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Trace elements status of white-tailed deer (Odocoileus virginianus) and moose (Alces alces) in Nova Scotia

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prepared for

the Nova Scotia Department of Natural Resources
and
the Canadian Cooperative Wildlife Health Centre

by

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Abstract

The province of Nova Scotia is considered to have two moose (*Alces alces*) populations. In 2003, the moose of the mainland area of the province were formally listed "ENDANGERED" under the Nova Scotia Endangered Species Act. To date, the specific causes of the Mainland moose population decline have not been determined. Trace element imbalances have been considered as a potential etiology for the population decline. Liver and kidney samples were collected from white-tailed deer (*Odocoileus virginianus*) and moose throughout Nova Scotia during the fall and winter 2000-01 to compare trace element concentrations between the two species, in relation to age, gender and location and to other areas. All samples were analysed for arsenic, cadmium, cobalt, copper, lead, manganese, nickel, selenium and zinc. Tissue concentrations of trace elements in deer and moose in Nova Scotia appear to be generally similar to levels reported in cervid populations elsewhere in North America and Europe with the exception of zinc and possibly cobalt which appear to be lower in Nova Scotia. Kidney cadmium concentrations are high in some Nova Scotia moose (geometric mean: 60.4 µg/g dry weight [95%CI: 40.3 - 90.6]), however, similar or higher concentrations have been reported in other regions. Relative to reference values for domestic cattle, cobalt, copper, manganese, selenium and zinc levels in some animals are deficient or marginally deficient. At the present time, there appears to be little supporting evidence that clinical deficiencies of any of these trace elements are occurring in Nova Scotia moose or deer populations. However, the possibility that marginal or deficient levels of these or other trace elements and high levels of cadmium may impact the health of individual animals either directly or through interactions with other factors (eg. infectious and non-infectious diseases, harsh environmental conditions, habitat limitations) cannot be dismissed. Recommendations for continued monitoring of trace element concentrations in these populations are made.

Acknowledgments

The data presented in this report was originally collected and compiled by staff of the Nova Scotia Department of Natural Resources. Trace element analyses of tissue samples were completed by Erin Roger through the Environmental Quality Laboratory, Environment Canada, Moncton, New Brunswick. Pierre-Yves Daoust and Scott McBurney from the Canadian Cooperative Wildlife Health Centre, Atlantic Region and Tony Nette from the Nova Scotia Department of Natural Resources provided helpful input and editorial comments.

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Introduction

The province of Nova Scotia is considered to have two moose (*Alces alces*) populations. The first population on Cape Breton Island experiences normal population growth and is harvested. It was supplemented by eighteen moose translocated from Alberta in 1947 and 1948 (Pulsifer and Nette, 1995), and, not surprisingly, subsequent genetic analysis (Broders et al., 1999) has demonstrated the population's genetic structure to be most closely related to that of moose from Alberta. The second population of moose is on the mainland of the province and is indigenous to the region, representative of eastern moose subspecies (*Alces alces americana*). This population has been in decline since the mid-1920's, and has been protected from legal hunting since 1981. In 2000, it was assigned a "RED" status which is defined to be a species at risk of extirpation or extinction under the General Status of Nova Scotia Wildlife Assessment Process. Following the completion of an independent commissioned status report (Parker, 2003) in October of 2003, moose of the mainland area of the province were formally listed "ENDANGERED" under the Nova Scotia Endangered Species Act.

To date, the specific causes of the Mainland moose population decline have not been determined. Factors influencing the size and distribution of moose populations in Nova Scotia are multiple and thought to include: disease (eg. parelaphostrongylosis), habitat suitability, illegal hunting and possibly predation of calves by black bear or coyotes (Beazley et al., 2005). Trace element imbalances have also been considered as a potential etiology for the population decline because such imbalances have been cited as possible contributing factors in population declines/mortality events of moose in other areas. Examples include possible copper deficiency in moose in Alaska (Flynn et al., 1977; O'Hara et al., 2001), copper deficiency and molybdenosis in moose in Sweden (Frank et al., 1994, 2000), and copper deficiency in northwestern Minnesota (Custer et al., 2004). Results from preliminary trace element analyses in moose kidney and liver tissues have raised the possibility of high cadmium and/or low cobalt levels as being contributing factors in the continued decline of moose population in Mainland Nova Scotia (Roger, 2002; Frank et al., 2004).

Historically, the white-tailed deer (*Odocoileus virginianus*) population in Nova Scotia has fluctuated (Pulsifer and Nette, 1995), but the current number of deer in the province is considered adequate. Liver and kidney samples were collected from deer and moose throughout Nova Scotia to compare trace element concentrations between the two species and in relation to other areas.

The objectives of this report were to:

1-organize and summarize trace element data collected from moose and deer in Nova Scotia

2-analyse data with respect to age, gender and location

3-compare levels found to reported reference values in order to identify possible deficiencies and/or toxicities

4-discuss findings in relation to other studies

5-identify recommendations for further studies or analyses.

Methods

Sample collection

Samples of liver from deer and liver and kidney from moose were collected from animals killed in vehicle accidents or by hunters, or animals killed and submitted to the Department of Natural Resources because of property intrusion or illness during the fall and winter 2000-01. Animals were aged by size (calves and fawns), tooth wear or tooth cementum analysis. Specific age data (in months) were available only for calves or fawns and those animals aged by tooth cementum analysis. Because specific age data were not available for all animals, where possible, animals were categorized by age as calves (moose) or fawns (deer) (<12 months), yearlings (12 to 24 months) and adults (>24 months) for descriptive and statistical analyses.

Specific location data (UTMs) were obtained for 51/54 deer and 20/48 moose. For comparisons between locations, deer were categorized according to three regions: Mainland East (ME), Mainland West (MW) and Cape Breton (CB) (Figure 1). Similar regions were used to categorize moose (Figure 2).

Analytical methods

All samples were analysed for trace element concentrations (arsenic, cadmium, cobalt, copper, lead, manganese, nickel, selenium, zinc) at the Environmental Quality Laboratory, Environment Canada, Moncton, New Brunswick in 2001. Samples were analysed using a Perkin Elmer Elan 6000 inductively coupled mass spectrophotometer (ICP-MS). The minimum detection limit was 0.25 µg/g for arsenic and selenium and 0.05 µg/g for cadmium, cobalt, copper, manganese, nickel, lead and zinc. Concentrations are reported in µg/g dry weight (dw).

Statistical analysis

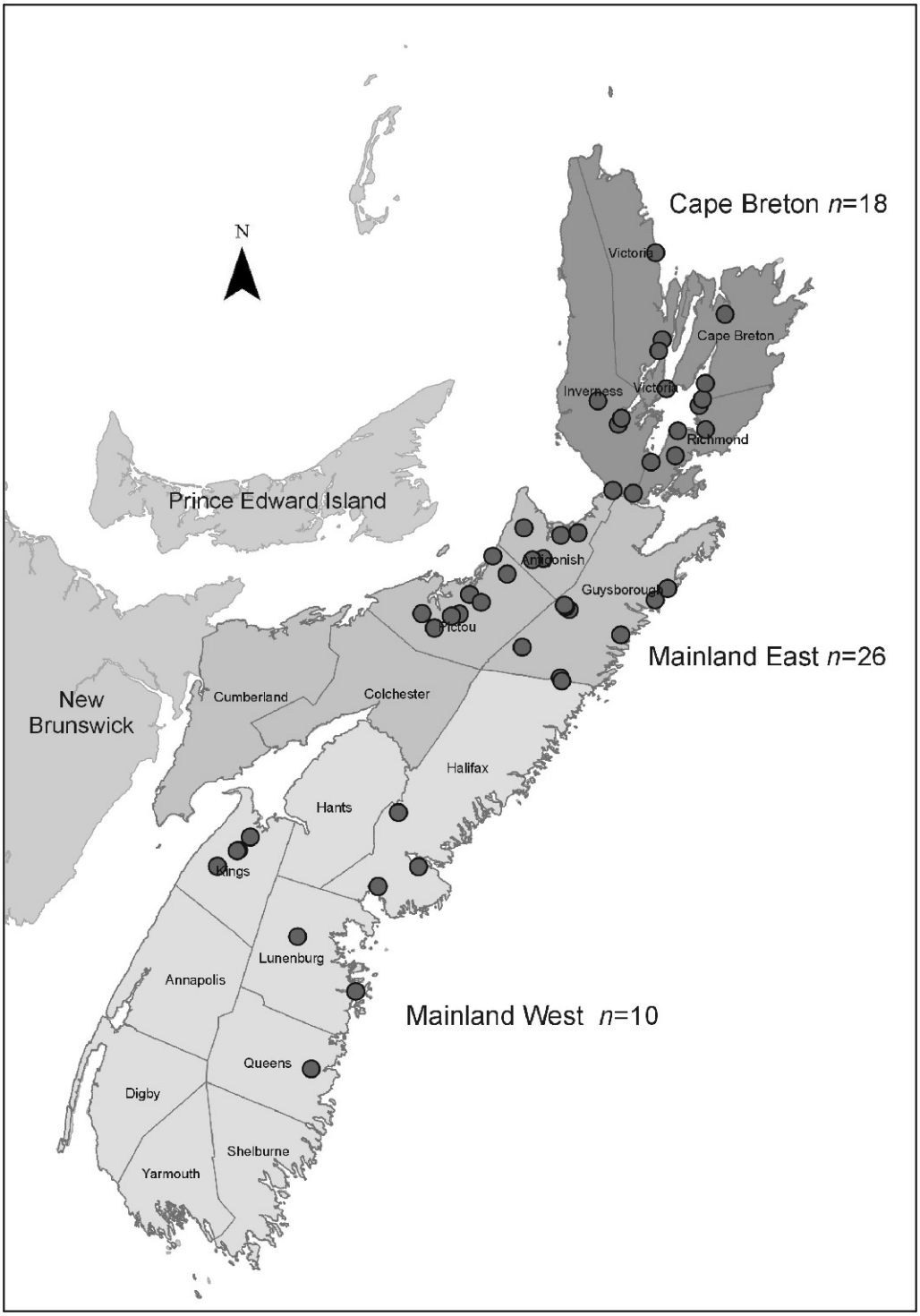
Trace element values less than the minimum detection limits (mdl) were replaced with ½ of the mdl. Geometric means and 95% confidence intervals (95% CI) and ranges were used for descriptive statistics.

