Toxic Element Concentration in the Atlantic Gannet Morus bassanus (Pelecaniformes, Sulidae) in Portugal

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Abstract The present study provides the first data on inorganic element levels in juvenile, subadult, and adult Atlantic gannets (Morus bassanus). Physiological and potentially toxic elements (As, Cd, Co, Cr, Cu, Hg, Mn, Pb, Se, and Zn) were assessed by ICP-MS in kidney, liver, muscle, and feathers of 31 gannets, including 18 juveniles, 7 subadults, and 6 adults. The effect of age and tissue on element accumulation was also assessed. Mercury was roughly above the minimum level for adverse effects in birds. A higher accumulation of Se and Cd was detected in kidney, Pb in feathers, and Mn in liver. Age was found to affect the accumulation of Cd, Co, Hg, Mn, Se, and Zn. Adults presented significantly lower levels of Mn, Se, and Zn than subadults. Linear positive relationships within tissues were detected involving Se-Cd and Se-Hg. Also, positive linear relationships were detected among kidney, liver, and muscle, with emphasis on relationships involving Cd, Hg, Se, and Zn, which may be indicative of analogous regulation mechanisms in those organs. Atlantic gannets occurring in the study area leave their reproduction sites as juveniles. During their development process, several molting cycles occur and thus the possible contamination risk by Hg should reflect levels in the development areas rather than contamination levels in reproduction areas. The present study provides basic information on multielement accumulation in Morus bassanus, which may help us to understand the behavior and toxicity of various elements in marine birds.

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Both anthropogenic activities and natural processes produce trace elements that occur in the marine environment. Bioaccumulation and/or biomagnification of such elements may occur in the food web and therefore top predators are liable to consume prey containing high levels of these potential toxic elements (Bearhop et al. [2000](#page-5-0)).

Monitoring the abundance and the environmental consequences of trace elements requires the use of bioindicators (Burger [1993\)](#page-5-0). Seabirds have been often used as biomonitors of element levels in the marine environment because of their high trophic position, bioaccumulation capacities, and resistance to toxic effects (ICES [2003](#page-6-0); Savinov et al. [2003\)](#page-6-0). Furthermore, the ecotoxicological assessment of seabirds is important because they are exposed to varying levels of contaminants and to different natural environmental stressors (e.g., adverse weather), which may produce physiological deficiencies (Forsyth [2001](#page-6-0)).

Among seabirds, Atlantic gannets (Morus bassanus, L.) may be considered as particularly appropriate biomonitors, considering their well-known general ecology and their stable populations. In fact, despite their severely reduced numbers in the past, Atlantic gannet populations have increased, especially since the 1950s (Nelson [2002\)](#page-6-0). In particular, the relatively high abundance of migratory nonreproductive gannets in Portuguese waters, mainly during winter, allows the collection of a relatively large amount of dead, stranded animals. In fact, mainly due to the gannet's ''plunge diver'' behavior and its incompatibility with some fishing gears and "ghost nets," gannets are the main reported avian bycatch in Portuguese waters (Dunn [1994\)](#page-5-0). Furthermore, while age is difficult to assess in most birds (Saeki et al. [2000\)](#page-6-0), Atlantic gannets may be divided into age classes according to the color of their plumage, thus allowing assessment of the age-dependent accumulation of trace elements.

This study presents, for the first time, data on element levels in Atlantic gannets in nonreproduction areas, according to bird tissue (kidney, liver, muscle, feathers) and age class (juveniles, subadults, adults).

Materials and Methods

Sample Collection, Storage, and Preparation

Between January 2003 and December 2006, 194 stranded Atlantic gannets were collected along the seashore between Ovar and Peniche, in the center of Portugal. Fresh carcasses were weighed, measured, and aged according to criteria adapted from Nelson [\(2002](#page-6-0)). Birds were grouped into three age classes: juveniles (first and second winters), subadults (third and fourth winters), and adults (fifth winter or older). During bird necropsies, two identical samples of each tissue (kidney, liver, and pectoral muscle) and caudal feathers were collected from 31 individuals (18 juveniles, 7 subadults, and 6 adults) selected randomly within each age class. The higher proportion of juveniles is due to the fact that juveniles were the most representative age class among the total number of collected birds. All samples were deep-frozen until posterior processing for analysis.

