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Essential and non-essential element concentrations in two sleeper shark species collected in arctic waters

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Patterns of essential and non-essential elements provide insight into sleeper shark biology and physiology.

Abstract

A number of elements/metals have increased in arctic biota and are of concern due to their potential toxicity. Most studies on elements in the Arctic have focused on marine mammals and seabirds, but concentrations in the Greenland shark (*Somniosus microcephalus*) and Pacific sleeper shark (*Somniosus pacificus*), the only two shark species known to regularly inhabit arctic waters, have never been reported. To address this data gap, concentrations and patterns of 25 elements were analyzed in liver of Greenland sharks collected about Cumberland Sound ($n = 24$) and Pacific sleeper sharks collected about Prince William Sound ($n = 14$). Several non-essential elements differed between species/locations, which could suggest geographical exposure differences or ecological (e.g., diet) differences between the species. Certain essential elements also differed between the two sleeper sharks, which may indicate different physiological requirements between these closely related shark species, although information on such relationships are lacking for sharks and fish.

Keywords: Metals; Greenland shark; Pacific sleeper shark; *Somniosus*; Hepatic tissue

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1. Introduction

Certain elements[1] have been highly studied in arctic invertebrates, freshwater fish, birds, and marine mammals due to concern over their toxicity and potential to biomagnify in arctic wildlife (Dietz et al., 1996; Riget et al., 2000). Mercury (Hg), for example, is frequently analyzed in arctic ecosystems, as it is associated with neurological damage in humans and biota (World Health Organization, 1990), is known to biomagnify (Campbell et al., 2005a; Dietz et al., 1996) and has achieved concentrations in some biota that warrant concern for both the wildlife and for humans whose diet includes these wildlife species (Fisk et al., 2003). Cesium, zinc, and rubidium have also been shown to biomagnify (Campbell et al., 2005a,b; Dietz et al., 1996). Despite concern over elements in arctic biota, there are limited data on elements in arctic marine fish, and no data for the only sharks, the Greenland shark, *Somniosus microcephalus*, and the Pacific sleeper shark, *Somniosus pacificus*, that routinely inhabit arctic waters (Bigelow and Schroeder, 1948; Benz et al., 2004). As high concentrations

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[1] The term "elements" is considered more appropriate than the more commonly used terms "metals" or "heavy metals" because the large suite of elements generated by modern analytical methods, and reported in this study, include both metallic and non-metallic members (Duffus, 2002).

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of organic contaminants (e.g., polychlorinated biphenyls (PCBS)) have been previously measured in the Greenland shark, likely due to the shark's high trophic level (Fisk et al., 2002), there is a need to address the concentrations of elements in the Greenland shark and its Pacific congener, the Pacific sleeper shark.

The Greenland and Pacific sleeper sharks are megafaunal vertebrates of Somniosidae, the sleeper sharks (Compagno, 1984). To date, Greenland sharks have been primarily reported from areas seasonally covered by ice in the North Arctic (Bigelow and Schroeder, 1948; Skomal and Benz, 2004), but have also been reported through more temperate areas in the Atlantic (Bigelow and Schroeder, 1948) and as far south as about the coast of Georgia, United States (Herdendorf and Berra, 1995). Pacific sleeper sharks have been reported from areas as far north as the Chukchi Sea (Benz et al., 2004) to as far south as approximately the southern coast of California, United States (Compagno, 1984). Based on stomach content data, the Greenland and Pacific sleeper sharks feed on a wide variety of taxa including invertebrates, fish, and marine mammals (Bigelow and Schroeder, 1948; Bright, 1959; Yang and Page, 1999), although the recent use of biochemical tools suggests that the species composition of the sharks' diet could vary between the species. For example, using stable isotope and stomach content analysis, the Greenland shark was shown to consume piscivorous fishes (e.g., turbot, *Reinhardtius hippoglossoides*) and piscivorous pinnipeds (e.g., ringed seal, *Phoca hispida*) (Fisk et al., 2002), whereas a recent study utilizing fatty acid analysis suggested that Pacific sleeper sharks off the coast of Alaska feed heavily on zooplanktonivorous fishes (e.g., Pacific herring, *Clupea pallasi*) and occasionally consume the blubber of filter-feeding cetaceans (i.e., baleen whales) (Schaufler et al., 2005). Although more detailed studies on the feeding behavior of these sharks are needed, these studies suggest that Pacific sleeper sharks could be feeding at a lower trophic level than Greenland sharks. The Greenland and Pacific sleeper sharks are the largest fish in arctic waters, apparently abundant (Bigelow and Schroeder, 1948; Hansen, 1963), and likely ecologically significant (Fisk et al., 2002; Taggart et al., 2005), however, there is a lack of data concerning their biology and life history. Further, the potentially high trophic level (Fisk et al., 2002) and long life (Hansen, 1963) of the Greenland shark could make it an excellent sentinel for contaminants in the arctic marine ecosystem.

Few previous studies have assessed element concentrations in sharks (Marcovecchio et al., 1991; Storelli et al., 2003; Domi et al., 2005), and most have only focused on four or five elements of concern due to their toxicity. Elements differ from most organic contaminants in that they occur naturally in the environment and can be classified as either essential (e.g., copper, zinc, manganese), which are necessary to an organism for life, or non-essential (e.g., arsenic, cadmium, mercury), which may be present in an organism, but serve no known purpose. Non-essential elements can sometimes behave like essential elements if regulated through the same processes, but are generally believed to be regulated less efficiently than essential elements (Kraemer et al., 2005). Therefore, interpreting concentrations of non-essential and biomagnifying elements (Hg and Rb) could provide information concerning diet and trophic level, whereas concentrations of non-essential elements that have not been shown to biomagnify could provide information on geographical exposure variations. Essential elements, which should be highly regulated, could be used to infer the physiological requirements of an organism. Very little is known about element regulation in marine fishes, and even less information exists concerning elasmobranchs (Grosell et al., 2003). Although the lack of data requires extreme conservation when drawing conclusions about shark physiology, further investigations into concentrations and patterns of certain essential elements in the Greenland shark and Pacific sleeper shark, or any species of shark, is a valid effort with potentially important implications.

This study addresses a data gap for elements in sharks and provides the first hepatic element data for the Greenland shark (collected in Cumberland Sound, Nuavut) and the Pacific sleeper shark (collected in Prince William Sound, Alaska) (Fig. 1), and has two main goals: (1) To assess within-species relationships of individual element concentration vs. shark length for all non-essential elements (which could be useful in determining the occurrence of any biomagnification) and essential elements (which could provide information concerning changes in physiological requirement with size), and (2) To make between-species comparisons of non-essential element concentrations to determine environmental and/or dietary exposure differences, and essential element concentrations to infer physiological differences between the species. The Greenland sharks were expected to exhibit higher concentrations of non-essential, biomagnifying elements (i.e., mercury and rubidium) based on the assumption that they feed at a higher trophic level than the Pacific sleeper sharks. Variances in other non-essential elements that are not known to biomagnify are expected to reflect environmental exposure differences. The Greenland and Pacific sleeper sharks were expected to have similar concentrations of essential elements based on their close phylogenetic relationship and therefore potentially similar physiologies.

