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Mercury contamination level is repeatable and predicted by wintering area in a long-distance migratory seabird☆

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ABSTRACT

The environmental presence of mercury has dramatically increased over the past century, leading to increased uptake, especially by top predators such as seabirds. Understanding the exact sources of contamination requires an individual-level approach, which is especially challenging for species that migrate. We took such an approach and located the wintering areas of 80 common terns (*Sterna hirundo*) through tracking, and, across years, collected feathers grown in those areas to assess their mercury levels using atomic absorption spectrometry. Although feathers of males and females did not differ in their mercury level, we found the average feather mercury level to be highest in birds wintering in the Canary Current (3.87 μg g^{-1}), medium in birds wintering in the Guinea Current (2.27 μg g^{-1}) and lowest in birds wintering in the Benguela Current (1.96 μg g^{-1}). Furthermore, we found considerable inter-annual fluctuations in feather mercury levels, a within-individual repeatability of 41%, that the mercury levels of 17% of feather samples exceeded the admitted toxicity threshold of 5 μg g^{-1}, and that the overall mean concentration of 3.4 μg g^{-1} exceeded that of other published reports for the species. Further studies therefore should assess whether these levels lead to individual-level carry-over effects on survival and reproductive performance.

1. Introduction

In addition to habitat destruction, overexploitation of natural resources and the introduction of invasive species, the release of pollutants into the environment affects wildlife on a global scale (Maxwell et al., 2016). As a non-degradable trace element, mercury (Hg) is a pollutant of particular concern. Although naturally occurring, most mercury present in the environment today is of anthropogenic origin (UN Environment, 2019). Global emission is increasing (UN Environment, 2019) and ongoing climate warming is predicted to exacerbate mercury levels, mainly due to the thawing of permafrost soils that contain substantial amounts of it (Schuster et al., 2018).

In sediments of marine ecosystems, bacterial action causes mercury to be methylated to its bioavailable and most toxic form, methylmercury (Obrist et al., 2018). Once in the food chain, methylmercury biomagnifies with each trophic level (Atwell et al., 1998), making organisms foraging at higher trophic positions particularly susceptible to detrimental effects on reproduction and physiology (Whitney and Cristol, 2017a). As a result, marine top predators have often been used as bioindicators for mercury pollution (e.g. Albert et al., 2019). As bioindicators for marine mercury pollution, seabirds have the advantage that they excrete parts of their ingested methylmercury via their feathers, potentially enabling relatively non-invasive sample collection (Monteiro and Furness, 1995). Feathers have been reported to contain 70–93% of birds' total mercury, making moult a major mercury excretion pathway (Whitney and Cristol, 2017b). Because the connection between blood and feather atrophies and is lost, mercury levels within the feather remain constant once feather development is completed (Appelquist et al., 1984). As such, feathers are an ideal tissue to assess levels of environmental pollutants in the area used for foraging during

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moult (e.g. Rutkowska et al., 2018).

In migrating seabirds, moult usually takes place in the wintering area (Bridge, 2006). So far, work on the wintering ecology of seabirds has shown that although the location of wintering areas seems mostly repeatable within individuals, birds from a single breeding population can use vastly different wintering areas (e.g. Kürten et al., 2022). If these wintering areas were to differ in their levels of mercury pollution, either consistently or among years, such consistent between-individual variation in wintering area could lead to within- and among-year variation in mercury contamination level of individual birds (e.g. Fleishman et al., 2019; Gatt et al., 2020; Albert et al., 2021), and therefore have wintering-area-specific carry-over effects on health, reproductive performance and/or survival (Fort et al., 2014; Lavoie et al., 2014). Whether variation in wintering area indeed results in variation in mercury pollution, however, is largely unknown (Fleishman et al., 2019; Albert et al., 2021).

Here, we therefore report on a 5-year study, in which we (i) collected 187 migratory tracks from 80 common terns (*Sterna hirundo*) breeding at a long-term study population and (ii) collected feathers grown at the wintering area to assess mercury concentrations of these birds. Our aim was to pinpoint sources of variation in mercury uptake, by (i) testing for effects of sex and wintering area on, as well as (ii) assessing annual variation in, and (iii) individual repeatability of, mercury levels.

