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Trace element concentrations in blood of harbor seals (*Phoca vitulina*) from the Wadden Sea

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Abstract

Concentrations of 23 elements (Be, Al, K, Ca, Ti, V, Cr, Mn, Fe, Co, Ni, Cu, Zn, As, Se, Rb, Sr, Mo, Pd, Cd, Sn, Pt, Pb) were evaluated in whole blood samples of live harbor seals (*Phoca vitulina*) from two different locations in the Wadden Sea, the Lorenzenplate in Germany, and the Danish island Rømø. Elemental blood levels were compared to data from literature of seals, other marine mammals and humans. While homeostatically controlled elements showed no differences, concentrations of As, Cr, Mn, Mo, Se, and V were higher than human levels. Furthermore, animals from both locations showed significant geographical differences in whole blood concentrations of Al, Mn, Cu, and Pt. These findings could be explained by differences in feeding areas. The element pattern was not affected by gender. In conclusion, these findings indicate an impact of the environment on biochemical blood parameters of the harbor seals. The significant differences of elements in blood samples of two groups of seals, which were associated with geographical variations

of prey support the use of element pattern in blood as tool for investigation of environmental impact on seals.

Keywords: Trace elements, Metals, Blood, ICP-MS, Harbor seals, North Sea

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1. Introduction

The harbor seal (*Phoca vitulina*) has been identified as model species to investigate effects of environmental contaminants on marine mammals by the Marine Mammal Commission (O´Shea et al., 1999; Reddy et al., 2001). Within the Trilateral Monitoring and Assessment Program (TMAP 2000) harbor seals at the Danish-German-Dutch Wadden Sea coast were investigated.

Numerous reports have documented the concentration of various elements in marine mammal tissues such as liver, kidney, muscle or blubber of post mortem examinated animals (O´Shea, 1999; Das et al., 2003). In Northern Europe, seals from different locations were investigated: Tissues of harbor, gray *(Halichoerus grypus)* and ringed seals (*Phoca hispida)* collected in various areas of the Baltic Sea and the Swedish west coast were investigated (Frank et al., 1992; Olsson et al., 1994; Ciesielski et al., 2006). Liver samples of harbor seals from Norwegian waters (Skaare et al., 1990), of common and gray seals *(Halichoerus grypus)* from waters around the British Isles (Law et al., 1991), the Welsh Coast and Irish Sea (Law et al. 1992) were analyzed. Trace elements were detected in tissues of gray seals from the

Faroe Islands (Bustamante et al., 2004), of ringed, harp (*Pagophilus groenlandicus*), and hodded seals (*Cystophora cristata*) from Greenland (Dietz et al., 1996). In conclusion all these studies showed local differences in element concentration of tissues from seals living in different polluted areas. But there was a lack of investigation of harbor seals from the Wadden Sea over the last years.

Evaluation of current contaminant effects on a wild marine mammal population requires samples collected from living animals. To describe the impact of metals on the immune system of free-ranging seals of the North Sea blood was investigated before (Pillet et al., 2000; Kakuschke et al., 2005). Feeding studies with different contaminated fish showed differences in immunological blood parameters in seals e.g. T-, B- lymphocytes, and natural killer cells (de Swart et al., 1996). The occurrence of mass mortalities among seals inhabiting contaminated marine areas have led to speculation about the possible involvement of immunosupression associated with environmental pollution (de Koeijer et al.; 1998). As top predators seals bioaccumulate persistant substances through their position in the food web. The harbor seals of the North Sea consume a vide variety of prey, mainly fish and cephalopods with geographical and seasonal differences in their diet (Hall et al., 1998; Pierce et al., 2003).

To asses the nutritional status, blood chemistry and haematological profiles were used as an index of foraging status or health (Trumble et al., 2006). Reference ranges have been reported for both free-ranging and captive harbor seals (Morgan et al., 1998*;* Griesel et al., 2006).

However, only few studies reported values for trace elements in blood of living seals for a minor number of metals. Therefore, the multi-element analysis of blood could be a useful method to combine the measurement of electrolytes as well as essential and toxic trace elements in living animals.

The aim of this study was to investigate blood of free-ranging harbor seals from the Wadden Sea and to evaluate whether element levels were affected by the habitats or differ with gender. Furthermore, it was aimed to find out whether these blood values could act as supporting parameters for the existing monitoring program to describe the status of the common seal population and to obtain hints for different polluted areas of the Wadden Sea.

2. Material and methods

2.1. Sample collection and preparation

Fresh whole blood samples were collected from 28 harbor seals. They were captured in by net during six campaigns in 2003 and 2004. 16 seals were examined and released in the German Wadden Sea at Lorenzenplate (54°30`N, 8°20`E) and 12 animals in the Danish Wadden Sea at Rømø (55°15´N, 8°30´E) (Fig. 1). Gender and standard length were recorded and blood samples were taken from each seal (Table 1). All animals were clinically examined. They were in normal nutritional status and no clinical symptoms or diseases were observed.

Blood samples were obtained from the epidural vertebral vein. Whole blood samples were collected in special Lithium Heparin monovettes for metal analysis (Sarstedt, Nümbrecht, Germany) and stored at -20 °C. For multi-element determination a microwave digestion system (MarsXpress, CEM GmbH, Kamp-Lintfort, Germany) was applied. 500 μL of whole blood was pipetted into perfluoralkoxy (PFA) vessels. 2000 μL sub-boiled nitric acid, 1000 μL of hydrogen peroxide, and 50 μL internal standard (Y=1 mg/L, Merck, Darmstadt, Germany) were added and heated in a three step program up to 180 °C.

