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Mercury concentrations in seabird tissues from Machias Seal Island, New Brunswick, Canada

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ABSTRACT

Mercury is a pervasive environmental contaminant, the anthropogenic portion of which is increasing globally, and in northeastern North America in particular. Seabirds frequently are used as indicators of the marine environment, including mercury contamination. We analysed paired samples for total mercury (Hg) concentrations in feathers and blood from adult and chick, albumen, and lipid-free yolk of seven seabirds breeding on Machias Seal Island, New Brunswick, Canada — Arctic Tern (*Sterna paradisaea*), Atlantic Puffin (*Fratercula arctica*), Common Eider (*Somateria mollissima*), Common Murre (*Uria aalge*), Common Tern (*Sterna hirundo*), Leach's Storm-petrel (*Oceanodroma leucorhoa*), and Razorbill (*Alca torda*). We also used stable-isotope ratios of carbon (δ^{13}C), and nitrogen (δ^{15}N) to evaluate the relationship between carbon source and trophic position and mercury. We found high Hg concentrations across tissue types in Leach's Storm-petrels, and Razorbills, with lower concentrations in other species, the lowest being in Common Eiders. Storm-petrels prey on mesopelagic fish that accumulate mercury, and Razorbills feed on larger, older fish that bioaccumulate heavy metals. Biomagnification of Hg, or the increase in Hg concentration with trophic position as measured by δ^{15}N, was significant and greater in albumen than other tissues, whereas in other tissues, δ^{15}N explained little of the overall variation in Hg concentration. Hg concentrations in egg components are higher on Machias Seal Island than other sites globally and in the Gulf of Maine region, but only for some species. Further detailed investigations are required to determine the cause of this trend.

1. Introduction

While naturally occurring (Nriagu and Pacyna, 1988; Nriagu, 1989), mercury (Hg) concentrations in the North Atlantic have been increasing over the last century (Appelquist et al., 1985; Thompson et al., 1992; Monteiro and Furness, 1997), particularly in northeastern North America (Perry et al., 2005). Seabirds are often used as indicators of marine ecosystem health (Cairns, 1987; Piatt et al., 2007) and frequently have been used to monitor Hg in marine environments by non-destructively sampling tissues such as feathers, blood, and eggs (e.g., Barrett et al., 1996).

Seabirds eliminate Hg through moulting and replacing feathers (Braune and Gaskin, 1987b; Monteiro and Furness, 2001), with smaller proportions of the total body Hg burden excreted into guano or eggs (Monteiro and Furness, 1995; Monteiro and Furness, 2001). Nevertheless, eggs often are used to monitor changes in Hg over time since they can easily be collected (N. Burgess, pers. comm., Goodale et al., in press). Nearly all Hg in seabird feathers, eggs and blood is organic methyl mercury (Sell et al., 1974; Thompson and Furness, 1989a,b; Bond and Diamond, 2009), the toxic and biologically active form (Weiner and Spry, 1996).

In general, Hg contamination occurs through seabirds' food and once ingested, Hg is transported through the blood (Kahle and Becker, 1999) to the liver where it can be demethylated and converted into inorganic Hg (Spalding et al., 2000; Burger and Gochfeld, 2002). The remaining methyl Hg remains as a body pool until it can be excreted during feather replacement or into eggs (Braune and Gaskin, 1987a; Monteiro and Furness, 2001; Rumbold et al., 2001; Bond and Diamond, 2009).

Naturally occurring stable-isotope ratios of carbon (δ^{13}C) and nitrogen (δ^{15}N) are used as continuous scales of trophic position (nitrogen) or carbon source (inshore δ^{13}C sources being more enriched in ^{13}C than offshore sources). Stable isotopes are useful in ecotoxicology studies because they provide continuous variables against which Hg concentrations can be related, provide information on the importance of different dietary sources of Hg using the same tissues as Hg analysis and can be non-destructively sampled (Jardine et al., 2006).

We examined total Hg and stable-isotope ratios in seven breeding seabirds — Arctic Tern (*Sterna paradisaea*), Atlantic Puffin (*Fratercula arctica*), Common Eider (*Somateria mollissima*), Common Murre (*Uria aalge*), Common Tern (*Sterna hirundo*), Leach's Storm-

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petrel (*Oceanodroma leucorhoa*), and Razorbill (*Alca torda*) — breeding on Machias Seal Island (MSI) in the lower Bay of Fundy.

Hg and some organic contaminants are monitored every four years in Atlantic Puffins and Leach's Storm-petrels from MSI and nearby Kent Island, respectively, by the Canadian Wildlife Service (N. Burgess, unpublished data), but there currently are no assessments of Hg for the other species in this area.

Monitoring programs are essential to tracking changes in Hg contamination over time, and programs have been in place in eastern Canada since the 1960's (e.g., Pearce et al., 1979), which has included collections at MSI. Biologically, MSI is more integrated with islands in the larger Gulf of Maine area (Breton et al., 2006; Devlin et al., 2008), which is an area where increasing Hg concentrations over the time have been documented (Evers et al., 2005; Perry et al., 2005).

The goals of this study were therefore two-fold: first, to examine the relationships between δ^{15}N and Hg in whole blood, albumen, lipid-free yolk (hereafter yolk) and feathers from a variety of species with differing foraging behaviours and life histories; and second, to compare Hg concentrations from MSI to concentrations from other regional and global sites from the published literature. In addition, this study will provide a comprehensive assessment of Hg concentrations in the breeding seabirds of the island to act as a comparison for future Hg studies.