Analytical Procedure

Feathers were triple-rinsed in Milli-Q water alternated with acetone, to remove loosely adherent external contamination (Burger [1993](#page-5-0)), and air-dried overnight. All samples were weighed $(\pm 150 \text{ mg})$ and digested in Teflon vessels with $HNO₃$ (2 ml) and $H₂O₂$ (1 ml) (Merck, Suprapure) at

90°C in an oven and left overnight. All material used in the digestion process was thoroughly acid-rinsed. After digestion, samples were diluted with Milli-Q water and then analyzed for As, Cd, Co, Cr, Cu, Hg, Mn, Se, Zn, and Pb in an inductively coupled plasma mass spectrometer (Perkin Elmer Elan 6000). The analytical procedure was checked using standard reference material, dogfish Squalus acanthias liver (DOLT-3) and muscle (DORM-2; National Research Council, Canada). Analytical blanks were prepared to determine the detection limits. The second set of identical samples from the same individuals was ovendried till constant weight in order to calculate the percentage of humidity in each sample, which allowed transforming wet weight-based results into dry weight values. On average, the humidity in samples was 72.03% in kidney, 69.50% in liver, 69.34% in muscle, and 25.98% in feathers.

Data Analysis

An analysis of variance (ANOVA) followed by the Tukey test and a set of linear regressions were used to detect differences and relations, respectively, between element levels according to tissue type and age class. ANOVA was carried out after $log(x+1)$ transformation. All analyses were performed in Prism 4.0 (GraphPad Software Inc). For all tests, a significance level of $p < 0.05$ was applied.

Results

With respect to the analytical procedure, considering the elements present in the respective reference material, ICP-MS analysis revealed accuracy rates ranging between 81.3% for Cu and 103.8% for Se. Average values (±standard errors) of element concentrations found in kidney, liver, muscle, and feathers of Atlantic gannets are summarized in Table [1.](#page-2-0) Zinc presented the highest values in all tissues.

Considering all 31 birds, element levels were found to vary according to tissue (ANOVA, $p < 0.0001$ for all elements), except for Cr ($p = 0.1082$). Kidney presented the highest levels of Se and Cd ($p < 0.001$ for all), Pb accumulated mostly in feathers ($p < 0.001$ for all tissue combinations), and Mn in liver ($p < 0.001$ compared to muscle and feathers, $p < 0.01$ with respect to kidney).

Element concentrations were generally higher in liver and kidney than in muscle. In muscle, there were significantly lower values of Hg and Mn (ANOVA, $p < 0.001$ for all tissues), whereas the lowest Co values were detected in liver ($p = 0.001$, $p = 0.01$, and $p < 0.05$, respectively, comparing liver to kidney, muscle, and

Table 1 Mean element concentration, μ g g⁻¹ dry weight (\pm standard error) in different tissues of Atlantic gannet

