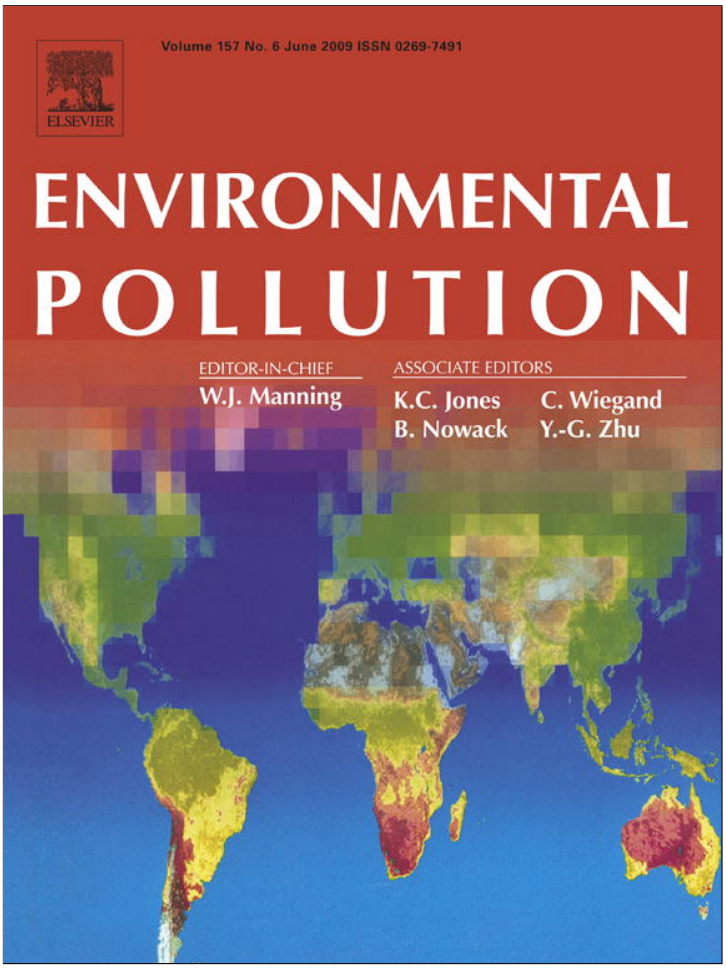

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Relationships between hepatic trace element concentrations, reproductive status, and body condition of female greater scaup

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Some female greater scaup initiate nesting with elevated hepatic concentrations of some trace elements, but adverse effects on condition and productivity are unlikely.

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ABSTRACT

We collected female greater scaup (*Aythya marila*) on the Yukon–Kuskokwim Delta, Alaska during two breeding seasons to determine if concentrations of 18 trace elements in livers and eggs were elevated and if hepatic concentrations correlated with body condition or affected reproductive status. Fifty-six percent, 5%, and 42% of females, respectively, had elevated hepatic cadmium (Cd: $>3\ \mu g\ g^{-1}$ dry weight [dw]), mercury (Hg: $>3\ \mu g\ g^{-1}$ dw), and selenium (Se: $>10\ \mu g\ g^{-1}$ dw). Somatic protein and lipid reserves were not correlated with hepatic Cd or Hg, but there was a weak negative correlation between protein and Se. Hepatic Cd, Hg, and Se were similar in females that had and had not initiated egg production. In a sample of six eggs, 33% and 100%, respectively, contained Se and Hg, but concentrations were below embryotoxicity thresholds. We conclude that trace element concentrations documented likely were not adversely impacting this study population.

1. Introduction

The long-term population decline of scaup (lesser [*Aythya affinis*] and greater [*Aythya marila*] scaup combined) in North America has raised concerns about effects of trace element acquisition on population dynamics (Austin et al., 2000; Afton and Anderson, 2001). The contaminant hypothesis suggests that acquired contaminants can adversely impact scaup productivity or survival and may have contributed to the long-term decline or are inhibiting population increase (Custer and Custer, 2000). Many studies have found that both lesser and greater scaup acquire and depart from major winter areas (Ohlendorf et al., 1986; Hoffman et al., 1998; Hothem et al., 1998; Cohen et al., 2000; Custer et al., 2000; Takekawa et al., 2002; Ware, 2008) and spring stopover sites (Custer and Custer, 2000; Custer et al., 2003; Anteau et al., 2007; Petrie et al., 2007) with elevated levels of several trace elements, such as mercury (Hg), cadmium (Cd), and selenium (Se).

Selenium in particular was deemed a trace element of concern for greater scaup after increasing and large numbers of these birds began to use the lower Great Lakes (LGL) during fall, winter, and spring in response to the establishment of zebra mussels (*Dreissena polymorpha*) that occurred during the late 1980s/early 1990s (Mitchell and Carlson, 1993; Custer and Custer, 1996, 2000). Dreissenid mussels have since become a major scaup prey item on the LGL and one that bioaccumulates Se (Badzinski and Petrie, 2006; Ware, 2008). Substantial numbers of greater scaup may thus acquire and depart the LGL for breeding areas with potentially problematic levels of Se (Petrie et al., 2007). Despite several studies of contaminants in non-breeding scaup, relatively little is known about Se and other trace element burdens in breeding females, particularly greater scaup, or how they might impact body condition or reproductive potential.

Lipid and protein reserves acquired prior to breeding can affect survival and reproductive performance in waterfowl (Alisauskas and Ankney, 1992), including scaup (Afton and Ankney, 1991; Esler et al., 2001; Gorman et al., 2008), and thus are linked to population dynamics. Studies of captive and wild waterbirds show increased liver or renal tissue concentrations of some trace elements can affect body condition (i.e., fat and protein reserves) (Hoffman et al., 1992; Scheuhammer et al., 1998; Eisler, 2000a,b; Wayland et al., 2002).

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Lipid reserves of spring-migrant lesser scaup, for example, decreased with increasing hepatic Cd burdens (Anteau et al., 2007), whereas those of wintering scaup species decreased with increasing hepatic zinc (Zn) concentrations (Takekawa et al., 2002). However, a recent study of greater scaup wintering on the LGL, where 99% of birds had elevated hepatic Se concentrations, found no Se-related effects on indices of oxidative stress, lipid or protein reserves (Ware, 2008). Similarly, DeVink et al. (2008b) found lipid and protein reserves were not correlated with hepatic Se burdens in lesser scaup collected from boreal breeding areas. No study, however, has evaluated links between trace element concentrations and body condition of female greater scaup during the breeding period.

The majority of greater scaup in North America breed within tundra habitats of western Alaska (Hodges et al., 1996). Although scaup nesting within the western boreal forest (mainly lesser scaup) have generally declined over the past three decades, long-term surveys show no decline in scaup (mainly greater scaup) nesting within tundra habitat of western Alaska (Afton and Anderson, 2001). Greater scaup nesting on the Yukon–Kuskokwim Delta (YKD), a major breeding area, have higher survival and productivity as compared to most boreal-nesting scaup (Flint et al., 2006). Long-term studies on the breeding ecology of YKD greater scaup showed no evidence of abnormally low survival nor any indications of reproductive failure (e.g., egg infertility) often associated with contaminant exposure (Flint et al., 2006). Contaminant burdens in females breeding on the YKD are thus likely below levels causing adverse effects on fitness. These earlier studies, however, did not evaluate links between trace element burdens and breeding propensity (probability of a sexually mature bird breeding in a given year) of greater scaup.

In this study, we examine the contaminant hypothesis by assessing trace element concentrations in female greater scaup throughout clutch formation. We evaluate maternal transfer of trace elements, specifically Hg and Se, to eggs. Finally, we test two predictions derived from the contaminants hypothesis: 1) strong evidence of negative correlations between liver concentrations of Cd, Hg, or Se and somatic fat and protein reserves and 2) females without developing ovarian follicles (non-developed) contain higher, and possibly elevated, hepatic concentrations of these three trace elements as compared to birds that initiated egg production via rapid follicle growth (RFG).