Trace elements detected in at least 20% of the samples by species and tissue (all trace elements measured except for arsenic and lead in deer liver and arsenic in moose liver) were included in univariate statistical analyses described below.

All transformations and analyses were carried out using statistical software (STATA v.8.0). Trace element concentrations were transformed (natural log [ln]) to meet assumptions of normality for the parametric statistical tests used. Non-parametric statistical tests were used for those variables for which ln transformation did not result in a normal distribution (cobalt and zinc concentrations in deer liver, and cadmium, manganese and lead concentrations in moose liver).

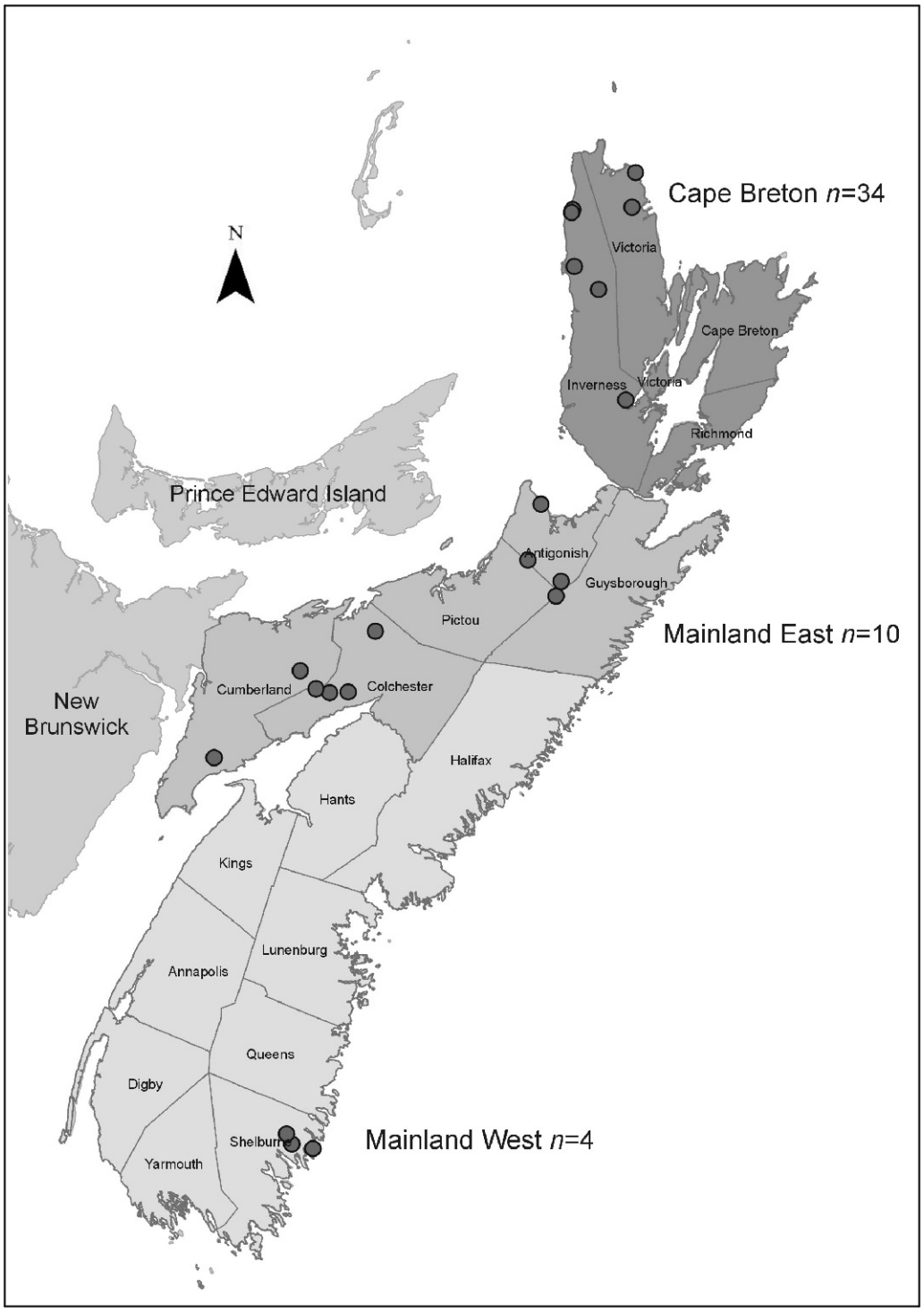
Simple associations among trace elements and demographic variables (gender, age, location) were examined using χ^2 statistics for categorical variables (gender, age group, location), pair-wise Pearson's correlation coefficients and *t*-tests and one-way ANOVA

Figure 1. Map of Nova Scotia illustrating locations of deer and number of deer sampled in each region.



Projection: Lambert Conformal Conic projection.

Figure 2. Map of Nova Scotia illustrating locations of moose and number of moose sampled in each region.



Projection: Lambert Conformal Conic projection.

(followed by Bonferroni's adjustments for pair-wise comparisons) for normally distributed continuous variables, and Spearman's correlation coefficients and Kruskal-Wallis test for non-normally distributed continuous variables. Differences between trace element concentrations in deer and moose liver samples were also examined. Results of $p<0.05$ were reported as statistically significant.

For the purposes of comparison, all concentrations reported in wet weight (ww) in other studies were converted to dry weight by estimating the moisture content of liver and kidney tissues as 71.4% (Puls, 1994), resulting in the conversion equation µg/g ww x 3.5= µg/g dw.

Results

Demographic data

Deer

Liver samples were collected from 54 white-tailed deer. Gender and age data were collected for 51/54 deer. Of these, 33 (65%) were female, 2 (4%) were fawns, 19 (37%) were yearlings, and 30 (59%) were adults. Specific age data (based on tooth cementum analysis) were collected for 34 deer and ages ranged from 8 months to 126 months (10.5 years). Geometric mean age was 23 months (95% CI: 20 - 27).

Of the 54 deer sampled, 18 (33%) were from Cape Breton, 26 (48%) from Mainland East and 10 (19%) from Mainland West.

The gender distribution was significantly different between locations (CB=59% male, ME=33% male, MW=0% male; $\chi^2=9.6$, df=2, $p<0.05$). No age group differences were found between locations ($\chi^2=3.1$, df=4, $p=0.54$) or genders ($\chi^2=4.48$, df=2, $p=0.11$).

Moose

Forty-eight liver samples and 21 kidney samples were collected from moose. Gender data were collected from 46/48 moose and 18 (39%) of these were female. Categorical age data were collected from 41/48 moose and 5 (12%) were calves, 11 (27%) were yearlings, and 25 (61%) were adults. Specific age data were collected from 36/48 moose and ages ranged from 4 months to 123 months (10.3 years). Geometric mean age was 29 months (95% CI: 21 - 39).

Of the 48 moose sampled, 34 (71%) were from Cape Breton, 10 (21%) from Mainland East and 4 (8%) from Mainland West.

Males were older than females. This gender difference between age groups was nearly significant (males: 1/5 calves, 6/10 yearlings, 19/25 adults) ($\chi^2=5.89$, df=2, $p=0.053$).

No age group differences ($\chi^2=8.92$, df=4, $p=0.063$) or gender differences ($\chi^2=2.47$, df=2, $p=0.29$) were found between locations.

Tissue prevalence and concentrations of trace elements

Tables 1, 2 and 3 provide summaries of the prevalence and concentrations of trace elements by location for deer liver, moose liver and moose kidney, respectively. Reference values for domestic cattle (*Bos taurus*) for marginal and toxic levels of each trace element

Table 1. Trace element concentrations in Nova Scotia deer liver by region.

