

## Investigation of Heavy Metals in a Large Mortality Event in Caribou of Northern Alaska

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**ABSTRACT.** We measured element concentrations (As, Cd, Cu, Pb, Zn, Fe) and body condition (gross and histologic endpoints) of animals from a caribou (*Rangifer tarandus*) mortality event that occurred in Alaska, in the area of Point Hope and Cape Thompson (including the Chariot site), in 1995. These were compared to results from hunter-killed caribou from reference sites (Barrow and Teshekpuk Lake, Alaska) and from the area of a mine (Red Dog Mine) to determine whether heavy metals had played a role in the mortality event or whether any elements were at concentrations of concern for human consumers. Starvation and malnutrition were major factors leading to death or severe weakness, as very little or no fat (very low body condition scores) and serous atrophy of fat (observed as watery contents of the marrow cavity, with no apparent fat, and histologically) were more prevalent in caribou associated with the mortality event than in hunter-killed animals from reference sites. Accumulation of hepatic (liver) hemosiderin in Kupffer cells (macrophages) was noted as an indicator of cachexia. Concentrations of lead in feces and liver, copper in the rumen contents, and arsenic in muscle were higher in caribou harvested near Red Dog Mine, as might be expected in that mineral-rich area, but were not at levels of concern for toxicoses. Kidney concentrations of cadmium, which increased significantly with increasing age, present a potential concern for human consumers, and this is an expected finding. We concluded that caribou had starved and that heavy metals had played no role in the mortality event. Further investigation of regional mineral differences is required to understand the sources and transport mechanisms that explain these findings and to properly address mining activity. Mortality events on the north slope of Alaska are common and likely involve starvation as described here, but in most cases they are not investigated, even though recent industrial activities have heightened concern among some local residents and wildlife managers.

**Key words:** Alaska, caribou, heavy metals, *Rangifer tarandus*, starvation

**RÉSUMÉ.** On a mesuré la concentration en éléments (As, Cd, Cu, Pb, Zn, Fe) et l'état corporel (points limites bruts et histologiques) de caribous (*Rangifer tarandus*) prélevés lors d'un épisode de mortalité qui s'est produit en 1995 en Alaska, dans la région de Point Hope et de Cape Thompson (y compris le site Chariot). On a comparé ces résultats à ceux de caribous tués par des chasseurs à des emplacements témoins (Barrow et Teshekpuk Lake, en Alaska) et à proximité d'une mine (Red Dog Mine) pour trouver si les métaux lourds avaient joué un rôle dans l'épisode de mortalité ou si la concentration d'un ou plusieurs éléments pouvait constituer un risque pour la consommation humaine. La famine et la malnutrition étaient des facteurs majeurs ayant causé la mort ou une extrême faiblesse, vu que la présence minimale ou l'absence de graisse (très basses notes d'état corporel) et une atrophie séreuse de la graisse (observée sous forme de contenu aqueux de la cavité médullaire, sans graisse visible, et à la suite de l'examen histologique) étaient plus courantes chez le caribou associé à l'épisode de mortalité que chez les animaux des emplacements témoins tués par les chasseurs. On a noté dans le foie une accumulation d'hémossidérine hépatique des cellules de Kupffer (cellules macrophages) témoignant d'une cachexie. La concentration de plomb dans les matières fécales et le foie, de cuivre dans le rumen et d'arsenic dans le tissu musculaire était plus élevée chez le caribou provenant de Red Dog Mine, comme on pouvait s'y attendre dans cette zone riche en minéraux, mais cette concentration n'atteignait pas un niveau pouvant provoquer des toxicoses. La concentration de cadmium dans le rein, qui augmentait de façon significative avec l'âge, pourrait constituer un risque pour la consommation humaine, ce qui n'est pas surprenant. On a conclu que les caribous étaient morts de faim et que les métaux lourds n'avaient joué aucun rôle dans l'épisode de mortalité. Il faudrait effectuer des recherches plus poussées sur les différences régionales en minéraux afin de comprendre les mécanismes d'origine et de transport qui expliquent ces résultats et

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d'aborder comme il le faut les activités minières. Les épisodes de mortalité sont courants sur le versant Nord de l'Alaska et sont probablement liés à la famine, comme le décrit cet article, mais dans la plupart des cas ils ne font pas l'objet d'une enquête, même si l'activité industrielle récente est un sujet qui préoccupe de plus en plus certains résidents et gestionnaires locaux de la faune.

Mots clés: Alaska, caribou, métaux lourds, *Rangifer tarandus*, famine

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## INTRODUCTION

The Western Arctic Herd (WAH) of caribou (*Rangifer tarandus*), approximately 450 000 animals (Dau, 1997), ranges over an area that extends from the western region of the north slope of Alaska and the Brooks Range southward to the Seward Peninsula, Alaska (Fig. 1). The Teshekpuk Lake caribou herd (TSHL) is composed of approximately 20 000 individuals, and portions of its range overlap with that of the WAH (Fig. 1).

The WAH population stopped increasing in size during the 1990s and local residents and biologists are concerned about a possible decrease (Dau, 1997). Recent industrial activities (i.e., mining) have added the concern that heavy metal contamination might affect the WAH caribou, or result in a food safety problem (i.e., contaminated meat). Large-scale mortality events (thousands of caribou found dead) have increased concern even more. A systematic, well-designed monitoring of the above issues was not in place, so a retrospective investigation was initiated.

In January 1995, the Alaska Department of Fish and Game received reports of dead and moribund WAH caribou from residents of Point Hope in northwestern Alaska (Fig. 1). Weak and dead caribou were observed and examined near the village of Point Hope. Responding to public concerns required a thorough examination of the caribou and the local environmental conditions temporally and spatially associated with the mortality event. The Red Dog Mine, near Cape Thompson, was a possible source of contamination. Thus, heavy metal toxicoses and food contamination were investigated. We sampled hunter-killed caribou from four reference sites (Barrow, Teshekpuk Lake, Red Dog Mine, and Anaktuvuk Pass) and compared them to caribou found dead at the sites of the mortality event, Point Hope (March 1995) and Chariot, located on Cape Thompson (summer 1995) (Fig. 1). One reference site, Teshekpuk Lake, represents a separate herd known as the Teshekpuk Lake caribou herd.