2.2. Analytical techniques

Measurements were performed by three different analytical methods: K, Ca, Fe, Cu, Zn, As, Se, Rb, and Sr were determined by Total-X-Ray-Fluorescence Spectrometry (Atomika TXRF 8030 C, CAMECA GmbH, Oberschleissheim, Germany). The spectrometer is equipped with a 3-kW X-ray tube which containes a Molybdenum-Tungsten (Mo-W) alloy anode and a double-multilayer monochromator. The Molybdenum Kα exitation was selected for detecting the elements. The counting time was set to 1000 s. Yttrium as internal standard was used to calculate all other element concentrations. Digested samples (20 μL) were pipetted onto quarz glass sample carriers and evaporated to dryness.

Trace elements with lower concentrations were determined by inductively coupled plasma-mass spectrometer. ^{9}Be, ^{27}Al, ^{51}V, ^{53}Cr, ^{55}Mn, ^{59}Co, ^{60}Ni, ^{98}Mo, ^{108}Pd, ^{114}Cd, ^{120}Sn, ^{195}Pt, and ^{208}Pb were analyzed using an Inductively Coupled Plasma Mass Spectrometer with a collision cell (Agilent 7500c ICP-MS, Agilent Technologies, Tokyo, Japan). The sample introduction system consists of a PFA micro-flow nebulizer (Elemental Scientific, Omaha, NE, USA) and an autosampler ASX 500 (CETAC Technologies Omaha, NE, USA). The standard mode was used for Be, Al, V, Mn, Co, Mo, Pd, and Pb. For the remaining elements better results were obtained with hydrogen used as cell gas with optimum flow-rate of 3.5 mL/min (Air Liquide, Düsseldorf, Germany). After digestion, samples were diluted by 20. Matrix effects and instrumental drift of the ICP-MS were corrected by using Yttrium as an internal standard. For calculation external calibration was made by diluted standard solutions (Merck, Darmstadt, Germany).

The level of Ti was determined by a high-resolution sector field ICP-MS (ELEMENT I, Thermo Finnigan MAT GmbH, Bremen, Germany). Since the Ca concentration in blood and serum is very high, the Ti determination via the most abundant isotope ^{48}Ti via high-resolution ICP-MS is not possible because of the isobaric interference by ^{48}Ca (0.2%). Additionally, ^{46}Ti is excluded for determination due to the presence of ^{46}Ca (0.03%). Finally, overlap with $^{50}V^+$ and $^{50}Cr^+$ ion signals made determination using the ^{50}Ti isotope impossible. Therefore, the level of Ti was determined via the isotopes ^{47}Ti and ^{49}Ti. Quantification was carried out using single standard addition as calibration technique.

All sample handling and analysis were carried out in clean rooms. High-purity deionized water purified with a Milli-Q analytical-reagent grade water-purification system (Millipore Ellix with Milli-Q Element, MILLIPORE, Billerica, MA, USA) and high-quality concentrated nitric acid (Merck Suprapur®) were used for the preparation of reagents and standards. To exclude contamination with metals from needles used for sampling the first milliliters of blood were abolished.

The accuracy of all results and the liability of the analytical procedures were checked with the reference material Clin Check® Whole Blood Control Level II, Lot. No./Ch.-B.:212 (Recipe, Chemicals+Instruments, Munich, Germany). The limits of detection were calculated according to DIN 32 645. Results for the certified elements Cd, Co, Cr, Mn, Ni, Pb, Se, and Zn were presented in Table 2. Within 10 measurements relative standard deviations (RSD) were in between 4-13 % and recoveries range from 73-126 %. For the other elements spiking experiments were applied with RSD 3-25 %.

Marine mammal reference material was measured in connection with the participance in the NIST/NOAA 2005 Interlaboratory Comparison Exercise for trace elements in marine mammals (Christopher et al. 2007). Pygmy sperm whale (*Kogia breviceps*) liver homogenate (QC03LH3) served as the control standard, while white-sided dolphin (*Lagenorhynchus acutus*) liver homogenate (QC04LH4) served as the unknown. The elements Ag, As, Cd, Co, Cs, Cu, Fe, Mn, Mo, Rb, Se, Sn, V, and Zn were determined in both materials. The control material was measured 3 times with RSD 6-14 % and recoveries 87-112 % (Table 3). Within 5 measurements, the results for the unknown material were within the consensus range with RSD 6-13 %.

2.3. Data analysis

To justify distributional assumption of the data the one-sided Kolmogorov-Smirnov Goodness-of-Fit Test was used. All elements were also checked by visual inspection. All data thus obtained with no Gaussian distribution were log transformed (base e) (elements Al, Cr, Co, Ni, Se, Pd, Pt, Pb). Be, V, Ti, Cd, and Sn were not tested because of unsufficient data. A three factorial analysis of covariance (ANCOVA) with replicates based on gender and location and the interaction of gender and location as covariances were used to identify differences for each element. To obtain exact significances exact double-sided P-values were calculated. To avoid age specific differences only data from adult harbor seals were considered. Since testing for differences between the two locations considering a total of 18 metal concentrations separately, Fisher`s Omnibus test was used to adjust P-values for multiple testing. Statistical significance was designated as $P < 0.05$. All the statistical tests were conducted using SPSS (vers. 12.0.1 for Windows).

