



Foraging ecology drives mercury contamination in chick gulls from the English Channel

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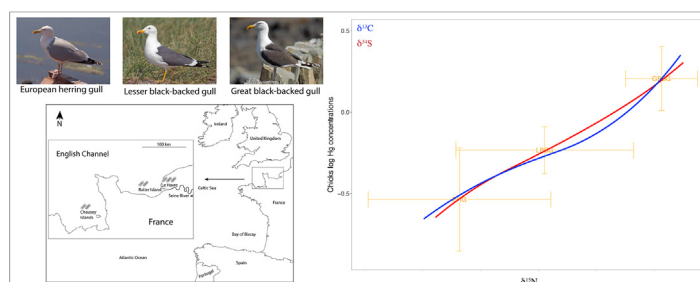
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HIGHLIGHTS

- Wide variations in Hg concentrations were found between the different species of gull.
- Hg concentrations were the highest in Great black-backed gulls.
- Hg exposure was different among the colonies of Great black-backed gull.
- Regardless of the breeding site, $\delta^{15}\text{N}$ explained Hg concentrations.
- Feeding habitat (proxied by $\delta^{34}\text{S}$) has a major influence on Hg concentrations.

GRAPHICAL ABSTRACT



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ABSTRACT

Although mercury (Hg) occurs naturally, human activity is currently the greatest source of release and the ocean receives Hg inputs by rivers and atmospheric deposition. Seabirds including chicks serve as valuable bioindicators of Hg contamination, reflecting local contamination around the colony. This study investigates the ecological drivers (trophic position and foraging habitat) influencing Hg concentrations in blood and feathers of chicks of three sympatric marine gull species. Chicks were sampled between 2015 and 2017 in the Seine Estuary, one of the most Hg contaminated rivers in Europe, and in the Normand-Breton Gulf (the Chausey Islands), 200 km west, as a reference site with limited contaminant inputs. The trophic status of the chicks was evaluated based on the relative abundance of stable isotopes ($\delta^{13}\text{C}$, $\delta^{15}\text{N}$ and $\delta^{34}\text{S}$). There was a tight correlation between Hg concentrations, as well as the abundance of stable isotopes, in blood and feathers. Great black-backed gull had the highest blood Hg concentrations of the species ($1.80 \pm 0.92 \mu\text{g}\cdot\text{g}^{-1}$ dry weight (dw)); the Lesser black-backed gull had intermediate concentrations ($0.61 \pm 0.18 \mu\text{g}\cdot\text{g}^{-1}$ dw); and the European herring gull had the lowest ($0.37 \pm 0.26 \mu\text{g}\cdot\text{g}^{-1}$ dw). Individuals with the highest trophic position showed consistently the highest Hg concentrations. The positive relationship between Hg concentrations and the feeding habitat (marine vs terrestrial) indicated that the main source of Hg for gulls in the English Channel is marine prey. This exposure led to relatively high Hg concentrations in Great black-backed gull, which may produce toxic effects to individuals with potential consequences for their populations.

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1. Introduction

Inorganic mercury (iHg), released by volcanism and soil erosion, occurs naturally in marine and coastal ecosystems (Chen et al., 2018). However, Hg is also released in large amounts by anthropogenic activities, the main sources being industry, the burning of fossil fuels and evaporation by artisanal gold mining (UNEP 2013; Eagles-Smith et al., 2018). Micro-organisms such as sulfato-reducing bacteria have the potential to transform iHg into methyl-mercury (MeHg) which is highly bioavailable for biota (Compeau and Barta, 1985). MeHg is also one of the most toxic metallic compounds (Wolfe et al., 1998), exerting deleterious effects, such as endocrine disruption and neurodevelopmental impairment in species at the top of the food webs (e.g., Wolfe et al., 1998; Dietz et al., 2013; Tartu et al., 2013). Unlike inorganic forms, MeHg efficiently biomagnifies along the food chains and bio-accumulates in organisms as a result of its high bioavailability and retention in biota (Dijkstra et al., 2013; Harding et al., 2018; Riisgård and Hansen, 1990). For these reasons, Hg contamination is a priority issue on a global scale for the United Nations, which adopted the Minamata Convention on Hg to protect human health and the environment from anthropogenic emissions (Gustin et al., 2016).

Seabirds are meso and top predators and therefore highly exposed to Hg due to bioaccumulation (concentrations increase over time in the body) and biomagnification (concentrations increase at each trophic level) processes. They are consequently good bioindicators of Hg contamination of marine ecosystems, and more particularly of their feeding habitats (e.g. coastal, pelagic, benthic) which may vary among the species feeding there (Furness and Camphuysen, 1997; Ochoa-Acuña et al., 2002; Carravieri et al., 2016). Investigating seabird Hg contamination is vital both for their conservation policies (Goutte et al., 2014a, 2014b) and for the monitoring of Hg contamination in the marine environment (UN Environment 2019). Since it allows non-lethal sampling, researchers often use blood and feathers to determine seabird exposure to Hg, although concentrations measured in these tissues depend on a variety of factors, such as the trophic position, the foraging range and the feeding habitat, and also the migration patterns (Carravieri et al., 2014; Cherel et al., 2018; Albert et al., 2019). This variability of factors means that it is the blood and feathers of chicks that reflect local contamination of the food webs more clearly than adults', since they are fed by their parents on prey caught in the vicinity of the nesting site (Blévin et al., 2013; Carravieri et al., 2020). These tissues also allow us to access to ecological information regarding birds' diets (i.e. the trophic position reflected by $\delta^{15}\text{N}$) and the origin of their prey (i.e. the feeding habitat reflected by both $\delta^{13}\text{C}$ and $\delta^{34}\text{S}$) (Cherel et al., 2018; Góngora et al., 2018).