Within whole blood, albumen and yolk the prediction is that there would be positive relationships between Hg and δ^{15}N for all species. A relationship between δ^{15}N and Hg in adult feathers was not expected as feather Hg is deposited from a body pool, and δ^{15}N is representative of the diet at the time of feather growth. Albumen is also predicted to have higher Hg concentrations than yolk because albumen has a high protein content (Magat and Sell, 1979), and Hg binds to disulphide bonds in protein (Crewther et al., 1965; Vallee and Ulmer, 1972; Sell et al., 1974).

2. Materials and methods

2.1. Study site & field collection

Machias Seal Island (44°30′N / 67°06′W, hereafter MSI) is a 9.5 ha treeless island that supports a diverse seabird community (Diamond and Devlin, 2003). In late July and early August of 2005 and 2006, we collected worn feathers, whole blood, and fresh whole eggs from all species of seabirds breeding on the island.

Whole eggs were collected as early into incubation as possible, and freshness was determined by flotation (fresh eggs sink in fresh water). Alcids and storm-petrels lay one egg per clutch, so there are no effects of laying sequence on Hg concentrations. For terns, clutches of only one egg were collected and for eiders, the longest egg was assumed to be the first egg (Robertson and Cooke, 1993). Whole eggs were covered in cellophane wrap and frozen in the field at −20 °C within 30 min. of collection, which does not alter isotopic composition (Gloutney and Hobson, 1998). Albumen and yolk were manually separated by removing the eggshell and membrane and cutting the frozen egg using a knife. All instruments and bench tops were sanitized between samples to minimize cross-contamination. Yolk and albumen were then placed in sterile plastic bags, thawed and manually homogenized by vigorous shaking for 30 s before being transferred to sterile glass vials.

Blood samples were taken from adults and chicks via the brachial vein using a needle and capillary tubes, placed in sterile glass vials, and frozen at −20 °C. Breast feathers (hereafter "feathers") were plucked from the same individuals, placed in sterile plastic bags and frozen. We chose worn feathers because they are grown in the winter, away from the breeding grounds (Pyle, 2008). We did not sex individuals, and make no inferences about sex differences in Hg concentrations (Becker et al., 2002).

We opportunistically collected prey items (whole fish or euphausiid shrimp) from regurgitating birds or from specimens dropped in the colony. Prey samples were identified, weighed, and measured, then frozen in sterile plastic bags.

2.2. Lab Analyses

Frozen prey items, and separated, homogenized albumen and yolk samples were freeze dried for 24–48 h in a Virtis Benchtop freeze-dryer. It is recommended that tissues with C:*N*>4.0 should be lipid-extracted (Post et al., 2007). Lipids, which are depleted in ^{13}C, were removed (Bligh and Dyer, 1959) from yolk and prey samples, eliminating this bias (Hobson, 1995; Post et al., 2007), and lipid extraction was not required for feathers, albumen, or blood (ALB, unpublished data; Kojadinovic et al., 2008). Approximately 2 ml of a 2:1 chloroform:methanol solution was used to wash the samples until the supernatant appeared clear, indicating that the majority of the lipids had been removed (Bligh and Dyer, 1959). Removing lipids has little effect on δ^{15}N values (Ricca et al., 2007). Samples were freeze-dried again for 24 h to remove residual traces of the chloroform:methanol solution and then powdered. Feathers were washed prior to analysis using a 0.25 M NaOH solution to remove external contamination (Bearhop et al., 2000b, 2002). Washing does not affect mercury concentrations as the mercury is bound to the disulfide bonds in the keratin (Crewther et al., 1965; Appelquist et al., 1984; Furness et al., 1986). Frozen blood samples were thawed, and 0.5 mL of 0.05% KOH solution was added to remove clots. The KOH solution does not alter mercury concentrations when compared to blanks (A.L.B., unpublished data), and by measuring the amount of blood prior to adding KOH it is possible to calculate blood Hg concentrations.

Total mercury was analysed at the Faculty of Forestry and Environmental Management, University of New Brunswick. Samples were loaded in a quartz boat and analysed in a Milestone DMA-80 direct mercury analyzer by atomic absorption spectrophotometry with a detection limit of 0.2 ng (Haynes et al., 2006). Internal duplicates and dogfish muscle standard (DORM-2, NRC Canada, certified total Hg: 4640 ppb) were used, in combination with blanks (deionised water), to calibrate the results in each run. Recovery of total mercury from DORM-2 was 87 ± 6.33% (mean ± standard deviation).

Freeze-dried and powdered egg components, prey items and whole feathers with 0.2 mg removed from the distal end for stable-isotope analysis were analyzed by placing 0.02–0.08 g in the boats. Results are expressed as parts per billion (ppb) fresh weight for blood and feathers, and dry weight for egg components. The amount of moisture in egg components can differ from year to year (Arnold et al., 1991), so reporting dry weight concentrations of mercury is recommended.

Stable isotopes were analyzed at the Stable Isotopes in Nature Laboratory (SINLAB) at the University of New Brunswick. Blood was freeze dried as described above prior to stable-isotope analysis. Approximately 0.2 mg of each sample was combusted in a Carlo Erba NC2500 elemental analyzer, and the resultant gases were delivered to a Finnigan Mat Delta XP mass spectrometer via continuous flow. Throughout analyses, internal repeats and standards (presented below as mean ± S.D.) were used. Values were corrected using International Atomic Energy Agency (IAEA) standards CH6 (δ^{13}C: −10.49 ± 0.21), N1 (δ^{15}N: 0.25 ± 0.29), and N2 (δ^{15}N: 20.54 ± 0.17). Internal lab standards of small-mouth bass muscle (δ^{13}C: −23.26 ± 0.12; δ^{15}N: 12.42 ± 0.12), bovine liver (δ^{13}C: −18.71 ± 0.12; δ^{15}N: 7.25 ± 0.18), acetanilide (2 batches, δ^{13}C: −33.60 ± 0.15, −33.13 ± 0.14; δ^{15}N: −3.16 ± 0.19, −1.18 ± 0.19), and nicotinamide (δ^{13}C: −34.20 ± 0.12; δ^{15}N: −1.77 ± 0.14) also were used. One standard deviation of sample duplicates within runs was <0.55‰ for both δ^{13}C and δ^{15}N.