Heavy metals and minerals are associated with the forage and browse of many ungulates, including caribou, and food is the likely pathway of exposure to heavy metals. Forage may have relatively high mineral levels, depending on the plant species, soil levels and conditions (pH), industrial activities, and other factors. Resultant levels in ungulates may be affected by level of exposure, body condition, age, and forage type. Levels of cadmium in caribou kidney have been considered potential human health problems in some parts of North America and other regions (AMAP, 1998; Aastrup et al., 2000; Elkin, 2001; Odsjö, 2002). Sources of minerals include licks and can be

anthropogenic (drilling mud, road dust) (Edwards and Gregory, 1991). To evaluate the role of heavy metal toxicoses in the mortality of found-dead caribou and in food safety, we measured elements in muscle, kidney, and liver (and on occasion in feces, bone, and rumen contents) of caribou from many regions of northern Alaska, including some of those examined after the 1995 mortality event on and near Cape Thompson, Alaska (O'Hara et al., 1999).

## MATERIALS AND METHODS

Aerial searches for caribou carcasses were conducted on 6–8 June 1995 from a Piper PA-18 airplane flying approximately 500 ft above ground. The area searched extended from Cape Krusenstern to Cape Lisburne within approximately 32 km of the Chukchi Sea coast (10 300–11 600 km<sup>2</sup>). Searches were delayed until June, when snow ablation was 90% complete. In areas with topographic relief, search lines followed drainages and ridgelines to maintain relatively constant elevation above ground and to provide the best view of tight valleys, riverbanks, etc. Transects spaced approximately 1.6 km (1 mi) apart were flown to ensure total coverage of flat areas; however, less than 20% of the total area was searched in this way. Residents of Kotzebue, Kivalina, and Point Hope, Alaska, were observers during these flights. Total mortality was estimated from the proportion of the area searched and our perception of carcass sightability. Approximately 90% of the total area was searched: wind prevented us from covering the northern 10% of the area. However, persistent snowfields present in the area probably reduced the percentage of total area actually viewed to 75–80%.

Our estimate of total mortality from the aerial searches was independently evaluated using radio-marked caribou. At the time of this mortality event, there were 126 functional radio collars in this herd of approximately 450 000 caribou (Dau, 1997). Thus, three radio-marked caribou represented approximately 10 000 caribou (one marked caribou for approximately every 3300 in the WAH, assuming random distribution throughout the herd). One of three radio-marked caribou in the area where this mortality event occurred had died the previous winter.

We were unable to directly evaluate sightability of carcasses; however, we estimate that sightability approached 100%. There are no trees in this area, shrubs are typically lower than 2 m and restricted to narrow riparian bands, vegetation is either absent or decumbent, and searches were conducted before the emergence of leaves.

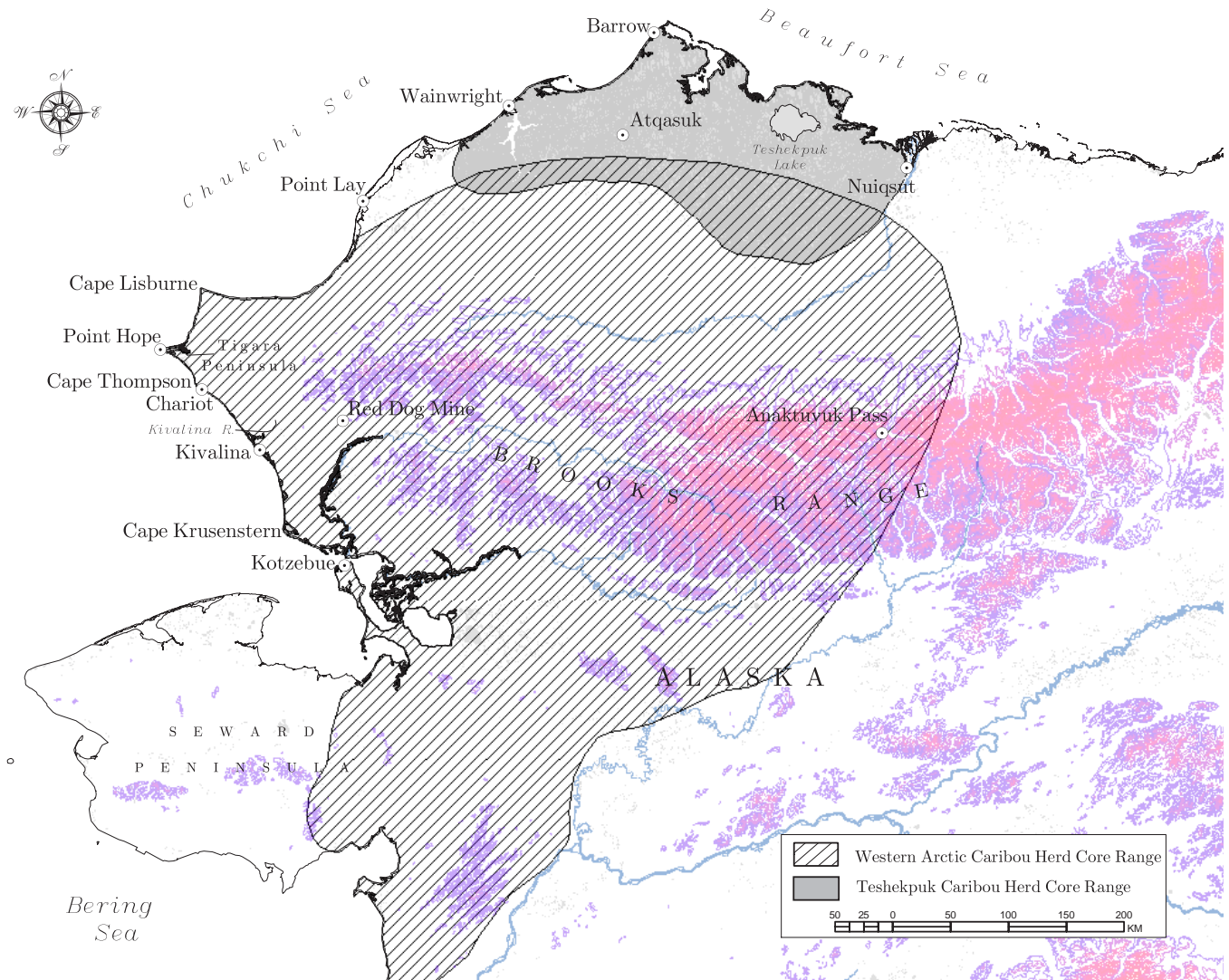


FIG. 1. Map of the study region, showing the general range of the Western Arctic Herd (WAH) of caribou (hatched area) and the sites (bold) where caribou were found dead or harvested and sampled. Caribou at Point Hope, Chariot, and Cape Thompson were found dead, and those at Teshekpuk Lake, Barrow, Anaktuvuk Pass, and Red Dog Mine were killed by hunters.