The French coasts of the English Channel are significantly affected by Hg contamination, in particular from the Seine River (Cossa et al., 2003; Meybeck et al., 2007). In this area, Hg contamination has been documented for organisms of low trophic levels (e.g. reaching $0.35 \mu\text{g}\cdot\text{g}^{-1}$ dw in shrimps; Cossa et al., 2003) and for marine mammals (e.g. reaching $55 \mu\text{g}\cdot\text{g}^{-1}$ dw in liver of harbour porpoises; Lahaye et al., 2007), but to the best of our knowledge, no data are available for seabirds in the literature. In that context, the main aim of this study was to investigate Hg concentrations in the blood and feathers of the chicks of different sympatric seabird species from sites influenced at different degrees by Hg and human activity. To this end, we focused on three gull species: the European herring gull (*Larus argentatus*), the Lesser black-backed gull (*L. fuscus graellsii*) and the Great black-backed

gull (*L. marinus*), all breeding along the French coast of the English Channel: in the Seine Estuary (two breeding sites influenced by high Hg levels from the Seine River and potential access to anthropogenic food) and in the Normand-Breton gulf (one breeding site uninfluenced by specific, known Hg sources and intense human activity). We checked the hypotheses that Hg contamination in birds depends on 1) the location of the colony, 2) the trophic position of the species and 3) the feeding habitat (marine vs terrestrial). We predicted that Hg concentrations are influenced by the trophic position of individuals (using $\delta^{15}\text{N}$ as a proxy) both at the interspecific and intraspecific levels, as a result of Hg biomagnification in food webs. We also predicted that Hg concentrations are related to the feeding habitat (using $\delta^{13}\text{C}$ and $\delta^{34}\text{S}$ as its proxies) with individuals feeding on marine prey having higher exposure as a result of higher methylation rates and longer food chains than in terrestrial systems (Lavoie et al., 2013).

2. Materials and methods

2.1. Sampling sites and sample collection

Fieldwork was carried out in three areas along the coast of the English Channel (Fig. 1). Two sampling sites were located in the Seine Estuary (highly urbanized and polluted area), approximately 8 km apart: the city of Le Havre ($\text{N}49^{\circ}29'$, $\text{E}0^{\circ}12'$), where nests were distributed across the city on the roofs of buildings, and the Ratier Island ($\text{N}49^{\circ}25'46''$, $\text{E}0^{\circ}08'7''$), where nests were built on the ground of an island located at the heart of the estuary. The third site was situated 200 km west of the Seine Estuary in the Normand-Breton Gulf and is at a good distance from industrial activity: the Chausey Islands ($\text{N}48^{\circ}53'$, $\text{W}1^{\circ}49'$), where nests were built on the ground on different islands).

Sampling occurred in the last week of June and first week of July of three consecutive years beginning in 2015 (Table 1). A total of 174 chicks (approximately two months old) of three species of gulls were captured. Herring gull (hereafter EHG) and Great black-backed gull (hereafter GBBG) were sampled in both the Seine Estuary and the Chausey Islands while Lesser black-backed gulls (hereafter LBBG) were sampled only in the City of Le Havre. These sympatric species differ significantly in their foraging ecology. Although they are all opportunistic, GBBG is clearly the most predatory, feeding on fish, invertebrates, refuse and other birds while EHG is the most plastic in food preferences and on disturbed environments easily shifts from offshore and onshore food to anthropogenic sources (Threlfall 1968; Ewins et al., 1994; Rail and Chapdelaine 2000).

From each bird, five ventral feathers were pulled and 2 mL of blood were collected from the brachial vein using a heparinized syringe and a 25G needle to determine total Hg concentrations, the relative abundance of carbon (C), nitrogen (N), and sulfur (S) isotopes, and the sex of the chicks. After collection, whole blood was cooled to 4°C and within less than 2 h of sampling, it was spun at 4000 rpm for 6 min at 4°C to separate red blood cells for Hg and isotope analyses and plasma for Persistent Organic Pollutant analyses (data not shown). For this study, only red blood cells were used which were stored frozen (-80°C) until laboratory analyses.

2.2. Molecular sexing

Molecular sexing was conducted at the Centre d'Etudes Biologiques de Chizé (CEBC), France. Chicks were sexed from blood cells by polymerase chain reaction amplification of part of two

