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Report on research activities carried out for determination of Mercury in different tissues of cetaceans

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Summary

Mercury concentration in different tissues of two dolphin species (*Stenella coreuleoalba* and *Tursiops truncatus*), stranded along the Italian coasts during the period 2000– 2009, were reported in order to assess Hg distribution patterns in different tissues. The highest concentrations of Hg were found in liver samples of both species (8.4-1752 mg/kg dw for *Stenella coeruleoalba* and 9.6-1404 mg/kg dw for *Tursiops truncatus*). Relative high mercury concentrations were reported also in kidney and lung tissues. The existence of Hg bioaccumulation process over time was also reported. The dataset documents the existence of different mechanisms of mercury bioaccumulation in the different tissues analyzed.

Introduction

Large amounts of organic and inorganic contaminants enter estuarine and coastal marine environments from natural and anthropogenic sources. Human activities have increased the flux of many naturally occurring chemicals, such as metals and petroleum hydrocarbons, into the ocean (Neff 2002). Trace metals are natural components of the hydrosphere and many are necessary, in minute quantities (e.g., arsenic, copper, iron, molybdenum, tin; Ward 1995), for the metabolism of marine organisms. However some trace elements, such as Hg, are considered highly toxic (Neff 2002). Mercury does not normally participate in metabolism and, at least in top predators, is accumulated throughout the entire life of an individual. Mercury can be transferred and accumulated through the food chain at even higher concentrations via a process known as bio-magnification (Bell & Pollard 1989). Dolphins are at the top of the marine food chain and accumulate large amounts of Hg as a result of bio-magnification of the trophic web. They, and generally all marine mammals, are considered sensitive and reliable tracers of environmental Hg contamination (Capelli et al. 2000). In this study Hg concentration in different tissues of two dolphin species (*S. coreuleoalba* and *T. truncatus*) stranded along the Italian coasts during the period 2000– 2009, were reported in order to assess Hg distribution patterns in the different tissues analyzed.

Materials and methods

Samples of muscle, liver, lung, kidney and heart were collected from 12 specimens of striped dolphin (*S. coeruleoalba*) and 12 specimens of bottlenose dolphin (*T. truncatus*) stranded along Italian coasts during the period 2000–2009 (Figure 1). The number of samples for each tissues and organs are shown in Table 1. Samples were wrapped in plastic bags and immediately stored at -20°C after collection. The total length of each specimen was measured before sample collection. Samples were

dried at 40°C for 48 h and homogenized in an agate mortar. About 0.25 g of each air-dried and homogenized sample were digested under pressure in 10 ml of ultra-grade HNO_3 in Teflon liners using a microwave oven CEM MARS-5 (Figure 2), for 4 h at 200 W and at T = 160 ± 5°C. Samples were prepared and analyzed with great caution to minimize contamination from air, glassware and reagents, all of which were of Suprapur quality. Mercury was determined by ICP-AES Varian Vista MPX (Figure 2) exciting analyte to form volatile hydride in the Hydrides Generation reactor VGA 76P, according to the method reported by Pohl (2004). Reagent blanks and duplicated samples (about 10% of the total number of samples) were used to check appropriateness and reproducibility of preparation and analytical procedures. A reference standard material, Tort-2 (National Research Council of Canada) was used to assess the accuracy (estimated between 85–93%) and precision (ranging between ±3–5%, RSD; n = 10) of analyses. Results of quality control show an excellent agreement with certified data (Certified: 0.27 ± 0.06 mg/kg dw; Found: 0.23 ± 0.07 mg/kg dw). The analytical precision, measured as relative standard deviation, was routinely between 5% and 6%, and never higher than 10%. All results were calculated with respect to dry weight (dw).

