EVALUATION OF SEABIRD EXPOSURE TO POP-LIKE (ORGANOTIN) COMPOUNDS IN COASTAL WATERS OF RIO DE JANEIRO STATE (BRAZIL) THROUGH TOTAL TIN CONCENTRATIONS IN INTEGUMENTARY SYSTEM

Padilha JA[1], Paiva TC[1], Castro RM[1], Malm O[1], Cunha LST[1], Collard F[2]*, Dorneles PR[1,2]

[1]Radioisotope Laboratory, Biophysics Institute, Federal University of Rio de Janeiro (UFRJ), Rio de Janeiro, Brazil, [2]Laboratory of Oceanology, University of Liege, Liege, Belgium

Introduction

Environmental contamination by organotin compounds (OTs) has received considerable attention due to the bioaccumulative nature of these substances. In fact, some investigations have drawn attention to a POP-like environmental behaviour of OTs, particularly triphenyltin (TPhT)[1]. Metals constitute a pollutant group that raises high environmental concern, especially the organic forms, as they are the most toxic chemical species[2]. Regarding tin specifically, the inorganic forms are virtually unabsorbed through the gastrointestinal mucosa of vertebrates. On the other hand, OTs are readily bioaccumulated and found in high concentrations in biota[3]. Therefore, it can be concluded being possible to evaluate which species / population of vertebrates would be more exposed to OTs through tissular concentrations of total tin (ΣSn), and the efficiency of this approach has already been demonstrated by our research team[3,4].

Regarding environmental contamination by heavy metals, there is a growing need to monitor oceanic and coastal areas and the use of sentinel species of environmental and human health is a valuable alternative. Seabirds have been used as sentinels of environmental contamination by toxic elements, due to their high longevity and high trophic positions occupied[5]. Integumentary structures, such as hair, scales and feathers are considered matrices of choice for the evaluation of exposure to organometallic compounds (OMCs), as they end up constituting excretory routes for these toxicants[6]. Additionally, feathers provide a nonlethal matrix for assessing the exposure of these animals to OMCs[5]. In addition, a relatively high proportion of the total body burden of OMCs is stored in feathers and strong correlations have been observed between levels of contaminants in this matrix and in the diet of seabirds[7,8].

Some OTs own great importance for industrial and agricultural activities, being used in many different processes[3]. However, the use that raised the highest environmental concern is related to the presence of OTs in inks applied to submerged parts of ships and floating platforms in order to prevent incrustation of molluscs and crustaceans (antifouling activity)[9]. Tributyltin (TBT) is highly toxic and persistent in the environment, generating enormous concern about its use as a biocide in industry and agriculture [9].

OTs have been used as active ingredients of antifouling paint formulations since the 1970s[10]. Due to their toxicity to non-target species, many developed nations adopted restrictions at the beginning of the 1980s; however, the global ban of the OT use in antifouling formulations only happened in 2008[10]. Developed nations use only tin-free antifouling paints nowadays; conversely, the same does not occur in all developing countries. The Brazilian Navy prohibited such use of OTs in the country; nevertheless, it is legal to buy TBT in Brazil. Theoretically, TBT is sold in Brazil for other purposes; nonetheless, the compound is still being used as antifouling agent in the country[11].

The exposure of birds and mammals to OTs can be monitored through chemical analyses of integumentary structures, since feathers and hair have been described as elimination routes for OMCs in species from those animal groups[12-14].

For comparison purposes, the concentrations of four other trace elements (Se, Mn, Cu and Cd) were determined in the same samples. To the authors' knowledge, the present study is the first scientific investigation on trace-element exposure of *Fregata magnificens* and *Sula leucogaster* from Cagarras Archipelago, Rio de Janeiro state (RJ), Brazil.

Materials and methods

Samples were obtained from individuals that nest in Cagarras Archipelago, which is located 4 km from the coast of Rio de Janeiro city. The nearness to "MegaCity 23" means also proximity to Guanabara Bay, an estuary considered the most dramatic example of man-made degradation along the Brazilian coast[3]. These islands are located 10 km south-westward from the entrance of this estuary. Guanabara Bay is surrounded by four cities (including Rio de Janeiro metropolitan area) with a total population of about 11million people. The estuary is bordered by 6000 industries, with more than 6000 additional industries in its drainage basin[15].

Tracking the radio transmitter information, attached to some frigatebirds by members of our research group, we were able to observe the activities of these seabirds. Then, it was possible for us to verify that the frigatebirds that nest in Cagarras Archipelago go to the Guanabara Bay for feeding on a daily basis.