2. Methods

2.1. Sampling

Greenland shark samples ($n = 24$, 14 males and 10 females) were collected as by-catch in the bottom long-line commercial turbot fisheries in Cumberland Sound in April of 1999 and 2000 and ranged in size from 234 cm to 322 cm (fork length) with a mean fork length of 278.0 ± 4.8 (mean ± 1 SE). All Pacific sleeper shark samples ($n = 14$, 7 males and 7 females) were collected as part of an Alaska Department of Fish and Game bottom long-line survey in Prince William Sound in September of 2000. The Pacific sleeper sharks ranged in size from 139 cm to 244 cm (fork length) with a mean fork length of 201.3 ± 8.9. The low sample sizes are due to the enigmatic nature of these sharks, in that they often occur at high latitudes and in deep water (Compagno, 1984; Herdendorf and Berra, 1995). Liver samples (left lobe) were collected from each shark shortly after death and were stored frozen in plastic bags until element analysis. Liver tissue typically has higher concentrations of elements

Fig. 1. Map showing sampling sites. Greenland shark samples ($n = 24$, 14 males and 10 females) were collected in Cumberland Sound and ranged in size from 234 cm to 322 cm (fork length). Pacific sleeper shark samples ($n = 14$, 7 males and 7 females) were collected in Prince William and ranged in size from 139 cm to 244 cm (fork length).

relative to muscle tissue (Dietz et al., 1998), and was therefore chosen for element analysis in an attempt to maximize the number of quantifiable elements.

2.2. Element analysis

Tissue samples were analyzed for elements at the National Laboratory for Environmental Testing (NLET) at the National Water Research Institute in Burlington, Canada (National Laboratory for Environmental Testing, 2003). Total mercury (Hg) in liver tissue (NLET Method 02-2802) was analyzed by cold vapour atomic absorption spectrometry (CVAAS). Analyses of 24 elements in liver samples (NLET Method 02-2705) were analyzed by inductively coupled plasma-Sector Field spectrometry (ICP-SFMS), with 22 elements analyzed at low resolution (silver (Ag), barium (Ba), cadmium (Cd), cobalt (Co), chromium (Cr), copper (Cu), gallium (Ga), lanthanum (La), lithium (Li), manganese (Mn), molybdenum (Mo), nickel (Ni), palladium (Pd), lead (Pb), platinum (Pt), rubidium (Rb), antimony (Sb), strontium (Sr), thallium (Tl), uranium (U), vanadium (V) and zinc (Zn) and the rest at high resolution (arsenic (As) and selenium (Se)). Instrumental detection limits (DL) for most elements is 0.001 μg/g, except for Sr (0.05 μg/g), Pt (0.01 μg/g), Pd (0.1 μg/g), Li (0.1 μg/g) and Sb (0.01 μg/g). Of the analyzed elements, Co, Cr, Cu, Mn, Mo, Ni, Se, and Zn were considered essential for the sharks based on their known necessity in the diets of fish (Lovell, 1998), and therefore, the remaining elements (Ag, As, Ba, Cd, Ga, Hg, La, Li, Pd, Pb, Pt, Rb, Sb, Sr, Tl, U, V) were considered non-essential. All elements were analyzed with wet tissue, and therefore are expressed as μg/g wet weight (μg/g, ww). NLET is certified by the Canadian Environmental Analytical Laboratory program of the Canadian Standards Association and participates in the QA program for the Northern Contaminants Program, with good results (Stokker, 2003).

2.3. Data analyses

Elements that were below DL in more than 30% of all samples (Ba, Cr, La, Li, Ni, Pb, Pd, Pt, Sb, Tl, U, and V) were excluded from the within-species (linear regression) and between-species (univariate and multivariate) statistical analyses. The data for Ag and Mn were arcsine transformed and concentrations of As, Cd, Co, Cu, Hg, and Zn were logarithmically transformed to reduce skewness and heterogeneity prior to analyses. Concentrations of Ga, Mo, Rb, Se, and Sr were left untransformed.

To investigate length/element relationships within-species, linear regressions were performed on individual element concentrations vs. length for each shark species separately.

Element concentrations between species were compared using univariate and multivariate statistics. Univariate analyses (general linear methods GLM, Type III Sum of Squares, SAS, version 8.2, Cary, NC) were performed on absolute element concentrations to determine the effects of individual element concentrations as a function of species and sex of the shark, and to determine any interaction between species and sex (GLM: ELEMENT = SPECIES + SEX + SPECIES × SEX). Length was left out of the GLM expression because linear regression analysis did not reveal any significant relationships between element concentration and length for either species, and also because length and species were confounding variables, due to the almost exclusive separation of the generally larger Greenland sharks and smaller Pacific sleeper sharks into two size classes. It should also be noted that because all Greenland sharks were collected in Cumberland Sound and all Pacific sleeper sharks were collected in Prince William Sound, the variable "species" could have also been defined as "location" in the GLM expression.

Multivariate ordinations (Principal component analyses PCA, CANOCO 4.5 for Windows, Ter Braak and Šmilauer, 1998) were performed both on logarithmically transformed absolute element concentrations and on element pattern (sample standardized by norm, similar to [element]/[ΣElements]). ΣElements only include the 13 elements quantified above the DL. The PCA assigns scores to the individual samples that are linear combinations of 13 elements, and are presented relative to their ordination axes (Fig. 2). Elements are presented as arrows pointing to the direction of increasing value. Details for diagram interpretation are described elsewhere (Ter Braak, 1995; Van Wijngaarden et al., 1995; Van den Brink and Ter Braak, 1999).

Molar Hg and Se relationships were investigated more closely, due to their known biochemical relationship (Dietz et al., 2000). Hg and Se concentrations (μg/g, ww) were converted to nmol/g as explained in Dietz et al. (2000). The means and standard deviation of molar Se:Hg ratios were calculated for each shark.

3. Results

Among all shark samples, Zn had the highest mean concentration, followed by As (Table 1). Means of element concentrations that were below detection limit, along with the number of samples above detection limit, are shown in Table 2. The within-species comparisons (linear regression) revealed that no element concentration was related to shark length for either the Greenland sharks ($r^2 = 0.0009–0.0856$, $p = 0.291–0.890$),