2. Materials and methods

2.1. Specimen collections

This study was conducted under permits from the U.S. Fish and Wildlife Service, the State of Alaska Department of Fish and Game, and the Yukon Delta National Wildlife Refuge and conformed to guidelines of the Canadian Committee on Animal Care (Simon Fraser University [SFU] Animal Care Permit 637B-02). Approximately two females were collected with a rifle or shotgun each day from 19 May to 21 June 2002 ($n = 58$), and 15 May to 19 June 2003 ($n = 54$) near the lower Kashunuk River (60°20′N, 165°35′W) on the YKD, Alaska. We also collected entire clutches of eggs from nests belonging to six collected females. Within 24 h of collection, reproductive tissues were dissected from carcasses. Carcasses and tissues were stored frozen in the field and later transported frozen to a laboratory at SFU, British Columbia, where they were stored at −20 °C until body composition analyses were conducted. For details on collections and carcass processing see Gorman et al. (2007, 2008).

2.2. Reproductive state, body size, and somatic nutrient reserve determinations

In the laboratory, carcasses were thawed and classified, based on ovarian follicle characteristics (presence, number, size, and mass) as described in Gorman et al. (2008), into the following reproductive groups: non-developed, RFG, laying, incubation, and renesting. Structural measurements (±0.1 mm using Vernier calipers) were made on each carcass, including right wing chord length, right tarsus length, culmen, bill width, bill nail width, bill nail length, and keel length. Measurements were used to quantify body size variation among females and control for possible size-related variation in somatic nutrient reserves in analyses (Sedinger et al., 1997).

Somatic nutrient (lipid, protein, and mineral) reserves of females were determined using standard proximate body composition analyses conducted as described in Gorman et al. (2008). In general, dry, homogenized carcass samples (±0.1 g) were placed in a modified Soxhlet apparatus and washed with petroleum ether to remove lipids then samples were dried to constant mass to determine lipid (g) content (Dobush et al., 1985; Ankney and Afton, 1988). Lean-dry carcass samples were then combusted in a muffle furnace to determine mineral (g of ash) and protein (g of ash-free, lean-dry tissue) content.

2.3. Trace element determinations

During dissections, samples (3 g) of liver tissue were excised from the right caudal lobe from each bird. Separate samples (5 g) of yolk and albumen were obtained from one unincubated egg chosen at random from each collected clutch ($n = 6$). Liver samples were wrapped in hexane rinsed foil, placed into a hexane rinsed glass vial and stored frozen. Wet yolk and albumen samples were placed in separate small plastic bags and stored frozen. Samples remained frozen until 2005 (2–3 years) when trace element analyses were performed at the Great Lakes Institute of Environmental Research in Windsor, Ontario.

All samples were analyzed for aluminum (Al), arsenic (As), calcium (Ca), Cd, cobalt (Co), chromium (Cr), copper (Cu), iron (Fe), potassium (K), magnesium (Mg), manganese (Mn), sodium (Na), nickel (Ni), lead (Pb), Se, vanadium (V), Zn, and total Hg according to Canadian Association of Environmental Analytical Laboratories-accredited standard operating procedures (Environment Canada, 1989); analytical methods are described in Mallory et al. (2004). The QA/QC protocols involved analyzing three method blanks (purified water), three certified biological reference tissues (DORM-2, LUTS-1, and DOLT-2; National Research Council, Canada), and sample blanks (2 randomly selected duplicates per batch) for each batch of 36 samples analyzed. Mean percent recoveries (range: 86.6–104.3%) of certified standards were within tolerance ranges for all trace elements except for Cr, which yielded a mean recovery of 241 ± 105%. Elemental concentrations were not corrected for percent recoveries. Method detection limits ($\mu g\, g^{-1}$ dry weight [dw]) were as follows: Al (4.52), As (1.32), Ca (2.04), Cd (0.06), Co (0.29), Cr (0.23), Cu (0.73), Fe (1.01), K (14.5), Mg (0.63), Mn (0.03), Na (3.76), Ni (0.25), Pb (0.51), Se (1.74), V (0.70), Zn (0.40), and Hg (0.01). Total egg trace element concentrations were estimated using data in Flint and Grand (1999). For use in wet mass conversions, water content of liver, egg albumen, and egg yolk averaged 68%, 69%, and 54%, respectively.

2.4. Statistical analyses

We identified analytes that had ≥50% detection rates and replaced non-detections with values half the detection limit. Data were log transformed to normalize error distributions. We used Multivariate Analysis of Variance (MANOVA) to test for differences in hepatic trace elements concentrations between years and among groups (PROC GLM, MANOVA option, SAS Institute, 2004). We used Wilks' λ as our test statistic and considered overall effects significant at $P \leq 0.05$. We made post hoc comparisons of means using Tukey–Kramer (T–K) tests. This analysis was made to provide summary statistics, which included geometric mean (GM), 95% confidence interval (CI), and range. We calculated the same summary statistics for trace element concentrations in yolk and albumen.

We conducted a principal components analysis on the correlation matrix of wing chord, tarsus, culmen, bill width, bill nail width, bill nail length, and keel length of non-developed and RFG females (PROC PRINCOMP, SAS Institute, 2004). The first principal component (PC1) explained 26% of variation in structural measurements. All eigenvector values were positive indicating PC1 was indexing bird body size. We regressed PC1 scores against somatic lipid and protein reserves to obtain residuals for each nutrient. We used these lipid and protein residuals (i.e., body size corrected values) in analyses of hepatic trace element concentrations.

We used a two-stage approach to develop and select best approximating models evaluating variation in hepatic concentrations of Se, Hg, and Cd, and to control for possible confounding effects, using data from the 64 females classified as either non-developed or RFG. We used Akaike's Information Criterion corrected for small sample sizes (AIC_c) to evaluate support for candidate models that included planned combinations of possible covariates and primary factors of interest. We obtained AIC_c values for all competing models in each stage of the model selection process (PROC MIXED, IC option, Method = ML, SAS Institute, 2004) and used those to rank (lowest to highest) models and calculate model weights (Burnham and Anderson, 2002).

For the first stage of the model selection process, we developed a set of candidate models to assess the importance of year (2002 vs. 2003) and date effects on hepatic trace element concentrations. We also considered liver dw as a covariate given that variation in size/mass of this organ can affect total trace element burdens within individuals (Rattner and Jehl, 1997). Because Se concentrations can have a moderating influence on Hg concentrations (Heinz, 1996), we included Se and Hg concentrations (log transformed) as covariates in appropriate analyses. We also included an intercept-only (null) model. These models were not designed as direct examinations of a priori hypotheses, but used to identify important covariates for subsequent analyses, thereby minimizing the number of models considered (Burnham and Anderson, 2002).

During the second stage of model selection, our main interest was to evaluate variation in hepatic Se, Hg, and Cd concentrations due to female reproductive status and nutrient reserves after controlling for sources of variation identified in stage one. We developed a set of models that included previously identified covariates, plus all combinations of the variables: lipid reserve (residuals), protein reserve (residuals), and female reproductive status (RFG vs. non-developed). We used model weights from this stage of model selection to calculate model-averaged parameter estimates (based on models ≤ 2 AIC_c), least squares (LS) means, standard errors (SE), or 95% confidence limits for effects of interest (Burnham and Anderson, 2002).

All statistical analyses were made using the SAS analytical software (SAS Institute, 2004). Geometric means are reported with 95% CI and parameter estimates (β) and LS means are reported ± 1 SE. Log-transformed concentrations were back-transformed for presentation in tables. Unless otherwise stated, all trace element concentrations are reported as $\mu g\, g^{-1}$ dw liver.