The seabird samples comprised wing feathers from frigatebirds (*Fregata magnificens*, *n*=14; 2 female, 1 male and 11 juveniles) and brown boobies (*Sula leucogaster*, *n*=27; 9 females, 11 males and 7 juveniles). Feathers were washed with EDTA 0.01% in order to remove external contamination, *i.e.*, the adsorbed trace elements. Before being solubilized, sampled feathers were dried at 50°C. The samples were digested with 2 mL of 65% HNO_3 in a screw-capped vessel, during 24 h. The vessel was then heated to 60 ºC for 120 min in a water bath. After cooling, the sample was made up to a known volume with high purity deionised water (18.2 MΩ cm) from a Milli-Q system. Quality control (QC) was carried out through the use of blanks, which were proceeded through in the same way as the samples; through the use of certified reference material (Dolt-4 from NRCC; Se, Mn, Cu and Cd); as well as by employing the standard addition method (Sn). Regarding the later QC procedures, recoveries from Dolt-4 and fortified extracts were always between the range of 90% and 110%. The temperature programs used and additional methodology details can be found elsewhere[3]. Tin, Cd, Se, Mn and Cu concentrations were determined by electrothermal atomic absorption spectrometry (ET-AAS), using an Analytic Jena spectrometer ZEEnit 60, equipped with Zeeman-effect background correction. Palladium nitrate was used as a matrix modifier and hence added to each solution to be analysed. For statistics, depending on data normality (Shapiro-Wilk's W test), parametric (Student's t-test and Pearson's correlation test) or non-parametric (Mann-Whitney U test and Spearman correlation test) tests were used.

Results and discussion

No significant correlations were found between ΣSn concentrations and biological parameters, which were the lengths of different anatomical structures and the weight of the birds. The anatomical structures used were tarsus, wing, tail and beak. For brown boobies, there was no significant difference between gender or age classes (adults and juveniles). Burger and Gochfeld[16] determined heavy metal concentrations in feathers of twelve seabird species from Midway Atoll (N. Atlantic) and two of these species were from the same taxonomic genera (*Fregata* sp., and *Sula* sp.) considered herewith. The ΣSn concentrations found in birds from Cagarras Archipelago (RJ) seem to be lower than those found in *Fregata minor* (Great Frigate) and *Sula sula* (Red-footed Booby) by Burger and Gochfeld[16] (Table 1). This apparent difference may be related to dissimilar degrees of environmental contamination by OTs in the two hemispheres, since the northern half of the planet was significantly more contaminated than the southern one[1].

Table 1. Mean, standard deviation and number (*n*) of analysed specimens (μg.Kg^{-1}, dry weight) found in the work of Burger and Gochfeld, 2000* and generated by the present study (individuals that nest in Cagarras Archipelago, Rio de Janeiro, Brazil).

Species	*n*	Cd	*n*	Se	*n*	Mn	*n*	Sn
*Sula sula**	12	51.3 ±5.58	12	2340 ±112	12	1460 ±314	12	2280 ±262
S. leucogaster	19	28.2 ±24.0	12	2343 ±1208	16	1559 ±884	5	257 ±85.7
*Fregata minor**	5	204 ±127	5	4540 ±1290	5	590 ±79.7	5	752 ±165
F. magnificens	19	33.1 ±20.4	8	1942 ±778	6	1701 ±618	5	260 ±117

Table 2 shows all elements measured in this study. Regarding the trace elements other than tin, no significant differences between males and females were found for adult brown boobies. Comparing adult and juvenile boobies, there were no significant differences for any of the targeted elements. This constituted surprising information, as higher levels were expected in adults as a result of the bioaccumulation process.

Regarding interspecific comparison (using juveniles of both species), higher Mn and Cu values were found in boobies than in frigatebirds (Fig.1). This may be related to the physiological peculiarities of each species or even to differences in diet. Using adults for the interspecific comparison, higher Cd concentrations were found in frigatebirds than in boobies. Brown boobies usually feed in deep, while frigatebirds snatch prey on the water surface. Considering exclusively this feeding behaviour, we would get to the conclusion that the brown booby not only presents a higher proportion of fish in its diet, but also preys on larger specimens than does the frigatebird[16]. By ingesting more and larger fish, boobies might be expected to accumulate higher levels of biomagnifying contaminants[16], as OTs, for example, but we have not found significant differences for ΣSn. However, it is worth mentioning that frigates have a kleptoparasitic behaviour, which is to disturb birds of other species (especially boobies) in order to make them regurgitate for stealing their food[17]. Therefore, the kleptoparasitism turns the diet of the two seabird species more similar. Despite of this similarity, frigatebirds had higher Cd levels. Cadmium concentrations are relatively high in marine organisms[18], but the explanation for the interspecific difference found between two bird species with such similar diets is not obvious. Cadmium accumulates with age[19], and this difference may be due to this variable. Adults of the two species may have belonged to different age classes. Although age could be an important factor, slight diet differences should not be rule out as possible explanation for the distinct Cd levels. It is well known that these seabird species feed on cephalopods in other areas, as well as that these invertebrates efficiently transfer cadmium to their predators[20]. However, there are no studies on the diet of these birds around Cagarras Archipelago for shedding further light on these differences.