The five major study sites for examining and sampling caribou are shown in bold type in Figure 1. The 1995 caribou mortality event occurred at Point Hope, Chariot, and the surrounding area. To establish a reference group for comparison, we examined caribou that had been killed by hunters near Red Dog Mine, Barrow, and Teshekpuk Lake and analyzed the carcasses to obtain mean body condition scores. These scores, at the time of year when caribou were examined, indicated that animals were generally healthy (O'Hara et al., 1999).

#### *Caribou Sampling, 1994–96*

Animals found dead or euthanatized by gunshot (at Point Hope and at Chariot and other areas on Cape Thompson) and animals killed by hunters (from Anaktuvuk Pass, Barrow, Point Hope, Teshekpuk Lake, and Red Dog Mine) were examined and sampled for this study. This

sampling introduces a distinction we cannot adequately address (i.e., hunter-killed versus found-dead animals). Caution is warranted in comparing heavy metal levels between these two groups, as tissue degradation over a longer period in the animals found dead may have had a minor effect on metal levels. Table 1 lists the locations, month and year of necropsies, and number of caribou examined and sampled at each location. Some animals were only examined (not sampled) if they showed evidence of being scavenged.

#### *Heavy Metal Analysis*

Frozen samples ( $-20^{\circ}\text{C}$ ) were sent in coolers by overnight express delivery to the Diagnostic Toxicology Laboratory at the College of Veterinary Medicine, Mississippi State University (Starkville), where they were placed in a freezer ( $-80^{\circ}\text{C}$ ) upon arrival. Heavy metal analyses were

TABLE 1. Location, number of caribou examined and sampled, and month and year of necropsies.

Location	Number Sampled (examined <sup>1</sup> )	Period and Year
Anaktuvuk Pass	10 (0)	March–April 1994
Point Hope	6 (6)	March 1995
Barrow	6 (4)	March–April 1995
Cape Thompson	65 (101)	June–July 1995
Teshkpuke Lake	9 (9)	July 1995
Red Dog Mine	15 (15)	March 1996
Total	111 (137)	

<sup>1</sup> Animals “examined” are all animals examined by any of the authors, including scavenged animals (which were not sampled) and animals partially or completely sampled. (The authors were unable to examine intact carcasses of two animals from which hunters had provided samples.)

conducted as in O'Hara et al. (1995). One gram of sample was digested and analyzed by Atomic Absorption (AA) Spectrophotometry (Perkin-Elmer 5000, Norwalk, Connecticut) using a graphite furnace (Perkin-Elmer HGA 500). Values were recorded as parts per million (ppm) wet weight (ww). Quality assurance and quality control were followed as in O'Hara et al. (1995).

### Aging

Caribou were aged by counting the cementum annuli of paired incisors. When these were not available, the first premolar was submitted, as in O'Hara et al. (1999). Teeth were submitted for aging with only species and season of death indicated; the matched pairs were not identified until after the teeth had been aged. A birth date of June 1 was assumed for all animals. Of 45 matched pairs of caribou teeth, 28 (62%) agreed in age, 12 (27%) disagreed by 1 year, four (9%) by 2 years, and one (2%) by 4 years. Age for matched teeth with conflicting age estimates was determined from the tooth with the best quality score or, if the quality scores were the same, from the mean age. Taking into account anticipated cementum layer variability within an animal and human error, these data indicate good age estimates.

### Body Condition Score

Body condition (BC) score was determined as outlined by Kistner et al. (1980). This technique, developed as a field method for evaluating physical condition of deer and previously used for caribou (O'Hara et al., 1999), is based on carcass fat at several sites (the cardiac, omental, and perirenal regions and subcutaneous regions of the tail, head, and brisket) and on the mass of muscle. The six fat depot sites are scored as 0 (no visible fat), 5 (slight quantities of fat), 10 (moderate fat), or 15 (heavy fat), and muscle mass is ranked as 0 (if carcass is “bony”) or 5 (full). Complete scores classify body condition as emaciated (0–10), poor (11–40), fair (41–70), good (71–80), and excellent (81–95). If possible, mean

scores should be calculated separately for sex and age-classes, but for most locations, our samples were too small to allow this. Even obtaining a complete BC score was not always possible, either because the time allowed for examination was limited or because scavenging had destroyed some of the regions to be examined. We also used bone marrow fat evaluation, another technique better suited for assessing very poor condition (Ransom, 1965; Neiland, 1970; Kistner et al., 1980), examining the depot sites where serous atrophy occurs in emaciated animals.

### Histology

Tissue samples as fresh as possible were collected and placed in neutral buffered 10% formalin (VWR Scientific Products, Brisbane, California). Tissues were submitted for trimming and placement in paraffin blocks for thin sectioning. Sections were mounted on glass slides and stained with hematoxylin and eosin (H and E). Binuclear hepatocyte density was estimated by counting these specific cell types in 10 high-power fields for each animal and calculating the mean number of binuclear hepatocytes per high-power field. The special stain for hemosiderin, periodic acid-Schiff (PAS), was used to characterize the pigment indicated by H and E staining.

### Statistics

Comparisons of element concentrations between locations was done with Analysis of Variance (ANOVA) and Fisher's Least Significant Difference (LSD), using the Statistical Analysis System (SAS, Chapel Hill, North Carolina). Correlations, coefficients (slopes), probability of slope different from 0, and  $r^2$  were obtained from simple linear regression using SAS and Microsoft Excel for Windows 95 (7.0). Differences were considered significant when  $p < 0.05$ .

## RESULTS

### *Aerial Survey of Carcasses from 1995 Mortality Event at Point Hope and Chariot*

During aerial searches, we observed 1120 caribou carcasses. Most carcasses occurred between the lower Kivalina River and the Tigara Peninsula. No carcasses were located more than 25 km inland from the Chukchi Sea coast, and the density of carcasses generally declined farther from the coast. Carcasses were distributed from valley bottoms to ridge tops, on all aspects of slope, and in all habitat types.