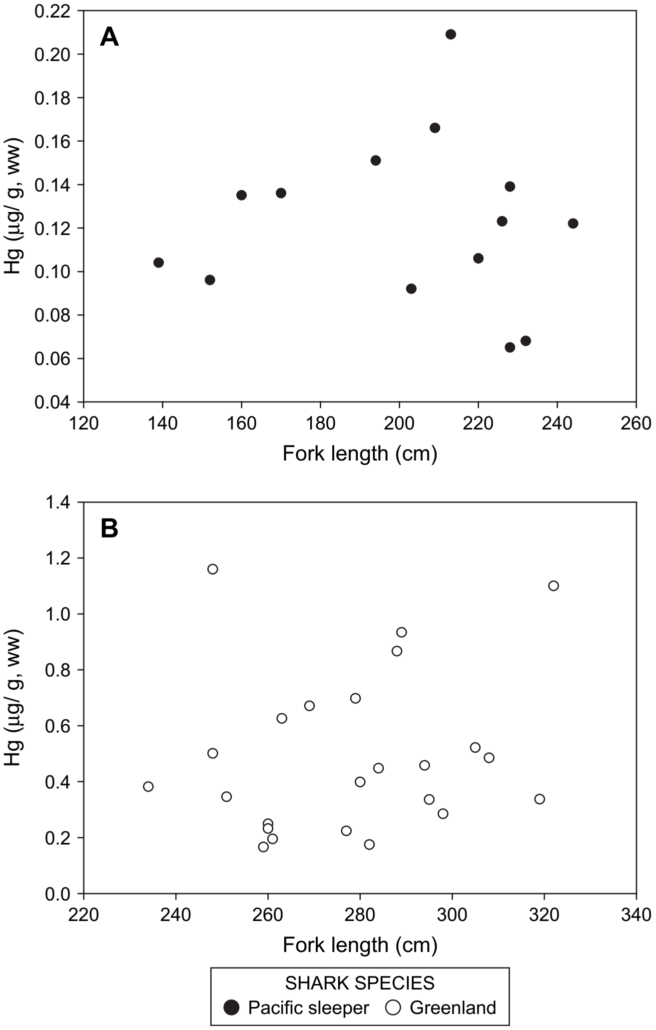

Fig. 2. Hg vs. fork lengths of individual (A) Pacific sleeper sharks (black circles) from Prince William Sound and (B) Greenland sharks (white circles) from Cumberland Sound.

which had a fork length range of 234–322 cm, or the Pacific sleeper sharks ($r^2 = 0.0076$–0.2320, $p = 0.088$–0.767), which had a fork length range of 139–244 cm. The Hg vs. length relationship is shown in Fig. 2A and B.

The GLM (univariate statistics) revealed sex was only significant for Rb ($F_{1,34} = 4.98$, $p = 0.0323$), with higher levels in females than males (0.44 ± 0.11, 0.38 ± 0.07 µg/g, ww, respectively), and there was no interaction between species and sex for any element. Several elements differed between the species based on GLM analysis of concentrations (Table 1): the non-essential elements Ag, As, Cd and Hg ($F_{1,34} = 4.47$–56.35, $p \leq 0.0001$–0.0420) and the essential element Cu ($F_{1,34} = 13.17$, $p = 0.0009$) were higher in Greenland sharks (Cumberland Sound) than Pacific sleeper sharks (Prince William Sound). Concentrations of the non-essential element Rb ($F_{1,34} = 6.05$, $p = 0.0192$), and the essential elements Co, Mn and Zn ($F_{1,34} = 6.35$–122.72, $p \leq 0.0001$–0.0166) were higher in Pacific sleeper sharks (Prince William Sound) than Greenland sharks (Cumberland Sound). The other elements (essential: Ga, Sr; non-essential: Mo, Se) did not differ between the species (all $p > 0.05$).

The PCA (multivariate statistics) of absolute element concentrations (Fig. 3A) provided similar results to the GLM analysis. One exception is that Zn did not differ between species based on the PCA of concentrations (Fig. 3A) but was significantly higher in Pacific Sleeper sharks based on the GLM results. The PCA of element concentrations also revealed four Pacific sleeper sharks that grouped out with the Greenland sharks, but overall, the variance of element concentrations between the species that were suggested by the GLM are supported by the PCA of absolute element concentrations.

The PCA of element pattern (Fig. 3B) revealed two clearly separated groups with less variability between the species than the PCA of element concentrations (Fig. 3A), as the four Pacific sleeper sharks observed to separate out in the PCA of element concentrations were not apparent in the analysis of pattern. PC1 and PC2 explained 95.5% of the total variability in element concentrations among samples on the PCA of pattern, and results were similar to the results of GLM, as Greenland sharks separated out due to higher Cd, Ag, Hg, As (non-essential elements) and Cu (essential element) concentrations, and Pacific sleeper sharks separated out due to higher concentrations of Zn, Co, Mn (essential elements) and Rb (non-essential element).

Table 1
Means (standard errors) for selected concentrations of elements (µg/g, ww) in liver tissue of Pacific sleeper sharks collected about Prince William Sound and Greenland sharks collected about Cumberland Sound

Species	*n*	Essential elements						
		Co*	Cu*	Mn*	Mo	Se	Zn*	
Pacific sleeper shark	14	0.027	0.833	0.427	0.039	0.542	10.39	
		(0.004)	(0.071)	(0.023)	(0.003)	(0.026)	(0.708)	
Greenland shark	24	0.018	1.754	0.179	0.033	0.516	6.898	
		(0.001)	(0.202)	(0.009)	(0.002)	(0.029)	(0.338)	
		Non-essential elements						
Species	*n*	Ag*	As*	Cd*	Ga	Rb*	Sr	Hg*
Pacific sleeper shark	14	0.048	5.364	2.638	0.005	0.454	0.241	0.122
		(0.014)	(0.149)	(0.354)	(2E-04)	(0.026)	(0.011)	(0.010)
Greenland shark	24	0.198	9.815	3.913	0.005	0.383	0.206	0.492
		(0.022)	(0.702)	(0.436)	(2E-04)	(0.016)	(0.200)	(0.058)

Elements marked with an asterisk (*) are significantly different between species at $p < 0.05$.

Table 2
Means and standard errors (SE) for concentrations of elements (μg/g, ww) in liver tissue of Pacific sleeper sharks collected about Prince William Sound and Greenland sharks collected about Cumberland Sound that were excluded from statistical analyses

Element	DL	Pacific sleeper sharks $n = 14$			Greenland sharks $n = 24$		
		n above DL	Mean	SE	*n* above DL	Mean	SE
Ba	0.005	11	0.015	0.03	13	0.011	0.002
Cr	0.050	0	–	–	0	–	–
La	0.001	0	–	–	11	0.001	0
Li	0.005	6	0.006	0	7	0.005	0
Ni	0.010	2	0.020–0.150	–	7	0.043	0.016
Pb	0.002	2	0.002–0.150	–	5	0.017	0.009
Pt	0.001	7	0.001	0	21	0.001	0
Sb	0.001	4	0.001	0	4	0.001	0
Tl	0.001	0	–	–	0	–	–
U	0.001	0	–	–	13	0.001	0
V	0.002	5	0.010	0.002	17	0.010	0.001

The detection limits (DL) (μg/g) and the number of samples above the DL, which were included in the mean calculation, are provided.

The mean Se:Hg molar ratio in the Pacific sleeper shark (12.36 ± 5.46) was higher than that in the Greenland shark (3.26 ± 1.45).

