Essential and non-essential elements in the bowhead whale: epidermis-based predictions of blubber, kidney, liver and muscle tissue concentrations

TODD M. O'HARA*+, CYD HANNS+, VICTORIA M. WOSHNER#, JUDY ZEH++, GERALD BRATTON** AND ROBERT TAYLOR**

Contact e-mail: fftmo@uaf.edu

ABSTRACT

Assessment of element concentrations in wildlife must address both nutritional and toxicological considerations. The liver, epidermis, muscle and kidney of the bowhead whale are rich in some essential and non-essential elements. Blubber tends to have lower concentrations of these elements. Various cetaceans have been evaluated for these elements using a variety of sample sources (live and dead stranded whales, bycaught animals, remote and capture-release biopsy techniques, hunter killed whales etc). One constant shared by these approaches is the sampling of epidermis and adjacent dermis (blubber). In this study, the ability of elemental concentrations in bowhead whale epidermal samples to predict the corresponding elemental concentrations in blubber, kidney, liver and muscle is investigated. Epidermal concentrations had no predictive value for copper (Cu), manganese (Mn), lead (Pb), selenium (Se) or zinc (Zn) in any of the other tissues evaluated, except that the epidermal measurement provided an upper bound for blubber concentration of Cu, Mn, Se and Zn. Epidermal concentrations of the four other elements considered were predictive for some other tissues. Arsenic (As) concentrations could be predicted in kidney, liver and muscle but not blubber, although the preponderance of samples with concentrations below the minimum level reported (MLR, also known as 'detection limit') and the small sample sizes that resulted from their omission suggest that these data should be interpreted with caution. Epidermal concentrations of cadmium (Cd) were strongly predictive for blubber and weakly predictive for muscle concentrations. Epidermal concentrations of mercury (Hg) were weakly predictive of blubber, liver and muscle concentrations. Epidermal concentrations of magnesium (Mg) were strongly predictive in blubber, kidney and liver but only weakly predictive in muscle. Thus epidermal biopsy cannot predict elemental concentrations in four key tissues in bowhead whales in most cases. Cobalt (Co) and molybdenum (Mo) were not detected in any epidermal samples. This inability of epidermal element concentrations to reflect concentrations in internal tissues is likely true for other mysticetes and perhaps for cetaceans in general. At a minimum, before using epidermal biopsies to predict internal tissue concentrations of elements, researchers must establish that a sound scientific basis exists for doing so. Such proof must be specific to the elements, species and tissues in question as well as based upon statistically adequate sample sizes.

KEYWORDS: BOWHEAD WHALE; ELEMENTS; EPIDERMIS; HEAVY METALS; TISSUES; STATISTICS

INTRODUCTION

The bowhead whale (*Balaena mysticetus*) is a large mysticete found in the Arctic waters of the Bering, Chukchi and Beaufort Seas (B-C-B stock) that feeds on marine invertebrates (Lowry, 1993). Native subsistence whalers hunt this species under regulation by the International Whaling Commission (IWC), National Oceanographic and Atmospheric Administration (NOAA) Fisheries Service (NMFS) and the Alaska Eskimo Whaling Commission (AEWC). This hunt provides a valuable opportunity to study nutrients and contaminants in a large number of what probably are healthy cetaceans.

Element interactions in the bowhead whale have previously been the subject of limited studies (Dehn *et al*., 2006; Woshner *et al*., 2001b). This paper reports on the prediction of element concentrations in blubber, liver, kidney and muscle from measurements of the elements in epidermis. Reports from numerous investigators have addressed element concentrations in various matrices and species of cetaceans from around the globe (André *et al*., 1991; Beck *et al*., 1997; Becker *et al*., 2000; Bustamante *et al*., 2003; Decataldo *et al*., 2004; Dehn *et al*., 2006; Fossi *et al*., 2004; Frodello *et al*., 2000; Fujise *et al*., 1988; Honda and Tatsukawa, 1983; Honda *et al*., 1983; Kuehl and Haebler, 1995; Kuehl *et al*., 1994; Kunito *et al*., 2002; Law *et al*., 1991; Mackey *et al*., 2003; Marcovecchio *et al*., 1990; Meador *et al*., 1999; Monaci *et al*., 1998; Nigro *et al*., 2002; O'Hara *et al*., 2003; Roditi-Elasar *et al*., 2003; Stein *et al*., 2003; Wagemann *et al*., 1996; Woods and Van Vleet, 1996; Yang *et al*., 2002).

Attempts to evaluate levels of contaminants (e.g. organochlorines and mercury) and nutrients (e.g. copper) in wildlife using non-lethal and/or minimally invasive procedures have met with varying degrees of success, depending upon the species, sampling design, endpoint measured, environmental/habitat conditions and matrix of interest. For example, the use of hair for assessing some essential elements (e.g. copper) and for detecting non-essential 'toxic' elements (e.g. mercury and arsenic) in various mammalian species is well established (Beckmen *et al*., 2002; Born *et al*., 1991; Flynn *et al*., 1975; Flynn *et al*., 1977; Frank *et al*., 1994; Gogan *et al*., 1989; Ikemoto *et al*., 2004; O'Hara *et al*., 2001; Underwood, 1977; Wiig *et al*., 1999). Thus, the suggestion that epidermis may be useful for evaluating the status of essential and non-essential elements in the animal more holistically (blubber, muscle, kidney and liver) has some scientific precedent. Epidermal biopsies (and underlying blubber) from free ranging cetaceans have been obtained remotely (i.e. using 'darts') and via capture, followed by surgical biopsy and release of the animal (Fossi *et al*., 2004; Hansen *et al*., 2004; Ylitalo *et al*., 2001). It is

* *Current address: Institute of Arctic Biology, University of Alaska Fairbanks, Fairbanks, AK 99775, USA.*
+ *Department of Wildlife Management, North Slope Borough, Barrow, AK 99723, USA.*
# *8607 Bromley Rd., Hillsborough, NC 27278, USA.*
++ *University of Washington, Department of Statistics, Box 354322, Seattle, WA 98195-4322, USA.*
** *Department of Veterinary Anatomy and Public Health, Texas A&M University, College Station, TX 77843, USA.*

important to emphasise that cetacean 'skin' is a food item and may be a target organ of concern or monitoring tool for mercury intoxication (Gauthier *et al*., 1998) and thus has merit as a sampled tissue regardless of the correlation of element concentrations or predictive capacity for other tissues that were tested in this study.

With respect to 'toxic' elements and xenobiotics in bowhead whales, there are global sources of contaminants in general (De Wit *et al*., 2004; Woshner *et al*., 2001b) and local activities of concern (Ford and Hasselbach, 2001; O'Hara *et al*., 2003), including Red Dog Mine, a zinc/lead/silver mine (162°49'04"W 68° 04'11"N). The proximity of the mine, port and haul road to the coast has caused concern among local communities regarding potential contamination of the environment. Oil activities within and surrounding Prudhoe Bay, Alaska, also raise concerns related to mobilisation of heavy metals. Aspects of the environmental chemistry and bioavailability of toxic elements have been addressed for some regions of the Southern Beaufort Sea and rivers draining this area (Trefry *et al*., 2003).

The suitability of epidermal biopsies were tested for their use in determining the essential and/or non-essential element status of bowhead whales by sampling five tissues (epidermis, blubber, liver, kidney and muscle) and evaluating whether animal-matched tissue concentrations of elements could be predicted from the epidermal sample. The accumulation of particular elements in marine mammal tissues varies with element and organ type, reflecting tissue-specific physiological mechanisms (Decataldo *et al*., 2004; Woshner *et al*., 2001a; Woshner *et al*., 2001b). Thus it was hypothesised that, for elements exhibiting organ-specific bioaccumulation, epidermis is unlikely to serve as a good indicator of either general status or specific internal organ concentration.

MATERIALS AND METHODS

Field sampling

The field sampling methods used in this study have been previously described (O'Hara *et al*., 1999) and some of the data used in this report, from 15 whales harvested in 1996-1997, have been published (Dehn *et al*., 2006; Woshner *et al*., 2001b). Table 1 provides the basic data on all 48 whales studied (data derived from Suydam *et al*., 2004; Suydam *et al*., 2003; Woshner *et al*., 2001b). Full-thickness blubber cores and various tissues (epidermis, kidney, liver and muscle) from bowhead whales were provided by native subsistence hunters in Barrow, Alaska, USA. Samples were collected by staff at the Department of Wildlife Management with the permission of the Alaska Eskimo Whaling Commission (AEWC) and Barrow Whaling Captains Association (Barrow, Alaska, USA). Epidermal and blubber cores from approximately the same location on each whale (dorsal midline, 1 meter caudal to the blowhole) were collected. Life history information was recorded from each whale harvested (body length, sex, etc.), see Table 1. Relationships among these parameters with respect to various elements have been described previously by Bratton *et al*. (1997), Woshner *et al*. (2001b), and Dehn *et al*. (2006) and are not repeated here. Samples were temporarily stored at −20°C at the Arctic Research Facility (Barrow, Alaska, USA) and temperature was maintained during transport to Texas A&M University (College Station, TX) via provision of the US Marine Mammal Protection Act (Permit No. 782-1399 and 932-1489-05).

