





EVALUATING THE EFFECTS OF NUTRITION, DISEASE, AND PREDATION ON CALF RECRUITMENT IN THE NORTHERN ALASKA PENINSULA CARIBOU HERD



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Executive Summary

The Northern Alaska Peninsula Caribou Herd is an important subsistence and recreational hunting resource in the southwest Alaska region. This herd has declined by approximately 88% since 1993, and calf recruitment will play a central role in the dynamics of this herd in the coming years. This project was initiated in the spring of 2005 to begin collecting data on the influence of nutrition, disease, and predation on calf production and survival. A parturition survey of 314 cows revealed an overall herd pregnancy rate of 57%, approximately 20% lower than the herd pregnancy rate in the mid-late 1990s. This may be an indicator of poor nutritional condition in this herd. The survival rate of 42 radio-marked calves was 43% during their first two weeks of life, which is comparable to rates reported for several stable or increasing herds of similar size in interior Alaska. However, the survival rate of radio-marked calves after two weeks of age was approximately 40-60% lower than rates reported for those same interior Alaska herds. The majority of calf mortality site investigations revealed evidence of scavenging or predation, and we hypothesize that calves may not be developing at a rate sufficient to decrease their vulnerability to predation after reaching the two-week-old milestone. Heavy parasite burdens and infectious disease exposure were detected in each of the eight calves and three adults that were examined during a health assessment of this herd. Clinical pathology indicated that parasites were adversely affecting the health of surveyed animals. Our preliminary appraisal of data collected in 2005 is that parasites and infectious diseases may be adversely impacting both calf production and survival in this herd. Data collection will continue and the project will be expanded in coming years to address this hypothesis.

Introduction

The Northern Alaska Peninsula Caribou (*Rangifer tarandus granti*) Herd (NAPCH) is historically the most important terrestrial subsistence resource on the Alaska Peninsula (Morris 1985, Morris 1987, Fall and Morris 1988), and was an important herd for recreational and trophy hunting from the 1950s until the 1990s (Sellers et al. 2003). The annual range of the migratory NAPCH spans the boundaries of Alaska Peninsula and Becharof National Wildlife Refuges, Aniakchak National Monument and Preserve, Katmai National Park, State of Alaska, and Native Corporation lands. This herd is in intensive management status with the Alaska Department of Fish and Game (ADFG). This herd is also listed in the enabling legislation for Alaska Peninsula and Becharof National Wildlife Refuges, as well as Aniakchak National Monument and Preserve, making it a trust wildlife species for the US Fish and Wildlife Service (FWS) and National Park Service (ANILCA Sections 201[1] and 302[1B, 2B]).

Post-parturition population counts of the NAPCH have declined by 88% since 1993, from approximately 16,500 to 2,000 animals (Figure 1). Herd managers have detected a delayed age of first reproduction in females and a decrease in calf recruitment in recent years, suggesting that poor nutritional condition may be a primary factor in the decline (Eberhardt 2002). Past studies have also pointed to nutrition as a factor in the decline, but have noted significant calf mortality from predation and disease (Sellers et al. 2003). Recently, subsistence users have focused on predation as a cause of the decline, and proposals were submitted to the State of Alaska Board of Game for more liberal wolf and bear seasons in 2005. These proposals resulted in the extension of the wolf hunting season by one month in 2005-06.

Results of sensitivity analyses of a population model including best estimates of female survival rates, female reproductive rates, and calf recruitment rates (Caswell 2001) indicate that calf recruitment will play a central role in the population dynamics of the NAPCH over the next

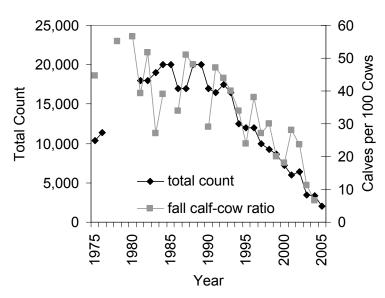


Figure 1. Population trend of the NAPCH as indexed by summer post-parturition counts and calf-cow ratios from fall composition counts.

few years. Estimated calf recruitment through summer has not exceeded 8% of the total population size since 2002 (Figure 1). The roles that nutritional condition, disease, and predation have played in this low recruitment rate are unclear.

Nutritional condition in ungulates can affect the entire calf recruitment process (ex. Cook et al. 2004). Nutritional condition affects the onset of maturity in young adult caribou and the probability of pregnancy in mature caribou (Adams and Dale 1998a), both of which influence the rate of calf production for a population. Poor female nutritional condition at the time of

breeding can affect the length of gestation and delay parturition in the following spring (Adams and Dale 1998b). Calves that are born later in the spring have lower growth and survival rates (Clutton-Brock et al. 1987, Guinness et al. 1978). Nutritional condition of females during gestation affects calf birth mass (Adams 2005) and the rate of development in calves during the summer months (Adams 2003), both of which in turn affect calf survival rates (Adams 2003, Adams et al. 1995a, Guinness et al. 1978). Evidence of poor nutritional condition was detected early in the decline of the NAPCH (Sellers et al. 2003), and nutritional condition has been suspected as the primary cause of the decline by managers since that time.

Disease can be defined as a disturbance that inhibits biological systems from maintaining homeostasis or health (Roffe and Work 2005). Disease can be caused by non-infectious agents such as toxins, genetics, traumatic injury, and neoplasia (cancer), or infectious agents such as bacteria, viruses, and other types of parasites. Empirical evidence suggests that parasites can regulate wild ungulate populations (Albon et al. 2002). Parasite infections can induce decreases in fecundity and body condition in adult females (Albon et al. 2002, Stein et al. 2002), as well as decreases in survival rates in young-of-the-year (Hudson et al. 1992) and older animals (Tompkins et al. 2001, Gulland et al. 1993). Infectious diseases can also dampen rates of increase in wild ungulate herds through effects on adult survival and fecundity (Jolles et al. 2005). Evidence of juvenile mortality from lungworms and/or pneumonia has been detected in the NAPCH (Sellers et al. 2003, Sellers et al. 1998), but the extent to which recruitment has been affected is unknown. Evidence of other disease presence in the NAPCH is limited (Zarnke 2000).

Wolf and bear predation on juveniles can affect calf recruitment in ungulates (Hayes et al. 2003, Gasaway et al. 1992). Predation has been identified as a major source of juvenile mortality in several Alaskan caribou populations (Valkenburg et al. 2002, Boertje and Gardner 2000, Adams et al. 1995a,b, Whitten et al. 1992). Specifically, predation on neonates very early in life limits recruitment, and predation rates tend to decline as caribou calves age and develop in herds with adequate nutritional reserves (Boertje and Gardner 2000, Valkenburg 1997, Adams et al. 1995a,b). To complicate the effects of predation, poor nutritional condition (Adams 2003, Adams et al. 1995a) and high parasite burdens (Moller 2005, Tompkins et al. 2001, Murray et al. 1997, Hudson et al. 1992) may increase the susceptibility of animals to predation. Significant juvenile mortality from bear and wolf predation has been identified in the NAPCH, though early calf mortality may be lower than in other areas of Alaska (Sellers et al. 2003). However, bear densities on the Alaska Peninsula are higher than most other areas around the world (Miller et al. 1997), so the potential for chronic effects of predation on NAPCH recruitment is high. Also, given the evidence indicating poor nutritional condition and disease presence in the NAPCH, the effects of predation on this herd may be amplified.

Reliable information on the effects of nutritional condition, disease, and predation on calf production and summer survival will be crucial in future management decisions for this herd. This information will be used to determine effective strategies for attempting to increase subsistence and recreational opportunities in the range of this herd, as well as in making decisions on the appropriate level of human harvest. A project was initiated in 2005 to address this need, and this report summarizes findings from the first year of work. Goals of the project during 2005 included (1) conduct a parturition survey to estimate pregnancy rates for the herd in general and various calving areas if possible; (2) capture, take body measurements from, radio-collar, and monitor a sample of up to 60 neonate caribou, spread between calving areas if possible and without previously collared dams, to estimate neonate survival rates and causes of mortality; (3) collect a sample of neonate and adult caribou for use in combination with

mortalities investigated in conjunction with goal (2) above to identify pathogens, parasites, and assess trace mineral status in the population; and (4) begin to coalesce this information to determine the relative roles of nutrition, disease, and predation in the recruitment process (i.e. pregnancy, parturition, and calf survival rates) for NAPCH caribou.