4. Discussion

4.1. Univariate and multivariate analysis of elements

Due to the increased risk of type II error (failure to reject a false null hypothesis) that occurs when individual elements are analyzed separately using repeated univariate analyses (Sokal and Rohlf, 1981), this study included both univariate (GLM) and multivariate (PCA) analyses. Univariate and multivariate analyses of element data in Greenland sharks and Pacific sleeper sharks yielded generally similar results, but the PCA of element concentrations did identify some differences in elements among four individual Pacific sleeper sharks. The variability in these individuals could potentially be explained by variable feeding events of the sharks, such as occasional cetacean/pinniped predation or scavenging (Smith and Baco, 2003; Schaufler et al., 2005). However, further research is needed to determine the relationship between shark element concentrations and feeding ecology before this conclusion can be made. It should also be noted that interpreting feeding behavior and bioaccumulation trends of elements based solely on hepatic element concentrations should be done with caution due to the high turnover and metabolic activity of liver tissue. The PCA of element pattern is useful because it normalizes the data and minimizes the effect of individual element concentrations, and relative to the concentrations PCA, the pattern PCA more clearly separates the samples into two groups based on either species identity or location. Thus the results of this study demonstrate that multivariate statistics can be a useful tool in element studies, often identifying underlying variability that is not obvious from univariate analysis, but also providing a means to use all element data in analyses when there is insufficient samples sizes for univariate statistics. Similar results and suggestions were made for statistical analysis of elements in seabirds (Borgå et al., 2006).

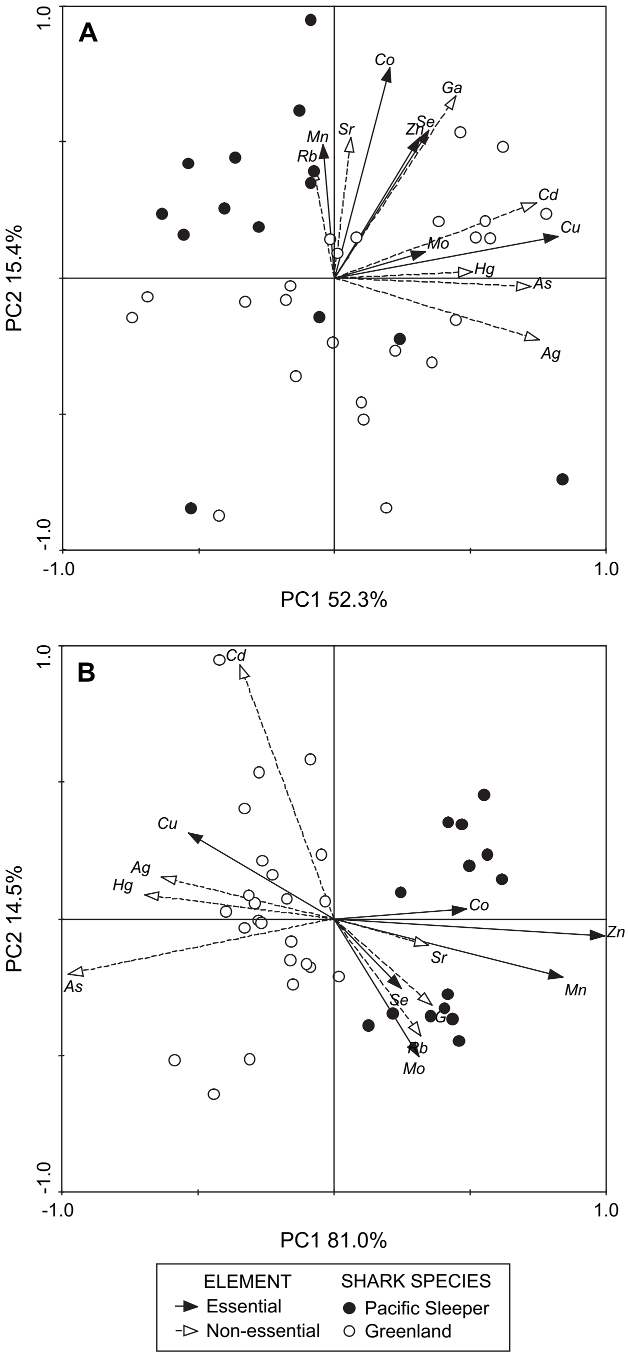

Fig. 3. Biplot of Prince William Sound Pacific sleeper sharks' (black circles) and Cumberland Sound Greenland sharks' (white circles) scores on the principal components (PC) extracted by principal component analyses (PCA), and the elements' loadings on the PCs. (A) Absolute element concentrations (log mg/kg) and (B) standardized element concentrations (pattern) in liver tissue. Symbols are individual sample's scores, and arrows are the respective element pointing in the direction of increasing value. Only elements that correlated more than 20% are shown in the ordination plot, as the others do not contribute much to sample separation in the ordination diagram.

4.2. Element/length relationships within the sleeper shark species

Linear regression analysis did not reveal any significant relationships between hepatic element concentrations and shark length, although element/length relationships have been previously demonstrated in other fishes (Riget et al., 2000; Adams and Onorato, 2005; Muir et al., 2005). Certain non-essential elements, especially Hg (Campbell et al., 2005a) and to a lesser degree Rb (Campbell et al., 2005b), are known to increase with increasing trophic level and with increasing length in fishes (Adams and Onorato, 2005; Muir et al., 2005) and one shark species (Storelli and Marcotrigiano, 2002). This is likely because fish generally increase in trophic level as they grow larger and are able to feed on larger prey. The lack of a mercury/length relationship (Fig. 2A and B) in the Greenland and Pacific sleeper sharks could be a result of the limited size range of sharks represented in this study. However, the lack of a mercury/length relationship could also indicate that these sharks are not necessarily increasing in trophic level as they grow larger (at least not within the size ranges sampled in this study), which is supported by the known scavenging behavior of Greenland sharks and Pacific sleeper sharks. For example, the Greenland shark that had the highest Hg concentration (1.16 ug/g, ww), and is therefore assumed to be feeding at the highest trophic level, had a fork length of 248 cm, which is at the low end of the range and below the mean of Greenland shark fork lengths sampled in this study. The scattered Hg concentrations along the length axis could also be reflective of the known omnivory of the Greenland and Pacific sleeper sharks, and could indicate variable marine mammal scavenging events. For example, it has been suggested, based on organic contaminants levels, that the frequency of marine mammal predation varies between individual Greenland sharks (Fisk et al., 2002), which would further explain the lack of a linear increase in Hg with shark length. The lack of a mercury/length relationship could also indicate that Hg does not bioaccumulate in the tissues of Greenland and Pacific sleeper sharks, although this explanation is highly unlikely because Hg has repeatedly been demonstrated to biomagnify. The impact of trophic position on Hg concentrations within the Greenland sharks and Pacific sleeper sharks is probably isolated from contributions of environmental Hg concentrations because both species were collected entirely in the same respective areas (i.e., all Greenland sharks were collected in Cumberland Sound and all Pacific sleeper sharks were collected in Prince William Sound), such that the environmental exposure is assumed to be the same for all individual representatives of each species.