Results are expressed as differences in isotopic ratios expressed in parts per thousand (‰) as compared to international standards; Pee Dee Belemnite (PDB) for carbon and atmospheric nitrogen (AIR) for nitrogen, as

$$\delta X = \left(\frac{R_{\text{sample}}}{R_{\text{std}}} - 1\right) \times 1000$$

where δX is either δ^{15}N or δ^{13}C and R_{sample} is the ratio of ^{15}N/^{14}N or ^{13}C/^{12}C respectively, and R_{std} is the ratio present in the international standard.

2.3. Statistical analysis

To test for factors influencing Hg concentrations, we used generalized linear models with a normal distribution and an identity-link function in SPSS 16.0.2 (SPSS Inc., Chicago). We based model selection on Akaike's Information Criteria adjusted for small sample sizes (AIC_c), and model structure fit ($QAIC_c$) when $\hat{c}>1.0$. We considered the model with the lowest AIC_c or $QAIC_c$ value to be the best fitting model to the data. First, we constructed a Null Model (intercept only) and then models that included additive terms and their interactions for species, age class, year, $\delta^{13}C$ and $\delta^{15}N$ (the Global Model). We then constructed biologically relevant models incorporating these terms. Models with ΔAIC_c or $\Delta QAIC_c>2$ were considered to have substantially less support, and overall model support was assessed using Akaike weights (Burnham and Anderson, 2002).

Finally, we used the estimated marginal means (EMMs) from the generalized linear model to examine differences among species and age classes. EMMs from the top-ranked model were used, and differences were considered significant if 95% confidence intervals did not overlap.

To test for direct relationships between $\delta^{15}N$ and Hg concentration, we used linear regressions, and to test for the amount of variation not attributed to trophic position that was accounted for by carbon source (inshore/offshore), we regressed $\delta^{13}C$ on $\delta^{15}N$-Hg regression residuals (Ricca et al., 2008). All tests were considered significant at $p<0.05$.

3. Results

3.1. Blood

After adjusting for $\hat{c}=2.96$, the best-fitting model included an effect of species and age, and less support for a year effect (Table 1). Based on estimated marginal means (EMMs), Common Eider adults had the lowest blood Hg concentration and Razorbills the greatest (Table 2). Among chicks, Razorbills had the greatest Hg concentrations, while Leach's Storm-petrels, Arctic Terns and Atlantic Puffins had lesser concentrations (Table 2).

The relationship between $\delta^{15}N$ and Hg was significant for adults (linear regression, $p=0.05$, $r^2=0.03$), but explained little variation. We used the slope of the $\delta^{15}N$-Hg regression as a biomagnification factor (BMF). For adult whole blood, the BMF was 0.22 (Fig. 1), and the regression of $\delta^{15}N$-Hg residuals against $\delta^{13}C$ was negative, but not significant ($p=0.33$). There was no significant relationship in chicks between $\delta^{15}N$ and Hg ($p=0.23$, $r^2=0.02$), and the slope was less than that in adults (0.08); however the $\delta^{15}N$-Hg residual regression on $\delta^{13}C$ was significant and positive ($p=0.004$, $r^2<0.11$).

Table 1
Summary of the model sets for feather and blood Hg concentrations.

Model	No. parameters	$QAIC_c$	$\Delta QAIC_c$	Akaike weight
Feathers ($\hat{c}=1.21$)				
S+A+SA	12	291.100	0.000	0.52
S+A	8	291.846	0.745	0.36
S+A+Y	10	294.106	3.006	0.11
S+A+Y+SA+SY+AY	21	298.625	7.525	0.01
$\delta^{15}N$	3	320.746	29.646	0.00
Null	2	331.207	40.107	0.00
Global	71	393.476	102.375	0.00
Blood ($\hat{c}=2.96$)				
S+A	9	430.422	0.000	0.47
S+A+Y	10	431.142	1.003	0.29
S+A+Y+$\delta^{15}N$	11	432.198	2.155	0.16
S+A+SA	14	430.604	3.733	0.07
S+A+Y+SA+SY+AY	21	432.926	10.299	0.00
S+Y	9	518.841	36.198	0.00
Null	2	548.628	43.765	0.00
$\delta^{15}N$	3	549.556	44.771	0.00
Global	82	516.143	117.678	0.00

The Global Model includes all factors and their interactions; the Null Model is an intercept-only model. Models are sorted by increasing $QAIC_c$ (quasi Akaike's information criteria corrected for small sample size) value with the most parsimonious model at the top. Akaike weight is the likelihood that a given model of the model set is the best approximation of the data. Model parameters abbreviations: S: species; A: age (adult or chick); Y: year.

Table 2
Estimated marginal mean (EMM) blood Hg concentrations for breeding seabirds on Machias Seal Island from 2005–2006 were derived from the top-ranked generalized linear model.

Species*	*n*	Mean [Hg] (ppb, wet wt.)	95% Confidence limit		Group
			Lower	Upper	
Adults					
COEI	4	51	16	158	A
LHSP	16	122	84	177	AB
ARTE	29	163	113	234	AB
ATPU	17	174	120	252	AB
COTE	24	242	163	358	BC
COMU	9	272	164	450	BC
RAZO	20	361	255	513	C
Chicks					
LHSP	19	23	16	33	A
ARTE	6	30	20	47	A
ATPU	18	32	23	47	A
COTE	2	45	28	74	AB
COMU	9	51	31	84	AB
RAZO	19	68	47	96	B

* ARTE: Arctic Tern; ATPU: Atlantic Puffin; COEI: Common Eider; COMU: Common Murre; COTE: Common Tern; LHSP: Leach's Storm-petrel; RAZO: Razorbill.
Differences were considered significant when confidence intervals do not overlap; species sharing the same Group letter are not significantly different from each other. Species with two letters are not significantly different from either group.