3. Results

3.1. Trace element content in blood

Concentration of 23 elements were measured in whole blood samples of 28 seals from two different locations. In Table 4 median values and concentration ranges of these elements are listed and ordered by their coefficient of variation (CV%). Mineral elements are in the order of 30-55 mg/L for Ca < K < 513-1137 mg/L for Fe and show minor variation in concentration level for all animals studied. The concentration of essential trace elements are in the range of 1.27 µg/L for Mo < Cr < Mn < Cu < Se < 4.6 mg/L for Zn. Potentially toxic elements show wide variation in element level concentration and their median values range from 0.09 µg/L for Sn < Be < Cd < Pb < Ni < 185 µg/L for As.

3.2. Differences in gender and locations

Our data revealed significant geographical differences ($\chi 2$=58.11,*df*=36, *p*=0.011) for elemental blood levels of the harbor seals of the Wadden Sea. No gender related differences were found (p>0.05) and there were no significant interactions between gender and location (p>0.05).

Animals caught at the Lorenzenplate showed higher element levels in blood for Mn (df=4.869, p =0.037) and Cu (df=4.844, p=0.038), whereas blood levels of Al (df=7.764, p=0.01) and Pt (df=5.625, p=0.026) were significantly higher for the animals from Rømø. Because less data were available for Cd and Ti, statistical evaluations were not possible. However, highest element values were also measured in seals from the isle of Rømø (Cd=3.10 µg/L, Ti=10.8 µg/L).

4. Discussion

Concentrations of metals in marine mammals are affected by a number of parameters, such as gender, age, geographical location, and prey type as well as changes in the environment over the last years. One part of this study compared element concentrations in seal blood with marine mammals and humans. Furthermore, differences between the two sampling locations were discussed.

4.1. Differences to other marine mammals and human blood levels

All major basic body functions of marine mammals noted to be comparable to terrestrial animals (King et al., 2001). So far only a few authors reported element blood values for whole blood of living harbor seals. Therefore concentrations measured in this study were compared to other marine mammals and humans (Table 5). It was possible to classificy elements in four groups.

The first group includes K, Ca, Fe, Cu, and Zn. These elements showed a very small range in concentration levels for all animals of this study. Their blood concentrations were believed to be homeostatically controlled and therefore their values varied little among and between species. Results for the elements K, Ca, Fe and Cu were comparable to studies of other seals (McConnell and Vaughan, 1983;

de Swart et al., 1995; Griesel et al., 2006), marine mammals (Nordøy and Thoresen, 2002; Larsen et al., 2002) and humans (Krachler and Irgolic, 1999; Alimonti et al., 2005). Though the Zn values (range 2.7-4.7 mg/L) were comparable to other marine mammals (Fujise et al., 1988; Baraj et al., 2001; Griesel et al., 2006), human values seem to be higher (mean 6.5 mg/L) (Sabbioni et al., 1992; Alimonti et al., 2005).

The values of the elements of a second group Al, Be, Ti, Co, Ni, Rb, Sr, Pd, Cd, and Pt are generally in the same order of magnitude of those reported for humans. While Dall´s porpoises showed comparable Ni and Cd levels (Fujise et al., 1988), other authors measured extremely high Cd values in marine mammals blood (Nielsen et al., 2000) an tissues *(*Dietz et el., 1998*;* Szefer et al., 2002). Higher levels of Cd in seal tissues of the Arctic in comparison with warmer, temperate regions as the Baltic Sea have been explained by diet (Fant et al., 2001). Crustaceans, the main diet for seals in the Arctic are rich in Cd. For the remaining elements of this group no literature on marine mammals could be found. Because the concentration of these trace elements show high inter-individual variability the distribution patterns may reveal the actual body burden of seals in the German and Danish Wadden Sea, and therefore, may reflect the exposure levels. High blood values for individual animals were measured for Al, Be and Pt.

Results for the elements Sn and Pb were lower than indicated normal human ranges. For Sn no literature value for blood is available for marine mammals. Pb levels in blood of other seals (Baraj et al., 2001), porpoises (Fujise et al., 1988) and whales (Nielsen et al., 2000) revealed the same range as the seals in this study. Other measurements demonstrated that marine mammals with relatively low environmental lead exposure have Pb levels within the predicted range of humans with similar low environmental lead exposure (Owen and Flegal, 1998). High Pb concentrations in human blood reflected the impact of traffic-induced lead emission

(Hashisho and El-Fandel, 2004) because humans are known to be exposed to Pb via the combustion of leaded gasoline (Rodamilans et al., 1996).

In contrast, the blood values of the last group V, Cr, Mn, As, Se, and Mo were significantly higher in seals than reported human levels. The concentration of As in the seal blood was 20 times higher than in human blood (Sabbioni et al., 2002; Daunderer, 2002). Though no literature values have been given for blood of marine mammals it is well known that marine biota tissue samples contain higher As concentration than terrestrial animals (Francesconi and Edmonds, 1998). Accumulation of As in tissues of seals was already observed (Goessler et al., 1998; Kubota et al., 2001).

While the concentration of V, Cr, Mn, and Se in blood of the seals in this study were ten times higher than reported human values (White et al., 1998; Daunderer, 2002; Alimonti et al., 2005), studies of other marine mammals showed comparable levels for Cr, Mn, and Se (Fujise et al., 1988; Nielsen et al., 2000; Baraj et al., 2001). No blood values for V in marine mammals could be found in literature but high V concentrations in tissues of seals were already reported (Saeki et al., 1999). The concentration of Mo in the seal blood is lightly higher than the Mo values of studied humans (Yoshida et al., 2006). No element levels for blood of marine mammals are reported.

As, Se, and Mo are present in high concentrations in sea water (Lavi and Alfassi, 1989; Schmolke et al., 2005). The high blood values of seals seem to reflect the natural adaptation to the marine ecosystem.