The lack of a relationship between concentrations of essential elements and fork lengths in either Greenland sharks or Pacific sleeper sharks suggests that the physiological requirements of essential elements do not vary with size, and therefore likely age, over the size ranges of sharks sampled in this study. Although the relationship between element requirements and physiology in fish is poorly understood, a negative relationship between Zn concentration and fish size has been previously observed, suggesting a decrease in Zn requirement with size/age (Halden et al., 2000; Zhang and Wang, 2005). As the documented symptoms of Zn deficiency in fish include depressed growth (Lovell, 1998), Zn could potentially be required in higher concentrations in smaller, rapidly growing fishes. In the Greenland and Pacific sleeper sharks, this trend either does not occur, or occurs in sharks smaller than the ones sampled in this study.

4.3. Differences in non-essential elements between the sleeper sharks

Based on similar results between the PCA and GLM, the concentrations of the non-essential elements Ag, As, Cd, Hg, and Rb differed between Greenland sharks (Cumberland Sound) and Pacific sleeper sharks (Prince William Sound), and all but Rb were higher in the Greenland sharks. Because non-essential elements are likely not regulated to a specific level, or regulated less efficiently than essential elements (Zhang and Wang, 2005), we suggest that variances in the non-essential element concentrations are indicative of environmental exposure differences between the locations. The higher concentrations of the biomagnifying element Hg in the Greenland sharks could also be attributed to their potentially higher trophic level relative to the Pacific sleeper sharks (Fisk et al., 2002; Schaufler et al., 2005). Cadmium, which has also been shown to exhibit trophic transfer in some species (Muir et al., 1992), was significantly higher in Greenland sharks from Cumberland Sound, which further suggests that the Greenland sharks are feeding at a higher trophic level than the Pacific sleeper sharks. However, higher concentrations of Hg and Cd in the Greenland sharks may also reflect greater concentrations of these elements in the environment where they were collected (i.e., Cumberland Sound) relative to where the Pacific sleeper sharks were collected (i.e., Prince William Sound). Cadmium concentrations increase from the western to the eastern North American arctic in marine mammals (Dietz et al., 1998), which is consistent with the trends seen in the Greenland and Pacific sleeper sharks. Spatial trends in Hg in the North American arctic are more variable (Dietz et al., 1998), and thus it is hard to assess differences in Hg levels in the sharks' ecosystems. Thus, a combination of a potentially higher trophic level and higher regional contaminants could explain the higher Cd and Hg concentrations in the Greenland sharks.

Higher concentrations of As and Ag were also found in the Greenland sharks, which is likely due to regional differences in exposure since these compounds have not been observed to biomagnify in food webs (Campbell et al., 2005a). Although Rb biomagnified in an arctic marine food web, but to a lesser degree than Hg (Campbell et al., 2005a,b), it was the only non-essential element that was higher in the smaller and potentially lower trophic level Pacific sleeper sharks. Rubidium therefore contradicts the trend observed in Hg, but this could be due to variation in Rb levels between locations, with higher levels in Prince William Sound than in Cumberland Sound. Unfortunately, information on Rb concentrations in the environment is scarce and conclusions about regional differences are not possible.

4.4. Differences in essential element concentrations between the sleeper sharks

The Greenland shark and Pacific sleeper shark were assumed to have similar physiologies and similar concentrations of essential elements in individuals representing both species were expected. It was shown in several arctic seabird species that the essential element concentrations in individuals belonging to the same species remained relatively constant across wide geographical distances (Borgå et al., 2006). In the Greenland and Pacific sleeper sharks, however, which belong to the same Genus but not the same species, only two essential element concentrations (Mo, Se) did not differ while the other four essential elements (Co, Cu, Mn, Zn) did differ between species. This suggests that either the metabolic regulation of essential elements is not highly efficient in these cold-blooded fishes, and that environmental levels determined the observed concentrations in the sharks, or that regulation is efficient, and the Greenland shark and Pacific sleeper shark have different physiological requirements. However, the lack of information and data for comparison, and the complicated behavior and fate of elements in the environment requires caution be taken when comparing element concentrations between species/locations.

4.5. Elements in sharks collected in arctic waters vs. sharks collected in more temperate waters

Of the few studies that have analyzed element concentrations in sharks, several report data for muscle tissue only (Domi et al., 2005; Turoczy et al., 2000) and cannot be used for comparisons to the hepatic element concentrations reported in this study because muscle and liver have been shown to vary in their element concentrations (Storelli et al., 2003; Campbell et al., 2005a; Borgå et al., 2006). Past studies that have analyzed hepatic tissue from sharks sampled about the coast of Argentina (Marcovecchio et al., 1991) and the Mediterranean Sea (Storelli et al., 2003; Storelli and Marcotrigiano, 2002), have generally reported higher mean concentrations of As, Cd, Cu, Hg, Pb, Se and Zn than those observed in the Greenland and Pacific sleeper sharks. For example, the Greenland sharks from Cumberland Sound and the Pacific sleeper sharks from Prince William Sound had lower mean hepatic Cd concentrations (3.9 ± 1.3 μg/g, ww and 2.6 ± 1.3 μg/g, ww, respectively) than three species of sharks sampled about the Argentinian coast: *Mustelus schmitti*, 5.92 ± 1.65; *Notorynchus* sp., 8.41 ± 0.32; *Schroederichthys bivius*, 7.93 ± 1.78 μg/g, ww (note that *S. bivius* is the current valid name for *Halaelurus bivius*) (Marcovecchio et al., 1991). A hammerhead shark species *Sphyrna zygaena*, from about the Mediterranean Sea, had an approximately five times higher hepatic Cd concentration (19.77 ± 1.29 μg/g, ww) than the sleeper sharks (Storelli et al., 2003). Similar to Cd results, the Greenland shark and Pacific sleeper shark had lower hepatic Hg concentrations than previously reported for sharks collected about Argentina, (*M. schmitti*: 0.79 ± 0.39; *S. bivius*: 2.26 ± 0.56; *Notorynchus* sp.: 2.11 ± 0.33 μg/g, ww) (Marcovecchio et al., 1991) and in *S. zygaena*, from the Mediterranean Sea (35.89 ± 3.58 μg/g, ww) (Storelli et al., 2003). The variance in Hg concentrations among the shark species collected about Argentina reported in Marcovecchio et al. (1991) could be a result of differences in feeding ecology or closer proximity to emission sources relative to the Greenland and Pacific sleeper sharks. For example, *S. bivius* and *Notorynchus* sp. feed on a higher percentage of fish and have higher Hg concentrations relative to *M. schmitti*, but because specific trophic positions were not reported, comparisons between the feeding ecology of the sleeper sharks to that of the sharks collected about Argentina are difficult. Size differences among shark species could also contribute to variance in element concentrations, but *S. zygaena* were of similar size to the sharks in this study and Marcovecchio et al. (1991) only report lengths for *M. schmitti*, which were much smaller than the Greenland and Pacific sleeper sharks.