3. Results

3.1. Hepatic trace element concentrations

Arsenic (% non-detection [ND] = 100%), Co (ND = 100%), Pb (ND = 98%, Max = 2.58 $\mu g\, g^{-1}$), Ni (ND = 87%, Max = 0.84 $\mu g\, g^{-1}$), and V (ND = 91%, Max = 1.35 $\mu g\, g^{-1}$) concentrations were detected infrequently or were at instrument detection limits, and thus not included in statistical analyses. Al, Cd, Ca, Cu, Cr, Fe, K, Mg, Mn, Hg, Se, Na, and Zn were detected in 100% of liver tissues.

Hepatic concentrations of several trace elements differed between years (MANOVA: Wilks' $\lambda = 0.30$, $P < 0.001$) and among reproductive groups (MANOVA: Wilks' $\lambda = 0.21$, $P < 0.001$). Tukey–Kramer tests indicated concentration of Al was higher in 2002 (GM = 350, CI = 280–437, range [R] = 64–7660 $\mu g\, g^{-1}$) than in 2003 (GM = 151, CI = 118–193, R = 70–305 $\mu g\, g^{-1}$), whereas K (2002: GM = 8320, CI = 8050–8590, R = 6570–10,900 $\mu g\, g^{-1}$ vs. 2003: 10,500, CI = 10,100–10,900, R = 8530–12,400 $\mu g\, g^{-1}$) and Na (2002: GM = 3380, CI = 3250–3520, R = 2450–4540 $\mu g\, g^{-1}$ vs. 2003: GM = 3690, CI = 3530–3850, R = 2550–4610 $\mu g\, g^{-1}$) were higher in 2003 as compared to 2002 ($P < 0.05$ in all pair-wise comparisons). Concentrations of Ca, Cd, Cu, Cr, Fe, K, Mg, Mn, Na, Hg, Se, and Zn differed among reproductive groups (Table 1).

Hepatic concentrations of Cd (>3 $\mu g\, g^{-1}$, Scheuhammer, 1987), Hg (>3 $\mu g\, g^{-1}$, Thompson, 1996), and Se (>10 $\mu g\, g^{-1}$, Heinz, 1996) were detected at levels in some individuals that are considered elevated, but average concentrations in most reproductive groups did not exceed those thresholds (Table 1). Overall, 56% and 42% of all individuals had elevated concentrations of Cd and Se, whereas only 5% (5 of 108) had elevated Hg concentrations; percentages of females with elevated levels of these trace elements also varied among groups (Table 1).

Table 1
Geometric mean concentrations, 95% confidence limits, ranges ($\mu g\, g^{-1}$ dw) of trace elements in liver tissues of female greater scaup (*Aythya marila*), plus percentages and numbers of birds with elevated concentrations of cadmium (>3 $\mu g\, g^{-1}$ dw), mercury (>3 $\mu g\, g^{-1}$ dw), and selenium (>10 $\mu g\, g^{-1}$ dw) collected during 2002 and 2003 breeding seasons at the Yukon–Kuskokwim Delta, Alaska.

Element	Reproductive group[a]				
	Non-developed ($n = 32$)	RFG[b] ($n = 32$)	Egg lay ($n = 30$)	Incubation ($n = 10$)	Renest ($n = 4$)
Calcium	329a 292–371 188–1030	301a 267–340 179–630	450b 398–509 257–1320	377ab 303–469 300–521	396ab 284–553 268–710
Cadmium[c]	2.96a 2.52–3.47 0.81–7.64 56% (18)	3.30a 2.82–3.86 1.43–9.41 59% (19)	2.74a 2.33–3.22 1.07–9.75 43% (13)	4.47b 3.35–5.97 2.25–7.74 90% (9)	2.40a 1.54–3.74 1.64–4.30 25% (1)
Chromium	1.10a 1.03–1.18 0.64–1.57	1.07a 1.00–1.14 0.68–1.34	1.24b 1.15–1.33 0.74–1.77	0.99a 0.88–1.12 0.67–1.30	1.04ab 0.86–1.26 0.86–1.25
Copper	58.9a 49.8–69.7 17.9–121	52.5a 44.2–61.6 18.1–133	32.1b 27.0–38.1 12.7–62.0	48.3ac 35.6–65.7 11.3–134	28.7bc 18.0–45.9 21.1–42.2
Iron	3240a 2730–3830 1170–10,900	3090a 2620–3650 1400–7110	2690a 2270–3200 1130–5910	4930b 3630–6690 2110–8660	3260ab 2040–5210 1990–5510
Potassium	9140a 8810–9480 6730–12,400	8750a 8440–9070 6570–11,300	9740b 9390–10,100 7150–11,100	9310ab 8710–9940 7440–10,200	9680ab 8750–10,700 8610–10,300
Magnesium	834a 806–862 711–1040	804a 778–831 664–977	911b 880–943 717–1170	821a 772–873 707–952	888ab 808–975 809–1030
Manganese	18.4a 16.4–20.6 9.84–27.7	19.4a 17.3–21.7 12.1–28.8	23.9bc 21.3–26.9 14.2–82.0	22.2ab 18.1–27.3 17.6–54.8	32.8c 24.0–45.0 16.3–77.9
Sodium	3630ab 3470–3790 2780–4610	3470a 3320–3620 2480–4230	3430a 3280–3590 2450–4540	3920b 3610–4250 2930–4300	3240a 2860–3670 2790–3540
Mercury[c]	1.00a 0.77–1.29 0.18–3.86 9% (3)	0.92a 0.72–1.19 0.17–3.21 6% (2)	0.54b 0.42–0.70 0.16–1.45 0% (0)	0.77ab 0.48–1.23 0.35–1.60 0% (0)	0.48ab 0.24–0.99 0.32–0.99 0% (0)
Selenium[c]	9.74ab 8.52–11.1 4.85–26.5 34% (11)	11.1b 9.76–12.7 4.13–25.8 63% (20)	9.00a 7.85–10.3 4.33–18.1 37% (11)	7.63a 5.99–9.73 4.11–10.6 20% (2)	7.91ab 5.45–11.5 6.17–10.9 25% (1)
Zinc	133a 124–142 105–164	117b 110–125 78.9–189	106c 99.1–114 73.8–223	144a 127–162 99.5–235	94.8c 78.8–114 84.7–106

[a] Different letters denote differences ($P \leq 0.05$) among reproductive groups.
[b] Rapid follicle growth initiated.
[c] Sources for elevated criteria: cadmium (Scheuhammer, 1987), mercury (Thompson, 1996), and selenium (Heinz, 1996).

3.2. Sources of variation in hepatic trace elements of RFG and non-developed females

3.2.1. Cadmium

The intercept-only model was identified as the best approximating model during the first stage of model selection. While the top model was 2.4 times better supported by the data than the second ranked model which consisted of only the date effect (Table 2), we chose to include the date covariate in subsequent models.

Within the second stage of model selection, the intercept-only ($\beta_0 = 0.4936 \pm 0.0254$) and date effect ($\beta = 0.0023 \pm 0.0034$) models were ranked as the best and second-best approximating models, respectively (Table 2). Date, however, was not deemed a major correlate because it only explained about 1% of the variation in hepatic Cd concentrations. Notably, we also found no support for effects of lipid reserve, protein reserve, and female status on Cd concentrations; addition of those factors in models did little to increase explained variation (2–3%).

3.2.2. Mercury

The first stage of model selection identified three top ranking models (all ≤ 2.0 AIC_c) that explained variation in hepatic Hg concentrations of females. The top-ranked model contained year, Se, and liver effects (Table 3). The second and third ranked models both contained the liver effect plus one additional effect each including Se and year, respectively. We included all three covariates in the second stage of model selection.

