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High levels of mercury and low levels of persistent organic pollutants in a tropical seabird in French Guiana, the Magnificent frigatebird, *Fregata magnificens*

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Abstract

In the present study, trace elements and persistent organic pollutants (POPs) were quantified from Magnificent frigatebirds (*Fregata magnificens*) breeding at a southern Atlantic island. Stable isotope ratio of carbon ($\delta^{13}C$) and nitrogen ($\delta^{15}N$) were also measured to infer the role of foraging habitat on the contamination. For another group from the same colony, GPS tracks were recorded to identify potential foraging areas where the birds may get contaminated. Fourteen trace elements were targeted as well as a total of 40 individual POPs, including organochlorine pesticides (OCPs), polychlorinated biphenyls (PCBs) and polybrominated diphenyl ethers (PBDEs). The concentration of Hg in the blood was up to 6 times higher in adults (5.81 ± 1.27 µg g^{-1} dw.) than in nestlings (0.99 ± 0.23 µg g^{-1} dw.). A similar pattern was found for POPs. ∑PCBs was the prevalent group both in adults (median 673, range 336 – 2801 pg g^{-1} ww.) and nestlings (median 41, range 19 – 232 pg g^{-1} ww.), followed by the sum of dichlorodiphenyltrichloroethanes and metabolities (∑DDTs), showing a median value of 220 (range 75 – 2342 pg g^{-1} ww.) in adults and 25 (range 13 – 206 pg g^{-1} ww.) in nestlings. The isotope data suggested that the accumulation of trace elements and POPs between adults and nestlings could be due to parental foraging in two different areas during incubation and chick rearing, respectively, or due to a shift in the feeding strategies along the breeding season. In conclusion, our work showed high Hg concentration in frigatebirds compared to non-contaminated seabird populations, while other trace elements showed lower values within the expected range in other seabird species. Finally, POP exposure was found generally lower than that previously measured in other seabird species.

Capsule abstract

In the present study we found high levels of mercury and low levels of persistent organic pollutants in a tropical seabird breeding in a protected area.

Keywords: trace elements, persistent organic pollutants, seabirds, contaminants, French Guiana.

Introduction

Since the last few decades, there has been a significant increase of trace element contamination of the environment and, among those trace elements, mercury (Hg) is a highly toxic non-essential metal. Overall, Hg derives from both natural and anthropogenic sources, but human activities have increased the global amount of circulating Hg. Once deposited in aquatic ecosystems, inorganic Hg is subject to biotic reactions (e.g. methylation) resulting in the production of methylmercury (Me-Hg). Me-Hg is the highly toxic form of Hg in organisms that assimilated it via food intake. Once incorporated in organisms, Me-Hg biomagnifies within food webs from lower to higher trophic levels. Hg has neurological and endocrinological effects and impacts reproduction, behaviour, development, and ultimately demography in humans and wildlife (Wolfe et al. 1998; Tan et al. 2009), especially in those species which occupy a high trophic level (e.g. seabirds), and are therefore potentially exposed to high contaminant loads (Frederick and Jayasena 2010; Tartu et al 2013; Goutte et al. 2014a). Birds are also vulnerable to other trace metals, particularly to non-essential trace elements such as silver (Ag), cadmium (Cd) and lead (Pb), which although much less studied (Burger 2008), have the potential to negatively affect reproduction, survival and growth (Scheuhammer 1987; Larison et al. 2000). High exposure to essential trace elements has been sometime associated with negative effects in birds (Sánchez-Virosta et al. 2015), but since wild birds are often exposed to a mixture of trace elements, it is generally difficult to demonstrate a causal link between environmental levels of specific compounds and health impairments (Burger 2008, Sánchez-Virosta et al. 2015). Similarly, several persistent organic pollutants (POPs), have been associated with many physiological, immune, endocrine, fitness and demographic consequences and with a decrease in the reproductive success (Bustnes et al. 2006; Verreault et al. 2010, Erikstad et al. 2013; Costantini et al. 2014). Although a long term study on the spatial and temporal trends of POPs revealed that these compounds are expected to decline in the Northern Hemisphere (Braune et al. 2005), they appear to still represent a potential threat to adult survival and thus for population dynamics (Goutte et al. 2015). Several studies on seabirds have focused their attention on the contamination in the polar regions (Goutte et al. 2014b; Goutte et al. 2015; Tartu et al. 2015; Bustnes et al. 2015), which are indeed considered a sink for Hg and organic pollutants (Gabrielsen and Henriksen 2001). Most of these contaminants, including Hg from coal burning sources and pesticides used in agriculture are primarily released from the industrialised areas, and their transport to the Arctic region occurs mainly via the atmosphere but also through large rivers and oceanic currents (Gabrielsen and Henriksen 2001). Compared to polar breeding sites, the level of knowledge is much less about contaminant exposure of seabirds in tropical regions. Moreover, since individual detection probabilities of seabirds at breeding colonies are generally high because of high overall site fidelity (Gauthier et al. 2012), and since long lived apex predators should be particularly exposed to persistent and biomagnifying contaminants (Rowe 2008), many seabirds species are ideal models to assess the physiological and behavioural effects of environmental pollution.

The main goal of this study was to investigate the presence of trace elements and POPs in a long-lived seabird, Magnificent frigatebirds (*Fregata magnificens*, hereafter frigatebirds) breeding at Grand Connétable Island, a small island of the coasts of French Guiana, which offers a unique situation to study contaminants in a multiple stressor framework. The assessment of POPs and toxic trace elements in tropical regions, which are well known for their complex ecosystem structure and their high biodiversity, is a significant environmental pollution issue. Information on POPs and trace elements is missing in high trophic level species in this region, and an assessment of contaminant exposure has been previously focussed only on Hg accumulation in humans and fish (Fréry et al. 2001; Fujimura et al. 2012). Moreover, since stable carbon and nitrogen isotope measurements have been successfully used to describe the trophodynamics of trace elements and POPs in marine ecosystems (Bearhop et al. 2000; Eulaers et al. 2014), stable isotopes were analysed to study the role of dietary contaminant pathway. Additionally, Global Positioning System (GPS) tracking was conducted on adult frigatebirds of this colony to identify the foraging areas and then the possible sources of the contamination during reproduction.

Material and methods

2.1 Sample collection

The field sampling was carried out in 2013 on Grand Connétable island, a protected area located off the Atlantic coast of South America (French Guiana, 4°49'30N; 51°56'00W). This island hosts a unique colony of Magnificent frigatebird that is considered one of the most important in South America, and represents the only breeding site for this seabird species in French Guiana (Dujardin and Tostain 1990). Breeding adults (n = 20, 11 females and 9 males during the incubation/early brooding stage) and 30 days old nestlings (n = 20) were captured by hand or with a nose at the end of a fishing rod (Chastel et al. 2005) on May 27th - 28th and June 25th, respectively. Adults and nestlings were not related to each other. Within few minutes after capture, 2 mL of blood were collected from the brachial vein using a heparinized syringe and a 25G needle. Samples were immediately put on ice and centrifuged in the field within less than 1 hour to separate plasma (to be used for POPs) and red blood cells (to be used for trace elements and stable isotopes). After centrifugation, both plasma and red blood cells were kept in dry ice until the end of the field work and, when at the laboratory, were kept in a -20 ^{0}C freezer until laboratory analysis.

2.2 Stable isotope analysis

The isotopic niche of frigatebirds was used as a proxy of their ecological niche, with δ^{13}C values of seabirds indicating foraging habitats and δ^{15}N values indicating trophic level (Newsome et al. 2007). The stable isotopic method is based on time-integrated assimilated food, with different tissues recording trophic information over different time scales. In the present study, δ^{13}C and δ^{15}N values were measured in red blood cells, which provide trophic information on a few weeks before sampling (Hobson & Clark 1993). Analyses were performed on lyophilised red blood cells of which 0.30 ± 0.05 mg subsamples were weighed in tin cups for stable isotope analyses. Isotopic analyses were performed at the Littoral Environnement et Sociétés (LIENSs) laboratory at the University of La Rochelle (France) with a Thermo Scientific Delta V Advantage mass spectrometer coupled to a Thermo Scientific Flash EA1112 elemental analyser. The results are expressed in the usual δ (‰) notation relative to the deviation from international reference standards (Pee Dee Belemnite for δ^{13}C and atmospheric nitrogen for δ^{15}N). Based on replicate measurements of internal laboratory standards, the experimental precision did not exceed ± 0.15 and ± 0.20‰ for δ^{13}C and δ^{15}N, respectively.

2.3 Contaminant analysis

2.3.1 Trace elements

The analysis of trace element concentrations was carried out by the Littoral Environnement et Sociétés (LIENSs) laboratory at the University of La Rochelle (France). Fourteen trace elements were analysed on lyophilized red blood cells. Total Hg was quantified with an Altec Advanced Mercury Analyzer AMA 254 spectrophotometer. Prior and after freeze-drying, blood samples were weighed to determine the percentage of water in blood, and aliquots ranging from 5 to 10 mg were analysed for quality assessment, as described in Bustamante et al. (2008). Arsenic (As), chromium (Cr), copper (Cu), iron (Fe), manganese (Mn), selenium (Se), and zinc (Zn) were analyzed using a Varian Vista-Pro ICP-OES and silver (Ag), cadmium (Cd), cobalt (Co), nickel (Ni), lead (Pb), and vanadium (V) using a Series II Thermo Fisher Scientific ICP-MS (aliquots mass: 50 − 200 mg dw.) as described in Bustamante et al. (2008). These elements were selected on 2 bases: a first set of non-essential elements (Ag, Cd, Hg and Pb) and a second set of essential trace elements whose metabolism is disrupted by the non-essential ones (Bustamante et al. 2008). Certified Reference Materials (CRM; dogfish liver DOLT-3, NRCC, and lobster hepatopancreas TORT-2, NRCC) were treated and analysed in the same way as the samples. Results were in good agreement with the certified values, and the standard deviations were low, proving good repeatability of the method. The results for CRMs displayed recoveries of the elements ranging from 88% to 116% (n = 10). All the results for trace elements are presented in absolute concentrations in μg g^{-1} dry weight (dw.).

2.3.2 POPs

The analysis of POPs was performed at the Toxicological Centre of the University of Antwerp (Belgium). The analytical protocol was based on the methods described earlier by Eulaers et al. (2011) and consisted in the processing of 1 mL of plasma by solid-phase extraction and clean-up on silica acidified with sulfuric acid (44% w/w). The protocol allowed for the analysis for 26 PCB congeners (CB 28, 49, 52, 74, 99, 101, 105, 118, 128, 138, 146, 153, 156, 170, 171, 174, 177, 180, 183, 187, 194, 196, 199, 203, 206, and 209), organochlorine pesticides (OCPs), amongst which dichlorodiphenyltrichloroethane (*p,p'*-DDT) and its metabolite dichlorodiphenyldichloroethylene (*p,p'*-DDE), hexachlorobenzene (HCB), cis-nonachlor (CN), trans-nonachlor (TN), oxychlordane (OxC), *β*- and *γ*-hexachlorocyclohexanes (HCHs), and 7 polybrominated diphenyl ethers (PBDEs: BDE 28, 47, 99, 100, 153, 154, and 183). Internal standards (CB 143, *ε*-HCH and BDE 77) were used to quantify the targeted compounds using gas chromatography (Agilent GC 6890, Palo Alto, CA, USA) coupled to mass spectrometry (Agilent MS 5973). Most PCB congeners, as well as *p,p'*-DDT and *p,p'*-DDE were separated using a HT-8 capillary column (30 m*0.22 mm*0.25 μm; SGE Analytical Science, Zulte, Belgium), with the mass spectrometer operated in electron impact ionization mode. The remaining PCB congeners, as well as HCB, CHLs (Chlordanes), HCHs, and PBDEs were separated using a DB-5 capillary column (30 m*0.25 mm*0.25 μm; J&W Scientific, Folsom, CA, USA) and the mass spectrometer was operated in electron capture negative ionization mode. Mean ± SD recoveries of the internal standards CB 143 and BDE 77 were 86 ± 6 % and 93 ± 10 %, respectively. Procedural blanks were analysed every 12th plasma sample, and plasma concentrations were corrected for average procedural blank values. The limit of quantification (LOQ) was compound-specifically set at 3*SD of the procedural blank concentration or, for compounds not detected in blanks, set at a 10:1 signal to noise ratio.