Methods

Study area

The Alaska Peninsula is bordered on the north by Bristol Bay and on the south by the Pacific Ocean. The Bristol Bay Coastal Plain consists of flat to rolling tundra, lakes, poorly-drained meadows, and wetlands. The Pacific side consists of cliffs, fjords, and sandy beaches. The Aleutian Mountain Range separates these two areas, and it is characterized by steep glaciated mountains, canyons, and several active volcanoes. Concentrated caribou calving is usually centered around the Sandy and Cinder Rivers on the Bristol Bay Coastal Plain, and dispersed calving occurs throughout the Aleutian Mountain Range (Figure 2). Caribou predators occur throughout the Alaska Peninsula, and include wolves (*Canis lupus*), brown bears (*Ursus arctos*), wolverines (*Gulo gulo*), coyotes (*Canis latrans*), bald eagles (*Haliaeetus leucocephalus*), and occasionally golden eagles (*Aquila chrysaetos*). Caribou hunting by humans has been gradually restricted since 1998, and was closed beginning with the 2005-06 season.

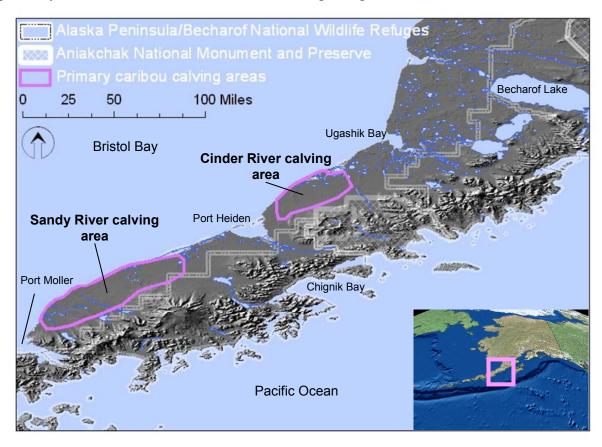


Figure 2. Concentrated caribou calving areas on the Alaska Peninsula. Calving area boundaries were generalized from Sellers et al. (2003).

Parturition survey

Between May 27 and June 1, 2005, we observed unmarked cows aged two years and older in the Cinder River and Sandy River calving areas to determine the NAPCH pregnancy rate. Yearling cows were excluded from the sample on the basis of body size. Cows were classified as pregnant or not pregnant using antler characteristics, udder distention, and/or neonate presence (Whitten 1995). All cows were classified from a Bellanca Scout fixedwing aircraft or Robinson R-44 helicopter.

Figure 3. ADFG King Salmon Area Biologist Lem Butler attempts to capture the newborn calf of a reluctant cow caribou.

Calf mortality

Between May 17 and May 26, three low-level reconnaissance flights were conducted using Aviat Husky and Found Bush Hawk airplanes to determine the general distribution of caribou. Previously deployed VHF and ARGOS satellite collars were used to direct these over flights. Beginning May 28, daily flights were conducted using Bellanca Scout and Aviat Husky airplanes and a Robinson R-44 helicopter to locate parturient cow caribou.

When post-parturient caribou were located, we attempted to capture all calves that appeared to be three days old or less. When a candidate calf was located, the helicopter landed as close as possible to the cow-calf pair, and one or two crew members were dropped off to capture the calf by hand (Figure 3). To avoid calf abandonment, we only captured dry calves (i.e. calves that were several hours old), and we avoided capturing calves that were in groups with other caribou. Also, each crew member wore clean latex gloves for each capture to avoid inter-calf scent transfer that may lead to abandonment (Adams et al. 1995b).

When calves were captured, their gender was determined and information was recorded on the condition of their umbilicus, hoof characteristics, and posture for use in estimating their age in days (Table 1). Each calf was weighed using a spring scale and a cloth sling that was disposed of after use to further prevent scent transfer. To estimate birth masses, calf weights were adjusted using estimated calf ages according to an equation developed from a large sample of calves in interior Alaska (Adams 2005). Calves were outfitted with VHF transmitters attached to elastic, expandable nylon collars designed to break off if the animal grows sufficiently large. Handling time for calves averaged less than one minute. The total time from when a calf was selected by the helicopter crew to when it was reunited with its dam after release averaged three minutes, excluding three captures during which the helicopter left before the animals reunited (all three of these cow-calf pairs were reunited when checked on the following day).

From May 28 through June 10, survival of marked calves north of Port Heiden was monitored daily, and survival of calves south of Port Heiden was monitored at approximately three-day intervals due to funding and logistical constraints. After June 10, all calves were monitored weekly through the first week in July, and then bi-weekly through the end of August. Prior to June 10, the time of death for calves could typically be pinpointed to within six hours.

After June 10, the date of death for calves was assumed to be midway between the date the mortality was detected and the date the animal was last located alive (Adams et al. 1995b). Following Adams et al. (1995b), we treated the first 15 days of life as the neonatal period for calves. The calf survival rates to 15 days of age and for ages 16 days and older were calculated using the Kaplan-Meier staggered entry procedure to account for calves that were not located during incomplete survey flights (Pollock et al. 1989).

Table 1. Criteria used for aging caribou calves, assuming that calves three or more days old could not be captured by hand. Table adapted from Adams et al. (1989).

Age	Criteria
1 Day (< 24 hrs)	-Running ability: Not, Barely, Wobbly
	-Umbilicus condition: Bloody, Wet
	-Hoof characteristics
	Color of rim: Light
	Hardness of rim: Soft, Partially Hard
	Hoof wear: Not, Lightly Worn
	-Posture
	Back: Hunched, Straight
	Hind legs: Bent and Splayed, Partially Straightened
2 Days (24-48 hrs)	-Running ability: Wobbly, Strong
, ,	-Umbilicus condition: Wet, Dry
	-Hoof characteristics
	Color of rim: Light, Dark
	Hardness of rim: Partially Hard, Hard
	Hoof wear: Lightly Worn, Worn
	-Posture
	Back: Straight
	Hind legs: Partially Straightened, Straight
3 Days (48-72 hrs)	-Running ability: Strong
, ,	-Umbilicus condition: Dry
	-Hoof characteristics
	Color of rim: Dark
	Hardness of rim: Partially Hard, Hard
	Hoof wear: Worn
	-Posture
	Back: Straight
	Hind legs: Straight

When a calf mortality was detected, the helicopter dropped a crew at the site to begin an investigation into the cause of death. Prior to June 10, this typically occurred within hours of the time of death. After a mortality site investigation during this time period, the carcass was transported to a field camp for a detailed necropsy. After June 10, calf deaths were investigated as soon as possible, which was within three weeks of the estimated date of death for most calves

that died during this period. For these investigations, a detailed necropsy was not performed because intact organs were usually not present.

Evidence accrued during site investigations and detailed necropsies was used to determine the proximate cause of death for each calf. When the cause of death was determined to be predation, several lines of evidence were used to distinguish between bear and wolf predation. A visual identification of a predator at the kill site was the primary indicator of the species of predator. If no predator was observed at the site, carcass remains (Figure 4), carcass caching patterns (Figure 5), hemorrhaging and puncture wound patterns (Figure 6), the spread of remains at kill sites (Figure 7), and predator tracks and/or hair (Figure 8) were used to determine the likely predator. Predator-killed calves located during 2005 were classified as bear kills, wolf kills, or unknown large carnivore kills if evidence was not present to distinguish between bear or wolf predation (species of predators other than bears or wolves were not detected), and were given a score of definite, probable, or possible predator kills by the field crew. Definite and probable predator kills are reported as predator kills; possible predator-killed calves are reported as uncertain cause of death. For mortalities investigated after June 10, on-site evidence of the cause of death was typically limited and dated. For this reason, evidence of predation was difficult to distinguish from evidence of scavenging.

For comparison with survival rate and cause of death information, wolf and bear encounter rates were estimated for calving areas north and south of Port Heiden during fixed-wing or helicopter flights in those areas. Flight start and stop times were noted, and the location and time were recorded for every bear and wolf seen during flights. Cub-of-the-year, yearling, and two-year-old bears, as well as wolf pups, were excluded from carnivore encounter rate calculations. Following the field season, the locations of all carnivore observations were plotted along with the locations of all marked calves. Minimum Convex Polygons (White and Garrott 1990) were created for calving areas north and south of Port Heiden based on all calf locations in those areas. Carnivore observations that were more than 20 kilometers from the border of these polygons were excluded from predator encounter rate calculations.

Parasites and diseases Field sample collection

Two cow caribou were captured and radio-collared for a separate project during field work reported on in this report. Samples were taken from these caribou for inclusion in analyses of diseases present in the NAPCH. Venipuncture was achieved in these caribou via the jugular vein using an 18 or 20 gauge, 1.5 inch needle and blood collected directly into Vacutainer® brand tubes (Becton Dickinson and Company, Franklin Lakes NJ) via a Vacutainer® adaptor. In addition, three adult caribou (two males and one female) were collected for diagnostic analyses. All edible meat was salvaged and donated to local village charities. Blood was obtained from these caribou immediately post mortem by cardiac venipuncture with a 60 ml syringe and an 18 gauge, three inch needle, then transferred to Vacutainer® tubes.

