LEGACY AND EMERGENT POPs IN THE GREENLAND SHARK *Somniosus microcephalus* AND IN ITS PREYS FROM NE GREENLAND FJORDS

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Introduction

The Greenland shark *Somniosus microcephalus* (Bloch and Schneider, 1801) is an interesting species from an ecotoxicologiacl point of view, being a long-lived and slow-growing[1] deep-sea shark at risk due to its longevity and food habits that may allow it to bioaccumulate toxic and persistent organic pollutants (POPs)[1-3]. The Greenland sharks are the largest fish in the Arctic sea-waters and due to their large size and biological features, their biology and life history are poorly known[3-4]. Ecotoxicological data are also very scarce.

The Greenland shark stomach content analysis indicate they feed on a wide variety of *taxa* including invertebrates, fish, and marine mammals[1, 4-7]. It was already reported as an excellent sentinel species for ecotoxicologial studies in the Arctic[2, 5] and a key species of the Arctic food web[7].

The aims of this study were to determine the concentrations of polychlorobyphenils (PCBs), polybromodiphenylethers (PBDEs), perfluorooctane sulfonate (PFOS) and perfluorooctanoic acid (PFOA), polychlorinated dibenzo-dioxins (PCDDs) and -furans (PCDFs), *o,p'*- and *p,p'* isomers of DDT, DDD and DDE, alpha-, beta-, gamma-, delta- isomers of HCHs, dieldrin, endrin, and HCB in the Greenland sharks and other fish and invertebrates from the pristine North-East Greenland fjords (Greenland Sea). Another aim of this study was to assess whether there is an increase of concentrations from lower to higher trophic levels.

Materials and methods

An aliquot of the whole body homogenates of invertebrates plus *T. nybelini* and the muscle of all the fish species were analysed; in addition, the stomach content homogenate of *L. fabricis*, *A glacialis*, and *A. hyperborea* were also analysed. All the organism were caught in the Kong Oscar Fjord, Greenland Sea (NE Greenland) in August 2010, during the scientific expedition TUNU-IV in the framework of the international program TEAM-Fish (previously TUNU-MAFIG) led by the University of Tromsø, Norway. Three specimens of Greenland shark were captured by long-line; the other organisms were caught by bottom trawling. The samples were labeled and stored at –20°C. Sharks were likely sexually immature; maturity in the Greenland sharks has been reported to be reached at total lengths of about 450 cm for females and about 300 cm for males[4].

PBDEs, PCBs, PCDDs, PCDFs, and chlorinated pesticides were analysed in 1-15 g of tissue following a method described elsewhere[8], including Soxhlet extraction, PowerPrep clean up and separation[9], high-resolution gaschromatography[8, 10]. After Soxhlet extraction, an aliquot of samples was used to determine lipid contents gravimetrically. PBDE, PCDD, PCDF and PCB congeners (ΣPBDEs=19, ΣPCDD=7, ΣPCDF=10 and ΣPCB=55 congeners) are represented by their IUPAC numbers throughout the text. Results are given as mean of three replicates on a wet weight basis (wet wt).

Concentrations of PFOS and PFOA in the samples (whole body or tissues) were measured using high performance liquid chromatography (HPLC) with electrospray ionization (ESI) tandem mass spectrometry[10-11]:

Data quality assurance and quality control protocols included matrix spikes, laboratory blanks, use of certified reference material (CRM) and continuing calibration verification. Matrix spikes were analyzed for each type of tissue sample. Blanks were analyzed with each set of five tissue samples as a check for possible laboratory contamination and interferences. Concentrations below the limit of detection (LOD) were treated as LOD/2 for calculations.

Results and discussion

Being a long-lived top predators, the Greenland shark can bioaccumulate toxic and persistent organic pollutants (POPs). It is an opportunistic feeder that may feed on a variety of items depending on availability and feeding grounds and many species listed in Table 1 may be its prey[1,7].