	Kidney			Liver			Muscle			Feathers		
	Juveniles	Subadults	Adults	Juveniles	Subadults	Adults	Juveniles	Subadults	Adults	Juveniles	Subadults	Adults
As	2.945	1.192	1.995	2.312	1.210	1.474	2.552	1.647	1.885	0.600	0.656	1.233
	(0.710)	(0.167)	(0.650)	(0.623)	(0.082)	(0.296)	(0.340)	(0.310)	(0.431)	(0.086)	(0.111)	(0.436)
C _d	4.045	13.340	35.209	0.752	2.000	4.259	0.077	0.105	0.381	0.122	0.107	0.064
	(1.324)	(3.402)	(4.588)	(0.243)	(0.533)	(0.938)	(0.024)	(0.016)	(0.098)	(0.034)	(0.034)	(0.016)
Co	0.210	0.109	0.077	0.059	0.045	0.034	0.081	0.452	0.185	0.137	0.075	0.073
	(0.029)	(0.035)	(0.013)	(0.005)	(0.011)	(0.005)	(0.022)	(0.205)	(0.056)	(0.039)	(0.013)	(0.015)
Cr	3.440	3.231	3.781	3.305	3.544	3.175	3.421	3.603	2.693	3.775	3.717	3.604
	(0.082)	(0.127)	(0.270)	(0.069)	(0.234)	(0.129)	(0.138)	(0.625)	(0.165)	(0.280)	(0.229)	(0.601)
Cu	15.826	18.329	23.612	17.623	22.500	25.528	15.991	21.084	22.614	11.826	14.603	17.516
	(0.995)	(3.374)	(4.167)	(2.051)	(3.575)	(3.442)	(1.551)	(5.748)	(4.924)	(1.258)	(2.237)	(10.306)
Hg	0.952	1.279	3.423	1.291	1.489	2.603	0.473	0.585	0.851	1.620	3.763	5.146
	(0.128)	(0.189)	(0.649)	(0.171)	(0.221)	(0.800)	(0.064)	(0.091)	(0.216)	(0.174)	(0.835)	(1.440)
Mn	6.103	5.379	6.606	10.901	10.208	11.837	1.821	1.889	1.855	7.934	22.038	3.043
	(0.591)	(0.772)	(0.617)	(1.178)	(1.517)	(1.353)	(0.167)	(0.330)	(0.307)	(1.834)	(9.851)	(0.625)
Ph	0.145	0.173	0.266	0.107	0.102	0.109	0.276	0.226	0.127	3.283	3.457	1.112
	(0.023)	(0.048)	(0.069)	(0.012)	(0.001)	(0.002)	(0.197)	(0.108)	(0.031)	(0.783)	(0.840)	(0.223)
Se	14.484	16.829	26.124	11.173	14.779	18.060	4.868	8.652	10.930	3.849	5.111	2.134
	(3.300)	(2.384)	(4.329)	(2.125)	(2.199)	(4.059)	(1.612)	(2.764)	(3.139)	(0.402)	(0.659)	(0.519)
Zn	92.457	109.858	132.443	140.680	143.642	256.954	83.039	91.056	84.714	159.680	278.562	96.984
	(5.961)	(12.299)	(15.173)	(23.291)	(32.643)	(80.432)	(7.138)	(19.280)	(19.255)	(10.137)	(69.118)	(5.953)

feathers). The lowest As ($p < 0.001$ for all) and Cu values $(p \lt 0.001$ compared to kidney and liver, $p \lt 0.01$ with respect to muscle) were detected in feathers. There were no significant differences between kidney and liver element levels, except for Cd, Se, and Co, which were present in higher amounts in kidney ($p < 0.001$ for all), and for Mn, which was present in higher amounts in liver $(p<0.01)$.

There were no significant differences in As, Cr, Cu, and Pb levels across age classes in any of the assessed tissues. There were, however, significant differences for the remaining elements (Table 2). While the highest element levels were found in adults for most of the elements, Co in kidney and Mn, Se, and Zn in feathers presented their lowest concentrations in adults.