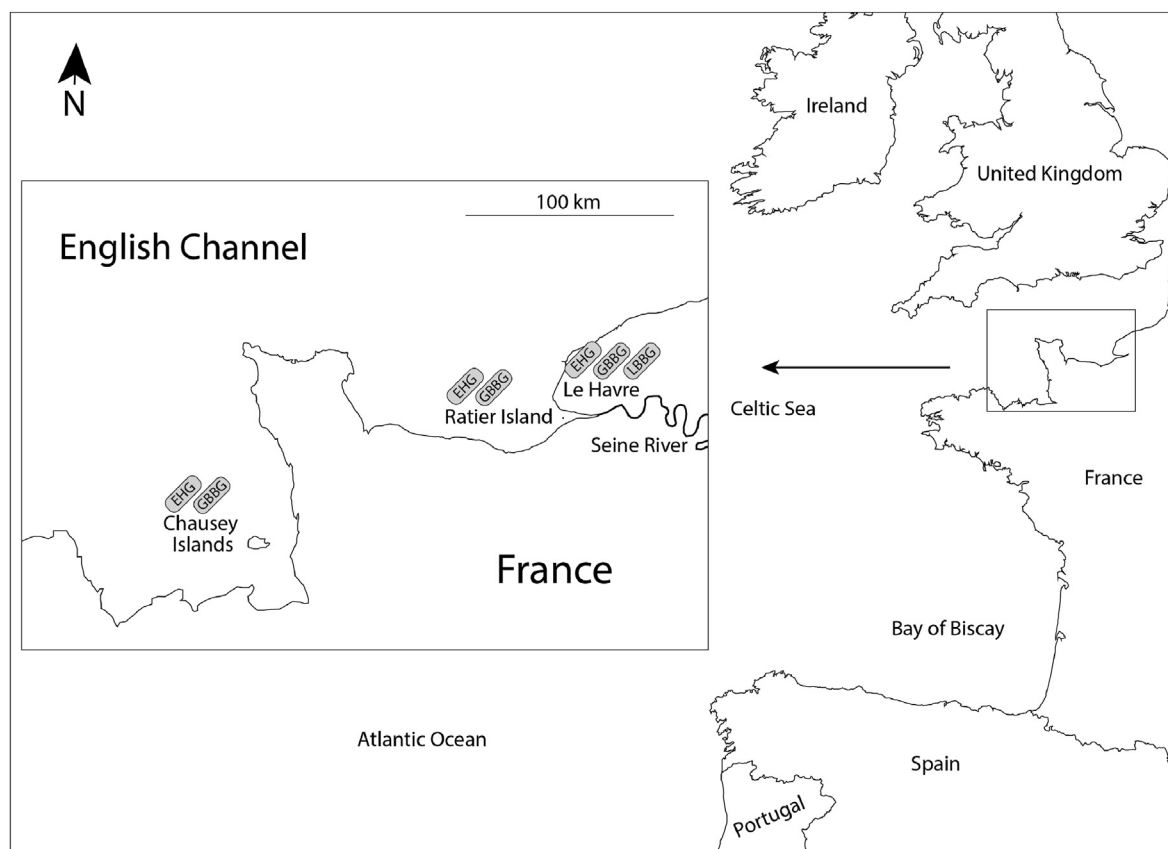


Fig. 1. Sampling sites along the French coast of the English Channel: The Chausey Islands and the Seine Estuary with the city of Le Havre and Ratier Island. EHG – European herring gull, GBBG – Great black-backed gull, LBBG – Lesser black-backed gull.

highly conserved genes (CHD) present in sexual chromosomes as described in Fridolfsson and Ellegren (1999).

2.3. Analysis of Hg and stable isotopes

Blood cell (hereafter blood) samples were freeze-dried for 48 h and ground to powder. The tips of the calami were removed from the feathers to avoid any contamination from the dermic papilla, and the feathers were cleaned with a solution of chloroform/methanol (2:1 v/v) followed by methanol rinsing. Clean feathers were dried for 48 h at 45 °C and cut to tiny pieces with steel scissors

for homogenization (Carravieri et al., 2013).

The relative abundance of stable isotopes (C, N and S) were measured in ~0.3 mg and ~0.6 mg aliquots of feathers and blood respectively, using a continuous flow mass spectrometer (Thermo Scientific Delta V Plus) coupled to an elemental analyzer (Thermo Scientific EA Flash, 2000). Isotopic data were presented as standard delta (δ) notation as parts per mil (‰) deviation relative to international standards (Vienna Pee Dee Belemnite for C, atmospheric N₂ for N, and Vienna-Canyon Diablo Troilite for S) in accordance with the formula: $\delta^{13}\text{C}$, $\delta^{15}\text{N}$ or $\delta^{34}\text{S} = [(R_{\text{sample}}/R_{\text{standard}}) - 1] \times 10^3$, where R is $^{13}\text{C}/^{12}\text{C}$, $^{15}\text{N}/^{14}\text{N}$ or $^{34}\text{S}/^{32}\text{S}$, respectively. Analytical

Table 1

Total mercury concentrations (mean \pm SD, $\mu\text{g}\cdot\text{g}^{-1}$ dw), $\delta^{13}\text{C}$, $\delta^{15}\text{N}$ and $\delta^{34}\text{S}$ values (mean \pm SD, ‰), in the blood of chick gulls studied in and outside the Seine Estuary.

Species		City of Le Havre	Ratier Island	Seine Estuary	Chausey Islands
EHG	Hg	0.32 \pm 0.25	0.47 \pm 0.32	0.37 \pm 0.28	0.37 \pm 0.18
	$\delta^{13}\text{C}$	−21.78 \pm 1.22	−21.86 \pm 1.15	−21.80 \pm 1.19	−20.21 \pm 0.62
	$\delta^{15}\text{N}$	10.17 \pm 1.53	10.51 \pm 1.48	10.28 \pm 1.51	12.10 \pm 0.84
	$\delta^{34}\text{S}$	9.63 \pm 2.52	9.62 \pm 1.90	9.62 \pm 2.33	14.16 \pm 1.26
	N	39	17	56	16
GBBG	Hg	1.56 \pm 0.74	1.38 \pm 0.27	1.48 \pm 0.60	3.10 \pm 0.92
	$\delta^{13}\text{C}$	−17.87 \pm 0.61	−18.22 \pm 1.12	−18.02 \pm 0.86	−17.40 \pm 0.45
	$\delta^{15}\text{N}$	14.73 \pm 0.70	15.39 \pm 0.17	15.00 \pm 0.63	15.50 \pm 0.38
	$\delta^{34}\text{S}$	13.76 \pm 1.02	13.03 \pm 1.19	13.46 \pm 1.14	16.93 \pm 0.48
	N	37	26	63	15
LBBG	Hg	0.61 \pm 0.18	—	0.61 \pm 0.18	—
	$\delta^{13}\text{C}$	−19.55 \pm 1.30	—	−19.55 \pm 1.30	—
	$\delta^{15}\text{N}$	13.04 \pm 1.54	—	13.04 \pm 1.54	—
	$\delta^{34}\text{S}$	12.10 \pm 1.58	—	12.10 \pm 1.58	—
	N	24	0	24	0

EHG: European herring gull; GBBG: Great black-backed gull; LBBG: Lesser black-backed gull.

precision for C, N and S isotopes was always better than 0.10, 0.15 and 0.20‰ respectively, for repeated measurements of laboratory standards: USGS-61 (Caffeine) and USGS-62 (Caffeine) for both C and N, and USGS-42 (human hair) and IAEA-S2 (silver sulfide) for S.