2.4 Global Positioning System (GPS) transmitters

During the breeding season of 2011, from July 5th to 7th, 12 brooding adults were equipped with GPS data loggers (Gipsy, Technosmart, Rome, Italy), which recorded GPS locations per second for 32 up to 85 h. GPS data loggers were taped to the back or tail feathers using Tesa© tape, and weighted ~20g, which represented <2% of the bird weight. Since birds needed to be recaptured to recover the GPS, data were recovered from 7 GPS units only.

2.5 Statistical analysis

A principal component analysis (PCA) based on correlation matrix with a direct oblimin factor rotation (i.e., oblique) solution was used to reduce the number of variables into a few representative variables explaining variability in metal accumulation. This approach was preferred instead of examining each metal or POP separately, because (i) concentrations are usually correlated with each other and (ii) this enabled us to reduce the number of statistical models because running many models may increase the chance for type II error. Trace elements and POPs with concentrations below the LOQ were replaced with a value equal to ½*LOQ. Ag, Cd, Co, Cr, Ni, and V had a concentration below the LOQ in all individuals and therefore were not included in the PCA on trace elements, while the other trace elements were quantified in all individuals. Among POPs, PBDEs group was not included in the PCA, since concentrations were below the LOQ for each individual. Then, compounds from the same class were grouped (PCBs, DDTs, and CHLs), and the PCA was applied. In addition, the suitability of the use of PCA to reduce data was tested through the Kaiser-Mayer-Olkin measure of sampling adequacy (K-M-O = 0.70 for trace elements and K-M-O = 0.60 for POPs) and the Bartlett's test of sphericity ($p < 0.01$ for both PCAs), showing the appropriate power of the PCA. After examination of the scree plot, the number of significant principal components was selected on the basis of the Kaiser criterion with eigenvalue higher than 1 (Kaiser 1960). According to Frontier (1976), eigenvalues are considered interpretable if they exceed eigenvalues generated by the broken-stick model, so the Broken Stick model performed with the PAST software (3.08 version) was utilized to underline which axes significantly explained variance in our data-set. To compare differences among adults and nestlings in the content of POP and trace elements, a parametric test was used when data were normally distributed, and non-parametric test were utilized when data were not normally distributed. The Spearman's rho test was used to test correlations among different POPs. Data on POPs have been reported as median value since they showed a wide range among samples, hence the mean value would have been an overestimation. Finally, the correlation among trace elements and stable isotope values and among POP groups and stable isotope values were also estimated, respectively, using the Spearman's rho correlation. Since in order to decrease Hg toxicity there should be an amount of Se available equal or higher than that of Hg so that the molar ratio of Se:Hg is greater than 1 (Raymond and Ralston 2009), the molar ratio Se:Hg was calculated using the formula "molar concentration (mol g^{-1} of dw.) = concentration (µg g^{-1} dw.) * 1000 / atomic weight (g mol^{-1})". All statistical analyses were performed using SPSS (22.0.0 version).

Results

3.1 Trace elements

Of the fourteen trace elements analysed, six had a concentration below the LOQ (Ag, Cd, Co, Cr, Ni and V) both in adults and nestlings while the remaining eight were quantifiable in all individuals, including both essential (As, Cu, Fe, Mn, Se, and Zn) and non-essential (Hg and Pb) elements (Table 1). Fe and Zn reported the highest concentrations among essential elements (2413 ± 68 in adults and 2330 ± 80 in nestlings for Fe, and 19.44 ± 0.90 in adults and 26.93 ± 2.95 in nestlings for Zn expressed as µg g^{-1} dw.). Notably, Hg had a quantifiable concentration in all individuals and showed the highest concentration among non-essential elements (5.81 ± 1.27 in adults and 0.99 ± 0.23 µg g^{-1} dw. in nestlings; Table 1 and S1). Blood concentrations of As, Fe, Pb and Se were significantly higher in adults than in nestlings ($p < 0.05$), while Mn and Zn were significantly higher in nestlings ($p < 0.01$), and Cu was similar between adults and nestlings ($p = 0.06$, Table 1). In particular, Hg showed significantly higher concentrations among the two groups, with adults showing a mean concentration of six times higher than the one for nestlings ($p < 0.01$). The Se:Hg molar ratio in adults (3.9) was almost 4 times lower than the one in nestlings (15). PCA reduced the targeted eight trace elements to three components (explaining 46.22%, 18.15% and 13.58% of the total variance, respectively) , while the Broken Stick model suggested to focus on PC1 only. As, Hg, Fe, Mn, and Zn were associated with the first axis (Figure 1), and according to the t-test, the age of the individuals (nestlings or adults) was a significant variable explaining the variation of trace elements along PC1 ($t = -10.70$, $p < 0.01$; Figure 2). In this scenario, 46.22 % of the total variance was explained by the differences in trace element concentrations between adults and nestlings. Adult females and males differed only for Mn (higher in females, $p = 0.03$) and Se (higher in males, $p = 0.03$) concentration.

3.2 POPs

Of the 40 POP compounds targeted, 15 were not detected in both adults and nestlings, and some congeners were below the LOQ for one group only (either adults or nestlings, Table 2). On average, ΣPCBs, ΣCHLs, ΣDDTs were higher in adults than in nestlings ($p < 0.01$), while HCBs ($p = 0.383$) and ΣHCHs ($p = 0.718$) were similar between adults and nestlings. PBDEs were not detected in any sample (Table 2). ∑PCBs was the most important group based in terms of concentration, showing a median (range) of 673 pg g^{-1} ww. (336 - 2801 pg g^{-1} ww.) in adults and 41 pg g^{-1} ww. (19 - 232 pg g^{-1} ww.) in nestlings, followed by ∑DDTs at 220 pg g^{-1} ww. (75 - 2342 pg g^{-1} ww.) in adults and 25 pg g^{-1} ww. (13 - 206 pg g^{-1} ww.) in nestlings. Among adults, the congeners CB 153, 268 pg g^{-1} ww. (114 - 869 pg g^{-1} ww.) and CB 180, 165 pg g^{-1} ww. (78 - 879 pg g^{-1} ww.), contributed most to the ΣPCBs (34% and 24%, respectively), while among nestlings, CB 153, 17 pg g^{-1} ww. (< 1.0 - 84 pg g^{-1} ww.) and CB 180, 8 pg g^{-1} ww. (3.0 - 40 pg g^{-1} ww.) contributed most to ∑PCBs (22 % and 11 %, respectively) (Table 2). Finally, there was a prevalence of heptaCBs (40 ± 9 % in adults and 25 ± 4 % in nestlings) and hexaCBs (46 ± 14 % in adults and 34 ± 9 % in nestlings; Figure S1). DDE was the only congener to differ significantly between adult males and females, with higher values in males ($p < 0.01$).

Spearman’s rho correlation coefficients among POP groups were positive between PCBs and CHLs ($r = 0.91$, $p < 0.01$), PCBs and DDTs ($r = 0.90$, $p < 0.01$), CHLs and DDTs ($r = 0.88$, $p < 0.01$), HCB and HCHs ($r = 0.44$, $p < 0.01$). Finally, the PCA reduced the targeted POPs to a number of two components (explaining 45.72% and 24.76% of the total variance, respectively), while Broken Stick model suggested PC1 as the only significant axis. The results of the PCA for the POP profiles are presented in Figure 3. The POPs profile significantly differed between adults and nestlings along the PC1 ($t = -11.13$, $p < 0.01$; Figure 4).

3.3 Stable isotopes and GPS

Differences between adults and nestlings for stable isotope values were significant for δ^{13}C (adults = –15.01 ± 0.11, nestlings = –15.19 ± 0.09; $p < 0.01$), while they were not different for δ^{15}N (adults = 13.37 ± 0.20, nestlings = 13.41 ± 0.28; $p = 0.99$; Figure 5).

Hg was significantly positively correlated to δ^{15}N in both adults ($r = 0.84$, $p < 0.01$) and nestlings ($r = 0.52$, $p = 0.02$). Moreover, in adults only, there was a significant positive correlation between δ^{15}N and As ($r = 0.66$, $p = 0.01$), and significant negative correlations between δ^{15}N and other trace elements were limited to Pb ($r = -0.52$, $p = 0.02$) and Zn ($r = -0.66$, $p < 0.02$). Adults also showed a positive correlation between δ^{13}C and Hg ($r = 0.51$, $p = 0.02$), while there were no significant correlations among stable isotope values and POPs both in adults and in nestlings.

Finally, GPS tracks showed that 6 out of 7 adults alternated long trips toward Brazilian coasts, south of the Grand Connétable colony, with short trips near the island (Figure 6). They showed a wide variance in the trips and, overall, covered an average distance per foraging roundtrip (one way and return) of 219.3 km with a standard deviation of 173.1 km, with the longest trip being 513.9 km and the shortest 5 km.

Discussion

4.1 Trace elements

Our results showed the presence of a high blood level of Hg in Magnificent frigatebirds breeding in French Guiana. In 2009, the National Forestry Office estimated that in French Guiana 1,333 km of watercourses and 12,000 hectares of tropical forest were directly affected by gold mining (Mansillon et al. 2009), and that the number of illegal mining sites was recently estimated between 500 and 900 (Tudesque et al. 2012). In addition, the changing geomorphology of the Amazon soil is an additional source of Hg (de Oliveira 2001), so that Hg has become a primary pollutant in the Amazonian basin (Roulet et al. 1999) and is a matter of great concern in French Guiana (Fujimura et al. 2012). Even so, an evaluation of its impact on local wildlife is, however, still missing. A previous study has shown how frigatebirds may move up to 1,400km away from the breeding colony outside the breeding season (Weimerskirch et al. 2006), and may therefore be contaminated far from French Guiana. However, the high Hg levels found in both adult and nestling frigatebirds suggest a contamination in the lower trophic levels from the coasts of French Guiana up to the upper Brazilian coasts, which includes the foraging areas of our study population during the breeding season (Figure 6). Since Hg biomagnifies within food webs (Lavoie et al. 2013), adults usually show higher concentrations than nestlings (Carravieri et al. 2014). Consistently, Hg was around six times higher in adults than nestlings (Table 1), and our results showed French Guiana frigatebirds to have values of Hg similar to highly Hg-contaminated species (e.g., *Diomedea exulans*, *Stercorarius skua*) (see Table S1). Such blood Hg concentrations have been associated with both a reduction of parental commitment (Tartu et al. 2016) and of the breeding success (Goutte et al. 2014a). In addition, similar Hg concentrations have been shown to interfere with several endocrine mechanisms (Tartu et al. 2013, 2014) and to increase oxidative stress (Costantini et al. 2014), a condition that may decrease reproductive success (Costantini 2014) and facilitate herpes infection (Sebastiano et al. 2016). In this scenario, it is important to take into consideration that Hg in the nestlings' red blood cells reflects Hg exposure since hatching as well as maternal Hg transfer through the eggs (Lewis et al. 1993), while adult Hg concentrations reflect the exposure since the last moult (Dauwe et al. 2003). So, the Hg content found in the blood of adults might be lower than actually is, since birds are able to excrete Hg in feathers (Dauwe et al. 2003).