3.2. Feathers

As with blood, the top-ranked model for feather Hg concentrations after correcting for $\hat{c}=1.21$ included an effect of species and age class, but also their interaction. A similar model with no interaction was also

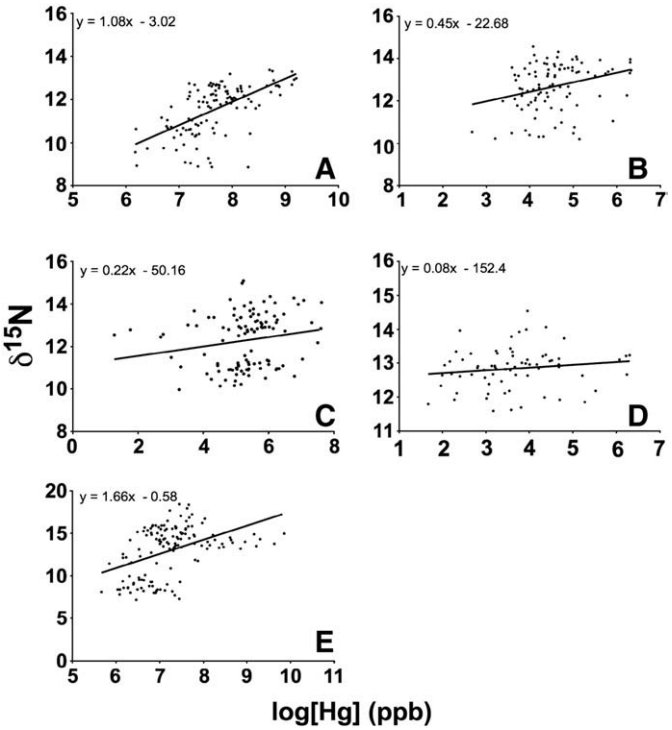

Fig. 1. Regressions of $\delta^{15}N$-Hg indicating biomagnification factors (BMF) for albumen (A), yolk (B), adult blood (C), chick blood (D) and adult feathers (E) for breeding seabird species from Machias Seal Island, 2005–2006.

Table 3
Estimated marginal mean (EMM) feather Hg concentrations for breeding seabirds on Machias Seal Island from 2005–2006 were derived from the top-ranked generalized linear model.

Species*	*n*	Mean [Hg] (ppb, fresh wt.)	95% Confidence limit		Group
			Lower	Upper	
Adults					
ARTE	36	741	490	1115	A
RAZO	21	1285	820	2014	A
ATPU	17	1411	877	2269	A
COEI	5	1539	385	6158	AB
COTE	28	1623	1047	2515	A
COMU	17	1652	787	3465	A
LHSP	15	7006	3880	12650	B
Chicks					
ARTE	2	989	247	3953	AB
ATPU	17	999	272	726	A
COMU	9	1136	591	2182	AB
COTE	3	1159	290	4634	AB
RAZO	16	1403	859	2290	AB
LHSP	20	1416	903	2220	AB

*Species abbreviations as in Table 2.
Differences were considered significant when confidence intervals do not overlap; species sharing the same Group letter are not significantly different from each other. Species with two letters are not significantly different from either group.

greatly supported by the data (Table 1). Based on EMMs, there were two groups with similar feather Hg concentrations: Leach's Storm-petrels with the greatest concentrations, and all other species with lesser concentrations than Leach's Storm-petrels, but no different from each other (Table 3). Atlantic Puffin chicks had lesser Hg concentrations than other species, while Razorbills and Leach's Storm-petrels had greater Hg concentrations than the four other species (Table 3).

Because we could not sample tern chicks in 2006 and we did not sample murre chicks in 2005, comparisons of chick feather Hg concentrations among species is difficult. Overall, Razorbills and Leach's Storm-petrels had the greatest chick feather Hg concentrations, while Atlantic Puffins had the lowest.

There was a significant relationship between δ^{15}N and Hg in adult feathers (linear regression, $p<0.001$, $r^2=0.20$), and it explained a relatively large amount of the overall variation in Hg as compared with other tissues; the slope was 1.66 (Fig. 1). The residuals from the δ^{15}N-Hg regression against δ^{13}C were not significant $p=0.66$).

Table 4
Summary of the model sets for albumen and yolk Hg concentrations.

Model	No. parameters	AIC_c	ΔAIC_c	Akaike weight
Albumen				
S	7	243.508	0.000	0.59
S + Y	8	245.765	2.258	0.19
S + δ^{15}N	8	245.785	2.278	0.19
δ^{15}N	2	249.729	6.222	0.03
S + Y + SY	13	256.854	13.346	0.00
Null	1	264.681	21.173	0.00
Y	2	266.567	23.060	0.00
Global	52	412.725	169.217	0.00
Yolk				
S	7	223.901	0.000	0.57
S + Y	8	225.678	1.777	0.24
S + δ^{15}N	8	226.294	2.393	0.17
δ^{15}N	2	231.817	7.916	0.01
Null	1	233.959	10.058	0.00
Y	2	234.537	10.636	0.00
S + Y + SY	13	236.388	12.488	0.00
Global	51	407.639	183.738	0.00

The Global Model includes all factors and their interactions; the Null Model is an intercept-only model. Models are sorted by increasing AIC_c (Akaike's information criteria corrected for small sample size) value with the most parsimonious model at the top. Akaike weight is the likelihood that a given model of the model set is the best approximation of the data. Model parameters abbreviations: S: species; Y: year.

