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CONCENTRATIONS AND INTERACTIONS OF SELECTED ESSENTIAL AND NON-ESSENTIAL ELEMENTS IN RINGED SEALS AND POLAR BEARS OF ARCTIC ALASKA

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ABSTRACT: In this study, we evaluated concentrations of twelve essential and non-essential elements (As, Cd, Co, Cu, Pb, Mg, Mn, Hg, Mo, Se, Ag, and Zn) in tissues of ringed seals (*Phoca hispida*) and polar bears (*Ursus maritimus*) of arctic Alaska (USA). All samples were collected between 1995–97 in conjunction with subsistence harvests. The essential elements are reported to help develop reference ranges for health status determination and to help assess known or suspected interactions affecting toxicoses of cadmium (Cd) and mercury (Hg). In some tissues, Cd, Hg, and selenium (Se) were present at concentrations that have been associated with toxicoses in some domestic animals. Nevertheless, tissue levels of all elements were within ranges that have been reported previously in other pinnipeds and polar bears. Significant associations included: Cd with Zn or Cu; Cu with Zn or Ag; and Hg with Se, Zn, or Cu. This study found hepatic Hg:Se molar ratios to be lower than unity and different between the two species. Based upon significant differences in mean tissue elemental concentrations for polar bear versus ringed seal, we concluded that biomagnification factors (bear/seal) were significant for: Cu in liver and muscle; Pb in kidney; Se in kidney and muscle; Zn in liver and muscle; and Hg in liver. Possible explanations for observed elemental correlations (i.e., interactions) and ancillary mechanisms of Cd and Hg detoxification are discussed.

Key words: Cadmium, elements, marine mammals, mercury, molar ratio, *Phoca hispida,* polar bear, ringed seal, selenium, *Ursus maritimus.*

INTRODUCTION

As described in Woshner et al. (2001), the Arctic has little human habitation, industry, or agriculture. Apprehension among Native people who consume pinnipeds and polar bear is in part due to the detection of anthropogenic contaminants (Wheatley and Parodis, 1995) in subsistence foods. Metals can accumulate in biotic and abiotic matrices (Pacyna and Winchester, 1990), and although cadmium (Cd) and mercury (Hg) occur naturally, human activities have increased the rate at which they and other elements are mobilized to and within the biosphere (Pacyna and Keeler, 1995). Cadmium and Hg occur at apparently elevated concentrations in marine mammals as compared to some domestic and laboratory animals. Consequently, they warrant investigation as potential risks to marine mammals or human consumers (Muir et al., 1992; Macdonald and Bewers, 1996). Some arctic species, especially marine mammals, may have evolved proficient physiologic mechanisms to detoxify and tolerate comparatively high concentrations of some potentially harmful metals (Dietz et al., 1998). Concentrations of metals in marine mammals, mechanisms of their accumulation and toxicity, as well as their elemental interactions with biological or ecological factors are poorly understood.

Findings of Koeman et al. (1973, 1975) and the association between Hg and Se levels in liver and brain of marine mammals are presented in Woshner et al. (2001). This relationship may serve as a protective mechanism against toxic effects of Hg by biotransformation of ingested methylmercury to a less toxic chemical form. Koeman's theory (Koeman et al., 1975) regarding equimolar Hg:Se ratios has been corroborated by data on tissues of polar bear and various pinnipeds (Smith and Armstrong, 1978; Braune et al., 1991; Dietz et al., 1995) with some variation

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from a 1:1 molar ratio noted (Wagemann et al., 1983; Hansen et al., 1990; Braune et al., 1991). Departures from unity have been interpreted as evidence that the mechanism for Hg detoxification has been overwhelmed or is not Se-dependent.

Dietz et al. (1995) noted that Hg and Cd tended to increase with age in liver and kidney of polar bears; and Zn and Se were positively correlated with age in kidney and liver, respectively. Cadmium levels in liver and kidney were lower in Greenlandic polar bears than in seals from the same region, and Hg in liver and kidney was significantly higher in bears with biomagnification factors (BMFs, bear/seal) of 1.99 and 11.6, respectively. Moreover, the mode of Hg accumulation in polar bears may differ from that of other marine mammals, with Hg reaching higher concentrations in kidney than in liver (Dietz et al., 1995).