A number of relationships between element concentrations was found in the soft tissues and feathers analyzed (Table [3](#page-3-0)). With respect to soft tissues, there were relationships among all element distributions except for Co, Cr, and Pb. However, with regard to feathers, only the amount of Hg in feathers was positively related to Hg in kidney (Table [3\)](#page-3-0). The amount of Hg in kidney increased as a linear function of the amount of Hg in the other soft tissues assessed. The same applies for Cd, Se, and Zn. There was also a significant relationship between the amount of Cu in muscle and kidney. Significant

Table 2 ANOVA and Tukey-test results indicating the significant effects of age class on element concentrations

	F	df	\boldsymbol{p}		\boldsymbol{p}
$[{\rm Cd}]_{\rm Kidney}$	16.130	2,28	< 0.0001	Juveniles \langle subadults	< 0.05
				Juveniles \lt adults	< 0.001
				Subadults \langle adults	< 0.05
$[Cd]_{\text{liver}}$	10.980	2,28	0.0003	Juveniles < subadults	< 0.05
				Juveniles \lt adults	< 0.001
$[Cd]_{Muscle}$	12.580	2,28	0.0001	Juveniles \lt adults	< 0.001
$[Co]_{Kidney}$	3.646	2,30	0.0382	Juveniles > adults	< 0.05
$[Co]_{Muscle}$	4.807	2,26	0.0167	Juveniles < subadults	< 0.05
$[Hg]_{\text{Kidney}}$	15.930	2,28	< 0.0001	Juveniles \lt adults	< 0.001
				Subadults \langle adults	< 0.05
$[Hg]_{\text{Features}}$	13.590	2,28	0.0001	Juveniles \lt adults	< 0.05
				Juveniles < subadults	< 0.01
$[Mn]_{\text{Features}}$	4.732	2,28	0.0170	Subadults > adults	< 0.05
$[Se]_{\text{Kidney}}$	4.399	2,28	0.0218	Juveniles \lt adults	< 0.05
$[Se]_{\text{Muscle}}$	4.518	2,28	0.0199	Juveniles \lt adults	< 0.05
$[Se]_{\text{Features}}$	6.463	2,28	0.0049	Juveniles $>$ adults	< 0.001
				Subadults > adults	< 0.001
$[Zn]_{\text{Kidney}}$	4.461	2,28	0.0208	Juveniles \lt adults	< 0.05
$[Zn]_{\text{Features}}$	11.390	2,26	0.0003	Juveniles < subadults	< 0.05
				Subadults > adults	< 0.001
				Juveniles $>$ adults	< 0.05

Table 3 Linear regressions relating element levels within tissues and levels of each element in different tissues: highly significant cases $(p \le 0.0001)$ associated with $r^2 > 50\%$