2. Methods

2.1. Study species and data collection

We studied the mercury concentration in feathers moulted at the wintering area by sampling known-sex common terns breeding at a monospecific colony located at the Banter See in Wilhelmshaven, at the German North Sea coast (53°30′40″ N, 08°06′20″ E). Common terns are Holarctic colonially breeding and long-distance migratory seabirds (Becker and Ludwigs, 2004). They display high site fidelity to their breeding grounds, as well as to their wintering areas, which are scattered along the West-to South-African coast and encompass the Canary, Guinea and Benguela Currents (Kürten et al., 2022). Common terns are relatively easy to catch during incubation and large enough to be equipped with a small tracking device, without this device having detectable detrimental effects on behaviour, reproductive performance or survival (Kürten et al., 2019). Moreover, common terns occupy a high trophic position and excrete mercury into their feathers, in which it does not degrade (Appelquist et al., 1984).

Between mid-May and early July 2017–2021, we caught 31, 42, 41, 48 and 51 focal common terns, respectively, with an electronically released drop trap, on average 14 ± 4 SD days after the first egg was laid. The captured birds were tagged with a light-level geolocator (Intigeo-C65, Migrate Technology, UK) after the geolocator of the previous year (if they carried one) was removed. The geolocator was attached to the leg using a 10 mm aluminium ring. The total mass of the ring, glue and geolocator was 1.6 g, i.e. 1.2% ± 0.1 SD of the body mass of the birds at tagging and well below the recommended threshold of 3% (Kenward, 2001). Before birds were released, five to ten feathers were collected from their back, as these feathers belong to the first ones grown at the onset of prenuptial moult just prior to spring migration (Dwight, 1901). Birds were released after an average handling time of 6 ± 3 SD minutes and all resumed incubation.

We set our geolocators to sample ambient light intensity every minute, with the maximum light intensity being stored every 5 min (mode 10, Migrate Technology). Stored light intensity data were analysed using the package "FLightR" (version 0.5.0, Rakhimberdiev et al., 2017) in R (version 4.0.3, R Core Team 2020) to extract the mean ± SD longitude and latitude of each wintering area (for details and R code, see Kürten et al., 2022). Based on the latitude, we assigned individuals to winter at either the (i) Canary Current (latitude >10°), (ii) Guinea Current (latitude 10-2°), or (iii) Benguela Current (latitude < -20°). Two individuals used wintering areas in two different currents, such that we used their location during moult (February) to assign them to their wintering current. Moreover, for two individuals, the detected wintering areas were located at the "border" of the Canary and Guinea Current, such that we prepared two datasets in which these individuals were assigned to one of the two currents. We present the results obtained from the analysis of the dataset in which the birds were assigned to winter at the Canary Current in the main text, those of the other analysis in Table S1. Finally, since repeatability analyses showed the birds to be highly site faithful to their wintering areas (Kürten et al., 2022), feather samples and tracking data were paired whether or not they were (n = 187) or were not (n = 26) collected in the same year.

Feather samples were alternately washed three times with distilled water and acetone. After each wash, they were cleaned in an ultrasonic bath (Sonorex Super RK510H, Bandelin, Germany) for 15 min. Mercury analysis was performed using a Direct Mercury Analyzer (DMA-80, MLS Mikrowellen-Labor-Systeme GmbH, Germany), an established tool for mercury quantification (e.g. Brasso et al., 2015) with a detection limit of 0.005 ng. To quantify total mercury concentrations, 1–2 feathers were weighed, wrapped in mercury-free aluminium foil and introduced into a nickel sample boat. Each sample was dried and thermally decomposed in a stream of oxygen at 750 °C (240 s) and catalytically converted (60 s). Released by the high temperature, gaseous mercury was trapped by gold amalgamation at 850 °C (12 s) and its level determined using atomic absorption spectrometry at 253.7 nm (30 s).

Mercury concentrations were quantified three times per sample in order to evaluate the sample repeatability (see below), which will reflect a combination of both the accuracy of the measurement and any biological variation among individually grown feathers.

2.2. Statistical analyses

Our dataset comprised 3 repeated measurements of 213 feather samples from 80 individuals - 39 males and 41 females - of which 54, 23 and 3 individuals overwintered at the Canary, Guinea and Benguela Current, respectively. The data were log-transformed to obtain a normal distribution (see Fig. S1).

The individual repeatability of mercury concentrations within years was calculated using a linear mixed model with year as a fixed effect and individual identity and sample ID as random intercepts, using the "rpt" function of the R package "rptR" (Stoffel et al., 2017). This repeatability was 0.75 ± 0.03 SE ($p < 0.001$). We then took the average of the three measurements within each year, and ran a second repeatability analysis, similar to the one described above, but excluding the random intercept of sample ID to calculate the individual repeatability of mercury concentrations among years.