4.2. Influence of location

Significant site-based differences in concentration of elements in blood of seals corroborates work in the context of the TMAP (Schmolke et al., 2005; Bakker et al., 2005) that indicated local differences in water quality and metal concentration in sediment and indicator species. Element levels of Cd, Cu, Hg, Pb, and Zn were determined to evaluate background concentrations in 12 subareas of the three riparian states. Highest Cd levels in blue mussels were found in the area of Rømø (DK1, Figure 1), whereas Cu levels in mussels and flounders were elevated at the Lorenzenplate (SH3, Figure 1) (Bakker et al., 2005). The defined Wadden Sea subareas of the Wadden Sea Quality Status Report (QSR) are congruent to the water body types defined for the Water Framework Directive (WFD) of the EU (Jekel, 2005).

Harbor seals are opportunistic feeders, however benthic species are the predominant prey of harbor seals in the Wadden Sea (Behrends, 1985; Sievers, 1989; Orthmann, 2000; Siebert et al., 2006). Because benthic fish are sedentary, they might reflect contamination of a distinct location (Cain et al., 2000). Since the uptake of metals by marine mammals is predominately dietary, it can be expected that regional differences in concentrations in prey species would be reflected in marine mammals. Such regional variations in diets are described in other studies (Brown et al., 2001; Pierce and Santos, 2003). The differences in blood values of seals from Lorenzenplate and Rømø and the differences to other species might be caused by different foraging habitats or behavior and/or individual specialization for particular prey. Differences in foraging habitates of the investigated seals were obtained by tagged seals in that area (Figure 2, Adelung and Müller, 2005). Seals from Rømø showed increased targed-oriented movements around 54°45´ - 55°15 N and 7°40´ - 8°10´E, compared to seals from the Lorenzenplate which seem to prefer an area around 54°10´ - 54°30 N and 8°00´ - 8°40´E.

The significant differences of elements in blood samples of two groups of seals, which were associated with geographical variations of prey support the use of blood composition as index for nutritional status and environmental impact on seals.

4.3. Summary and conclusion

As mammals living in coastal areas and as top predators, harbor seals are markedly influenced by the status of the environment. Measuring element levels in blood of free ranging animals, trace element levels are more appropriate to reflect the current exposure situation associated with specific localization. The results of this study indicate the use of seal blood to monitor the influence of different conditions or contaminations of the Wadden Sea on living marine mammals. Further studies with more animals of different areas are needed to evaluate the use of particular elements in blood as biomarkers.

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Tables 1 - 5

Table 1

Haul out sites and biometry of live harbor seals (*Phoca vitulina*) sampled in the Wadden Sea, from 2003 to 2004

Table 2

Trace element concentration in reference material Recipe Clin Check® Whole Blood Control Level II.

Table 3

Trace element concentration in Pygmy sperm whale (*Kogia breviceps*) liver, results of the NIST/NOAA Interlaboratory Comparison Exercise 2005

Table 4

Element concentration (µg/L) in whole blood of harbor seals (*Phoca vitulina*) of the Wadden Sea, ordered by coefficient of variation (CV%)

Table 5

Element levels in whole blood of harbor seals of this study compared to other marine mammals and humans

Figures 1, 2

Fig. 1. The 12 sub-areas of the Wadden Sea Quality Status Report (QSR, Bakker et al., 2005). Seals catched at the different sub-areas★ (southern part of DK2 and southern part of SH3) showed significant differences in elemental blood composition in this study.

Fig. 2. At sea-distribution of tagged seals assert differences in foraging habitats for seals from the two different haul-out sites Lorenzenplate and Rømø (Adelung and Müller, 2005).

Tables to:

Trace element concentrations in blood of harbor seals (*Phoca vitulina*) from the Wadden Sea

Simone Griesel , Antje Kakuschke, Ursula Siebert, Andreas Prange

Tables 1 - 5

Table 1

Haul out sites and biometry of live harbor seals (*Phoca vitulina*) sampled in the Wadden Sea, from 2003 to 2004

location	gender	number	lenght (cm)	weight (kg)
L	m	12	144 - 177	53 - 100
	f	4	153 - 160	60 - 66
R	m	9	149 - 179	60 - 106
	f	3	148 - 173	64 - 91
		28		

L = Lorenzenplate, R = Rømø, m= male, f= female

Table 2

Trace element concentration in reference material Recipe Clin Check® Whole Blood Control Level II.

	Certified Recipe			Measured (*n*=10)			
Element	Mean value	Control range		Mean	± SD	% RSD	Recovery %
Cd	4.3	3.5	5.1	3.42	0.210	6.1	73 - 86
Co	4.7	3.7	5.7	5.06	0.251	5.0	99 -114
Cr	7.1	6.0	8.2	7.26	0.872	12.1	82 -121
Hg	15	12	18	n.a.			
Mn	27	22	32	31.3	1.29	4.1	110 -126
Ni	8.3	6.4	10.2	8.24	1.06	12.8	80 -113
Pb	304	258	350	272	11.3	4.2	84 - 96
Se	171	137	205	166	18.9	11.4	82 -116
Zn	2449	1954	2939	2611	236	9.1	90 -125

values are given in µg/L wet weight

n.a.= not analysed

Table 3

Trace element concentration in Pygmy sperm whale (*Kogia breviceps*) liver, results of the NIST/NOAA Interlaboratory Comparison Exercise 2005

