2004). On the other hand, BDE-183 was found in less than 8% of the total analyzed samples. This congener is rarely detected in fish tissues. This can be attributed to its low intake rate due to its scarce solubility in water (Wurl et al., 2006), and/or its debromination in the intestine to BDE-154, which in turn accumulates (Stapleton et al., 2004b). Remaining PBDE congeners were <LOQ (Table 1).

Fig. 4 shows the contribution of each congener to the total PBDE load in the different species and tissues, in which the following order was observed: BDE-47 > BDE-99 > BDE-100 > BDE-28 > BDE-154 ~ BDE-153. The sum of BDE-47 and BDE-99 contributed to ca. 93% of the total PBDE load (Fig. 4S, Supplementary material). This pattern is comparable to commercial mixtures (e.g. 70-5DE Bromkal), in which BDE-47 and

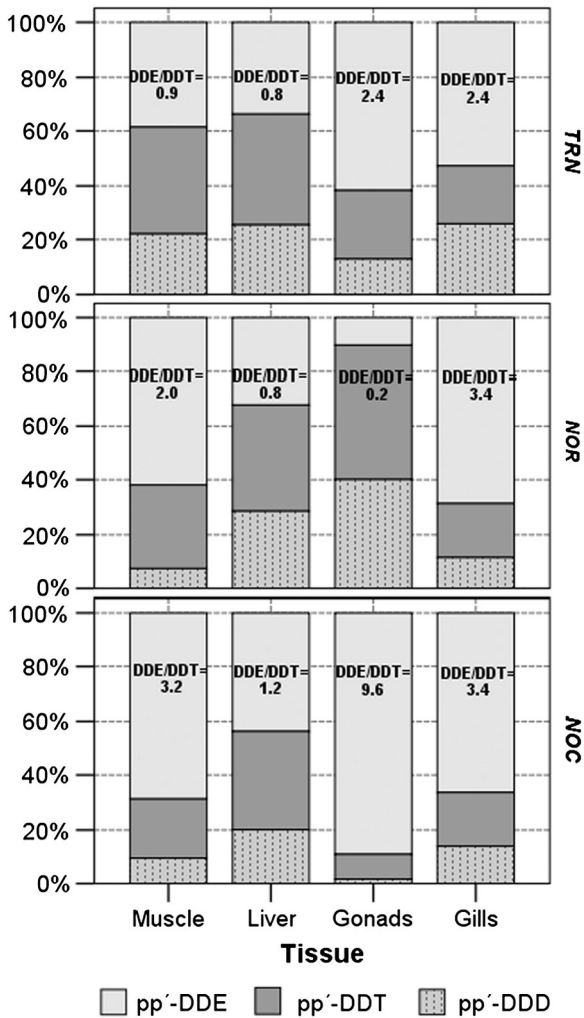


Fig. 3. Composition (percentage) of DDT and metabolites in tissues of the analyzed notothenioid species.