Trace element	Region *n*	Geometric mean µg/g dw (95% confidence interval) [range] %above minimum detection limit Cape Breton 18	Mainland East 26	Mainland West 10	overall 54	Reference values [c] marginal levels	toxic levels
Arsenic		na [<0.25 - 0.32]	na [<0.25 - 0.60]	na [<0.25 - 0.45]	na [<0.25 - 0.60] 13.0%	na	>24.5
Cadmium		1.6 (1.0 - 2.5)[a] [0.1 - 6.5]	1.0 (0.6 - 1.8)[a] [0.05 - 28.1]	0.6 (0.3 - 1.5)[a] [0.1 - 5.9]	1.1 (0.8 - 1.5) [0.05 - 28.1] 100%	na	>175
Cobalt		0.15 (0.11 - 0.20)[a] [<0.05 - 0.41]	0.16 (0.13 - 0.19)[a] [0.05 - 0.27]	0.14 (0.09 - 0.23)[a] [<0.05 - 0.31]	0.15 (0.13 - 0.17) [<0.05 - 0.41] 96.3%	0.06 marginal	>17.5
Copper		135.8 (77.6 - 237.7)[a] [2.0 - 344.4]	135.0 (83.4 - 218.5)[a] [2.4 - 506.3]	77.5 (34.3 - 175.0)[a] [3.7 - 207.6]	122.0 (88.9 - 167.5) [2.0 - 506.3] 100%	<35 deficient 35 - 87.5 marginal	>875
Manganese		11.6 (8.6 - 15.7)[a] [1.8 - 27.9]	11.5 (9.6 - 13.7) [a] [3.0 - 27.8]	12.1 (8.3 - 17.7)[a] [3.8 - 25.2]	11.6 (10.1 - 13.4) [1.8 - 27.9] 100%	<3.5 deficient 3.5 - 10.3 marginal	
Nickel		0.57 (0.31 - 1.04)[a] [<0.05 - 4.03]	0.54 (0.32 - 0.92)[a] [<0.05 - 4.46]	0.66 (0.30 - 1.44)[a] [0.08 - 2.55]	0.57 - 0.41 - 0.80) [<0.05 0 4.46] 94.4%	<0.035 marginal	
Lead		na [<0.05 - 0.25]	na [<0.05 - 0.51]	na [<0.05 - 0.35]	na [<0.05 - 0.51] 14.8%	na	>17.5
Selenium		1.7 (1.5 - 2.1)[a] [1.1 - 3.2]	1.3 (1.1 - 1.6)[b] [0.6 - 4.0]	1.2 (0.9 - 1.5)[b] [0.6 - 1.8]	1.4 (1.2 - 1.6) [0.6 - 4.0] 100%	<0.6 deficient 0.6 - 0.88 marginal	4.4 - 24.5
Zinc		68.1 (60.0 - 77.3)[a] [33.3 - 100.1]	81.2 (68.0 - 97.0[a] [41.5 - 406.0]	100.6 (67.5 - 150.0)[a] [71.1 - 461.9]	79.7 (70.9 - 89.6) [33.3 - 461.9] 100%	<70 deficient 70 - 87.5 marginal	

[a,b] For each trace element, regional means having different superscripts were significantly different , $p<0.05$.

[c] Marginal and toxic reference values for cattle in µg/g dw (Puls, 1994). Concentrations converted from wet weight based on estimate of 71.4% moisture (conversion factor of 3.5).

Table 2. Trace element concentrations in Nova Scotia moose liver by region.

Trace element	Region	Geometric mean µg/g dw (95% confidence interval) [range] %above minimum detection limit				Reference values [c]	
		Cape Breton	Mainland East	Mainland West	overall	marginal levels	toxic levels
	n	34	10	4	48		
Arsenic		na [<0.25 - 1.42]	na [<0.25 - <0.25]	na [<0.25 - 0.34]	na [<0.25 - 1.42] 6.3%	na	>24.5
Cadmium		5.2 (3.8 - 7.0)[a] [0.1 - 13.1]	4.6 (1.1 - 19.1)[a] [<0.05 - 23.6]	27.5 (12.4 - 60.7)[b] [17.9 - 51.9]	5.8 (4.0 - 8.3) [<0.05 - 51.9] 97.9%	na	>175
Cobalt		0.07 (0.06 - 0.09)[a] [<0.05 - 0.24]	0.14 (0.08 - 0.26)[b] [<0.05 - 0.39]	0.06 (0.02 - 0.17)[a,b] [<0.05 - 0.1]	0.08 (0.07 - 0.10) [<0.05 - 0.39] 85.4%	0.06 marginal	>17.5
Copper		308.3 (225.4 - 421.8)[a] [2.9 - 625.5]	134.7 (59.2 - 306.8)[a] [6.4 - 334.1]	213.4 (42.2 - 1078.5)[a] [56.3 - 595.1]	251.6 (188.2 - 336.5) [2.9 - 625.5] 100%	<35 deficient 35 - 87.5 marginal	>875
Manganese		8.6 (6.6 - 11.3)[a] [0.3 - 19.3]	9.9 (4.5 - 21.7)[a] [0.7 - 57.8]	13.0 (4.7 - 35.8)[a] [5.4 - 22.6]	9.2 (7.2 - 11.7) [0.32 - 57.8] 100%	<3.5 deficient 3.5 - 10.3 marginal	
Nickel [d]		0.65 (0.52 - 0.80)[b] [0.15 - 5.24]	0.08 (0.04 - 0.13)[a] [<0.05 - 0.26]	0.18 (0.01 - 3.60)[a] [<0.05 - 1.04]	0.37 (0.26 - 0.52) [<0.05 - 5.24] 93.8%	<0.035 marginal	
Lead		0.07 (0.05 - 0.11)[a] [<0.05 - 2.69]	0.06 (0.03 - 0.11)[a] [<0.05 - 0.25]	0.05 (0.01 - 0.20)[a] [<0.05 - 0.11]	0.07 (0.05 - 0.09) [<0.05 - 2.69] 56.3%	na	>17.5
Selenium		0.77 (0.65 - 0.90)[a] [0.37 - 3.57]	0.57 (0.39 - 0.82)[a] [0.30 - 2.00]	0.88 (0.32 - 2.47)[a] [0.37 - 1.58]	0.73 (0.63 - 0.84) [0.29 - 3.57] 100%	<0.6 deficient 0.6 - 0.88 marginal	4.4 - 24.5
Zinc		71.4 (53.6 - 95.1)[a] [9.5 - 476.9]	75.4 (58.6 - 97.0)[a] [44.3 - 134.7]	111.9 (48.5 - 258.3)[a] [53.1 - 181.6]	75.0 (60.7 - 92.6) [9.5 - 476.9] 100%	<70 deficient 70 - 87.5 marginal	

[a,b] For each trace element, regional means having different superscripts were significantly different , $p<0.05$.

[c] Marginal and toxic reference values for cattle in µg/g dw (Puls, 1994). Concentrations converted from wet weight based on estimate of 71.4% moisture (conversion factor of 3.5).

[d] One outlier (44.5 µg/g dw) was dropped for statistical analyses; Cape Breton *n*=33 and overall *n*=47.

Table 3. Trace element concentrations in Nova Scotia moose kidney by region.

Trace element	Geometric mean µg/g dw (95% confidence interval) [range] %above minimum detection limit				Reference values [c]	
Region	Cape Breton	Mainland East	Mainland West	overall	marginal levels	toxic levels
n	10	7	4	21		
Arsenic	0.28 (0.11 - 0.43)[a] [<0.25 - 0.59]	<0.25 [a] [<0.25 - <0.25]	<0.25 [a] [<0.25 - <0.25]	0.19 (0.15 - 0.24) [<0.25 - 0.59] 33.3%	na	>17.5
Cadmium	49.5 (34.6 - 70.9)[a] [31.9 - 160.8]	45.0 (17.0 - 119.2)[a] [14.3 - 283.4]	166.6 (48.5 - 572.1)[b] [76.4 - 346.1]	60.4 (40.3 - 90.6) [14.3 - 346.1] 100%	na	>350
Cobalt	0.11 (0.09 - 0.15)[a] [0.07 - 0.19]	0.12 (0.08 - 0.18)[a] [0.08 - 0.21]	0.05 (0.02 - 0.13)[a] [<0.05 - 0.08]	0.10 (0.08 - 0.13) [<0.05 - 0.21] 95.2%	<0.05 marginal	>105
Copper	14.5 (13.4 - 15.7)[a] [12.5 - 17.5]	17.0 (14.1 - 20.6)[a,b] [13.1 - 23.6]	19.0 (15.3 - 23.6)[b] [16.5 - 22.6]	16.1 (14.8 - 17.5) [12.5 - 23.6] 100%		
Manganese	14.3 (10.8 - 19.0)[a] [9.3 - 29.7]	11.5 (9.4 - 13.9)[a] [8.5 - 15.4]	12.9 (8.8 - 19.0)[a] [9.3 - 15.9]	13.0 (11.3 - 15.1) [8.5 - 29.7] 100%	3.3 - 4.2 marginal	
Nickel	1.37 (0.72 - 2.60)[b] [0.30 - 5.76]	0.21 (0.16 - 0.28)[a] [0.13 - 0.35]	0.33 (0.06 - 1.88)[a] [0.13 - 0.94]	0.56 (0.33 - 0.95) [0.13 - 5.76] 100%	<0.035	
Lead	0.03 (0.02 - 0.04)[a] [<0.05 - 0.09]	0.11 (0.08 - 0.16)[a] [0.07 - 0.18]	0.07 (0.01 - 0.43)[a] [<0.05 - 0.19]	0.05 (0.04 - 0.08) [<0.05 - 0.19] 47.6%	na	>17.5
Selenium	3.7 (3.2 - 4.2)[a] [2.6 - 5.3]	2.0 (1.8 - 2.3)[a] [1.8 - 2.5]	2.9 (2.0 - 4.1)[a] [2.1 - 3.4]	2.9 (2.5 - 3.3) [1.8 - 5.3] 100%	<2.1 marginal	8.8 - 17.5
Zinc	82.1 (71.5 - 94.2)[a] [68.6 - 131.1]	106.6 (85.9 - 132.3)[a] [76.0 - 145.2]	144.1 (115.4 - 180.1)[a] [117.5 - 159.5]	99.7 (87.4 - 113.7) [68.6 - 159.5] 100%	56 - 70 deficient/marginal	455 - 1680