The following PCB congeners were quantified: IUPAC nos. 28, 37, 52, 95, 101, 99, 110, 123, 118, 114, 105, 151, 149, 145, 153, 138, 128, 167, 156, 137, 187, 183, 177, 180, 170, 189 and most of them were detected in the

shark samples (57.82 ng/g wet wt); only PCBs 28, 52, 95, 101, 99, and 138 (= Σ6PCBs) were found in the invertebrates and other fish. Only those PCBs detected in most of the samples were considered for comparisons. The Σ6PCBs concentrations were higher in the Greenland shark muscle (18.26 ng/g wet wt) > C. *reinhardti* (5.02 ng/g wet wt) > stomach content of *A. hyperborea* (1.4454 ng/g we wt) > *B. saida* (0.355 ng/g wet wt) (Table 1). The concentrations found in the plasma of Greenland shark from Svalbard (Norway) collected in 2008-2009[12] (36.3±24.6 ng/g wet wt) were of the same order of magnitude of those detected in the shark analyzed in this study.

The ΣPBDE concentrations were 1365±1845 pg/g wet wt in the shark muscle; BDE66 was 499.13 pg/g wet wt, BDE77 was 2127.45 pg/g wet wt, and BDE119 was 3.97 pg/g wet wt. The comparison between the species was based on BDEs 47, 100, and 99 (= Σ3PBDEs), the only congeners detected in the other organisms (Figure 1): BDE47 made up most of the residue in the shark, while BDEs 99 and 100 were major contributors to the ΣPBDEs in the other species of fish and invertebrates. The BDE47 concentrations were lower in *Arctogadus glacialis* < Amphipods < *Amblyraja hyperborean* < *Somniosus microcephalus*, and the Σ3PBDEs in *Amblyraja hyperborea* < *Somniosus microcephalus* << other species (1-2 order of magnitude lower). BDE47 is a major constituents of technical mixtures and it bioaccumulates largely in fishes[10] and biota[13]. It is the most abundant congener among the Arctic fishes, marine birds and mammals, and humans (Inuit women)[14-15]. Ikonomu et al.[16] reported that debromination processes may occur in organisms following two different pathways: BDE154 → BDE99 → BDE47 and BDE153 → BDE100 → BDE49; congeners nos. 138, 153, 154, 156 were below LOD in all our samples and BDE99, a major constituent of Bromkal mixtures, made up less than 5% of BDE residue in our samples.

Among PCDDs/DFs, only two congeners were detected in the shark muscle: the 1,2,3,6,7,8-HxCDF was 2.24 pg/g and the 2,3,7,8-TCDF was 50.84 pg/g. All the other PCDD/DF congeners were below LOD in the shark muscle and in the other organisms.

PFOS concentrations were below LOD in all the samples except in the amphipods (1.28 ng/g wet wt) and the PFOA ranged from < LOD to 14.46 ng/g wet wt (Table 1). PFOA ranged from <LOD to 14.46 in *G. argentatus* and the pattern was Ascidians < Amphipods < *Liparis fabricis* < *Arctogadus glacialis* < Sea star < Shrimp female < Shrimp male < *Careproctus reinhardti* < Ophiuroides < Bivalves < *Leptagonus decagonus* < *Gaidropsarus argentatus.*

The chlorinated pesticides were below LOD in all the samples, except the shark muscle, where the following chemicals were detected: HCB was 0.04 ng/g, gamma-HCH was 0.012 ng/g, o,p'-DDE was 0.22±0.2 ng/g, p,p'-DDT was 0.14±0.19 ng/g, and p,p'-DDE was 0.17±0.12 ng/g. The presence of the DDT at detectable levels in the top predator, the Greenland shark, is interesting because it may be due to the current use of this pesticide in those countries where the malaria disease is still endemic.