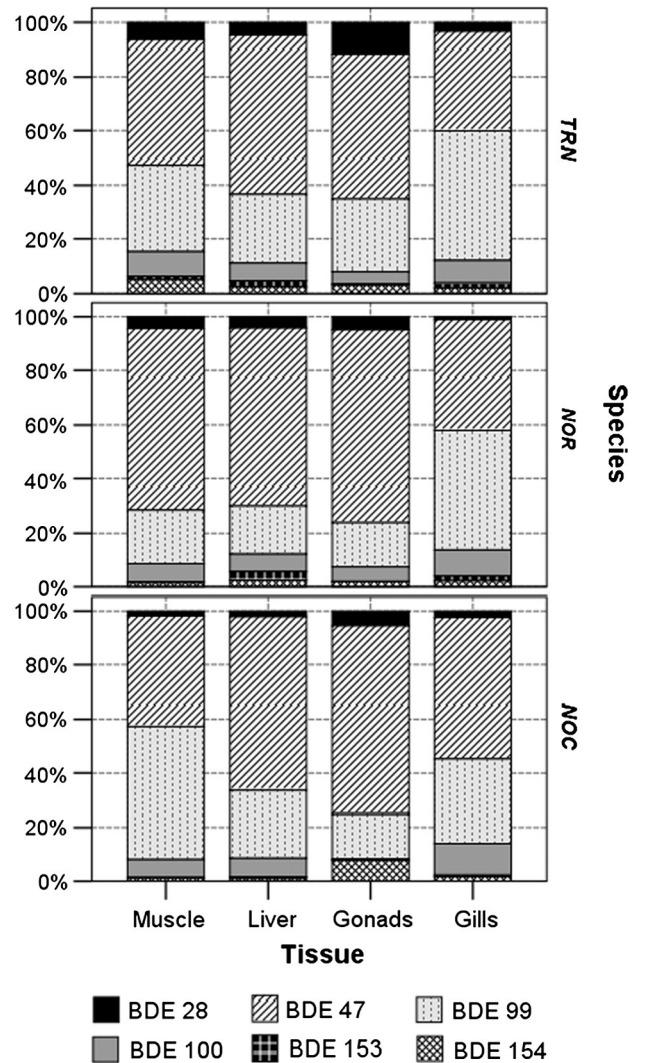


Fig. 4. Composition (percentage) of PBDE congeners in tissues of analyzed notothenioid species.

-99 are ca. 70% of the formulation (Ikonomou et al., 2002b). Additionally, the relative abundance of BDE-47 was consistent with previous reports of fish from other regions of the world (Corsolini et al., 2008; Vives et al., 2004; Voorspoels et al., 2003). Reported high levels may be either due to an elevated uptake rate or a debromination of BDE-99 (Stapleton et al., 2004a). Higher levels of BDE-28 would be expected due to its higher volatility, water solubility, and bioaccumulative potential (Watanabe and Sakai, 2003). However, the low contribution of BDE-28 to the PBDE pattern distribution may be explained due to its low ratio in technical mixtures (0.11% of BDE-28) (de Boer et al., 2000).

PBDE congeners levels presented significant differences among all analyzed samples (KW BDE-28: $X^2 = 7.34$, $p = 0.02$; BDE-47: $X^2 = 23.90$, $p < 0.00$; BDE-100: $X^2 = 13.87$, $p < 0.00$; BDE-99: $X^2 = 9.77$, $p < 0.00$; BDE-154: $X^2 = 22.87$, $p < 0.00$), except for BDE-153 and -183 (KW $X^2 = 2.53$, $p = 0.28$ and $X^2 = 0.92$, $p = 0.63$, respectively). The presence of highest brominated congeners in Antarctica suggests recent exposure to the octa- and deca-BDE commercial mixture and/or BDE-209 debromination processes (Shaw et al., 2012). Because of its large size (MW 959) BDE-209 mainly originates from contamination by local sources such as touristic or research activities instead of global fractionation process (Goutte et al., 2013). Local release of congeners resulting from BDE-209 might generate a similar exposure stage for the three species, leading thus to comparable BDE-153 and -183 concentrations among them.

The highest Σ PBDE levels were found in NOC and NOR specimens (153 and 127 ng g^{-1} l.w., respectively), while concentrations in TRN were one order of magnitude below (30 ng g^{-1} l.w.; Table 1). A suitable explanation could be inter-specific differences in selective metabolism of individual congeners and their diet (Wolkers et al., 2004). Some PBDE congeners may present higher biomagnification potentials, leading to a differential and selective enrichment in the studied species (Gustafsson et al., 1999).

Median PBDEs levels in tissues significantly differed among species (KW muscle: $X^2 = 8.17$, $p = 0.01$; liver: $X^2 = 12.91$, $p < 0.00$; gills: $X^2 = 15.88$, $p < 0.00$), except for gonads (KW $X^2 = 0.404$, $p = 0.817$). Main differences were found in liver tissue of NOC in comparison with NOR and TRN (Wxon W = 1788.5, $p < 0.00$; W = 5880, $p < 0.00$, respectively). The lowest Σ PBDE levels were found in muscle and gonads (21 and 15 ng g^{-1} l.w., all species combined), while gills and liver reported the highest (146 ng g^{-1} and 128 ng g^{-1} l.w.). These results suggest that not only dietary intakes contribute to the accumulation of PBDEs in the studied fish species, but also respiratory function. Considering the benthic habits of these species, the observed results are to be expected, since these fish species are more exposed to the PBDE adsorbed on suspended particles, as well as on seabed (Ondarza et al., 2011). Thus, water pumped through the gills could be a route of exposure for TRN and NOR species, while the euriphagous diet of NOC may represent the main intake of PBDE congeners.