		F	$r^2 \left(% \right)$
$[\mathbb{C}\mathrm{d}]_{\mathrm{Kidney}}$	$[Hg]_{\text{Kidney}}$	74.472	73.4
	$[\mathrm{Zn}]_{\mathrm{Kidney}}$	37.410	58.1
	$[Se]_{\text{Kidney}}$	56.828	68.7
$[\mathrm{Cu}]_{\mathrm{Kidney}}$	$[Zn]_{\text{Kidney}}$	67.017	72.1
	$[\mathrm{Hg}]_{\mathrm{Kidney}}$	34.882	55.5
$[Zn]_{\text{Kidney}}$	$[\mathrm{Hg}]_{\mathrm{Kidney}}$	28.200	51.1
$[\mathrm{Se}]_{\mathrm{Kidney}}$	$[\mathrm{Hg}]_{\mathrm{Kidney}}$	32.791	54.8
$[Zn]_{\text{Liver}}$	$[\rm{Hg}]_{\rm{Liver}}$	101.308	79.0
	$[Cd]_{\rm{Liver}}$	55.083	66.3
$[\mathbf{Mn}]_{\mathrm{Liver}}$	$[Pb]_{\text{Liver}}$	25.502	61.5
	$[\mathrm{Hg}]_{\mathrm{Liver}}$	37.923	59.3
$[\mathrm{Se}]_{\mathrm{Liver}}$	$[Cd]_{\rm{Liver}}$	77.608	74.9
	$[\mathrm{Hg}]_{\mathrm{Liver}}$	46.970	63.5
$[\mathbf{C} \mathbf{d}]_{\mathrm{Liver}}$	$[\mathrm{Hg}]_{\mathrm{Liver}}$	75.222	72.2
$[\mathrm{Cu}]_\mathrm{Muscle}$	$[\mathrm{Hg}]_\mathrm{Muscle}$	35.638	57.8
	$[Zn]_{\text{Muscle}}$	39.073	60.0
	$[\mathrm{Se}]_\mathrm{Muscle}$	68.800	71.1
$[\mathbf{C}d]_\text{Muscle}$	$[\mathrm{Hg}]_\mathrm{Muscle}$	40.872	60.2
	$[\mathrm{Se}]_\mathrm{Muscle}$	29.631	51.4
$[\mathrm{Se}]_\mathrm{Muscle}$	$[\mathrm{Zn}]_\mathrm{Muscle}$	30.843	54.3
	$[\mathrm{Hg}]_\mathrm{Muscle}$	77.446	74.2
$[\mathrm{Zn}]_{\mathrm{Features}}$	$\mbox{[Cu]}_{\rm Feathers}$	52.829	66.2
	$\mbox{[Mn]}_{\rm Features}$	203.445	87.5
$[\mathbf{C} \mathbf{r}]_{\text{Features}}$	$[\mathrm{As}]_{\mathrm{Feathers}}$	58.441	68.4
	${\rm [Cu]_{Feathers}}$	44.562	61.4
$[\mathbf{Mn}]_{\text{Features}}$	${\rm [Cu]_{Features}}$	66.380	71.1
$[\mathrm{Hg}]_\mathrm{Muscle}$	$[\mathrm{Hg}]_{\mathrm{Kidney}}$	31.851	52.3
$[\mathrm{Zn}]_\mathrm{Muscle}$	$[\mathbf{Zn}]_{\mathbf{Kidney}}$	46.824	63.4
$[\mathrm{Cu}]_\mathrm{Muscle}$	$[\mathrm{Cu}]_{\mathrm{Kidney}}$	60.560	69.2
$[\mathrm{Se}]_\mathrm{Muscle}$	$[\mathrm{Se}]_{\mathrm{Kidney}}$	56.298	70.6
$[\mathbf{C}d]_\text{Muscle}$	$[\mathsf{Cd}]_{\mathsf{Kidney}}$	72.158	72.8
$[\mathrm{Hg}]_\mathrm{Muscle}$	$[\mathrm{Hg}]_{\mathrm{Liver}}$	109.945	80.3
$[\mathrm{Zn}]_\mathrm{Muscle}$	$[Zn]_{\text{Liver}}$	40.835	61.1
$[\mathrm{Se}]_\mathrm{Muscle}$	$[\mathrm{Se}]_{\mathrm{Liver}}$	49.461	63.9
$[\text{Cd}]_\text{Muscle}$	$[Cd]_{\rm{Liver}}$	83.366	74.9
$[\rm{Hg}]_{\rm{Liver}}$	$[\rm{Hg}]_{\rm{Kidney}}$	53.423	64.8
$[\mathbf{Mn}]_{\mathrm{Liver}}$	$[\mathbf{Mn}]_{\mathbf{Kidney}}$	43.519	60.0
$[Zn]_{\text{Liver}}$	$[Zn]_{\text{Kidney}}$	71.354	71.8
$[\rm{As}]_{\rm{Liver}}$	$[\mathrm{As}]_{\mathrm{Kidney}}$	91.709	76.6
$[\mathrm{Se}]_{\mathrm{Liver}}$	$[\mathrm{Se}]_{\mathrm{Kidney}}$	123.394	82.6
$[Cd]_{\rm{Liver}}$	$[{\rm Cd}]_{\rm Kidney}$	164.343	85.4
$[\mathrm{Hg}]_{\mathrm{Features}}$	$[Hg]_{\text{Kidney}}$	53.350	68.1

relationships were also found between the levels of Mn in liver and kidney and between the levels of As in liver and kidney.