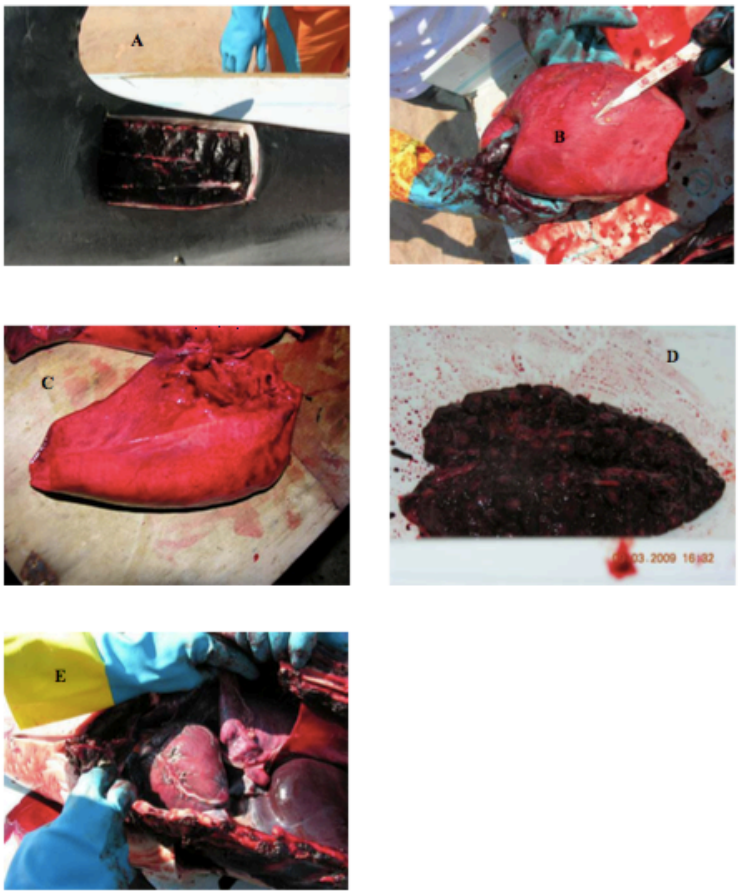

Figure 1: Sample collection of muscle (A), liver (B), lung (C), kidney (D) and heart (E) from cetacean body.

Figure 2: microwave oven CEM MARS-5 and ICP-AES Varian Vista MPX used for Hg determination.

Results and discussions

Mercury concentrations in tissues of the studied organisms are shown in Table 1.

ID	Stranding site	Length (cm)	Muscle (n = 10)	Lung (n = 8)	Kidney (n = 8)	Heart (n = 7)	Liver (n = 10)
Stenella coeruleoalba	N = 12						
Sc 1	Mazara del Vallo (Sicily Channel)	85	0.94	2.69	0.37		
Sc 2	Mazara del Vallo (Sicily Channel)	190	16.60	71.00		5.50	455.60
Sc 3	Mazara del Vallo (Sicily Channel)	182	13.58	5.64	24.59	9.85	97.38
Sc 4	Mazara del Vallo (Sicily Channel)	131	4.99	1.79	10.02	0.00	22.50
Sc 5	Mazara del Vallo (Sicily Channel)	98	0.00	2.42		1.12	15.30
Sc 6	Mazara del Vallo (Sicily Channel)	103	1.52	3.04	4.47	4.86	8.48
Sc 7	Mazara del Vallo (Sicily Channel)	173	11.75	11.29	23.66	11.12	51.43
Sc 8	Mazara del Vallo (Sicily Channel)	110		7.62	22.48	11.94	89.99
Sc 9	Monte Argentario (Thyrrenian sea)	131	4.45		8.04		21.19
Sc 10	Viareggio - (Thyrrenian sea)	110	36.62				90.54
Sc 11	Viareggio - (Thyrrenian sea)	192	116.54				1752.20
Sc 12	Orbetello - (Thyrrenian sea)	91			13.08		
Tursiops truncatus	N = 12		Muscle (n = 10)	Lung (n = 5)	Kidney (n = 11)	Heart (n = 4)	Liver (n = 9)
Tt 1	Mazara del Vallo (Sicily Channel)	100		0.00	0.00	0.07	
Tt 2	Mazara del Vallo (Sicily Channel)	225	19.97	62.56		2.47	181.00
Tt 3	Mazara del Vallo (Sicily Channel)	180	10.70	34.4	29.0		70.13
Tt 4	Mazara del Vallo (Sicily Channel)	270	17.09	20.87	21.46	13.99	261.68
Tt 5	Mazara del Vallo (Sicily Channel)	130	0.00	0.00	3.60	0.00	9.60
Tt 6	Jesolo (Adriatic sea)	285	35.92		60.03		570.49
Tt 7	Ravenna (Adriatic sea)	136	5.26		3.25		
Tt 8	Monte Argentario (Thyrrenian sea)	276			56.66		1404.92
Tt 9	Napoli - (Thyrrenian sea)	213	14.35		15.38		
Tt 10	Marciana Marina - (Thyrrenian sea)	150	1.21		9.37		17.48
Tt 11	Viareggio - (Thyrrenian sea)	265	25.56		30.73		453.49
Tt 12	Jesolo (Adriatic sea)	150	4.75		10.98		23.03

Table 1: Sites of stranding, length and Hg concentrations (mg/kg dw) in tissues of *Stenella coeruleoalba* and *Tursiops truncatus*.

There is a wide range of Hg concentrations especially in liver samples (8.4–1752 mg/kg dw for *S. coeruleoalba* and 9.6–1404 mg/kg dw for *T. truncatus*). Lower values and narrow ranges of concentrations were found in the other tissues, particularly in muscle and heart samples of both species. Liver shows the highest concentrations of Hg in all analyzed specimens, followed by kidney (Figure 3). The lowest values were found in samples of muscle and heart of *T. truncatus* and in samples of lung and heart of *S. coreuleoalba*. Mercury concentration is highly variable within species but, on average, higher in organs of *S. coeruleoalba*. Positive significant correlations emerge between Hg concentrations and length in all tissues of *S. coeruleoalba* (except for heart samples). In regards to specimens of *T. truncatus*, positive significant correlation was found between Hg concentrations and length in liver, kidney and muscle samples.