Table 5
Estimated marginal mean (EMM) albumen Hg concentrations for breeding seabirds on Machias Seal Island from 2005–2006 were derived from the top-ranked generalized linear model.

Species*	*n*	Mean [Hg] (ppb, dry wt.)	95% Confidence limit		Group
			Lower	Upper	
COEI	18	1246	785	1978	A
COTE	13	1290	749	2222	A
ARTE	20	1468	947	2276	A
ATPU	20	2154	1374	3377	A
COMU	10	2353	1266	4373	AB
RAZO	20	3013	1944	4671	AB
LHSP	17	6392	3974	10283	B

*Species abbreviations as in Table 2.
Differences were considered significant when confidence intervals do not overlap; species sharing the same Group letter are not significantly different from each other. Species with two letters are not significantly different from either group.

3.3. Albumen

The models fit the data such that correction model fit using $\hat{c}$ was not required. The top-ranked model for predicting albumen Hg concentrations only included a species effect; no other model received substantial support (Table 4). Based on EMMs, Leach's Storm-petrels had greater Hg concentrations than other species, followed by Razorbills and Common Murres. All species, except Leach's Storm-petrels, had similar Hg concentrations, however (Table 5).

The relationship between δ^{15}N and albumen Hg concentration was significant (linear regression, $p<0.0001$, $r^2=0.36$), with a positive slope of 1.08 (Fig. 1). Regression of the residuals from the δ^{15}N-Hg regression against δ^{13}C was not significant ($p=0.21$, $r^2=0.01$).

3.4. Yolk

Similar to albumen, no correction using $\hat{c}$ was needed, and the model that best predicted yolk Hg concentrations only included a year effect, but the model with both species and year effects received some support as well (Table 4). EMMs predicted the same groups of species based on yolk Hg concentrations as for albumen Hg concentrations (Table 6).

There was a significant positive relationship between δ^{15}N and Hg for yolk (linear regression, $p=0.003$), but little overall variation was explained ($r^2=0.08$). The slope of 0.45 was less than that of albumen, but greater than that of adult whole blood (Fig. 1). The regression of δ^{15}N-Hg residuals on δ^{13}C was significantly positive ($p=0.04$, $r^2=0.04$), indicating that species foraging more inshore tended to have greater Hg concentrations (Fig. 2).

Table 6
Estimated marginal mean (EMM) yolk Hg concentrations for breeding seabirds on Machias Seal Island from 2005–2006 were derived from the top-ranked generalized linear model.

Species*	*n*	Mean [Hg] (ppb, dry wt.)	95% Confidence limit		Group
			Lower	Upper	
COTE	12	150	85	264	A
ATPU	19	184	116	293	A
ARTE	17	205	89	329	A
COEI	15	214	129	355	A
COMU	10	259	139	482	AB
RAZO	20	359	232	557	AB
LHSP	11	787	436	1422	B

*Species abbreviations as in Table 2.
Differences were considered significant when confidence intervals do not overlap; species sharing the same Group letter are not significantly different from each other. Species with two letters are not significantly different from either group.

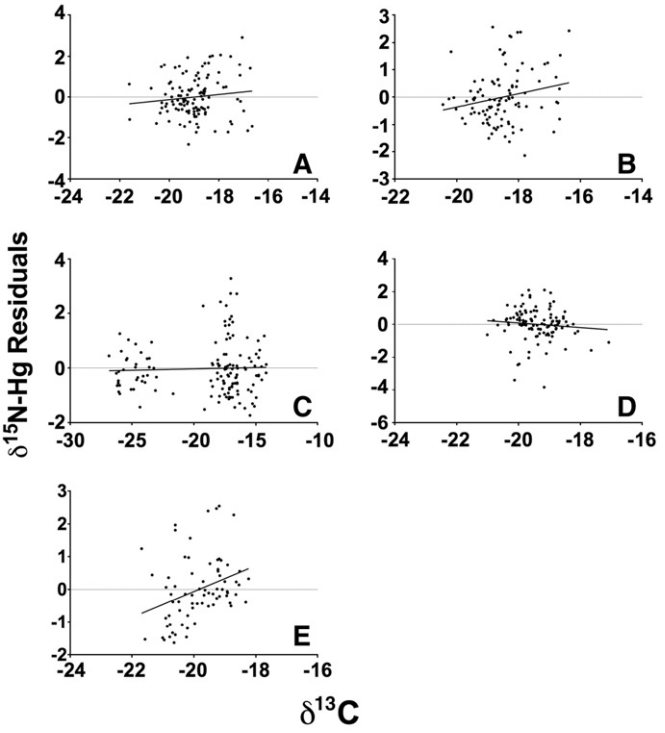

Fig. 2. Plots of the residuals from δ^{15}N-Hg regression and δ^{13}C indicating variation explained by carbon source in albumen (A), yolk (B), adult feathers (C), adult blood (D) and chick blood (E) from breeding seabird species from Machias Seal Island, 2005–2006.

We found that both δ^{15}N and δ^{13}C were correlated between yolk and albumen (δ^{13}C: $r=0.873$, δ^{15}N: $r=0.898$), but that yolk had a consistently lower δ^{15}N (paired t-test, $p<0.001$) and a lower δ^{13}C ($p<0.001$).

3.5. Prey

Prey model AIC_c values did not require correction using $\hat{c}$. The model including differences in Hg concentrations by species was the top-ranked model, although the model including species and year effects also received some support (Table 7). Of the prey species commonly observed, Northern Krill (*Meganyctiphanes norvegica*) had the greatest Hg concentrations, at concentrations similar for this species in the Mediterranean (Minganti et al., 1996). Atlantic herring (*Clupea harengus*), Hake/Four-bearded Rockling *(Urophycis tennuis, Merluccius bilinearis* and *Enchelyopus cimbrius,* White Hake, Silver Hake and Four-bearded Rockling, indistinguishable in the field) had similar Hg concentrations, although hake had significantly lesser Hg concentrations than euphausiids (Table 8). Pre-metamorphosis 0-group fish ("larval fish") consisting of one Atlantic Herring and two hake were pooled for this analysis.