In this study, we compare metal levels and interactions in ringed seals and polar bears of Alaska. Major concerns regarding element levels in these species include potential toxic effects to wildlife and human consumers. We present what may be considered "normal" or reference ranges of essential elements in apparently healthy (as determined by gross and histologic examination of tissue; Woshner, 2000) subsistence-harvested animals. This "normal range" can be used when a mineral deficiency is suspected, for comparison to other species, and to identify interactions that may counter toxicoses (i.e., Se and Hg). A better understanding of essential elements in arctic pinnipeds and polar bears will help to improve health assessments and disease investigations.

The objectives of this study were to (1) evaluate concentrations of selected essential and non-essential elements in tissues of ringed seals and polar bears harvested by Native Alaskan subsistence hunters; (2) to determine whether these element levels differ with tissue type, gender, species, etc.; (3) evaluate elemental interactions; and (4) report "normal ranges" of essential elements.

MATERIALS AND METHODS

General

During 1995–97, subsistence-harvested polar bears (n = 24) and ringed seals (n = 17) were opportunistically sampled in the Barrow (71°17′N, 156°47′W), Alaska area. Tissues we collected were liver, kidney, muscle, and blubber (subcutaneous fat of polar bears). Tissues were processed as in Woshner et al. (2001a). Frozen samples were express delivered (24–48 hr) to College Station, Texas (USA) for archiving and analyses under appropriate National Marine Fisheries Service (Washington, D.C., USA) permits or U.S. Fish and Wildlife Service (Washington, D.C.) letters of authorization.

Elemental analyses

Tissue digestion and metals analysis procedures, including limits of detection, have been described (Woshner et al., 2001). However, it is worth noting here that total Hg was determined by two methods: Total Hg measured directly (THg) and/or calculated total Hg (SHg), determined by summing quantities of monomethylmercury (MHg) and divalent Hg [Hg(II)] (i.e., SHg = Hg(II) + MHg] (Woshner et al., 2001). All elemental data were reported on a wet weight (ww) basis.

Statistical analysis

Student's t-test was used to compare hepatic SHg concentrations and SHg:Se molar ratios between species, to test hepatic SHg:Se ratios against a theoretical 1:1 ratio, and to compare elemental concentrations between genders within species. With the exception of renal Cu in ringed seals, significant differences were not observed between genders, so data were pooled for analysis. One-way Analysis of Variance (ANOVA) with Tukey's post-hoc test tested for differences in elemental concentrations between tissues within species, and between species within tissue, with ringed seals and polar bears of this study analyzed in conjunction with two cetacean species (Woshner et al., 2001). All statistical analyses were done using Systat (SPSS, Inc., Chicago, Illinois USA; 1998 version) computer software with a P-value of <0.05 considered significant, as in Woshner et al. (2001). Biomagnification factors (BMFs) were determined by dividing the mean concentration of a given element in a specified tissue of polar bears by that in ringed seals.

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TABLE 1. Concentrations (μg/g ww) of selected elements in tissues of Alaskan ringed seals.

	As	Cd	Co	Cu	Pb	Mg	Mn	THg	Mo	Se	Zn
Liver[a]											
Mean	0.11[1]	5.27[1]	0.04[1]	9.06[1]	0.03[1]	354.8[1]	7.14	3.52[1]	0.74[1]	7.16[1]	49.38[1]
SD[b]	0.07	3.21	0.03	3.98	0.04	73.4	1.87	5.07	0.17	5.77	9.40
n	17	17	17	17	17	17	17	16	17	17	17
Kidney											
Mean	0.14[1]	26.01[2]	0.06[1]	7.84[1]	0.01[1]	254.7[2]	2.84[2]	0.50[2]	0.20[2]	2.51[2]	46.36[1]
SD	0.05	15.59	0.03	4.53	0.02	47.9	1.06	0.28	0.04	0.97	12.86
n	17	17	17	17	17	17	17	16	17	17	17
Muscle											
Mean	0.09[1]	0.09[3]	0.01[2]	1.33[3]	0.01[1]	367.1[1]	0.37[3]	0.22[2]	—	0.28[2]	35.38[2]
SD	0.03	0.06	0.00	0.38	0.01	53.5	0.08	0.33		0.09	8.76
n	11	11	11	11	11	16	11	11	11	16	16
Blubber											
Mean	1.44[2]	0.02[3]	0.01[2]	0.38[2]	0.01[1]	30.3[3]	0.07[3]	0.002[2]	—	0.21[2]	3.75[3]
SD	0.49	0.01	0.01	0.18	0.01	4.34	0.03	0.001		0.11	0.94
n	16	11	11	16	11	11	11	11	11	16	11

[a] Liver levels (SD) of Ag, Hg(II), MHg, and SHg were 0.12 (0.13), 2.62 (3.11), 0.15 (0.06), and 2.77 (3.12) μg/g ww.
[b] Standard deviation.
The same superscript number ([1,2,3]) indicates mean level for that element did not differ significantly among tissue(s) as determined by one-way ANOVA with Tukey's post-hoc test.