To test whether mercury concentrations differed between birds wintering in the three currents, we ran a linear mixed model with average mercury concentration as a dependent variable. This model included individual identity as a random intercept to account for the non-independence of repeated observations on the same bird across years. As fixed effects, we included *wintering area* as a 3-level, *sex* as a 2-level, and *year* as a 5-level class variable. We also included all two-way interactions between our three fixed effects, then used a stepwise backwards elimination procedure to obtain a minimally adequate model. The model was run with the "lmer" function of the R package "lme4" (Bates et al., 2015). P-values were obtained using the "lmerTest" R package (Kuznetsova et al., 2017), the level of significance was set to $p < 0.05$.

Data were visualised using the R package "ggplot2" (Wickham, 2016) and the program QGIS (QGIS Development Team, 2021) by making use of the "heatmap" function. Mercury concentrations are presented as means ± standard error (SE).

Table 1
Results from the final linear mixed model testing whether (log-transformed) average feather mercury levels of common terns differ between ocean currents used for wintering (Canary Current as a reference), years (2017 as a reference) and the sexes (males as a reference). Significant effects (i.e. $p < 0.05$) are presented in bold.

	Estimate ±SE	*t*	*p*
Intercept	7.932 ± 0.079	100.683	**< 0.001**
Guinea Current	−0.536 ± 0.076	−7.043	**< 0.001**
Benguela Current	−0.883 ± 0.159	−5.555	**< 0.001**
Year (2018)	0.363 ± 0.086	4.223	**< 0.001**
Year (2019)	0.082 ± 0.087	0.953	0.342
Year (2020)	0.197 ± 0.086	2.300	0.023
Year (2021)	0.370 ± 0.086	4.282	**< 0.001**
Sex (female)	0.066 ± 0.066	1.008	0.317

3. Results and discussion

Mercury concentrations in feathers across all 213 samples collected from 80 individual common terns ranged between 0.77 and 9.87 μg g^{-1}, with a mean of 3.4 (±0.11) μg g^{-1}. These concentrations varied with the wintering area of the birds (Table 1): individuals wintering in the Canary Current had significantly higher mercury concentrations (3.87 ± 0.13 μg g^{-1}, n = 153 samples of 54 individuals) than those wintering in the Guinea Current (2.27 ± 0.14 μg g^{-1}, n = 50 samples of 23 individuals), which in turn had significantly higher mercury concentrations than those wintering in the Benguela Current (1.96 ± 0.46 μg g^{-1}, n = 10 samples of 3 individuals; Fig. 1). Such spatial variation was reported in other seabirds as well (e.g. Lavoie et al., 2015; Watanuki et al., 2016; Fleishman et al., 2019; Gatt et al., 2020; Albert et al., 2021) and may reflect spatial variation in contamination levels of wintering areas (e.g. Brasso et al., 2015; Fleishman et al., 2019; Dodino et al., 2022). Alternatively or additionally, however, the spatial variation in mercury concentrations in feathers may be caused by (i) individuals occupying different trophic positions at the different wintering areas (e.g. Gatt et al., 2020) and/or (ii) selective disappearance of the most contaminated individuals migrating to the Guinea or Benguela Current. The latter would be a potential explanation not only for the decreasing mercury levels with increasing distance from the breeding area, but also for the small number of common terns found to migrate to these distant wintering areas. Further studies, for example employing stable isotope analysis to assess the trophic position of individual birds, or assessing mercury levels of water and prey species in the different wintering areas, could shed light on the matter.

We also found major fluctuations in feather mercury concentrations among years (Table 1): annual concentrations were higher in 2018, 2020 and 2021 (3.96 ± 0.26, 3.33 ± 0.22 and 3.81 ± 0.25 μg g^{-1}, n = 42, 48 and 51 samples, respectively) than in 2017 and 2019 (2.61 ± 0.20 and 3.00 ± 0.21 μg g^{-1}, n = 31 and 41 samples, respectively; Fig. 2a). Such fluctuations may reflect climatic variation affecting environmental mercury levels (Foster et al., 2019), since mercury occurrence in the ocean has, for example, been found to correlate with sea surface temperature (Schartup et al., 2019).