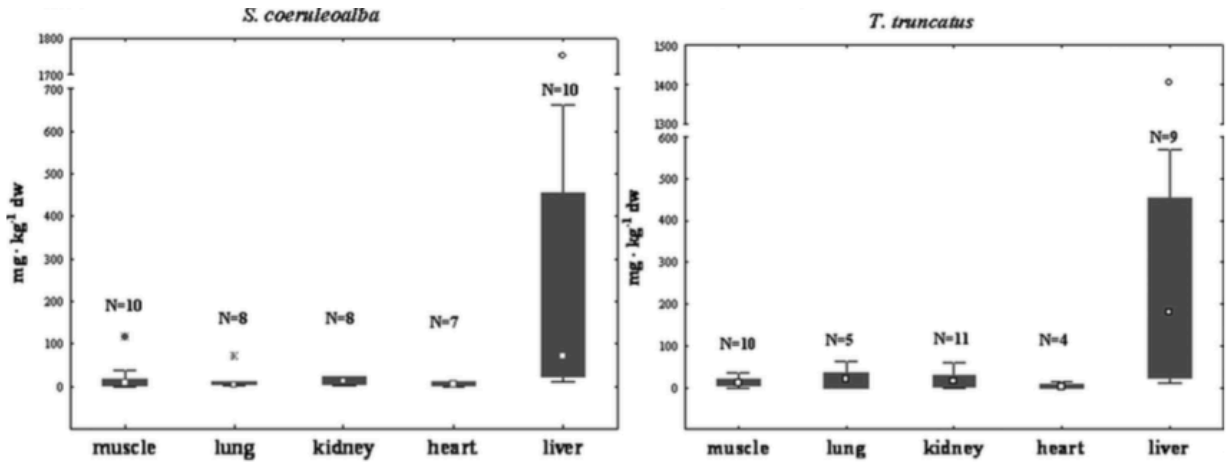

Figure 3: Mercury concentrations in tissues of *Stenella coeruleoalba* and *Tursiops truncatus.*

The high hepatic Hg concentrations are generally related to the role played by the liver in terms of pollutant bio-transformation (Thomson 1990). Specifically, demethylation, namely the transformation of organic Hg (methylmercury) into the less toxic inorganic form, is believed to occur in the liver. Mercury is stored in a permanent and continuous manner in cetacean liver under the form of insoluble mercury selenides. The selenium compounds help protect the organism from the oxidative damages due to Hg (Cuvin-Aralar & Furness 1991). Relatively high concentrations of Hg were found in the analyzed kidney samples in both species. High concentrations of Hg in kidneys have also been previously reported by different authors (e.g., Leonzio et al. 1992; Augier et al. 1993). This is probably due to the fact that the kidney stores a non negligible fraction of the metal and is also involved in the process of elimination. Relatively high concentrations of Hg were found in lung samples of adult specimens of *T. truncatus* (Table 1). Augier et al. (1993), while evaluating the different sources of Hg for dolphins, hypothesized that Hg could penetrate from the atmosphere into the lungs increasing the effect of accumulation in this organ. Also Rawson et al. (1995) found relative high concentrations of Hg compounds in the respiratory system of bottlenose dolphins and short finned pilot whales. The finding of high concentrations of Hg has raised the question about its potential toxicity. Chronic Hg accumulation was associated with liver abnormalities observed in stranded bottlenose dolphins from the Atlantic. Mercury would have inhibited the activity of lysosomal digestive enzymes and, therefore, reduced degradation of proteins, leading to excessive accumulation of lipofuscin within cells and finally cell death (Rawson et al. 1995). The limit of Hg tolerance of mammalian's liver was established by Wagemann and Muir (1984) and ranges between 100–400 mg/kg dw. In this study Hg concentration of 1752 mg/kg and 1404 mg/kg dw were found in liver of a specimen of *S. coeruleoalba* and *T. truncatus* respectively.

Conclusions

In regards to the distribution of Hg in the different analyzed tissues, the reported dataset is substantially comparable with those reported by other authors worldwide. In particular, liver systematically shows the highest concentrations of Hg for both *S. coeruleoalba* and *T. truncatus* species with evident high potential for toxic elements accumulation in this organ. Also, in accordance with previous authors, we reported the existence of Hg bioaccumulation process over time. This dataset documents the existence of different mechanisms of mercury bioaccumulation in different cetacean tissues.

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