Table 7
Summary of the model sets for seabird prey species Hg concentrations.

Model	No. parameters	AIC_c	ΔAIC_c	Akaike weights
S	4	170.452	0.000	0.34
S + Y	5	171.531	1.079	0.20
S + δ^{15}N	5	172.131	1.679	0.15
δ^{15}N	2	173.096	2.644	0.09
Y	2	173.420	2.968	0.08
S + Y + δ^{15}N	6	173.678	3.226	0.07
Null	1	174.078	3.626	0.06
S + Y + SY	8	176.987	6.535	0.01
Global	25	234.323	63.871	0.00

The Global Model includes all factors and their interactions; the Null Model is an intercept-only model. Models are sorted by increasing AIC_c (Akaike's information criteria corrected for small sample size) value with the most parsimonious model at the top. Akaike weight is the likelihood that a given model of the model set is the best approximation of the data. Model parameters abbreviations: S: species; Y: year.

Table 8
Estimated marginal mean (EMM) prey species Hg concentrations for breeding seabirds on Machias Seal Island from 2005–2006 were derived from the top-ranked generalized linear model.

Species*	n	Mean [Hg] (ppb, wet wt.)	95% Confidence limit		Group
			Lower	Upper	
Hake	32	13	9	18	A
Larval fish	3	13	4	42	AB
Atlantic herring	16	17	11	28	AB
Euphausiid shrimp	12	38	21	67	B

Differences were considered significant when confidence intervals do not overlap; species sharing the same Group letter are not significantly different from each other. Species with two letters are not significantly different from either group.

3.6. Comparisons with other sites

Calculated whole egg Hg concentrations are presented in Table 9. Significant differences in Hg concentrations in whole eggs between MSI and other sites are indicated in Table 10. Arctic Terns on MSI had significantly greater egg Hg concentrations than those on Petit Manan Island, Maine, 63 km to the southwest (paired t-test, $p=0.0096$), as did Common Eiders (paired t-test, $p=0.033$). Atlantic Puffin and Common Tern Hg concentrations did not differ between the two islands (paired t-test, $p=0.37$ and 0.18 respectively).

In 2005, Leach's Storm-petrels on MSI had greater egg Hg concentrations in whole eggs than those sampled from Kent Island in 2004 (paired t-test, $p=0.0095$) but samples from MSI in 2006 were not significantly different (paired t-test, $p=0.48$). Finally, Razorbills on MSI had significantly greater Hg concentrations in eggs than those on Matinicus Rock, Maine (paired t-test, $p=0.0002$). Additional global comparisons are presented in Table 10.

4. Discussion

4.1. Hg concentrations in seabird tissue

In general, Leach's Storm-petrels had among the greatest Hg concentrations, with the exception of blood. This could imply that Leach's Storm-petrels are more efficient at excreting mercury via feathers and eggs than other species. While storm-petrel diet in

Table 9
Mean Hg concentrations in whole eggs (parts per billion (ppb)) and conversions for dry to wet weights for seabirds from MSI in 2005 and 2006.

Species*	Year	n	Hg concentrations in eggs (ppb dry wt ± S.D.)	Hg concentrations in eggs (ppb wet wt. ± S.D.)
ARTE	2005	8	1030 ± 311	203 ± 49
ARTE	2006	9	1155 ± 312	236 ± 71
ATPU	2005	10	1048 ± 185	229 ± 61
ATPU	2006	9	1390 ± 3934	282 ± 79
COEI	2005	9	724 ± 454.	322 ± 180
COEI	2006	6	1243 ± 644	420 ± 339
COMU	2006	10	1574 ± 283	247 ± 61
COTE	2005	8	1016 ± 256.	126 ± 36
COTE	2006	5	797 ± 253	116 ± 32
LHSP	2005	8	4201 ± 1147	1070 ± 299
LHSP	2006	3	3222 ± 1302	703 ± 496
RAZO	2005	10	1771 ± 290	466 ± 168
RAZO	2006	10	2037 ± 623	455 ± 108

*Species abbreviations as in Table 2.
Whole egg Hg concentrations were calculated as the mean of albumen and yolk Hg concentrations weighted to their proportion of total egg mass without the shell and membrane.

Table 10
Summary of mean whole egg mercury concentrations taken from this study and the literature for seabirds breeding on MSI.