Within the second stage of model selection, the top-ranked model only contained the three variables (year, Se, and liver) identified in the first phase of model selection. The top-ranked model was 1.8 times better supported by the data than the second ranked model, which also contained the lipid reserve effect (Table 3). Support for a lipid reserve effect was not strong because adding that

Table 2
Number of parameters (K), Akaike's Information Criterion values adjusted for small sample size (AIC_c), differences in AIC_c value from top approximating model (ΔAIC_c), and model weights (w_i) used to rank models containing factors hypothesized to correlate with liver cadmium concentrations ($\mu g\,g^{-1}$ dw; log transformed) in female greater scaup collected during late May through mid June 2002 and 2003 at the Yukon–Kuskokwim Delta, Alaska.

Candidate model[a]	K	AIC_c	ΔAIC_c	w_i
Stage 1 – initial covariate selection				
β_0	2	−18.91	0.00	0.40
$\beta_0+\beta_1$(DATE)	3	−17.14	1.76	0.17
$\beta_0+\beta_1$(LIV)	3	−16.71	2.19	0.13
$\beta_0+\beta_1$(YR)	3	−16.71	2.20	0.13
$\beta_0+\beta_1$(DATE)+β_2(LIV)	4	−14.89	4.02	0.05
$\beta_0+\beta_1$(DATE)+β_2(YR)	4	−14.87	4.04	0.05
$\beta_0+\beta_1$(LIV)+β_2(YR)	4	−14.45	4.46	0.04
$\beta_0+\beta_1$(DATE)+β_2(LIV)+β_3(YR)	5	−12.53	6.38	0.02
Stage 2 – assess status and nutrient reserve effects				
β_0	2	−18.91	0.00	0.42
$\beta_0+\beta_1$(DATE)	3	−17.14	1.76	0.18
$\beta_0+\beta_1$(DATE)+β_2(LIP)	4	−16.56	2.35	0.13
$\beta_0+\beta_1$(DATE)+β_2(PRO)	4	−16.06	2.85	0.10
$\beta_0+\beta_1$(DATE)+β_2(STAT)	4	−15.48	3.42	0.08
$\beta_0+\beta_1$(DATE)+β_2(LIP)+β_3(STAT)	5	−14.23	4.67	0.04
$\beta_0+\beta_1$(DATE)+β_2(PRO)+β_3(STAT)	5	−13.98	4.93	0.04
$\beta_0+\beta_1$(DATE)+β_2(LIP)+β_3(PRO)+β_4(STAT)	6	−12.14	6.77	0.01

[a] DATE (Julian date on which female was collected); LIV (liver dw [g]); YR (class variable denoting females collected in year 2002 or 2003); STAT (class variable denoting female status as non-developed [rapid follicle growth not initiated] or RFG [rapid follicle growth initiated]); PRO (protein reserves corrected for variation due to body size [residuals]); LIP (lipid reserves corrected for variation due to body size [residuals]).

Table 3
Number of parameters (K), Akaike's Information Criterion values adjusted for small sample size (AIC_c), differences in AIC_c value from top approximating model (ΔAIC_c), and model weights (w_i) used to rank models containing factors hypothesized to correlate with liver mercury concentrations ($\mu g\,g^{-1}$ dw; log transformed) in female greater scaup collected during late May through mid June 2002 and 2003 at the Yukon–Kuskokwim Delta, Alaska.

Candidate model[a]	K	AIC_c	ΔAIC_c	w_i
Stage 1 – initial covariate selection				
$\beta_0+\beta_1$(Se)+β_2(LIV)+β_3(YR)	5	49.11	0.00	0.29
$\beta_0+\beta_1$(Se)+β_2(LIV)	4	50.03	0.92	0.19
$\beta_0+\beta_1$(LIV)+β_2(YR)	4	50.67	1.56	0.14
$\beta_0+\beta_1$(Se)+β_2(LIV)+β_3(YR)+β_4(DATE)	6	51.27	2.16	0.10
$\beta_0+\beta_1$(LIV)	3	51.87	2.76	0.07
$\beta_0+\beta_1$(Se)+β_2(LIV)+β_3(DATE)	5	52.35	3.25	0.06
$\beta_0+\beta_1$(LIV)+β_2(YR)+β_3(DATE)	5	52.81	3.70	0.05
$\beta_0+\beta_1$(Se)	3	53.78	4.67	0.03
$\beta_0+\beta_1$(LIV)+β_2(DATE)	4	54.14	5.03	0.02
$\beta_0+\beta_1$(Se)+β_2(YR)	4	54.72	5.61	0.02
$\beta_0+\beta_1$(Se)+β_2(DATE)	4	55.92	6.81	0.01
β_0	2	56.24	7.13	0.01
$\beta_0+\beta_1$(Se)+β_2(YR)+β_3(DATE)	5	56.70	7.59	0.01
$\beta_0+\beta_1$(YR)	3	57.02	7.91	0.01
$\beta_0+\beta_1$(DATE)	3	58.35	9.24	0.00
$\beta_0+\beta_1$(YR)+β_2(DATE)	4	59.00	9.89	0.00
Stage 2 – assess status and nutrient reserve effects				
$\beta_0+\beta_1$(Se)+β_2(LIV)+β_3(YR)	5	49.11	0.00	0.38
$\beta_0+\beta_1$(Se)+β_2(LIV)+β_3(YR)+β_4(LIP)	6	50.31	1.20	0.21
$\beta_0+\beta_1$(Se)+β_2(LIV)+β_3(YR)+β_4(PRO)	6	51.41	2.30	0.12
$\beta_0+\beta_1$(Se)+β_2(LIV)+β_3(YR)+β_4(STAT)	6	51.50	2.39	0.11
$\beta_0+\beta_1$(Se)+β_2(LIV)+β_3(YR)+β_4(LIP)+β_5(STAT)	7	52.73	3.62	0.06
$\beta_0+\beta_1$(Se)+β_2(LIV)+β_3(YR)+β_4(LIP)+β_5(PRO)	7	52.83	3.72	0.06
$\beta_0+\beta_1$(Se)+β_2(LIV)+β_3(YR)+β_4(PRO)+β_5(STAT)	7	53.91	4.80	0.03
$\beta_0+\beta_1$(Se)+β_2(LIV)+β_3(YR)+β_4(LIP)+β_5(PRO)+β_6(STAT)	8	55.35	6.24	0.02
β_0	2	56.24	7.13	0.01

[a] Se (log transformed liver selenium concentrations [$\mu g\,g^{-1}$ dw]); DATE (Julian date on which female was collected); LIV (liver dw [g]); YR (class variable denoting females collected in year 2002 or 2003); STAT (class variable denoting female status as non-developed [rapid follicle growth not initiated] or RFG [rapid follicle growth initiated]); PRO (protein reserves corrected for variation due to body size [residuals]); LIP (lipid reserves corrected for variation due to body size [residuals]).

effect to the covariates-only model explained only about 1% more variation in Hg concentrations.

Model-averaged parameter estimates indicated positive correlation ($\beta = 0.4571 \pm 0.2255$) between hepatic concentrations of Se and Hg. Liver dw decreased with increasing Hg concentrations in female greater scaup ($\beta = -0.0884 \pm 0.0335$). Model-averaged Hg concentration (LS mean, lower–upper 95% CL) was slightly higher in 2002 (1.18, 0.88–1.57 $\mu g\,g^{-1}$) than in 2003 (0.82, 0.64–1.04 $\mu g\,g^{-1}$); although 95% CIs did overlap slightly. Lipid reserves tended to decrease with increasing Hg concentrations ($\beta = -0.0014 \pm 0.0013$), but the SE for this estimate was large.