[a,b] For each trace element, regional means having different superscripts were significantly different , $p<0.05$.

[c] Marginal and toxic reference values for cattle in µg/g dw (Puls, 1994). Concentrations converted from wet weight based on estimate of 71.4% moisture (conversion factor of 3.5).

are also listed where applicable (Puls, 1994).

Detectable levels of cadmium, copper, manganese, selenium and zinc were found in all deer liver samples and detectable levels of cobalt and nickel in greater than 50% of samples. Less than 20% of samples had detectable levels of arsenic and lead (Table 1).

Detectable levels of copper, manganese, selenium and zinc were found in all moose liver samples. Detectable levels of cadmium, cobalt, nickel and lead were found in greater than 50% of liver samples and arsenic in only 6% of liver samples (Table 2). Only 47 liver samples were used for calculations involving nickel concentrations, as one sample was dropped because of an erroneously high nickel value (44.5 µg/g dw).

Detectable levels of cadmium, copper, manganese, nickel, selenium and zinc were found in all moose kidney samples. Detectable levels of cobalt were found in 95% and detectable levels of arsenic and lead in less than 50% of kidney samples (Table 3).

Determination of trace element deficiency and/or toxicity

In general, based on reference values for domestic cattle, concentrations of arsenic, copper, lead and nickel in deer and moose tissues were within normal ranges (Tables 1, 2 and 3) (Puls, 1994). Results for other trace elements are listed below.

Univariate analyses

In deer, univariate analyses showed that liver manganese concentrations varied with gender and liver selenium concentrations varied with age, gender and location. Detailed results are listed under each trace element. No differences were seen in gender, age or location with respect to liver cadmium, cobalt, copper, nickel or zinc concentrations.

In moose, univariate analyses showed that cadmium concentrations varied with location (kidney and liver) and gender (kidney). Cobalt concentrations varied with age (kidney) and location (liver). Copper concentrations varied with age (liver) and location (kidney). Manganese concentrations varied with gender (liver). Nickel concentrations varied with location (kidney and liver). Detailed results are listed under each trace element. No differences were seen in gender, age or location with respect to lead, selenium, zinc (liver or kidney) or arsenic (kidney) concentrations.

Cadmium

Liver cadmium concentrations were higher in moose compared to deer ($p<0.001$) and within moose, kidney concentrations were higher than liver (kidney geometric mean: 60.4 µg/g dw [95%CI: 40.3 - 90.6]; liver: 8.4 µg/g dw [95%CI: 5.8 - 12.3], $n=21$).

Relative to reference values for domestic cattle, cadmium concentrations in deer and moose were below the chronic toxicity level (liver: 175 µg/g dw; kidney: 350 µg/g dw) (converted from ww) (Puls, 1994).

Mainland West moose had significantly higher liver and kidney cadmium concentrations compared to Mainland East or Cape Breton moose (Tables 2 and 3, respectively).

Female moose had higher kidney cadmium concentrations than males (male geometric mean: 41.1 µg/g dw [95%CI: 29.7 - 57.3]; female geometric mean: 92.4 µg/g dw [95% CI: 43.5 - 196.0]) ($p<0.05$).

Table 4 shows kidney and liver cadmium concentrations by age group. No significant differences were found between age groups with respect to cadmium concentrations in deer or moose. Similarly, no association was found between age (in months) and cadmium in either species (moose liver: r=0.08, moose kidney: r=0.40; deer liver: r=0.25) (p>0.05).

Cobalt

Liver cobalt concentrations were higher in deer compared to moose (p<0.001). In relation to reference values for domestic cattle, 4/54 (7.4%) of deer liver samples and 14/48 (29.2%) of moose liver samples were marginally deficient (Figure 1), and 2/21 (9.5%) of moose kidney samples were deficient ($\leq$ 0.05 µg/g dw) (Puls, 1994).

Mainland East moose had higher liver cobalt concentrations than Cape Breton moose (Table 2). There was a significant negative association between age (in months) and cobalt concentrations in kidney (Pearson's r=-0.53, p<0.05): as age increased, kidney cobalt concentrations decreased.

Copper

Liver copper concentrations were higher in moose compared to deer (p<0.001). In relation to reference values for domestic cattle, 4/54 (7.4%) and 2/54 (3.7%) of deer liver samples were deficient and marginally deficient, respectively, and 2/48 (4.2%) and 1/48 (2.1%) of moose liver samples were deficient and marginally deficient, respectively (Figure 1).

In moose, kidney copper concentrations were significantly higher in Mainland West moose compared to Cape Breton moose (Table 3). Yearling moose had significantly lower copper concentrations in liver (geometric mean: 95.2 µg/g [95%CI: 31.4 - 288.5]) compared to adult moose (geometric mean: 338.4 µg/g dw [95% CI: 280.6 - 408.0]) (p<0.05). Liver copper concentrations in yearlings were also lower than in calves (geometric mean: 299.5 µg/g dw [95%CI:194.4 - 461.5]), however, the difference was not statistically significant (p= 0.08).

Manganese

Liver manganese concentrations were higher in deer compared to moose (p<0.05). In relation to reference values for domestic cattle, 2/54 (3.7%) and 15/54 (27.8%) of deer liver samples were deficient and marginally deficient, respectively, and 2/48 (4.2%) and 20/48 (41.7%) of moose liver samples were deficient and marginally deficient, respectively (Figure 1).

In moose, females had significantly higher liver manganese concentrations (geometric mean: 12.9 µg/g dw [95%CI: 11.1 - 14.9]) compared to males (geometric mean: 9.1 µg/g dw [95% CI: 6.7 - 12.5]) (p<0.05).

Nickel

In moose, liver and kidney nickel concentrations were significantly higher in Cape Breton compared to Mainland East and Mainland West (Tables 2 and 3).

Table 4. Cadmium concentrations (µg/g dry weight) in deer liver and in moose liver and kidney by age group.

species	tissue		calf <12 months	yearling 12 - 24 months	adult >24 months	overall
deer	liver	geom. mean (95% CI) [a]	-	0.8 (0.4 - 1.5)	1.2 (0.8 - 1.9)	1.1 (0.8 - 1.5)
		min	0.64	0.05	0.1	0.05
		max	4.1	6.5	28.1	28.1
		n [b]	2	19	30	54
moose	liver	geom. mean (95% CI)	5.3 (3.4 - 8.3)	3.2 (0.7 - 13.8)	6.6 (4.9 - 8.9)	5.8 (4.0 - 8.3)
		min	3.3	<0.05	1.2	<0.05
		max	7.4	19	32.1	51.9
		n	5	11	25	48
moose	kidney	geom. mean (95% CI)	-	54.4 (25.0 - 83.7)	62.3 (34.0 - 114.0)	60.4 (40.3 - 90.6)
		min	16.6	14.3	27	14.3
		max	83.3	86.8	303.1	346.1
		n	2	6	11	21

[a] geometric mean (95%confidence interval): mean is calculated for sample sizes greater than 2, for sample sizes of 2, minimum and maximum values are listed.

[b] Age data were not collected in all animals. The number of animals listed in the column 'overall' include those for which age data were collected as well as those lacking age data.

Selenium

Liver selenium concentrations were higher in deer compared to moose ($p<0.001$). In relation to reference values for domestic cattle, 2/54 (3.7%) and 6/54 (11.1%) of deer liver samples were deficient and marginally deficient, respectively, 19/48 (39.6%) and 15/48 (31.3%) of moose liver samples were deficient and marginally deficient, respectively (Figure 1), and 6/21 (28.6%) of moose kidney samples were deficient or marginally deficient (<2.1 µg/g dw) (Puls 1994).