Table 2. Concentrations (mean ±standard deviation) of Cd, Se, Cu, Mn and Sn in feathers from *Fregata magnificens* and *Sula leucogaster* (μg.Kg^{-1} dry weight), considering individuals that nest in Cagarras Archipelago, Rio de Janeiro, Brazil.

Species	***n***	**Cd**	***n***	**Se**	***n***	**Cu**	***n***	**Mn**	***n***	**Sn**
Fregata magnificens										
Male	1	115	1	2201	1	7535		-		-
Female	2	66.7 ±1.24	1	2550 ±494	2	8245 ±1095	2	5143 ±1785	2	320
Juvenile	11	30.7 ±13.67	8	1749 ±739	8	1404 ±833	6	1721 ±618	6	233 ±110
Species	***n***	**Cd**	***n***	**Se**	***n***	**Cu**	***n***	**Mn**	***n***	**Sn**
Sula leucogaster										
Male	11	32.5 ±30.5	6	2455 ±1396	9	7980 ±3274	8	1563 ±828	2	264 ±25.8
Female	9	22.8 ±12.2	6	2231 ±1109	6	8613 ±3650	6	1732 ±1104	3	175 ±34.1
Juvenile	7	32.0 ±31.5	6	1870 ±1619	7	5061 ±2307	7	3839 ±3537	3	257 ±85.7

Additional research on assimilation of persistent and bioaccumulative toxicants (PBTs) by *Sula leucogaster* and *Fregata magnificens* from Cagarras Archipelagos is of utmost importance. The same holds for the components of the food web on which these predatory birds rely. These additional studies would allow a better understanding on PBT bioavailability, trophic flow, and exportation from the contaminated estuaries of Rio de Janeiro state. Regarding OTs specifically, our results suggest both species are equally exposed to OTs in the waters around Cagarras Archipelago, Rio de Janeiro, Brazil.

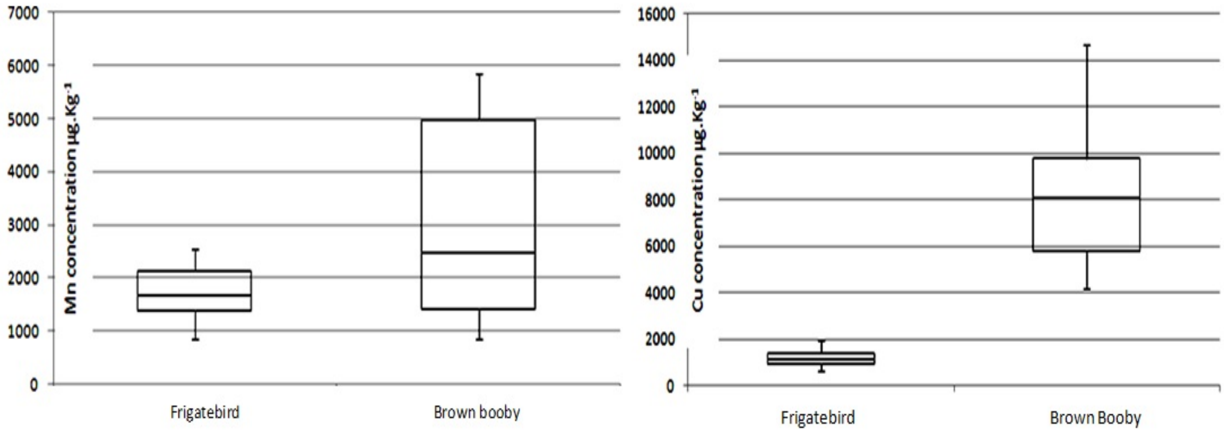

Fig 1. Levels of manganese and copper (µg.Kg^{-1} dry weight) in feathers of juveniles from both species (*Fregata magnificens*, frigatebird; and *Sula leucogaster*, brown booby).

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