Table 1

Whale identification number (ID), date landed (dd/mm/yy), length (m) and sex of bowhead whales landed by Alaskan Eskimos during the 1996/97, and 2002/03 subsistence hunts evaluated in this study.

Whale ID	Date landed	Sex	Length (meters)
96B1	25/04/96	F	8.46
96B2	03/05/96	F	7.65
96B3	05/05/96	F	7.63
96B4	24/05/96	F	14.38
96B5	29/05//96	F	14.9
96B22	24/09/96	M	11.63
96B23	26/09/96	M	7.59
96B24	26/09/96	F	10.87
97B1	04/05/97	M	10
97B3	07/05/97	F	16.97
97B5	10/05/97	F	10.08
97B6	12/05/97	F	8.3
97B7	12/05/97	F	13.15
97B8	15/05/97	F	13.9
97B10	04/06/97	F	16.71
02B1	03/05/02	F	11.7
02B2	10/05/02	F	16.7
02B3	30/05/02	F	19.2
02B4	30/09/02	F	8.6
02B5	01/10/02	F	8.5
02B6	03/10/02	M	9.0
02B7	03/10/02	M	8.0
02B8	03/10/02	F	6.8
02B9	10/10/02	F	7.5
02B10	10/10/02	M	9.5
02B11	15/10/02	F	8.1
02B13	15/10/02	M	9.6
02B14	18/10/02	F	8.5
02B15	18/10/02	M	8.8
02B16	19/10/02	M	8.3
02B17	19/10/02	F	9.3
02B21	22/10/02	F	10.0
02B22	25/10/02	F	8.1
03B1	19/04/03	F	9.1
03B2	03/05/03	M	13.8
03B3	07/05/03	F	9.0
03B4	08/05/03	M	13.4
03B5	08/05/03	M	7.7
03B6	09/05/03	F	13.9
03B7	12/05/03	M	12.8
03B8	24/05/03	M	14.9
03B9	25/05/03	F	16.4
03B11	08/10/03	F	8.7
03B12	09/10/03	F	11.2
03B13	09/10/03	M	11.9
03B14	09/10/03	M	11.1
03B15	14/10/03	F	12.5
03B16	14/10/03	F	10.1

The first two numbers of the ID indicate the year landed; the letter the village where landed (B = Barrow); and the final number(s) indicate the sequence in which it was landed (1=first) for that calendar year. Sample size 1996=8; 1997=7; 2002=18; 2003=15. The lengths reported are taken from Suydam *et al.* (2003; 2004) and Woshner *et al.* (2001b).

Minerals and metals analysis

Upon receipt in the laboratory, sample integrity was evaluated and samples were immediately transferred to secure freezers for storage until processed. All tissues were stored at −60°C until analysis. Tissues were thawed, homogenised by chopping in plastic weigh boats and microwave digested as previously described (Woshner *et al*., 2001b). Samples were run in sets of 20 along with standard reference materials, SRMs (bovine liver 1577b, Dogfish liver Dolt 2, and Dogfish muscle Dorm 2), a blank, a blank spike (Lab Control Sample-LCS), a sample duplicate for each sample type, and a sample spike for each sample type. Two SRMs were used per sample set with the intent to bracket the expected analyte levels in the tissues being considered. In general, each type of tissue was digested

separately, with pooling of tissue types for digestion only carried out when less than 20 tissues of a given type were received. Following digestion, all samples and QA (quality assurance) samples were diluted to 20ml with 18meg ohm water, a final weight was taken and the samples were density corrected. Digestates were stored tightly sealed until analysis was complete. All analytical work was generally completed within a month after digestion. Table 2 summarises the procedures used in this study.

Elements were analysed by atomic absorption spectrophotometry (AAS) at Texas A&M University (TAMU) Trace Element Research Laboratory (TERL) as in Woshner *et al*. (2001b) for those samples collected in 1996-1997, employing a strict quality assurance/quality control protocol with appropriate standard reference materials, sample duplicates, blanks and spiked samples. The following elements were analysed: arsenic (As), cadmium (Cd), cobalt (Co), copper (Cu), mercury (Hg), magnesium (Mg), manganese (Mn), molybdenum (Mo), lead (Pb), selenium (Se) and zinc (Zn). Neither Co nor Mo was detected in any epidermal sample, so they are not considered further. The remaining elements were detected in more than half of the samples for at least two tissues. Co, As, Mg, Mn, Pb, and Se were analysed using a *Perkin-Elmer* (Norwalk, Connecticut, USA) model SIMAA 6000 graphite furnace atomic absorption spectrophotometer (GFAAS) equipped with an AS-60 autosampler and Zeeman background correction (Perkin-Elmer); Cd and Zn were determined using a *Perkin-Elmer* instrument model 306 flame AAS; total Hg was determined via cold vapor AAS (AAS-CVG) using a Thermo-Jarrell Ash (Franklin, Massachusetts, USA) model S-11 AAS with a *Thermo-Jarrell Ash* AVA-440 atomic vapor accessory. The minimum level reported (MLR, also known as 'detection limit') for Hg was 0.001 μg/g. For total Hg analysis (at the Texas Veterinary Diagnostic Laboratory, Amarillo, Texas USA), wet tissue samples (1996-1997) were weighed and transferred to 250ml quartz volumetric digestion tubes. Sample digestion and analysis followed Korsrud *et al*. (1985) with minor modifications as reported in Woshner *et al*. (2001b). See Bratton *et al*. (1997) and Woshner *et al*. (2001b) for details. The minimum level reported for all elements except Hg was 0.01μg/g.

For samples collected in 2002-2003, subsample preparation and digestion followed the method previously described by Woshner *et al*. (2001b) and the analyses is similar to that reported by Dehn *et al*. (2006). Total mercury was determined using a cold-vapor atomic absorption spectrometry (CVAAS) method with a *Cetac* 7500 QuickTrace Hg analyser. Hg^{2+} was converted to Hg^0 by reduction with $SnCl_2$, purged from the digested sample with a stream of argon gas, and swept into a thermally-stabilised absorption cell. Concentrations were determined by absorbance peak height using commercial calibration standards.

Inductively coupled plasma-mass spectroscopy (ICP-MS) is a hyphenated technique in which a high-temperature, radio frequency argon plasma provides ions that are measured by a mass-specific detector. TERL uses a *Perkin-Elmer/Sciex* DRC-2 ICP-MS instrument equipped with a quadrupole detector. This instrument also employs a dynamic reaction cell to remove molecular ion interferences. Digested samples were diluted to a final strength of 2% nitric acid. The aqueous samples were introduced to the plasma using a peristaltic pump, concentric nebuliser, and cyclonic spray chamber. Internal standards were added to all samples to compensate for viscosity differences and slight variations in instrument performance related to sample matrix effects. The instrument was calibrated using a blank and three external standards. Calibration was evaluated using both a low standard and an independent check standard (NIST SRM 1640). Data were collected in dual pulse/analog mode using the instrument's autolens feature to maximise sensitivity at each target mass.

Digested samples were diluted to 20ml with trace metal free water and run directly on a *Spectro* CirOS axial inductively coupled plasma optical emission spectrometer (ICP-OES) (Spectro AI, Fitchburg, MA) for Cu, Mg, Mn, and Zn. Additional aliquots of the digestates were then diluted further by a factor of ten and run on a *Perkin-Elmer/Sciex* DRC-2 ICP-MS (Perkin-Elmer, Norwalk, CT) for As, Cd, Co, Mo, and Pb. A separate aliquot was diluted ten-fold in 3N HCL and analysed for Se using a *PSA* Millennium HG- Atomic Fluorescence Spectrometer (AFS) system (PSA Analytical Deerfield Beach, FL). A final aliquot of the original digestate was diluted by a factor of five in 7% v/v HCL and analysed for Hg using a *Cetac* M7500 CVAAS system (Cetac Technologies, Omaha, NE).

QA was considered in compliance when recovery for SRMs, blank spikes, and sample spikes was +/-20% for minerals/metals that were very low and close to baseline levels and +/-10% for mineral/metals that were considerably above baseline levels. Duplications were considered acceptable when +/-10% and the analyte was significantly above the baseline. Blanks were considered acceptable if <0.0001 ppm.

Statistical analysis

Summary statistics were generated using *Microsoft* Excel (Microsoft Corporation, 1999) and S-PLUS (Venables and Ripley, 1999). Plots of element concentrations (μg/g wet weight) in blubber, kidney, liver and muscle tissues versus

Table 2

Summary of the procedures used in this study.