3.2.3. Selenium

Within the first stage of the model selection process, the highest ranked model for variation in hepatic Se concentrations ($w_i = 0.33$) contained only the Hg effect (Table 4). Further, Hg was included in the next three highest ranking models, all of which were >2 AIC_c from the best approximating model, plus the intercept-only model was not in the probable best set of models (i.e., $\Delta AIC_c > 2$). As a result, Hg was included as a covariate in all subsequent candidate models evaluating nutrient reserve and female status effects.

Within the second stage of model selection, the highest ranked model again contained only the Hg effect, but female status and protein reserve effects (plus Hg) also were included in the set of probable models (Table 4). Thus, female status and protein reserve effects also were supported by these data.

Model-averaged parameter estimates indicated positive correlation between hepatic concentrations of Hg and Se ($\beta = 0.1268 \pm 0.0604$), plus a slight negative correlation between protein residuals of female greater scaup and Se concentrations ($\beta = -0.0030 \pm 0.0024$). Females that initiated RFG had 11.3 $\mu g\,g^{-1}$ (95% CI: 9.82–12.9 $\mu g\,g^{-1}$), whereas non-developed females had 9.59 $\mu g\,g^{-1}$ (8.36–11.0 $\mu g\,g^{-1}$) Se in liver tissue.

3.3. Trace element concentrations in egg constituents

In both dry egg yolk and albumen, concentrations of Al, As, Cd, Co, Ni, Pb, and V were below detection limits in 100% ($n = 6$) of samples. Ca, Co, Fe, K, Mg, Mn, Hg, Na, and Zn were detected in 100% of albumen and yolk samples (Table 5). Concentrations of Cr were below detection limits in 5 of 6 albumen samples, but detected in 100% of yolk samples (GM = 0.70 $\mu g\,g^{-1}$). Se was not detected in yolk samples, but was detected in 2 of 6 (33%) albumen samples at 3.18 and 3.45 $\mu g\,g^{-1}$.

4. Discussion

Several trace elements that can be problematic for avian species, including Ar, Co, Cr, Ni, Pb, and V, either were at background levels (Eisler, 2000a,b) or were not detected in liver tissues of female greater scaup. The only trace elements that were consistently detected above background concentrations in greater scaup liver tissues were Cd, Hg, and Se. We thus conclude that all other trace elements considered in our analyses would likely have no effect on our study population.

4.1. Cadmium

Hepatic Cd concentrations in 56% of greater scaup in this study were above the 3 $\mu g\,g^{-1}$ background level proposed by Scheuhammer (1987) for freshwater duck species. All Cd concentrations

Table 4
Number of parameters (K), Akaike's Information Criterion values adjusted for small sample size (AIC_c), differences in AIC_c value from top approximating model (ΔAIC_c), and model weights (w_i) used to rank models containing factors hypothesized to correlate with liver selenium concentrations ($\mu g\,g^{-1}$ dw; log transformed) in female greater scaup collected during late May through mid June 2002 and 2003 at the Yukon–Kuskokwim Delta, Alaska.

Candidate model[a]	K	AIC_c	ΔAIC_c	w_i
Stage 1 – initial covariate selection				
$\beta_0+\beta_1$(Hg)	3	−35.36	0.00	0.33
$\beta_0+\beta_1$(Hg) + β_2(DATE)	4	−33.17	2.19	0.11
$\beta_0+\beta_1$(Hg) + β_2(LIV)	4	−33.09	2.27	0.10
$\beta_0+\beta_1$(Hg) + β_2(YR)	4	−33.08	2.28	0.10
β_0	2	−32.91	2.45	0.10
$\beta_0+\beta_1$(LIV)	3	−31.25	4.11	0.04
$\beta_0+\beta_1$(Hg) + β_2(DATE) + β_3(LIV)	5	−30.83	4.53	0.03
$\beta_0+\beta_1$(Hg) + β_2(DATE) + β_3(YR)	5	−30.82	4.53	0.03
$\beta_0+\beta_1$(YR)	3	−30.78	4.58	0.03
$\beta_0+\beta_1$(DATE)	3	−30.75	4.61	0.03
$\beta_0+\beta_1$(Hg) + β_2(YR) + β_3(LIV)	5	−30.73	4.63	0.03
$\beta_0+\beta_1$(YR) + β_2(LIV)	4	−29.17	6.19	0.01
$\beta_0+\beta_1$(DATE) + β_2(LIV)	4	−29.04	6.32	0.01
$\beta_0+\beta_1$(YR) + β_2(DATE)	4	−28.53	6.83	0.01
$\beta_0+\beta_1$(Hg) + β_2(DATE) + β_3(YR) + β_4(LIV)	6	−28.39	6.97	0.01
$\beta_0+\beta_1$(DATE) + β_2(YR) + β_3(LIV)	5	−26.85	8.50	0.00
Stage 2 – assess status and nutrient reserve effects				
$\beta_0+\beta_1$(Hg)	3	−35.36	0.00	0.21
$\beta_0+\beta_1$(Hg) + β_2(STAT)	4	−35.21	0.15	0.20
$\beta_0+\beta_1$(Hg) + β_2(STAT) + β_3(PRO)	5	−34.92	0.44	0.17
$\beta_0+\beta_1$(Hg) + β_2(PRO)	4	−34.16	1.20	0.12
$\beta_0+\beta_1$(Hg) + β_2(LIP)	4	−33.28	2.08	0.07
$\beta_0+\beta_1$(Hg) + β_2(STAT) + β_3(LIP)	5	−32.96	2.40	0.06
β_0	2	−32.91	2.45	0.06
$\beta_0+\beta_1$(Hg) + β_2(PRO) + β_3(LIP)	5	−32.70	2.66	0.06
$\beta_0+\beta_1$(Hg) + β_2(STAT) + β_3(PRO) + β_4(LIP)	6	−32.50	2.86	0.05

[a] Hg (log transformed liver mercury concentrations [$\mu g\,g^{-1}$ dw]); DATE (Julian date on which female was collected); LIV (liver dw [g]); YR (class variable denoting females collected in year 2002 or 2003); STAT (class variable denoting female status as non-developed [rapid follicle growth not initiated] or RFG [rapid follicle growth initiated]); PRO (protein reserves corrected for variation due to body size [residuals]); LIP (lipid reserves corrected for variation due to body size [residuals]).

Table 5
Geometric mean concentrations, 95% confidence limits, and ranges ($\mu g\,g^{-1}$ dw) of trace elements in egg constituents (albumen and yolk) and livers of six female greater scaup (*Aythya marila*) collected during 2002 and 2003 breeding seasons at the Yukon–Kuskokwim Delta, Alaska.

Element	Albumen	Yolk	Liver[a]
Calcium	795 (620–1020) (554–1030)	2510 (2180–2900) (2030–2890)	415 (340–505) (342–541)
Chromium	<dl[b] – (0.28)	0.70 (0.51–0.96) (0.37–0.85)	1.03 (0.88–1.22) (0.82–1.30)
Copper	7.25 (5.89–8.92) (5.07–8.64)	2.59 (2.07–3.24) (1.94–3.40)	40.7 (19.9–83.4) (12.3–81.6)
Iron	3.35 (1.89–5.92) (1.68–6.88)	175 (143–213) (133–229)	4480 (2430–8250) (7440–9730)
Potassium	8410 (6430–11,000) (5520–10,500)	2300 (2020–2630) (1930–2800)	8090 (7270–9000) (7440–9730)
Magnesium	719 (574–900) (521–987)	274 (223–336) (214–357)	786 (732–843) (707–857)
Manganese	0.14 (0.10–0.20) (0.09–0.20)	2.86 (2.24–3.66) (2.24–4.00)	19.0 (17.7–20.5) (17.6–24.4)
Sodium	6110 (4320–8640) (4110–8920)	1920 (1640–2250) (1470–2280)	3980 (3700–4290) (3640–4300)
Selenium	<dl[c] – (3.18–3.45)	<dl[d] – –	8.18 (4.68–14.3) (4.11–18.1)
Zinc	4.27 (2.59–7.04) (1.83–6.22)	60.0 (53.8–66.9) (51.6–67.4)	139 (109–176) (108–209)
Mercury	0.62 (0.51–0.76) (0.45–0.81)	0.02 (<0.01–1.08) (0.01–0.08)	0.85 (0.57–1.27) (0.53–1.60)