3.4. HCHs

The Σ HCH concentrations ranged from 1.3 to 37 ng g⁻¹ l.w. (Table 1). While γ -HCH was detected in all samples being up to 69% of the total HCH load, the α -HCH and β -HCH contributions were 15% and 16%, respectively (Fig. 5S, Supplementary material). Contrary to PBDEs, the ratio among levels of HCH isomers found in tissues does not reflect the isomers in the commercial product, lindane (Kutz et al., 1991). This could be explained by the differences in physicochemical properties and persistence of HCH-isomers, what determine their bio-degradation rates (Phillips et al., 2005). The α -HCH isomer may bioaccumulate, while β -HCH is stable to enzymatic degradation, thus it is more persistent in biota and can biomagnify (Cipro et al., 2010; Tanabe et al., 1997). Additionally, lindane (γ -HCH, 99% purity) is still permitted for human health pharmaceutical applications toward head lice control and scabies as second line treatment (UNEP, 2009). Considering thus the degradation rate trend of HCH isomers ($\beta > \gamma > \alpha$) (Kouras et al., 1998), the highest values of γ -HCH may indicate recent contamination with lindane (UNEP, 2009).

The highest concentrations of Σ HCH in the studied fish species were found in NOC (61 ng g⁻¹ l.w.), followed by NOR (50 ng g⁻¹ l.w.). TRN had the lowest levels (32 ng g⁻¹ l.w.), which are 50% lower than for NOC (Table 1). Even when no significant differences among fish species was observed for each HCH isomer ($p > 0.05$), a comparable pattern was observed among species when the HCH isomeric profile was analyzed in tissues (Fig. 5). It is noteworthy that only β -HCH and γ -HCH isomers were detected in liver tissue, with prevalence of β -HCH (Table 1). This

fact may reflect its bioaccumulative nature and resistance to enzymatic degradation (Tanabe et al., 1997).

Statistical analysis revealed that concentrations of HCH isomers differed in muscle and gills tissues (KW $X^2 = 6.364$, $p = 0.042$ and $X^2 = 6.614$, $p = 0.037$, respectively), while for liver and gonads were comparable among the studied fish specimens (KW $X^2 = 0.39$, $p = 0.823$ and $X^2 = 5.96$, $p = 0.05$, respectively). The highest value of Σ HCH was found in gonads (81 ng g⁻¹ l.w.), followed by gills (39 ng g⁻¹ l.w.) and muscle (14 ng g⁻¹ l.w.). Comparable results were previously reported in other fish species (*Rutilus rutilus*, *Abramis brama*, *Leuciscus idus*) (Singh and Canario, 2004; Singh and Singh, 2008; Tomza-Marciniak and Witczak, 2010). These results suggest that gonads are the main organ for accumulating HCH residues, thus resulting in a potential threat to the reproductive system of the organism. Singh and Singh (2008) reported that, during reproductive phase of catfishes and carps, HCHs are transferred from liver to ovary, causing reproductive disorders (Singh and Singh, 2008).

3.5. Reported POPs levels in Antarctic fish species

In Antarctic fish, POPs levels and metabolites were reported mainly in muscle and liver tissue. Available data includes seventeen fish species from five Antarctic regions (King George Is., Elephant Is., Weddel Sea, Ross Sea and Adelie Land). Fig. 1 shows the Antarctic regions reported in bibliographic data, which were plotted in Fig. 6 and presented in Table 3S. The whole dataset was summarized in Table 3S of the Supplementary Material section. In order to get a quick view of the bulk of information, data was analyzed based on $RM C_s^{POP}$ parameter (described in Materials and methods section), and was plotted in Fig. 6. PBDE and HCH information was plotted together due to the scarce HCH data. The reference line at $\log_{10}(RM C_s^{POP}) = 0$ is indicative of those values comparable to those found for TRN. Relative mean concentrations plotted in Fig. 6 showed higher dispersion for $RM C_s^{PCB}$ values, followed by $RM C_s^{PBDE}$ and $RM C_s^{HCH}$; while $RM C_s^{DDT}$ did not show significant dispersion considering all analyzed fish species, tissue types, and regions. The significant dispersion of $RM C_s^{PCB}$ along the time, sampled areas, and fish species considered in Fig. 6, may suggest that PCBs burden in Antarctic area have still not reached a steady state (Borghesi et al., 2008; Borghesi et al., 2009; Cipro et al., 2013; Corsolini et al., 2005; Corsolini et al., 2006; Goerke et al., 2004; Goutte et al., 2013; Weber and Goerke, 2003). This could be due to reservoirs of POPs (soils and snow/ices) in Polar Regions can be remobilized due to decreasing primary emissions or due to climate change-driven warmer conditions (Cabrerizo et al., 2013). On the other hand, DDT levels were comparable with those reported for TRN, independently of the analyzed fish species, tissue types, and regions (Cipro et al., 2013; Corsolini et al., 2005; Corsolini et al., 2006; Goerke et al., 2004; Weber and Goerke, 2003). Additionally, these results suggest that while DDT is still present in Antarctica, p,p' -DDT is becoming less prevalent, resulting in an increased ratio of its metabolite, p,p' -DDE, in the fish tissues. All $RM C_s^{PBDE}$ showed values above TRN reference, independently of tissue type, fish species or analyzed region. Although the number of reports is scarce for generalizing the observations, it is interesting to mention that $RM C_s^{PBDE}$ values obtained for fish species from King George Is. were slightly higher than those from Ross Sea or Adelie Land (Borghesi et al., 2008; Borghesi et al., 2009; Cipro et al., 2013; Corsolini et al., 2006; Goutte et al., 2013). HCH were only reported in TRN, NOC and NOR species from Ross Sea and King George Is., respectively (Cipro et al., 2013; Corsolini et al., 2006). Resulted $RM C_s^{HCH}$ values for liver as well muscle tissues for NOC and NOR from King George Is. were comparable between those species and TRN.