From the number of carcasses observed, the proportion of the area searched, and our perception of carcass sightability, we estimated that 2000–3000 caribou had died during this mortality event. This number is consistent with an independent mortality estimate based on the fact

TABLE 2. Mean, standard deviation (SD), and range of element concentrations (ppm) in liver of caribou from northern Alaska, by element, for each sampling location and for total sample.

Locations <sup>1</sup> N <sup>2</sup>	AKP 10	Barrow 6	Chariot 18	Point Hope 6	RDM 15	TSHL 9	Total 64
Arsenic (As)							
Mean (SD)	0.21 (0.11)	0.02 (0.02)	0.31 (0.5)	0.10 (0.14)	0.30 (0.13)	0.32 (0.32)	0.25 (0.32)
Range	0.05–0.43	0.005–0.06	0.005–1.36	0.005–0.33	0.06–0.51	0.07–1.11	0.005–1.36
Cadmium (Cd)							
Mean (SD)	0.70 (0.42)	1.27 (0.48)	1.15 (0.80)	1.87 (0.51) <sup>3</sup>	0.43 (0.18)	1.20 (0.54)	1.00 (0.68)
Range	0.14–1.59	0.87–2.10	0.38–4.00	1.4–2.60	0.11–0.70	0.56–2.52	0.11–4.00
Copper (Cu)							
Mean (SD)	29.2 (13.1) <sup>3</sup>	66.0 (37.7)	68.8 (47.0)	85.5 (22.5)	100.4 (76.5)	6.35 (4.43) <sup>3</sup>	62.5 (55.8)
Range	11.8–51.9	31.0–140.0	4.30–170.0	54.0–110.0	6.15–225.7	3.55–17.70	3.55–225.7
Lead (Pb)							
Mean (SD)	0.19 (0.06)	1.08 (0.35)	1.42 (0.60) <sup>3</sup>	0.73 (0.21)	1.65 (0.60) <sup>3</sup>	0.77 (0.29)	1.09 (0.68)
Range	0.13–0.29	0.70–1.60	0.43–2.70	0.41–1.00	0.82–2.88	0.11–1.11	0.11–2.88
Zinc (Zn)							
Mean (SD)	48.4 (15.8)	48.2 (30.7)	58.7 (32.1) <sup>3</sup>	77.3 (37.5) <sup>3</sup>	22.4 (4.6)	34.4 (6.2)	45.9 (28.3)
Range	28.3–75.2	27.0–110.0	4.0–160.0	38.0–140.0	17.2–33.5	23.2–41.7	4.0–160.0
Iron (Fe)							
Mean (SD)	363.6 (141.1)	1201.7 <sup>3</sup> (1336.4)	1502.7 <sup>3</sup> (777.4)	1537.3 <sup>3</sup> (844.3)	363.3 (188.6)	126.2 (59.2)	839.1 (848.4)
Range	166.8–551.3	350.0–3900.0	260.0–3120.0	44.0–2640	51.1–659.5	47.2–218.0	44.0–3900.0

<sup>1</sup> Locations: Anaktuvuk Pass (AKP), Chariot (represents animals from Cape Thompson), Red Dog Mine (RDM), and Teshekpuk Lake (TSHL).

<sup>2</sup> N = Number of individuals analyzed.

<sup>3</sup> Analysis of Variance (ANOVA) indicated significant difference ( $p < 0.05$ ) for that element compared to other locations.

that one of three radio-collared WAH caribou wintering in this area died during the 1994–95 winter. Although a crude approximation, this also suggests that roughly 3000 caribou perished during this event.

#### *Examination of Carcasses Found Dead at Point Hope and Cape Thompson*

In March 1995, six caribou were examined near Point Hope. One was found dead, and five were found in moribund condition and euthanized. All were in emaciated to very poor body condition (BC score 0 to 15, mean of 5). Five animals showed no evidence of body fat, and marrow cavity examination indicated serous atrophy (watery contents instead of fat). Tissues from these animals were examined histologically. Three were females and one was pregnant.

In June–July 1995, 101 caribou were examined (22 at Chariot in early June and 79 in the area of Cape Thompson in early July), and 65 of these were partially or completely sampled (Table 1). The 22 caribou at Chariot were found dead in a curled posture or “sleeping position.” All were examined and found to be in various states of post-mortem condition. We sampled only carcasses found farther inland that remained intact. (Additional scavenged caribou found near the beach were unsuitable for evaluation, and no data about them were recorded.) All 22 carcasses showed evidence of malnourishment and starvation: rocks in the abomasum, lack of visceral and body fat, and serous atrophy of bone marrow. Tissues from intact animals were collected and submitted for heavy metal analyses (Tables 2, 3, 4, 5, and 6). Tissues were not suitable for histologic examination. The 79 animals found in early July were also

in varying states of post-mortem condition. Four had apparently been killed by hunters, as evidenced by removal of the head.

These 101 carcasses were 58 females, 35 males, and 8 animals of unknown gender, ranging in age from 0 to 12 years (mean age: 5.6 years). They included 66 adults, 22 subadults, and 11 calves (age not recorded for 2). Of 12 females examined for a fetus, three (25%) were pregnant. Only 95 carcasses were considered suitable for evaluation. Of these, all but the four killed by hunters (91 animals, or 96%) were classified as emaciated because they had no body fat and empty marrow cavities. Small rocks (5–8 mm diameter) were found in the abomasa of 26 of the 31 caribou whose stomachs were examined. The presence on the skulls of many mature males of fully calcified, velvet-free antlers with a partially developed abscission layer suggests that the caribou began dying in mid to late October.

#### *Examination of Caribou Killed by Hunters*

Caribou taken in March 1994 by hunters near Anaktuvuk Pass were not thoroughly examined, but their tissues were analyzed for heavy metals. During March and April 1995, six hunter-killed caribou in Barrow were sampled for contaminants. Four of these were thoroughly examined (BC scores: 5 to 45), three were determined to be in fair body condition, and one animal was reported to be “lame” (BC score: 5) and was killed for humane reasons. All were adults: three males, one female, and two of unknown sex (intact carcass not available). The female was not pregnant. During July 1995, nine apparently healthy caribou

TABLE 3. Mean, standard deviation (SD), and range of element concentrations (ppm) in kidney of caribou from northern Alaska, by element, for each sampling location and for total sample.