RESULTS

Ringed seals

With respect to comparisons of weight, body length and individual tissue elemental concentrations, no significant differences were observed between sexes except renal mean Cu level was greater in males (10.34 μg/g ww) than females (5.66 μg/g). Mean As was highest in blubber (1.44 μg/g ww) as compared to other tissues (Table 1). Mean Cd levels were highest in kidney (26.01 μg/g ww), followed by liver (5.72 μg/g ww) (Table 1). Mean Se and Hg concentrations were greatest in liver (7.16 μg/g for Se, 3.53 μg/g for Hg), followed by kidney (2.51 μg/g for Se, 0.50 μg/g for Hg) (Table 1). Mean levels of Co, Cu, Pb and Zn were not significantly different in liver versus kidney; but Mg, Mn and Mo were significantly higher in liver (Table 1). In general, hepatic calculated total mercury [SHg, which is the sum of divalent mercury, Hg(II), and methylmercury, MHg] was lower than total mercury measured directly (THg) by a mean difference of 0.75 μg/g, although not necessarily on a case-by-case basis. The mean fraction of THg accounted for by hepatic MHg was 10.8%, with a range of 1.1% to 25.4%. Additionally, %MHg in liver was negatively correlated with: body weight ($r = -0.52$, $P = 0.04$); THg ($r = -0.75$, $P = 0.001$); Hg(II) ($r = -0.84$, $P < 0.001$); and SHg ($r = -0.82$, $P < 0.001$). Significant correlations between variables within tissue are given in Table 2. Linear regression parameters of Se versus various forms of Hg (μmol/Kg), and of Se versus the summed molar quantity of THg and Ag are shown in Table 3. Among these regressions, all those in liver and that for Se vs. total Hg in kidney were significant, while all regressions in muscle and blubber were not. In terms of correlation coefficients associated with linear regressions in which Se was the dependent variable, the highest r^2 values observed were for hepatic tissue: 0.80, 0.80, and 0.78, associated with the independent variables THg + Ag (μmol/Kg), THg (μmol/Kg), and HgD (THg − SHg), respectively (Table 3). The mean THg:Se ratio in livers of ringed seals was 0.14 (stan-

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TABLE 2. Pearson's correlation coefficients between selected significantly correlated ($P < 0.05$) variables[a] in Alaskan ringed seals.

	r	P	n		r	P	n
Liver				Kidney			
Lgth[b] vs[c] Wgt[d]	0.90	<0.001	16	Cu vs Wgt	−0.54	0.031	16
Hg(II) vs Wgt	0.53	0.034	16	Cd vs THg	0.64	0.007	16
Hg(II) vs THg	0.94	<0.001	16	Cd vs Zn	0.87	<0.001	17
SHg vs Wgt	0.55	0.028	16	Se vs THg	0.59	0.016	16
SHg vs THg	0.95	<0.001	16	THg vs Zn	0.71	0.002	16
SHg vs Hg(II)	1.0	<0.001	16	Blubber			
Se vs THg	0.67	0.004	16	Cu vs As	0.65	0.006	16
Se vs Hg(II)	0.55	0.027	16	Se vs As	0.59	0.015	16
Se vs SHg	0.59	0.017	16	Se vs Cu	0.81	<0.001	16
Ag vs Cu	0.82	<0.001	17	Muscle			
Ag vs THg	0.59	0.015	16	THg vs Wgt	−0.63	0.04	17
Zn vs As	0.64	0.006	17	THg vs Lgth	−0.64	0.034	17
Zn vs Cd	0.55	0.022	17	Se vs Cu	0.55	0.027	16

[a] Data were ranked prior to analysis.
[b] Lgth is body length, tip of snout to base of tail.
[c] vs = versus.
[d] Body weight.

dard deviation = 0.11; range 0.01 to 0.40; $n = 16$), which was significantly different from 1:1 (Student's $t = -30.663$; $P < 0.001$).