Total Hg (hereafter Hg) concentrations were measured in blood and feathers using an atomic absorption spectrophotometer (Altec AMA-254) on aliquots weighing 2–5 mg as described by [Chouvelon et al. \(2009\)](#). Briefly, analysis included burning the organic matrix at a temperature of 750 °C, bounding Hg evaporated by amalgamation on a gold trap, freeing Hg from amalgamate by heating it and finally measuring the absorption at 253.65 nm. All analyses were repeated 2–3 times until an SD for two measurements below 10% was acquired. The mean of these two measurements, expressed as $\mu\text{g}\cdot\text{g}^{-1}$ dry weight (dw), was considered for further statistical analyses. The accuracy and reproducibility of Hg measurements were assessed by the analyses of blanks and certified reference material (CRM) TORT-2 Lobster Hepatopancreas (NRC, Canada; certified mercury concentration: $0.27 \pm 0.06 \mu\text{g}\cdot\text{g}^{-1}$ dw). The CRM were analyzed at the beginning and at the end of each analytical cycle, and every 10 samples ([Bustamante et al., 2008](#)). The mass of the CRM was adjusted to represent an amount of Hg similar to that in the samples. Our measured values for the CRM were $0.25 \pm 0.01 \mu\text{g}\cdot\text{g}^{-1}$ dw ($n = 42$) showing a recovery of $93 \pm 2\%$. Blanks were analyzed at the beginning of each set of samples and the quantification limit of the method was 0.01 ng Hg.

2.4. Statistical analysis

All the statistical procedures and tests were run with R 3.5.2 ([R Core Team, 2018](#)) using Tidyverse ([Wickham, 2019](#)) and AICcmodavg packages ([Mazerolle, 2019](#)). The significance level was established at $\alpha = 0.05$. Prior to statistical analyses, data were scrutinized graphically to identify their distribution. Since the distribution of Hg concentrations was right-skewed, the concentrations were used further as log-transformed. To establish the final form of statistical protocol, we ran a preliminary data inquiry. As we found no statistical differences in any variable between the sexes (one-way ANOVA) and the study did not focus particularly on temporal trends, we pooled the data for both factors across their levels. In respect to obvious differences between species, we ran separate analyses for each species studied.

We ran two separate analyses: on a smaller-scale (within the Seine Estuary) using data from all three species studied from the city of Le Havre and Ratier Island, and on a larger-scale using data from EHG and GBBG from the Seine Estuary (pooled) and the Chausey Islands (excluding LBBG, which was collected solely in the Seine Estuary). In both cases, general linear models (GLM) were used with log Hg concentrations as dependent variable, site as discrete explanatory variable and isotopic values as continuous explanatory variables. All the continuous explanatory variables ($\delta^{13}\text{C}$, $\delta^{15}\text{N}$, $\delta^{34}\text{S}$) were tightly correlated in most cases ([Table S1](#)), so separate GLM models with particular isotopes were built. The analysis was done separately for each species. We built a set of models and used Akaike's Information Criterion corrected for small sample sizes (AICc) to identify the best models.

We also used $\delta^{13}\text{C}$, $\delta^{15}\text{N}$ and $\delta^{34}\text{S}$ values to study the isotopic space used by each species at the three breeding sites, with Stable Isotope Bayesian Ellipses in R package (SIBER) ([Jackson et al., 2011](#)). The standard ellipses were drawn containing approximately 40% of the data (level = 0.4). The standard ellipse area corrected for small sample size (SEAc) was calculated. Posterior estimates of the Bayesian standard ellipse area (SEAb) were then used to test for differences in the isotopic niche width.

3. Results

Hg concentrations were higher than the limit of detection in all the samples analyzed. Mean Hg concentrations were higher in chick feathers ($0.41\text{--}14.49 \mu\text{g}\cdot\text{g}^{-1}$ dw) than in chick blood ($0.05\text{--}4.30 \mu\text{g}\cdot\text{g}^{-1}$ dw), and both variables were tightly correlated ([Fig. S1](#); [Table S1](#)) as were the ratios of stable isotopes ([Table S1](#)). All further analyses on Hg and stable isotopes values were therefore carried out on blood only to avoid repetitions. The data set obtained for feathers is presented in the Supplementary Material ([Table S2](#)).

3.1. Comparisons within the Seine Estuary

In the Seine Estuary, Hg concentrations in EHG ([Table 1](#)) were positively related to $\delta^{15}\text{N}$ and influenced by the site factor ($p < 0.01$ and $p = 0.03$ respectively; [Fig. 2](#), [Table 2](#)). For GBBG, higher values on Ratier Island (contrarily to EHG) were explained by the same factors (for both $p < 0.01$). Since for GBBG correlations between $\delta^{13}\text{C}$, $\delta^{15}\text{N}$ and $\delta^{34}\text{S}$ isotopic values were low ([Table S5](#)), additional models including all the isotopes were constructed. The best model confirmed former observations that Hg concentrations were significantly influenced by $\delta^{15}\text{N}$ and site only ($p < 0.01$ and $p = 0.05$, respectively). In the case of LBBG, Hg concentrations appeared to be best explained by $\delta^{13}\text{C}$ (slope = 0.09, intercept = 1.60, $p < 0.01$; [Fig. 2](#), [Table 2](#)).

The highest Hg concentrations among chick gulls were noted in the blood of GBBG, regardless of the site, with $1.56 \pm 0.74 \mu\text{g}\cdot\text{g}^{-1}$ dw in the city of Le Havre and $1.38 \pm 0.27 \mu\text{g}\cdot\text{g}^{-1}$ dw on Ratier Island. In contrast, EHG showed the lowest concentrations at both sites with $0.33 \pm 0.25 \mu\text{g}\cdot\text{g}^{-1}$ dw in the city of Le Havre and $0.47 \pm 0.32 \mu\text{g}\cdot\text{g}^{-1}$ dw on Ratier Island. For the single colony of LBBG situated in the city of Le Havre, Hg concentrations ($0.61 \pm 0.18 \mu\text{g}\cdot\text{g}^{-1}$ dw) were intermediate between the two other species ([Table 1](#)).