Concerning essential elements, several studies have reported shark hepatic Zn concentrations (Marcovecchio et al., 1991; Storelli et al., 2003). Zn is of interest because it has been suggested to biomagnify (Campbell et al., 2005a,b), although it appears to be efficiently regulated by fish (Kraemer et al., 2005; Zhang and Wang, 2005). The variance in Zn concentrations observed in the food web could be attributed to different physiological requirements between invertebrates and vertebrates (i.e., higher Zn requirements in higher trophic level vertebrates), instead of biomagnification. Hepatic Zn concentrations in the Greenland shark and Pacific sleeper shark were compared with previously published hepatic concentrations of Zn in four different species of shark (Marcovecchio et al., 1991; Storelli et al., 2003) and in two mammal species (Campbell et al., 2005a) (Fig. 4). This comparison reveals a casual observation that the different species of sharks, and the sharks and mammals, have different tissue concentrations, and likely different physiological requirements of Zn. The Zn concentrations also appear to be similar within, but different among, orders of sharks (Squaliformes, Carcharhiniformes, Hexanchiformes). Although this is preliminary data and no conclusions can be drawn, the comparison is nonetheless interesting.

4.6. Hg–Se relationships in sleeper sharks

The mean Se:Hg molar ratios in the Pacific sleeper shark (12.36 ± 5.46) and Greenland shark (3.26 ± 1.45) reflect the similar Se concentrations between the two species (0.54 ± 0.03, 0.52 ± 0.03 μg/g, ww, respectively) and the higher Hg concentrations in the Greenland shark. A 1:1 molar ratio of Se:Hg has been suggested to occur due to the potential of Se to mitigate the toxicity of Hg, but is most often observed only in marine mammals with very high Hg concentrations (above approximately 2.0 μg/g, ww) (Braune et al., 1991; Dietz et al., 2000). Organisms with lower Hg concentrations relative to marine mammals, such as seabirds and fish, generally have excess concentrations of Se relative to Hg, resulting in Se:Hg ratios larger than 1 (Dietz et al., 2000; Campbell et al., 2005a; Borgå et al., 2006). For example, previously reported mean hepatic Se:Hg ratios include: 16.3 ± 15.2 for a species of cat shark

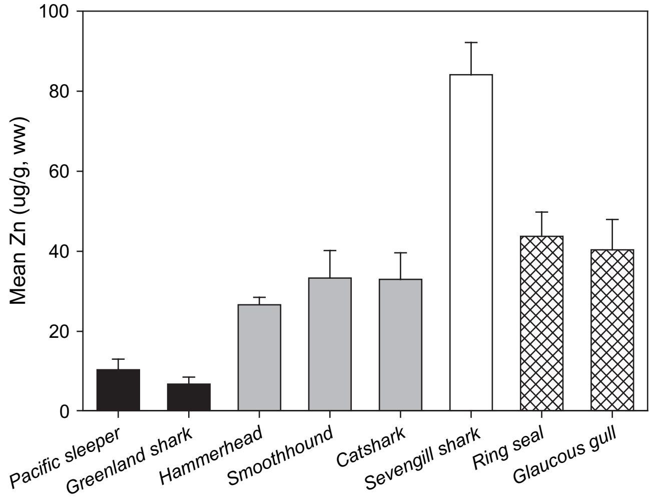

Fig. 4. A comparison of hepatic Zn concentrations (mean ± SD) among several shark species, including: Pacific sleeper shark (*Somniosus pacificus*), Greenland shark (*Somniosus microcephalus*), smooth hammerhead shark (*Sphyrna zygaena*) (Storelli et al., 2003), narrownose smooth-hound (*Mustelus schmitti*) (Marcovecchio et al., 1991), narrowmouthed cat shark (*Schroederichthys bivius*) (Marcovecchio et al., 1991), sevengill shark (*Notorynchus* sp.) (Marcovecchio et al., 1991); and two mammal species: ring seal (*Phoca hispida*) and glaucous gull (*Larus hyperboreus*) (Campbell et al., 2005a). The shark species belong to order Squaliformes (black bars), order Carchirhiniformes (grey bars), or order Hexanchiformes (white bar), and the ring seal and glaucous gull belong to the class Mammalia (hatched bars).

(*Galeus melastomus*) (Storelli and Marcotrigiano, 2002), 26.3 ± 44.1 for several species of seabird, and 198 ± 296 for teleost fishes (Dietz et al., 2000). However, a mean Se:Hg ratio of 1.14 ± 0.272 was reported for polar bears (Dietz et al., 2000), suggesting that high Hg concentrations (such as those observed in polar bears) induces a Se detoxification action, and that the Se:Hg ratio might tend towards 1:1 as Hg concentrations increase towards some species-specific threshold level (Storelli and Marcotrigiano, 2002). Although the Hg concentrations in the Greenland shark were substantially lower than the Hg concentrations previously reported for marine mammals and scavenging seabirds, the mean molar Se:Hg ratio in the Greenland shark is more similar to reported values for the ringed seal (*P. hispida*) (2.2 ± 2.8) and the glaucous gull (*Larus hyperboreus*) (3.7 ± 1.4) (Campbell et al., 2005a) than to values reported for other fish (Dietz et al., 2000) and one shark species (Storelli and Marcotrigiano, 2002). This relatively low Se:Hg ratio could indicate a low Hg threshold (i.e., a Se detoxification response that is potentially activated at low Hg concentrations) for the Greenland shark, even though its Hg concentration seems low relative to other mammals and southerly fishes and sharks. However, it is difficult to draw conclusions about shark physiology, as both the natural levels and "normal" concentrations of Se and Hg in the sleeper sharks are not known.

4.7. Comparison of Hg in sharks collected in arctic waters to other arctic species

Due to the potential toxicity of Hg and its ability to biomagnify, Hg concentrations have been analyzed in several arctic organisms. Mean Hg concentrations were higher in the Greenland shark (0.49 ± 0.06 μg/g ww) than in arctic cod, *Boreogadus saida*, liver samples collected about Greenland (0.015 ± 0.002 μg/g, ww) (Campbell et al., 2005a), and in other arctic areas summarized by Muir et al. (1992) (0.01 ± 0.01–0.04 ± 0.02 μg/g, ww), which is likely due to the higher trophic level of the sharks. However, the mean Hg concentrations in the Greenland and Pacific sleeper sharks were much lower than previously reported hepatic Hg concentrations for the ringed seal, *P. hispida*, (22.51 ± 16.87 μg/g, ww) and the scavenging glaucous gull, *L. hyperboreus*, (2.92 ± 1.06 μg/g, ww) collected about Greenland (Campbell et al., 2005a), despite the fact that these species probably all feed at similar trophic levels (Fisk et al., 2002). Lower concentrations of Hg in Greenland sharks compared to marine mammals and seabirds is a contradiction to what has been observed for biomagnifying organic contaminants such as PCBs and dichloro-diphenyl-trichloroethane (DDT), the levels of which are much higher in Greenland sharks compared to ringed seals (Fisk et al., 2002). This discrepancy is most likely due to varying half lives between Hg and organic contaminants and provides insights on the feeding ecology of the shark species. Organic contaminants, such as PCBs, would have very long half lives, potentially decades, in large cold water fish species like Greenland sharks and Pacific sleeper sharks (Fisk et al., 2002), which are likely much longer than for Hg. Thus the occasional predation or scavenging of a marine mammal would result in a significantly higher exposure and accumulation of Hg and organic contaminants than from feeding on invertebrates or fish, and the increased concentrations of the organic contaminants would remain in the sharks much longer than the Hg. Thus, lower Hg concentrations in the shark compared to the seals and seabirds suggest that on average Greenland sharks feed at a lower trophic level, but higher organic contaminants levels suggest that at times the sharks feed at a higher trophic level. Concentrations of mercury in the liver of flathead sole, *Hippoglossoides elassodon*, (0.32 ± 0.1 μg/g, dw) (Meador et al., 2005) collected about Prince William Sound are similar to Hg concentrations in Pacific sleeper shark livers (0.12 ± 0.01 μg/g, ww), if the liver of the flathead sole is assumed to be approximately 50% water and most of the Hg in fish is assumed to be methylmercury. Concentrations of Hg in livers of harbor seals, *Phoca vitulina*, (range = 0.4–72 μg/g, ww, *n* = 23) from the Gulf of Alaska/Prince William Sound area (Miles et al., 1992) are much higher than those of Pacific Sleeper sharks. These results suggest that the Pacific sleeper sharks feed at a lower trophic level than seals, even though Pacific sleeper sharks occasionally consume marine mammals (Bright, 1959). However, evidence from fatty acids suggests that the marine mammals consumed by Pacific sleeper sharks are baleen whales, which are likely at a lower trophic level than piscivorous seals (Schaufler et al., 2005).