Although POPs levels in liver and/or muscle may be useful for estimative purposes, it is interesting to consider the results of this work, which show different accumulation pattern among the organs for the studied specimens. Our results showed that PCBs, DDT and HCH highest levels were found in gonads, while PBDEs in gills. Additionally, POPs

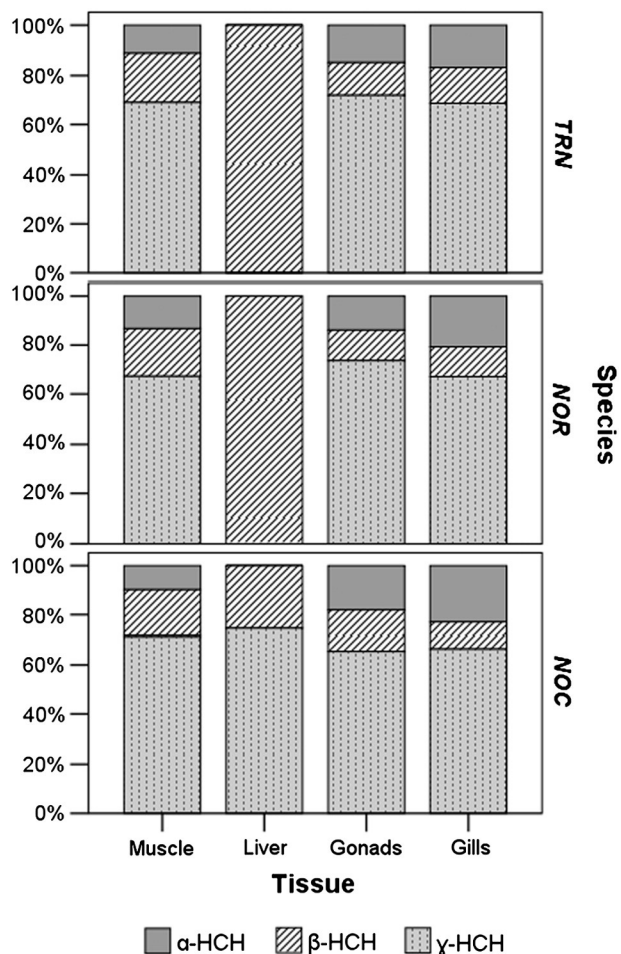


Fig. 5. Composition (percentage) of HCH isomers in tissues of analyzed notothenioid species.