Liver selenium concentrations in Cape Breton deer were significantly higher than in Mainland East and Mainland West deer (Table 1). There was also a significant positive association between age (in months) and selenium concentrations in liver (r=0.49, $p<0.05$). This association approached significance when the deer were grouped by age category (fawn, yearling and adult) (one-way ANOVA, F=3.08, $p=0.06$). Male deer had significantly higher concentrations of selenium in liver (geometric mean: 1.70 µg/g dw [95%CI: 1.33 - 2.17]) compared to females (1.28 µg/g dw [95% CI: 1.13 - 1.45]), ($p<0.05$).

Zinc

In relation to reference values for domestic cattle, 20/54 (37.0%) and 19/54 (35.2%) of deer liver samples were deficient and marginally deficient, respectively, and 28/48 (58.3%) and 6/48 (12.5%) of moose liver samples were deficient and marginally deficient, respectively (Figure 1).

Discussion

Based on tissue concentrations, evidence for widespread deficiencies or toxicities of trace elements in Nova Scotia moose or deer is lacking. However, relative to reference values for cattle, cobalt, copper, manganese, selenium and zinc levels in some animals are deficient or marginally deficient. These findings must be interpreted with caution as species differences are known to exist for trace elements and reference values for moose, deer or other free-ranging cervids are not available. In general, the levels of these trace elements are within ranges reported for other cervid populations in North America and Europe with the exception of liver zinc concentrations in deer and moose and possibly liver cobalt concentrations in deer and moose. Only one other published report for cobalt concentrations in a small number (n=24) of moose from Sweden was found for comparison.

The range of normal trace element values for wildlife species is generally unknown and many trace elements have been shown to vary with age, gender, season of collection and health status. Discussions for each trace element of concern include: tissue concentrations and relevant demographic differences found in relation to levels in cervid populations elsewhere; clinical signs of deficiency in domestic ruminants or wildlife species where reported; and tissue samples of particular diagnostic value. General conclusions and recommendations follow.

Cobalt

Frank et al. (2004) cited cobalt/vitamin B12 deficiency as a contributing factor in observed “moose sickness” in Nova Scotia. Blood and liver samples were collected from 17 affected moose in 1998 - 2000. However, it is unclear from the published article what, if

any, clinical signs or lesions the sampled moose exhibited. The diagnosis of cobalt/vitamin B12 deficiency appears to have been based on historical reports of diseased animals with signs of weakness, emaciation and neurological lesions. Frank et al. (2004) reported median cobalt concentrations of 0.09 µg/g dw (range: 0.01 - 0.29) in liver and 0.06 µg/g dw (range: 0.01 - 0.19) in kidneys (converted from ww). Similar concentrations were found in apparently healthy moose and deer in this study (Tables 1, 2 and 3).

Based on criteria for domestic cattle (Puls, 1994; Radostits et al., 2000), of the moose livers sampled by Frank et al. (2004), 7/17 had marginal or deficient levels of cobalt (<0.02 µg/g wet weight) and 9/17 had marginal or deficient levels of vitamin B12 (<0.25 µg/g wet weight). Based on these same criteria, deficient levels of cobalt were also found in 7% of deer livers and 29% of moose livers in the present study. Overall, liver cobalt concentrations were higher in deer compared to moose in the present study.

Frank et al. (2004) found liver cobalt concentrations in Nova Scotia moose to be lower than liver cobalt concentrations in moose from Sweden and Alaska (unpublished data). Liver cobalt concentrations in apparently healthy moose (median 0.42 µg/g dw [range: 0.26 - 0.60]) and moose thought to be copper deficient (median 0.46 µg/g dw [range: 0.29 - 0.68]) (converted from ww) from Sweden (Frank et al., 2000) were higher than levels found in both moose and deer in the present study. We were unable to find any other published reports of cobalt or vitamin B12 concentrations in free-ranging cervid populations for comparison.

Frank et al. (2004) cites McBurney et al. (2001) to support the hypothesis that cobalt/vitamin B12 deficiency is a problem in the Nova Scotia moose population. However, the moose McBurney et al. (2001) described with neurological disease of uncertain etiology were in excellent body condition in contrast to the emaciation described by Frank et al. (2004). Also, any emaciated moose diagnosed with neurological disease in Nova Scotia has had parelaphostrongylosis and meningoencephalitis to account for their debilitated physical condition (S. McBurney, pers comm). Therefore, without other supporting evidence, the case for cobalt/vitamin B12 deficiency in Nova Scotia moose presented by Frank et al. (2004) appears incomplete.

Cobalt is required for the synthesis of vitamin B12. In domestic ruminants, cobalt deficiency results in inappetence and loss of body weight, emaciation, weakness, decreased growth, unthrifty appearance, diarrhea, and anemia (Smith, 1990; Radostits et al., 2000). Young sheep (*Ovis aries*) and cattle are more seriously affected than adults. In sheep, cobalt deficiency is associated with white liver disease. Cobalt deficiency in cattle and sheep occurs only on soils deficient in cobalt. Both liver cobalt and vitamin B12 levels are felt to be valuable diagnostic tests (Smith, 1990; Puls, 1994; Radostits et al., 2000). Liver vitamin B12 concentration is closely associated with cobalt status in ruminants, however, the effects of starvation can tend to increase tissue vitamin B12 concentrations (Smith, 1990). Thus, tissue vitamin B12 concentrations may appear normal with cobalt deficiency in anorectic animals. Methylmalonic acid (MMA) levels in plasma and urine are also useful diagnostically. An elevated plasma concentration of MMA is an early indicator of functional vitamin B12 deficiency (Puls, 1994; Radostits et al., 2000).

Although the evidence for cobalt/vitamin B12 deficiency in Nova Scotia moose cited by Frank et al. (2004) is lacking, certainly, the possibility of cobalt/vitamin B12 deficiency

should not be entirely dismissed. The continued collection of serum and liver samples from both healthy and diseased animals for cobalt and vitamin B12 determination is recommended in moose throughout the province. As cobalt deficiency in ruminants is characterized in part by emaciation, the determination of body condition at the time of sample collection is strongly recommended.

Copper

Apparent or possible copper deficiency has been reported in free-ranging moose from Alaska (Flynn et al., 1977; O'Hara et al., 2001), northwest Minnesota (Custer et al., 2004) and Sweden (Frank et al., 1994; Frank, 1998).

In general, liver copper concentrations in this study appeared to be adequate to high, although a small number of moose and deer had deficient or marginally deficient liver copper concentrations relative to reference values for domestic cattle. Among age groups, yearlings had lower liver copper concentrations than adults.

Comparable levels of copper were found in other studies on apparently healthy moose and caribou (*Rangifer tarandus*) elsewhere. Froslie et al. (1984) found that moose liver mean concentrations from various locations in Norway ranged from 80.5 to 354 µg/g dw (converted from ww). Caribou and reindeer (*Rangifer tarandus*) from Greenland (liver geometric means: 76.3 to 248.5 µg/g dw [converted from ww]) (Aastrup et al., 2000), caribou from northern Alaska (liver geometric means: 19.4 to 289.8 µg/g dw; kidney geometric means: 11.9 to 45.5 µg/g dw) (O'Hara et al., 2003) and caribou from the Northwest Territories (liver means: 51.9 to 120.8 µg/g dw; kidney means: 27.8 to 49.7 µg/g dw) (Elkin and Bethke, 1995) also had concentrations comparable to those of moose and deer in this study.

In moose with apparent copper deficiency, mean concentrations were much lower than concentrations found in moose and deer in this study (Frank et al., 1994; O'Hara et al., 2001; Custer et al., 2004). Frank (1998) reported decreased liver copper concentrations secondary to increased liver concentrations of molybdenum in Swedish moose.

In a case report of an adult cow moose exhibiting severely incoordinated and ataxic movements in Sweden, liver (13.7 µg/g dw) and kidney (7.7 µg/g dw) (converted from ww) concentrations were very low (Rehbinder and Petersson, 1994).

Free-ranging deer in Texas and Tule elk (*Cervus elaphus nannodes*) in California had antler deformities associated with low hepatic copper levels (Gogan et al., 1988). Flynn et al. (1977) found faulty hoof keratinization and reduced reproductive rates in a subpopulation of Alaskan moose with low levels of copper in blood and hair samples. Specific clinical signs of copper deficiency in individual animals were not noted by Custer et al. (2004), however, reduced liver copper concentrations in bog and forest habitats in northwestern Minnesota coincided with reduced calf-to-cow ratios in these areas compared to agriculture and prairie habitats. In Sweden, clinical signs of moose disease reportably associated with copper deficiency and/or molybdenosis include diarrhea, anorexia, emaciation, osteoporosis and loss of hair colour (Frank et al., 1994; Frank,1998). O'Hara et al. (2001) found moose in the Colville River area to be deficient in copper and other minerals based on hair, whole blood and serum samples, however, due to the remote location, the sampled animals were not examined for clinical signs or lesions associated

with the apparent copper deficiency.