5. Summary

This study presents the first element data reported for the only two shark species known to regularly inhabit arctic waters. The within-species comparison revealed that no non-essential element concentration was related to length in either the Greenland shark or the Pacific sleeper shark, which could suggest a lack of trophic level increase in sharks within the size range sampled in this study. No changes in physiological requirements with size were apparent based on the lack of significant essential element vs. length relationships in either species. Univariate and multivariate statistics suggested that the Greenland sharks had higher levels of certain non-essential elements relative to the Pacific sleeper sharks, likely due to exposure variances (environmental and dietary). The variance in essential elements between the species could suggest that the Greenland and Pacific sleeper sharks have different physiological requirements. The Greenland and Pacific sleeper sharks had lower non-essential and essential element concentrations than those previously reported for sharks inhabiting lower latitudes. However, the mean molar Se:Hg ratio in the Greenland shark was lower than that observed in more southerly fishes, which could indicate a lower Hg threshold for the Greenland shark relative to other fishes. Baseline element data is important because the Greenland shark and Pacific sleeper shark are likely ecologically significant in the arctic marine ecosystem, and could be sentinel species based on their feeding behavior. Further, very little element data exists for sharks and there is a need to better understand the trophic transfer, assimilation, and elimination of both non-essential and essential elements, which could be useful in not only addressing concerns over toxicity, but could also help in answering basic questions about shark life history and physiology.

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References

Adams, D.H., Onorato, G.V., 2005. Mercury concentrations in red drum, *Sciaenops ocellatus*, from estuarine and offshore waters of Florida. Marine Pollution Bulletin 50 (3), 291–300.

Benz, G.B., Hocking, R., Kowunna Sr., A., Bullard, S.A., George, J.C., 2004. A second species of Arctic shark: Pacific sleeper shark *Somniosus pacificus* from Point Hope, Alaska. Polar Biology 27, 250–252.

Bigelow, H.B., Schroeder, W.C., 1948. Sharks. In: Tee-Van, J., Breder, C.M., Hildebrand, S.F., Parr, A.E., Schroeder, W.C. (Eds.), Fishes of the Western North Atlantic. Part 1. Lancelets, Cyclostomes, Sharks. Memoir Sears Foundation for Marine Research, New Haven, pp. 514–526.

Borgå, K., Campbell, L., Gabrielsen, G.W., Norstrom, R.J., Muir, D.C.G., Fisk, A.T., 2006. Regional and species specific bioaccumulation of major and trace elements in arctic seabirds. Environmental Toxicology and Chemistry 25, 2927–2936.

Braune, B.M., Norstrom, R.J., Wong, M.P., Collins, B.T., Lee, J., 1991. Geographical distribution of metals in livers of polar bears from the northwest territories, Canada. Science of the Total Environment 100, 283–299.

Bright, D.B., 1959. The occurrence and food of the sleeper shark, *Somniosus pacificus*, in a central Alaska Bay. Copeia 1, 76–77.

Campbell, L.M., Norstrom, R.J., Hobson, K.A., Muir, D.C.G., Backus, S., Fisk, A.T., 2005a. Mercury and other trace elements in a pelagic Arctic marine food web (Northwater Polynya, Baffin Bay). Science of the Total Environment 351-352, 247–263.

Campbell, L.M., Fisk, A.T., Wang, X., Köck, G., Muir, D.C.G., 2005b. Evidence of biomagnification of rubidium in aquatic and marine food webs. Canadian Journal of Fisheries and Aquatic Sciences 62, 1161–1167.

Compagno, L.J.V., 1984. Sharks of the world. An annotated and illustrated catalogue of shark species known to date, part 1: Hexanchiformes to Lamniformes. FAO Fisheries Synopsis, 125. In: FAO Species Catalogue of the World, vol. 4.

Dietz, R., Riget, F., Johansen, P., 1996. Lead, cadmium, mercury and selenium in Greenland marine animals. Science of the Total Environment 186, 67–93.

Dietz, R., Pacyna, J., Thomas, D.J., 1998. Heavy metals. In: AMAP Assessment Report: Arctic Pollution Issues. Arctic Monitoring and Assessment Program, Oslo, Norway, pp. 373–524.

Dietz, R., Riget, F., Born, E.W., 2000. An assessment of selenium to mercury in Greenland marine animals. Science of the Total Environment 245, 15–24.

Domi, N., Bouquegneau, J.M., Das, K., 2005. Feeding ecology of five commercial shark species of the Celtic Sea through stable isotope and trace metal analysis. Marine Envrionmental Research 60, 551–569.

Duffus, J.H., 2002. "Heavy metals" – a meaningless term? IUPAC Technical Report. Pure and Applied Chemistry 74 (5), 793–807.

Fisk, A.T., Tittlemier, S.A., Pranschke, J.L., Norstrom, R.J., 2002. Using anthropogenic contaminants and stable isotopes to assess the feeding ecology of Greenland shark. Ecology 83, 2162–2172.

Fisk, A.T., Muir, D.C.G., Hobbs, K., Borg, H., Braune, B., Burgess, N., Culhane, M., de Wit, C., Evans, M., Hickie, B., Hoekstra, P., Kuzyk, Z., Kwan, M., Lockhart, L., Macdonald, C., Norstrom, R., Outridge, P., Roach, P., Stern, G., Wayland, M., Stokker, Y., 2003. Contaminant levels, trends and effects in the biological environment, Part I. In: Fisk, A.T., Muir, D.C.G., Hobbs, K. (Eds.), Contaminant Levels, Trends and Effects in the Biological Environment. Canadian Arctic Contaminants Assessment Report II. Indian and Northern Affairs Canada, Ottawa.