Element	Recommended NIST/NOAA Mean value ± SD		Measured (*n*=3) Mean	± SD	% RSD	Recovery %
Ag	0.088	0.007	0.089	0.005	5.6	99 - 103
As	0.398	0.037	0.386	0.053	13.7	87 - 102
Cd	5.94	0.38	6.20	0.52	8.4	99 - 109
Co	0.071	0.003	0.072	0.004	9.8	95 - 107
Cs	0.0079	0.0003	0.0077	0.0007	10.3	88 - 104
Cu	2.74	0.19	2.751	0.21	7.6	95 - 106
Fe	694	45	675	72	10.7	90 - 102
Hg	3.65	0.1	n.a.			
Mn	1.43	0.015	1.40	0.16	11.4	89 - 106
Mo	0.211	0.07	0.209	0.012	5.7	98 - 101
Rb	1.61	1.18	1.63	0.14	8.6	97 - 107
Se	7.87	0.024	9.11	0.98	10.8	98 - 104
Sn	0.094	0.024	0.093	0.008	8.6	93 - 106
V	0.037	1.7	0.037	0.004	10.9	89 - 112
Zn	21.12	19.5	21.1	2.0	9.5	96 - 106

values are given in mg/kg wet weight

n.a.= not analysed

Table 4

Element concentration (µg/L) in whole blood of harbor seals (*Phoca vitulina*) of the Wadden Sea, ordered by coefficient of variation (CV%)

	Lorenzenplate (D), n=16		RømØ (DK), n=12		CV %, n=28
	Median	Range	Median	Range	
K	161 mg/L	131 - 197 mg/L	155 mg/L	138 - 183 mg/L	13
Zn	3436	2905 - 4568	3420	2730 - 4349	13
Ca	41.7 mg/L	29.8 - 55.0 mg/L	41.6 mg/L	32.7 - 52.9 mg/L	15
Cu [a]	878	604 - 1371	770	527 - 986	19(L), 17(R)
Fe	760 mg/L	520 - 1137 mg/L	738 mg/L	599 - 936 mg/L	20
Sr	42	25 - 63	47.0	34 - 70	25
Rb	70	52 - 149	72.5	52 - 99	35
Mn [a]	95.7	67.4 - 151	79.1	67.7 - 105	27(L), 16(R)
Se	899	591 - 2261	940	518 - 1372	42
V	0.82	<0.05 - 1.30	- [c]	<0.05 - 1.06	52
As	169	42 - 592	185	118 - 316	62
Mo	5.30	1.52 - 22.8	6.16	1.27 - 14.9	66
Ti [b]	1.70	1.21 - 4.08	- [c]	1.1 - 10.8	84
Sn	0.09	<0.06 - 0.47	- [c]	<0.06 - 0.16	156
Pb	0.98	<0.02 - 1.82	0.40	0.02 - 4.52	178
Cr	6.97	1.52 - 21.54	11.02	1.88 - 84.9	211
Ni	2.34	0.94 - 9.48	3.72	<0.38 - 25.7	251
Pt [a]	0.19	0.09 - 0.47	0.96	<0.04 - 8.30	66(L), 319(R)
Co	0.51	0.16 - 4 40	0.53	<0.02 - 7.56	348
Be	- [c]	<0.08 - 1.80	- [c]	<0.08 - 0.18	410
Cd	- [c]	<0.12 - 1.06	0.14	<0.12 - 3.10	488
Pd	0.24	<0.12 - 3.75	0.41	<0.12 - 5.00	530
Al [a]	3.34	<0.17 - 126	36.8	3.97 - 499	1192(L), 382(R)

Values are given in µg/L unless otherwise noted

R=Rømø, L=Lorenzenplate, n= sample size

a: significant local differences

b: for Ti nL=10, nR=4,

c: no median calculated because of unsufficient data

Table 5

Element levels in whole blood of harbor seals of this study compared to other marine mammals and humans

Element	Marine Mamals Blood concentration	Species, Source	Human Blood concentration	Source
comparable to other marine mammals and humans, homeostaticaly controlled elements				
K	160 ± 19 mg/L (= 4.09 mmo/L) 110 - 199 mg/L n=85	Habor Seal *Phoca vitulina* This study, includes data from Griesel et al. (2006)	P: 137 - 180 mg/L normal range	Alimonti et al. (2005)
	S: 141 - 164 mg/L n=22	Habor Seal *Phoca vitulina* De Swart et al. (1995)		
	S: 109 - 180 mgl/L n=11	Habor Seal *Phoca vitulina* McConnell et al. (1983)		
	S: 207 ± 23 mg/L n=14	Harp seal *Phoca groenlandica* Nordøy et al. (2002)		
	152 - 227 mg/L n=20	Northern elephant seal *Mirounga* Larsen et al. (2002)		
	S: 172 - 242 mg/L n=20	Northern elephant seal *Mirounga* Larsen et al. (2002)		
Ca	42.9 ± 8.3 mg/L (=1.07 mmol/L) 21.6 - 61.9 mg/L n=85	Habor Seal *Phoca vitulina* This study, includes data from Griesel et al. (2006)	66.3 ± 10.8 mg/L n=110 S: 63.1 ± 5.9 mg/L n=110	Alimonti et al. (2005) Alimonti et al. (2005)
	S: 92 - 108 mg/L n=11	Habor Seal *Phoca vitulina* De Swart et al. (1995)		
	S: 96 - 104 mg/L n=22	Habor Seal *Phoca vitulina* McConnell et al. (1983)		