Species*	Year	Site	Hg (ppm) ± SD wet wt.	Source	*p* value
ARTE	2005	Machias Seal Island, NB	0.20 ± 0.05	This study	
ARTE	2006	Machias Seal Island, NB	0.24 ± 0.07	This study	
ARTE	1993	Petit Manan Island., ME	0.10 ± 0.02	Meirzykowski et al., 2001	0.0015
ATPU	2005	Machias Seal Island, NB	0.23 ± 0.06	This study	
ATPU	2006	Machias Seal Island, NB	0.28 ± 0.08	This study	
ATPU	1992	Machias Seal Island, NB	0.24 ± 0.02	N. Burgess, pers. comm.	
ATPU	1996	Machias Seal Island, NB	0.27 ± 0.03	N. Burgess, pers. comm.	
ATPU	2000	Machias Seal Island, NB	0.22 ± 0.02	N. Burgess, pers. comm.	
ATPU	2004	Machias Seal Island, NB	0.21 ± 0.02	N. Burgess, pers. comm.	
ATPU	2004	Petit Manan Island., ME	0.17	Merizykowski et al., 2005	
ATPU	1993	N. Norway	0.22 ± 0.01	Barrett et al., 1996	0.72
ATPU	1993	N. Norway	0.10 ± 0.02	Barrett et al., 1996	0.0005
ATPU	1993	N. Norway	0.06 ± 0.02	Barrett et al., 1996	0.0001
COMU	2006	Machias Seal Island, NB	0.25 ± 0.06	This study	
COMU	2000	Bogoslof, AK	0.03 ± 0.01	Day et al., 2006	0.0001
COMU	2001	St. Lazaria, AK	0.15 ± 0.04	Day et al., 2006	0.0004
COMU	1994	Farallones, CA	0.17 ± 0.06	Jarman et al., 1996	0.0055
COMU	1993	N. Norway	0.10 ± 0.04	Barrett et al., 1996	0.0001
COMU	1993	N. Norway	0.08 ± 0.01	Barrett et al., 1996	0.0001
COTE	2005	Machias Seal Island, NB	0.13 ± 0.04	This study	
COTE	2006	Machias Seal Island, NB	0.12 ± 0.03	This study	
COTE	2004	Stratton Island, ME	0.12 ± 0.02	Merizykowski et al., 2005	
COTE	2004	Jenny Island, ME	0.10 ± 0.03	Merizykowski et al., 2005	0.25
COTE	2004	Pond Island, ME	0.15 ± 0.04	Merizykowski et al., 2005	0.31
COTE	2004	Metinic Island, ME	0.10 ± 0.05	Merizykowski et al., 2005	0.43
COTE	2004	E. Egg Rock, ME	0.12 ± 0.02	Merizykowski et al., 2005	
COTE	2004	Petit Manan Island, ME	0.10 ± 0.02	Merizykowski et al., 2005	0.14
COTE	93–95	Azores	1.0 ± 0.2**	Monteiro et al., 1999	
COTE	93–95	Azores	1.5 ± 0.5**	Monteiro et al., 1999	
COTE	93–95	Azores	1.1 ± 0.5**	Monteiro et al., 1999	
COEI	2005	Machias Seal Island, NB	0.32 ± 0.18	This study	
COEI	2006	Machias Seal Island, NB	0.42 ± 0.34	This study	
COEI	2004	Metinic Island, ME	0.10 ± 0.03	Merizykowski et al., 2005	0.0370
COEI	2004	Petit Manan Island, ME	0.14 ± 0.04	Merizykowski et al., 2005	0.0332
COEI	1993	N. Norway	0.06 ± 0.02	Barrett et al., 1996	0.0082
LHSP	2005	Machias Seal Island, NB	1.07 ± 0.30	This study	
LHSP	2006	Machias Seal Island, NB	0.70 ± 0.50	This study	
LHSP	1992	Kent Island, NB	0.54 ± 0.03	N. Burgess, pers. comm.	0.61
LHSP	1996	Kent Island, NB	0.58 ± 0.07	N. Burgess, pers. comm.	0.70
LHSP	2000	Kent Island, NB	0.61 ± 0.05	N. Burgess, pers. comm.	0.77
LHSP	2004	Kent Island, NB	0.48 ± 0.05	N. Burgess, pers. comm.	0.48
RAZO	2005	Machias Seal Island, NB	0.47 ± 0.17	This study	
RAZO	2006	Machias Seal Island, NB	0.45 ± 0.11	This study	
RAZO	2005	Matinicus Rock, ME	0.14 ± 0.05	Goodale et al., in press	0.0001
RAZO	1993	N. Norway	0.18 ± 0.04	Barrett et al., 1996	0.0002

Results are given as parts per million on a wet weight basis unless otherwise indicated. *P* values are between the indicated study and the closest Hg value from MSI and are based on paired *t*-tests.
*Species abbreviations as Table 2.
**Dry weight.

eastern Canada is poorly known, they feed on a mixture of euphausiids and myctophid fishes (lanternfish) (Linton, 1979; Hedd and Montevecchi, 2006), and other mesopelagic fishes which are known to accumulate heavy metal toxins (Monteiro et al., 1996). Goodale et al. (in press) suggest that Leach's Storm-petrels would serve as useful bioindicators of global Hg trends because of their pelagic foraging habits and mesopelagic prey.

Both of the larger alcids (Common Murres and Razorbills) and Common Terns had the greatest blood Hg concentrations among adults and chicks. This suggests that these three species are feeding on prey of similar Hg concentrations or there are differences in the metabolic processes that remove Hg from the blood stream. Evidence from conventional diet studies (Minich, 2007) and stable isotope analysis (Bond, 2007) suggests that while there is some evidence of overlap among species on MSI, this is not consistent across years. Thus, neither of these hypotheses can be ruled out at present.

Overall, albumen Hg concentrations were 10 times higher than yolk Hg concentrations, and this was consistent for all species. For both egg components, Leach's Storm-petrels, followed by Razorbills and Common Murres, consistently had the greatest Hg concentrations, while eiders and terns were consistently less. Hg binds to disulfide bonds in protein, and because albumen has much greater protein content than yolk, higher Hg concentrations are expected in albumen.

Razorbills and murres dive deeper than puffins or terns (Piatt and Nettleship, 1987) and tend to eat larger, and therefore older, fish (Bond et al., 2007), which could have greater Hg concentrations than younger epipelagic fishes (Weiner and Spry, 1996). We did not, however, find any correlation between prey Hg concentration and length, fresh mass, dried mass, percent moisture or δ^{15}N (all $p > 0.07$). Prey δ^{15}N, however, did have a significantly positive correlation with length ($r = 0.32$, $p = 0.012$). This suggests a physiological difference between Razorbills and other sampled seabird species.