Grosell, M., Wood, C.M., Walsh, P.J., 2003. Copper homeostasis and toxicity in the elasmobranch *Raja erinacea* and the teleost *Myoxocephalus octodecemspinosus* during exposure to elevated water-borne copper. Comparative Biochemistry and Physiology Part C: Toxicology and pharmacology 135, 179–190.

Halden, N.M., Mejia, S.R., Babaluk, J.A., Reist, J.D., Kristofferson, A.H., Campbell, J.L., Teesdale, W.J., 2000. Oscillatory zinc distribution in Arctic char (*Salvelinus alpinus*) otoliths: the result of biology or environment? Fisheries Research 46, 289–298.

Hansen, P.M., 1963. Tagging experiments with the Greenland Shark (*Somniosus microcephalus* (Bloch and Schneider)) in Subarea 1. International Commission for the Northwest Atlantic Fisheries Special Publication 4, 172–175.

Herdendorf, C.E., Berra, T.M., 1995. A Greenland shark from the wreck of the SS Central America at 2200 meters. Transactions of the American Fisheries Society 124, 950–953.

Kraemer, L.D., Campbell, P.G.C., Hare, L., 2005. Dynamics of Cd, Cu and Zn accumulation in organs and sub-cellular fractions in field transplanted juvenile yellow perch (*Perca flavescens*). Environmental Pollution 138, 324–337.

Lovell, R.T., 1998. Nutrition and Feeding of Fish, Second ed. Kluwer Academic Publishers, Boston.

Marcovecchio, J.E., Moreno, V.J., Perez, A., 1991. Metal accumulation in tissues of sharks from the Bahia Blanca estuary, Argentina. Marine Environmental Research 31, 263–274.

Meador, J.P., Ernest, D.W., Kagley, A.N., 2005. A comparison of the non-essential elements cadmium, mercury, and lead found in fish and sediment form Alaska and California. Science of the Total Environment 339, 189–205.

Miles, K.A., Calkins, D.G., Coon, N.S., 1992. Toxic elements and organochlorines in harbor seals (*Phoca vitulina richardsi*), Kodiak, Alaska, USA. Bulletin of Environmental Contamination and Toxicology 48, 727–732.

Muir, D.C.G., Wagemann, R., Hargrave, B.T., Thomas, D.J., Peakall, D.B., Norstrom, R.J., 1992. Arctic marine ecosystem contamination. Science of the Total Environment 122, 75–134.

Muir, D.C.G., Wang, X.W., Bright, D., Lockhart, L., Kock, G., 2005. Spatial and temporal trends of mercury and other metals in landlocked char from lakes in the Canadian Arctic archipelago. Science of the Total Environment 351, 464–478.

National Laboratory for Environmental Testing, 2003. NLET Schedule of Services. National Water Research Institute, Environment Canada, Burlington, Ontario.

Riget, F., Dietz, R., Johansen, P., Asmund, G., 2000. Lead, cadmium, mercury and selenium in Greenland marine biota and sediments during AMAP phase 1. Science of the Total Environment 245, 3–14.

Schaufler, L., Heintz, R., Sigler, M., Hulbert, L., 2005. Fatty acid composition of sleeper shark (*Somniosus pacificus*) liver and muscle reveals nutritional dependence on planktonivores. International Council for the Exploration of the Sea: Conference Manuscript N:05.

Skomal, G.B., Benz, G.W., 2004. Ultrasonic tracking of Greenland sharks, *Somniosus microcephalus*, under Arctic ice. Marine Biology 145, 489–498.

Smith, C.R., Baco, A.R., 2003. Ecology of whale falls at the deep-sea floor. Oceanography and Marine Biology 41, 311–354.

Sokal, R.R., Rohlf, F.J., 1981. Biometry. Freeman, New York.

Stokker, Y., 2003. Part II. Interlaboratory Quality Assurance for the Northern Contaminants Program. In: Fisk, A.T., Hobbs, K., Muir, D.C.G. (Eds.), Contaminant Levels, Trends and Effects in the Biological Environment. Canadian Arctic Contaminants Assessment Report II. Northern Contaminants Program, Department of Indian and Northern Affairs Canada, Ottawa, Canada, pp. 111–127.

Storelli, M.M., Marcotrigiano, G.O., 2002. Mercury speciation and relationship between mercury and selenium in liver of *Galeus melastomus* from the Mediterranean Sea. Bulletin of Environmental Contamination and Toxicology 69, 516–522.

Storelli, M.M., Ceci, E., Storelli, A., Marcotrigiano, G.O., 2003. Polychlorinated biphenyl, heavy metal and methylmercury residues in hammerhead sharks: contaminant status and assessment. Marine Pollution Bulletin 46, 1035–1048.

Taggart, S.J., Andrews, A.G., Mondragon, J., Mathews, E.A., 2005. Co-occurrence of Pacific sleeper sharks *Somniosus pacificus* and harbor seals *Phoca vitulina* in Glacier Bay. Alaska Fishery Research Bulletin 11 (2), 113–117.

Ter Braak, C.J.F., 1995. Ordination. In: Jongman, R.G.H., Ter Braak, C.J.F., Van Tongeren, O.F.R. (Eds.), Data Analysis in Community and Landscape Ecology. Cambridge University Press, Cambridge, UK, pp. 91–173.

Ter Braak, C.J.F., Šmilauer, P., 1998. CANOCO Reference Manual and User's Guide to CANOCO for Windows: Software for CANOCO Community Ordination. Version 4. Microcomputer Power, Ithaca, NY, USA.

Turoczy, N.J., Laurenson, L.J.B., Allinson, G., Nishikawa, M., Lambert, D.F., Smith, C., Cottier, J.P.E., Irvine, S.B., Stagnitti, F., 2000. Observations on metal concentrations in three species of shark (*Deania calcea*, *Centroscymnus crepidater*, and *Centroscymnus owstoni*) from Southeastern Australian Waters. Journal of Agricultural and Food Chemistry 48, 4357–4364.

Van den Brink, P.J., Ter Braak, C.J.F., 1999. Principal response curves: analysis of time-dependent multivariate responses of biological community to stress. Environmental Toxicology and Chemistry 18, 138–148.

Van Wijngaarden, R.P.A., Van Den Brink, P.J., Oude Voshaar, J.H., Leeuwangh, P., 1995. Ordination techniques for analyzing response of biological communities to toxic stress in experimental ecosystems. Ecotoxicology 4, 61–77.

World Health Organization, 1990. IPCS Environmental Health Criteria 101: Methylmercury. International Programme of Chemical Safety. World Health Organization, Geneva, Switzerland, 144 pp.

Yang, M., Page, B.N., 1999. Diet of Pacific sleeper shark, *Somniosus pacificus*, in the Gulf of Alaska. Fisheries Bulletin 97, 406–409.

Zhang, L., Wang, W., 2005. Effects of Zn pre-exposure on Cd and Zn bioaccumulation and metallothionein levels in two species of marine fish. Aquatic Toxicology 73, 353–369.