	S: 104 ± 4 mg/L	Harp seal *Phoca groenlandica*		
	n=14	Nordøy et al. (2002)		
Fe	733 ± 86 mg/L	Habor Seal *Phoca vitulina*	550 ± 61 mg/L	Alimonti et al. (2005)
	485 - 1136 mg/L	This study,	n=110	
	n=85	includes data from Griesel et al. (2006)	500 - 1000 mg/L	Krachler&Irgolic (1999)
	829 - 915 µg/g	Dall`s porpoise *Phocoenoides dalli*		
	n=2	Fujise et al. (1988)		
	700 µg/g	Southern elephant seal *Mirounga leonina*		
	n=6	Baraj et al. (2001)		
	497 - 119 µg/g	Weddel seal *Leptonychotes weddellii*		
	n=3	Yamamoto et al. (1987)		
Cu	527 - 1371µg/L	Habor Seal *Phoca vitulina*	938 ± 141 µg/L	Alimonti et al. (2005)
	n=28	This study	n=110	
	821 ± 123 µg/L	Habor Seal *Phoca vitulina*		
	n=81	Griesel et al. (2006)		
	0.71 µg/g; 0.76 µg/g	Dall`s porpoise *Phocoenoides dalli*		
	n=2	Fujise et al. (1988)		
	1.04 µg/g	Southern elephant seal *Mirounga leonina*		
	n=6	Baraj et al. (2001)		
	822 µg/L	Gray seal *Halichoerus grypus*		
		Kakuschke et al. (2006)		
	0.11 - 5.05 µg/g	Weddel seal *Leptonychotes weddellii*		
	n=3	Yamamoto et al. (1987)		
Zn	3.4 ± 0.5 mg/L	Habor Seal *Phoca vitulina*	6.7 ± 0.9 mg/L	Alimonti et al. (2005)
	2.6 - 6.2 mg/L	This study,	n=110	
	n=85	includes data from Griesel et al. (2006)	6.3 mg/L	Sabbioni et al. (1992)
	3.9; 4.6 µg/g	Dall`s porpoise *Phocoenoides dalli*	n=502	
	n=2	Fujise et al. (1988)		
	3.13 µg/g	Southern elephant seal *Mirounga leonina*		
	n=6	Baraj et al. (2001)		
	3.15 mg/L	Gray seal *Halichoerus grypus*		

		Kakuschke et al. (2006)		
	4.02 - 17.2 µg/g n=3	Weddel seal *Leptonychotes weddellii* Yamamoto et al. (1987)		
comparable to humans, high inter individual variability				
Al	< 0.17 - 500 µg/L n=28	Habor Seal *Phoca vitulina* This study	17.0 ± 9.4 µg/L n=110	Alimonti et al. (2005)
			S/P: 3.72 µg/L n=8	Cornelis et al (1994)
Be	< 0.08 - 1.80 µg/L n=28	Habor Seal *Phoca vitulina* This study	0.42 ± 0.2 µg/L n=110	Alimonti et al. (2005)
Ti	1.13 - 10.9 µg/L n=14	Habor Seal *Phoca vitulina* This study	11.2 µg/L n=6	Bockmann et al. (2000)
Co	0.52 µg/L < 0.02 - 7.56 µg/L n=28	Habor Seal *Phoca vitulina* This study	0.12 ± 0.08 µg/L n=110	Alimonti et al. (2005)
			< 0.9 µg/L normal range	Daunderer (2002)
			0.39 ± 0.13 µg/L n=441	Sabbioni et al. (1992)
Ni	2.41 µg/L <0.38 - 25.74 µg/L n=28	Habor Seal *Phoca vitulina* This study	0.89 ± 0.6 µg/L n=110	Alimonti et al. (2005)
			< 3.3 µg/L normal range	Daunderer (2002)
	≤ 0.05 µg/g n=2	Dall`s porpoise *Phocoenoides dalli* Fujise et al. (1988)	0.34 - 2.3 µg/L n=130	Templeton et al. (1994)
Rb	78 µg/L 33 - 149 µg/L n=85	Habor Seal *Phoca vitulina* This study, includes data from Griesel et al. (2006)	78 – 317 µg/L normal range	Daunderer (2002)
Sr	46 µg/L 17 - 95 µg/L n=85	Habor Seal *Phoca vitulina* This study, includes data from Griesel et al. (2006)	27.3 ± 11.8 µg/L n=110	Alimonti et al. (2005)
			< 19.8 µg/L normal range	Daunderer (2002)