The PCA showed that the high Hg concentration is coupled with high levels of As, Fe, and Se and low levels of Mn and Zn, while Cu and Pb did not show a related pattern (Figure 1). However, for some trace elements such as Cu, Fe, Mn, and Pb, concentrations were very low as compared to literature values in other seabird species (Summers et al. 2014; Carravieri et al. 2014). Interestingly, among non-essential trace elements, Ag and Cd were below the LOQ for every sample, Pb concentrations were very low, while Hg was the only non-essential trace element with high concentrations.

A previous study has underlined that As concentrations varied widely among different tissues, being higher in liver and muscle tissues, and varied with the age of the organism, geographic location, and proximity to anthropogenic activities (Eisler 1988). In birds, inorganic As is considered highly toxic in comparison with organic compounds of this element and may disrupt reproduction, and trigger sub-lethal effects or even induce individual’s death (Eisler 1994; Kunito et al. 2008). However, marine animals have only a limited ability to bioaccumulate inorganic arsenic from solution (Neff 1997), so As concentrations in living organisms are generally low (Braune and Noble 2009), and concentrations of As in frigatebirds are much lower than the threshold levels of other seabirds, and therefore should not represent a threat for this population (Eisler 1994).

In contrast to non-essential elements, Zn is an essential micronutrient and its deficiency has been associated to an increase in oxidative stress and DNA damage, and a decrease in antioxidant defences (Song et al. 2009). Zn is also one of the main component of metallothioneins, a group of proteins which play an essential role in heavy metal detoxification (Siscar et al. 2013). A comparison among tissues and different species is difficult to interpret, but Zn content showed concentrations similar to other seabird species (Carvalho et al. 2013; Fromant et al. 2016). However, the PCA has underlined a strong lowering of the Zn content in the individuals with higher levels of Hg. As a result, since Zn has a stimulatory action on the immune response, further studies are warranted in order to clarify if the decrease in Zn content with the increase in Hg might reduce the immune competence of this seabird.

In a different way, Se, besides being an essential constituent of selenoproteins utilised as a cofactor for reduction of glutathione peroxidases, (Beckett and Arthur 2005) is also important for the detoxification of Hg exposure. In fact, previous studies have emphasized the "protective effect" of Se on Hg toxicity (Raymond and Ralston 2009). Its protective effect was initially presumed to involve Se sequestration of Hg, thereby preventing its harmful effects. However, as more has become understood about Se physiology, the mechanism of MeHg/Hg toxicity and the mechanism of Se protective effect have also become clear. The high affinity between Hg and Se results in Hg binding to Se (Ralston and Raymond 2010), with the consequent generation of mercuric selenide (HgSe), which is well known to be a non-toxic form in marine mammals and birds (Nigro and Leonzion 1996; Ikemoto et al. 2004). In order to be able to decrease Hg toxicity, there should be an amount of Se available higher than that of Hg so that the molar ratio of Se:Hg is greater than 1 (Raymond and Ralston 2009). Since the molar ratio was 3.9 for adults and 15 for nestlings, and since the PCA has shown that individuals with high levels of Hg tend to have higher levels of Se, it is likely that Se is contributing to the detoxification of Hg (Sørmo et al. 2011), preventing from Hg toxic effects more in nestlings than in adults. However, Se in blood, which was higher in males, was lower compared to other seabirds (Fromant et al. 2016), and further investigation is needed to understand if the amount of this essential trace element is adequate to contribute to the organisms' physiological functions.

4.2 POPs

To the best of our knowledge, no studies have previously described plasma POP levels in Magnificent frigatebirds or more generally in French Guiana seabirds. In Mexico, a study of Magnificent frigatebirds eggs detected low levels of OCPs and PCBs (Trefry et al. 2013). In French Guiana only one study has investigated whole blood levels of OCPs and PCBs in the eggs of leatherback turtles *Dermochelys coriacea*, and found low levels (Girlet et al. 2010). As mentioned earlier, most seabird POP studies have been conducted on polar and especially Arctic species. In the present study, POP concentrations were generally much lower than what has been found previously in polar seabirds (e.g. Tartu et al. 2015a). The contamination with organochlorine compounds and their metabolites can lead to lethal as well as sub-lethal effects in wildlife (Beyer et al. 1996). In particular, DDTs are highly relevant for apex seabirds, since they are associated with eggshell thinning and thus reduced reproductive success (Beyer et al. 1996). Our results pointed out that the *p,p'*-DDT content was below the LOQ, while *p,p'*-DDE showed a median value of 220 pg g^{-1} ww. in adults (being higher in males), and 25 pg g^{-1} ww. in nestlings, respectively, which are much lower than other top predator seabirds (Bustnes et al. 2006). POPs have never been measured in frigatebird plasma and a reliable comparison with other tissues cannot be made since different tissues show different toxicodynamics.

Comparisons with other species showed PCB 153 and other PCBs, HCB and *p,p'*-DDE to be similar to low contaminated populations of common eider *Somateria mollissima* in the sub-Arctic and high Arctic regions (Bustnes et al. 2012; Fenstad et al. 2014), but much lower than in moderately POPs-contaminated Antarctic seabirds, like the snow petrel (*Pagodroma nivea*; Tartu et al. 2015a). Although ∑PCBs show higher concentrations than the other chemical classes, with a median of 673 pg g^{-1} ww. in adults and 41 pg g^{-1} ww. in nestlings, these concentrations are much lower than those reported in the blood of 7 polar seabirds among which the extremely contaminated Glaucous gull *Larus hyperboreus* (Tartu et al. 2015b). In polar seabirds specifically, high PCB contamination is associated with concentration from dozens (mean 47,000 pg g^{-1} ww., Tartu et al. 2015a) up to hundreds of times higher (mean 448,700 pg g^{-1} ww., Bustnes et al. 2006) than those we found in frigatebirds from French Guiana.

4.3 Stable isotopes and GPS

Differences and similarities in trace elements and POPs between nestlings and adults may be explained by trophic ecology. For example, nestlings may differ from adults in δ^{15}N if they are fed on a different diet or a different trophic level (Overman and Parrish 2001). This explanation is supported by several studies on seabirds (Hobson 1993; Schmutz and Hobson 1998), which found that adults provide to their offspring a food different from that they feed on. One way to increase energy gain per unit time of nestlings would be to increase the size of the fish caught for the nestlings, a strategy that has been recorded in other seabirds (Bugge et al. 2011). However, our results do not support this hypothesis. The similar stable nitrogen isotope values between nestlings and adults suggest that they feed on similar trophic level prey.

On the other hand, the stable carbon isotope values in the present study showed adults to have significantly higher δ^{13}C values than nestlings (Figure 5). A latitudinal decline in δ^{13}C values has been documented in marine mammals and seabirds (Kelly 2000), and studies have shown patterns which might suggest a decreasing δ^{13}C from the coast to the open sea (Eulaers et al. 2014), but information of such stratification in French Guiana is not available. Since at this stage of development, frigatebird nestlings are not able to fly, the stable isotope values in nestlings reflect the prey provided by the adults. Hence, the different carbon stable isotope values between adults and nestlings might be explained in two different ways: (a) adults may get their food in a different feeding area than where they forage for their nestlings (GPS tracks of the breeding season 2011 showed how most adults alternated short trips, mostly to the north, with more long trips in the direction of the Brazilian coasts) (Figure 6); (b) adults may have changed their feeding strategies between the incubation stage and the chick rearing period. In fact, since δ^{13}C in seabird red blood cells reflects up to three-four weeks before the blood sampling (Hobson and Clark 1992), δ^{13}C in adults might have reflected the foraging habitat during the incubation period. In addition, δ^{13}C in nestlings might have reflected the foraging habitat during the beginning of the chick rearing, since nestlings were around 30 days of age. However, differences in the carbon composition are significant from the statistical point of view, but studies are needed to clarify if such difference can be ecologically significant, and if it can be related to the differences in the trace element concentrations.

Conclusions

Although our study provided the first evidence of the presence of POPs in French Guiana frigatebirds, PCB and DDT concentrations were generally lower compared to those found in other seabird species, especially in polar seabirds, and they are not likely to be a threat for this population. However, even if concentrations of these pollutants are low, they may have a combined effect with trace elements and especially Hg. Our study clearly shows that this frigatebird population is bearing high Hg burden, and there is an urgent need to evaluate whether increased blood Hg concentrations may affect endocrine and fitness aspects in this top predator bird, as has been documented in other seabird species. Other essential and non-essential trace elements showed different accumulation in adults and nestlings, but values were in the range of previous studies on other seabirds. Since the trophic position did not differ between adults and nestlings (same nitrogen isotope value), an explanation for the different POPs and metal profiles between adults and nestlings might lie with the foraging area of adults (carbon isotope values), which appeared to change over the breeding season. Furthermore, in our study population, a previous study has reported the occurrence of herpes virus outbreaks in this colony (De Thoisy et al. 2009), which is causing high mortality of nestlings. These herpes virus outbreaks make this population a highly relevant biological model for investigating the interactions between pollutant exposure and impact of virus activity of population viability. Indeed, previous studies have underlined that there might be a strong relation among exposure to trace elements and virus infections (Koller 1975; Gainer 1977), and, more specifically, Hg is highly suspected to aggravate herpes simplex virus-2 infection in mice (Christensen et al. 1996). Our data indicate that future studies may be warranted to better understand if the herpes virus outbreaks in this population are favoured by the high Hg contamination.

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References

Bearhop S., Waldron S., Thompson D. and Furness R. 2000 Bioamplification of mercury in Great Skua *Catharacta skus* nestlings: the influence of trophic status as determined by stable isotopes signatures of blood and feathers. Marine Pollution Bulletin 40, 181-185.

Beckett G.J. and Arthur J.R. 2005 Selenium and endocrine systems. J. Endocrinol. 183, 455-465.

Beyer W.N., Heinz G.H. and Redmon-Norwood A.W. 1996 Environmental Contaminants in Wildlife: Interpreting Tissue Concentrations. CRC press.

Braune B.M., Outridge P.M., Fisk A.T., Muir D.C.G., Helm P.A., Hobbs K., Hoekstra P.F., Kuzyk Z.A., Kwan M., Letcher R.J., Lockhart W.L., Norstrom R.J., Stern G.A. and Stirling I. 2005 Persistent organic pollutants and mercury in marine biota of the Canadian Arctic: an overview of spatial and temporal trends. Sci. Total Environ. 351, 4e56.

Braune B.M. and Noble D.G. 2009 Environmental contaminants in Canadian shorebirds. Environ. Monit. Assess. 148, 185-204.

Bugge J., Barrett R.T. and Pedersen T. 2011 Optimal foraging in chick-raising Common Guillemots Uria aalge. Journal of Ornithology 152, 253–259.

Burger J. 2008 Assessment and management of risk to wildlife from cadmium. Science of The Total Environment 389, 37‑45.

Bustamante P., Gonzàlez A.F., Rocha F., Miramand P. and Guerra A. 2008 Metal and mettaloid concentrations in the giant squid *Architeuthis dux* from Iberian waters. Mar. Environ. Res. 66, 278-287.