Discussion

Element Levels and Tissue Distribution

There were virtually no data on toxic elements in Morus bassanus, and overall, there are relatively few data on toxic element contamination in seabirds occurring on the Atlantic coast of the Iberian Peninsula (Pérez-López et al. [2006](#page-6-0)). In this context, the present study assesses, for the first time, the levels of 10 elements in Atlantic gannets collected along the central coast of Portugal. Considering worldwide data on other seabirds, in general, the levels of essential and nonessential elements in gannet tissues are within previously reported ranges (Honda et al. [1990;](#page-6-0) Kim et al. [1998;](#page-6-0) Savinov et al. [2003;](#page-6-0) Nam et al. [2005](#page-6-0)).

Mercury and cadmium are generally considered to be the most likely metals to give rise to pollution problems in marine ecosystems (e.g., Dietz et al. [1996\)](#page-5-0). Values reported for these toxic elements in seabirds vary widely among different studies, according to the individuals' feeding ecology, intensity, and time of exposure in foraging areas, and also according to their physiological and biochemical characteristics (Savinov et al. [2003](#page-6-0); Kojadinovic et al. [2007a\)](#page-6-0). In fact, seabirds that feed offshore revealed a higher rate of mercury accumulation, and moreover, higher heavy metal levels are consistently described in pelagic seabirds compared to levels detected in inshore feeders, unless there is a known local pollution source (Nisbet [1994](#page-6-0)).

As expected, the highest concentration of mercury was found in feathers because Hg excreted through feathers is generally higher than the amount of Hg existing in food taken by marine birds (Lee [1989](#page-6-0)). A range between 5 and 40 μ g g⁻¹ d.w. Hg in feathers may be associated with adverse effects (Eisler [1987\)](#page-5-0), although higher levels should be tolerated by marine birds. Also, tolerance ranges should vary between seabird groups because Hg in birds may depend on the types of molting cycles (Honda et al. [1986](#page-6-0)). Nonetheless, in the present study, the average concentration of Hg in feathers from adult birds was 5.15 μ g g⁻¹ and the maximum Hg value found in an individual adult bird in feathers was 11.77 μ g g⁻¹. These Hg levels may be an early warning indication of a Hg contamination risk for the Atlantic gannet population occurring in the study area.

Cadmium levels were significantly lower than the accepted threshold level (Furness [1996\)](#page-6-0) for biological effects in birds. However, the Cd concentration in kidneys of adult birds (35.21 μ g g⁻¹) was higher than that registered in cormorants from Japan (6.83 μ g g⁻¹) by Saeki et al. [\(2000](#page-6-0)). Such differences may indicate the importance of different diets on toxic element variability considering its major role as a contamination path in marine vertebrates (Kim et al. [1998;](#page-6-0) Stewart et al. [1999\)](#page-6-0). In fact, birds that feed

mainly on squid tend to accumulate high Cd levels, whereas those that feed mainly on fish have higher Hg levels (Muirhead and Furness [1988;](#page-6-0) Honda et al. [1990](#page-6-0)), and therefore a future analysis of gannet diet in the study area may help to clarify the importance of prey type in the levels of toxic elements found in these predators. The detected preferential accumulation of cadmium in kidney is a usual trend in seabirds (Dietz et al. [1996;](#page-5-0) Nam et al. [2005](#page-6-0); Kojadinovic et al. [2007a](#page-6-0)), and according to Scheuhammer [\(1987](#page-6-0)), a higher level of Cd in kidney compared to liver usually indicates chronic exposure to low Cd concentrations.

Generally, seabirds exhibit low lead levels (Norheim [1987\)](#page-6-0). In the present study, Pb burdens in feathers were higher than those in soft tissues, probably as a result of direct atmospheric deposition onto the feather surface (Kim et al. [1998\)](#page-6-0). With respect to arsenic, levels were within the ranges usually reported in other studies and were below values known to produce toxic effects ($> 50 \mu g g^{-1}$ d.w.) in seabirds (Neff [1997](#page-6-0)).