3.2. Comparisons along the French coast of the English Channel

Comparisons between the Seine Estuary and the Chausey Islands show that Hg concentrations were positively related to $\delta^{15}\text{N}$ and additionally influenced by the site factor ($p < 0.01$ for both, [Table 3](#)). Regardless of the site, EHG had consistently lower Hg concentrations in the blood than GBBG ([Table 1](#)). For EHG, Hg reached similar concentrations at both sites ($0.37 \mu\text{g}\cdot\text{g}^{-1}$ dw), but for GBBG concentrations measured on the Chausey Islands were significantly higher (3.10 vs $1.48 \mu\text{g}\cdot\text{g}^{-1}$ dw) than in the Seine Estuary ([Fig. 2](#), [Table 1](#)).

3.3. Isotopic niches

The values for the three isotopes considered were lower in the blood of EHG than the other species at all sites, while values measured for GBBG were consistently the highest. The stable isotope values in the blood of LBBG fell between the two other species ([Fig. 2](#), [Table 1](#)). The relationships between isotope values had different strengths depending on the species and the sites ([Fig. 3](#) and [Fig. S2](#)). With the exception of GBBG chicks from Ratier Island, stable isotope values were correlated positively. The biggest SEAb were generally observed in EHG in Ratier Island (mean 9.72%) followed by LBBG in the city of Le Havre (mean 6.69% ; [Table 4](#)). All the differences for particular relationships among all the sites (one-way ANOVAs $p < 0.01$) and all the species (one-way ANOVAs $p < 0.01$) were statistically significant ([Table 4](#)).

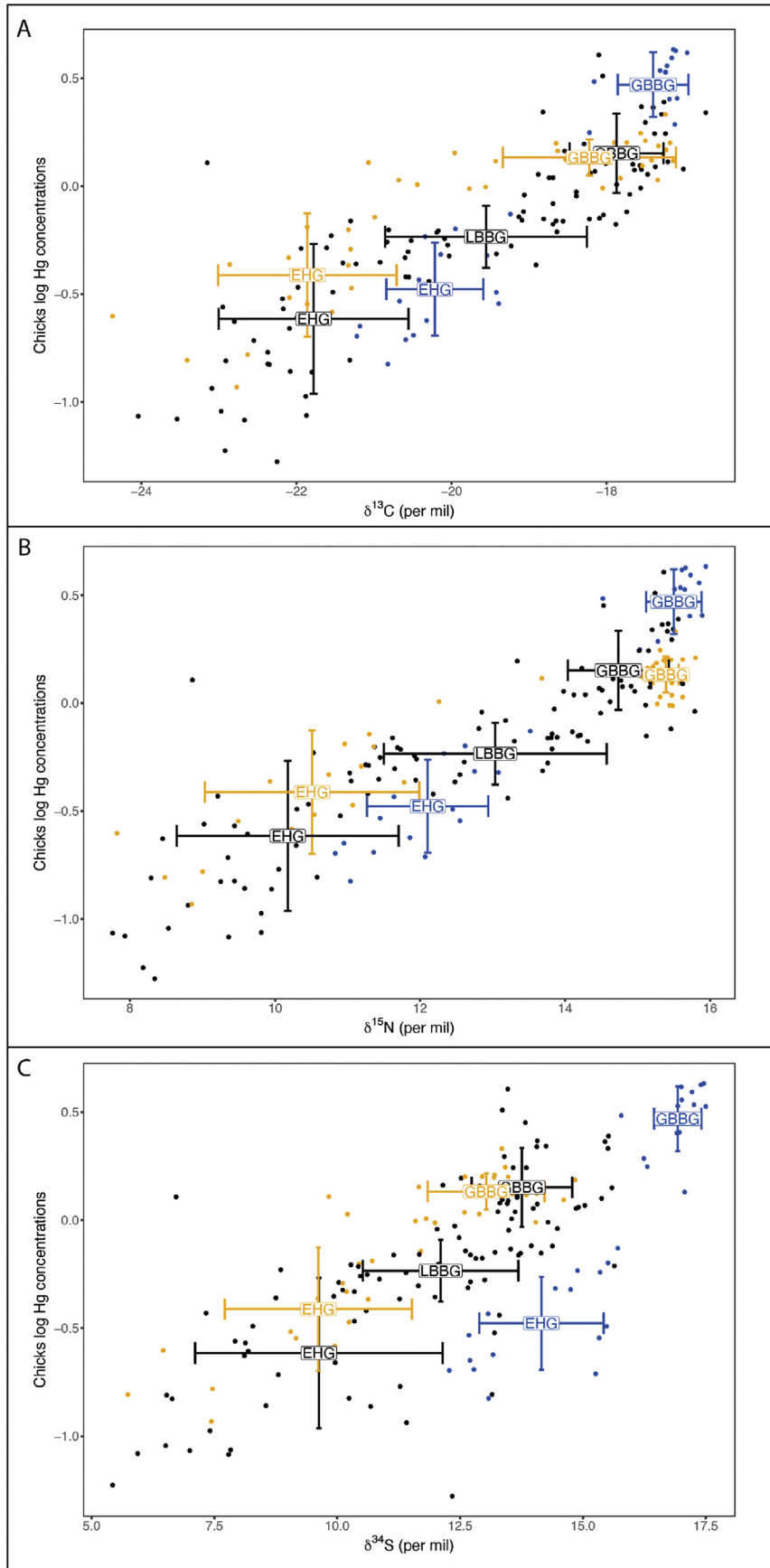


Fig. 2. Hg concentrations in blood as a function of $\delta^{13}\text{C}$ (A), $\delta^{15}\text{N}$ (B) and $\delta^{34}\text{S}$ (C) values in chick gulls from the French coast of the English Channel (EHG – European herring gull $N = 72$, GBBG – Great black-backed gull $N = 78$, LBBG – Lesser black-backed gull $N = 24$; blue – the Chausey Islands, black – the city of Le Havre, orange – Ratier Island). (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

Table 2Parameters of GLM and LM (AICc followed by R² adjusted) explaining blood Hg concentrations in chick gulls studied within the Seine Estuary.