Polar bears

Mean As levels were relatively low in all tissues; the highest mean As concentration occurred in liver (0.09 μg/g ww), followed by subcutaneous fat (0.07 μg/g ww) (Table 4). The mean Cd concentration in kidney was 8.69 μg/g ww, which was at least 15 times greater than in the next highest tissue, liver (Table 4). Renal tissue also had the highest mean concentration of Pb (0.29 μg/g ww). The mean SHg in liver was 14.22 μg/g ww with a mean of 5.5% in the form of MHg. In muscle, SHg averaged 0.09 μg/g with a mean level of 72.7% present as MHg. Element interactions based on correlations between variables within tissues are given in Table 5. Linear regression parameters of Se versus various forms of Hg and of Se versus the summed molar quantify of total Hg and Ag are shown in Table 3. All of these simple regressions were significant in both liver and muscle. The hepatic mean SHg:Se molar ratio in polar bears was 0.68 (standard deviation = 0.28; range 0.17 to 1.46; $n = 24$), which was significantly different from unity (Student's $t = -5.651$; $P < 0.001$). In summary, mean Cu, Mn, Mo, Ag, Zn, and As levels were highest in liver versus kidney; Cd, Co, Pb were higher in kidney, and Se was not significantly different (Table 4). Magnesium was greatest in muscle.

Species comparison

Mean Cu and Zn levels in liver and muscle were significantly greater in polar bears than in ringed seals. Cadmium in liver, kidney and muscle of ringed seals was significantly greater than in polar bears. Renal Cu and Zn were significantly greater in ringed seals, while renal Pb was greater in polar bears. Mean Se in polar bear muscle and kidney was significantly higher than in ringed seals. Mean SHg levels were significantly higher in liver of polar bears than in ringed seals, and hepatic mean SHg:Se ratio also was significantly higher in polar bears (Student's $t = -8.705$; $P < 0.001$). Biomagnification factors (BMFs, with * indicating a factor with

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TABLE 3. Linear regression parameters for Hg[a], Ag[a] and Se[a] in tissues of Alaskan ringed seals and polar bears.

Tissue	Variables	*n*	β_1-Coeff[b]	r^2	*P* (regression)
Ringed Seal					
Liver	Se[a] vs[c] THg[a]	16	2.62	0.803	<0.001
	Se[a] vs THg[a] + Ag[a]	16	2.54	0.804	<0.001
	Se (ppm) vs HgD[d]	16	1.49	0.781	<0.001
Kidney	Se[a] vs THg[a]	16	2.65	0.361	0.014
Polar bear					
Liver	Se[a] vs SHg[a,e]	24	1.58	0.380	0.001
	Se[a] vs SHg[a,e] + Ag[a]	24	1.55	0.376	0.001
	Se[a] vs Hg(II)[a]	24	1.68	0.401	0.001
	Se[a] vs MHg[a]	24	34.88	0.192	0.032
Muscle[d]	Se[a] vs SHg[a,e]	23	3.21	0.372	0.002
	Se[a] vs Hg(II)[a]	23	5.69	0.292	0.008
	Se[a] vs MHg[a]	23	3.86	0.250	0.015

[a] Expressed on a molar basis (μmol/Kg).
[b] β_1 coefficient.
[c] vs = versus.
[d] Mercury level difference, HgD = total Hg (THg) − sum Hg (SHg) (all in ppm).
[e] SHg was used rather than THg which was not determined for polar bears.

TABLE 4. Concentrations (μg/g ww) of selected elements in tissues of Alaskan polar bears.