Years collected	Number of whales	Procedure	Elements*	Published
1996-97	15	Atomic absorption spectrophotometry (AAS)	Arsenic (As), cadmium (Cd), cobalt (Co), Copper (Cu), mercury (Hg), magnesium (Mg), manganese (Mn), molybdenum (Mo), lead (Pb), selenium (Se) and zinc (Zn)	Woshner *et al.* (2001b); Dehn *et al.* (2006)
2002-03	33	Hg via AAS Others via ICP (see next column)	Mercury via cold-vapour atomic absorption spectrometry (CVAAS). Inductively coupled plasma-mass spectrometry (ICP-MS). Perkin-Elmer/Sciex DRC-2 ICP-MS for As, Cd, Co, Mo and Pb. Using ICP - optical emission spectroscopy (ICP-OES) (Spectro AI, Fitchburg, MA) for Cu, Mg, Mn and Zn	This paper
Total	48			

the corresponding concentration in epidermal tissue (E), whale body length in meters (length) and sex of the whale were used to explore relationships. The plots indicated linear relationships, so simple linear regressions and multiple regressions with E, length and sex as potential predictors of concentrations in blubber, kidney, liver and muscle were used. Length was used as a surrogate for age, recognising that it is a poor surrogate, particularly for physically mature whales. However, ages of most of the sampled whales were unknown. Plots and regressions for each tissue only include whales in which both E and the concentration in that tissue ≥MLR. This could provide a misleading assessment of predictive power for those elements with some tissue concentrations <MLR in some samples (As, Cd, Hg, Mn and Pb). For elements with both E<MLR and concentrations <MLR in the tissue being predicted using E, cross-tabulations were calculated giving the number of whales with concentrations: (1) <MLR; (2) ≥MLR but less than the median concentration among all samples with concentrations ≥MLR; and (3) greater than that median concentration. Independence of row and column classifications was evaluated by Fisher's exact test. If the null hypothesis of independence is not rejected, this provides evidence against a strong association between E and the concentration in the other tissue. However, if the number of samples available for the test is small, failure to reject the null hypothesis may be due to a lack of power.

The best regression for each element in each tissue was chosen by minimising Akaike's Information Criterion (AIC) (Akaike, 1973) among regressions involving E, length and sex. A model with either too few or too many predictors has poor predictive ability. AIC is designed to identify the best predictive model, with neither poor predictive ability owing to omission of relevant predictors, nor excess variability due to inclusion of extraneous predictors. AIC penalises added predictors less severely than other selection criteria such as likelihood ratio tests (Venables and Ripley, 1999), so it sometimes selects unnecessary predictors. The main utility of the best regression result is to assess further whether E is a more useful predictor of the element's internal organ concentration than length and sex, which are much easier to measure.

Both the regression with E as the only predictor and the best regression were evaluated using R^2, the percentage of the variability in the concentration being predicted that is explained by the regression. The regression is characterised as having no predictive value if R^2 ≤35%, weakly predictive if R^2 is 36% to 55%, moderately predictive if R^2 is 56% to 75% and strongly predictive if R^2>75%. However, these characterisations are sometimes tempered by patterns in the cross-tabulations described above or the results of the Fisher's exact tests.

RESULTS AND DISCUSSION

General results, with comparisons to other species

Of the elements evaluated in this study, Mg, Mn, Cu, Zn and Se are considered essential (Table 3), while As, Cd, Pb and Hg have no known function in mammals and are considered non-essential (Table 4) and are sometimes referred to as the 'toxic' elements. A basic mammalian need for an element was used to define 'essential', even if the element had not been specifically evaluated for cetaceans. This assumption may be somewhat inappropriate considering the many known morphologic and physiologic differences between cetaceans and terrestrial mammals. The non-essential elements important for toxicological assessment in the Arctic food chain include Cd, Hg and Pb (AMAP, 1998; 2002). All of the elements assayed in this study were below concentrations associated with toxic effects in domestic animals (Puls, 1994), although this extrapolation between terrestrial and marine mammals may be misleading (André *et al*., 1991; Beck *et al*., 1997; Bustamante *et al*., 2003; Decataldo *et al*., 2004; Frodello *et al*., 2000; Honda *et al*., 1983).

Compared to other species of northern Alaska, the bowhead whales used in this study had much lower tissue concentrations of Hg, in agreement with the reports of Dehn *et al*. (2006), Woshner *et al*. (2001b) and Bratton *et al*. (1997). It is well described that element-element interactions with length (age) exist, particularly for Hg and Se in marine mammals, especially odontocetes (Honda and Tatsukawa, 1983; Honda *et al*., 1983). Thus considering these interactions in a tissue specific manner by species for age (length) and sex is very important. The details of these relationships are not reviewed in this report since they have been documented elsewhere (note citations above).

Among marine mammals, Cd appears to be higher in species with an invertebrate-based diet (e.g. similar to walrus), and the Cd concentrations reported here are similar to concentrations previously reported for bowhead whales (Bratton *et al*., 1997; Dehn *et al*., 2006; Woshner *et al*., 2001a; Woshner *et al*., 2001b). Bowhead whales, like other mammalian species, have been shown to accumulate Cd with age in the liver (Honda and Tatsukawa, 1983; Honda *et al*., 1983) and particularly in the kidney, with variations in the rate of accumulation occurring by region (Aastrup *et al*., 2000; Cooper *et al*., 2000; Elkin and Bethke, 1995; Gamberg and Scheuhammer, 1994; Honda and Tatsukawa, 1983; Honda *et al*., 1983; Larter and Nagy, 2000; O'Hara *et al*., 2001; O'Hara *et al*., 2003; O'Hara *et al*., 1999; Woshner *et al*., 2001a; Woshner *et al*., 2001b).

For both Cd and As, over half the epidermal samples had E<MLR (Table 4), resulting in sample sizes <20 for the regression analyses. Although regression results are discussed for these elements, they are omitted from Table 5 for this reason. Blubber was less frequently sampled than the other tissues, resulting in sample sizes <20 for all elements. This is included in Table 5, but the small sample sizes mandate caution in interpreting blubber results.

Concentrations of essential elements

Magnesium

All 48 epidermal samples had concentrations of Mg above the MLR. The best single predictor of Mg in all four other tissues is E, which is weakly predictive for muscle but strongly predictive for the other three. The best regression for Mg, as determined by AIC, always includes E. In addition, length was also included for all tissues except blubber. In kidney, liver and muscle tissue, concentrations of Mg increase with E and decrease with length. The best regression for blubber, kidney and liver explains 83-90% of the variability in Mg in these tissues. Even the best regression for muscle is only moderately predictive, explaining 57% of the variability of Mg. Based on Puls (1994), the Mg status of mammals may best be assessed using other tissues (e.g. blood) and/or fluids (plasma, urine, etc.). The role of cetacean epidermis in Mg elemental dynamics and tissue tropism requires further investigation.

Manganese

Manganese was detected in fewer than half of the 2002/03 epidermal and blubber samples analysed using ICP-OES, while low Mn levels were detected in these tissues by AAS

Table 3

Summary statistics for essential elements for bowhead whales sampled in 1996/97 (analysed using AA*) and 2002/03 (analysed using ICP or AFS**) in epidermis, liver, kidney, muscle and blubber. All statistics are based on the *N* samples with concentrations ≥MLR***.