Bustnes J.O., Tveraa T., Henden J.A., Varpe Ø., Janssen K. and Skaare J.U. 2006 Organochlorines in Antarctic and Arctic avian top predators: a comparison between the south polar skua and two species of Northern Hemisphere gulls. Environ. Sci. Technol. 40, 2826–2831.

Bustnes J.O., Moe B., Hanssen S.A., Herzke D., Fenstad A.A., Nordstad T., Borga K. and Gabrielsen G.W. 2012 Temporal dynamics of circulating persistent organic pollutants in a fasting seabird under different environmental conditions. Environ. Sci. Technol. 46, 10287-10294.

Bustnes J.O., Bourgeon S., Leat E.H.K., Magnusdóttir E., Strøm H., Hanssen S.A., Petersen A., Olafsdóttir K., Borgå K., Gabrielsen G.W. and Furness R.W. 2015 Multiple Stressors in a top predator seabird: potential ecological consequences of environmental contaminants, population health and breeding conditions. PLoS One 10(7): e0131769.

Carravieri A., Bustamante P., Tartu S., Meillère A., Labadie P., Budzinski H., Peluhet L., Barbraud C., Weimerskirch H., Chastel O. and Cherel Y. 2014 Wandering albratrosses document latitudinal variations in the transfer of persistent organic pollutants and mercury to southern ocean predators. Environmental Science and Technology 48, 14746-14755.

Carvalho P.C., Bugoni L., McGill R.A. and Bianchini A. 2013 Metal and selenium concentrations in blood and feathers of petrels of the genus Procellaria. Environ. Toxicol. Chem. 32, 1641-1648.

Chastel O, Barbraud C, Weimerskirch H, Lormee H, Lacroix A, Tostain O. 2005. High levels of LH and testosterone in a tropical seabird with an elaborate courtship display. General and Comparative Endocrinology 140, 33-40.

Cherel Y. and Hobson K.A. 2007 Geographical variation in carbon stable isotope signatures of marine predators: a tool to investigate their foraging areas in the Southern Ocean. Mar. Ecol. Prog. Ser. 329, 281-287.

Christensen M.M., Ellermann-Eriksen S., Rungby J. and Morgensen S.C. 1996 Influence of mercury chloride on resistance to generalized infection with herpes simplex virus type 2 in mice. Toxicology 114, 57-66.

Costantini D. 2014 Oxidative Stress and Hormesis in Evolutionary Ecology and Physiology. Springer-Verlag, Berlin and Heidelberg SBN 978-3-642-54663-1,pp. 13-20.

Costantini D., Meillere A., Carravieri A., Lecomte V., Sorci G., Faivre B., Weimerskirch H., Bustamante P., Labadie P., Budzinski H. and Chastel O. 2014 Oxidative stress in relation to reproduction, contaminants, gender and age in a long-lived seabird. Oecologia 175, 1107–1116.

Dauwe T., Bervoets L., Pinxten R., Blust R. and Eens M. 2003 Variation of heavy metals within and among feathers of birds of prey: effect of molt and external contamination. Environmental pollution 124, 429-436.

de Oliveira S.M.B., Melfi A.J., Fostier A.H., Forti M.C., Favaro D.I.T. and Boulet R. 2001 Soils as an important sink for mercury in the Amazon, Water Air Soil Pollut. 26, 321-337.

de Thoisy B., Lavergne A., Semelin J., Pouliquen J.F., Blanchard F., Hansen E., and Lacoste V. 2009 Outbreaks of diseases possibly due to a natural avian herpesvirus infection in a colony of young Magnificent Frigatebirds (*Fregata magnificens*) in French Guiana. Journal of Wildlife diseases 45, 802-807.

Dujardin J.L. and Tostain O. 1990 Les oiseaux de mer nicheurs de Guyane Française. Alauda 58, 107-134.

Eisler R. 1988 Arsenic hazards to fish, wildlife, and invertebrates: a synoptic review. Biological Report 85 (1.12).

Eisler R. 1994 A review of arsenic hazards to plants and animals with emphasis on fishery and wildlife resources. John Wiley & Sons, Inc. New York.

Erikstad K.E., Sandvik T., Reinertsen T.K., Bustnes J.O. and Strom H. 2013 Persistent organic pollution in a high-artic top predator: sex dependent thresholds in adult survival. Proc. Roy. Soc. London Ser. B 280(1769)20131483.

Eulaers I., Covaci A., Hofman J., Nygard T., Halley D.J., Pinxten R., Eens M. and Jaspers V.L.B. 2011 A comparison of non-destructive sampling strategies to assess the exposure of white-tailed eagle nestling (*Haliaeetus albicilla*) to persistent organic pollutants. Sci. Total Environ. 410, 258-265.

Eulaers I., Jaspers V.L.B., Halley D.J., Lepoint G., Nygård T., Pinxten R., Covaci A. and Eens M. 2014 Brominated and phosphorus flame retardants in White-tailed Eagle *Haliaeetus albicilla* nestlings: Bioaccumulation and associations with dietary proxies (δ13C, δ15N and δ34S). Sci. Total Environ. 478, 48-57.

Fenstad A.A., Jenssen B.M., Moe B., Hanssen S.A., Bingham C., Herzke D., Bustnes J.O. and Krokje A. 2014 DNA double-strand breaks in relation to persistent organic pollutants in a fasting seabird. Ecotoxicology and Environmental Safety 106, 68-75.

Frederick P. and Jayasena N. 2010 Altered pairing behaviour and reproductive success in white ibises exposed to environmentally relevant concentrations of methylmercury. Proceedings of the Royal Society B 278, 1851–1857.

Fréry N., Maury-Brachet R., Maillot E., Deheeger M., de Mérona B. and Boudou A. 2001 Gold-mining activities and mercury contamination of native amerindian communities in French Guiana: key role of fish in dietary uptake. Environ. Health Perspect. 109, 449-456.

Fromant A., Carravieri A., Bustamante P., Labadie P., Budzinski H., Peluhet L., Churlaud C., Chastel O. and Cherel Y. 2016 Wide range of metallic and organic contaminants in various tissues of the antartic prion, a planktonophagous seabird from the Southern Ocean. Sci. Total Environ. 544, 754-764.

Frontier S. 1976 Étude de la décroissance des valeurs propres dans une analyze en composantes principales: comparison avec le modèle de baton brisé. J. Exp. Mar. Biol. Ecol. 25, 67-75.

Fujimura M., Matsuyama A., Harvard J.P., Bourdineaud J.P. and Nakamura K. 2012 Mercury contamination in humans in upper Maroni, French Guiana between 2004 and 2009. Bull. Environ. Contam. Toxicol. 88, 135-139.

Gabrielsen G.W. and Henriksen E. 2001 Persistent organic pollutants in Arctic animals in the Barents Sea area and at Svalbard: levels and effects. Mem. Natl. Inst. Polar Res. 54, 349e364.

Gainer J.H. 1977 Effects of heavy metals and the deficiency of zinc on mortality rates in mice infected with encephalomyocarditis virus. Am. J. Vet. Res. 38, 869-672.

Girlet E., Das K., Thomé J.P., Girondot M. 2010 Maternal transfer of chlorinated contaminants in the leatherback turtles, *Dermochelys coriacea*, nesting in French Guiana. Chemosphere 79, 720–726.

Goutte A., Bustamante P., Barbraud C., Delord K., Weimerskirch H. and Chastel O. 2014a. Demographic responses to mercury exposure in two closely-related Antarctic top predators. Ecology 95, 1075-1086.

Goutte A., Barbraud C., Meillère A., Carravieri A., Bustamante P., Labadie P., Budzinski H., Delord K., Cherel Y., Weimerskirch H. and Chastel O. 2014b Demographic consequences of heavy metals and persistent organic pollutants in a vulnerable long-lived bird, the wandering albatross. Proceedings of the Royal Society B 281(1787), 20133313.

Goutte A., Barbraud C., Herzke D., Bustamante P., Angelier F., Tartu S., Clément-Chastel C., Moe B., Bech C., Gabrielsen G.W., Bustnes J.O. and Chastel O. 2015 Survival rate and breeding outputs in a high Arctic seabird exposed to legacy persistent organic pollutants and mercury. Environmental Pollution 200, 1-9.

Hobson K.A. 1993 Trophic relationship among high Artic seabirds: insights from tissue-dependent stable-isotope models. Mar. Ecol. Progr. Ser. 95, 7-18.

Hobson K.A. and Clark R.G. 1992 Assessing avian diets using stable isotopes I: turnover of 13C in tissues. Condor 94, 181–188.

Hobson K.A. and Clark R.G. 1993 Turnover of 13C in cellular and plasma fractions of blood: Implications for nondestructive sampling in avian dietary studies. Auk 110, 638−641

Ikemoto T., Kunito T., Tanaka H., Baba N., Miyazaky N. and Tanabe S. 2004 Detoxification mechanism of heavy metals in marine mammals and seabirds: interaction of selenium with mercury, silver, copper, zinc, and cadmium in liver. Arch. Environ. Contam. Toxicol. 47, 402-413.

Kaiser H.F. 1960 The Application of Electronic Computers to Factor Analysis. Educational and Psychological Measurement 20, 141-151.

Kelly J.F. 2000 Stable isotopes of carbon and nitrogen in the study of avian and mammalian trophic ecology. Can. J. Zool. 78, 1-27.

Koller L.D. 1975 Methylmercury: effect on oncogenic and non-oncogenic viruses in mice. Am. J. Vet. Res. 36, 1501-1504.

Kunito T., Kubota R., Fujihara J., Agusa T. and Tanabe S. 2008 Arsenic in marine mammals, seabirds, and sea turtles. Rev. Environ. Contam. Toxicol. 195, 31-69.Martoja R. and Berry J.P. 1981 Identification of tiemannite as a probable product of delmethylation of mercury by selenium in cetaceans. Vie Milieu 30, 7-10.

Larison J.R., Likens G.E., Fitzpatrick J.W. and Crock J.G. 2000 Cadmium toxicity among wildlife in the Colorado Rocky Mountains. Nature 406, 181-183.

Lavoie R.A., Jardine T.D., Chumchal M.M., Kidd K.A. and Campbell L.M. 2013 Biomagnification of mercury in aquatic food webs: a worldwide meta-analysis. Environ. Sci. Technol. 47(23): 13385-94.

Lewis S.A., Becker P.H. and Furness R.W. 1993 Mercury levels in eggs, tissues, and feathers of herring gulls *Larus argentatus* from the German Wadden Sea Coast. Environ. Pollut. 80,293-299.

Mansillon Y., Allain Y.M., de Chalvron J.G., Hirtzman P. 2009 Proposition de schéma departmental d'orientation minière de la Guyane, rapport au president de la République. Ministère de l'environnement, de l'énergie et de la mer.

Neff J.M. 1997 Ecotoxicology of arsenic in the marine environment. Environ. Toxicol. Chem 16, 917-927.

Newsome S.D., Martinez del Rio C., Bearhop S., and Phillips D.L. 2007 A niche for isotopic ecology. Front. Ecol. Environ. 5, 429−436

Nigro M. and Leonzio C. 1996 Intracellular storage of mercury and selenium in different marine vertebrates. Marine Ecology Progress Series 135, 137-143.

Overman N.C. and Parrish D.L. 2001 Stable isotope composition of walleye: δ^{15}N accumulation with age and area specific differences in δ^{13}C. Can. J. Fish. Aquat. Sci. 58, 1253-1260.

Raymond L.J. and Ralston N.V.C. 2009 Selenium's importance in regulatory issues regarding mercury. Fuel Process. Technol. 90, 1333–1338.