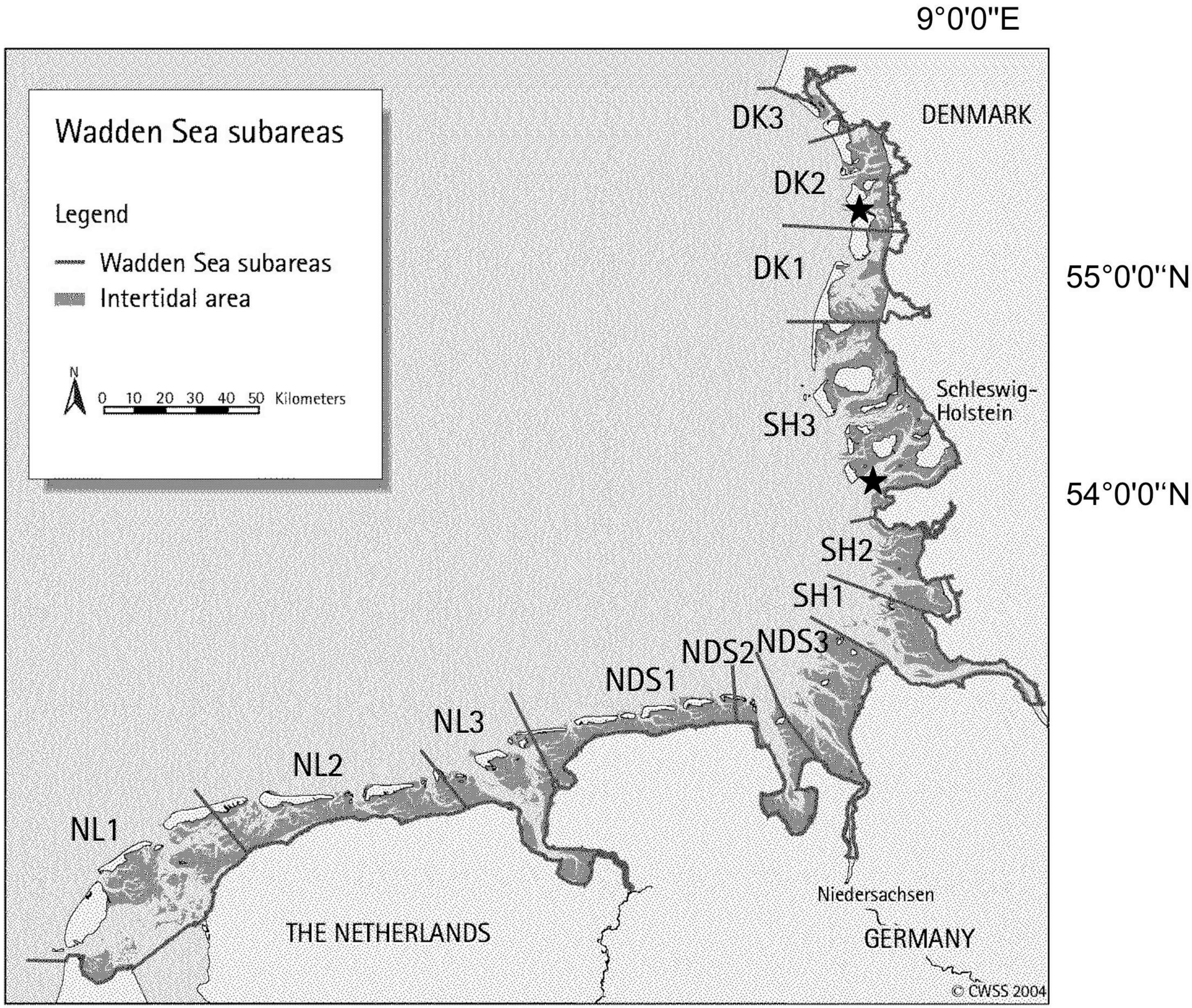
Pd	0.25 µg/L	Habor Seal *Phoca vitulina*	< 0.4 µg/L	Daunderer (2002)
	< 0.12 - 5.00 µg/L n=28	This study	normal range	
Cd	< 0.12 - 3.10 µg/L n=28	Habor Seal *Phoca vitulina* This study	< 1.7 µg/L normal range	Daunderer (2002)
	3.8 ng/g n=6	Southern elephant seal *Mirounga leonina* Baraj et al. (2001)	0.16 - 3.2 µg/L n=210	White and Sabbioni (1998)
	9.2 - 33.4 µg/L n=6	Pilot Whale *Globicephala melas* Nielsen et al. (2000)	1.2 µg/L n=143	Cornelis et al (1994)
	930 - 31 100 µg/L n=4	Sperm Whale *Physeter catodon* Nielsen et al. (2000)	0.6 ± 0.3 µg/L n=900	Sabbioni et al. (1992)
	ER: 123 µg/L; P: 50.4 µg/L n=25	Pilot Whale *Globicephala melas* Caurant & Amiard-Triquet (1995)		
	ER: 38.4 µg/L; P: 5.4 µg/L n=15	Pilot Whale *Globicephala melas* Caurant & Amiard-Triquet (1995)		
	0.16 µg/g; 0.49 µg/g n=2	Dall`s porpoise *Phocoenoides dalli* Fujise et al. (1988)		
	<0.005 - 0.01 n=3	Weddel seal *Leptonychotes weddellii* Yamamoto et al. (1987)		
Pt	< 0.04 - 8.30 µg/L n=28	Habor Seal *Phoca vitulina* This study	< 0.2 µg/L normal range	Daunderer (2002)
			0.13 - 0.25 µg/L n=22	Farago et al. (1998)
lower than human levels				
Sn	< 0.06 - 0.47 µg/L n=28	Habor Seal *Phoca vitulina* This study	1.5 ± 0.6 µg/L n=110	Alimonti et al. (2005)
			< 2.0 µg/L normal range	Daunderer (2002)
Pb	0.73 µg/L	Habor Seal *Phoca vitulina*	39.5 ± 20.2 µg/L	Alimonti et al. (2005)

	<0.02 - 4.52	This study	n=110	
	n=28		< 150 µg/L	Daunderer (2002)
			normal range	
			5.0 - 132 µg/L	White and Sabbioni (1998)
			n=214	
			157.7 µg/L	Sabbioni et al. (1992)
			n=959	
higher than human levels				
V	0.88 µg/L	Habor Seal *Phoca vitulina*	0.09 ± 0.05 µg/L	Alimonti et al. (2005)
	<0.05 - 3.06	This study	n=110	
	n=28			
Cr	8.74 µg/L	Habor Seal *Phoca vitulina*	0.44 ± 0.27µg/L	Alimonti et al. (2005)
	1.52 - 84.9	This study	n=110	
	n=28		< 0.7 µg/L	Daunderer (2002)
	7.1 ng/g	Southern elephant seal *Mirounga leonina*	normal range	
	n=6	Baraj et al. (2001)	0.1 - 0.6 µg/L	White and Sabbioni (1998)
			n=134	
Mn	67 - 151 µg/L	Habor Seal *Phoca vitulina*	7.7 ± 3.1 µg/L	Alimonti et al. (2005)
	n=28	This study	n=110	
	≤ 0.17 µg/g	Dall`s porpoise *Phocoenoides dalli*	7 - 10 µg/L	Daunderer (2002)
	n=2	Fujise et al. (1988)	normal range	
			1.5 - 22 µg/L	White and Sabbioni (1998)
			n=206	
As	175 µg/L	Habor Seal *Phoca vitulina*	< 10 µg/L	Daunderer (2002)
	42 - 592	This study	normal range	
	n=28		7.9 µg/L	Sabbioni et al. (1992)
			n=470	
Se	885 µg/L	Habor Seal *Phoca vitulina*	73 - 165 µg/L	Daunderer (2002)
	518 - 2261	This study,	normal range	
	n=85	includes data from Griesel et al. (2006)	59 - 158 µg/L	White and Sabbioni (1998)