	Cu-AA	Cu-ICP	Mg-AA	Mg-ICP	Mn-AA	Mn-ICP	Se-AA	Se- AFS	Zn-AA	Zn-ICP
Epidermis										
Mean	0.53	0.34	521	172	0.056	0.075	0.75	0.64	15.2	12.5
SD	0.10	0.09	41	17	0.023	0.048	0.36	0.14	2.1	1.8
Minimum	0.37	0.22	448	136	0.030	0.037	0.14	0.39	11.1	9.9
Maximum	0.75	0.72	604	202	0.100	0.229	1.32	0.86	19.2	18.7
Geometric mean	0.52	0.33	520	171	0.053	0.066	0.64	0.63	15.0	12.3
Total samples	15	33	15	33	8	33	15	33	15	33
Liver										
Mean	11.02	4.91	298	123	2.07	1.24	1.98	1.07	39.3	34.5
SD	13.35	1.42	44	19	0.61	0.48	0.40	0.33	14.7	11.1
Minimum	2.53	3.08	220	91	1.39	0.45	1.39	0.50	19.7	23.6
Maximum	54.00	8.96	396	178	2.96	2.43	2.56	1.79	76.4	65.1
Geometric mean	7.33	4.74	295	122	1.99	1.15	1.95	1.02	37.0	33.0
Total samples	14	34	14	34	14	34	14	34	14	34
Kidney										
Mean	2.35	1.65	298	91	0.53	0.36	1.78	1.29	34.8	21.1
SD	0.69	0.26	108	10	0.18	0.08	0.40	0.27	9.8	8.4
Minimum	1.40	1.13	142	72	0.30	0.20	0.95	0.77	23.0	12.7
Maximum	3.29	2.29	480	132	0.87	0.55	2.32	2.04	56.3	57.2
Geometric mean	2.25	1.63	278	91	0.50	0.35	1.73	1.26	33.6	19.9
Total samples	14	33	14	33	14	33	14	33	14	33
Muscle										
Mean	0.90	0.57	468	232	0.17	0.12	0.14	0.20	51.6	36.3
SD	0.22	0.10	220	23	0.08	0.04	0.03	0.03	12.3	9.1
Minimum	0.53	0.36	203	180	0.07	0.05	0.08	0.13	33.2	24.7
Maximum	1.25	0.76	753	268	0.30	0.18	0.17	0.25	76.6	62.8
Geometric mean	0.87	0.56	416	231	0.15	0.11	0.14	0.19	50.3	35.4
Total samples	15	33	15	33	8	33	15	33	15	33
Blubber										
Mean	0.12	0.13	47.0	14.5	0.020	0.036	0.09	0.10	5.51	0.93
SD	0.04	0.02	11.1	3.2	0.007	NA	0.04	0.03	1.61	0.16
Minimum	0.06	0.10	33.0	9.4	0.010	0.036	0.05	0.06	3.85	0.71
Maximum	0.18	0.16	64.0	18.6	0.030	0.036	0.17	0.14	7.55	1.16
Geometric mean	0.12	0.13	46.0	14.2	0.019	0.036	0.08	0.10	5.32	0.92
Total samples	12	6	5	6	5	6	12	6	5	6

*AA = atomic absorption spectrophotometry (AAS). **ICP = inductively coupled plasma - mass spectroscopy (ICP-MS) or inductively coupled plasma - optical emission spectroscopy (ICP-OES), or Atomic Fluorescence Spectrometry (AFS). MLR = minimum level reported. ***All samples reported in this Table were > MLR, except Mn-ICP has 55% (18) and 83% (5) of epidermis and blubber <MLR, respectively.

(Woshner *et al*., 2001b). Epidermal Mn is of no value as a predictor of Mn in any of the other four tissues. In no case is it chosen as a predictor by AIC. The mean (intercept) is the best predictor for kidney, liver and muscle. Although sex is weakly predictive for blubber, only six whales with Mn reported in both epidermal and blubber tissue were included in this analysis. A larger sample would be needed to determine whether sex is really a useful predictor of Mn in blubber.

In all 11 whales with both blubber and epidermal samples analysed, blubber concentration of Mn was lower than epidermal. This suggests that epidermal concentration could provide an upper bound for blubber concentration of Mn in bowheads with epidermis but not blubber sampled. However, Table 3 suggests that this would considerably overestimate blubber concentration.

Copper, Selenium and Zinc

Copper is an essential element reported to be at higher concentrations in foetal and neonatal bowhead whales that decreases with length (age) (Bratton *et al*., 1997; Woshner *et al*., 2001b) and Cu and Zn occur at similar concentrations in odontocetes (Decataldo *et al*., 2004; Honda and Tatsukawa, 1983; Woods and Van Vleet, 1996). However, the Se concentration tends to be much higher in odontocetes (Kuehl and Haebler, 1995; Kuehl *et al*., 1994; Mackey *et al*., 2003). The best regressions for Cu in blubber, kidney and muscle in Table 5 support these reports.

Concentrations of Cu, Se and Zn were above the MLR in all epidermal samples. Nevertheless, E had no predictive value for concentrations of these elements in other tissues, despite the fact that AIC occasionally included E as a predictor for a given element-tissue combination. Among regression analyses for these three elements, even the best regression that included E (for Cu in muscle) accounted for only 25% of the variability (Table 5). However, as was the case for Mn, all blubber concentrations of Cu, Se and Zn were below the corresponding epidermal concentrations, so epidermal concentrations may constitute an upper bound for blubber concentrations with respect to these three elements.

Concentrations of non-essential elements

Cadmium

Cadmium occurs at lower concentrations in the epidermis than in the other tissues examined, with E<MLR in 24 (59%) of the 41 epidermal samples analysed (Table 4),

Table 4

Summary statistics for non-essential elements for bowhead whales sampled in 1996/97 (analysed using AA*) and 2002/03 (analysed using ICP**) in epidermis, liver, kidney, muscle and blubber. All statistics except the medians are based on the *N* samples with concentrations ≥MLR.

	As-AA	As-ICP	Cd-AA	Cd-ICP	Pb-AA	Pb-ICP	Hg-AA	Hg-AA#
Epidermis								
Mean	0.078	0.491	0.013	0.010	0.012	0.008	0.007	0.017
SD	0.049	0.091	0.006	0.009	0.005	0.003	0.004	0.010
Minimum	0.020	0.380	0.010	0.004	0.010	0.004	0.003	0.004
Maximum	0.160	0.660	0.020	0.039	0.020	0.016	0.014	0.037
Geometric mean	0.063	0.485	0.013	0.008	0.012	0.007	0.006	0.014
N ≥ MLR	15	7	3	14	4	19	7	29
N < MLR	0	26	5	19	4	14	1	4
Total samples	15	33	8	33	8	33	8	33
Median of total samples	0.070	<MLR	<MLR	<MLR	<MLR	0.006	0.005	0.016
Liver								
Mean	0.122	0.538	7.86	9.47	0.015	0.015	0.029	0.051
SD	0.029	0.146	9.16	11.14	0.007	0.007	0.015	0.039
Minimum	0.080	0.380	0.04	0.28	0.010	0.006	0.004	0.009
Maximum	0.160	0.820	30.50	42.20	0.030	0.030	0.052	0.194
Geometric mean	0.118	0.520	1.99	3.99	0.014	0.014	0.024	0.038
N ≥ MLR	13	13	14	34	11	34	14	34
N < MLR	1	21	0	0	3	0	0	0
Total samples	14	34	14	34	14	34	14	34
Median of total samples	0.120	<MLR	4.900	5.850	0.010	0.013	0.031	0.051
Kidney								
Mean	0.089	0.453	20.84	13.95	0.020	0.008	0.022	0.032
SD	0.071	0.055	22.15	15.01	0.017	0.003	0.012	0.032
Minimum	0.020	0.400	0.10	0.47	0.010	0.005	0.006	0.003
Maximum	0.240	0.510	62.18	70.20	0.040	0.015	0.050	0.180
Geometric mean	0.064	0.451	6.17	6.31	0.016	0.008	0.018	0.022
N ≥ MLR	14	3	14	33	3	21	14	33
N < MLR	0	30	0	0	11	12	0	0
Total samples	14	33	14	33	14	33	14	33
Median of total samples	0.050	<MLR	12.38	14.10	<MLR	0.005	0.022	0.029
Muscle								
Mean	0.029	0.608	0.100	0.044	NA	0.016	0.013	0.020
SD	0.015	0.187	0.116	0.057	NA	0.017	0.008	0.012
Minimum	0.010	0.410	0.010	0.007	NA	0.006	0.001	0.003
Maximum	0.050	0.830	0.300	0.212	NA	0.066	0.021	0.040
Geometric mean	0.025	0.584	0.057	0.026	NA	0.012	0.009	0.016
N ≥ MLR	8	5	5	22	0	15	8	33
N < MLR	0	28	3	11	8	18	0	0
Total samples	8	33	8	33	8	33	8	33
Median of total samples	0.030	<MLR	0.020	0.016	<MLR	<MLR	0.013	0.022
Blubber								
Mean	1.376	1.305	0.038	0.012	0.020	0.008	0.002	0.006
SD	0.355	0.366	0.017	0.002	0.007	0.003	0.001	0.001
Minimum	0.770	0.770	0.020	0.009	0.010	0.006	0.001	0.005
Maximum	1.920	1.770	0.060	0.015	0.030	0.012	0.003	0.008
Geometric mean	1.328	1.258	0.035	0.011	0.019	0.008	0.002	0.006
N ≥ MLR	12	6	4	5	5	4	3	5
N < MLR	0	0	1	1	0	2	2	1
Total samples	12	6	5	6	5	6	5	6
Median of total samples	1.430	1.325	0.030	0.011	0.020	0.006	0.001	0.006

As=arsenic; Cd=cadmium; Pb=lead; Hg=mercury; *AA=atomic absorption spectrophotometry (AAS); **ICP=inductively coupled plasma - mass spectroscopy (ICP-MS) or inductively coupled plasma - optical emission spectroscopy (ICP-OES); MLR=minimum level reported; NA=not available. #=2002/03 samples for Hg.

compromising prediction of Cd concentrations in other tissues using E. For E≥MLR for Cd (Table 5, Fig. 1), E was strongly predictive of blubber concentration and weakly predictive of muscle concentration, but had no predictive value for kidney or liver concentrations. Two of three whales with E<MLR also had blubber concentration <MLR, but the third had blubber concentration in the highest category. However, the sample sizes are too small to give statistically significant results. Fisher's exact test did not reject the null hypothesis of independence of epidermal and blubber Cd concentration levels, suggesting that the regression result for blubber should be interpreted with particular caution.