Locations <sup>1</sup> N <sup>2</sup>	AKP 10	Barrow 6	Chariot 19	Point Hope 6	RDM 14	TSHL 9	Total 64
Arsenic (As)							
Mean (SD)	0.39 (0.07)	0.03 (0.03)	0.78 (1.13)	0.08 (0.11)	0.34 (0.2)	0.27 (0.19)	0.42 (0.67)
Range	0.27–0.51	0.005–0.06	0.01–4.2	0.01–0.24	0.001–0.74	0.08–0.69	0.001–4.2
Cadmium (Cd)							
Mean (SD)	3.32 (1.49)	7.92 (12.38) <sup>3</sup>	2.13 (1.01)	5.25 (1.85)	2.3 (1.05)	3.15 (1.46)	3.33 (4.07)
Range	2.0–6.13	1.5–33.0	0.64–4.5	2.3–7.8	0.54–4.32	0.6–5.85	0.54–33.0
Copper (Cu)							
Mean (SD)	4.65 (0.33)	5.95 (1.45)	20.3 (36.6)	6.8 (1.94)	22.0 (17.1)	3.58 (1.18)	13.3 (22.6)
Range	4.06–5.19	3.5–7.7	2.18–140.0	4.4–9.7	2.64–41.3	1.71–5.71	1.71–140.0
Lead (Pb)							
Mean (SD)	0.06 (0.03)	0.29 (0.19)	0.77 (0.49)	0.43 (0.28)	10.0 (33.4)	0.76 (0.46)	2.61 (15.7)
Range	0.03–0.13	0.05–0.5	0.04–2.3	0.15–0.86	0.25–126.1	0.19–1.79	0.03–126.1
Zinc (Zn)							
Mean (SD)	35.5 (11.6)	26.8 (3.92)	44.2 (35.7)	35.0 (7.56)	16.8 (2.27)	26.6 (5.72)	31.9 (22.4)
Range	20.8–56.0	20.0–32.0	21.5–180.0	25.0–48.0	13.7–21.0	20.6–40.2	13.7–180.0
Iron (Fe)							
Mean (SD)	38.7 (7.11)	95.0 (57.5)	441.0 <sup>3</sup> (522.7)	158.3 (34.3)	55.5 (26.7)	40.1 (13.9)	178.5 (330.6)
Range	27.6–52.0	10.0–180.0	70.0–2300.0	110.0–200.0	30.4–112.9	26.8–67.6	10.0–2300.0

<sup>1</sup> Locations: Anaktuvuk Pass (AKP), Chariot (represents animals from Cape Thompson region), Red Dog Mine (RDM), and Teshekpuk Lake (TSHL).

<sup>2</sup> N = Number of individuals analyzed.

<sup>3</sup> Analysis of Variance (ANOVA) indicated significant difference ( $p < 0.05$ ) for that element compared to other locations.

(one nonpregnant female with no calf and eight males), ranging in age from 4 to 12 years (mean age: 7.9 years), were shot near Teshekpuk Lake and sampled (Table 1). This group was determined to be in fair body condition (mean BC score: 50). During March 1996, local residents harvested 15 apparently healthy adult caribou (eight males and seven females; mean age: 5.4 years) just southwest of the Red Dog Mine. This group was determined to be in fair body condition (mean BC score: 42). Four of the seven females (57%) were pregnant.

### Metals and Minerals

Tables 2 (liver), 3 (kidney), 4 (muscle), and 5 (rumen contents) list the arithmetic mean, standard deviation (SD), and range for the metal concentrations detected in caribou from the various locations and for those in all caribou analyzed. Table 6 presents the geometric means for the metal levels detected in caribou from the various locations. The comparisons between locations were done with ANOVA and Fisher's LSD.

**Age and Metals:** Linear regression analyses indicated no significant relationship between metal concentration and age except for concentration of cadmium in kidney (coefficient [slope] = 0.211 ppm/year,  $p < 0.001$ ,  $r^2 = 0.35$ , correlation coefficient = 0.59).

**Metals by Location:** ANOVA comparison of locations and mean concentrations within each matrix of metals revealed some significant differences, and these are indicated in Tables 2, 3, 4, and 5. Figures in parentheses below refer to mean concentrations (in ppm), and all differences reported here are significant ( $p < 0.05$ ). For cadmium,

mean cadmium levels were higher in kidney (7.92 ppm) and muscle (0.48 ppm) from Barrow; higher in liver (1.87 ppm) and rumen contents (0.98 ppm) from Point Hope; and lowest in muscle (0.006 ppm) from Red Dog Mine. For copper, significant differences were also noted for some locations. In liver, the lowest copper levels were detected in Anaktuvuk Pass (29.2 ppm) and Teshekpuk Lake (6.35 ppm) caribou, the easternmost animals studied. In muscle, copper levels were highest in Teshekpuk Lake animals (13.8 ppm). Copper in rumen contents was highest in Red Dog Mine caribou (11.0 ppm). Lead concentrations differed significantly by location; in liver, the highest lead levels were in caribou from Red Dog Mine (1.65 ppm) and Cape Thompson (1.42 ppm). In muscle, lead levels were higher in animals from Cape Thompson (0.78 ppm) and Teshekpuk Lake (0.72 ppm). Note that evaluation of lead concentrations in hunter-killed animals must be done with caution, as these animals may contain lead fragments from bullets. Arsenic levels were slightly higher in muscle of Red Dog Mine caribou (0.55 ppm). Zinc levels were highest in kidney from Cape Thompson (44.2 ppm) and in liver from Cape Thompson (58.7 ppm) and Point Hope (77.3 ppm) animals. Iron levels were highest in kidney from Cape Thompson caribou (441.0 ppm). In liver, iron levels were higher in Cape Thompson, Barrow, and Point Hope animals than in caribou from Anaktuvuk Pass, Red Dog Mine, and Teshekpuk Lake.

### Histopathology

The hunter-killed caribou from Point Hope had marked serous atrophy of fat consistent with their gross lesions and

TABLE 4. Mean, standard deviation (SD), and range of element concentrations (ppm) in muscle of caribou from northern Alaska, by element, for each sampling location and for total sample.