[a] Geometric mean concentration from females that laid eggs used in analyses.
[b] Below detection limit in 5 of 6 samples.
[c] Below detection limit in 4 of 6 samples.
[d] Below detection limit in all 6 samples.

were, however, well below hepatic concentrations associated with altered metabolism in mallards (*Anas platyrhynchos*) (436 $\mu g\,g^{-1}$ in Di Giulio and Scanlon, 1984) and general guidelines proposed for Cd poisoning in birds (>40 $\mu g\,g^{-1}$ wet weight [ww] in Furness, 1996; 10 $\mu g\,g^{-1}$ ww in Eisler, 2000a). Cd concentrations in females we collected during the breeding season were similar to those reported for greater scaup at several major wintering locales within the Long Island Sound region in northeastern US (0.88–4.36 $\mu g\,g^{-1}$ in Cohen et al., 2000) and at San Francisco Bay – California (Ohlendorf et al., 1986; ~0.36–2.93 $\mu g\,g^{-1}$ [converted from kidney tissue] derived from data in Cohen et al., 2000; Takekawa et al., 2002), but slightly higher than those for fall- and spring-staging (<0.15–1.99 $\mu g\,g^{-1}$ in Petrie et al., 2007) and wintering (1–3 $\mu g\,g^{-1}$ in Custer and Custer, 2000; Custer et al., 2003) scaup at the lower Great Lakes.

Because we did not observe a negative correlation between somatic lipid or protein reserves and Cd concentrations of non-developed and RFG females, it is unlikely that Cd was limiting endogenous energy reserves used during the breeding season. However, the highest levels of Cd were found during incubation when females use stored reserves to meet some of their metabolic demands (Flint, 2003; Gorman et al., 2008). Increases in hepatic Cd concentration post egg production have been observed in other species and may be related to lipid, protein, and calcium mobilization during reproduction (Stewart et al., 1994; Scheuhammer, 1996). Contrary to our findings, lipid reserves and hepatic Cd concentrations were negatively correlated in spring-migrant lesser scaup collected throughout the US upper Midwest, suggesting that Cd may be a trace element of concern, particularly during nutritionally or energetically stressful periods such as migration (Anteau et al., 2007).

Although a high proportion of greater scaup had Cd concentrations exceeding background levels, we suspect concentrations affecting this species, on average, must be higher than those in these birds because they were not negatively correlated with condition indices nor were they substantially higher in females without ovarian follicle development. Experimental studies are needed to establish species-specific tissue thresholds and biological endpoints to evaluate effects of Cd exposure on behavior, reproduction, and health of this species.

4.2. Mercury

Only 3% of female greater scaup in this study had elevated (>3 $\mu g\,g^{-1}$) hepatic Hg concentrations, plus all individuals were well below levels associated with reproductive or survival effects in other bird species (Thompson, 1996; Eisler, 2000a). Hepatic Hg concentrations in females we collected did vary slightly between years, but concentrations did not exceed background levels in either year. Other studies of greater scaup also have mostly reported background concentrations of Hg at spring and fall stopover

sites (Petrie et al., 2007) and at some wintering areas along the east coast of the US (Cohen et al., 2000). However, greater scaup wintering at San Francisco Bay, California contained higher Hg concentrations than did females we collected on breeding grounds in Alaska (Ohlendorf et al., 1986; Hothem et al., 1998; Takekawa et al., 2002) and body mass of greater scaup wintering at San Francisco Bay decreased with increasing hepatic Hg concentrations (Hoffman et al., 1998). Our results, however, did not support an adverse effect of Hg on female body condition nor on ability of females to initiate RFG.

4.3. Selenium

Se is a semi-metallic trace element that is required for biological processes, but has a relatively narrow threshold between normal and toxic levels, both of which vary considerably among species (Heinz, 1996). A sizeable percentage of female greater scaup we collected had liver Se concentrations exceeding the 10 $\mu g\,g^{-1}$ threshold associated with reproductive impairment in mallards (Heinz, 1996). Although 42% of females had elevated Se burdens, none exceeded the 33 $\mu g\,g^{-1}$ threshold associated with physiological impairment in mallards (Heinz, 1996), plus average concentrations in females generally did not exceed background levels. While Se burdens in some of our females exceeded levels deemed problematic for mallards, sensitivity to this trace element may vary among species and within avian families (Skorupa, 1998; Eisler, 2000b). For example, Wilson et al. (2007) reported a negative effect of elevated Se on egg viability (>7% inviable) for common eiders (*Somateria mollissima*) nesting on the YKD, whereas estimates of egg viability for greater scaup (<2% inviable) at our YKD study area (P.L. Flint, Unpublished results) were similar to values previously reported for other diving ducks (Johnson et al., 1992). Given most greater scaup spend the non-breeding season in marine environments which are generally Se-enriched (Haygarth, 1994), they, like sea ducks (Grand et al., 2002; Wayland et al., 2003), may have evolved higher tolerances to Se burdens than mallards and other freshwater waterfowl (Skorupa, 1998). Further, Se is eliminated quickly from the body after consumption of enriched food is reduced (Heinz et al., 1990; Grand et al., 2002; DeVink et al., 2008a). Our results show at least some females (2 of 6) transferred Se to their eggs although at very low levels. Thus, some greater scaup transport Se acquired at major wintering and spring-staging areas, many of which are located in highly urbanized and industrialized regions of North America (Kessel et al., 2002), to their northern breeding grounds.

Contrary to many other waterfowl (Alisauskas and Ankney, 1992), female greater scaup on the YKD did not use endogenous lipid and protein reserves to produce eggs (Gorman et al., 2008), but used reserves as energy for maintenance during incubation (Flint, 2003; Gorman et al., 2008). Body condition is a function of both lipid and protein reserves, with lipid often considered the more important component for survival during prolonged energy deficits (Lindstrom and Piersma, 1993). Thus, lack of any correlation between Se and female lipid reserves, combined with only a very slight reduction in protein reserves in females with higher Se burdens in this population, suggests that the observed Se concentrations had little, if any, impact on ability to store or later mobilize energy for maintenance or survival.

Our finding that lipid reserves were unrelated to liver Se burdens in greater scaup females does not support the hypothesis that Se is adversely affecting aspects of reproduction that are related to body condition. Similarly, Anteau et al. (2007) did not find evidence of an inhibitive effect of hepatic selenium on lipid reserves in spring-migrant lesser scaup. DeVink et al. (2008b) reported similar results for both lipid and protein reserves and concluded that Se had little impact on reproduction in lesser scaup. Captive studies of lesser scaup fed Se-enriched diets also did not support Se-related cross-seasonal impacts on body mass, egg teratogenicity, clutch initiation dates, or breeding probability (DeVink et al., 2008a). In this study, hepatic Se concentrations in females that had initiated RFG were similar to those in non-developed females, further implying that these concentrations of Se were likely insufficient to impact breeding propensity, thus reproduction, of female greater scaup on the YKD.