In general, our results confirmed the low contamination of organisms from one of the last pristine and remote areas of the world. The Greenland shark showed higher concentrations respect to the other species including its preys. Its peculiar biological and ecological features make it a vulnerable species to bioaccumulation of toxic and persistent contaminants. Concentrations in the other fish species were low, but some chemicals showed unexpected concentrations. Further studies will be focused on the confirmation of these results and on the toxic risk assessment for the Greenland shark and its trophic web, taking into account that it was listed as 'near threatened' in 2011 by the IUCN Red List of Threatened Species[17], given to the shark's slow growth rate.

Table 1: Concentrations of Σ6PCBs (sum of IUPAC nos. 52 + 37 + 95 + 101 + 138), PFOA, PFOS (ng/g wet wt), BDE47, BDE100, BDE99 (pg/g wet wt) in the Greenland shark and other organisms from NE Greenland Fjords (Greenland Sea) [TL= trophic level (fishbase.org); Lip% = lipid content expressed as percentage; n.d. = not detected). Other BDE congeners as well as chlorinated pesticides and PCDDs/DFs were <LOD and are not showed in the table.

	Tissue	TL	n	Lip %	ΣPCBs	PFOA	PFOS	BDE47	BDE100	BDE99
Amphipods	whole		6	25.8	<0.001	1.12	1.28	10.8	6.4	5.0
Shrimp male	whole		10	14.1	<0.001	6.69	<0.5	6.3	4.0	9.1
Shrimp female	whole		10	12.6	<0.001	2.89	<0.5	2.9	5.4	4.2
Sea star	whole		10	21.6	<0.001	2.14	<0.5	1.0	14.5	19.3
Ophiuroides	whole		1	8.5	<0.001	10.56	<0.5	5.6	6.1	1.5
Gasteropods	whole		5	12.7	<0.001	<0.5	<0.5	0.8	<0.1	4.7
Octopus	whole		5	20.8	0.292	<0.5	<0.5	2.01	11.25	7.59
Bivalves	whole		26	22.1	0.27	11.69	<0.5	5.7	19.0	8.3
Ascidians	whole		1	21.5	<0.001	0.68	<0.5	1.4	9.7	13.5
Leptagonus decagonus	muscle	3.21	5	24.6	<0.001	12.46	<0.5	2.2	18.6	<0.1
Triglops nybelini	whole	3.29	4	22.7	<0.001	<0.5	<0.5	3.1	4.1	8.8
Liparis fabricis	stomach content	3.33	4	25.6	0.13	1.12	<0.5	0.9	10.8	17.9
	muscle		4	17.8	0.21	10.29	<0.5	<0.1	3.2	<0.1
Gaidropsarus argentatus	muscle	3.39	5	14.2	0.096	14.46	<0.5	1.1	3.1	<0.1
Careproctus reinhardti	muscle	3.75	3	23.6	<0.001	7.11	<0.5	1.1	15.7	<0.1
Arctogadus glacialis	stomach content	3.82	4	90.2	<0.001	1.52	<0.5	10.5	38.9	<0.1
	muscle		4	15.4	<0.001	9.89	<0.5	1.1	<0.1	<0.1
Amblyraja hyperborea	stomach content	3.84	1	15.2	1.445	<0.5	<0.5	11.4	125.6	10.6
	muscle		1	20.5	<0.001	<0.5	<0.5	2.4	3.7	<0.1
Boreogadus saida	muscle	3.1	30	15.6±4	0.355	n.d.	n.d.	0.47	<0.1	<0.1
Somniosus microcephalus	white muscle	4.22	3	24±2	18.26	n.d.	n.d.	419	<0.1	7.05