It is not surprising that E was a poor predictor for kidney and liver Cd concentration. These internal organs are well known to accumulate Cd with age and are rarely at concentrations <MLR (Bratton *et al.*, 1997; Roditi-Elasar *et al.*, 2003; Wagemann *et al.*, 1996; Woods and Van Vleet, 1996; Woshner *et al.*, 2001a; Woshner *et al.*, 2001b). The best regression for both of these organs had length as the

only predictor. However, although length was a strong predictor for liver concentration of Cd (R^2=81%), it had no predictive value for kidney concentration (R^2=18%). Fig. 1 shows why kidney Cd concentration cannot be predicted well. There are two whales of adult length with extremely high kidney concentrations of Cd; these whales may have been very old (possibly not well accounted for with length), accounting for the large amount of Cd they have accumulated in their kidneys.

Mercury

Only five epidermal samples had E<MLR (Table 4). An adequate number of samples were available for kidney, liver and muscle (Table 5). With a regression value of R^2=30%, E had no predictive value for kidney concentration of Hg, and it was only weakly predictive in the other tissues. Epidermal prediction of Hg concentration in internal tissues might prove more useful in toothed cetaceans that are known to accumulate much higher levels of Hg in liver, kidney, muscle and epidermis (André *et al*., 1991; Beck *et al*., 1997; Becker *et al*., 2000; Bustamante *et al*., 2003; Yang *et al*., 2002) than bowhead whales (Woshner *et al*., 2001b). The best regressions for bowhead whales, which increase R^2 slightly, all include E as a predictor and also include sex for blubber (females have higher Hg concentrations) and length for kidney, liver and muscle (longer whales have higher Hg concentrations). The best regressions are still only weakly predictive for Hg in kidney and liver and moderately predictive in blubber and muscle.

Lead

Eighteen (44%) of 41 epidermal samples analysed for Pb had E<MLR, resulting in small sample sizes for regression analyses. Table 5 indicates that epidermal Pb is of no value for predicting Pb in other tissues as $R^2 \leq 21\%$ for Pb. The hypothesis of independence of E and concentration in the other tissue is never rejected by Fisher's exact test. For blubber and liver, none of the potential predictors (E, length, sex) is judged by AIC to be useful for prediction. For kidney and muscle, $R^2 \leq 29\%$ even for the models chosen by AIC

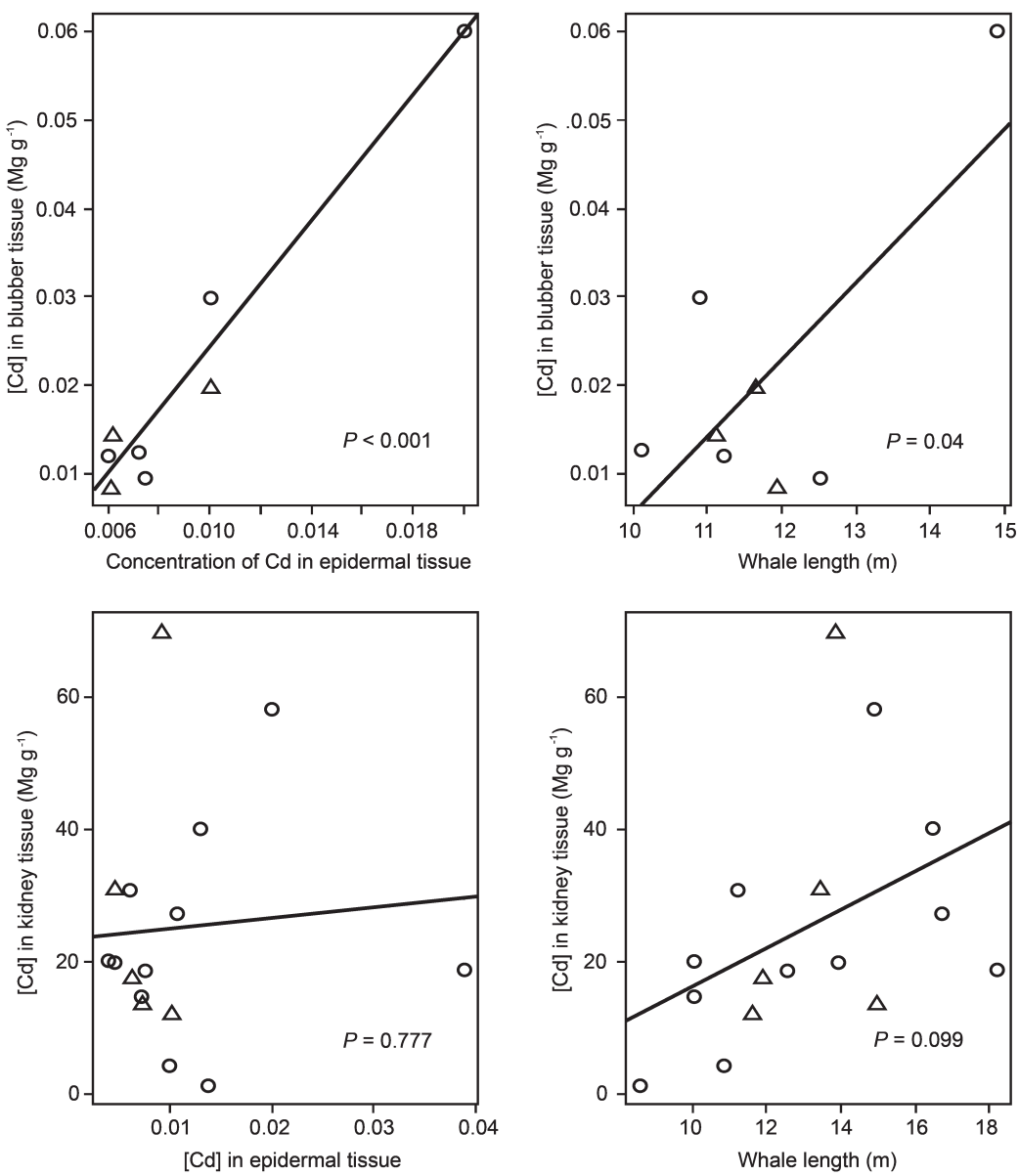

Fig. 1. Concentration (μg g^{-1}) of cadmium (Cd) in blubber and kidney tissue versus the corresponding epidermal concentrations and whale length. Males are denoted by triangles and females by circles. A least squares regression line with its *P*-value is shown on each plot.

Table 5

Evaluation of predictability of concentrations (µg/g wet weight) of elements in blubber, kidney, liver and muscle from the corresponding epidermal concentrations (E), whale length in meters (Length) and Sex (0 for males, 1 for females) in bowhead whales. Sample size is the number of whales with concentration ≥ MLR in both the epidermal sample and the other tissue sample; these are the whales on which the regression results for the element and tissue are based. The percentage of the variability of the element in the tissue explained by the regression is denoted by R^2. Standard errors (SE) of regression coefficients are given in parentheses after the coefficients. Best regression denotes the regression model chosen by Akaike's information criterion (AIC); NA for a coefficient means it was not selected by AIC. When AIC chose the mean (Intercept) as the best model, R^2 = 0% is used to indicate that none of E, Length or Sex was useful for predicting concentration of the element in the tissue.