4.4. Egg concentrations

Our analysis of eggs laid by six females show many trace elements, including Hg and Se, were transferred to eggs. Mercury was detected in all eggs, whereas selenium was less prevalent (2 of 6 eggs; 33%) but indicated maternal transfer of this trace element occurs in some females. Assuming eggshell contained minimal concentrations (≤1% total egg burdens) of these trace elements (Burger, 1994), we estimated total egg concentrations of Hg (≤0.2 $\mu g\,g^{-1}$) and Se (≤1.2 $\mu g\,g^{-1}$) in this sample would be well below concentrations (Hg = ~6.0 $\mu g\,g^{-1}$; Se = ~9.0 $\mu g\,g^{-1}$) associated with reduced hatchability and embryonic deformity (Heinz, 1996; Thompson, 1996). Also, Hg and Se concentrations in these greater scaup eggs were within the range of values reported for eggs of lesser scaup (Fox et al., 2005; DeVink et al., 2008b). Our assessment of trace element concentrations and their prevalence in this, albeit a small, sample of eggs suggests little cause for concern regarding Hg and Se (or other trace element) impacts on egg viability or hatchling development in this species. This conclusion is further supported by field observations that inviable eggs are a rare occurrence in this population (P.L. Flint, Unpublished results).

5. Conclusions

Much of the current understanding of Se and other trace element impacts on scaup and other waterfowl has resulted from experimental studies of captive mallards (Gasaway and Buss, 1972; Heinz et al., 1983, 1989, 1990) and correlation-based field studies of scaup (e.g., Takekawa et al., 2002; Anteau et al., 2007; DeVink et al., 2008b; this study). Because of the potential interspecific variability in sensitivity to trace element concentrations, differences in toxicity of various forms of elements, and potential interactions among trace elements (Eisler, 2000a,b), direct tests of the contaminants hypothesis will require experimental studies on wild-strain and captive scaup (e.g., DeVink et al., 2008a) to identify thresholds and establish cause and effect relationships between exposure and behavior, reproduction, and health/survival. Based on results of this one study, we cannot universally reject the hypothesis that trace elements, including Se, Hg, and Cd, can adversely affect greater scaup reproduction. However, if effects proposed by the contaminants hypothesis do exist in the wild, exposure concentrations would have to exceed those we documented in female greater scaup collected at the YKD.

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References

Afton, A.D., Anderson, M.G., 2001. Declining scaup populations: a retrospective analysis of long-term population and harvest survey data. Journal of Wildlife Management 65, 781–796.

Afton, A.D., Ankney, C.D., 1991. Nutrient-reserve dynamics of breeding lesser scaup: a test of competing hypotheses. Condor 93, 89–97.

Alisauskas, R.T., Ankney, C.D., 1992. The cost of egg laying and its relationship to nutrient reserves in waterfowl. In: Batt, B.D.J., Afton, A.D., Anderson, M.G., Ankney, C.D., Johnson, D.H., Kadlec, J.A., Krapu, G.L. (Eds.), Ecology and Management of Breeding Waterfowl. University of Minnesota Press, Minneapolis, pp. 30–61.

Ankney, C.D., Afton, A.D., 1988. Bioenergetics of breeding northern shovelers: diet, nutrient reserves, clutch size, and incubation. Condor 90, 459–472.

Anteau, M.J., Afton, A.D., Custer, C.M., Custer, T.W., 2007. Relationships of cadmium, mercury, and selenium with nutrient reserves of female lesser scaup (*Aythya affinis*) during winter and spring migration. Environmental Toxicology and Chemistry 26, 515–520.

Austin, J.E., Afton, A.D., Anderson, M.G., Clark, R.G., Custer, C.M., Lawrence, J.S., Pollard, J.B., Ringelman, J.K., 2000. Declining scaup populations: issues, hypotheses, and research needs. Wildlife Society Bulletin 28, 254–263.

Badzinski, S.S., Petrie, S.A., 2006. Diets of lesser and greater scaup during autumn and spring on the lower Great Lakes. Wildlife Society Bulletin 34, 664–674.

Burger, J., 1994. Heavy metals in avian eggshells: another excretion method. Journal of Toxicology and Environmental Health 41, 207–220.

Burnham, K.P., Anderson, D.R., 2002. Model Selection and Multi-model Inference: a Practical Information Theoretic Approach, second ed. Springer-Verlag, New York.

Cohen, J.B., Barclay, J.S., Major, A.R., Fisher, J.P., 2000. Wintering greater scaup as biomonitors of metal contamination in federal wildlife refuges in the Long Island region. Archives of Environmental Contamination and Toxicology 38, 83–92.

Custer, C.M., Custer, T.W., 1996. Food habits of diving ducks in the Great Lakes after the zebra mussel invasion. Journal of Field Ornithology 67, 86–99.

Custer, C.M., Custer, T.W., 2000. Organochlorine and trace element contamination in wintering and migrating diving ducks in the southern Great Lakes, USA, since the zebra mussel invasion. Environmental Toxicology and Chemistry 19, 2821–2829.

Custer, C.M., Custer, T.W., Anteau, M.J., Afton, A.D., Wooten, D.E., 2003. Trace elements in lesser scaup (*Aythya affinis*) from the Mississippi flyway. Ecotoxicology 12, 47–54.

Custer, T.W., Custer, C.M., Hines, R.K., Sparks, D.W., 2000. Trace elements, organochlorines, polycyclic aromatic hydrocarbons, dioxins, and furans in lesser scaup wintering on the Indiana Harbor Canal. Environmental Pollution 110, 469–482.

DeVink, J.M.A., Clark, R.G., Slattery, S.M., Scheuhammer, T.M., 2008a. Effects of dietary selenium on reproduction and body mass of captive lesser scaup. Environmental Toxicology and Chemistry 27, 471–477.

DeVink, J.M.A., Clark, R.G., Slattery, S.M., Wayland, M., 2008b. Is selenium affecting body condition and reproduction in boreal breeding scaup, scoters, and ring-necked ducks? Environmental Pollution 152, 116–122.

Di Giulio, R.T., Scanlon, P.F., 1984. Sublethal effects of cadmium ingestion on mallard ducks. Archives of Environmental Contamination and Toxicology 13, 765–771.

Dobush, G.R., Ankney, C.D., Krementz, D.G., 1985. The effect of apparatus, extraction time, and solvent type on lipid extractions of snow geese. Canadian Journal of Zoology 63, 1917–1920.

Eisler, R., 2000a. Handbook of Chemical Risk Assessment: Health Hazards to Humans, Plants, and Animals. In: Metals, vol. 1. Lewis Publishers, New York.

Eisler, R., 2000b. Handbook of Chemical Risk Assessment: Health Hazards to Humans, Plants, and Animals. In: Metalloids, Radiation, Cumulative Index to Chemicals and Species, vol. 3. Lewis Publishers, New York.

Environment Canada, 1989. Analytical Methods Manual. In: Group 2, Metals and Organometallics, vol. 2. Water Quality Branch, Environment Canada, Ottawa.

Esler, D., Grand, J.B., Afton, A.D., 2001. Intraspecific variation in nutrient reserve use during clutch formation by lesser scaup. Condor 103, 810–820.

Flint, P.L., 2003. Incubation behavior of greater scaup on the Yukon–Kuskokwim Delta, Alaska. Wildfowl 54, 97–105.

Flint, P.L., Grand, J.B., 1999. Patterns of variation in size and composition of greater scaup eggs: are they related? Wilson Bulletin 111, 465–471.

Flint, P.L., Grand, J.B., Fondell, T.F., Morse, J.A., 2006. Population dynamics of greater scaup breeding on the Yukon–Kuskokwim Delta, Alaska. Wildlife Monographs 162, 1–22.

Fox, G.A., MacCluskie, M.C., Brook, R.W., 2005. Are current contaminant concentrations in eggs and breeding female lesser scaup of concern? Condor 107, 50–61.

Furness, R.W., 1996. Cadmium in birds. In: Beyer, N.W., Heinz, G.H., Redmon-Norwood, A.W. (Eds.), Environmental Contaminants in Wildlife: Interpreting Tissue Concentrations. Lewis Publishers, New York, pp. 389–404.

Gasaway, W.C., Buss, I.O., 1972. Zinc toxicity in the mallard duck. Journal of Wildlife Management 36, 1107–1117.