Four types of Vacutainer® tubes were filled during each blood collection: serum separator tube (SSTTM), EDTA anti-coagulated tube, sodium heparin trace mineral tube and plain trace mineral tube. The SSTTM and trace mineral plain tubes were chilled immediately after collection, allowed to clot for at least one hour and then centrifuged for 15 minutes. The serum was aspirated, aliquoted into cyrovials (Nalgene Company, Rochester NY) and immediately frozen on dry ice and stored at -50°C. A single five ml EDTA tube was collected for hematology. A one ml aliquot of whole blood was transferred from the heparinized trace mineral tube and

frozen in a cryovial. The tube was then centrifuged for 15 minutes and the plasma harvested and transferred in to a five ml Falcon® tube and frozen.



Figure 4. Typical remains at bear and wolf kill sites. Bears tended to eat all internal organs and the majority of bones (a). Remains at bear kill sites were commonly limited to hooves, skull cap fragments, miscellaneous bone fragments, and lower mandibles. Wolves tended to eat abdominal organs and muscle tissue first (b). Remains at wolf kill sites commonly included brain (brain case still intact), long bones, muscle tissue, and occasionally thoracic organs.



Figure 5. Wolf carcass caching pattern. Wolves buried parts of the carcass in neat caches that were difficult to find. The vegetation was pulled over the carcass, leaving the area looking untouched (a). With the aid of radio-telemetry, crews were able to find the carcass cache underneath the vegetation (b). Often we found half of a calf in the cache; the other half had been consumed by wolves. Bears are also known to cache unconsumed portions of kills, but the caching pattern is different. A bear cache looks like a large pile of grass and plant material; all of the material in a small area around the carcass is typically scraped up and piled on top of the carcass (no picture available).

Neonate samples

In facilities located at a field camp, detailed necropsies were conducted on the carcasses of deceased radio-collared calves as well as on a sample of four calves euthanized for diagnostic analyses. Gross examinations included morphometrics and examination of all body systems with narrative description of abnormalities, injuries, and other lesions. Whenever possible, a



Figure 6. Wolf and bear hemorrhaging and puncture wound patterns. Wolves typically left canine puncture wounds that were spread five centimeters or less (a). Hemorrhaging patterns and puncture wounds on the remains of wolf kills were localized such that bite patterns in specific areas of the body could be deciphered (b). Hemorrhaging patterns from bears were more diffuse than those from wolves, often resulted from swatting the neonate with the paw, and often spanned the length of the thorax and abdomen (c). Wound patters on calves killed by bears often included broken ribs and/or hemorrhaging on the internal organs (d). We did not examine any calf carcasses with punctures from bear canines, but we assume that such puncture wounds would be six centimeters or greater in spread.

preliminary diagnosis as to the manner of death was given. An intensive systemic collection of ten tissues were taken by sterile technique and transferred into individual sterile Whirlpac® bags for bacterial and viral culture when the carcass condition was suitable (i.e. recently deceased). Tissues collected included spleen, liver, kidney, lung, thoracic and gastrointestinal lymph nodes, and intestinal tract. These tissues were snap frozen on dry ice in the field and during shipping to storage facilities. These tissues were stored in an ultra-cold freezer until shipped to the laboratory for culture. Caudate liver lobe, kidney, and bone were collected and frozen for trace minerals analysis. A representative section of each tissue was collected and placed in ten percent neutral buffered formalin for histopathological analysis. Samples for parasitologic examination were collected, including lung, abomasal wall, and feces (or intestinal content). Additional fecal samples were collected for other diagnostic tests.



Figure 7. Spread of remains at a wolf kill site. Wolves often scattered carcasses. This probably occurs when several wolves are present at a kill site. The remains pictured in (a) and (b) were from the same calf and were located several hundred yards apart. Wolves were seen carrying pieces of this carcass when the mortality was first detected. Bears tend to eat entire carcasses without moving or scattering remains. The few remains that were found on most bear kills were commonly in small, discrete areas.



Figure 8. Bear scat (a) and wolf track (b) at kill sites. If no remains were found, time was spent looking for hair, scat, or tracks at kill sites.

Adult samples

Complete necropsies were conducted on adult carcasses in the field immediately post mortem. Gross examinations included morphometrics and examinations of all body systems, and tissues were collected using the techniques described for neonates above. Fourteen tissues were collected by sterile technique for bacterial and microbiology culture, including brain, spleen, liver, kidney, lung, thoracic and gastrointestinal lymph nodes, reproductive and intestinal tract tissue. Hair, caudate liver lobe, kidney and bone were collected and frozen for trace minerals analysis. Samples for parasitologic examination were collected as described above and were reserved for future diagnostic testing. A tooth was also removed and air-dried for aging. Bone marrow was collected and frozen for fat analysis.

Hematology

Hematocrit was determined by measuring packed red cells as a percent of blood volume after centrifugation in a microhematocrit tube at 10,000 x g for five minutes. Total protein was

Table 2. Dilution protocol used to develop stock standard for use in analyses of hemoglobin (Hb) levels in caribou.

Standard number	[Hb] g/dL	Hb standard (ul)	Drabkin's reagent (ml)
1	24	10	4
2	18	10	2.5
3	12	10	4
4	6	10	8
5	0	0	1

determined from plasma with a handheld refractometer (AO Scientific Instruments, Buffalo NY 14215). The total leukocyte count was determined manually using the Unopette® system (Becton-Dickinson and

Company, Franklin Lakes, NJ 07417) and a hemacytometer (Bright-Line, AO Scientific Instruments, Buffalo, NY 14215). The differential cell counts were made manually under oil immersion compound microscopy at 100x magnification on fresh blood smears stained with Wright's-Giemsa type stain (Dip Quick Stain, Jorgensen Laboratories, Loveland CO 80538) by identifying 100 leukocytes based on morphology. A single operator conducted all differential cell counts to maintain consistency.

To obtain reticulocyte counts, fresh whole blood was diluted one to one with new methylene blue stain (Jorgensen Laboratories, Loveland CO 80538) and allowed to sit for ten minutes before making a smear to count reticulocytes. Once completed, slides were examined under oil immersion at 100x magnification and 100 red cells were counted to determine a reticulocyte percentage and then corrected for the normal hematocrit and reported as corrected reticulocyte percentage.

In the field, 10μ l of whole blood was transferred into 2.5 ml Drabkin's reagent for preservation for hemoglobin determination. The assay was performed at the ADFG Marine Mammal Physiology Laboratory using the standard laboratory protocol. Briefly, potassium ferrocyanide oxidizes hemoglobin and its derivatives to methemoglobin. Potassium cyanide reacts with methemoglobin producing a more stable form of methemoglobin (Pointe Scientific Inc.; technical sheet P803-H7504-01, rev.1/02). Total blood hemoglobin measured as methemoglobin absorbs maximally at λ =540nm.

A stock standard was prepared by the addition of one ml of distilled water to Point Scientific Hemoglobin lyophilized standard. The resulting solution achieves a concentration of hemoglobin of 2400 g/dL. The stock standard was diluted (Table 2) and placed at room temperature for three minutes. Absorbance was determined on standards and samples using a SpectronicTM HeliosTM Gamma UV-Vis Spectrophotometer (Thermo Electron Corporation, San Jose, CA 95134). A regression equation from the standards was used to determine sample hemoglobin concentration.

Erythrocyte (red blood cell) indices were calculated from the hematocrit (PCV), red cell count (RBC), and hemoglobin (Hb) concentrations as follows:

Mean corpuscular volume (MCV) femtoliters = $PCV \times 10/RBC$ (106)

Mean corpuscular hemoglobin (MCH) picrograms = Hb x 10 / RBC (106)

Mean corpuscular hemoglobin concentration (MCHC) grams/ $dL = Hb \times 100/PCV$.

Parasitology

An attempt was made to obtain feces per rectum from adults that were immobilized and released. In addition, random fecal samples were collected off the ground during carcass

collections if they appeared to be relatively fresh. At necropsy, feces were collected from the rectum or descending colon as available. Fecal samples were frozen until analysis.

Zinc sulfate fecal floatation for the identification of parasitic ova was conducted using a sub sample of the previously frozen feces (less than two g) utilizing Ova Float Zn 118 (Butler, Dublin, OH 43017). After 10-15 minutes of floatation, wet mounted slides were examined under a compound microscope. Quantitative floatation of fecal samples was performed at the Mississippi State College of Veterinary Medicine, and the number and type of eggs per gram of feces was determined.