Models	EHG	GBBG	LBBG
$\delta^{13}\text{C}$	18.87; 0.35	-62.31; 0.10	-50.11; 0.71
$\delta^{13}\text{C}$ + site	13.01; 0.42	-60.03; 0.08	—
$\delta^{13}\text{C}$ + site(+interactions)	15.30; 0.41	-61.27; 0.12	—
$\delta^{15}\text{N}$	1.45; 0.52	-66.89; 0.16	-43.44; 0.62
$\delta^{15}\text{N}$ + site	-1.33; 0.55	-73.15; 0.25	—
$\delta^{15}\text{N}$ + site(+interactions)	1.00; 0.55	-71.51; 0.25	—
$\delta^{34}\text{S}$	28.14; 0.23	-61.84; 0.09	-35.81; 0.48
$\delta^{34}\text{S}$ + site	24.46; 0.29	-59.67; 0.08	—
$\delta^{34}\text{S}$ + site(+interactions)	23.12; 0.33	-58.96; 0.08	—
$\delta^{13}\text{C}+\delta^{15}\text{N}+\delta^{34}\text{S}$	—	-68.55; 0.21	—
$\delta^{13}\text{C}+\delta^{15}\text{N}+\delta^{34}\text{S}$ + site	—	-70.46; 0.25	—
$\delta^{13}\text{C}+\delta^{15}\text{N}+\delta^{34}\text{S}$ + site (+all interactions)	—	-39.83; 0.13	—

Bolds indicate the best models according to AICc values. EHG: European herring gull; GBBG: Great black-backed gull; LBBG: Lesser black-backed gull.

Table 3Parameters of GLM (AICc followed by R² adjusted)* explaining blood Hg concentrations in chick gulls studied along the French coast of the English Channel.

Models	EHG	GBBG
site	43.56; 0.00	-70.47; 0.42
$\delta^{13}\text{C}$	16.74; 0.31	-44.41; 0.19
$\delta^{13}\text{C}$ + site	13.33; 0.35	-79.08; 0.49
$\delta^{13}\text{C}$ + site + all interactions	15.37; 0.34	-80.65; 0.51
$\delta^{15}\text{N}$	-2.09; 0.47	-49.96; 0.25
$\delta^{15}\text{N}$ + site	-10.34; 0.53	-83.55; 0.52
$\delta^{15}\text{N}$ + site + all interactions	-8.53; 0.53	-82.44; 0.52
$\delta^{34}\text{S}$	28.59; 0.18	-73.08; 0.44
$\delta^{34}\text{S}$ + site	24.54; 0.24	-76.88; 0.48
$\delta^{34}\text{S}$ + site + all interactions	26.38; 0.24	-76.26; 0.48

Bold indicates the best models according to AICc values. EHG: European herring gull; GBBG: Great black-backed gull; LBBG: Lesser black-backed gull.

4. Discussion

We found that, regardless of the site, Hg concentrations were highest in GBBG and lowest in EHG. In the Seine Estuary, there was no clear relationship between the location of the colony and Hg concentrations (e.g. the higher mean was noted for EHG in the city of Le Havre, but for GBBG on Ratier Island), suggesting that the feeding habits of the parents are more important than the sampling site of the chicks in this respect. Along the French coast of the English Channel and in the Seine Estuary, $\delta^{15}\text{N}$ and the site were equally important factors in explaining Hg concentrations. The only exception was LBBG, whose Hg concentrations were best explained by $\delta^{13}\text{C}$. This study is the first to report Hg concentrations in the chicks of these species from the French coasts and to make the link with their trophic ecology.

4.1. Hg contamination of the gull community

In seabirds, food represents the main pathway of exposure to Hg, so these predators reflect the Hg contamination of their food webs (Lavoie et al., 2013). Regardless of the geographic area, high Hg concentrations in seabirds are frequent in both adults and chicks (Carravieri et al. 2014, 2020). Studies from the same area, however, tended to reveal lower concentrations in chicks than in adults (Carravieri et al., 2014; Mallory et al., 2018; Kucharska et al., 2019), so comparison should be made within the same age group. Overall, the literature offers little information on chicks, so comparisons are usually compelled to include different species. Importantly, because chicks are fed with prey captured in the vicinity of the colony, Hg concentrations in either their blood or their feathers reflect local contamination (Blévin et al., 2013). In this study, Hg

concentrations in blood and feathers were strongly correlated, indicating that both tissues are relevant in surveys of Hg exposure. In the present study, only blood Hg concentrations will be further discussed.

Generally, chick blood Hg concentrations remain below $1 \mu\text{g}\cdot\text{g}^{-1}$ dw, but those in our study were higher. For instance, Hg concentrations in the blood of the American Herring gull chicks (*Larus smithsonianus*) from New York Bight (USA) reached different values in different years with 0.32 and $0.42 \mu\text{g}\cdot\text{g}^{-1}$ dw (recalculated as others from ww to dw based on 79.13% of water content; Eagles-Smith et al., 2008; Burger and Gochfeld, 1997). Hg concentrations in the blood of California gull chicks (*Larus californicus*) from San Francisco Bay (USA) were comparable with $0.34 \mu\text{g}\cdot\text{g}^{-1}$ dw (Peterson et al., 2017). Common gull chicks (*Larus canus*) from different parts of Germany revealed Hg concentrations in blood ranging from 0.28 to $0.40 \mu\text{g}\cdot\text{g}^{-1}$ dw (Kahle and Becker, 1999). In a study of 13 species of birds from the southern Indian Ocean (Carravieri et al., 2020), Hg concentrations in chicks' blood also remained below that of GBBG chicks from the Chausey Islands. These comparisons suggest that Hg concentrations noted along the French coast of the English Channel, especially for GBBG, were high and due to the high toxicity of Hg already documented, should be compared with toxicity data.