Roulet M., Lucotte M., Farella N., Serique G., Coelho H., Sousa-Passos C.J., da Silva J.E., de Andrade S.P., Mergler D., Guimaraes J.R.D. and Amorim M. 1999 Effects of recent human colonization on the presence of mercury in Amazonian ecosystems. Water Air Soil Pollut 112, 297-313.

Rowe C.L. 2008 The calamity of so long life: life histories, contaminants, and potential emerging threats to long -lived vertebrates. Bioscience 58, 623-631.

Sánchez-Virosta P., Espín S., García-Fernanández A.J. and Eeva T. 2015 A review on exposure and effects of arsenic in passerine birds. Sci. Tot. Environ. 512-513, 506-525.

Scheuhammer A.M. 1987 The chronic toxicity of aluminium, cadmium, mercury, and lead in birds: a review. Environmental Pollution 46, 263-295.

Schmutz J.A. and Hobson K.A. 1998 Geographic, temporal and age-specific variation in diets of glaucous gulls in Western Alaska. Condor 100, 119-130.

Sebastiano M., Chastel O., de Thoisy B., Eens M. and Costantini D. 2016 Oxidative stress favours herpes virus infection in vertebrates: a meta-analysis. Current Zoology (in press).

Siscar R., Koenig S., Torreblanca A. and Solè M. 2013 The role of metallothionein and selenium in metal detoxification in the liver of deep-sea fish from the NW Mediterranean Sea. Sci. Total Environ. 466-467, 898-905

Song Y., Leonard S.W., Traber M.G. and Ho E. 2009 Zinc deficiency affects DNA damage, oxidative stress, antioxidant defences, and DNA repair in rats. Journal of Nutrition 139, 1626-1631.

Sørmo E.G., Ciesielski T.M., Øverjordet I.B., Lierhagen S., Eggen G.S., Berg T. and Jenssen B.M. 2011 Selenium moderates mercury toxicity in free-ranging freshwater fish. Environ. Sci. Technol. 45, 6561–6566.

Summers C.F., Bowerman W.W., Parsons N., Chao W.Y. and Bridges W.C.Jr. 2014 Lead and Cadmium in the Blood of nine species of seabirds, Marion Island, South Africa. Bull. Environ. Contam. Toxicol. 93, 417-422.

Tan S.W., Meiller J.C., and Mahaffey K.R.. 2009 The endocrine effects of mercury in humans and wildlife. Critica Reviews in Toxicology 39, 228–269.

Tartu S., Goutte A., Bustamante P., Angelier F., Moe B., Clément-Chastel C., Bech C., Gabrielsen G.W., Bustnes J.O. and Chastel O. 2013 To breed or not to breed: endocrine response to mercury contamination by an arctic seabird. Biology Letters 9:20130317.

Tartu S., Bustamante P., Goutte A., Cherel Y., Weimerskirch H., Bustnes J.O. and Chastel O. 2014 Age-related mercury contamination and relationship with luteinizing hormone in a long-lived Antartic bird. PLoS One 29, 9(7).

Tartu S., Angelier F., Wingfield J.C., Bustamante P., Labadie P., Budzinski H., Weimerskirch H., Bustnes J.O. and Chastel O. 2015a Corticosterone, prolactin and egg neglect behavior in relation to mercury and legacy POPs in a long-lived Antarctic bird. Sci. Total Environ. 505, 180-188.

Tartu S., Angelier F., Bustnes J.O., Moe B., Hanssen S.A., Herzke D., Gabrielsen G.W., Verboven N., Verreault J., Labadie P., Budzinski H., Wingfield J.C., Chastel O. 2015b Polychlorinated biphenyl (PCB) exposure and adrenocortical function in seven polar seabirds. Environmental Pollution 197, 173-180.

Tartu S., Bustamante P., Angelier F., Lendvai A.Z., Moe B., Blévin P., Bech C., Gabrielsen G.W., Bustnes J.O. and Chastel O. 2016 Mercury exposure, stress and prolactin secretion in an Arctic seabird: an experimental study. Functional Ecology DOI:10.1111/1365-2435.12534.

Thompson D.R. and Furness R.W. 1989 The chemical form of mercury stored in South Atlantic seabirds. Environ. Pollut. 60, 305-317.

Trefry S.A., Diamond W.A., Spencer N.C., Mallory M.L. 2013 Contaminants in Magnificent frigatebird eggs from Barbuda, West Indies. Marine Pollution Bulletin 75, 317-321.

Tudesque L., Grenouillet G., Gevrey M., Khazraie K., Brosse S. 2012 Influence of small-scale gold mining on French Guiana streams: Are diatom assemblages valid disturbance sensors? Ecological Indicators 14, 100-106.

Verreault J, Bustnes J.O., Gabrielsen G.W. 2010 The Svalbard glaucous gull (Larus hyperboreus) as bioindicator species in the Norwegian Arctic: a review of 35 years of contaminant research. Rev. Environ. Contam. Toxicol. 205 , 77–116. (doi:10. 1007/978-1-4419-5623-1_2).

Weimerskirch H., Le Corre M., Marsac F., Barbraud C., Tostain O. and Chastel O. 2006 Postbreeding movements of frigatebirds tracked with satellite telemetry. The Condor 108, 220-225.

Wolfe M.F., Schwarzbach S. and Sulaiman R.A. 1998 Effects of mercury on wildlife: a comprehensive review. Environmental Toxicology and Chemistry 17, 146–160.

Figure captions

Figure 1. Plot of the PC1 correlation coefficients on trace elements. **Correlation is significant at the 0.01 level (2-tailed). *Correlation is significant at the 0.05 level (2-tailed).

Figure 2. Scatter plot of the principal component analysis for trace elements for adult individuals (black squares) and nestlings (red circles).

Figure 3. Plot of the PC1 correlation coefficients on POPs. **Correlation is significant at the 0.01 level (2-tailed).

Figure 4. Scatter plot of the principal component analysis for POPs for adult individuals (black squares) and nestlings (red circles).

Figure 5. Stable carbon and nitrogen isotope values (mean ± SD) of red blood cells of adults and nestlings of the Magnificent frigatebird from French Guiana.

Figure 6. GPS tracks of seven Magnificent frigatebirds adults recorded during the breeding season of 2011. The white dotted line represents the political border between French Guiana and Brazil.

Figures

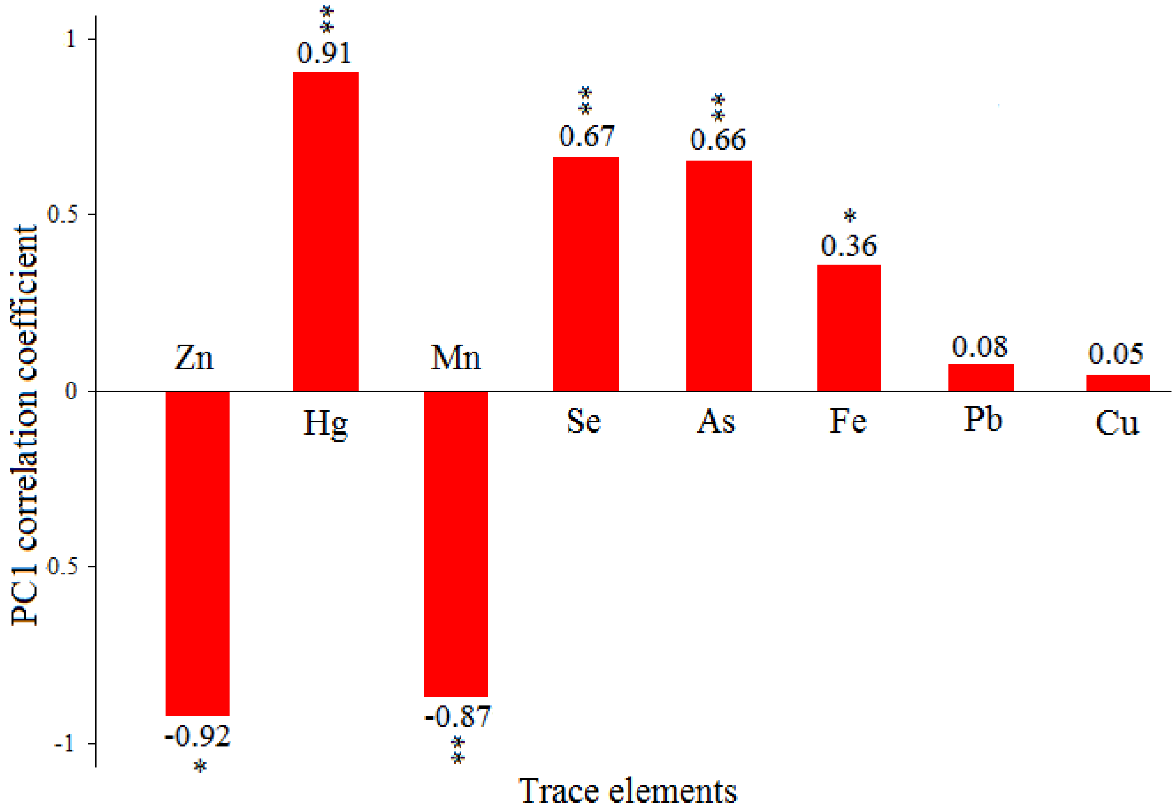
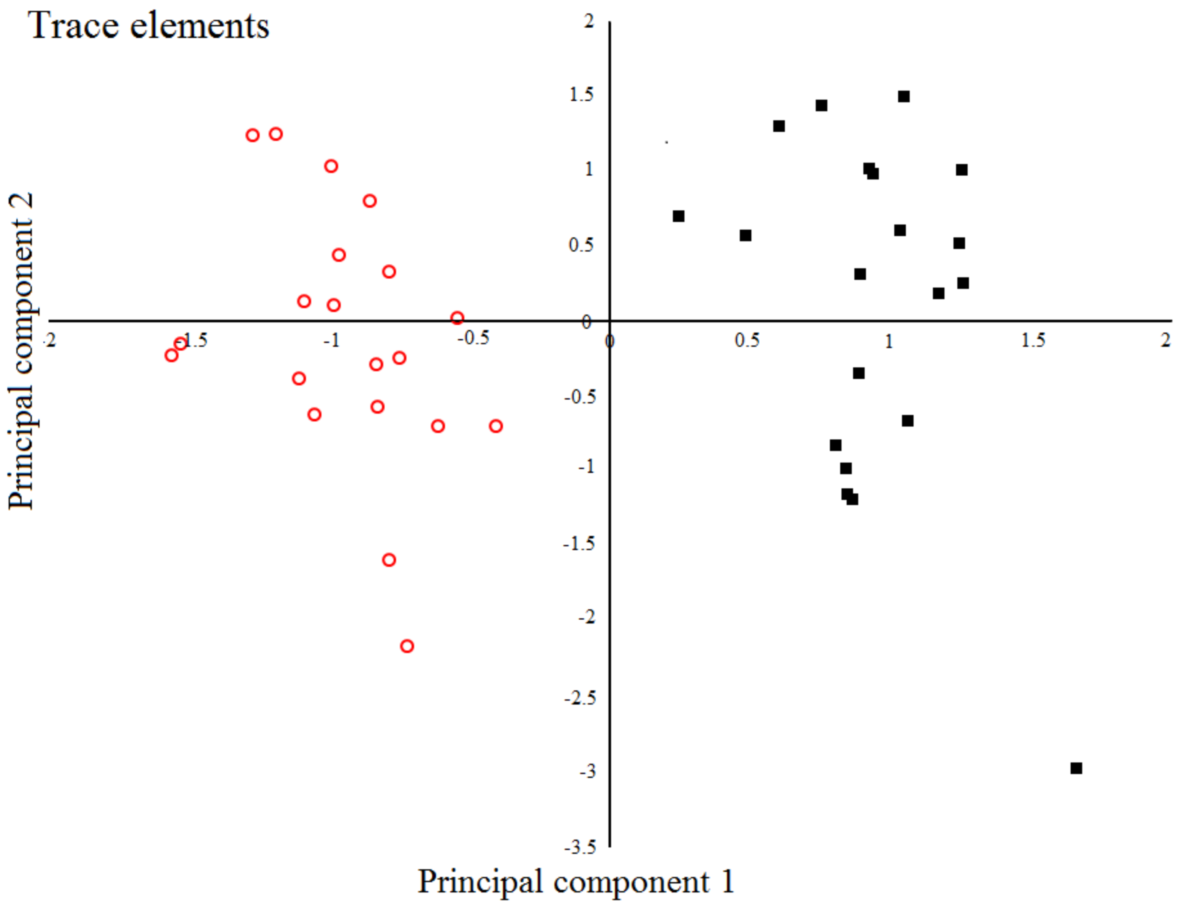