The Quantitative Beaker Baermann Technique was used to recover live larvae (Forrester and Lankester 1997). An envelope of nylon screen with a single layer of gauze was prepared and stapled on two sides. An approximate five g sample of feces was positioned in the screen envelope on top of the gauze and then placed horizontally submerged in a beaker containing 200 ml of tepid water. After 18 to 24 hours, the envelope was removed with forceps without disturbing the sediment. The contents of the container were allowed to settle for five minutes before further processing. The supernatant was then aspirated until approximately 30 ml of fluid remained. The contents of the beaker were swirled to mix and divided between two 15 ml centrifuge tubes. The tubes were centrifuged for ten minutes at 1500 rpm. The supernatant was then aspirated until two-four ml remained. The sample was next resuspended and the two tubes' contents combined. The sample was then centrifuged again. All but the last two ml of the tube contents were aspirated from the meniscus. The sediment was resuspended in the tube and transferred with a Pasteur pipette to glass slides. The sediment was examined under a compound microscope at 40x magnification. Larvae were differentiated into two types (i.e. dorsal-spined versus *Protostrongylus* sp.). The total numbers of larvae of each type were divided by number of grams of feces (usually five) to calculate the number of larvae per gram of feces.

Adult *Ostertagia* sp. and other abomasal nematodes (*Marshallagia* sp., *Telardorsagia* sp., and others) were recovered as follows. The contents and mucosa of the thawed abomasal walls were washed three times with a small amount of water in a bucket. Two aliquots of the material were then fixed in 10% formalin. The remaining unfixed contents were examined under a dissecting microscope and all adult nematodes were enumerated, collected and fixed in 80% alcohol for morphologic and molecular identification. Definitive identification of several species will be conducted at the National Parasite Collection and Animal Parasitic Disease Laboratory of the U.S. Department of Agriculture. Molecular techniques were applied to detect *Cryptospordium* and *Giardia* at Mississippi State University.

Pathology

Tissues fixed in 10% neutral buffered formalin were processed by routine paraffin imbedding, sectioned at six microns and stained with hematoxylin-eosin at Histology Consulting Services. Microscopic sections were examined and histopathologic diagnoses reported by Alaska Veterinary Pathology Services. Viral and bacterial cultures were conducted at Washington State Animal Disease Diagnostic Laboratory.

Serology

Serology analysis was conducted on 39 NAPCH serum samples collected during routine capture operations from 2001-2004 and ten NAPCH samples collected during this project in June 2005. Serologic testing for Bovine Respiratory Syncytial Virus (BRSV), Infectious Bovine Rhinotracheitis (IBR), Bovine Viral Diarrhea (BVD), Parainfluenza-3 (PI3), *Leptospirosis* 5

serovars, EHD, Toxoplasmosis, and *Neospora* were conducted at the Wyoming Veterinary Diagnostic Laboratory. Additional assays on retrospective sera for *Neospora* and Toxoplasmosis were conducted at the University of Tennessee College of Veterinary Medicine. *Brucella* serology was conducted at the University of Alaska, Fairbanks. *Brucella* testing consisted of both the Brucellosis Card Test (Becton Dickinson, Cockeysville, MD 21030), and a standard plate antigen test (Animal and Plant Health Inspection Service National Veterinary Services Laboratories, Ames, IA 50010). Both are macroscopic agglutination procedures. In the card test, 0.03 ml of serum is placed in a teardrop-shaped test area on the diagnostic card, then two drops (0.03 ml each) of *Brucella* antigen are placed on the card, not touching the serum. Using a stirrer, the antigen and serum are mixed and spread over the teardrop area. The card is then rocked for four minutes and observed. Specimens showing the characteristic agglutination pattern are classified as positive, and those with dispersed or no clumps are classified as negative.

For the standard plate antigen test, 10, 20, 40, and 80 μ L of serum were pipetted onto separate squares of an etched glass plate. Thirty μ L of standard plate antigen was then added to each square and thoroughly mixed. The plate was rotated four times and incubated for four minutes under cover to prevent drying. After four minutes, the plate was rotated again four times and incubated for another four minutes. After the second incubation, the plate was observed for degree of agglutination over indirect light.

Haptoglobins

Levels of serum haptoglobins were examined as another potential measure of sub clinical inflammation. Haptoglobin analysis was conducted on 39 NAPCH serum samples collected during routine capture operations from 2001-2004 and ten NAPCH samples collected during this project in June 2005. Serum haptoglobin levels were determined using a commercial gel electrophoresis kit and haptoglobin-hemoglobin binding assay (Titan Gel High Resolution Protein Kit #3040, Helena Laboratories, Beaumont TX 77704). A 14.5% solution of reindeer hemoglobin was obtained by dilution of washed red blood cells that had been lysed in sterile water and by freezing. Two µl of the 14.5% hemoglobin solution was added to each 38 µl of test sample serum in a microcentrifuge vial and mixed by vortex. Two µl of the mixture was placed on the gel previously prepared according to kit instructions. Electrophoresis was conducted for 45 minutes at 105 volts. Gels were stained with an o-dianisidine solution for ten minutes and rinsed three times according to the kit specifications. Haptoglobins were recorded as positive (having a band) or negative (no band) for each animal.

Blood Urea Nitrogen

Assays for Blood Urea Nitrogen (BUN) were performed at the ADFG Marine Mammal Physiology Laboratory according to the manufacturers instructions for Sigma Urea Nitrogen Kit # 640A (Sigma-Aldrich Inc. St. Louis MO 63103). Samples were diluted one to one with deionized (DI) water (150 µl sample + 150 µl water) and the kit standard was diluted from 30 mg/dL to one to one as well. The spectrophotometer (SpectronicTM HeliosTM Gamma UV-Vis Spectrophotometer, Thermo Electron Corporation, San Jose, CA, 95134) was warmed up for 30 minutes at 570 nm. The Urease Buffer reagent was reconstituted with 30 ml DI water. Two blanks, three standards, and the samples were prepared in duplicate. Into each tube, the following was added: 125 µl Urease solution, 10 µl DI water for blank, and standard (one to one) or sample (one to one). The resulting solutions were gently mixed by vortex. After standing for 15-20

minutes at room temperature, the following were added in turn and mixed after each addition: $250 \mu l$ phenol nitroprusside solution, $250 \mu l$ Alkaline hypochlorite solution, and 1.25 ml DI water. After standing for 20 to 30 minutes at room temperature under a hood, the samples were vortex mixed, and absorbance at 570 nm was read. BUN is calculated as follows: BUN (mM) = (Sample A – Blank A)/(Standard A- Blank A) * 10.7.

Bone Marrow

As an additional indicator of nutritional condition, bone marrow fat content of NAPCH caribou was determined and compared to samples available from elsewhere in Alaska. Previously frozen bone marrow samples were weighed in a tared 12 oz plastic cup. The cups were held at 65 degrees Celcius in an incubator for 48 hours and then re-weighed. Samples were then returned to the incubator for an additional 24 hours and a final weight was obtained. The final dry weight was used to determine the water loss and calculate the percent fat from the remaining mass less the connective tissue (Neiland 1970).

Trace Minerals

Trace mineral analysis will be conducted in the Wildlife Toxicology Laboratory at the University of Alaska Fairbanks. Blood, serum and plasma will be analyzed from the live captured animals and liver, kidney and bone from carcasses. Analyses will include selenium, cadmium, iron, copper, molybdenum, and lead levels.

Results

Parturition survey

A total of 314 caribou were classified during parturition survey flights. The overall estimated pregnancy rate was 57% for cow age two and older (Table 3). The pregnancy rate for caribou south of Port Heiden was slightly higher than the pregnancy rate for caribou north of Port Heiden (Table 3).

Table 3. Caribou cow parturition class	ssification summary.
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Parturition classification	North of Port Heiden	South of Port Heiden	Total
Cows with calves	21	31	52
Cows with hard antlers	3	55	58
Cows with distended udders	22	46	68
Cows with no evidence of pregnancy	65	71	136
Total number of cows classified	111	203	314
Estimated pregnacy rate (≥ 2 yrs old)	41%	65%	57%

Calf mortality

Forty-two calves were captured and radio-collared to monitor calf mortality. Most calves were captured and relocated in the Cinder River and Sandy River calving areas, but several calves were also captured and relocated in areas of dispersed calving (Figure 9). No cow abandonments resulting from capture operations were detected.