	As	Cd	Cu	Pb	Mg	Mn	Se	Ag	Zn	HgII[b]	MHg[c]	SHg[d]
Liver[a]												
Mean	0.09^1	0.47^1	30.0^1	0.08^1	371.0^1	5.13^1	9.33^1	0.17^1	78.64^1	13.21^1	0.49^1	14.22^1
SD[e]	0.06	0.22	10.00	0.07	50.89	0.89	13.00	0.08	19.11	12.43	0.42	12.92
n	24	24	24	24	24	24	24	24	24	24	24	24
Kidney[a]												
Mean	0.02^2	8.69^2	3.39^2	0.29^2	247.0^2	1.80^2	12.99^1	0.01^2	39.60^2	NA[g]	NA[g]	NA[g]
SD	0.03	5.05	0.71	0.19	46.82	0.53	10.58	0.01	8.71	—	—	—
n	19	24	24	24	24	24	24	19	24	—	—	—
Muscle												
Mean	ND[f]	0.01^3	2.97^2	0.02^1	495.3^3	0.28^3	0.54^2	ND[f]	64.08^3	0.03^2	0.07^2	0.09^2
SD		0.02	1.01	0.02	103.5	0.08	0.15		10.94	0.04	0.05	0.07
n	18	18	23	23	23	23	23	18	23	23	23	23
Blubber												
Mean	0.07^1	ND[f]	0.27^3	ND[f]	27.94^4	0.05^4	0.04^2	0.01^2	1.96^4	NA[g]	NA[g]	NA[g]
SD	0.03		0.13		9.64	0.02	0.02	0.00	1.08	—	—	—
n	11	11	14	14	14	11	11	11	14	—	—	—

[a] Co levels (SD) were 0.01 (0.01) and 0.05 (0.03) in liver and kidney, respectively; Mo levels (SD) were 0.83 (0.23) and 0.39 (0.08) in liver and kidney, respectively.
[b] Divalent (+2) mercury.
[c] Monomethylmercury.
[d] Sum of Hg = Hg(II) + MHg.
[e] Standard deviation.
[f] Not detected in greater than 50% of the samples.
[g] Not analyzed.
The same superscript number (^{1,2,3}) indicates mean level for that element was not significantly different among tissue(s) as determined by one-way ANOVA with Tukey's post-hoc test.

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TABLE 5. Pearson's correlation coefficients between selected significantly correlated variables[a] ($P < 0.05$) in Alaskan polar bears.

	r	P	n		r	P	n
Liver				Liver			
Cu vs[b] Cd	0.60	0.002	24	Se vs Cd	0.69	<0.001	24
Hg(II) vs Cd	0.72	<0.001	24	Se vs Cu	0.46	0.028	24
Hg(II) vs Cu	0.49	0.015	24	Se vs Hg(II)	0.98	0.011	24
MHg vs Cd	0.57	0.004	24	Se vs MHg	0.53	0.007	24
MHg vs Hg(II)	0.54	0.006	24	Se vs SHg	0.97	<0.001	24
SHg vs Cd	0.72	<0.001	24	Kidney			
SHg vs Cu	0.45	0.028	24	Se vs Cd	0.82	<0.001	24
SHg vs Hg(II)	0.99	<0.001	24	Zn vs Cd	0.44	0.034	24
SHg vs MHg	0.54	0.003	24	Muscle			
Ag vs Cd	0.61	0.002	24	Hg(II) vs Cu	0.52	0.011	23
Ag vs Cu	0.71	<0.001	24	SHg vs Hg(II)	0.59	0.003	23
Ag vs Hg(II)	0.67	<0.001	24	SHg vs MHg	0.95	<0.001	23
Ag vs SHg	0.63	0.001	24	Blubber			
Ag vs Se	0.67	<0.001	24	Zn vs Cu	0.92	<0.001	14
Zn vs MHg	−0.46	0.025	24				

[a] Data were ranked prior to analysis.
[b] vs = versus.

a significantly greater mean level in polar bears) in liver, kidney, and muscle, respectively, between ringed seals and polar bears were 8.2×10^{-2}, 0.33 and 0.11 for Cd; 3.31*, 0.43 and 2.23* for Cu; 2.67, 29.0* and 2.0 for Pb; 1.30, 5.17* and 1.93* for Se; and 1.59*, 0.85 and 1.81* for Zn. The BMF for SHg in liver was 5.13*.

DISCUSSION

General

Ringed seals and polar bears are important subsistence resources to Alaskan Natives, as well as middle (ringed seal) and upper (polar bear) trophic level representatives of a food web, with ringed seal comprising a majority of the polar bear's diet (Stirling and McEwan, 1975; Norstrom et al., 1986). Ringed seals are middle trophic level feeders, relying upon crustaceans (amphipods, euphausiids and mysids) and fish, particularly Arctic cod (*Boreogadus saida*; Norstrom and Muir, 1994). Polar bears feed predominantly upon ringed seals, bearded seals (*Erignathus barbatus*) and when the opportunity arises, carrion (Stirling and McEwan, 1975; Norstrom et al., 1986). Polar bears are known to scavenge carcasses of subsistence-harvested whales and beach cast marine mammals. As apical predators, polar bears may be particularly prone to amassing high body burdens of persistent contaminants.