In domestic ruminants, copper deficiency is defined as primary when the diet is deficient in copper or secondary when copper absorption or metabolism is adversely affected (Smith, 1990). In general, young animals and fetuses are more susceptible to copper deficiency than adults and cattle are more susceptible than sheep (Smith, 1990; Radostits et al., 2000). Signs of copper deficiency in cattle include: unthriftiness, loss of milk production, anemia, change in hair coat colour in adults as well as poor growth and increased susceptibility to bone fractures in calves. Diarrhea can be seen in secondary copper deficiency (in association with increased molybdenum and/or sulfur). In sheep, abnormalities of the wool are the first observed signs. In unweaned lambs, severe incoordination and paresis resulting in recumbency (termed “enzootic ataxia”) can be seen. Osteoporosis can be seen in less affected lambs.

In farmed red deer (*Cervus elaphus*), neurological disease characterized by ataxia, swaying of the hindquarters and eventual loss of ability to use the hind legs can be seen in animals of all ages. Spinal cord demyelination and midbrain neuronal degeneration are characteristic of this manifestation of copper deficiency in red deer (Radostits et al., 2000).

Liver concentrations are more informative for copper status than kidney concentrations. Hair levels can also be used to help in diagnosis of copper deficiency (Puls, 1994; Radostits et al., 2000). Serum copper levels alone are difficult to interpret. Plasma ceruloplasmin levels have been shown to correlate strongly with serum copper levels in cattle and sheep (Radostits et al., 2000).

Several trace elements have been shown to interact with copper including molybdenum, sulfur, iron, and zinc (high dietary levels of these elements can lead to secondary copper deficiency). Copper and selenium deficiencies frequently occur concurrently, and high dietary cadmium may also interact with copper (Puls, 1994; Smith, 1990; Radostitis et al., 2000).

Notably, yearling moose in this study had lower liver copper concentrations than adults, however levels considered deficient were found in only a small number of animals. Since antler and hoof deformities have been reported in Nova Scotian moose (S. McBurney pers comm), continued monitoring of the copper status of healthy and diseased moose, particularly yearlings, is recommended. Where copper deficiency is suspected (such as with abnormal hoof keratinization or antler deformities), analysis of liver samples for sulfur and molybdenum may aid in diagnosis and interpretation of copper results.

Zinc

Zinc concentrations in Nova Scotia moose and deer appear on the lower end of, or lower than, the range of values reported in free-ranging cervids elsewhere. In relation to reference values for domestic cattle, approximately 70% of deer and moose livers in this study had deficient or marginal concentrations of zinc.

Zinc concentrations from moose and deer in this study were in the lower end of the range found in moose from Norway (liver means: 73.5 to 112.0 µg/g dw [converted from ww]) (Froslie et al., 1984), in caribou and reindeer from Greenland (liver geometric means: 81.2 to 103.3 µg/g dw [converted from ww]) (Aastrup et al., 2000), in caribou from northern Alaska (liver geometric means: 77.0 to 246.4 µg/g dw; kidney geometric means: 58.1 to

132.0 µg/g dw [converted from ww]) (O'Hara et al., 2003) and in caribou from the Northwest Territories (liver means: 75.8 to 114.1 µg/g dw; kidney means: 96.8 to 123.5 µg/g dw) (Elkin and Bethke, 1995).

Zinc concentrations from moose and deer in this study were lower than those found in apparently healthy moose (liver mean: 256.6 µg/g dw; kidney mean: 117.2 µg/g dw) and copper-deficient moose (liver zinc mean: 232.9 µg/g dw; kidney zinc mean: 150.5 µg/g dw [converted from ww]) from northern Alaska (O'Hara et al., 2001), in apparently copper-deficient moose in northwestern Minnesota (liver geometric means:167 µg/g dw in agriculture and prairie habitat and 219 µg/g dw in bog and forest habitat) (Custer et al, 2004) and in apparently healthy moose (liver median concentration: 119.4 µg/g dw [converted from ww]) from Sweden (Frank et al., 2000). Frank et al. (2004) found higher median concentrations in Nova Scotia moose (161 µg/g dw in liver n=17 and 175 µg/g dw in kidneys n=16 [converted from ww]) compared to moose and deer in this study.

In naturally occurring zinc deficiency in domestic cattle, hair loss and the development of thick, wrinkled skin are the primary clinical manifestations. Affected animals are often stunted in growth and below average body condition. Experimental zinc deficiency has also resulted in stiffness, swelling of the joints and oral ulcers and hemorrhages (Radostits et al., 2000). Reduced hoof strength has also been reported (Puls, 1994).

Serum and liver zinc levels are useful in the diagnosis of zinc deficiency in domestic animals. Hair zinc levels are also useful but are not considered to be sufficiently accurate as an indicator of zinc status (Radostits et al., 2000).

Zinc interacts with other trace elements including cadmium and copper. For example, high dietary copper reduces zinc in liver (Puls, 1994). Overall, it appears that tissue zinc concentrations in Nova Scotia moose and deer are at the low end of, or lower than, the range of values reported elsewhere. It is unclear whether the levels are sufficiently low to result in clinical signs in individual animals or at the population level. Continued monitoring of zinc levels in deer and moose in Nova Scotia is recommended, particularly in diseased animals.

Manganese

Based on reference values for domestic cattle, approximately 30% of deer liver samples and 45% of moose liver samples had deficient or marginally deficient manganese concentrations. However, it appears unlikely that manganese concentrations are truly deficient in deer and moose in Nova Scotia. Manganese concentrations in deer and moose in this study were similar to those of apparently healthy caribou from the Northwest Territories (liver means: 8.6 to 15.9 µg/g dw; kidney means: 9.0 to 18.6 µg/g dw) (Elkin and Bethke, 1995), moose from northwestern Minnesota (liver geometric means: 7.9 and 8.0 µg/g dw) (Custer et al, 2004) and apparently healthy moose (liver median: 15.0 µg/g dw) and moose affected with a 'mysterious' disease (liver median: 14.0 µg/g dw [converted from ww]) from Sweden (Frank et al., 2000).

In cattle, manganese deficiency generally manifests as skeletal abnormalities such as calves with congenital limb deformities. Other common syndromes in cattle include infertility and calves with poor growth, dry hair coat and loss of coat colour (Radostits et al.,

2000). In domestic animals, no simple, single diagnostic test has been found to detect manganese deficiency. Hair and serum levels of manganese have been used to evaluate body stores, however, hair levels in female cattle drop at calving and low levels of serum manganese have been reported in normal cattle. Therefore, such samples may be of limited value as diagnostic tests (Radostits et al., 2000), particularly in free-ranging populations in which the range of normal values is not known.

Selenium

Overall, tissue selenium concentrations in deer and moose in this study were similar to those reported in other cervid populations elsewhere. In this study, moose had lower liver selenium concentrations than deer. Based on reference values commonly reported for cattle, 15% of deer liver samples were deficient or marginal compared to 70% of moose liver samples (Puls, 1994; Radostits et al., 2000).

Selenium concentrations in deer and moose in this study were similar to those of caribou and reindeer from Greenland (liver geometric means: 0.30 to 3.4 µg/g dw [converted from ww]) (Aastrup et al., 2000), moose from Norway (liver means: 0.28 to 3.2 µg/g dw [converted form ww]) (Froslie et al, 1984), moose from northwestern Minnesota (liver geometric means:1.2 and 2.7 µg/g dw) (Custer et al, 2004) and from Sweden (liver medians: 0.33 to 1.02 µg/g dw [converted from ww]) (Galgan and Frank, 1995).

Selenium is an essential nutrient and the role of selenium in animal health is complex. Diseases due to selenium deficiency are recognized in livestock worldwide, generally in association with selenium-deficient soil (Radostits et al., 2000). Selenium deficiency in young animals can manifest as nutritional muscular dystrophy resulting in weakness, muscle stiffness, inability to stand and possibly sudden death (Radostits et al., 2000). Selenium deficiency has also been associated with decreased resistance to infectious diseases, ill-thrift in sheep and retained fetal membranes in cattle (Radostits et al., 2000). Selenium is reported to interact with a variety of other trace elements (eg. copper).

Soils throughout eastern North America, including Nova Scotia, are known to be generally deficient in selenium. It is unclear what effect subclinical deficiencies of selenium may have on the health of wildlife in these areas either directly or through interaction with other trace elements. Further exploration of these relationships is beyond the scope of this report. Where selenium deficiency is suspected (clinical signs observed) concentrations in liver are a better indicator of selenium status than those in kidneys.

Cadmium

Cadmium concentrations in moose kidney analysed in this study are among the highest reported for the species. Although the highest concentrations found were in adults from the Mainland West region, calves and yearlings from all regions sampled had high kidney concentrations when compared to those found in young animals in other studies.

There has been considerable effort made to document tissue cadmium concentrations in moose, deer and other wildlife species throughout North America and Europe for a variety of reasons including: concerns about environmental contamination; potential human health risk related to consumption of organ meats from these animals; and,

to a lesser extent, potential biological effects from cadmium toxicity in the animals.

In this discussion, concentrations and demographic differences found are assessed in relation to levels elsewhere and to reported threshold levels for biological effects in animals.