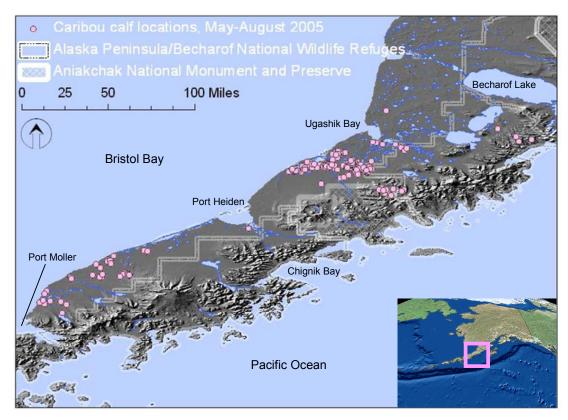


Figure 9. Calf capture and aerial telemetry locations during May-August of 2005.

The estimated birth mass of calves was 7.72 kg (se=0.23 kg, n=27) and 7.78 kg (se=0.34 kg, n=15) for males and females, respectively (Figure 10). Most projects do not account for the age of calves at the time of measurement, making direct comparisons of calf masses in the NAPCH to masses of calves in other herds difficult. However, the 2005 NAPCH estimated calf masses were comparable to the average recorded during the period from 1987-1997 in the Denali Caribou Herd, where calf age was accounted for (Figure 10).

Calf survival to 15 days of age was estimated as 0.43

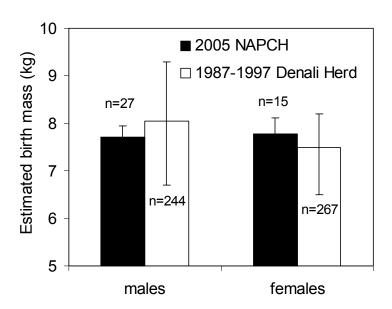


Figure 10. Estimated birth masses of NAPCH caribou in 2005 compared to the Denali Caribou Herd from 1987-1997 (data from Adams 2005). Error bars for the NAPCH data represent 1 SE, and error bars for the Denali Herd represent the range of means observed during that project.

(se=0.08), and survival of calves age 16 days and older, through August 31, was estimated as 0.19 (se=0.10, Figure 11). Calf survival to 15 days is comparable to that reported for stable and increasing interior Alaska caribou herds of relatively small size (i.e. less than 25,000 animals), but survival of calves age 16 days and older was much lower than that reported for those same interior Alaska herds (Figure 11).

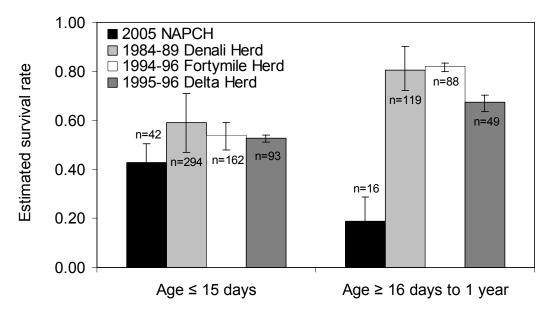


Figure 11. Estimated Kaplan- Meier survival rates of NAPCH calves in 2005 compared to survival rates of small, stable or increasing interior Alaskan herds. The reporting period for the 2005 NAPCH data is May 28- August 31. The Denali Herd numbered approximately 2500 animals and was increasing at approximately 6-8% annually during the time period depicted (data from Adams et al. 1995a,b). The Fortymile Herd numbered approximately 23,000 animals and was increasing at approximately 11% annually during the time period depicted (data from Boertje and Gardner 1998). For the Fortymile Herd, calf deaths reported during the months of May-June are included in the age ≤15 days category, and calf deaths reported during the months of July-October are included in the age ≥16 days to 1 year category (see Boertje and Gardner 1998). The Delta Herd numbered approximately 2600 animals and was approximately stable during the period depicted (data from Valkenburg 1997). For the Delta Herd, calf deaths reported during the period from May 15 - June 15 are included in the age ≤15 days category, and calf deaths reported during the period from June 16 - September 30 are included in the age ≥16 days to 1 year category (see Valkenburg 1997). Error bars for the NAPCH data represent 1 SE; error bars for other herds represent the range of estimates observed in those studies.

A total of 22 calf deaths were investigated prior to June 10, and 11 deaths were investigated from June 10 through mid August. For calves aged 15 days or fewer, the primary cause of death was predation (Figure 12). Bear predation (n=9 calves) was as common as wolf predation (n=9 calves) for this age bracket. For calves aged 16 days and older, predation and scavenging could not be clearly distinguished as causes of death due to incomplete and dated evidence, so these causes of death were lumped into one category. For this age bracket, bear

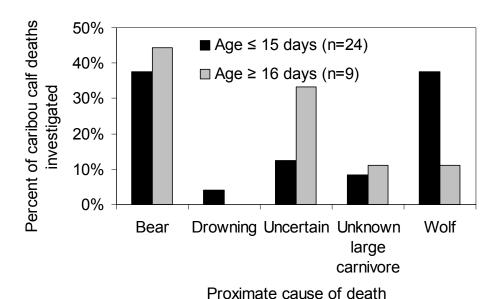


Figure 12. Proximate causes of death for 33 NAPCH calves as determined from mortality site investigations and detailed necropsies. For calves age ≥16 days, predation could not be distinguished from scavenging. Mortality site investigations in the bear, unknown large carnivore, and wolf categories for this age bracket should be interpreted as either predation or scavenging.

scavenging/ predation was more common (n=4) than wolf scavenging/ predation (n=1), but uncertain causes of death were also common (n=3). Predator encounter rates were monitored on 27 flights north of Port Heiden and on eight flights south of Port Heiden A total of 88 bears and 31 wolves were seen on these flights. All carnivore observations

were within 20 km of the Minimum Convex Polygons surrounding calf locations, so no carnivore observations were excluded from predator encounter rate calculations. Prior to June 10, when most of the predator-attributed calf deaths were detected, the predator encounter rate south of Port Heiden was approximately double the predator encounter rate north of Port Heiden (Figure 13). Correspondingly, the survival of caribou neonates to 15 days of age, which approximately coincides with the period prior to June 10, was twice as high north of Port Heiden as south of Port Heiden (Figure 13).

Parasites and diseases Neonate Pathology

Four calves that died during the calf mortality portion of this project were suitable for histopathologic examination. One of these calves had no unexpected histologic lesions. One calf, which died of massive trauma to the chest and inguinal areas, had a chronic mild, multifocal, degenerative cardiomyopathy (histiocytic myocarditis) with areas of myofiber loss. Since the calf was only four days old and the lesion is chronic, it may be due to a toxic or nutritional cause. One calf terminally inhaled pond water, but there was no evidence of inflammatory disease. The final calf possibly had an unusual amount of embryonic rest cells in the ventricles and white matter of the brain (a second opinion is pending), but this may be within normal limits for a young animal. The same calf also had mild lymphocytic inflammation around the bile ducts, which may be due to blockage, parasite migration, an autoimmune disorder, or a drainage reaction.

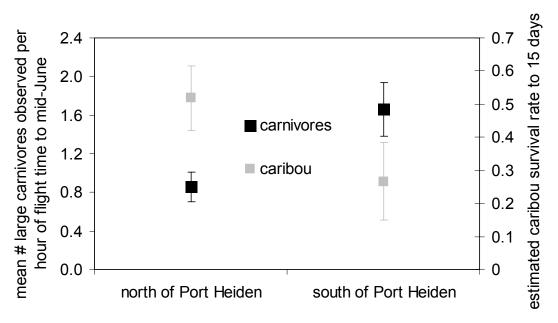


Figure 13. Predator encounter rate prior to mid-June compared to caribou neonate survival north and south of Port Heiden. Note the inverse relationship; in the area where predators were more frequently encountered, survival of caribou neonates was lower. Error bars represent 1 SE.

Of four calves euthanized for diagnostics, two had no significant histologic lesions in any of the survey tissues examined (left ventricle, abomasum, liver, kidney, lung), and one had a mild eosinophilic enteritis/colitis and very mild interstitial nephritis. The remaining collected calf had a mild bronchitis and bronchiolitis with possible intracytoplasmic and intranuclear inclusion bodies. The lung was sent for viral culture for PI3 or BRSV and results were negative. It is unusual to see these lesions in a neonate and further diagnostic testing is being pursued including PCR and immunohistochemistry.

Overall, five of the eight calves examined had a mild vacuolar hepatopathy, which is not unexpected in neonates, giving the cytoplasm a moth-eaten appearance.