Gorman, K.B., Esler, D., Flint, P.L., Williams, T.D., 2008. Nutrient-reserve dynamics during egg production by female greater scaup (*Aythya marila*): relationships with timing of reproduction. Auk 125, 384–394.

Gorman, K.B., Flint, P.L., Esler, D., Williams, T.D., 2007. Ovarian follicle dynamics of female greater scaup during egg production. Journal of Field Ornithology 78, 64–73.

Grand, J.B., Franson, J.C., Flint, P.L., Petersen, M.R., 2002. Concentrations of trace elements in eggs and blood of spectacled and common eiders on the Yukon–Kuskokwim Delta, Alaska, USA. Environmental Toxicology and Chemistry 21, 1673–1678.

Haygarth, P.M., 1994. Global importance and cycling of selenium. In: Frankenberger Jr., W.T., Benson, S. (Eds.), Selenium in the Environment. Marcel Dekker, New York, pp. 1–27.

Heinz, G.H., 1996. Selenium in birds. In: Beyer, W.N., Heinz, G.H., Redmon-Norwood, A.W. (Eds.), Environmental Contaminants in Wildlife: Interpreting Tissue Concentrations. Lewis Publishers, Boca Raton, Florida, pp. 447–458.

Heinz, G.H., Haseltine, S.D., Sileo, L., 1983. Altered avoidance behavior of young black ducks fed cadmium. Environmental Toxicology and Chemistry 2, 419–421.

Heinz, G.H., Hoffman, D.J., Gold, L.G., 1989. Impaired reproduction of mallards fed an organic form of selenium. Journal of Wildlife Management 53, 418–428.

Heinz, G.H., Pendleton, G.W., Krynitsky, A.J., Gold, L.G., 1990. Selenium accumulation and elimination in mallards. Archives of Environmental Contamination and Toxicology 19, 374–379.

Hodges, J.I., King, J.G., Conant, B., Hanson, H.A., 1996. Aerial Surveys of Waterbirds in Alaska 1957–94: Population Trends and Observer Variability. National Biological Service Information and Technology Report 4.

Hoffman, D.J., Ohlendorf, H.M., Marn, C.M., Pendleton, G.W., 1998. Association of mercury and selenium with altered glutathione metabolism and oxidative stress in diving ducks from the San Francisco Bay region, USA. Environmental Toxicology and Chemistry 17, 167–172.

Hoffman, D.J., Sanderson, C.J., LeCaptain, L.J., Cromartie, E., Pendleton, G.W., 1992. Interactive effects of arsenate, selenium, and dietary-protein on survival, growth, and physiology in mallard ducklings. Archives of Environmental Contamination and Toxicology 22, 55–62.

Hothem, R.L., Lonzarich, D.G., Takekawa, J.E., Ohlendorf, H.M., 1998. Contaminants in wintering canvasbacks and scaups from San Francisco Bay, California. Environmental Monitoring and Assessment 50, 67–84.

Johnson, D.H., Nichols, J.D., Schwartz, M.D., 1992. Population dynamics of breeding waterfowl. In: Batt, B.D.J., Afton, A.D., Anderson, M.G., Ankney, C.D., Johnson, D.H., Kadlec, J.A., Krapu, G.L. (Eds.), Ecology and Management of Breeding Waterfowl. University of Minnesota Press, Minneapolis, Minnesota, pp. 446–485.

Kessel, B.D., Rocque, D.A., Barclay, J.S., 2002. Greater scaup (*Aythya marila*). In: Poole, A., Gill, F. (Eds.), The Birds of North America, No. 650, Academy of Natural Sciences and American Ornithologists' Union, Philadelphia, Pennsylvania and Washington, D.C.

Lindstrom, A., Piersma, T., 1993. Mass changes in migrating birds: the evidence for fat and protein storage reexamined. Ibis 135, 70–78.

Mallory, M.L., Wayland, M., Braune, B.M., Drouillard, K.G., 2004. Trace elements in marine birds, arctic hare and ringed seals breeding near Qikiqtarjuaq, Nunavut, Canada. Marine Pollution Bulletin 49, 135–141.

Mitchell, C.A., Carlson, J., 1993. Lesser scaup forage on zebra mussels at Cook Nuclear Plant, Michigan. Journal of Field Ornithology 64, 219–222.

Ohlendorf, H.M., Lowe, R.W., Kelly, P.R., Harvey, T.E., Stafford, C.J., 1986. Selenium and heavy metals in San-Francisco Bay diving ducks. Journal of Wildlife Management 50, 64–70.

Petrie, S.A., Badzinski, S.S., Drouillard, K.G., 2007. Contaminants in lesser and greater scaup staging on the lower Great Lakes. Archives of Environmental Contamination and Toxicology 52, 580–589.

Rattner, B.A., Jehl, J.R., 1997. Dramatic fluctuations in liver mass and metal content of eared grebes (*Podiceps nigricollis*) during autumnal migration. Bulletin of Environmental Contamination and Toxicology 59, 337–343.

SAS Institute, 2004. SAS OnlineDoc® 9.1.3. SAS Institute Inc., Cary, North Carolina.

Sedinger, J.S., Ankney, C.D., Alisauskas, R.T., 1997. Refined methods for assessment of nutrient reserve use and regulation of clutch size. Condor 99, 836–840.

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

Scheuhammer, A.M., 1996. Influence of reduced dietary calcium on the accumulation and effects of lead, cadmium and aluminum in birds. Environmental Pollution 94, 337–343.

Scheuhammer, A.M., Wong, A.H.K., Bond, D., 1998. Mercury and selenium accumulation in common loons (*Gavia immer*) and common mergansers (*Mergus merganser*) from eastern Canada. Environmental Toxicology and Chemistry 17, 197–201.

Skorupa, J.P., 1998. Selenium poisoning of fish and wildlife in nature: lessons from twelve real-world examples. In: Frankenburger, W.T., Engburg, R.A. (Eds.), Environmental Chemistry of Selenium. Marcel Dekker, New York, pp. 315–354.

Stewart, F.M., Thompson, D.R., Furness, R.W., Harrison, N., 1994. Seasonal variation in heavy metal levels in tissues of common guillemots, *Uria aalge*, from Northwest Scotland. Archives of Environmental Contamination and Toxicology 27, 168–175.

Takekawa, J.Y., Wainwright-De La Cruz, S.E., Hothem, R.L., Yee, J., 2002. Relating body condition to inorganic contaminant concentrations of diving ducks wintering in coastal California. Archives of Environmental Contamination and Toxicology 42, 60–70.

Thompson, D.R., 1996. Mercury in birds and terrestrial mammals. In: Beyer, W.N., Heinz, G.H., Redmon-Norwood, A.W. (Eds.), Environmental Contaminants in Wildlife: Interpreting Tissue Concentrations. Lewis Publishers, Boca Raton, Florida, pp. 341–356.

Ware, L.L., 2008. Selenium Uptake and Effects in Greater Scaup (*Aythya marila*) Wintering on Western Lake Ontario. M.Sc. thesis. University of Western Ontario, London, Ontario, Canada.

Wayland, M., Gilchrist, H.G., Marchant, T., Keating, J., Smits, J.E., 2002. Immune function, stress response, and body condition in arctic-breeding common eiders in relation to cadmium, mercury, and selenium concentrations. Environmental Research 90, 47–60.

Wayland, M., Smits, J.E., Gilchrist, H.G., Marchant, T., Keating, J., 2003. Biomarker responses in nesting common eiders in the Canadian arctic in relation to tissue cadmium, mercury and selenium concentrations. Ecotoxicology 12, 225–237.

Wilson, H.M., Flint, P.L., Powell, A.N., 2007. Coupling contaminants with demography: effects of lead and selenium in Pacific common eiders. Environmental Toxicology and Chemistry 26, 1410–1417.