4.2. Comparison of Hg concentrations in eggs

Many studies examining egg Hg concentrations do so by analysing a homogenate of yolk and albumen. In order to make comparisons with our study, we reconstructed whole egg Hg concentrations using masses of yolk and albumen and their respective Hg concentrations (Table 9). Statistically, some MSI seabirds have greater Hg than

conspecifics in the larger Gulf of Maine region (Arctic Terns, Common Eiders, and Razorbills) while MSI seabird Hg concentrations are greater than those of conspecifics globally in Common Murres (whose distribution in the Gulf of Maine is restricted, Ainley et al., 2002), and some Atlantic Puffins. Whole egg Hg concentrations in some species are consistent across their range (Common Terns, Leach's Storm-petrels), while others species' whole egg Hg concentrations show significant differences. We believe this to be an effect of inadequate sampling across all species' ranges and our interpretation of seabird Hg concentrations would be greatly improved by additional information on Hg concentrations in seabird prey species. We have no reason to suspect that anthropogenic Hg input is greater at MSI than other sites in the Gulf of Maine, and so a detailed analysis of spatial Hg patterns in seabird tissues throughout the region would be required to fully understand differences in Hg concentrations in both whole eggs and their constituent tissues.

4.3. Biomagnification and Hg source

A variety of studies have found a strong positive relationship between δ^{15}N and Hg concentrations in marine mammals (Das et al., 2003; Braune et al., 2005) and seabirds (Jarman et al., 1996; Atwell et al., 1998; Bearhop et al., 2000b), yet other seabird studies have found no significant relationship (Bearhop et al., 2000a; Nisbet et al., 2002). Our study found a significantly positive, but weak, relationship between δ^{15}N and Hg concentrations in adult blood and yolk, and a stronger δ^{15}N-Hg relationship in adult feathers and albumen. In seabirds, Hg is accumulated and stored in internal tissues as a "body pool" and excreted during feather replacement or egg laying (Braune and Gaskin, 1987b). Hg in feathers can therefore represent a much longer accumulation period than stable isotopes, as isotopes are incorporated into feathers and only represent the diet during growth (Hobson and Clark, 1992). Feather δ^{15}N and Hg values will therefore represent fundamentally different periods of incorporation, and there is little biological meaning to any correlation (Thompson et al., 1998; Bearhop et al., 2000a).

A relationship between δ^{15}N and Hg concentration is expected for both egg components (albumen and yolk) in six of the species considered here, since they are income breeders, using locally-derived nutrients for egg production. Since locally-derived nutrients are incorporated into egg components, Hg concentrations in albumen and yolk likely represent local contaminant concentrations. A similar positive relationship between δ^{15}N and Hg concentration is expected for whole blood since δ^{15}N values integrate diet over the previous 2–3 weeks (Hobson and Clark, 1993), and Hg is mobilized in the blood prior to storage or detoxification (Kahle and Becker, 1999).

In yolk and chick blood, the regression of δ^{13}C on residuals from the δ^{15}N-Hg regression was significant and in both cases, the relationship was positive, meaning that more inshore sources had greater Hg concentrations when trophic position was accounted for. This is in contrast to seabirds from the Aleutian Islands, Alaska, where the opposite was true — offshore δ^{13}C values were related to greater Hg concentrations once trophic position was considered (Ricca et al., 2008). Euphausiid shrimp δ^{13}C ratios were not significantly different from other common fish prey (Bond, 2007), and so would not account for this effect. In both cases, the amount of variation explained by δ^{15}N and δ^{13}C was small (combined r^2 for yolk: 0.12, for chick blood: 0.13), indicating that physiological or other factors play a much larger role than trophic position or carbon source in determining Hg concentrations.

Chick blood had the lowest biomagnification factor (BMF), likely because most Hg was being deposited into the growing feathers (Evers et al., 2005). Adult feathers had the highest BMF, but the amount of Hg is not necessarily related to diet (and hence δ^{15}N), but rather to the timing and duration of moult (Stewart et al., 1999; Bearhop et al., 2000a). Moult duration, the period of time during which feathers present during the breeding season were grown, can range from 30–274 days (Ainley et al., 1976; Harris and Yule, 1977; Harris and Wanless, 1990; Voelker, 1997; Gaston and Jones, 1998; Nisbet, 2002; Spear and Ainley, 2007; Pyle and Howell, 2008), but in many cases, these are rough approximations; there is little accurate information about the duration of moult in seabirds. Adult blood and egg component BMFs are more biologically meaningful as they represent integration of both diet and Hg over a shorter period and incorporate methyl mercury prior to detoxification. Our BMF values for blood (0.22) and yolk (0.45) are similar to those found in other marine predators (Campbell et al., 2005; Ricca et al., 2008), while that of albumen was much higher (1.08). Hg preferentially binds to albumen proteins (Magat and Sell, 1979), so greater Hg concentrations are expected over other tissues with lower protein content (i.e., yolk or whole blood). We found that albumen Hg concentrations were generally greater than yolk Hg concentrations by a factor of 10, and since both tissues integrate diet information over short periods (Hobson and Clark, 1992), they are expected to have similar stable isotope values, which we did not find. We believe the differences between yolk and albumen in δ^{13}C and could be because lipids were extracted from yolk and not albumen. Lipid extraction can affect δ^{15}N values (e.g., Ricca et al., 2007) and further study of the effect of chemical extraction on yolk δ^{15}N is required. Differences in δ^{15}N could be reflective of fine-scale temporal variation in diet during the time of egg formation. Seabirds are known to exploit temporally variable food sources (e.g., Cheng and Harrison, 1983; Huettmann et al., 2005; Bond et al., 2007, in press). Further information on the temporal variability of Hg concentrations in albumen, and yolk are required to determine which, if either, would make a suitable long-term indicator of Hg in seabirds.

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