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Mercury in wintering seabirds, an aggravating factor to winter wrecks?

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HIGHLIGHTS

- We investigated the role played by Hg in an unprecedented seabird winter wreck.
- Stranded seabirds show the highest Hg concentrations measured in these species.
- Hg is a major aggravating stress factor contributing to enhanced seabird winter mortality.
- Total blood can be used as a predictor of Hg contamination in other seabird tissues.

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ABSTRACT

Every year, thousands of seabirds are cast ashore and are found dead along the coasts of North America and Western Europe. These massive mortality events called ‘winter wrecks’ have generally been attributed to harsh climatic conditions and prolonged storms which affect bird energy balance and impact their body condition. Nevertheless, additional stress factors, such as contaminant body burden, could potentially cumulate to energy constraints and actively contribute to winter wrecks. However, the role played by these additional factors in seabird massive winter mortality has received little attention to date. In February/March 2014, an unprecedented seabird wreck occurred along the Atlantic French coasts during which >43,000 seabirds were found dead. By analyzing mercury (Hg) concentrations in various tissues collected on stranded birds, we tested the hypothesis that Hg played a significant role in this mortality. More specifically, we aimed to (1) describe Hg contamination in wintering seabirds found along the French coasts in 2014, and (2) determine if Hg concentrations measured in some vital organs such as kidney and brain reached toxicity thresholds that could have led to deleterious effects and to an enhanced mortality. We found some of the highest Hg levels ever reported in Atlantic puffins, common guillemots, razorbills and kittiwakes. Measured concentrations ranged from 0.8 to 3.6 $\mu g \cdot g^{-1}$ of dry weight in brain, 1.3 to 7.2 $\mu g \cdot g^{-1}$ in muscle, 2.5 to 13.5 $\mu g \cdot g^{-1}$ in kidney, 2.9 to 18.6 $\mu g \cdot g^{-1}$ in blood and from 3.1 to 19.5 $\mu g \cdot g^{-1}$ in liver. Hg concentrations in liver and brain were generally below the estimated acute toxicity levels. However, kidney concentrations were not different than those measured in the liver, and above levels associated to renal sub-lethal effects, suggesting a potential Hg poisoning. We concluded that although Hg was not directly responsible for the high observed mortality, it has been a major aggravating stress factor for emaciated birds already on the edge. Importantly, this study also demonstrated that total blood, which can be non-lethally collected in seabirds, can be used as a predictor of Hg contamination in other tissues.

1. Introduction

Every year, thousands of seabirds cast ashore and are found dead along the coasts of North America and Western Europe (e.g., Piatt and Van Pelt, 1997; Gaston, 2004; Harris and Wanless, 2013). These massive mortality events called ‘winter wrecks’ have generally been attributed to harsh climatic conditions and prolonged storms which affect birds foraging efficiency and enhance thermoregulatory costs, thereby affecting their energy balance, impacting their body conditions and ultimately increasing their mortality (Fort et al., 2009). Hence, alcids which have little energy reserves and cannot survive longer than 3–4 days without foraging (Gaston, 1983; Gaston and Jones, 1998) are the first affected by starvation and the most exposed to harsh conditions. However, other

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additional factors could cumulate to bird energy constraints and actively contribute to winter wrecks. For instance parasite load or diseases could affect individual immune system and therefore enhance their sensitivity to energy constraints (Bulté et al., 2012). Contaminant body burden could also be a factor playing a significant role in these massive mortality events. Indeed, through an acute toxicity (see Wolfe et al., 1998), by affecting bird physiology and behavior (Tan et al., 2009) or by increasing energy requirements to sustain detoxification mechanisms (Lucia et al., 2012), contaminants could directly or indirectly impact their survival. However, very little is known about the role played by these additional factors in seabird winter wrecks (Debacker et al., 2000, 2001a). A better understanding of cumulative effects would benefit to the conservation of these vulnerable species during a critical period of their life-cycle.

Among contaminants present in the marine environment which could affect seabird mortality, the study of mercury (Hg) is of particular interest. Hg, and most particularly methyl-Hg, is indeed a powerful neurotoxicant in marine top-predators (e.g., Wolfe et al., 1998; Dietz et al., 2013) which can, through brain lesions or endocrine disruptive effects, affect seabird behavior and ultimately shape their survival (Wolfe et al., 1998; Tartu et al., 2013; Goutte et al., 2014a, 2014b). High Hg concentrations in other tissues (e.g., kidney and liver) could as well cause acute poisoning and impact seabird survival (Wolfe et al., 1998). Elevated concentrations of Hg were previously found in stranded wintering seabirds from the Northern Hemisphere (Debacker et al., 1997; Joiris et al., 1997). Hence, some authors suggested that Hg could be an additional stress factor that could partly result in the death of birds (Debacker et al., 2000).

In February and March 2014, an unprecedented seabird wreck occurred along the Atlantic French coasts. Within 6 weeks, a total of >43,000 seabirds had been found dead or extremely weakened on beaches (Farque, 2014). Atlantic puffins (*Fratercula arctica*) and common guillemots (*Uria aalge*), two species of the alcid family, were the most affected, representing 66% and 27% of stranded birds, respectively (Farque, 2014). Almost all birds were emaciated; starvation was designated as the main cause of this massive mortality. However, as dozens of birds of several species were collected on beaches or kept once dead in wildlife centers, they represented a unique opportunity to investigate further Hg contamination in wintering seabirds, and its potential role in this winter wreck.