Figure 1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Principal component 1
2
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1
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-1
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-2
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-1
-0.5
0
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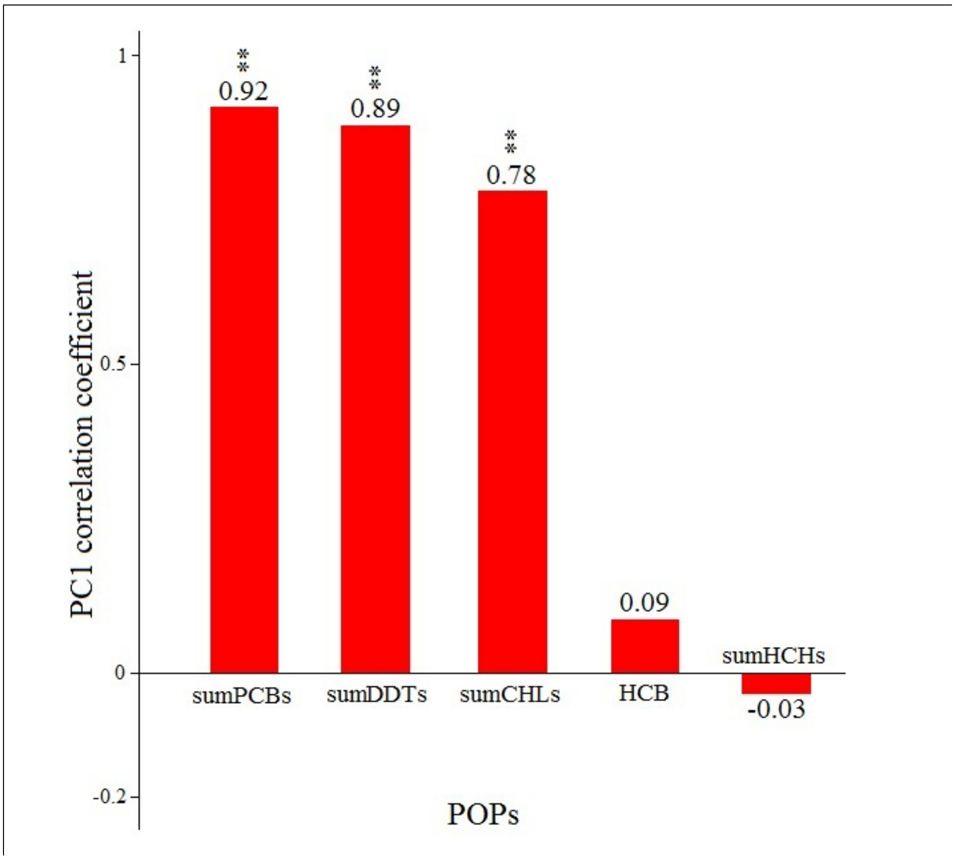
1
1.5
2

Figure 2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**
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**
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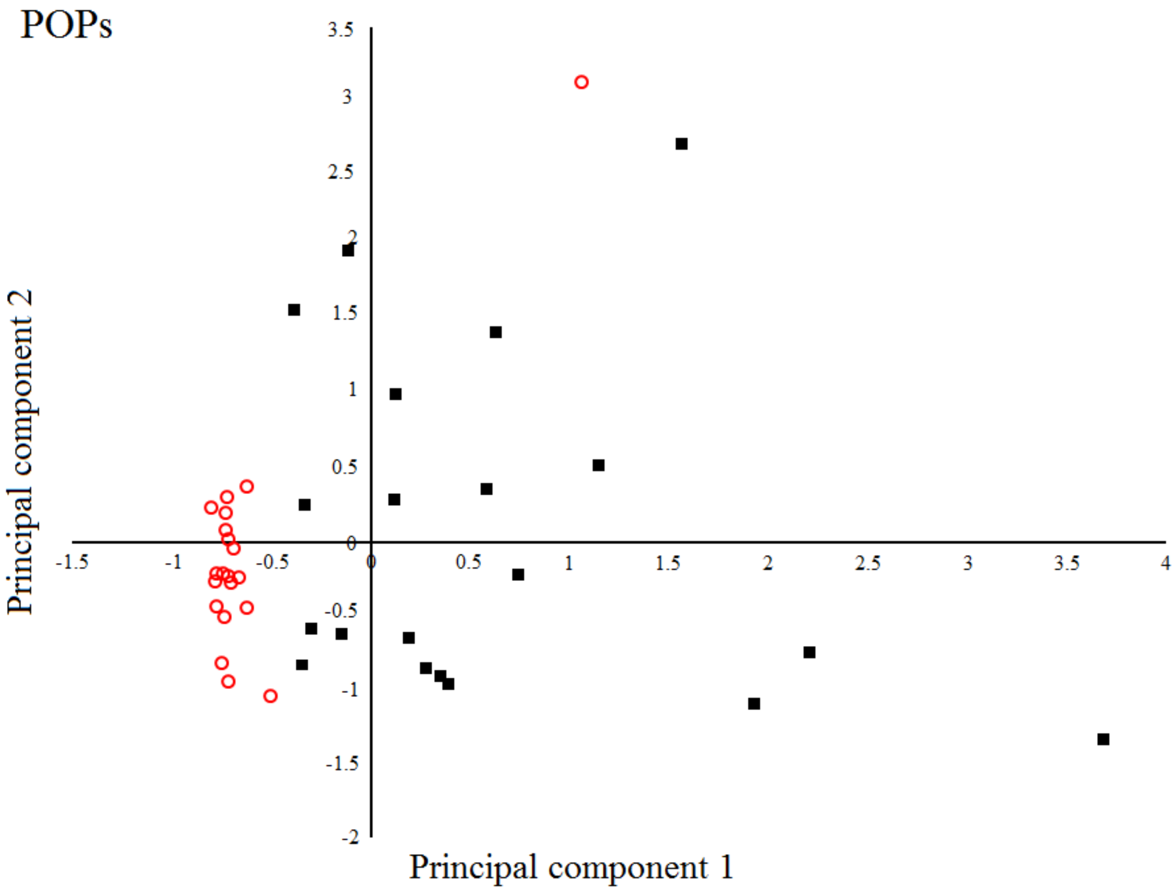
sumCHLs
HCB
sumHCHs
POPs

Figure 3



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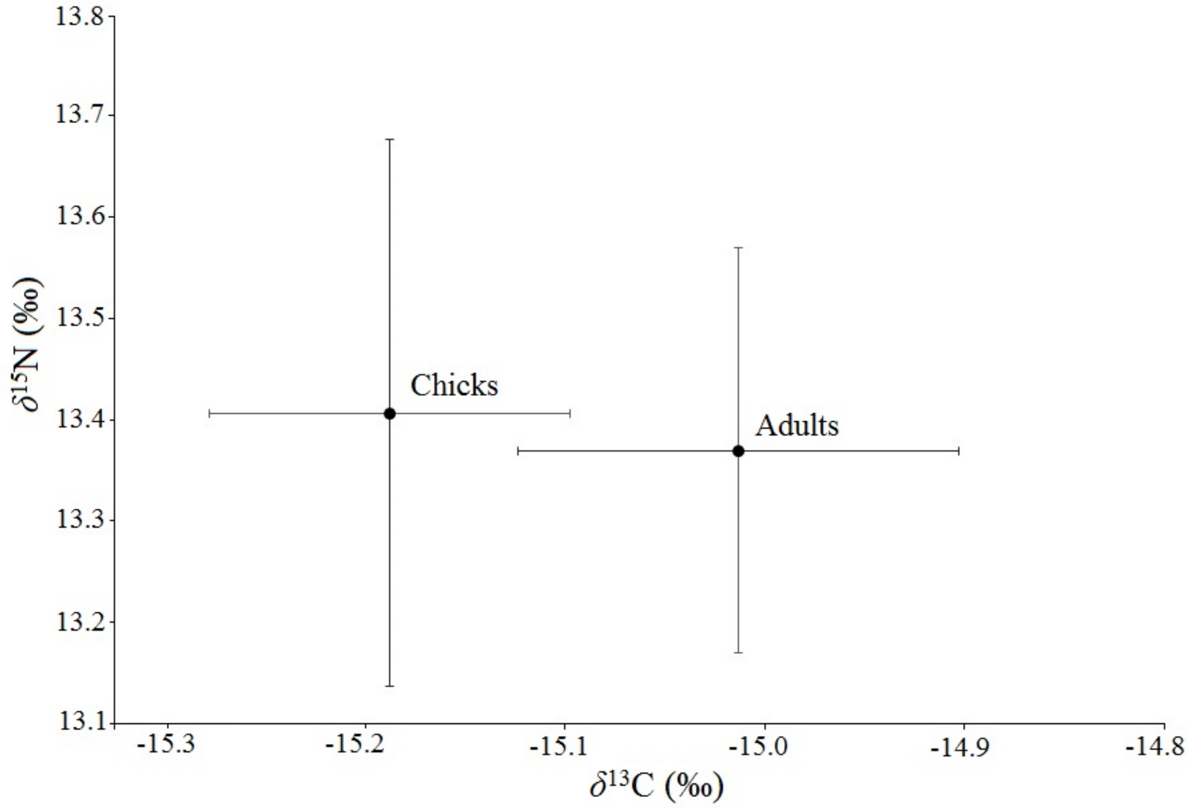
754 Figure 4


13.8
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δ^{15}N (‰)
Chicks
Adults
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-15.2
-15.1
-15.0
-14.9
-14.8
δ^{13}C (‰)