	665 - 1449 µg/L	Pilot whale *Globicephala melas*	n=219	
	n=6	Nielsen et al. (2000)	107 µg/L	Sabbioni et al. (1992)
	324 - 729 µg/L	Sperm whale *Physeter catodon*	n=455	
	n=4	Nielsen et al. (2000)		
Mo	5.04 µg/L	Habor Seal *Phoca vitulina*	3.1 ± 1.6 µg/L	Alimonti et al. (2005)
	1.27 - 15	This study	n=110	
	n=28		0.5 - 1.8 µg/L	Daunderer (2002)
			normal range	

1 Values are given in µg/L unless otherwise noted

2 ER= erythrocytes, P=plasma, S=serum, R=Rømø, L=Lorenzenplate, n= sample size



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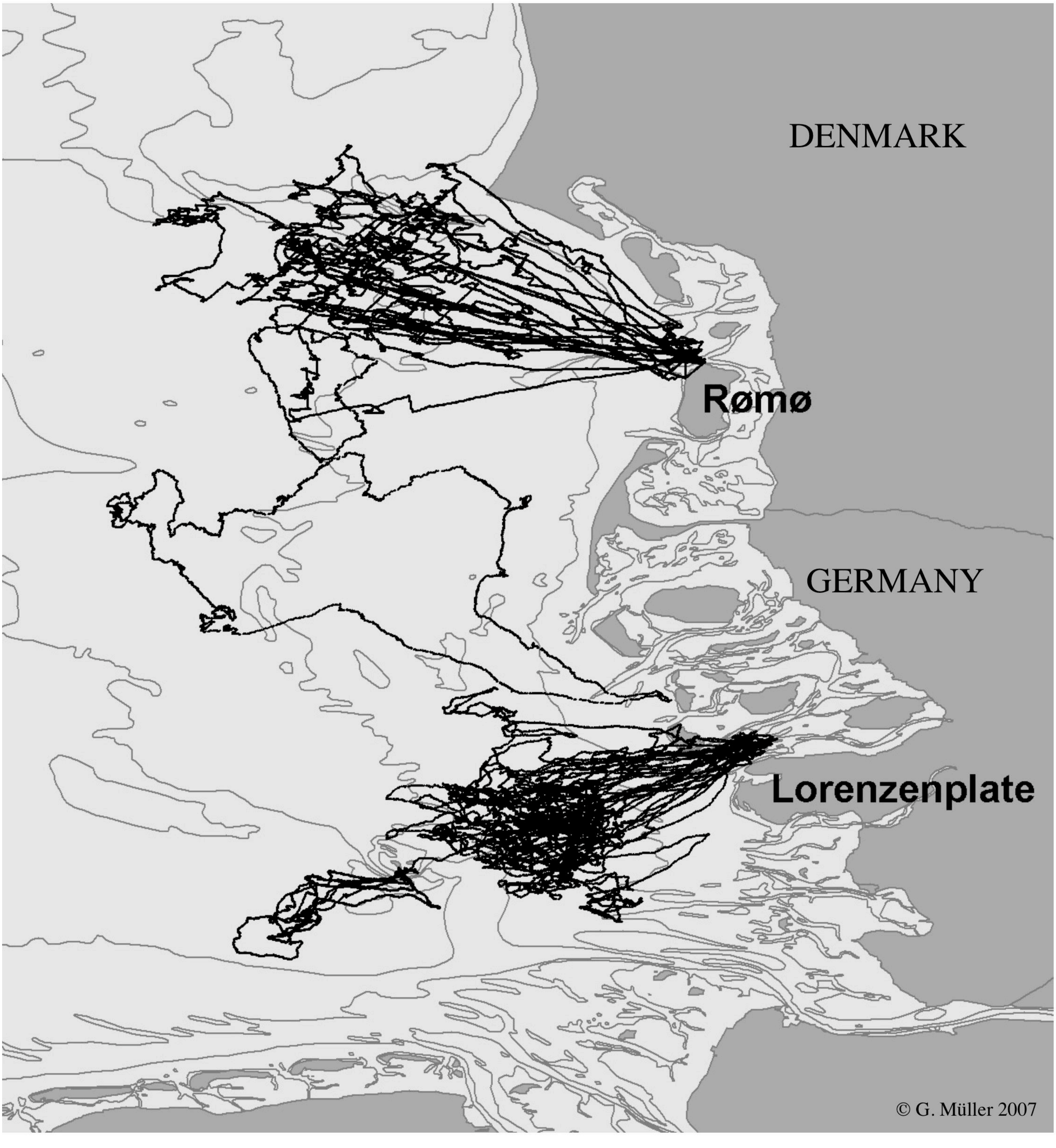
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