		Blubber	Kidney	Liver	Muscle
Mercury (Hg)	Sample size	8	35	36	36
Regression on E:	R^2	49%	30%	39%	55%
Coefficients:	Intercept (SE)	0.0005 (0.0019)	0.0085 (0.0079)	0.0179 (0.0088)	0.0076 (0.0023)
	E (SE)	0.3071 (0.1284)	1.6404 (0.4352)	2.2781 (0.4839)	0.8127 (0.1260)
Best regression:	R^2	70%	40%	54%	60%
Coefficients:	Intercept (SE)	-0.0008 (0.0017)	-0.0263 (0.0165)	-0.0324 (0.0174)	-0.0014 (0.0049)
	E (SE)	0.3213 (0.1085)	1.5838 (0.4089)	2.1958 (0.4289)	0.7979 (0.1205)
	Length (SE)	NA	0.0032 (0.0014)	0.0047 (0.0015)	0.0008 (0.0004)
	Sex (SE)	0.0021 (0.0011)	NA	NA	NA
Lead (Pb)					
	Sample size	7	15	22	8
Regression on E:	R^2	21%	10%	3%	6%
Coefficients:	Intercept (SE)	0.0045 (0.0081)	0.0069 (0.0017)	0.0138 (0.0031)	0.0049 (0.0136)
	E (SE)	0.8283 (0.7096)	0.2205 (0.1875)	0.2595 (0.3273)	0.8485 (1.3519)
Best regression:	R^2	0%	29%	0%	27%
Coefficients:	Intercept (SE)	0.0132 (0.0033)	0.0043 (0.0021)	0.0160 (0.0013)	0.0528 (0.0269)
	E (SE)	NA	0.3097 (0.1794)	NA	NA
	Length (SE)	NA	NA	NA	-0.0040 (0.0027)
	Sex (SE)	NA	0.0027 (0.0015)	NA	NA
Magnesium (Mg)					
	Sample size	11	46	47	47
Regression on E:	R^2	89%	78%	88%	52%
Coefficients:	Intercept (SE)	-2.438 (4.115)	-13.178 (15.407)	40.201 (8.690)	103.309 (33.955)
	E (SE)	0.095 (0.011)	0.600 (0.048)	0.487 (0.027)	0.717 (0.104)
Best regression:	R^2	89%	83%	90%	57%
Coefficients:	Intercept (SE)	-2.438 (4.115)	67.365 (27.330)	84.320 (15.792)	239.984 (64.499)
	E (SE)	0.095 (0.011)	0.622 (0.043)	0.499 (0.025)	0.744 (0.099)
	Length (SE)	NA	-7.903 (2.313)	-4.321 (1.339)	-13.140 (5.372)
	Sex (SE)	NA	NA	NA	NA
Manganese (Mn)					
	Sample size	6	22	23	23
Regression on E:	R^2	25%	<1%	2%	<1%
Coefficients:	Intercept (SE)	0.0097 (0.0117)	0.450 (0.066)	1.817 (0.296)	0.146 (0.023)
	E (SE)	0.1935 (0.1663)	0.363 (0.841)	-2.273 (3.738)	-0.007 (0.285)
Best regression:	R^2	52%	0%	0%	0%
Coefficients:	Intercept (SE)	0.0287 (0.0041)	0.474 (0.034)	1.662 (0.148)	0.145 (0.011)
	E (SE)	NA	NA	NA	NA
	Length (SE)	NA	NA	NA	NA
	Sex (SE)	-0.0120 (0.0057)	NA	NA	NA
Copper (Cu)					
	Sample size	18	46	47	47
Regression on E:	R^2	3%	3%	9%	15%
Coefficients:	Intercept (SE)	0.1064 (0.0311)	1.553 (0.262)	-0.370 (3.587)	0.4115 (0.0988)
	E (SE)	0.0417 (0.0626)	0.785 (0.628)	17.825 (8.667)	0.6498 (0.2351)
Best regression:	R^2	47%	15%	13%	25%
Coefficients:	Intercept (SE)	0.1331 (0.0277)	2.010 (0.313)	1.559 (3.736)	0.5966 (0.1209)
	E (SE)	NA	1.392 (0.648)	18.918 (8.556)	0.8444 (0.2371)
	Length (SE)	-0.0037 (0.0024)	-0.064 (0.027)	NA	-0.0240 (0.0099)
	Sex (SE)	0.0518 (0.0142)	NA	-3.578 (2.271)	NA
Selenium (Se)					
	Sample size	18	46	47	47
Regression on E:	R^2	3%	19%	21%	<1%
Coefficients:	Intercept (SE)	0.0758 (0.0264)	0.9651 (0.1561)	0.6228 (0.2179)	0.1738 (0.0169)
	E (SE)	0.0215 (0.0309)	0.7007 (0.2177)	1.0663 (0.3040)	0.0063 (0.0235)
Best regression:	R^2	0%	19%	30%	5%
Coefficients:	Intercept (SE)	0.0934 (0.0078)	0.9651 (0.1561)	0.2799 (0.2516)	0.1896 (0.0093)
	E (SE)	NA	0.7007 (0.2177)	NA	NA
	Length (SE)	NA	NA	0.0971 (0.0221)	NA
	Sex (SE)	NA	NA	NA	-0.0169 (0.0113)

Cont.

Table 5 cont.

		Blubber	Kidney	Liver	Muscle
Zinc (Zn)					
	Sample size	11	46	47	47
Regression on E:	R^2	5%	<1%	<1%	12%
	Coefficients:				
	Intercept (SE)	-0.305 (4.711)	20.387 (9.528)	42.472 (10.836)	15.813 (10.007)
	E (SE)	0.236 (0.330)	0.374 (0.706)	-0.497 (0.805)	1.862 (0.739)
Best regression:	R^2	19%	20%	35%	12%
Coefficients:	Intercept (SE)	-2.886 (4.182)	8.292 (5.369)	9.700 (5.566)	15.813 (10.007)
	E (SE)	NA	NA	NA	1.862 (0.739)
	Length (SE)	0.520 (0.363)	1.557 (0.471)	2.386 (0.489)	NA
	Sex (SE)	NA	NA	NA	NA

and E is included only in the model for kidney. This may be because means and standard deviations of Pb concentration are low in all tissues, with Pb undetectable in many samples (Table 4). Low Pb levels for many cetaceans have previously been reported (Dehn *et al*., 2006; Meador *et al*., 1999; Woshner *et al*., 2001b).

Arsenic

Arsenic was reported (Table 4) with E<MLR in 26 (54%) of the 48 epidermal samples because few whales in the 2002/03 dataset had concentrations high enough to be measured by ICP-MS. It was similarly low in all other tissues, except blubber. Comparing means between the two analytical techniques is inappropriate when greater than 50% of the samples are below detection; the difference in MLR is driving the difference in means by allowing more 1996/97 samples to have reported low concentrations and thus a lower reported mean.

Based on the 12 whales with E≥MLR and As also measured in blubber, none of the three predictor variables is of value as a predictor of As in blubber. When E≥MLR, E is the best single predictor of As in kidney, liver and muscle, explaining ≥79% of the variability in the concentration of As, with higher values of E associated with higher concentrations in those tissues. E is the only predictor of As in the kidney chosen by AIC. Length is added to E in the best regressions for liver and muscle chosen by AIC. The above regressions for those tissues are greatly influenced by the one (in the case of kidney and muscle) or two (in the case of liver) whales with concentrations of As high enough to be detected using ICP-MS; these concentrations are all higher than those in the AAS dataset. The relatively large number of samples with E<MLR but a concentration in kidney, liver or muscle at or above the median, or vice-versa, suggests that E might not be strongly predictive for these tissues. For example, there are 15 whales with both liver concentration and E<MLR, but 16 whales with E<MLR and high liver concentration or vice-versa. More samples with concentrations >MLR in all tissues are needed before firm conclusions can be drawn concerning the predictive power of epidermal As concentration for its concentration in other tissues.

CONCLUSIONS

Using concentrations of elements in epidermal tissue to predict corresponding elemental concentrations in other tissues (blubber, kidney, liver and muscle) does not appear to be a sound method based on our evaluation of bowhead whale samples from 1996, 1997, 2002 and 2003 (providing a sample size of 40 or more whales for all tissues except blubber). Epidermal concentration had no predictive value for Cu, Mn, Pb, Se or Zn in any of the other tissues evaluated. Epidermal concentrations of the four other elements considered were predictive for some other tissues. Arsenic could be predicted in kidney, liver and muscle, but not blubber, although the distribution of samples with concentrations below the MLR and the small sample sizes suggest that these results should be interpreted with caution. Epidermal concentrations of Cd were strongly predictive for blubber and weakly predictive for muscle concentrations. Epidermal concentrations of Hg were weakly predictive of blubber, liver and muscle concentrations. Epidermal concentrations of Mg were strongly predictive in blubber, kidney and liver but only weakly predictive in muscle. Thus, if investigators wish to develop an understanding of concentrations of essential and toxic elements in non-epidermal tissues, this cannot be accomplished via epidermal biopsy alone. One could monitor epidermis for its own sake (e.g. to assess whether it accumulates elements of toxicologic concern or could be a tissue affected by mineral deficiency) and might detect temporal or spatial trends. However, it has been shown here that epidermal biopsy cannot predict concentrations in four key tissues in bowhead whales in most cases. This ineffectiveness of epidermal element concentrations to reflect concentrations in internal tissues is likely true for other mysticetes and perhaps for cetaceans in general. At a minimum, before using epidermal biopsies to predict internal tissue concentrations of elements, researchers must establish that a sound scientific basis exists for doing so. Such proof must be specific to the elements, species and tissues in question as well as based upon statistically adequate sample sizes.