Figure 5

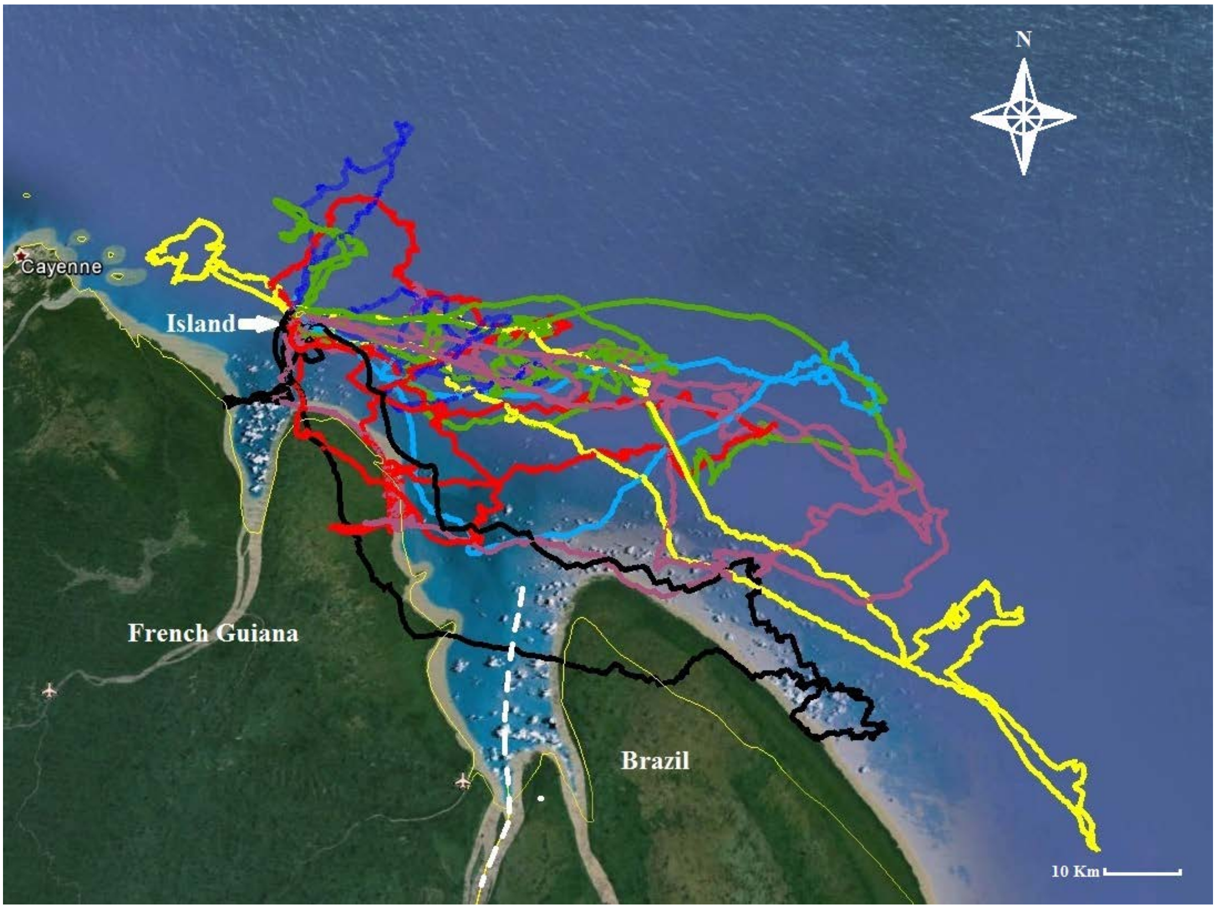

Figure 6

Tables

Table 1. Concentrations (µg g^{-1} dw.) of trace elements in red blood cells of adult and nestling Magnificent frigatebirds. df = detection frequency.

	Adults			**Nestlings**			***P***
	mean ± SD	median (range)	df (%)	mean ± SD	median (range)	df (%)	
Non-essential trace elements							
Ag	-	-	0	-	-	0	-
Cd	-	-	0	-	-	0	-
Hg	5.81 ± 1.27	5.62 (3.78 – 7.83)	100	0.99 ± 0.23	0.96 (0.68 – 1.68)	100	< **0.01**
Pb	0.02 ± 0.01	0.02 (0.02 – 0.04)	100	0.02 ± 0.005	0.02 (0.01 – 0.03)	100	< **0.01**
Essential trace elements							
As	2.35 ± 1.44	2.15 (0.58 – 7.33)	100	1.55 ± 0.67	1.51 (0.67 – 3.61)	100	**0.04**
Co	-	-	0	-	-	0	-
Cr	-	-	0	-	-	0	-
Cu	0.78 ± 0.07	0.80 (0.65 – 0.90)	100	0.74 ± 0.07	0.73 (0.60 – 0.86)	100	0.06
Fe	2413 ± 68	2411 (2235 – 2503)	100	2330 ± 80	2337 (2146– 2477)	100	< **0.01**
Mn	0.12 ± 0.03	0.11 (0.09 – 0.19)	100	0.21 ± 0.05	0.19 (0.13 – 0.19)	100	< **0.01**
Ni	-	-	0	-	-	0	-
Se	9.09 ± 1.91	8.74 (6.67 – 13.09)	100	5.75 ± 0.63	5.82 (4.57 – 6.57)	100	< **0.01**
Zn	19.44 ± 0.90	19.36 (18.29 – 22.08)	100	26.93 ± 2.95	26.80 (22.49 – 32.62)	100	< **0.01**
V	-	-	0	-	-	0	-

Table 2. POP concentrations in adults and nestlings of Magnificent frigatebirds for all congeners analysed. Concentrations are expressed as pg g^{-1} of wet weight. ND = not detected.

Congener	**Adults**		**Nestlings**		**P**
	median (range)	**mean ± SD**	**median (range)**	**mean ± SD**	
CB 28	ND	ND	ND	ND	ND
CB 52	ND	ND	ND	ND	ND
CB 49	ND	ND	ND	ND	ND
CB 74	ND	ND	ND	ND	ND
CB 101	ND	ND	ND	ND	ND
CB 99	<4 (<4 - 68)	7 ± 16	ND	ND	ND
CB 105	<2 (<2 - 29)	45 ± 6	ND	ND	ND
CB 118	22 (6 - 122)	29 ± 25	<1 (<1 - 8)	2.15 ± 1.88	<0.01
CB 128	<1 (<1 - 4)	1 ± 1	ND	ND	ND
CB 138	56 (26 - 277)	78 ± 60	6 (2 - 38)	7 ± 7	<0.01
CB 146	28 (<1 - 134)	35 ± 38	ND	ND	ND
CB 153	268 (114 - 869)	333 ± 211	17 (<1 - 84)	19 ± 16	<0.01
CB 156	7 (<1 - 21)	9 ± 5	ND	ND	ND
CB 170	47 (23 - 217)	68 ± 50	<3 (<1 - 13)	3 ± 3	<0.01
CB 171	<1 (<1 - 2)	1 ± <1	ND	ND	ND
CB 174	ND	ND	ND	ND	ND
CB 177	<1 (<1 - 4)	1 ± <1	ND	ND	ND
CB 180	165 (78 - 879)	240 ± 189	8 (3 - 40)	10 ± 8	<0.01
CB 183	32 (14 - 122)	42 ± 31	<1 (<1 - 11)	2 ± 2	<0.01
CB 187	34 (15 - 141)	43 ± 33	3 (<2 - 25)	4 ± 5	<0.01
CB 194	21 (7 - 154)	32 ± 32	<1 (<1 - 3)	1 ± <1	<0.01
CB 196/203	23 (7 - 108)	30 ± 25	<1 (<1 - 6)	1 ± 1	<0.01
CB 199	9 (<4 - 30)	11 ± 8	<1 (<1 - 3)	1 ± <1	<0.01
CB 206	<1 (<1 - 14)	<3 ± 3	ND	ND	ND
CB 209	ND	ND	ND	ND	ND
ΣPCBs	673 (336 - 2801)	967 ± 688	41 (19 - 232)	51 ± 44	<0.01
OxC	<1 (<1 - 7)	<2 ± <2	ND	ND	ND
TN	11 (5 - 16)	10 ± 3	<3 (<2 - 32)	4 ± 7	<0.01
CN	<1 (<1 - 5)	<2 ± 1	<1 (<1 - 5)	1 ± <1	0.10
ΣCHLs	14 (7 - 22)	14 ± 5	4 (3 - 37)	6 ± 7	<0.01
HCB	7 (2 - 41)	12 ± 11	11 (<2 - 33)	11 ± 6	0.38

***p,p’*-DDE**	220 (75 - 2342)	426 ± 561	25 (13 - 206)	40 ± 45	<0.01
***p,p’*-DDT**	ND	ND	ND	ND	ND
ΣDDTs	220 (75 - 2342)	426 ± 561	25 (13 - 206)	40 ± 45	<0.01
***β*-HCH**	2 (2 - 19)	8 ± 6	2 (2 - 11)	3 ± 2	0.38
***γ*-HCH**	2 (2 - 82)	8 ± 18	12 (2 - 20)	11 ± 7	0.01
ΣHCHs	14 (5 - 84)	16 ± 18	14 (5- 23)	14 ± 7	0.72
					ND
BDE 28	ND	ND	ND	ND	ND
BDE 47	ND	ND	ND	ND	ND
BDE 100	ND	ND	ND	ND	ND
BDE 99	ND	ND	ND	ND	ND
BDE 154	ND	ND	ND	ND	ND
BDE 153	ND	ND	ND	ND	ND
BDE 183	ND	ND	ND	ND	ND
ΣPBDEs	ND	ND	ND	ND	ND

SUPPORTING INFORMATION

High levels of mercury and low levels of persistent organic pollutants in a tropical seabird in French Guiana, the Magnificent frigatebird, *Fregata magnificens*.

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Table S1. Total Hg concentrations (mean ± SD) in the blood of seabirds worldwide. Data are mean ± SD of µg g^{-1} of dry weight or wet weight. Please note that this table is not exhaustive, but is a comparison of selected studies on seabirds, both in contaminated and non-contaminated areas, in order to have an idea of the amount of total Hg in frigatebirds.