By analyzing various tissues collected on stranded birds belonging to four different species, and by comparing Hg concentrations with those measured in healthy birds, the present study had the following objectives: (1) describe Hg contamination in stranded seabirds wintering in the Bay of Biscay; (2) test the hypothesis that Hg concentrations measured in some vital organs such as kidney and brain reached toxicity levels that could have led to deleterious effects and to enhanced bird mortality; and (3) determine if whole blood samples could be used as a predictor of Hg contamination in other seabird tissues. Indeed, while blood can be easily and non-lethally collected in seabirds, measurements of contaminant levels in other tissues are, for ethical issues, more limited. Understanding if/how blood can be used as surrogate to evaluate the contamination of other tissues is therefore essential.

2. Materials and methods

2.1. Seabird winter wreck 2014

For this study, 43 seabird carcasses were collected on the Isle of Rhé (46°1′N 1°2′W) and on Oléron Island (45.9°N 1.3°W) during February and March 2014, and kept frozen at −20 °C until analyses. They belonged to four species: Atlantic puffin (n = 15), common guillemot (n = 13), razorbill (*Alca torda*; n = 7) and black-legged kittiwake (*Rissa tridactyla*; n = 8). Part of these birds was collected freshly dead on beaches (n = 15) while another part (n = 28) was found alive by walkers and brought to a Wildlife Care Center where they died within 24 h. We assumed that these few hours spent at the Care Center did not affect Hg concentrations in bird tissues. During dissections, sex was defined by visual inspection of gonads and age of all individuals was determined following plumage patterns or morphological criteria (Pyle, 2009). In the present study, only adult birds were analyzed. The entire liver, kidney and brain were collected as well as some pectoral muscle and whole blood (hereafter 'blood') samples (from the cardiac clot). All tissues were lyophilized for 48 h, ground into powder and homogenized prior to Hg analyses. Feathers were not analyzed in this study since they reflected a different period than internal organs and were therefore not directly comparable. Hg that has accumulated in body tissues is excreted to feathers when birds are molting and Hg concentrations in feathers are thus considered to be an indicator of contamination between two molting sequences rather than a shorter-term contamination as in other tissues (Furness et al., 1986; Agusa et al., 2005).

Body condition of birds was evaluated through (1) the thickness of their pectoral muscle (Lindström et al., 2000; Moseley et al., 2012) and (2) the calculation of a body condition index (ratio of liver to kidney mass; Wenzel and Adelung, 1996; Debacker et al., 2000). Thickness of pectoral muscles was determined using a needle pricked transversally (at a 90° angle) in the right-side muscle, along the keel of the sternum (1 cm from the keel and at mid-length of the keel) (Erbacher, 2012).

2.2. Wintering seabirds in standard, healthy, body condition

In April 2006, 21 razorbills were found freshly dead on the beach in the southern part of the Bay of Biscay (43°03′N, 01°03′W). Contrary to razorbills collected in 2014 that were highly emaciated (see Results), these birds presented external signs of probable by-catch suggesting that they had drowned in fishing gears. We therefore assumed these birds to be in standard, healthy, body conditions at the time of death, and hereafter, they are referred to as 'healthy birds'. Razorbills collected in 2006 were used to compare body conditions as well as the mass of each organ and their Hg contamination levels between stranded (2014) and healthy (2006) razorbills.

2.3. Mercury analyses

Total Hg (hereafter termed Hg) concentrations were measured in each tissue at the Littoral Environnement et Sociétés laboratory (LIENSs, La Rochelle, France) using an Advanced Mercury Analyzer spectrophotometer (Altec AMA 254) as described in Bustamante et al. (2006). Analyses were repeated two to four times for each sample until the relative standard deviation for two samples was <10%. The mean Hg concentrations for those two measurements were then considered for statistical analyses. To ensure the accuracy of measurements, a certified reference material was used (Lobster Hepatopancreas Tort-2; NRC, Canada; Hg concentration of 0.27 ± 0.06 $\mu g \cdot g^{-1}$ of dry weight (dw)) and measured every 10 samples. The average measured value was 0.28 ± 0.02 $\mu g \cdot g^{-1}$ of dw (n = 105). Additionally, blanks were run at the beginning of each sample set. The detection limit of the method was 0.005 $\mu g \cdot g^{-1}$ of dw.

2.4. Statistical analyses

Statistics were computed using R version 3.0.2 (R Development Core Team 2011). Hg data were log-transformed to comply with parametric assumptions of normality and homoscedasticity. Differences in Hg concentrations between tissues were tested for each species using repeated measures ANOVA followed by paired *t*-test. When sphericity was not met, we used the Greenhouse–Geisser adjustment, by multiplying the numerator and denominator degrees of freedom by ε, to calculate adjusted p-values (Greenhouse and Geisser, 1959). Differences in morphometrics, Hg concentrations and total Hg content in each organ between stranded (2014) and healthy (2006) razorbills were tested using Student's *t* tests. Relationships of Hg concentrations between blood and other tissues were calculated for both emaciated (2014) and healthy (2006) birds. ANCOVA were used to test for a potential effect of bird state on the