Locations <sup>1</sup> N <sup>2</sup>	AKP 10	Barrow 6	Chariot 31	Point Hope 6	RDM 13	TSHL 9	Total 75
Arsenic (As)							
Mean (SD)	0.42 (0.1)	0.06 (0.08)	0.09 (0.09)	0.19 (0.18)	0.55 (0.120) <sup>3</sup>	0.12 (0.07)	0.22 (0.21)
Range	0.29–0.56	0.005–0.20	0.03–0.47	0.005–0.36	0.29–0.71	0.08–0.27	0.01–0.70
Cadmium (Cd)							
Mean (SD)	0.03 (0.02)	0.48 (0.17) <sup>3</sup>	0.14 (0.10)	0.38 (0.03)	0.006 (0.007) <sup>3</sup>	0.30 (0.1)	0.16 (0.17)
Range	0.01–0.06	0.29–0.71	0.02–0.41	0.35–0.41	0.001–0.029	0.17–0.47	0.001–0.71
Copper (Cu)							
Mean (SD)	3.96 (0.43)	3.90 (1.16)	2.24 (0.76)	3.50 (1.37)	2.71 (0.58)	13.8 (10.5) <sup>3</sup>	4.17 (5.06)
Range	3.47–4.69	2.40–5.50	1.33–4.46	2.20–6.10	2.11–3.87	3.36–31.8	1.33–31.8
Lead (Pb)							
Mean (SD)	0.03 (0.03)	0.35 (0.21)	0.78 (0.54) <sup>3</sup>	0.23 (0.20)	0.05 (0.03)	0.72 (0.23) <sup>3</sup>	0.47 (0.49)
Range	0.002–0.09	0.03–0.64	0.02–2.91	0.02–0.56	0.005–0.08	0.46–1.10	0.002–2.91
Zinc (Zn)							
Mean (SD)	29.6 (8.8)	40.3 (27.4)	32.1 (9.9)	50.7 (54.2)	33.0 (9.11)	29.2 (13.7)	33.7 (19.1)
Range	17.8–44.3	21.0–90.0	15.7–57.3	22.0–160.0	19.9–44.4	18.7–64.6	15.7–160.0
Iron (Fe)							
Mean (SD)	48.3 (6.59)	72.5 (25.6)	141.7 (332.4)	124.5 (36.8)	44.5 (5.81)	54.2 (10.7)	95.0 (216.6)
Range	31.3–54.0	40.0–110.0	29.1–1910.0	87.0–180.0	31.4–56.2	30.0–65.4	29.1–1910.0

<sup>1</sup> Locations: Anaktuvuk Pass (AKP), Chariot (represents animals from Cape Thompson region), Red Dog Mine (RDM), and Teshekpuk Lake (TSHL).

<sup>2</sup> N = Number of individuals analyzed.

<sup>3</sup> Analysis of Variance (ANOVA) indicated significant difference ( $p < 0.05$ ) for that element compared to other locations.

extremely poor body condition. This pattern was not evident in hunter-killed caribou from the reference sites. Of the four Point Hope animals that provided liver samples suitable for microscopic examination, three had a larger amount of pigment in the Kupffer cells than the other caribou examined (from Barrow, Teshekpuk Lake, and Red Dog Mine). Special staining (i.e., PAS) indicates that the pigment is primarily hemosiderin.

The binuclear hepatocytes observed were not correlated with age ( $p > 0.05$ ), and the mean number of binuclear hepatocytes was 6.0 per high-power field. Mild interstitial lymphoplasmacytic nephritis was present in many animals (9 of the 23 with suitable [not autolyzed or traumatized] kidney tissue sections). No histologic lesions in Point Hope animals indicated a toxic or infectious etiology, and the gross and histologic findings (as compared with findings for reference animals) strongly supported malnutrition and starvation.

## DISCUSSION

Some Western Arctic Herd caribou winter, although not every year, within 25 km of the Chukchi Sea, between Cape Lisburne and Cape Krusenstern. This area is chronically windy and characterized by low snow cover and decumbent, alpine vegetation with low plant biomass. The topography near Cape Thompson, roughly the geographic center of the highest mortality zone, makes it especially vulnerable to high wind.

The investigation of weak and found-dead caribou near Point Hope and found-dead caribou near Chariot in 1995

included aerial surveys, necropsy of many carcasses (both found-dead and euthanatized animals), gross and histologic examination of fresh tissues, chemical analyses (elements), aging, and similar examination and analyses of caribou from reference or control groups (Barrow, Teshekpuk Lake) and animals from the Red Dog Mine region. Much of the evidence indicates that the caribou examined near Point Hope (March 1995) and Cape Thompson (June–July 1995) were suffering from malnutrition or had succumbed to starvation. In the process of this evaluation, we made many observations concerning element levels, changes due to starvation, and differences based on location of sampling and age. We describe these observations and discuss starvation and its diagnoses and the absence of any evidence (element levels in tissue or presence of lesions) to indicate heavy metal toxicoses or unusual contamination (i.e., food safety concerns).

### Body Condition (BC)

Subcutaneous fat is present in most deer only when body condition is good to excellent (Kistner et al., 1980). The general sequence of fat deposition in deer is first in the coronary groove (heart base), then in the pericardium, omentum, and perirenal area, and finally in subcutaneous areas (Harris, 1945; Kistner et al., 1980) including those of the rump and brisket. Fat mobilization typically occurs in the reverse order, and during this process, the fat turns yellow or red. In emaciated animals, serous atrophy occurs at these depot sites and in the bone marrow cavity. Animals in suitable post-mortem condition were histologically evaluated in our study to assess nutritional status.

TABLE 5. Mean, standard deviation (SD), and range of element concentrations (ppm) in rumen contents of caribou from northern Alaska, by element, for each sampling location and for total sample.