Many significant correlations among our data have associated P-values of <0.01 or <0.001, which are not likely to be due to chance. Moreover, we scrutinized data for common patterns that would implicate elemental interactions across tissues and species.

Arsenic

In comparison to other tissues, As concentrations were highest in blubber of ringed seals, probably connoting that it is in a non-toxic, organic form, with levels similar to those observed in belugas (Woshner et al., 2001a).

Lead

Caution must be exercised in interpretation of Pb levels in animals taken using lead ammunition as was the case for animals in this study. Nevertheless, all of the tissues in all species examined had very

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low Pb levels, with the highest concentrations observed in polar bear kidney. Ringed seals displayed higher Pb levels in liver as compared to kidney, similar to cetaceans (Woshner et al., 2001); the reverse was true in polar bears, which would appear to be more like terrestrial mammals in their organ distribution of Pb. The BMF for renal Pb was 29.0, the largest BMF calculated in this study. Overall, Pb concentrations in polar bears and ringed seals were well below concentrations likely to be associated with toxic effects (Puls, 1994).

Cadmium, copper, zinc

In polar bears, renal Cd concentration was approximately 20 times greater than liver Cd, while in ringed seals Cd levels in these two tissues were much less disparate, with mean kidney Cd concentrations being approximately four-fold higher than in liver, similar to observations in cetaceans (Woshner et al., 2001). The significantly higher Cd levels in liver, muscle and kidney of ringed seals as compared to polar bears connote that the BMF for this metal is significantly less than unity (with the hepatic BMF for Cd of 8.2×10^{-2} the lowest BMF calculated in this study). Therefore, it is unlikely that Cd would biomagnify in or present a toxicologic risk to polar bears. Although Cd in these two tissues was elevated in ringed seals compared to what are considered normal levels in domestic species such as dogs and cattle (Puls, 1994), Cd concentrations found in this study are well within ranges previously reported for arctic marine mammals (Hansen et al., 1990; Wagemann et al., 1996; Dietz et al., 1996, 1998). While a renal cortical concentration of 200 μg/g ww has been acceded as the critical threshold for chronic Cd toxicosis, our results, as well as most previous studies, have documented Cd concentrations in combined cortical and medullary tissue (Woshner et al., 2001), and thus underestimate cortical Cd concentrations by about 25% (Dietz et al., 1998). A histopathological examination of kidney in conjunction with chemical analyses for ringed seals divided into three groups on the basis of kidney Cd levels indicated no lesions typical of chronic renal Cd toxicosis (Dietz et al., 1998). Histopathologic examinations of ringed seals, beluga whales, and bowhead whales performed together with the present study would appear to support this assertion, because lesions characteristic of chronic Cd toxicosis were not observed (Woshner, 2000).

Cadmium demonstrated a significant positive correlation with Se in kidney of polar bears, as it did in liver of bowhead whales and liver and kidney of beluga whales (Woshner et al., 2001) but did not correlate significantly with Se in any tissues of ringed seals. While Se has been shown to ameliorate toxic effects of Cd in laboratory animals (Ridlington and Whanger, 1981; Wahba et al., 1993; Rana and Verma, 1996), its association with Cd in polar bears (as well as in beluga and bowhead whales; Woshner et al., 2001) might result from coincidental accumulation of these two elements with age. If physiologic association of Cd and Se were a generally occurring phenomenon among arctic marine mammals, one would expect Cd and Se to have been correlated in ringed seals, which had Cd concentrations in liver and kidney similar to bowhead whales (Woshner et al., 2001). Additionally, Cd was positively correlated with Hg in kidney of ringed seal and liver of polar bear, probably as a consequence of mutual accretion with age. Because of these findings, it seems doubtful that Se plays a major role in Cd detoxification in these species; nevertheless, the possibility cannot be ruled out.