There were no significant differences in chromium levels among tissues, in accordance with results obtained in great cormorants (Nam et al. [2005\)](#page-6-0). Although Cr produces neurotoxic effects in birds, information about this element in the marine environment is scarce (Burger and Gochfeld [1995\)](#page-5-0). The feathers' Cr level in the present study was higher than those registered for other relatively large sea bird species by Burger and Gochfeld ([2000a](#page-5-0)). Nonetheless, average Cr levels in Morus bassanus in liver and kidney (3.3 and 3.5 μ g g⁻¹ d.w., respectively) do not reach the level of adverse effects proposed by Eisler ([1986\)](#page-5-0) for internal tissues (4.0 μ g g⁻¹).

Selenium presented the highest levels in kidney, in agreement with previous works (Dietz et al. [1996;](#page-5-0) Kim et al. [1998\)](#page-6-0). Levels of Se in liver (4.2 μ g g⁻¹ w.w.) and kidney (5.1 µg g^{-1} w.w.) in the present study were roughly half the accepted threshold levels for adverse biological effects in birds, 9 μ g g⁻¹ w.w. (Heinz [1996](#page-6-0)) and 10 μ g g⁻¹ w.w., respectively (Thomson et al. [1996\)](#page-6-0). Thus, this level of Se is probably beneficial, considering its previously described importance in the antioxidant system (Norheim [1987\)](#page-6-0) and in other toxic element detoxification processes (e.g., Ikemoto et al. [2004\)](#page-6-0).

Age Influence

In the present study, levels of Hg and Cd increased with age. Such age-dependent increments are a well-documented occurrence in many seabirds (Yamamoto et al. [1996;](#page-6-0) Saeki et al. [2000\)](#page-6-0), mainly in feathers (Walsh [1990](#page-6-0); Burger and Gochfeld [2000b](#page-5-0)). However, data on concentration rates for other elements are scarce, particularly with respect to element concentrations in soft tissues according to age class.

With respect to Hg, the concentration in feathers of adult birds was higher than that in juveniles and renal Hg was higher in adults than juveniles and subadults. It is known that Hg accumulates with age in birds, even though they have a molting system for Hg removal (Furness and Hutton [1979](#page-6-0); Honda et al. [1986\)](#page-6-0). The significant increase in Hg in kidney from nonadults to adults indicates that Hg is accumulating in the internal organs via the trophic chain, i.e., Hg intake exceeds excretion by molting. Cadmium concentration in adults was higher in kidney, liver, and muscle. The relatively long half-lives of Hg and Cd, their capacity to bind to metallothioneins, and their difficult excretion from living organisms contribute to these results.

The levels of Cr, Cu, As, and Pb were not influenced by age in any of the analyzed tissues. The Co concentration decreased with increasing age in kidney but not in other tissues, while Mn in feathers was lower in adults than in subadults, possibly as a result of different metabolization/ excretion processes. Selenium levels showed an increase with age in kidney, liver, and muscle, probably as a result of its role in the detoxification of Cd and Hg (Ikemoto et al. [2004](#page-6-0), Kojadinovic et al. [2007b](#page-6-0)). On the contrary, Se levels in feathers were lowest in adults, in disagreement with other studies (Yamamoto et al. [1996](#page-6-0); Burger and Gochfeld [2000a\)](#page-5-0). Renal Zn levels increased significantly with age from juveniles to adults, whereas Zn levels in feathers showed a particular pattern of accumulation, being higher in subadults than in adults.