Neonate Hematology

A total of four samples were analyzed for leukocyte parameters from NAPCH neonate caribou (Table 4). In general, leukocytes were depressed in NAPCH caribou compared to reference ranges. Erythrocyte samples were analyzed from four NAPCH calves for most indices, though sample were only available from a single calf for some indices (Table 5). Data were limited for these indices, and generalizations are not possible.

Neonate Parasitology

No parasites were seen grossly in calves during necropsies, however a number of parasite eggs and larvae were detected during fecal exams (Table 6). Additionally, fecal samples from nine calves were analyzed by molecular methods for *Cryptosporidium* and *Giardia*, and all samples were negative.

Table 4. Mean ± SE, median, and minimum to maximum ranges of leukocyte parameters in NAPCH neonates compared with means and reference ranges (Mean±2SD) for normal, newborn captive reindeer calves age 0-7 days (J. Blake, unpublished data). Bold values indicate parameters for caribou and reindeer for which 95% confidence intervals do not overlap.

	NAPO	CH Calves	Reinde	Reindeer Calves		
	1-3 mo.,	n = 4 samples	n	= 20		
_	Mean ±SE	Median (Min-Max)	Mean ±SE	Reference Range		
Total Leukocytes/µl	2,284 ±130	2,206 (1,225-3,500)	5,085 ±437	1,171-9,000		
Neutrophils:Lymphocytes	1.9 ±0.13	2.1 (0.4-3.2)	2.6 ±0.31	0.9-7.0		
Neutrophils/µl	1,026 ±150	1,408 (294-2,380)	2,957 ±451	0-6,994		
Band neutrophils/µl	64 ±6.58	66 (0-124)	2 ±2.23	0-22		
Lymphocytes/µl	644 ±18.5	649 (472-809)	1,579 ±172	35-3,124		
Monocytes/µI	158 ±9.9	137 (79-279)	161 ±48	0-656		
Eosinophils/µI	61 ±5.15	48 (25-124)	31 ±12	0-139		
Basophils/µl	0	0	3 ±1.78	0-19		

Table 5. Mean ± SE, median, and minimum to maximum ranges of erythrocyte indices in NAPCH neonates compared with means and reference ranges (Mean±2SD) from normal, newborn captive reindeer calves age 0-7 days (J. Blake, unpublished data).

	NAPC	H Calves	Reinde	Reindeer calves		
	n = 4		n	n = 25		
	Mean ±SE Median (Min-Max)		Mean ±SE	Reference Range		
Total protein g/dl	5.8 ±0.35	5.8 (5.2-6.2)	5.3 ±0.12	4.1-6.6		
Hematocrit (Hct) %	35.4 ±11.5	27.4 (26.7-60.0)	35 ±0.86	30.8-39.3		
Hemaglobin (g/dl)	13.1 ±3.39	14.5 (7.8-17.2)	12.76 ±0.28	9.9-15.6		
RBC (x106/ul)	8.3*		7.6 ±0.18	5.7-9.5		
CRP	2.7 ±0.98	2.5 (1.4-4.6)	na			
MCV (fl)	72.7*		45 ±0.34	41.6-48.4		
MCH (pg)	20.8*		16.9 ±0.12	15.7-18.1		
MCHC (g/dl)	28.4*		37.8 ±0.28	34.9-40.6		

^{*} Only one individual had a red cell count available for calculation of indicies

Adult Pathology

Three adult caribou from the Northern Alaska Peninsula Herd were selected for euthanasia for detailed disease assessment. The first adult (ADFG necropsy #106878, approximately 2 year old male) was in poor body condition and heavily parasitized. Grossly visible parasites included *Setaria* sp, *Sarcocystis* sp., nasal bots (*Cephenemyia trompe*), *Taenia hydatigena*, *Echinococcus granulosus*, rumen flukes, and abomasal lesions consistent with *Ostertagia* sp. During histopathologic examination, it was evident that there were large numbers of eosinophils throughout the tissues examined, indicative of a heavy parasite infestation. There were eosinophils in the lymph nodes, spleen, lung (with macrophages, plasma cells, and lymphocytes), an eosinophilic and lymphocytic reaction of the lamina propria of the rumen (with a fluke on the surface), an eosinophilic reaction in the pericardium (along with intramyofibrillar protozoal cysts), and an eosinophilic reaction in the abomasal wall, along with encysted nematode larvae (most likely *Ostertagia*). There was also a mild meningoencephalitis in the brainstem, which was determined to be caused by *Sarcocystis* sp. The kidneys showed a mild interstitial nephritis, and special stains are pending to rule out Leptospirosis. Finally, there was

CRP = Corrected Reticulocyte Percentage

MCV = Mean Corpuscular Volume

MCH = Mean Corpuscular Hemaglobin

MCHC = Mean Corpuscular Hemaglogin Content

Total number of positive samples Parelaphostrongylus 4 6 1 sp. larvae Beaker Baermann test Ostertagia sp. larvae Protostrongyloid larvae ယ Marshallagia Flotation Trichostrongyle Coccidia test (minimum) Monezia Cestode

Table 6. Fecal examination results for parasite larvae from NAPCH neonates. Eight neonates were surveyed for each parasite

species

an area in the mesentery that contained what was consistent with a hemangioma, a benign tumor that is an incidental finding.

The second animal (ADFG necropsy #106879, approximately 10 year old female) was emaciated and limping. She was also heavily parasitized; parasites included warbles (Oedemagena tarandi), nasal bots, Sarcocystis, Taenia krabbei, Taenia hydatigena, rumen flukes, Ostertagia sp. and lungworm (likely Dictyocaulus sp.). She had areas of alopecia (hair loss with skin and hair discoloration) on the caudolateral thighs, feet, legs, and perineum. There was also evidence of diarrhea (perineal staining), a swollen hock, evidence of gastrointestinal inflammation, and possibly metritis. Much of the histologic findings were similar to the first animal, showing evidence of heavy parasitism, especially in the lungs. This animal also had lesions consistent with enteritis, and had a rumen fluke embedded in the duodenal wall (this is an abnormal location and can cause pathology). The areas of alopecia were associated with hyperplastic dermatitis, and most likely were due to biting insects and the resultant scratching. Interestingly, this animal had several areas of inflammation in the brain, along with basophilic inclusion bodies, which were caused by a Sarcocystis. There was mild interstitial nephritis in this animal as well as chronic active metritis. A culture of the uterus yielded many Echinococcus coli and moderate growth of *Enterococcus* sp. Culture of the uterus and serology were negative for Brucella sp. Examination of the hock remains to be undertaken.

The final animal (ADFG necropsy #106883, adult male) was in poor body condition, had some alopecia (hairloss), and was seen lagging behind the other animals and stopping to cough while being pursued by the helicopter. Gross necropsy revealed chronic pleuritis, enlarged lymph nodes, a pancreatic mass, and heavy parasitism (*Ostertagia* sp., *Sarcocystis* sp., *Echinococcus* cysts, and rumen flukes). Histologically there was evidence of heavy parasite load, as in the other adults. In the gastrointestinal tract, there was evidence of parasitic infection (nematode eggs and *Balantidium*-type organisms), along with damage to the mucosal surfaces and an eosinophilic infiltrate. A granuloma was also present on the liver. This animal also had frequent areas of inflammation in the brain (perivascular cuffs of lymphocytes, plasma cells, and macrophages), and some free red blood cells in the meninges. This was a nonspecific reaction caused by a *Sarcocystis*. Curiously, the thyroid was inactive.

To summarize, these animals were heavily parasitized, all three had infections of *Ostertagia* sp., high numbers of *Sarcocystis* in the skeletal muscles including the abdominal muscles and diaphragm, and rumen fluke infections ranging from moderate to severe. Two of these animals had severe nasal bots, *Taenia hydatigena* cysts in the liver, and *Echinococcus granulosus* cysts in the lungs. Two of these animals also had a mild interstitial nephritis, which may not be uncommon in caribou,

however it may also be Leptospirosis. They also each had varying degrees of meningoencephalitis, caused by a *Sarcocystis*. The *Sarcocystis* species that are known to cause meningoencephalitis in North America require virginia oppossums (*Didelphis virginiana*) as intermediate hosts. As this species is not present in Alaska, there either must be a different intermediate host, or perhaps this is a species of *Sarcocystis* that was not previously known to cause meningoencephalitis.

Adult Hematology

Adult NAPCH caribou had low total leukocyte counts, as well as low neutrophil, lymphocyte, and eosinophil counts, compared to a reference wild caribou herd (Table 7). In light of the heavy parasitism and chronic disease state of these individuals, these leukgrams indicate a poor immune response to the ongoing infections. Thus, the adults surveyed were either immunosuppressed or suffering from immune system exhaustion from the chronic stress caused by parasites.