Species	Location	Adults	Nestlings/Juv.	Weight	Reference
Fregata magnificens	French Guiana	5.81 ± 1.27	0.99 ± 0.23	dw	This study
Pagodroma nivea	Antarctica	2.70 ± 1.10		dw	Tartu et al. 2014
Diomedea exulans	(Indian Ocean)	10.70 ± 0.50		dw	Goutte et al. 2014b
Pachyptila desolata	Georgia	0.53 ± 0.21		dw	Anderson et al. 2009
Halobaena caerulea	Georgia	0.56 ± 0.28		dw	Anderson et al. 2009
Pelacanoides urinatrix	Georgia	0.31 ± 0.15		dw	Anderson et al. 2009
Pelacanoides georgicus	Georgia	0.41 ± 0.14		dw	Anderson et al. 2009
Procellaria aequinoctialis	Georgia	5.37 ± 1.18		dw	Anderson et al. 2009
Macronectes giganteus	Georgia	2.74 ± 1.05		dw	Anderson et al. 2009
Thalassarche melanophrys	Georgia	4.38 ± 1.1		dw	Anderson et al. 2009
Thalassarche chrysostoma	Georgia	6.57 ± 1.11		dw	Anderson et al. 2009
Macronectes halli	Georgia	3.93 ± 1.37		dw	Anderson et al. 2009
Diomedea exulans	Georgia	11.15 ± 3.38		dw	Anderson et al. 2009
Diomedea exulans	Georgia	9.57 ± 4.29	0.84 ± 0.36	dw	Tavares et al. 2013
Sterna hirundo	**USA**	**0.36 ± 0.40**	**0.04 ± 0.05**	**ww**	**Nisbet 2002**
Catharacta maccormicki	Antarctica	2.15 ± 0.17		dw	Goutte et al. 2014a
Catharacta lonnbergi	Antarctica	8.22 ± 0.24		dw	Goutte et al. 2014a
Ptychoramphus aleuticus	Canada	0.63 ± 0.07	0.23 ± 0.07	dw	Hipfner et al 2011
Cerorhinca monocerata	Canada	1.75 ± 0.11	0.41 ± 0.12	dw	Hipfner et al 2011
Rissa tridactyla	Norway	1.80		dw	Tartu et al. 2013
Oceanodroma leucorhoa	**USA**	**0.54 ± 0.37**	**0.03 ± 0.04**	**ww**	**Huntington et al. 1996**
Sterna hirundo	USA	0.44 ± 0.26		ww	Gochfeld 1980
Rissa tridactyla	Norway	2.33 ± 0.44		dw	Goutte et al. 2015
Diomedea exulans	(Indian Ocean)	7.70 ± 3.60		dw	Carravieri et al. 2014
Fratercula arctica	**(Middle Europe)**	**7.12 ± 2.58**		**dw**	**Fort et al. 2015**
Uria aalge	**(Middle Europe)**	**6.32 ± 5.17**		**dw**	**Fort et al. 2015**
Rissa tridactyla	**(Middle Europe)**	**8.58 ± 2.82**		**dw**	**Fort et al. 2015**
Alca torda	**(Middle Europe)**	**9.38 ± 4.12**		**dw**	**Fort et al. 2015**
Somateria mollissima	USA	0.05		ww	Bond and Diamond 2009
Sterna paradisaea	USA	0.16 ± 0.03		ww	Bond and Diamond 2009
Oceanodroma leucorhoa	USA	0.12 ± 0.02		ww	Bond and Diamond 2009
Fratercula arctica	USA	0.17 ± 0.03		ww	Bond and Diamond 2009
Sterna hirundo	USA	0.24 ± 0.04		ww	Bond and Diamond 2009
Uria aalge	USA	0.27 ± 0.05		ww	Bond and Diamond 2009
Alca torda	USA	0.36 ± 0.07		ww	Bond and Diamond 2009
Macronectes halli	**Georgia**	**2.9 ± 0.97**		**dw**	**Gonzales-Solis et al. 2002**
Macronectes giganteus	**Georgia**	**4.9 ± 5.1**		**dw**	**Gonzales-Solis et al. 2002**
Sterna forsteri	USA		0.33 ± 0.01	ww	Ackerman et al. 2008
Stercorarius skua	(Northern Europe)	3.67 ± 2.03	0.27 ± 0.16	dw	Bearhop et al. 2000
Stercorarius skua	Scotland	7.37 ± 3.1	1.12 ± 0.41	dw	Bearhop et al. 2000

Procellaria aequinoctialis	**Brazil**	**3.20 ± 3.67**		**dw**	**Carvalho et al. 2013**
Procellaria conspicillata	**Brazil**	**3.41 ± 2.14**		**dw**	**Carvalho et al. 2013**
Recurvirostra americana	USA	1.49 ± 0.13	2.02 ± 0.29	dw	Eagles-Smith et al. 2008
Himantopus mexicanus	USA	5.05 ± 0.46	0.99 ± 0.09	dw	Eagles-Smith et al. 2008
Hydroprogne caspia	USA	6.83 ± 0.89		dw	Eagles-Smith et al. 2008
Sterna forsteri	USA	7.06 ± 0.62	1.71 ± 0.18	dw	Eagles-Smith et al. 2008
Acaena adscendens	**(Indian Ocean)**	**0.67 ± 0.11**		**dw**	**Fromant et al. 2016**

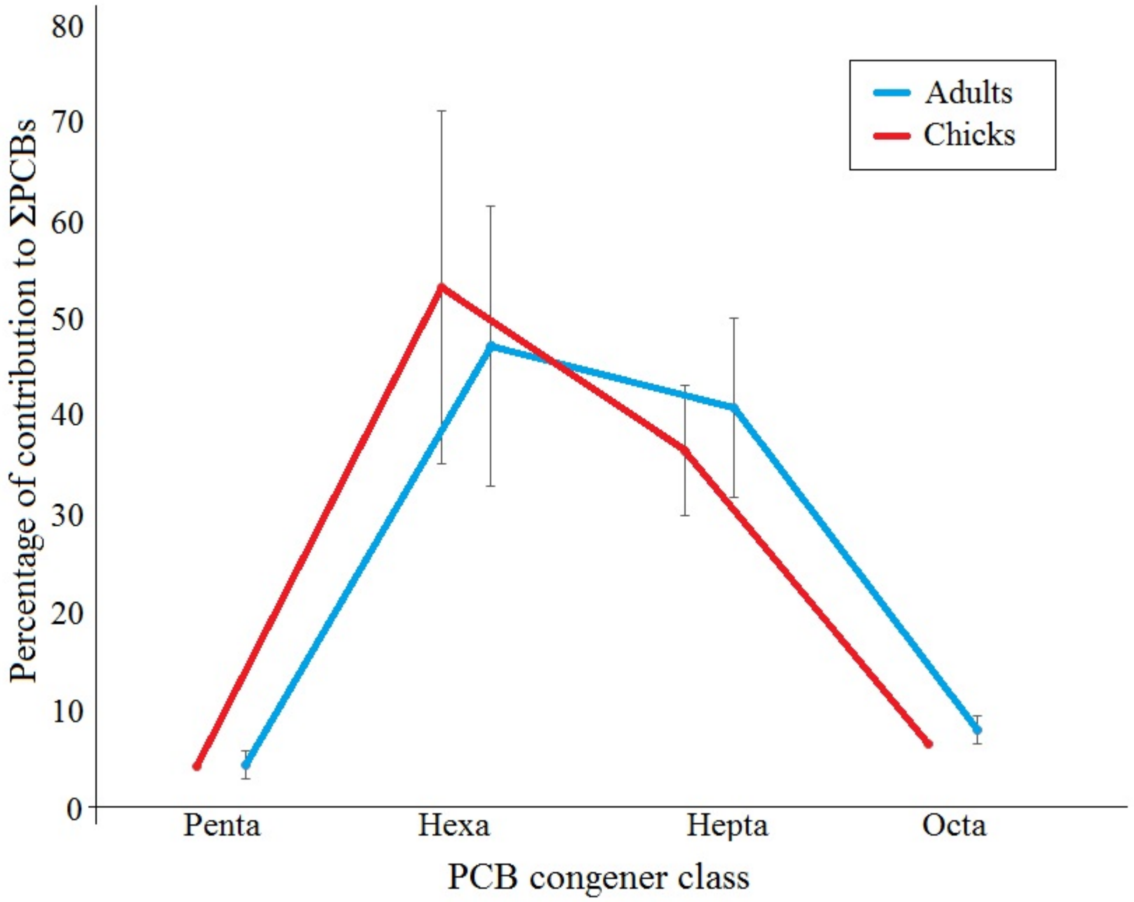

Figure S1. Contribution of congener classes to the ∑PCBs in adults (blue line) and nestlings (red line). Data are presented as mean ± SD.

References

Ackerman J.T., Eagles-Smith C.A., Takekawa J.Y. and Iverson S.A. 2008 Survival of postfledging Forster's terns in relation to mercury exposure in San Francisco Bay. Ecotoxicology 17, 789-801.

Anderson O., Phillips R., McDonald R., Shore R., McGill R. and Bearhop S. 2009 Influence of trophic position and foraging range on mercury levels within a seabird community. Marine Ecology Progress Series 375, 277–288 doi:10.3354/meps07784.

Bearhop S., Waldron S., Thompson D. and Furness R. 2000 Bioamplification of mercury in Great Skua *Catharacta skus* chicks: the influence of thropic status as determined by stable isotopes signatures of blood and feathers. Marine Pollution Bulletin 40, 181-185.

Bond A.L. and Diamond A.W. 2009 Mercury concentrations in seabird tissues from Machias Seal Island, New Brunswick, Canada. Sci. Total Environ. 407, 4330-4347.

Carvalho P.C., Bugoni L., McGill R.A. and Bianchini A. 2013 Metal and selenium concentrations in blood and feathers of petrels of the genus Procellaria. Environ. Toxicol. Chem. 32, 1641-1648.

Eagles-Smith C.A., Ackerman J.T., Adelsbach T.L., Takekawa J.Y., Miles A.K. and Keister R.A. 2008 Mercury correlations among six tissues for four waterbird species breeding in San Francisco Bay, California, USA. Environ. Toxicol. Chem. 27, 2136-2153.

Fort J., Lacoue-Labarthe T., Nguyen H.L., Boué A., Spitz J. and Bustamante P. 2015 Mercury in wintering seabirds, an aggravating factor to winter wrecks? Sci. Total Environ. 527-528, 448-454.

Fromant A., Carravieri A., Bustamante P., Labadie P., Budzinski H., Peluhet L., Churlaud C., Chastel O. And Cherel Y. 2016 Wide range of metallic and organic contaminants in various tissues of the Antarctic prion, a planktonophagous seabird from the Southern Ocean. Sci. Total Environ. 544, 754-764.

Gochfeld M. 1980 Tissue distribution of mercury in normal and abnormal young Common Terns. Marine Pollution Bulletin 11, 362-366.

Gonzales-Solis J., Sanpera C. and Ruix X. 2002 Metals and selenium as bioindicators of geographic and trophic segregation in giant petrels Macronectes spp. Mar. Ecol. Prog. Ser. 244, 257-264.

Goutte A., Bustamante P., Barbraud C., Delord K., Weimerskirch H. and Chastel O. 2014a Demographic responses to mercury exposure in two closely related Antartic top predators. Ecology 95, 1075-1086.

Goutte A., Barbraud C., Meillère A., Carravieri A., Bustamante P., Labadie P., Budzinsky H., Delord K., Cherel Y., Weimerskirch H. and Chastel O. 2014b Demographic consequences of heavy metals and persistent organic pollutants in a vulnerable long-lived bird, the wandering albatross. Proceeding of the Royal Society B 281, 20133313.

Goutte A., Barbraud C., Herzke D., Bustamante P., Angelier F., Tartu S., Clement-Chastel C., Moe B., Bech C., Gabrielsen G.W., Bustnes J.O. and Chastel O. 2015 Survival rate and breeding outputs in a high Artic seabird exposed to legacy persistent organic pollutants and mercury. Environ. Pollut. 200, 1-9.

Hipfner J.M., Hobson K.A. and Elliott J.E. 2011 Ecological factors differentially affect mercury levels in two species of sympatric marine birds of the North Pacific. Science of the Total Environment 409, 1328-1335.

Huntington C.E., Butler R.G. and Mauck R.A. 1996 Leach’s stormpetrel. In: The Birds of North America Online, Poole A (editor), Ithaca, NY: Cornell Laboratory of Ornithology; retrieved from The Birds of North America Online database.

Nisbet I.C.T. 2002 Common tern. In: The Birds of North America Online, Poole A (editor), Ithaca, NY: Cornell Laboratory of Ornithology; retrieved from The Birds of North America Online database.

Tartu S., Goutte A., Bustamante P., Angelier F., Moe B., Clément-Chastel C., Bech C., Gabrielsen G.W., Bustnes J.O. and Chastel O. 2013 To breed or not to breed: endocrine response to mercury contamination by an Arctic seabird. Biology Letters 9. doi:10.1098/rsbl.2013.0317.

Tartu S., Bustamante P., Goutte A., Cherel Y., Weimerskirch H., Bustnes J.O. and Chastel O. 2014 Age-related mercury contamination and relationship with luteinizing hormone in a long-lived Antartic bird. PLoS One 29, 9(7).

Tavares S., Xavier J.C., Pjillips R.A., Pereira M.E. and Pardal M.A. 2013 Influence of age, sex and breeding status on mercury accumulation patterns in the wandering albatross *Diomeda exulans*. Environmental Pollution 181, 315-320.