ACKNOWLEDGEMENTS

This study was funded through the North Slope Borough by the US Dept of Commerce, NOAA award #NA170Z2054, a Coastal Impact Assistance Program (CIAP) award. The authors gratefully acknowledge the generous provision of bowhead whale samples by the whale-hunting captains and crews (Barrow Whaling Captains Association, Barrow, Alaska) and the cooperation of the Alaska Eskimo Whaling Commission (AEWC, Barrow, Alaska, USA). In particular, we recognise Mr. Elijah Rock (Point Hope, Alaska) for requesting that we include nutrients analyses in tissue chemical composition studies of bowhead whales. Sample collection and logistics related to the shipment of samples to analytical laboratories were conducted by numerous NSB Dept. of Wildlife Management staff with special thanks to B. Akootchook and J.C. George. We thank T. Rowles (Marine Mammal Health and Stranding Response Program,

Washington, DC), S. Moore and M. Gosho (NOAA, Seattle, WA) for assistance in acquiring the necessary permits for collection and shipping (US Marine Mammal Protection Act Permit No. 782-1399 and 932-1489-05).

REFERENCES

Aastrup, P., Riget, F., Dietz, R. and Asmund, G. 2000. Lead, zinc, cadmium, mercury, selenium and copper in Greenland caribou and reindeer (*Rangifer tarandus*). *Sci. Total Environ.* 245: 149-59.

Akaike, H. 1973. Information theory and an extension of the maximum likelihood principle. pp.267-81. *In*: Petran, B.N. and Csaáki, F. (eds). *International Symposium on Information Theory*. Akademia Kiado, Budapest, Hungary. 451pp.

Arctic Monitoring and Assessment Programme. 1998. *AMAP Assessment Report: Arctic Pollution Issues*. Arctic Monitoring and Assessment Programme (AMAP), Oslo, Norway. xii+859pp.

Arctic Monitoring and Assessment Programme. 2002. *Arctic Pollution 2002*, Oslo, Norway. 111pp.

André, J.M., Boudou, A. and Ribeyre, F. 1991. Mercury accumulation in Delphinidae. *Water Air Soil Pollut.* 56: 187-201.

Beck, K.M., Fair, P., McFee, W. and Wolf, D.L. 1997. Heavy metals in livers of bottlenose dolphins stranded along the South Carolina coast. *Mar. Poll. Bull.* 34: 734-39.

Becker, P.R., Krahn, M., Mackey, E.A., Demiralp, R., Schantz, M., Epstein, M.S., Donais, M.K., Porter, B.J., Muir, D.C.G. and Wise, S.A. 2000. Concentrations of polychlorinated biphenyls (PCBs), chlorinated pesticides, and heavy metals and other elements in tissues of belugas, *Delphinapterus leucas*, from Cook Inlet, Alaska. *Mar. Fish. Rev.* 62: 81-98.

Beckmen, K., Duffy, L.K., Zhang, X. and Pitcher, K.W. 2002. Mercury concentrations in the fur of Steller sea lions and nothern fur seals from Alaska. *Mar. Poll. Bull.* 44: 1130-35.

Born, E.W., Renzoni, A. and Dietz, R. 1991. Total mercury in hair of polar bears (*Ursus maritimus*) from Greenland and Svalbard. *Polar Res.* 9(2): 113-20.

Bratton, G., Flory, W., Spainhour, C.B. and Haubold, E.M. 1997. Assessment of selected heavy metals in liver, kidney, muscle, blubber and visceral fat of Eskimo harvested bowhead whales *Balaena mysticetus* from Alaska's north coast. Final report submitted to the North Slope Borough Dept. of Wildlife Management, Barrow, Alaska (unpublished). 233pp.

Bustamante, P., Garrigue, C., Breau, L., Caurant, F., Dabin, W., Greaves, J. and Dodemont, R. 2003. Trace elements in two odontocete species (*Kogia breviceps* and *Globicephala macrorhynchus*) stranded in New Caledonia (South Pacific). *Environ. Pollut.* 124: 263-71.

Cooper, L.W., Larsen, I.L., O'Hara, T.M., Dolvin, S., Woshner, V. and Cota, G.F. 2000. Radionuclide contaminant burdens in Arctic marine mammals harvested during subsistence hunting. *J. Arctic Inst. N. America* 53(2): 174-82.

De Wit, C.A., Fisk, A.T., Hobbs, K.E., Muir, D.C.G., Kallenborn, R., Krahn, M., Norstrom, R. and Skaare, J. 2004. Persistent organochlorine pollutants. *In*: De Wit, C.A., Fisk, A.T., Hobbs, K.E. and Muir, D.C.G. (eds). *Arctic Monitoring and Assessment Programme (AMAP) II*, Oslo, Norway.

Decataldo, A., Di Leo, A., Giandomenico, S. and Cardellicchio, N. 2004. Association of metals (mercury, cadmium and zinc) with metallothionein-like proteins in storage organs of stranded dolphins from the Mediterranean Sea (southern Italy). *J. Environ. Monit.* 6: 361-67.

Dehn, L.A., Follman, E., Rosa, C., Duffy, L.K., Thomas, D.L., Bratton, G.R., Taylor, R.J. and O'Hara, T.M. 2006. Stable isotope and trace element status of subsistence hunted bowhead (*Balaena mysticetus*) and beluga whales (*Delphinapterus leucas*) in Alaska and gray whales (*Eschrichtius robustus*) in Chukotka. *Mar. Poll. Bull.* 52: 301-19. [Available on web].

Elkin, B.T. and Bethke, R.W. 1995. Environmental contaminants in caribou in the Northwest Territories, Canada. *Sci. Total Environ.* 160/161: 307-21.

Flynn, A., Franzmann, A.W. and Arneson, P.D. 1975. Sequential hair shaft as an indicator of prior mineralization in the Alaskan moose. *J. Anim. Sci.* 41: 906-10.

Flynn, A., Franzmann, A.W., Arneson, P.D. and Oldemeyer, J.L. 1977. Indications of copper deficiency in a sub-population of Alaskan moose. *J. Nutr.* 107: 1182-89.

Ford, J. and Hasselbach, L. 2001. Heavy metals in mosses and soils on six transects along the Red Dog Mine haul road. Alaska National Park Service No. NPS/AR/NRTR-2001/38. [Available from: *www.nps.gov/akso*].

Fossi, M.C., Marsili, L., Lauriano, G., Fortuna, C., Canese, S., Ancora, S., Leonzio, C., Romeo, T., Merino, R., Abad, E. and Jimenez, B. 2004. Assessment of toxicological status of a SW Mediterranean segment population of striped dolphin (*Stenella coeruleoalba*) using skin biopsy. *Mar. Environ. Res.* 58(2-5): 269-74.

Frank, I.A., Galgan, V. and Petersson, L.R. 1994. Secondary copper deficiency, chromium deficiency and trace element imbalance in the moose: effect of anthropogenic activity. *Ambio* 23: 315-17.

Frodello, J.P., Romeo, M. and Viale, D. 2000. Distribution of mercury in the organs and tissues of five toothed whale species of the Mediterranean. *Environ. Pollut.* 108: 447-52.

Fujise, Y., Honda, K., Tatsukawa, R. and Mishima, S. 1988. Tissue distribution of heavy metals in Dall's porpoise in the northwestern Pacific. *Mar. Poll. Bull.* 19(5): 226-30.

Gamberg, M. and Scheuhammer, A.M. 1994. Cadmium in caribou and muskoxen from the Canadian Yukon and Northwest Territories. *Sci. Total Environ.* 143: 221-34.

Gauthier, J.M., Dubeau, H. and Rassart, E. 1998. Mercury-induced micronuclei in skin fibroblasts of beluga whales. *Environ. Toxicol. Chem.* 17(12): 2487-93.

Gogan, P.J.P., Jessup, D.A. and Akeson, M. 1989. Copper deficiency in tule elk at Point Reyes, California. *J. Range Manage.* 42: 233-38.

Hansen, L.J., Schwacke, L.H., Mitchum, G.B., Hohn, A.A., Wells, R.S., Zolman, E.S. and Fair, P.A. 2004. Geographic variation in polychlorinated biphenyls and organochlorine pesticide concentrations in the blubber of bottlenose dolphins from the US Atlantic coast. *Sci. Total Environ.* 319: 147-72.

Honda, K. and Tatsukawa, R. 1983. Distribution of cadmium and zinc in tissues and organs, and their age related changes in striped dolphins, *Stenella coeruleoalba*. *Arch. Environ. Contam. Toxicol.* 15: 543-50.

Honda, K., Tatsukawa, R., Itano, K., Miyazaki, N. and Fujiyama, T. 1983. Heavy metal concentrations in muscle, liver and kidney tissue of striped dolphin, *Stenella coeruleoalba*, and their variations with body length, weight, age and sex. *Agric. Biol. Chem.* 47(6): 1,219-28.