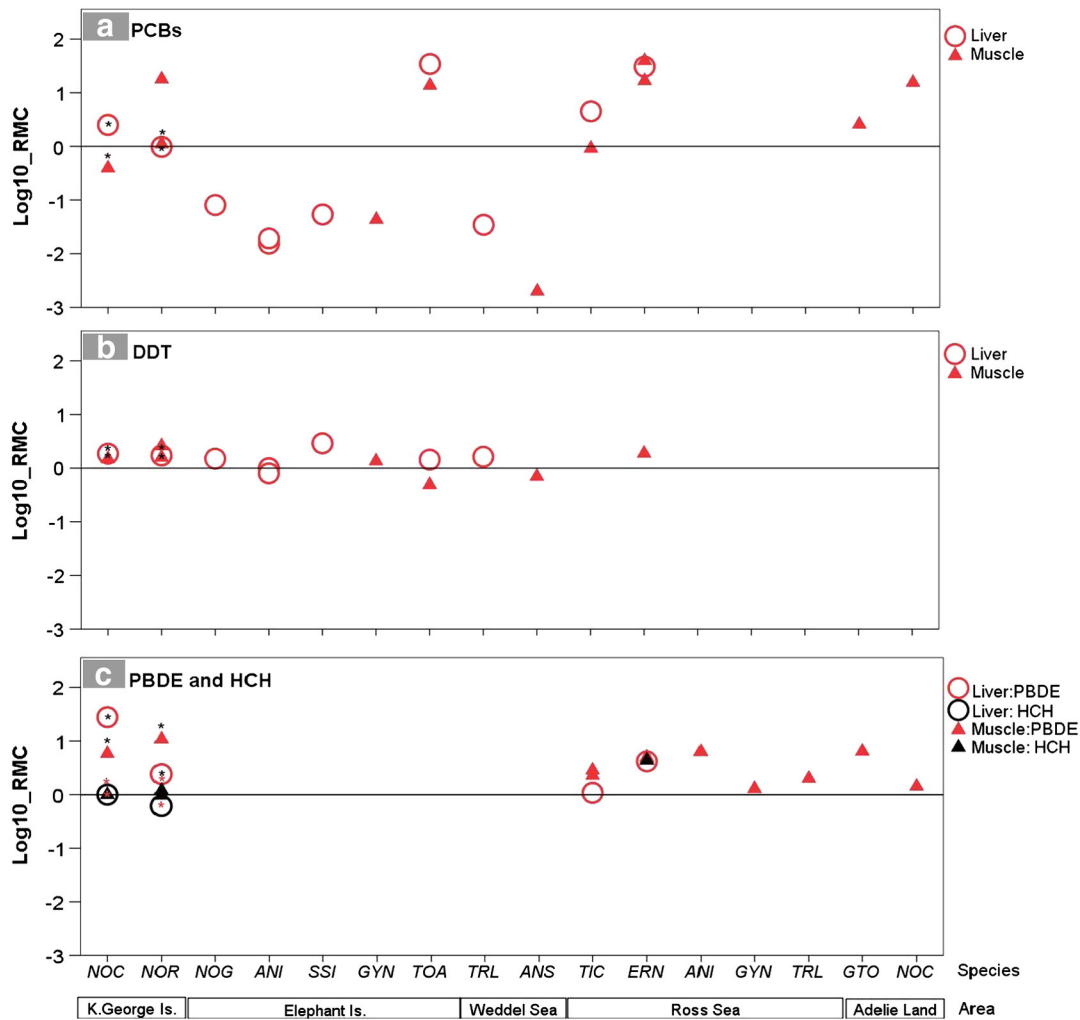


Fig. 6. Relative mean concentration value (RMC_{POP}^{rel}) for PCB (a), DDT (b), PBDE and HCH (c) reported in Antarctic fish. TRN is the referent species and it is represented by the zero line. Asterisk ** indicate the species analyzed in the present work. NOC: *Notothenia coriiceps*, NOR: *Notothenia rossii*, NOG: *Gobionotothen gibberifrons*, ANI: *Chamsocephalus gunnari*, SSI: *Chaenocephalus aceratus*, GYN: *Gymnoscopelus nicholsi*, TOA: *Dissostichus mawsoni*, TRL: *Trematomus eulepidotus*, ANS: *Pleuragramma antarcticum*, TIC: *Chionodraco hamatus*, ERN: *Trematomus bernacchii*, GTO: *Pagothenia borchgrevinki*.

here reported may suggest that NOR and NOC are the most susceptible to accumulate the studied POPs. POPs accumulation in TRN could be attenuated by the ecology of the species, which is less linked to the benthic community in the bottom, with a higher tendency of migratory habits in the water column throughout its life. Thus, this observation strengthens the suggestion of considering a broad context when estimating POPs accumulation in Antarctic fish specimens, since it could be considering a non-representative organ of the addressed problem and/or fish species without comparable habitats. As mentioned previously, the accumulation pattern may be conditioned not only by the physicochemical characteristic of the target POP, but also by the biology and ecology of the studied specimens.

Acknowledgments

This research was carried out under an agreement between the Instituto Antártico Argentino and the Instituto Argentino de Nivología, Glaciología y Ciencias Ambientales (IANIGLA). Field and laboratory works were supported by Consejo Nacional de Investigaciones Científica y Técnicas (CONICET), Agencia Nacional de Promoción Científica y Tecnológica (ANPCyT PICT: 2012-2429, PICTO: 0100-2010), Universidad Nacional de Cuyo (06/M062), Instituto Antártico Argentino, University of Antwerp, and Organization for the Prohibition of Chemical

Weapons (OPCW). The fish samples were collected by E. Moreira, C. Bellisio and L. Vila.

We are grateful to University of Antwerp for the work developed in those laboratories. J. Altamirano and B. Lana acknowledge the provision of Erasmus Mundus fellowships for their scientific visits to the Toxicological Center, University of Antwerp, Belgium.