As found in other studies, levels of cadmium in deer liver were lower than those found in moose liver (Crête et al, 1987; Glooschenko et al., 1988). Suggested reasons for this difference include: different dietary habits of moose and deer and higher quantity of forage consumed by the larger moose.

In this study, kidney and liver cadmium concentrations were higher in Mainland West moose compared to Mainland East moose and Cape Breton moose. However, this result should be interpreted with caution as the number of Mainland West moose sampled was small (n=4). Regional differences in cadmium levels were also seen in moose in Ontario (Glooschenko et al., 1988), Norway (Froslie et al., 1986; Scanlon et al., 1986), Québec (Crête et al, 1987; Paré et al., 1999) and northwestern Minnesota (Custer et al., 2004), but not in moose from Newfoundland (Brazil and Ferguson, 1989) or Maine (Scanlon et al., 1986).

Elsewhere, differences in buffering capacity of the soil and the degree of environmental acidification have been cited as potential explanations for regional differences seen in the degree of cadmium contamination in free-living moose (Froslie et al., 1986; Crête et al, 1987; Glooschenko et al., 1988). However, other factors such as the proximity of a site to sources of industrial pollution (eg. smelters) (Scanlon et al., 1986; Paré et al.,1999), the degree of natural mineralization of soils (Dietz et al., 1998; Larison et al., 2000), and the composition and diversity of forage species in an area (Ohlson and Staaland, 2001) may be as important as, or more important than, environmental acidification on the degree of cadmium accumulation in wildlife (Crête et al, 1987; Glooschenko et al., 1988, Scheuhammer 1991). For example, increased access to plants such as *Salix* spp. which naturally accumulate high concentrations of cadmium can influence the degree of cadmium accumulation in herbivore species (Dietz et al., 1998; Larison et al., 2000; Ohlson and Staaland, 2001). Other habitat differences may also play a role in regional variation in cadmium levels. Custer et al. (2004) found higher levels of cadmium in livers of moose from agricultural areas of northwestern Minnesota compared to moose from bog and forest habitats.

A detailed discussion of bedrock composition throughout Nova Scotia in relation to cadmium levels can be found in Roger (2002).

Because cadmium accumulates over time and has a long biological half life (Dietz et al., 1998), positive correlations between age of the animal and cadmium concentration in kidney and/or liver have been fairly consistent findings in many studies in deer, moose and other free-ranging ungulates (Froslie et al., 1986; Scanlon et al., 1986; Crête et al, 1987; Glooschenko et al., 1988; Brazil and Ferguson 1989; Gamberg and Scheuhammer, 1994; Paré et al., 1999; Gustafson et al., 2000; O'Hara et al., 2001; Custer et al., 2004). In this study, however, no correlation was seen between age (when measured by months or when categorized by age group) and cadmium concentrations in moose or deer. Although the highest concentrations found were in adults, the variability within each age group was high (Table 4).

Gender differences in tissue cadmium concentrations were also seen in moose from Maine (Scanlon et al., 1986), Québec (Crête et al, 1987) and New England (Gustafson et al., 2000) where lower mean concentrations were found in older females compared to older males. Those findings are in contrast to this study where females had higher mean kidney cadmium concentrations than males. Gamberg and Scheuhammer (1994) found that cadmium levels in caribou and muskoxen (*Ovibos moschatus)* were higher in spring compared to fall. Crête et al. (1989) also found higher tissue cadmium levels in caribou in winter compared to fall.

As discussed in a number of studies, age, gender, location, species and season have all been found to influence cadmium concentrations. Thus, direct comparison of tissue concentrations among studies can be difficult. However, in order to put the tissue concentrations found in this study in a context relative to other regions, the mean concentrations of cadmium found are compared to those in white-tailed deer and moose from elsewhere in North America and Europe (Tables 5 and 6, respectively).

Liver cadmium concentrations in deer were similar to those found in white-tailed deer in other regions of eastern North America (Table 5).

Overall mean concentrations in liver and kidney from moose in Nova Scotia (this study) are higher than those reported in Newfoundland, Minnesota (liver), Maine and Norway. Overall mean concentrations are similar to those in Québec and below the maximum mean concentrations reported in New England (liver), New Brunswick, Ontario and Alaska (liver and kidney) (Table 6). The highest concentration found in this study was 346.1 µg/g dw in kidney. Similarly high levels have also been reported in moose in western Québec (440.1 µg/g dw) and concentrations of cadmium as high as 1380 µg/g dw have been found in kidney tissue from moose in the Yukon (M. Gamberg, pers comm).

Although the number of young animals sampled in this study was low, kidney cadmium concentrations in moose calves and yearlings are among the highest reported for these age groups. Comparable levels of kidney cadmium were reported in moose calves and yearlings sampled from the area surrounding the community of Rouyn-Noranda, Québec, which contains a copper smelter (calves 14.9 - 47.3 µg/g dw; standard error [SE]: 3.9 - 30.9; yearlings 60.0 - 100.1 µg/g dw; SE: 9.7 - 31.7) (Paré et al., 1999). Mean kidney cadmium concentrations in adults from that area were higher (141.9 - 173.4 µg/g dw; SE:22.1 - 26.9) than levels found in this study (Table 4). Paré et al. (1999) attributed the high level of cadmium contamination found in moose of all ages surrounding Rouyn-Noranda, Québec, to the natural mineralization of the area and historical and recent human contributions of cadmium to the environment.

The relatively elevated kidney cadmium concentrations found in calves and yearlings from all regions of Nova Scotia is difficult to interpret and may be a factor of small sample size. Explanations offered in the literature for high levels of cadmium in certain groups of animals include: proximity to a point source of pollution (eg. smelter) (Paré et al., 1999); increased reliance of the animals on cadmium-accumulating plants such as *Salix* spp. (Larison et al., 2000); high natural level of mineralization in the soil (Larison et al., 2000; Dietz et al., 1998); regional differences in levels of atmospheric fallout of cadmium and/or acidification of the environment resulting in increased bioavailability of cadmium (Froslie et al., 1986; Dietz et al., 1998). Because cadmium bioaccumulates over time, older animals

Table 5. Liver concentrations of cadmium reported in white-tailed deer from eastern North America.

Region	Year	*n*	Concentration [a]	Range [a]	Reference	Notes [c]
Nova Scotia	2000	54	2.2 (1.1 - 3.2) 1.1 (0.8 - 1.5)	0.05 - 28.1	this study	mean (95% CI) geometric mean (95% CI)
New Brunswick [b]	1986-87	123	2.6 (2.2) min 8.3 (7.3) max		Ecobichon et al., 1988	mean (SD) by county
Québec	1985	181	1.0 (0.1) min 2.6 (0.2) max		Crête et al., 1987	mean (SE) by gender and location
Ontario [b]	1984-85	243	0.4 (0.1) min 6.7 (1.4) max		Glooschenko et al., 1988	mean (SE) by age and location
Manitoba	1986-89	105	0.6 (0.5)	0.1-1.8	Crichton and Paquet, 2000	mean (SD)
New Jersey	1986-87	86	4.0 2.0	0.1 - 23.2	Stansley et al., 1991	mean geometric mean

[a] Concentration in µg/g dry weight.
[b] Concentrations converted from wet weight based on estimate of % moisture of 71.4% (conversion factor of 3.5)
[c] Overall mean values are listed where available, otherwise minimum and maximum mean values for specific groups of animals as presented in the reference studies are listed; CI=confidence interval; SD=standard deviation; SE=standard error.

Table 6. Liver and kidney concentrations of cadmium reported in moose from North America and Norway.

Region	Year	Liver			Kidney			Reference	Notes [c]
		n	Concentration [a]	Range	*n*	Concentration [a]	Range		
Nova Scotia	2000	48	8.9 (6.3 - 11.6) 5.7 (3.9 - 8.2)	0.05 - 51.9	21	91.6 (46.9 - 136.2) 60.4 (40.3 - 90.6)	14.3 - 346.1	this study	mean (95%CI) geom. mean (95% CI)
New Brunswick [b]	1986-87	84	5.8 (4.2) min 64.4 (33.3) max		84	31.8 (23.6) min 225.8 (137.2) max		Ecobichon et al.,1988	mean (SD) by county
Newfoundland [b]	1987-88	79	3.6 (3.0)		88	19.3 (18.9)		Brazil and Ferguson, 1989	mean (SD?)
Ontario [b]	1984-85	187	1.4 (0.7) min 20.0 (4.6) max		197	2.1 (0.4) min 179.9 (18.6) max		Glooschenko et al., 1988	mean (SE) by age group
western Québec	1986	474	11.2 (0.7)	0.43 - 232.1	276	72.4 (3.9)	0.01 - 440.1	Paré et al., 1999	mean (SE)
Québec	1985	306	2.9 (0.4) min 15.9 (3.5) max		125	31.8 (8.0) min 100.0 (12.0) max		Crête et al., 1987	mean (SE) by gender and location
Manitoba	1986-89	228	1.2 (0.9)	0.01 - 4.7	228	6.8 (6.0)	0.2 - 38.0	Crichton and Paquet, 2000	mean (SD)
Maine	1983	160	5.6 (0.6)		22	26.8 (4.3)		Scanlon et al., 1986	mean(SE)
New England [b]	1988-98	253	2.3 (4.1) min 17.0 (3.7) max	0.7 - 56.6	103	31.3 (4.4) min 41.8 (4.0) max		Gustafson et al., 2000	geom. mean (SE) by age, gender
Minnesota	1998-99	79	1.4 (0.3) min 2.4 (0.5) max					Custer et al., 2004	geom. mean (SE) by location
Alaska [b]	1995	22	11.0 (8.8) min 41.6 max		22	75.6 (72.8) min 255.9 max		O'Hara et al., 2001	mean (SD) by location
Telemark, Norway	1983-84	48	1.3 (0.2) min 2.1 (0.3) max		47	8.4 (1.2) min 20.5 (2.6) max		Scanlon et al., 1986	mean (SE) by location