Adult NAPCH caribou had high hematocrits, but not total protein, while erythrocyte counts were low compared to a reference wild caribou herd (Table 8). Low erythrocyte counts with increased Mean Corpuscle Volume (MCV) and Mean Corpuscle Hemoglobin concentration (MCH) may indicate that the NAPCH animals were suffering from macrocytic hypochromic anemia, which usually results from chronic blood loss. High hematocrits in combination with low erythrocyte counts is indicative of dehydration. In a state of dehydration, the total protein is usually increased because of the loss of plasma water. The lack of an increase in the total protein in these NAPCH adults is likely due to a loss of plasma protein or low production of plasma protein. A loss in total protein could be related to the same chronic blood loss that appears to be causing anemia in these animals. Low production of plasma protein could be caused by a poor nutritional plane or energy required to fight diseases. These animals were all in very poor body condition with no fat reserves. The lack of increased total protein while the animals are dehydrated is likely a result of both blood loss due to high numbers of blood-sucking parasites and low protein production associated with either low protein intake or high muscle catabolism for energy production.

Table 7. Mean ± SE, median, and minimum to maximum ranges of leukocyte parameters in adult NAPCH caribou compared with means and reference ranges (Mean±2SD) from free-ranging arctic caribou from the Teshekpuk Lake Herd (TSH) collected in June-July 2005 (K. Beckmen, unpublished data). Bold values indicate parameters for the NAPCH and TSH for which 95% confidence intervals do not overlap.

	NAPCH Adults n = 6 Mean ±SE Median (Min-Max)		TSH Adults n = 17		
-			Mean ±SE		
Total Leukocytes/µl	5,679 ±1209	4,675 (2,825-11,625)	9,442 ±599	4500-14384	
Neutrophils:Lymphocytes	0.9 ±0.24	0.8 (0.4-1.7)	0.8 ±0.12	0-1.8	
Neutrophils/µl	1,296 ±104	1,219 (1,007-1,737)	2,042 ±168	654-3430	
Band neutrophils/µl	51 ±17	59 (0-97)	152 ±33	0-430	
Lymphocytes/µl	1,706 ±347	1,518 (841-2,790)	3,011 ±301	525-5497	
Monocytes/µl	369 ±77	438 (77-581)	701 ±116	0-1661	
Eosinophils/µl	2,238 ±953	1,406 (994-6,975)	3,624 ±390	400-6848	
Basophils/µI	0	33 (32-34)	0		

Table 8. Mean ± SE, median, and minimum to maximum ranges of erythrocyte indices in adult NAPCH caribou compared with means and reference ranges (Mean±2SD) from free-ranging arctic caribou from the Teshekpuk Lake Herd (TSH) collected in June-July 2005 (K. Beckmen, unpublished data). Bold values indicate parameters for the NAPCH and TSH for which 95% confidence intervals do not overlap.

	NAPCH Adults n = 6			TSH Adults n = 17		
•	Mean ±SE Median (Min-Max)		Mean ±SE	Reference Range		
Total protein g/dl	6.4 ±0.16	6.6 (5.7-6.7)	6.5 ±0.14	5.3-7.7		
Hematocrit (Hct) %	54.6 ±2.6	55.1 (44.8-62.0)	46.1 ±0.97	38.1-54.1		
Hemaglobin (g/dl)	17.8 ±0.89	18.0 (14.6-20.30)	16.8 ±0.70	11.0-22.6		
RBC (x10 ⁶ /ul)	8.5 ±0.82	9.2 (5.8-10.4)	10.3 ±0.24	8.3-12.3		
CRP (%)	1.5 ±0.44	1.4 (0-3.1)	na			
MCV (fl)	66.7 ±5.3	61.8 (55.4-89.9)	45.0 ±0.70	39.2-50.8		
MCH (pg)	21.7 ±1.55	20.1 18.7-28.3)	16.0 ±1.35	4.8-27.2		
MCHC (g/dl)	32.7 ±0.28	32.7 (31.5-33.7)	36.3 ±1.06	27.5-45.1		

CRP = Corrected Reticulocyte Percentage

MCV = Mean Corpuscular Volume

MCH = Mean Corpuscular Hemaglobin

MCHC = Mean Corpuscular Hemaglogin Content

Adult Parasitology

Large numbers of parasites were detected during gross and fecal examinations (Table 9, Table 10). Confirmation of results from the National Parasite Collection and Animal Parasitic Disease Laboratory of the U.S. Department of Agriculture for the species of adult and larval parasites detected are expected in October 2005. Feces from the seven random adult fecals and the three adult collected animals were all negative for *Cryptosporidium* and *Giardia*.

Serology

Between 1999 and 2001, NAPCH caribou had a remarkable emergence in antibody prevalence for BRSV, IBR, PI-3, and *Neospora* (Table 11). The first three are viral diseases that comprise the bovine respiratory complex. In cattle, these agents are rarely fatal by themselves. They can establish relatively mild viral infection of the lungs (pneumonia or pneumonitis). More importantly, they damage the lungs and provide an opportunity for bacterial infections to become established, especially in the presence of lungworms. The bacterial infections can then progress into more serious and potentially fatal pneumonia. The positive serology for the parasites *Neospora* and *Toxoplasma* are of interest because they may indicate exposure to domestic animals. However, seropositive caribou have been detected elsewhere in the state and at this time we have not established pathology associated with these parasites in caribou.

Haptoglobins

Haptoglobin levels were compared with hemogram values to confirm an expected correlation with indicators of an inflammatory leukogram. However, all NAPCH caribou samples were negative for haptoglobins.

Blood Urea Nitrogen

Concentrations of BUN were determined in the 4 calf and 6 adult NAPCH serum samples available from 2005 and compared with 17 neonatal captive reindeer and 61 4-16 month old interior herd caribou from 2004. The NAPCH calves had lower mean BUN than reindeer calves,

6.6 mg/dL (SE= 1.3, range 5.2-8.0) vs. 20.8 mg/dL (SE= 10.5, range 8 – 50) respectively. The NAPCH adults had mean BUN concentrations higher than the 4 to 16 month old Interior caribou, 7.7 mg/dL (SE= 2.03, range 4.5-10.0) vs. 2.2 mg/dL (SE=0.9, range 1.2 – 4.8) respectively. This is likely an additional indicator of dehydration rather than decreased renal function.

Bone Marrow

Bone marrow was analyzed from the three euthanized adult NAPCH caribou. The mean fat content in marrow from NAPCH caribou was 56.9% (SE=22.6, range 31.6-75.1), compared to 60.3 % (SE=18.6, range 32.7-93.3) from ten caribou marrow samples available from elsewhere in Alaska. While the mean percent fat content was slightly lower in NAPCH caribou, 95% confidence intervals for the two groups overlapped almost entirely.

Trace Minerals

Trace mineral analyses are ongoing and results should be available by the end of 2005.

Discussion

The primary goal during the first year of this project was to begin collecting data that will be useful in determining the influences of nutritional condition, disease, and predation on recruitment in the NAPCH.

Final conclusions should not be drawn solely from data collected in 2005. Research in other areas has highlighted how variable caribou populations and the factors that influence them are, and how long it takes to gain insight into complex ecological relationships between caribou and other aspects of the ecosystems that they inhabit (Adams 2005, Valkenburg et al. 2002, Boertje and Gardner 1998). In spite of this,

Table 10. Fecal examination results for parasite larvae from NAPCH adults, including three necropsied animals and sever random fecal samples (ten adult fecal samples were surveyed for each parasite species) Parelaphostrongylus 4 6 1 sp. larvae Ostertagia larvae Protostrongyloid Marshallagia Trichostrongyle Coccidia Monezia Cestode

Total number of

positive samples

PCCICO.											
	Abomasal Lesions	Ostertagia sp.	Setaria cervi	Abomasal Ostertagia Setaria Sarcocystis Lesions sp. cervi sp.	Nasal Bots (<i>Cephenemyia</i> <i>trompe</i>)	Nasal Bots Warbles Lungworm Taenia (Cephenemyia (Oedemagena (Dictyocaulus hydatiger trompe) tarandi) sp.) cyst	Lungworm (<i>Dictyocaulus</i> sp.)	Taenia hydatigena cyst	Echinococcus granulosus cyst	Rumen Flukes Taenia (Paramphistomes krabbe sp.) cyst	Taenia krabbei cyst
otal number of		7		(7	trompe)	tarandi)	sp.)	cyst		sp.)	cyst
positive	ω	ω	_	ω	2	_	_	2	2	ω	_

identifications

Table 9. Parasite survey results from detailed necropsies of adult NAPCH caribou. Three adults were surveyed for each parasite

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Table 11. Results of surveillance [as positive/total (% prevalence)] of NAPCH serum samples for selected disease agents compared to samples from free-ranging arctic caribou from the Teshekpuk Lake Herd (TSH) collected during 1986-2001. Data from 1982-1999 NAPCH and 1986-2000 TSH caribou are from Zarnke (2000), and data from 2001 TSH caribou are from K. Beckmen (unpublished data). Bold values indicate agents with high prevalence rates in the NAPCH in recent years.