Relationships Among Tissues and Metals

In the present study, regression analyses expose strong relationships between elements, which are generally in agreement with those reported in other studies (Honda et al. [1990](#page-6-0); Kim et al. [1998](#page-6-0); Nam et al. [2005\)](#page-6-0). Element levels in seabirds and other marine vertebrates are controlled by regulation mechanisms, such as detoxification by binding to metallothioneins (MTs) and insolubilization of metals as mineral concretions (Ikemoto et al. [2004](#page-6-0); Kojadinovic et al. [2007b](#page-6-0)). Metallothioneins are involved in the homeostasis of Zn and Cu, and both MTs and Se are involved in the detoxification of Cd and Hg (Ikemoto et al. [2004](#page-6-0); Nam et al. [2005](#page-6-0); Barbieri et al. [2007\)](#page-5-0). The strong relationships between element levels may suggest common uptake and storage pathways or similar regulation and detoxification processes.

The positive relations detected between Se and Cd in liver and in kidney were also described by Kim et al. [\(1998](#page-6-0)) and Furness and Hutton ([1979\)](#page-6-0), respectively. The latter had already suggested the possible role of Se in Hg and Cd detoxification mechanisms.

Levels of MTs were not assessed in this study, but earlier works showed that Cd, Zn, Hg, and Cu bind to MTs and are often found to accumulate in parallel (Kojadinovic et al. [2007b\)](#page-6-0). In fact, positive linear regressions were found between these elements (Hg-Cd, Hg-Zn, Hg-Cu, Zn-Cu, and Zn-Cd in kidney and Hg-Zn, Hg-Cd, and Zn-Cu in liver), also in accordance with Barjaktarovic et al. (2002). These relations may be explained by relatively similar pathways of metabolism and storage for these elements (Kojadinovic et al. [2007a](#page-6-0)), such as those involving binding to MTs. The Cd-Zn relationship in livers of Gannets had already been detected in other sea birds (Elliott et al. [1992](#page-6-0); Kim et al. [1998](#page-6-0)), suggesting that higher hepatic zinc levels are likely to be associated with MT induction by chronic Cd accumulation. Considering the Cu-Zn relationships, these elements are metabolically regulated in seabird tissues, and external factors, such as short-term fasting or starvation after mobilization of stored energy, are known to influence the metabolism of these particular elements (Debacker et al. 2000).

The analysis of the distribution of each element shows many positive relationships between soft tissues but not between soft tissues and feathers. Metal concentrations in feathers are a result of metal levels in circulation at the time of feather formation. These levels may be due to ongoing exposure (through food or water) or they may also result from the mobilization of metals stored in other tissues (e.g., Furness et al. [1986\)](#page-6-0). On the other hand, levels in feathers may reflect external contamination by deposition, particularly with respect to Pb and Cd, which cannot be entirely removed by washing procedures (Furness and Camphuysen [1997](#page-6-0)).

Only Hg, Cd, Se, and Zn show a significant positive relation for all of the soft tissues analyzed (kidney, liver, and muscle), indicating an identical pattern of metabolism and storage. However, considering that many contaminants have been found to covary (Fisk et al. [2005](#page-6-0) and that some correlations may be due to chance (Kim et al. [1998\)](#page-6-0), further studies are needed on the interactions between metals and their biologic significance, in particular concerning biosynthesis of metal-binding proteins and the antagonistic effects between elements.

There was a positive relationship between Hg in feathers and in kidney. Considering that over 90% of a bird's total body burden of mercury may be sequestered into feathers during molt (Burger 1993), this relationship confirms the suitability of feathers as biomonitors for mercury contamination.

Exposure of migratory birds to contaminants is determined by their migration patterns, which can extend across entire hemispheres between breeding and wintering grounds (Pérez-López et al. [2006](#page-6-0)). Burger and Gochfeld (2000b) emphasized the need for data on the same species in a wide range of geographical areas and for data comparing many different species, representing different trophic levels, from the same general area. Apart from identifying a possible contamination risk by Hg for the Atlantic gannet population occurring in the study area, the present study provides basic information on multielement accumulation in Morus bassanus, which may help us to understand the behavior and toxicity of various elements in marine birds.

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