Appendix A. Supplementary data

Supplementary data to this article can be found online at <http://dx.doi.org/10.1016/j.scitotenv.2014.08.033>.

References

Ael EV, Covaci A, Blust R, Bervoets L. Persistent organic pollutants in the Scheldt estuary: environmental distribution and bioaccumulation. *Environ Int* 2012;48:17–27.
 Ali N, Malik RN, Mehdi T, Eqani SAMAS, Javeed A, Neels H, et al. Organohalogenated contaminants (OHCs) in the serum and hair of pet cats and dogs: Biosentinels of indoor pollution. *Sci Total Environ* 2013;449:29–36.
 Ainley DG, Pauly D. Fishing down the food web of the Antarctic continental shelf and slope. *Polar Rec* 2014;50:92–107.
 Bargagli R. Antarctic ecosystems: environmental contamination, climate change, and human impact, vol. 175 New York: Springer; 2005.
 Barrera-Oro E. The role of fish in the Antarctic marine food web: differences between inshore and offshore waters in the southern Scotia Arc and west Antarctic Peninsula. *Antarct Sci* 2002;14:293–309.

- Barrera-Oro E. Analysis of dietary overlap in Antarctic fish (Notothenioidei) from the South Shetland Islands: no evidence of food competition. *Polar Biol* 2003;26:631–7.
- Borghesi N, Corsolini S, Focardi S. Levels of polybrominated diphenyl ethers (PBDEs) and organochlorine pollutants in two species of Antarctic fish (*Chionodraco hamatus* and *Trematomus bernacchii*). *Chemosphere* 2008;73:155–60.
- Borghesi N, Corsolini S, Leonards P, Brandsma S, de Boer J, Focardi S. Polybrominated diphenyl ether contamination levels in fish from the Antarctic and the Mediterranean Sea. *Chemosphere* 2009;77:693–8.
- Cabrerizo A, Dachs J, Barceló D, Jones KC. Climatic and biogeochemical controls on the remobilization and reservoirs of persistent organic pollutants in Antarctica. *Environ Sci Technol* 2013;47:4299–306.
- Casaux RJ, Mazzotta AS, Barrera-Oro E. Seasonal aspects of the biology and diet of near-shore nototheniid fish at Potter Cove, South Shetland Islands, Antarctica. *Polar Biol* 1990;11:63–72.
- Cipro CVZ, Taniguchi S, Montone RC. Occurrence of organochlorine compounds in *Euphausia superba* and unhatched eggs of *Pygoscelis* genus penguins from Admiralty Bay (King George Island, Antarctica) and estimation of biomagnification factors. *Chemosphere* 2010;78:767–71.
- Cipro CVZ, Colabuono FI, Taniguchi S, Montone RC. Persistent organic pollutants in bird, fish and invertebrate samples from King George Island, Antarctica. *Antarct Sci* 2013;25:545–52.
- Corsolini S, Ademollo N, Romeo T, Olmastroni S, Focardi S. Persistent organic pollutants in some species of a Ross Sea pelagic trophic web. *Antarct Sci* 2003;15:95–104.
- Corsolini S, Ademollo N, Romeo T, Greco S, Focardi S. Persistent organic pollutants in edible fish: a human and environmental health problem. *Microchem J* 2005;79:115–23.
- Corsolini S, Covaci A, Ademollo N, Focardi S, Schepens P. Occurrence of organochlorine pesticides (OCPs) and their enantiomeric signatures, and concentrations of polybrominated diphenyl ethers (PBDEs) in the Adélie penguin food web, Antarctica. *Environ Pollut* 2006;140:371–82.
- Corsolini S, Guerranti C, Perra G, Focardi S. Polybrominated diphenyl ethers, perfluorinated compounds and chlorinated pesticides in swordfish (*Xiphias gladius*) from the Mediterranean Sea. *Environ Sci Technol* 2008;42:4344–9.
- Covaci A, Gheorghe A, Hulea O, Schepens P. Levels and distribution of organochlorine pesticides, polychlorinated biphenyls and polybrominated diphenyl ethers in sediments and biota from the Danube Delta, Romania. *Environ Pollut* 2006;140:136–49.
- Covaci A, Voorspoels S, Roossens L, Jacobs W, Blust R, Neels H. Polybrominated diphenyl ethers (PBDEs) and polychlorinated biphenyls (PCBs) in human liver and adipose tissue samples from Belgium. *Chemosphere* 2008;73:170–5.