Feather mercury concentrations did not differ between male (3.40 ± 0.15 μg g^{-1}, n = 108 samples) and female (3.40 ± 0.17 μg g^{-1}, n = 105 samples; Table 1 and Fig. 2b) common terns. This fits well with findings of males and females not differing in their wintering areas (Kürten et al., 2022), and suggests that they also may not differ in their prey selection at the wintering areas, although very little is known about the sex-specificity of foraging behaviour and prey assortment of common terns wintering in West and South Africa. It also fits well with findings from other studies assessing mercury concentrations in feathers of various seabird species, which mostly did not find differences between male and female birds either (e.g. Brasso et al., 2015; Fleishman et al., 2019). The few studies that did find sex-specific feather mercury concentrations in seabirds attributed this either to sex-specific foraging areas and trophic positions (Mills et al., 2020) or to a long-lasting effect of mercury reduction via egg laying in females (Ramos et al., 2009).

Taking into account the annual variation in mercury concentration,

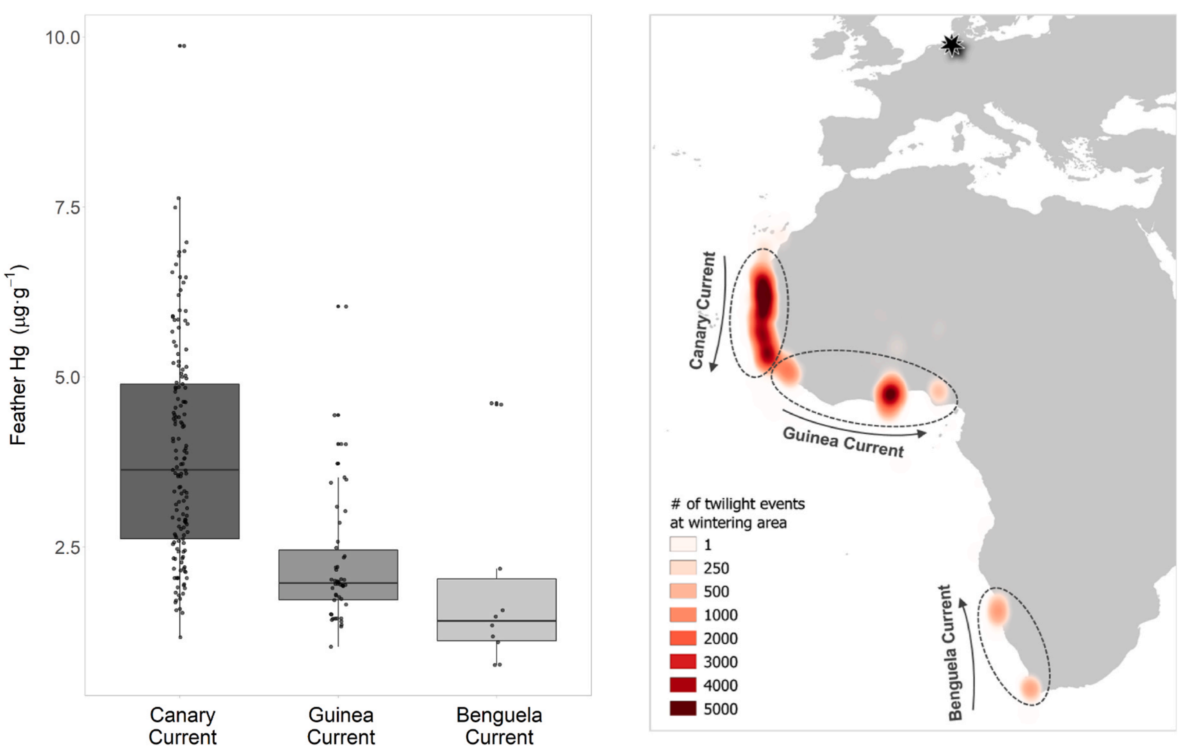

Fig. 1. Mean (±SE) feather mercury concentrations of 80 common terns wintering in the Canary, Guinea or Benguela Current. Wintering areas are based on the individuals' locations at daily twilights from one week after the estimated arrival at, up to one week prior to the estimated departure from, the wintering area. Heatmaps were produced in QGIS using quartic kernel density with a 2° radius. The star marks the breeding colony.