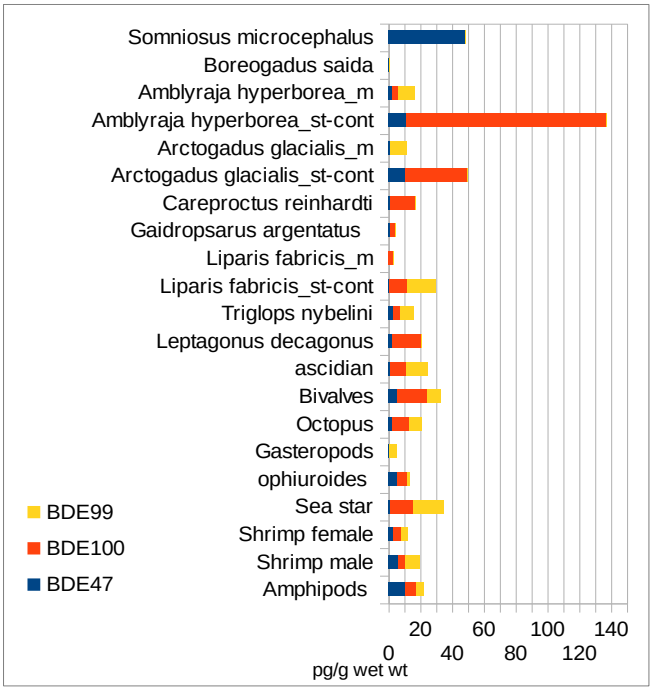

Figure 1: Concentrations of BDE47, 99, 100 (pg/g wet wt) and PFOS, PFOA (ng/g wet wt) in the organisms.

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References:
1. Fisk AT, Tittlemier S, Pranschke J, Norstrom RJ (2002); *Ecology* 83: 2162-2172
2. Strid A, Jörundsdóttir H, Päpke O, Svavarsson J, Bergman Å (2007) *Mar. Pollut. Bull.* 54: 1514–1522
3. MacNeil MA, McMeans BC, Hussey NE, Vecsei P, Svavarsson J, *et al.* (2012) *J. Fish. Biol.* 80: 991–1018
4. Yano K, Stevens JD, Compagno LJV (2007) *J. Fish Biol.* 70: 374-390
5. McMeans BC, Svavarsson J, Dennard S, Fisk AT (2010) *Can. J. Fish. Aquatic Sci.* 67: 1428–1438
6. Leclerc LM, Lydersen C, Haug T, Glover KA, Fisk AT, Kovacs KM (2011) *Polar Res.* 30: 7342, doi:10.3402/polar.v30i0.7342.
7. Nielsen J., Hedeholm R.B., Simon M., Steffensen J.F., 2014. *Polar Biol*. 37, 37–46.
8. Corsolini S, Borghesi N, Schiavone A, Focardi S (2006) *Environ. Sci. Pollut. Res*. 14;421–429
9. Pirard C, Focant JF, De Pauw E (2002) *Bioanal. Chem*. 372: 373-381
10. Corsolini S, Guerranti C, Perra G, Focardi S (2008) *Environ. Sci. Technol*. 42(12) : 4344 – 4349
11. Hansen KJ, Clemen LA, Ellefson ME, Johnson HO (2001) *Environ. Sci. Technol. 35:* 766–70
12. Molde K, Ciesielski TM, Fisk AT, Lydersen C, Kovacs KM, Sørmo EG, Jenssen BM (2013) *Sci. Tot. Environ*. 442: 445-454
13. de Boer J, de Boer K, Boon JP (2000) In: Paasivirta, J (Ed) The Handbook of Environmental Chemistry. Springer, Berlin pp. 61–95
14. Vorkamp K, Christensen JH, Glasius M, Riget FF (2004) *Mar. Pollut. Bull*. 48: 111–121
15. Kelly BC, Ikonomou MG, Blair JD, Gobas FAPC (2008) *Sci. Total Environ.* 401 : 60–72
16. Ikonomou MG, Rayne S, Fischer M (2002) *Chemosphere* 46(5): 649–663
17. Kyne PM, Sherrill-Mix SA, Burgess GH (2006) *Somniosus microcephalus*. In: IUCN 2013. IUCN Red List of Threatened Species. Version 2013.1. <www.iucnredlist.org>. Downloaded on 01 October 2013.