- de Boer J, de Boer K, Boon JP. New types of persistent halogenated compounds. The handbook of environmental chemistry. Springer; 2000. p. 61–95.
- Eastman JT, Barrera Oro E. Buoyancy studies of three morphs of the Antarctic fish *Trematomus newnesi* (Nototheniidae) from the South Shetland Islands. *Polar Biol* 2010;33:823–31.
- Goerke H, Weber K, Bornemann H, Ramdohr S, Pláňtj J. Increasing levels and biomagnification of persistent organic pollutants (POPs) in Antarctic biota. *Mar Pollut Bull* 2004;48:295–302.
- Goutte A, Chevreuril M, Alliot F, Chastel O, Chery Y, Eléaume M, et al. Persistent organic pollutants in benthic and pelagic organisms off Adélie Land, Antarctica. *Mar Pollut Bull* 2013;77:82–9.
- Gustafsson K, Björk M, Burreau S, Gilek M. Bioaccumulation kinetics of brominated flame retardants (polybrominated diphenyl ethers) in blue mussels (*Mytilus edulis*). *Environ Toxicol Chem* 1999;18:1218–24.
- Haglund PS, Zook DR, Buser HR, Hu J. Identification and quantification of polybrominated diphenyl ethers and methoxy-polybrominated diphenyl ethers in Baltic biota. *Environ Sci Technol* 1997;31:3281–7.
- Ikonoumou MG, Rayne S, Addison RF. Exponential increases of the brominated flame retardants, polybrominated diphenyl ethers, in the Canadian Arctic from 1981 to 2000. *Environ Sci Technol* 2002a;36:1886–92.
- Ikonoumou MG, Rayne S, Fischer M, Fernandez MP, Cretney W. Occurrence and congener profiles of polybrominated diphenyl ethers (PBDEs) in environmental samples from coastal British Columbia, Canada. *Chemosphere* 2002b;46:649–63.
- Jaspers VLB, Sonne C, Soler-Rodríguez F, Boertmann D, Dietz R, Eens M, et al. Persistent organic pollutants and methoxylated polybrominated diphenyl ethers in different tissues of white-tailed eagles (*Haliaeetus albicilla*) from West Greenland. *Environ Pollut* 2013;175:137–46.
- Kock KH. Antarctic fish and fisheries. New York: Cambridge University Press; 1992.
- Kock KH, Kellermann A. Reproduction in Antarctic Nototheniid fish. *Antarct Sci* 1991;3:125–50.
- Kouras A, Zouboulis A, Samara C, Kouimtzi T. Removal of pesticides from aqueous solutions by combined physicochemical processes — the behaviour of lindane. *Environ Pollut* 1998;103:193–202.
- Kutz FW, Wood PH, Bottimore DP. Organochlorine pesticides and polychlorinated biphenyls in human adipose tissue. *Rev Environ Contam Toxicol* 1991;120:1–82.
- Montone RC, Taniguchi S, Weber RR. PCBs in the atmosphere of King George Island, Antarctica. *Sci Total Environ* 2003;308:167–73.
- Mormede S, Davies IM. Horizontal and vertical distribution of organic contaminants in deep-sea fish species. *Chemosphere* 2003;50:563–74.
- Ondarza PM, Gonzalez M, Fillmann G, Miglioranza KSB. Polybrominated diphenyl ethers and organochlorine compound levels in brown trout (*Salmo trutta*) from Andean Patagonia, Argentina. *Chemosphere* 2011;83:1597–602.
- Paasivirta J, Sinkkonen S, Mikkelsen P, Rantio T, Wania F. Estimation of vapor pressures, solubilities and Henry's law constants of selected persistent organic pollutants as functions of temperature. *Chemosphere* 1999;39:811–32.
- Phillips TM, Seeh AG, Lee H, Trevors JT. Biodegradation of hexachlorocyclohexane (HCH) by microorganisms. *Biodegradation* 2005;16:363–92.
- Roossens L, Dirlu AC, Goemans G, Belpaire C, Gheorghe A, Neels H, et al. Brominated flame retardants and polychlorinated biphenyls in fish from the river Scheldt, Belgium. *Environ Int* 2008;34:976–83.
- Shaw SD, Berger ML, Weijs L, Covaci A. Tissue-specific accumulation of polybrominated diphenyl ethers (PBDEs) including deca-BDE and hexabromocyclododecanes (HBCDs) in harbor seals from the northwest Atlantic. *Environ Int* 2012;44:1–6.
- Singh PB, Canario AVM. Reproductive endocrine disruption in the freshwater catfish, *Heteropneustes fossilis*, in response to the pesticide gamma-hexachlorocyclohexane. *Ecotoxicol Environ Saf* 2004;58:77–83.