[a] All concentrations in µg/g dry weight;

[b] Concentrations converted from wet weight based on estimate of % moisture of 71.4% (conversion factor of 3.5)

[c] Overall mean values are listed where available, otherwise minimum and maximum mean values for specific groups of animals as presented in the reference studies are listed; CI=confidence interval; geom. mean=geometric mean; SD=standard deviation; SE=standard error.

tend to accumulate more cadmium in their tissues compared to younger animals, thus, the geographical or geological explanations for increased tissue cadmium concentrations (mineral-rich soil, environmental acidification, point source of pollution) should affect all ages of animals in an area equally. Dietary differences between younger and older animals may be a plausible explanation, however, willow, a known cadmium accumulator, is a preferred browse species for moose (Renecker and Schwartz, 1998) and therefore would presumably be preferentially consumed by moose of all ages.

Although an interesting discussion, on a practical level, a greater sample size of calves and yearlings would be necessary to confirm the relatively high levels of cadmium in these age groups. As well, in relation to potential biological effects in these young cohorts, it is unlikely that these concentrations are impacting the health of these animals because much higher concentrations have been reported in moose with no observable biological effects.

The primary target of chronic cadmium toxicity in mammals and birds is the kidney (Scheuhammer, 1987; Alden and Frith, 1991). The earliest light microscopic change in this organ is proximal tubular necrosis (Alden and Frith, 1991). In addition to the renal toxicity, there is also evidence that exposure to cadmium can result in disturbances in calcium balance and decreases in bone density (Taylor et al., 1999). Although large mammals such as ungulates and seals have been shown to have some of the highest levels of cadmium recorded in wildlife (Muir et al., 1997), studies documenting biological effects associated with high cadmium concentrations in tissues in wildlife populations are rare, and a specific threshold level for cadmium toxicity in most wildlife species, including deer and moose, has not been determined.

In experimental animals, the threshold for renal tubular injury has been shown to vary with species and route of exposure (Alden and Frith, 1991). The threshold for significant renal tubular damage in mammals and birds is generally reported as 100-200 µg/g ww (approximately 350-700 µg/g dw) (Cooke and Johnson, 1996). However, lower threshold concentrations have also been published. Based on a review of the literature on experimental and domestic mammals, Outridge et al.(1994) reported a threshold concentration of 30 µg/g ww (approximately 105 µg/g dw) for kidney damage in mammals. More recently, microscopic and biochemical evidence of damage has been reported in rats (*Rattus* sp.) with experimental chronic cadmium exposure at renal concentrations as low as 2 - 4 µg/g ww (7 - 14 µg/g dw) (Brzóska et al., 2003).

Among free-ranging wildlife species, Larison et al. (2000) examined white-tailed ptarmigan (*Lagopus leucurus*) from the Colorado ore belt and found that those with renal cadmium concentrations >100 µg/g ww (approximately 350 µg/g dw) had histopathological evidence of renal injury. Ptarmigan with renal cadmium concentrations above this level also had reduced concentrations of skeletal calcium compared to controls.

In a study conducted on ringed seals (*Phoca hispida*) from Greenland, Sonne-Hansen et al. (2000) found no evidence of cadmium-induced renal toxicity or skeletal demineralization. They found histopathological damage in kidneys of 10% of the animals but concluded that these changes were not obviously cadmium-induced as lesions were found in animals of various ages and with various renal cadmium concentrations. Renal cadmium concentrations of the seals examined ranged from 0 - 868 µg/g dw (converted

from wet weight).

Among published studies on cadmium in ungulates, evidence for biological effects with high concentrations is lacking. Paré et al. (1999) found no lesions characteristic of renal disease in 33 kidney samples with a mean cadmium concentrations of 123.1 (+/- 17.98) µg/g dw submitted for histopathological examination. O'Hara et al. (2003) found no histopathologic evidence of renal lesions in caribou kidneys with cadmium concentrations of 1.9 - 115.5 µg/g dw (converted from wet weight) from northern Alaska. Kidneys from two moose sampled in this study (cadmium concentrations 96.1 and 346.2 µg/g dw) were submitted to the Atlantic Veterinary College Diagnostic Services for histopathological examination and no evidence of renal proximal tubular lesions was found, although autolysis and freezing artifacts may have masked subtle changes in the tissues (S. McBurney, pers comm).

Although evidence for biological effects due to cadmium exposure in wildlife species is lacking, the possibility that elevated cadmium concentrations in individual animals may lead to subclinical or clinical disease cannot be dismissed.

Continued monitoring of moose tissue cadmium concentrations in conjunction with detailed health assessment of individuals including histopathological examination of kidneys is recommended.

Conclusions

A good foundation of trace element data has been collected for deer and moose in Nova Scotia, however, some age groups and regions are under-represented. Tissue concentrations of trace elements in deer and moose in Nova Scotia appear to be generally similar to levels reported elsewhere in North America and Europe with the exception of zinc and possibly cobalt. Liver concentrations of zinc in both deer and moose are at the low end of, or lower than, the range of values reported elsewhere for moose and caribou. Liver cobalt concentrations in moose and deer were lower than levels reported for moose in Sweden. No other published reports of cobalt or vitamin B12 concentrations in free-ranging cervid populations were found for comparison.

Kidney cadmium concentrations are high in some Nova Scotia moose, however, similar or higher concentrations have been reported in other regions. To date, individual or population-level health effects in relation to elevated tissue cadmium levels have not been reported in free-ranging ungulates in North America including Nova Scotia.

In relation to reported reference values for domestic cattle, tissue levels of trace elements in deer and moose were generally within normal ranges, however, marginal or deficient levels of selenium, copper, cobalt and zinc were found in some animals. Deficiencies in these trace elements can lead to clinical signs in domestic animals. At the present time, there appears to be little supporting evidence that clinical deficiencies of any of these trace elements are occurring in Nova Scotia moose or deer populations. However, the possibility that marginal or deficient levels of these or other trace elements and high levels of cadmium may impact the health of individual animals either directly or through interactions with other factors (eg. infectious and non-infectious diseases, harsh environmental conditions, habitat limitations) cannot be dismissed.

Recommendations

Recommendations are aimed primarily at moose because of the endangered status of the Mainland population, however, because deficient and marginal levels of some trace elements were also found in deer, most of the recommendations (with the exception of those related to cadmium levels) could also apply to the deer population.

In order to help establish “normal” values for these populations and to identify possible health effects, if any, in relation to trace element deficiencies or toxicities (cadmium), continued monitoring of tissue concentrations of selected trace elements is recommended.

Specifically, in dead animals, additional samples of liver tissue (for selenium, copper, zinc and cobalt [+/- vitamin B12] analysis) and kidney tissue (for cadmium analysis) should be collected. In fresh carcasses, a sample of kidney tissue fixed in formalin for histopathological examination should also be taken.

For each animal sampled, demographic data including location, age (as specific as possible), gender and species are essential. As many disease conditions related to trace element deficiencies can lead to loss of body condition or emaciation, for each animal sampled, body condition should be assessed.

The timely necropsy of obviously diseased animals is also strongly recommended.

If live animals are being handled, serum samples can be taken for selenium, zinc, copper and cobalt (+/- vitamin B12) analysis. Hair samples for zinc and copper analysis may also be useful. The collection of detailed demographic data along with an evaluation of body condition are also important in live animals.

In general, increased representation of yearlings and calves/fawns, particularly for liver copper and kidney cadmium concentrations is recommended. This would help to ascertain whether concentrations found in the younger cohorts in this study are true representations of cadmium and copper distribution in these age groups in the larger population.

If possible, the collection of additional samples from Mainland moose is recommended to determine whether the regional differences seen in the animals sampled are in fact present.

It is important to note that most tissue and serum samples can be archived (frozen) for analysis at a later date pending results of additional monitoring of the populations. In addition, factors that impact the health of wildlife are complex and often interrelated, thus, it cannot be overemphasized that there is little value in collecting trace element data alone without supporting demographic and health information from each animal sampled. This supporting data is essential for meaningful interpretation of trace element data.

As some regional and age differences were found in individual trace element concentrations, additional statistical analyses of the collected data may be useful (eg. principal components analysis) to further explore interactions among trace elements and demographic variables.

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