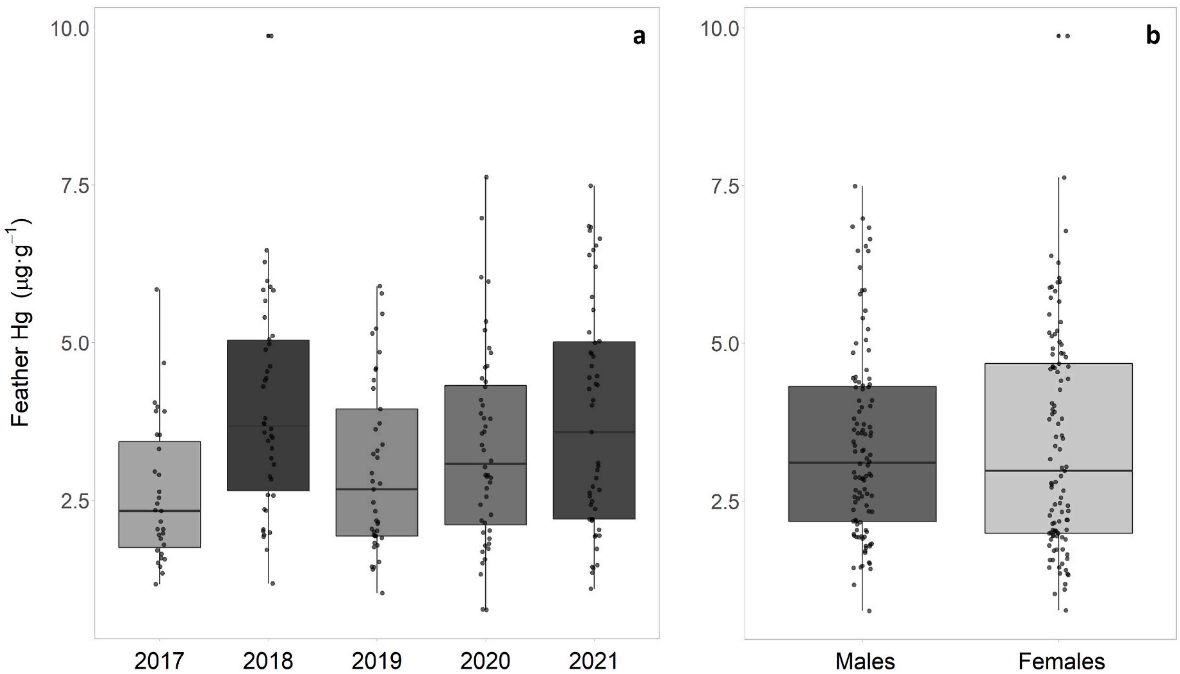

Fig. 2. Mean (±SE) feather mercury concentrations of common terns across years (a) and in relation to sex (b).

we found the individual repeatability of feather mercury concentration across years, which has rarely been assessed, to equal 41% ± 8 SE ($p < 0.001$). This was substantially lower than the within-year repeatability of 75% ± 3 SE (see Methods), which itself shows that mercury measurement in our feather samples is rather reliable and only to 25% subject to a combination of measurement error and biology (e.g. sequence of moult and thus mercury deposition in feathers). Since the repeatability of the wintering area itself is extremely high (Kürten et al., 2022), the 'missing' repeatability of 75–41 = 34% may reflect within-individual variation in diet composition or trophic level among years.

Overall, at 3.4 μg g^{-1}, the mean mercury concentration we detected in our common tern feather samples is below the admitted toxicity threshold of 5 μg g^{-1}, above which reproductive disorders have been reported for birds (Eisler, 1987). Nevertheless, 36 of our samples (c. 17%) did exceed this threshold, which, together with the fact that the mean feather mercury concentration we observed also exceeds that observed in previous studies of other common tern populations (Burger et al., 1994; Bracey et al., 2020) and several other bird species (Albert et al., 2019), does raise concern. Studies examining the suitability of the threshold, however, are scarce and in little auks (*Alle alle*), feather mercury levels have, for example, been found to be negatively associated with egg size even below the threshold (Fort et al., 2014). Further studies therefore are needed to determine whether mercury uptake during the non-breeding period affects survival, and/or interacts with mercury uptake at the breeding site and has carry-over effects on reproductive performance (Fort et al., 2014; Lavoie et al., 2014).

Ethics approval and consent to participate

The study was performed under licenses of the city of Wilhelmshaven and the Lower Saxony State Office for Consumer Protection and Food Safety, Germany.

CRediT author statement

Justine Bertram: Investigation, Writing - Original Draft, Visualization, Formal analysis. **Nathalie Kürten**: Investigation, Visualization, Writing - Review & Editing, Funding acquisition, Formal analysis. **Coraline Bichet**: Writing - Review & Editing, Formal analysis. **Peter J. Schupp**: Resources. **Sandra Bouwhuis**: Conceptualization, Project administration, Supervision, Writing - Review & Editing.

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envpol.2022.120107.

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