- Singh PB, Singh V. Pesticide bioaccumulation and plasma sex steroids in fishes during breeding phase from north India. *Environ Toxicol Pharmacol* 2008;25:342–50.
- Sladen WJL, Menzie CM, Reichel WL. DDT residues in Adélie penguins and a crabeater seal from Antarctica. *Nature* 1966;210:670–3.
- Stapleton HM, Alaei M, Letcher RJ, Baker JE. Debromination of the flame retardant decabromodiphenyl ether by juvenile carp (*Cyprinus carpio*) following dietary exposure. *Environ Sci Technol* 2004a;38:112–9.
- Stapleton HM, Letcher RJ, Baker JE. Debromination of polybrominated diphenyl ether congeners BDE 99 and BDE 183 in the intestinal tract of the common carp (*Cyprinus carpio*). *Environ Sci Technol* 2004b;38:1054–61.
- Storelli MM, Losada S, Marcotrigiano GO, Roossens L, Barone G, Neels H, et al. Polychlorinated biphenyl and organochlorine pesticide contamination signatures in deep-sea fish from the Mediterranean Sea. *Environ Res* 2009;109:851–6.
- Tanabe S, Madhusree B, Öztürk AA, Tatsukawa R, Miyazaki N, Özdamar E, et al. Persistent organochlorine residues in harbour porpoise (*Phocoena phocoena*) from the Black Sea. *Mar Pollut Bull* 1997;34:338–47.
- Tatton JOG, Ruzicka JHA. Organochlorine pesticides in Antarctica. *Nature* 1967;215:346–8.
- Tomza-Marciniak A, Witczak A. Distribution of endocrine-disrupting pesticides in water and fish from the Oder River, Poland. *Acta Ichthyol Piscat* 2010;40:1–9.
- UNEP. Stockholm convention on persistent organic pollutants; 2009.
- UNEP. Polychlorinated biphenyls (PCBs): uses and environmental releases; 2013.
- van den Brink NW, Riddle MJ, van den Heuvel-Greve M, van Franeker JA. Contrasting time trends of organic contaminants in Antarctic pelagic and benthic food webs. *Mar Pollut Bull* 2011;62:128–32.
- Vives I, Grimalt JO, Lacorte S, Guillaumon M, Barceló D, Rosseland BO. Polybromodiphenyl ether flame retardants in fish from lakes in European high mountains and Greenland. *Environ Sci Technol* 2004;38:2338–44.
- Vives I, Grimalt JO, Ventura M, Catalan J, Rosseland BO. Age dependence of the accumulation of organochlorine pollutants in brown trout (*Salmo trutta*) from a remote high mountain lake (Redó, Pyrenees). *Environ Pollut* 2005;133:343–50.
- Voorspoels S, Covaci A, Schepens P. Polybrominated diphenyl ethers in marine species from the Belgian North Sea and the Western Scheldt Estuary: levels, profiles, and distribution. *Environ Sci Technol* 2003;37:4348–57.
- Wania F, Daly GL. Estimating the contribution of degradation in air and deposition to the deep sea to the global loss of PCBs. *Atmos Environ* 2002;36:5581–93.
- Wania F, Mackay D. Tracking the distribution of persistent organic pollutants. *Environ Sci Technol* 1996;30:390A–7A.
- Watanabe I, Sakai SI. Environmental release and behavior of brominated flame retardants. *Environ Int* 2003;29:665–82.
- Weber K, Goerke H. Persistent organic pollutants (POPs) in Antarctic fish: levels, patterns, changes. *Chemosphere* 2003;53:667–78.
- Weijs L, van Elk C, Das K, Blust R, Covaci A. Persistent organic pollutants and methoxylated PBDEs in harbour porpoises from the North Sea from 1990 until 2008: young wildlife at risk? *Sci Total Environ* 2010;409:228–37.
- Wolkers H, Van Bavel B, Derocher AE, Wiig Ø, Kovacs KM, Lydersen C, et al. Congener-specific accumulation and food chain transfer of polybrominated diphenyl ethers in two Arctic food chains. *Environ Sci Technol* 2004;38:1667–74.
- Wurl O, Lam PKS, Obbard JP. Occurrence and distribution of polybrominated diphenyl ethers (PBDEs) in the dissolved and suspended phases of the sea-surface microlayer and seawater in Hong Kong, China. *Chemosphere* 2006;65:1660–6.
- Yogui GT, de Oliveira Santos MC, Montone RC. Chlorinated pesticides and polychlorinated biphenyls in marine tucuxi dolphins (*Sotalia fluviatilis*) from the Cananéia estuary, southeastern Brazil. *Sci Total Environ* 2003;312:67–78.