Locations N <sup>2</sup>	Barrow 4	Chariot <sup>1</sup> 11	Point Hope 5	Red Dog Mine 10	Total 30
Arsenic (As)					
Mean (SD)	0.18 (0.22)	1.06 (0.41)	0.70 (0.50)	1.37 (2.09)	0.92 (0.92)
Range	0.005–0.42	0.49–1.90	0.05–1.30	0.24–4.50	0.005–4.50
Cadmium (Cd)					
Mean (SD)	0.40 (0.24)	0.47 (0.03)	0.98 (0.96) <sup>3</sup>	0.06 (0.03)	0.41 (0.48)
Range	0.06–0.62	0.42–0.52	0.50–2.70	0.03–0.14	0.03–2.70
Copper (Cu)					
Mean (SD)	3.37 (4.41)	2.1 (0.18)	1.6 (0.38)	11.0 (0.73) <sup>3</sup>	5.15 (4.49)
Range	1.10–9.99	1.80–2.40	1.20–2.10	9.53–11.7	1.10–11.7
Lead (Pb)					
Mean (SD)	0.6 (0.78)	2.93 (1.33)	1.34 (0.93)	2.85 (1.47)	2.39 (1.49)
Range	0.12–1.50	0.02–5.40	0.20–2.50	1.03–4.69	0.02–5.40
Zinc (Zn)					
Mean	20.0 (5.38)	31.8 (3.74)	22.2 (7.46)	26.2 (5.72)	26.8 (6.74)
Range	13.9–25.0	26.0–38.0	16.0–35.0	18.0–38.4	13.9–38.4
Iron (Fe)					
Mean	132.6 (34.8)	891.8 (469.4)	786.6 (456.9)	165.8 (112.4)	531.1 (487.2)
Range	90.0–175.0	210.0–1600.0	330.0–1420.0	21.6–370.3	21.6–1600.0

<sup>1</sup> Chariot (represents animals from Cape Thompson region).

<sup>2</sup> N = Number of individuals analyzed.

<sup>3</sup> Analysis of Variance (ANOVA) indicated significant difference ( $p < 0.05$ ) for that element compared to other locations.

The severe weather (cold, wind, precipitation) common in this area of the Arctic creates high metabolic demands (Kistner et al., 1980) that, combined with malnutrition, may cause death before total exhaustion of fat reserves (Cheatum, 1949; deCalesta et al., 1975). Beginning in late October 1994, a series of severe winter storms with high winds, freezing rain, and snow moved through northwestern Alaska at the rate of one per week until early January. Individual storms lasted 1–4 days, and windchill temperatures occasionally approached  $-70^{\circ}\text{C}$  ( $-100^{\circ}\text{F}$ ).

For nonhibernating Arctic wildlife species, winter and early spring are the most challenging periods for survival, mostly because of a limited food supply and harsh conditions. Food intake may be insufficient, and deer may enter a negative energy balance. Endogenous fat becomes the main source of energy (Wolkers et al., 1994), resulting in annual body-mass and fat cycles that correspond closely to seasons (Worden and Pekins, 1995). Long-term dietary restrictions are characterized by a decreasing body mass (reduction of muscles) and low levels of bone marrow fat, indicators of extreme undernourishment (Suttie, 1983; Torbit et al., 1988). These changes in condition of *Rangifer* are reflected in blood (Soveri et al., 1992), ciliated fauna of the rumen, and liver histology (Soveri, 1993).

We speculate that caribou emigrating from their summer range during October 1994 were in poor condition, lacking adequate fat reserves to survive the storms, and were probably prevented from accessing the limited vegetation in this area by ground-fast ice and wind-hardened snow. From aerial surveys and radio-collar data, we estimate that approximately 30% (roughly 3000 of 10 000) of the caribou that wintered in this area died. Many were

found in a “sleeping position,” which has been described in cases of starvation in deer (Rausch, 1950).

Much of the evidence suggests that starvation and malnourishment, combined with severe weather, caused the 1994–95 caribou mortality in the Cape Thompson area. This evidence is convincing, but it is not definitive. Gross (whole-body) examination of carcasses indicated essentially no fat was present in key depot sites, and this is reflected in the very low BC scores. Detection during histologic exam of serous atrophy of fat in the marrow cavity and of tissues supports the conclusion of severe emaciation for animals examined in Point Hope. Other histologic evidence indicating a lack of caloric intake was the presence of larger than normal accumulations of hemosiderin in Kupffer cells of the liver, which is known as a marker of cachexia or emaciation (Rausch, 1950).

#### *Element Concentrations*

Elemental analysis (As, Cd, Cr, Cu, Pb, Zn, Fe) was conducted on several caribou matrices. We compared element concentrations by location, since regional differences in natural sources of elements are known to exist, and industrial activity (i.e., Red Dog Mine) could potentially mobilize elements to enter the food chain and affect wildlife and human health. Possible bias should be considered for these comparisons because we sampled found-dead and hunter-killed caribou. Compromised caribou, or specific age cohorts, are more likely to be found dead, and certain caribou may be preferred by hunters, or simply more accessible to them. The Red Dog Mine area caribou had slightly higher arsenic concentrations in muscle as compared to other caribou, but compared to livestock the



TABLE 6. Geometric means for element concentrations (ppm, ww) in liver, kidney, muscle, rumen contents, bone and feces for caribou from northern Alaska, by element and location of sampling.

Location/tissue	Arsenic	Cadmium	Copper	Lead	Zinc	Iron
Barrow						
Liver	0.02	1.20	59.3	1.04	42.8	852.6
Kidney	0.02	3.94	5.78	0.22	26.6	71.3
Muscle	0.02	0.45	3.75	0.26	34.5	68.7
Rumen	0.06	0.30	1.99	0.31	19.4	129.1
Point Hope						
Liver	0.03	1.81	82.8	0.70	70.4	967.0
Kidney	0.02	4.92	6.54	0.36	34.3	155.1
Muscle	0.07	0.38	3.32	0.15	37.2	120.3
Rumen	0.46	0.76	1.57	0.97	21.4	683.7
Bone	0.08	0.08	1.65	1.37	44.0	53.9
Chariot						
Liver	0.04	1.00	47.62	1.30	48.8	1236.5
Kidney	0.10	1.90	8.28	0.60	37.7	279.4
Muscle	0.08	0.10	2.14	0.58	30.6	80.3
Rumen	0.99	0.47	2.09	1.96	31.6	754.4
Teshekpuk Lake						
Liver	0.23	1.12	5.53	0.67	33.8	113.9
Kidney	0.22	2.74	3.40	0.65	26.1	38.2
Muscle	0.11	0.28	10.73	0.69	27.3	53.0
Red Dog Mine						
Liver	0.27	0.39	63.3	1.54	22.0	307.0
Kidney	0.21	2.02	13.0	1.20	16.6	50.7
Muscle	0.53	0.01	2.66	0.04	31.7	44.2
Feces	0.14	0.06	11.06	6.18	39.1	299.0
Rumen	0.61	0.06	11.0	2.48	25.7	127.7
Anaktuvuk Pass						
Liver	0.19	0.59	26.2	0.18	46.1	336.5
Kidney	0.38	3.07	4.64	0.06	33.9	38.1
Muscle	0.41	0.02	3.94	0.02	28.4	47.8