		NAP	СН	TSH
Agent -	1982-1999	2001-2004	2005	1986-2001
Bovine Respiratory Synchial Virus	0/189 (0%)	20/34 (59%)	2/10 (20%)	1/58 (2%)
Brucella suis	0/177 (0%)	0/39 (0%)	0/10 (0%)	2/77 (3%)
Bovine Viral Diarrhea	0/89 (0%)	0/35 (0%)	0/10 (0%)	9/52 (17%)
Enzootic Hemorhagic Disease	0/194 (0%)	0/34 (0%)	0/10 (0%)	0/63 (0%)
Infectious Bovine Rhinotracheitis	0/188 (0%)	13/35 (37%)	7/10 (70%)	0/60 (0%)
Parainfluenza-3	0/145 (0%)	9/39 (23%)	7/10 (70%)	0/60 (0%)
Leptospirosis	0/185 (0%)	0/34 (0%)	0/10 (0%)	0/45 (0%)
Neospora	na	4/31 (13%)	0/10 (0%)	1/4 (25%)
Toxoplasma	0%	0/31 (0%)	1/10 (10%)	0/5 (0%)

management needs and variable annual agency budgets require that some comparisons and preliminary conclusions be drawn. These conclusions are tentative, and data interpretations are likely to change as more data accumulate.

Birth masses and pregnancy rates tend to decline in caribou populations that are in poor nutritional condition (Valkenburg et al. 2002). Birth mass and pregnancy rate data collected in 2005 from the NAPCH were somewhat ambivalent regarding the nutritional condition of the herd. NAPCH birth masses were approximately average compared to data spanning increasing and decreasing phases of the Denali Caribou Herd (Figure 10, Adams 2005). This may indicate that NAPCH caribou are not in peak condition, nor are they in poor condition. Comparisons with herds other than the Denali Herd are not straightforward because caribou neonatal growth rates are typically not accounted for. Most caribou calves can be captured by hand during their first three days of life (Adams et al. 1989), and growth can exceed 10% of body mass during this period (Adams 2005). Comparisons of mass between caribou herds are also problematic in general due to baseline differences between mass and growth rates between herds (Valkenburg et al. 2002). This suggests that more data are required to document the range of variability in NAPCH birth masses before accurate extrapolations can be made using the relationship between birth masses and adult female nutritional condition.

Parturition survey data from 2005 provides more substantial evidence for poor nutritional condition in NAPCH caribou. The estimated overall pregnancy rate for the NAPCH was 57%, which is low for an Alaskan caribou herd (Valkenburg et al. 2002). Due to our methods, two-and three-year-old cows are included in our parturition sample, and these age classes have lower pregnancy rates than older caribou (Adams and Dale 1998a). This may cause a low bias in our pregnancy rate estimate. However, due to low recruitment rates in recent years (Figure 1), the proportion of cows in the NAPCH that are two- and three-year-olds should be minimal, especially given the additional mortality that occurs in the yearling and two-year-old age classes. Further, NAPCH pregnancy rates have been estimated in previous years using the same methodology employed here, resulting in pregnancy rate estimates more than 20% higher than those recorded for 2005 (Sellers et al. 2003). Those previous estimates were marked by a similar spatial pattern to that observed in 2005, in which the pregnancy rate appeared to be higher south of Port Heiden than north of Port Heiden (Table 3, Sellers et al. 2003). While the cause of this

pattern is unknown, it adds credence to the notion that the low pregnancy rate observed in 2005 is in fact a real trend.

The low NAPCH pregnancy rate suggests that the herd may be in poor nutritional condition, in concurrence with previous hypotheses regarding the cause of the NAPCH decline (Sellers et al. 2003). For other caribou herds, poor nutritional condition in cows has been documented to result from poor range conditions (Valkenburg 1997) and severe winters (Adams and Dale 1998a). We did not collect data on these potential causes during 2005. However, another potential cause of poor nutritional condition in the NAPCH is the heavy parasitism detected in 2005. Hematologic results from the live-captured and collected caribou support the conclusion that disease and parasites are adversely affecting the health of NAPCH caribou. The total circulating leukocytes as well as the subpopulations of neutrophils, lymphocytes, and eosinophils, which are all blood cells involved in fighting diseases and parasites, were depressed in NAPCH adult caribou (Table 7). This pattern of depressed immune system function was also present in NAPCH neonates (Table 4). NAPCH adult blood samples also had decreased numbers of erythrocytes, which are blood cells involved in respiration and metabolism (Table 8). NACPH erythrocytes were large and had low concentrations of hemoglobin, which is indicative of chronic anemia when considering the low erythrocyte counts. Further, NAPCH hematocrits were increased while total protein counts were slightly low, which may indicate that NAPCH caribou are somewhat dehydrated and that their abomasum function has been compromised.

Several of the parasites detected in the NAPCH may have population-level effects. In particular, *Ostertagia* sp. has been shown to decrease food intake, body condition, growth, and pregnancy rates in reindeer populations (Stein et al. 2002, Arneberg et al. 1996). The effects of this parasite alone are likely strong enough to regulate wild reindeer populations (Albon et al. 2002). Respiratory diseases may decrease survival and pregnancy rates, as they have been documented to do in other ungulate species (Jolles et al. 2005). Lastly, *Toxoplasma nativa* and *Neospora sp.* may contribute to increased fetal loss during gestation and the birth of weak calves with low survival and development rates.

Survival data from marked calves also supports the notion that NAPCH calves may have poor development rates. The primary cause of death detected for calves was predation (Figure 12), indicating that predators play a role in the survival of NAPCH calves. This pattern is consistent with every caribou herd where calf survival rates have been studied (ex. Arthur and Del Vecchio 2003, Valkenburg et al. 2002, Boertje and Gardner 1998, Adams et al. 1995a,b). We detected more than 50% mortality during the first 15 days of life in NAPCH caribou, which is common even in stable and increasing caribou herds (Figure 11). This mortality was caused predominately by predation (Figure 12), which is also common in stable and increasing caribou herds (Valkenburg et al. 2002, Boertje and Gardner 1998, Adams et al. 1995a,b). During this period, we also found that predation rates were higher in areas where predator encounter rates were higher (Figure 13).

In stable or increasing herds, survival rates of calves typically increase dramatically beginning at approximately 16 days of age (Adams et al. 1995a,b), and can approach survival rates of adult animals (Boertje and Gardner 1998, Adams et al. 1995b). This pattern may reflect decreasing vulnerability of calves to predation as they rapidly grow and develop motor and sensory skills (Adams 2003, Adams et al. 1995a,b). In 2005, NAPCH calf survival rates for animals age 16 days or older were approximately 40-60% lower than survival rates of similarage calves in stable or increasing herds of similar size (Figure 11). While causes of death for calves age 16 days and older should be interpreted with caution due to larger elapsed time

periods between the dates of death and the date of site investigations, the majority of mortality site investigations conducted for animals in this age bracket revealed evidence of scavenging or predation, primarily by brown bears (Figure 12). If these animals were killed by predators, this may indicate that calves are not developing at a rate sufficient to decrease their vulnerability to predation.

A pattern of high late-summer mortality of caribou calves due to predation has been detected in at least one other decreasing caribou herd of similar size to the NAPCH (Adams et al. 1995a). In this caribou herd, the late summer calf mortality pattern was attributed to poor development rates of calves resulting from poor nutritional condition of cows following severe winter conditions (Adams 2003). We hypothesize that the pattern of high late-summer calf mortality observed in the NAPCH in 2005 can also be attributed to poor development rates of calves resulting from poor nutritional condition of cows. While poor nutritional condition in cows can be caused by a multitude of factors including depleted range (Valkenburg et al. 2002) and environmental conditions in winter and summer (Adams 2005, Cook et al. 2004), we hypothesize that NAPCH cows may be in poor condition due to heavy parasite burdens.

To evaluate these hypotheses, FWS, ADFG, and United States Geological Survey biologists will begin a collaborative project in the fall of 2005. This project will involve the experimental decreases in parasite burdens from a sample of NAPCH cows to evaluate the influence of parasites on nutritional condition, health, pregnancy rates, and calf recruitment. This project will provide information necessary for effective NAPCH management strategies, and will be reported on in coming years.

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