Toxicity thresholds are usually established for adult birds and are further extrapolated to include nestlings. Such inferences, however, may be biased, since chicks have a different efficiency of physiological processes and may thus not react to Hg toxicity in the same way as adults. Laboratory and field studies of adult birds indicated that concentrations above $14.4 \mu\text{g}\cdot\text{g}^{-1}$ dw (recalculated from $3.0 \mu\text{g}\cdot\text{g}^{-1}$ ww) or averaging $2 \mu\text{g}\cdot\text{g}^{-1}$ dw in common loons (*Gavia immer*) and male snow petrels (*Pagodroma nivea*), respectively, induce adverse effects (Evers et al., 2008; Tartu et al., 2015), highlighting that sensitivity to Hg is species-specific (Heinz et al., 2009). Even concentrations as low as $1 \mu\text{g}\cdot\text{g}^{-1}$ dw in blood may induce toxic effects by, for instance, a negative relationship with thiobarbituric acid activity (TBARs) (Custer et al., 2000). Because 88% of the GBBG chicks from the Seine Estuary and the Chausey islands have blood concentrations above this value, a question of the toxicological effects of Hg on these individuals arises. More generally, the impact of Hg contamination on the physiology, reproduction, survival and population dynamics of *Larus* gull populations highlights the need for further investigations in order to determine the biomarkers of effects and assess the toxicological impacts.

4.2. Comparisons in the Seine Estuary and farther afield

In the Seine Estuary environmental conditions were very similar

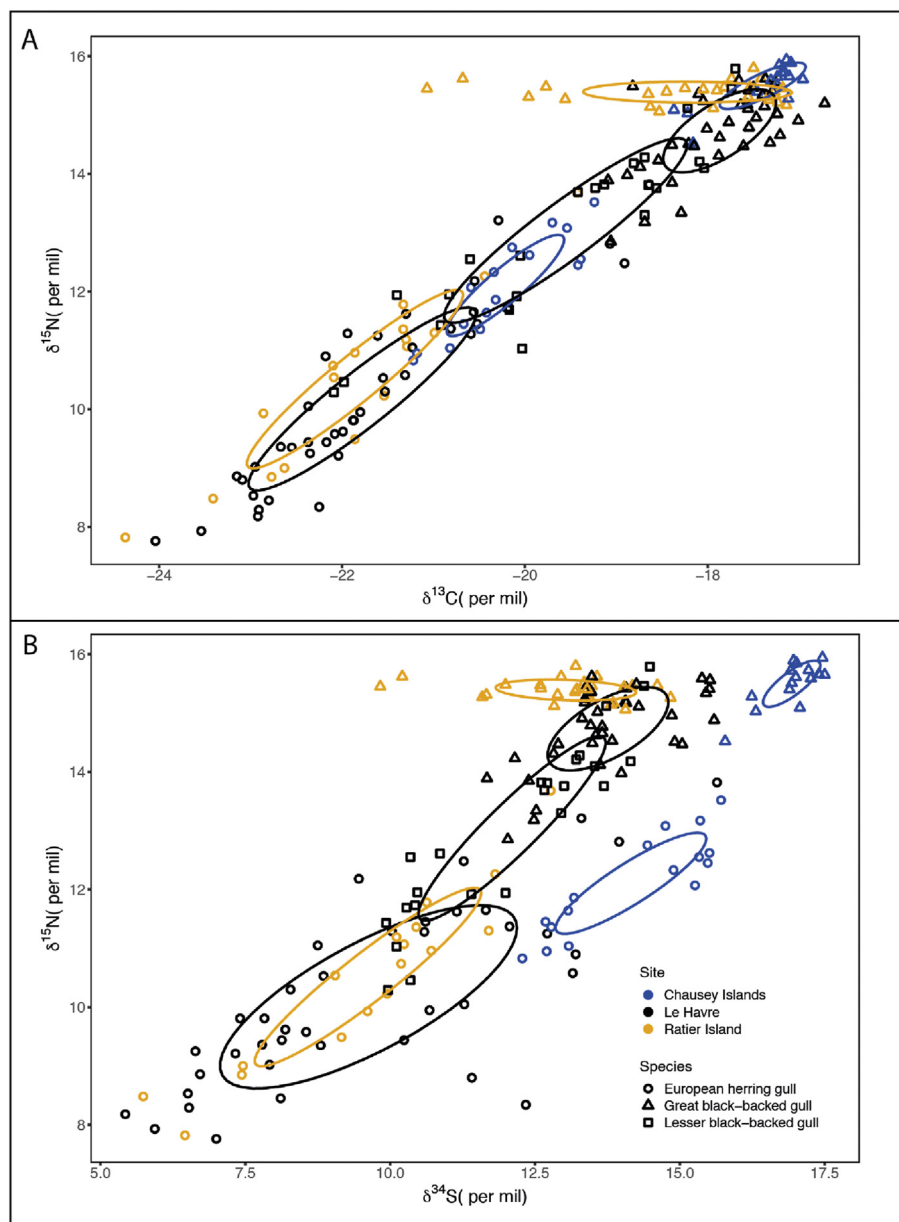


Fig. 3. Relationship between $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ (A) and $\delta^{34}\text{S}$ (B) isotopic values for all the species at all the sites studied, with isotopic-niche standard ellipses.