Cadmium correlated positively with Zn in kidney of both species, as well as in liver of ringed seals. Other associations commonly observed in various tissues were Cd with Cu, and Cu with Zn or Ag; these elements were also frequently correlated with Hg and Se. It has been shown repeatedly that Cd and Hg both accumulate

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with age (Norstrom et al., 1986; Braune et al., 1991; Muir et al., 1992; Wagemann et al., 1996), and that the chief mode of detoxification of Hg in most marine mammals species is through formation of insoluble Hg-Se complexes in liver (Martoja and Berry, 1980; Rawson et al., 1995; Nigro and Leonzio, 1996). As discussed in Woshner et al. (2001), intercorrelations of the aforementioned metals could result both from mutual binding to inducible MTH, as well as coincidental accumulation with age via non-MTH processes.

Braune et al. (1991) noted positive correlations between Cd, Zn, Cu and Ag in polar bears, which they attributed to common binding of these elements by MTH. They also observed high hepatic Cu levels, which they suggested might indicate a physiological Cu requirement peculiar to the polar bear. In the present study, mean hepatic Cu in polar bears was statistically higher than that of ringed seals (BMF = 3.31), but was well within normal ranges reported for some domestic terrestrial species, whereas the seals had mean liver Cu concentrations which would be considered below the normal range of dogs or cattle (Puls, 1994). Nevertheless, levels in this study were in agreement with previously published data in pinniped species (Wagemann et al., 1996). The levels of essential elements, like Cu, are reported here for apparently healthy animals and should be useful in cases where a deficiency is suspected.

The liver mean total Hg:Se molar ratio in polar bears was approximately 5 times that in their primary food source, ringed seals. This finding is incongruous with the pattern of Cd disposition between the two species and the explanation for it proffered by previous researchers, who suggested that some polar bears may only consume the blubber layer (low Cd and Hg concentrations), thus not consuming the portion of the carcass in which most metal contaminants tend to be found (Stirling and McEwan, 1975; Dietz et al., 1995). These data also suggest that, unlike cetaceans and pinnipeds, in which Se may serve as the primary vehicle for Hg detoxification, polar bears may, like some terrestrial mammals, rely more on MTH for this function. Induction of MTH by Hg would, in turn, account for the relatively elevated levels of Cu and Zn in the liver of this species. Dietz et al. (1995) reinforced the supposition that, unlike other marine mammals in which the highest Hg concentrations are in liver, the polar bear accumulates Hg primarily in kidney as do some terrestrial mammals. Therefore, the dissimilarity noted here between tissue distribution of Hg in polar bears and ringed seals may be attributable to differences in metal metabolism or toxicodistribution as opposed to the partial seal consumption theory.

In addition to liver, Zn concentrations also were significantly higher in polar bear muscle compared to ringed seals. Wagemann et al. (1996) found mean Zn levels to be similar across muscle, liver and kidney within species (narwhal, belugas, and ringed seals), and Zn (and Cu) levels tended to track those of Cd. While we observed similar associations among Zn, Cd, and Cu within species, differences in mean Zn levels between tissues were more marked. This is most likely because Alaskan animals have a lower Cd burden compared to the Canadian animals, mirroring the geologic gradient of Cd, which decreases from east to west across North America (Wagemann et al., 1996).

Silver

Uniformly low Ag levels were observed across tissues of ringed seals and polar bears. In liver of polar bears and ringed seals Ag was positively correlated with Se, Cd, and various forms of Hg. A positive correlation between hepatic Ag and Cu was observed in both species examined. The associations between Ag and the aforementioned elements might be explained by common binding to MTH or other sulfur-rich molecules.

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Selenium and mercury

Mean Se concentrations in liver and kidney tissue of ringed seals and polar bears were consistent with levels associated with toxicosis in domestic species such as cattle and dogs (Puls, 1994), although well within ranges previously reported for these species of marine mammals (Norstrom et al., 1986; Braune et al., 1991; Wagemann et al., 1996). Among ringed seals, hepatic Se concentrations were approximately 3-fold higher than levels in kidney. In contrast, the highest mean Se concentration in polar bear tissues occurred in kidney. Dietz et al. (1995) reported higher concentrations of Hg in kidney as compared to liver of polar bears; this is similar to terrestrial species, and contrary to the pattern of tissue distribution among cetaceans and pinnipeds.