level is not considered elevated (Puls, 1988). Mean cadmium concentration was significantly higher in kidney and muscle for caribou from Barrow, and higher in liver and rumen contents for caribou from Point Hope. Cadmium concentrations are known to increase with age in kidney (as shown in this study) and occasionally in liver (Gamberg and Scheuhammer, 1994). The concentrations reported here are considered high for most livestock (except horses) but are within the range for caribou from other regions (AMAP, 1998). The concentration of cadmium in kidney of some animals exceeds a recommended critical tissue concentration of 30 mg/kg ww (Outridge et al., 1994), but neither gross nor histologic assessment found evidence of lesions. The renal cadmium concentrations reported here (mean: 3.33 ppm ww; range: 0.54–33.0 ppm) are not the highest known for caribou in North America, but the Canadian Northern Contaminants Program has advised that consumption of caribou kidney be limited to one meal (a single kidney) per week, or approximately 50 meals per year (AMAP, 1998; Aastrup et al., 2000; Elkin, 2001; Odsjö, 2002). We cannot provide sound consumption advice because our data on rates at which humans consume caribou organs are very limited, but we detected no metal at unexpected concentrations in caribou tissues.

Low levels of copper in liver were detected in both Anaktuvuk Pass and Teshekpuk Lake animals. The latter

had levels below 10 ppm (ww), the suggested deficiency level of copper in liver for cattle, sheep (Puls, 1988), and moose (Flynn et al., 1977; Frank et al., 1994), but above the 3.5 ppm (ww) recommended for horses (Puls, 1988). The concentration of copper in rumen contents was highest for Red Dog Mine caribou. Adequate dietary levels for cattle and sheep are considered to be 10 ppm (dry weight), and toxicosis requires copper levels much higher than those reported here.

Lead concentrations in liver of less than 1.0 ppm (ww) are considered normal for cattle and sheep, and levels are not considered toxic until greater than 5.0 to 10.0 ppm (ww) (Puls, 1988). Muscle lead levels of 0.1 to 0.3 ppm (ww) are considered normal for cattle. Mean liver lead concentrations were highest (> 1.0 ppm, ww) for caribou from Red Dog Mine and Cape Thompson, and mean muscle lead levels were highest (> 0.3 ppm, ww) for Cape Thompson and Teshekpuk Lake. However, these concentrations were well below those known to result in toxicoses. Mean fecal lead level for Red Dog Mine caribou was within the normal range for cattle (2.0 to 35.0 ppm, ww), except for one animal with 38.0 ppm (ww) in the feces. Toxic levels of lead in feces of cattle usually exceed 200 ppm (ww) (Puls, 1988). Mean levels of lead in rumen content were 2.93 ppm lead (ww) for Cape Thompson and 2.85 ppm (ww) for Red Dog Mine. A normal cattle diet contains less than 1.0 ppm, and toxic levels are greater than 100 ppm (Puls, 1988). Thus, although caribou near Red Dog Mine and Cape Thompson are exposed to higher lead levels than caribou in the other regions studied, exposure is not at levels of concern for cattle. However, we must stress that only a small sample represents the area adjacent to the mine. In hunter-killed caribou, fragmentation from lead bullets can contaminate some tissues, but this is unlikely for rumen contents or feces if the projectile did not penetrate the stomach or intestine. The source of lead could be natural (the region is rich in minerals) or a result of human activities (mining, road dust, atmospheric deposition). No reliable conclusion identifying the lead source(s) can be drawn from this study design. Future evaluation of lead exposure would require a larger sample and a more focused experimental design that includes analysis of forage and resident (nonmigratory) wildlife.

Levels of zinc (ww) were higher in kidney of Cape Thompson caribou (44.2 ppm), and in liver of Cape Thompson (58.7 ppm) and Point Hope (77.3 ppm) caribou compared to other caribou studied. In cattle, adequate levels of zinc are 25–100 ppm for liver and 18–20 ppm (ww) for kidney, and zinc levels are not considered “elevated” (not necessarily toxic) until they reach 300–500 ppm in liver and 50–140 ppm in kidney. The same general ranges apply to goats, pigs, and other species (Puls, 1988). The concentrations of zinc in these caribou appear to be of no concern. Mean kidney iron level was highest in Cape Thompson caribou (441 ppm) as compared to other caribou, and this level is considered high. However, the post-mortem congestion and hemosiderin accumulation

evidenced in the histologic examination of Point Hope starving animals would lead one to expect increased iron levels. Mean liver iron levels were considered high (> 1000 ppm) for Cape Thompson, Barrow, and Point Hope, but the range (variability) was great (Puls, 1988). Blood congestion (pooling) and starvation can increase iron levels (Rausch, 1950). High concentrations of aluminum, manganese, nickel, and cadmium in liver and kidney of caribou from other regions have previously been reported and may be associated with regional sources (mostly natural) (Elkin and Bethke, 1995) or other sources such as licks (Heard and Williams, 1990). The element levels in the caribou studied indicate no concern for toxicoses. Only renal cadmium levels represent a potential concern for human consumers, and this is an expected finding.

### *Histologic Findings*

The significant serous atrophy of fat (indicating starvation) observed in caribou from the Point Hope area confirmed gross visual findings that essentially no fat remained. This was not observed in caribou examined from reference sites. Another histologic indicator of nutritional stress was the higher degree of hemosiderosis (pigment in Kupffer cells). Hemosiderin occurs commonly in all species in the liver and spleen, and its observed abundance suggests excess hemolytic activity relative to the rate of reutilization of iron. Hemosiderosis is seen in hemolytic anemia, in copper-deficiency anemia, in cachexia (likely here), and can be seen in periacinal zones in severe chronic passive congestion. In this case, it was most likely due to cachexia or emaciation (Rausch, 1950).

In conclusion, gross and histologic observations indicate that the majority of caribou found dead in NW Alaska (Point Hope and Cape Thompson) in 1995 died from starvation/malnutrition. Exposure to certain elements and their residue levels in animal tissue show regional differences. Increased levels of copper, lead, and arsenic occur near Red Dog Mine and in surrounding areas, but there is no evidence of metal toxicoses in caribou of the area. Renal cadmium levels need further scrutiny from the perspective of human consumption.

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