Table 4

Mean areas of the isotopic-niche ellipses (SEAb obtained using the Bayesian inference) for gull chicks from all the sites studied (all expressed as ‰²).

	Chausey Islands		City of Le Havre			Ratier Island	
	EHG	GBBG	EHG	GBBG	LBBG	EHG	GBBG
$\delta^{13}\text{C} \sim \delta^{15}\text{N}$	1.09 ^{a#}	0.32 ^{b#}	2.77 ^{a*}	0.77^{b*}	4.11^c	4.77^{a&}	0.44 ^{b†}
$\delta^{34}\text{S} \sim \delta^{15}\text{N}$	2.34 ^{a#}	0.38 ^{b#}	7.34 ^{a*}	1.19^{b*}	6.69^c	9.72^{a&}	0.45 ^{b†}
$\delta^{13}\text{C} \sim \delta^{34}\text{S}$	2.28 ^{a#}	0.48 ^{b#}	6.24^{a*}	1.24 ^{b*}	6.30^c	4.82 ^{a&}	1.59^{b†}

Bold indicates the greatest values for the species. EHG: European herring gull; GBBG: Great black-backed gull; LBBG: Lesser black-backed gull. Different letters indicate significant differences between species within the given site (ANOVA or *t*-test, $p < 0.05$). Different symbols indicate significant differences between sites within a given species (ANOVA, $p < 0.05$).

at both sites (the city of Le Havre and Ratier Island), so differences observed were clearly linked to discrepancies between the species. Both study sites in the Seine Estuary are also close enough to

suspect reasonably that birds from both colonies may forage in the same locations. Interestingly, GBBG displayed the highest Hg concentrations, followed by LBBG and EHG. These results are consistent with previous observations of GBBG and EHG along the Baltic coast of Poland (Szumilo-Pilarska et al., 2016) and with the ecology of these species. Even both species are generalist as explained previously, GBBG also feeds other birds (e.g., Gilliland et al., 2004) and competes with LBBG and EHG, and is suspected of contributing to their decline (e.g. Rome and Ellis 2004). Although we did not investigate the prey fed to gull chicks directly, $\delta^{15}\text{N}$ values and standard ellipse shapes (Fig. 3, Table 4) confirm that GBBG tend to feed at higher trophic levels than LBBG and EHG. Moreover, $\delta^{34}\text{S}$ values also indicate that GBBG specialize more on marine prey than the other species (Hobson 1999). Along with the trophic position (proxied with $\delta^{15}\text{N}$), we observed an increase in Hg concentrations in the blood (Fig. 2). This indicates that Hg was biomagnified in the gull community of the English Channel.

Larger-scale comparison mainly included the aspects of urbanization and spatial pollution. Part of the study was carried out in highly urbanized areas within the Seine Estuary. The Seine River is also one of the most polluted rivers in Europe with decreasing, but still significant contamination with Hg (Hamzeh et al., 2013). The Chausey Islands are on the other hand a remote area with no direct sources of Hg, where seabird exposure is likely to be linked to their marine food. In contrast to our prediction (i.e. higher Hg concentrations in birds from the Seine Estuary), Hg concentrations in EHGs were comparable in the two regions and GBBG was significantly higher on the Chausey Islands. The relatively small SEAB and high blood $\delta^{34}\text{S}$ values in birds from the Chausey Islands indicate that birds breeding at this latter site occupied a much smaller trophic niche, and thus fed on a smaller range of prey and within more restricted habitats. These differences most likely reflect the lower diversity of food and feeding grounds available in the Chausey Islands, limited to marine prey (Tables 1, 4 and Fig. 3). In contrast, in the Seine Estuary, where a large mosaic of habitats are available along the terrestrial-marine continuum, EHGs seems to feed on more species of coastal prey (indicated by lower $\delta^{34}\text{S}$ values) and GBBG on prey lower in the food chain (indicated by lower $\delta^{15}\text{N}$ values) (Fig. 3). This explains higher Hg concentrations in GBBG on the Chausey Islands, since prey and habitat choices are known to influence Hg exposure, with marine diet and higher trophic levels leading to enhanced exposure (Ochoa-Acuña et al., 2002).

5. Conclusions

Thanks to the examination of the chicks of three different gull species from three colonies on the French coast of the English Channel, we provide the first data for gull chick contamination with Hg in the region and geographic variations comparing a contaminated site (the Seine Estuary) and a reference site (the Chausey Islands). Irrespective of the site, GBBG consistently showed the highest Hg concentrations. As Hg concentrations in blood were often above the toxicity threshold, they may produce toxic effects, deserving of further investigations. With the exception of LBBG, $\delta^{15}\text{N}$ was the most important factor explaining Hg concentrations in the Seine Estuary, meaning that the exposure to Hg is closely linked to the trophic position of the birds. A wider perspective showed that, as well as site and trophic position, the feeding habitat indicated by $\delta^{34}\text{S}$ values, also appear to be an important driver in Hg exposure. Considering the Seine Estuary alone and the French coast of the English Channel as a whole, the biomagnification of Hg was observed in both cases. Chicks' exposure to Hg was clearly driven by the feeding ecology of the species studied.

Author statement

Lukasz J. Binkowski: Conceptualization, Formal analysis, Writing - original draft, Visualization, Writing - review & editing. Jérôme Fort: Conceptualization, Supervision, Fieldwork, Formal analysis, Validation, Writing - review & editing. Maud Brault-Favrou: Methodology, Investigation, Writing - review & editing. Fabrice Gallien: Fieldwork, Writing - review & editing. Gilles Le Guillou: Fieldwork, Writing - review & editing. Olivier Chastel: Methodology, Writing - review & editing. Paco Bustamante: Conceptualization, Supervision, Project administration, Fieldwork, Funding acquisition, Validation, Writing - review & editing.

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.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.chemosphere.2020.128622>.

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