The presence of Hg-Se complexes has been demonstrated in liver tissue of marine mammals, although the precise structure, function, and distribution of these complexes is not known (Martoja and Berry, 1980; Pelletier, 1985; Nigro and Leonzio, 1996), and other tissues have not been addressed. In controlled experiments, Se has been proven to mitigate toxic effects of Hg (Stillings et al., 1974; Ridlington and Whanger, 1981; Cuvin-Aralar and Furness, 1991), Cd (Ridlington and Whanger, 1981; Rana and Verma, 1996) and Ag (Ridlington and Whanger, 1981; Wagner et al., 1975). It has been proposed that the ameliorative action of Se towards these three metals occurs via antioxidant activity because other antioxidants such as reduced glutathione (GSH) and vitamin E offer similar protection (Rana and Verma, 1996), or because these metals deplete the Se-dependent enzyme glutathione peroxidase (GSH-Px; Wagner et al., 1975; Sidhu et al., 1993). Evidence also exists for Se acting as an antioxidant through formation of metal selenide complexes, in particular for Ag (Aaseth et al., 1981) and Hg (Björkman et al., 1995).

In liver of ringed seal and polar bear, total Hg was present at concentrations that would be considered high or even toxic among domestic species (Puls, 1994). However, levels were consistent with those previously reported for marine mammals (Koeman et al., 1973, 1975; Dietz et al., 1990; Braune et al., 1991; Wagemann et al., 1996). The majority of Hg in the liver (~90%) of ringed seals and polar bears was inorganic [Hg(II)]. Polar bears exhibited a lower proportion of MHg in muscle tissue as compared to Alaskan beluga whales (Woshner et al., 2001), with a mean of 72.7% as a percentage of SHg. In polar bears, Hg(II), MHg and SHg were positively correlated with Se in liver. Hepatic Hg(II) was more closely associated with Cu than was SHg, which might support a relationship to induction of MTH, as might the positive association between Cu and Hg(II) in polar bear muscle. In ringed seals, THg, Hg(II) and SHg were positively correlated with Se in liver. In ringed seal kidney, THg was positively correlated with Cd and Zn, as well as with Se, association of THg with the former two elements probably being due to mutual accumulation with age.

The 1:1 molar relationship purported to be characteristic of the association between hepatic Hg and Se in marine mammals was not borne out in the present research. Regression analyses of hepatic Hg versus Se expressed in molar quantities for both species evaluated revealed slopes below one, and mean total Hg:Se ratios were significantly less than unity. Various researchers have presented information supporting and refuting the consistency of a 1:1 ratio in tissues of marine mammals, as recapitulated in Woshner et al. (2001). Findings of the present study do not counter the recognized relationship of Hg with Se, or the premise that this association could be the primary mode of Hg detoxification in marine mammals. However, the variability of the Hg:Se ratio within and between species suggests that these elements might occur in a consistent proportion only when a physiologic threshold has

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been surpassed, or that adherence to this ratio is not a physiologic necessity. Ancillary detoxification mechanisms for Hg may exist, particularly in the polar bear which, not surprisingly, appears to resemble terrestrial mammals more closely than pinnipeds or cetaceans in metal toxicodisposition. If this is the case, polar bears, which appear to bioaccumulate Hg, but not Cd, may be more vulnerable to toxic effects of Hg than pinniped or cetacean species. Hepatic Hg levels were significantly greater in polar bears, which had a relatively high BMF for hepatic SHg (5.13). Consequently it is likely that polar bears accumulate Hg above levels found in their prey, especially given that the tissue primarily responsible for Hg accretion in polar bears appears to be the kidney, rather than liver (Dietz et al., 1995). While frequently applied to comparisons of toxicant concentrations in a single tissue between species (as we have done here), we emphasize that the term BMF ideally applies to a ratio of body burdens, or some other measure of metal burden not susceptible to between species differences in tissue tropism.

In conclusion, interactions between Hg and Se, and between Cd, Zn and Cu appear prevalent in many tissue types of polar bears and ringed seals. Variations in tissue tropism for some elements (Hg, Pb, Se) between species were also apparent, suggesting that major metabolic and/or physiologic differences exist between ringed seals and polar bears with respect to some elements. Results of the present study indicate that a better understanding of the interactions between essential and non-essential elements and impacts in these two species will necessitate more comprehensive research of mercury speciation, metal localization at the cellular level, coordinated assessment of health of marine mammals by gross and histological examination, and physiologic/behavioral assessments. Some of these issues form the focus of our on-going research and present us with great challenges in understanding the toxicodisposition, interactions, essentiality and potential adverse effects of these elements in arctic marine mammals.

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