Ikemoto, T., Kunito, T., Watanabe, I., Yasunaga, G., Baba, N., Miyazaki, N., Petrov, E. and Tanabe, S. 2004. Comparison of trace element accumulation in Baikal seals (*Pusa sibirica*), Caspian seals (*Pusa caspica*) and northern fur seals (*Callorhinus ursinus*). *Environ. Pollut.* 127(1): 83-97.

Korsrud, G.O., Meldrum, J.B., Slaiisbury, C.D., Houlahan, B.J., Saschenbrecker, P.W. and Tittiger, F. 1985. Trace element levels in liver and kidney from cattle, swine and poultry slaughtered in Canada. *Can. J. Comp. Med.* 49: 159-63.

Kuehl, D.W. and Haebler, R. 1995. Organochlorine, organobromine, metal and selenium residues in bottlenose dolphins (*Tursiops truncatus*) collected during an unusual mortality event in the Gulf of Mexico. *Arch. Environ. Contam. Toxicol.* 28: 494-99.

Kuehl, D.W., Haebler, R. and Potter, C. 1994. Coplanar PCB and metal residues in dolphins from the US Atlantic coast including Atlantic bottlenose obtained during the 1987/88 mass mortality. *Chemosphere* 28: 1,245-53.

Kunito, T., Watanabe, I., Yasunaga, G., Fujise, Y. and Tanabe, S. 2002. Using trace elements in skin to discriminate the populations of minke whales in the southern hemisphere. *Mar. Environ. Res.* 53: 175-97.

Larter, N.C. and Nagy, J.A. 2000. A comparison of heavy metal levels in the kidneys of High Arctic and mainland caribou populations in the Northwest Territories of Canada. *Sci. Total Environ.* 246: 109-19.

Law, R.J., Fileman, C.F., Hopkins, A.D., Baker, J.R., Harwood, J., Jackson, D.B., Kennedy, S., Martin, A.R. and Morris, R.J. 1991. Concentrations of trace metals in the livers of marine mammals (seals, porpoises and dolphins) from waters around the British Isles. *Mar. Poll. Bull.* 22(4): 183-91.

Lowry, L. 1993. Foods and feeding ecology. pp.201-38. *In*: Burns, J.J., Montague, J.J. and Cowles, C.J. (eds). *The Bowhead Whale*. Society for Marine Mammalogy, Lawrence, Kansas. xxxvi+787.

Mackey, E.A., Oflaz, R.D., Epstein, M.S., Buehler, B., Porter, B.J., Rowles, T., Wise, S.A. and Becker, P.R. 2003. Elemental composition of liver and kidney tissues of rough-toothed dolphins (*Steno bredanensis*). *Arch. Environ. Contam. Toxicol.* 22: 523-32.

Marcovecchio, J.E., Moreno, V.J., Bastida, R.O., Gerpe, M.S. and Rodríguez, D.H. 1990. Tissue distribution of heavy metals in small cetaceans from the southwestern Atlantic Ocean. *Mar. Poll. Bull.* 21(6): 299-304.

Meador, J.P., Ernest, D.W., Hohn, A., Tilbury, K.L., Gorzelany, J., Worthy, G. and Stein, J.E. 1999. Comparison of elements in bottlenose dolphins stranded on the beaches of Texas and Florida in the Gulf of Mexico over a one-year period. *Arch. Environ. Contam. Toxicol.* 36: 87-98.

Microsoft Corporation. 1999. *Microsoft Excel 2000*. Microsoft Corporation, Redmond, WA.

Monaci, F., Borrell, A., Leonzio, C., Marsili, L. and Calzada, N. 1998. Trace elements in striped dolphins (*Stenella coeruleoalba*) from the western Mediterranean. *Environ. Pollut*. 99(1): 61-8.

Nigro, M., Campana, A., Lanzillotta, E. and Ferrara, R. 2002. Mercury exposure and eliminiation rates in captive bottlenose dolphins. *Mar. Poll. Bull*. 44: 1071-75.

O'Hara, T.M., Carroll, G., Barboza, P., Mueller, K., Blake, J.B., Woshner, V. and Willetto, C. 2001. Mineral and heavy metal status as related to a mortality event and poor recruitment in a moose population in Alaska. *J. Wildl. Dis*. 37(3): 509-22.

O'Hara, T.M., George, J.C., Blake, J., Burek, K., Carroll, G., Dau, J., Bennett, L., McCoy, C.P., Gerard, P. and Woshner, V. 2003. Investigation of heavy metals in a large mortality event in caribou of northern Alaska. *Arctic* 56(2): 125-35.

O'Hara, T.M., Krahn, M.M., Boyd, D., Becker, P.R. and Philo, M. 1999. Organochlorine contaminant levels in eskimo harvested bowhead whales of Arctic Alaska. *J. Wildl. Dis*. 35(4): 741-52.

Puls, R. 1994. *Mineral Levels in Animal Health: Diagnostic Data*. Sherpa International, Clearbrook, British Columbia, Canada.

Roditi-Elasar, M., Kerem, D., Hornung, H., Kress, N., Shohan-Frider, E., Goffman, O. and Spanier, E. 2003. Heavy metal levels in bottlenose and striped dolphins off the Mediterranean coast of Israel. *Mar. Poll. Bull*. 46: 491-521.

Stein, J.E., Tilbury, K.L., Meador, J.P., Gorzelany, J., Worthy, G. and Krahn, M. 2003. Ecotoxicological investigations of bottlenose dolphin (*Tursiops truncatus*) strandings: accumulation of persistent organic chemicals and metals. pp.458-88. *In*: Vos, J.G., Bossart, G.D., Fournier, D. and O'Shea, T.J. (eds). *Toxicology of Marine Mammals*. Taylor and Francis, New York.

Suydam, R., George, J.C., O'Hara, T.M., Hanns, C. and Sheffield, G. 2004. Subsistence harvest of bowhead whales (*Balaena mysticetus*) by Alaskan Eskimos during 2003. Paper SC/56/BRG11 presented to the IWC Scientific Committee, July 2004, Sorrento, Italy (unpublished). 5pp. [Paper available from the Office of this Journal].

Suydam, R., George, J.C., O'Hara, T.M. and Sheffield, G. 2003. Subsistence harvest of bowhead whales by Alaskan Eskimos during 2002. Paper SC/55/BRG5 presented to the IWC Scientific Committee, May 2003, Berlin (unpublished). 5pp. [Paper available from the Office of this Journal].

Trefry, J.H., Rember, R.D., Trocine, R.P. and Brown, J.S. 2003. Trace metals in sediments near offshore oil exploration and production sites in the Alaskan Arctic. *Environ. Geol*. 45(2): 149-60.

Underwood, E.J. 1977. *Trace Elements in Human and Animal Nutrition*. Academic Press, New York. 196pp.

Venables, W.N. and Ripley, B.D. 1999. *Modern Applied Statistics with S-Plus*. 3rd ed. Springer-Verlag Inc., New York. xi+501pp.

Wagemann, R., Innes, S. and Richard, P.R. 1996. Overview and regional and temporal differences of heavy metals in Arctic whales and ringed seals in the Canadian Arctic. *Sci. Total Environ.* 186: 41-66.

Wiig, Ø., Renzoni, A. and Gjertz, I. 1999. Levels of cadmium and mercury in the hair of Atlantic walruses (*Odobenus rosmarus rosmarus*) from Svalbard, Norway. *Polar Biol*. 21(5): 343-46.

Woods, C.M. and Van Vleet, E.S. 1996. Copper, cadmium and zinc in liver, kidney and muscle tissues of bottlenose dolphins (*Tursiops truncatus*) stranded in Florida. *Mar. Poll. Bull*. 32(12): 886-89.

Woshner, V.M., O'Hara, T.M., Bratton, G.R. and Beasley, V.R. 2001a. Concentrations and interactions of selected essential and non-essential elements in ringed seals and polar bears of Arctic Alaska. *J. Wildl. Dis*. 37(4): 711-21

Woshner, V.R., O'Hara, T.M., Bratton, G.R., Suydam, R.S. and Beasley, V.T. 2001b. Concentrations and interactions of selected essential and non-essential elements in bowhead and beluga whales of Arctic Alaska. *J. Wildl. Dis*. 37(4): 633-710.

Yang, Y., Kunito, T., Tanabe, S., Amano, M. and Miyazaki, N. 2002. Trace elements in skin of Dall's porpoises (*Phocoenoides dalli*) from the northern waters of Japan: an evaluation for utilization as non-lethal tracers. *Mar. Poll. Bull*. 45: 230-36.

Ylitalo, G.M., Matkin, C.O., Buzitis, J., Krahn, M.M., Jones, L.L., Rowles, T. and Stein, J.E. 2001. Influence of life-history parameters on organochlorine concentrations in free-ranging killer whales (*Orcinus orca*) from Prince William Sound, AK. *Sci. Total Environ.* 281: 183-203.

Date received: April 2006
Date accepted: February 2007