Alaska Department of Fish and Game Wildlife Restoration Grant

Grant Number:	W-33	Segment Number:	9
Project Number:	18.74		
Project Title:	Wildlife Health and Disease	Surveillance in Alaska	

Appendix 1. Abstracts from meetings and conferences in chronological order

 55th Annual Meeting American Association of Veterinary Parasitologists – Atlanta, GA – 31 Jul - 2 Aug, 2010. Toxicant-Parasite Interactions: The Role of Macroparasites In Mercury Dynamics Within The Gastrointestinal Tract Of Mammalian Hosts.

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Abstract

Certain intestinal macroparasites have been shown to bioaccumulate heavy metals (e.g. Pb, Cd) at significantly higher concentrations than that of their fish hosts. We, therefore, hypothesize that toxicant-parasite interactions within the host intestine may have a positive effect on overall host-health. Thus, the objective of this ongoing study is to assess the ability of gastrointestinal macroparasites to bioaccumulate mercury, and to elucidate their role in mercury distribution and biotransformation within the host. Intestinal tracts were processed from Alaskan Gray wolves (Canis *lupus*). Macroparasites were removed and weighed, and nematodes were enumerated; additionally, host luminal contents and various tissue samples were collected for total mercury (THg) analysis. Prevalence of cestodes and ascarids in the 89 intestinal tracts examined was 61.8% (55/89) and 19.1% (17/89), respectively. Nine wolves contained both nematodes and ascarids, out of 63 parasitized animals (14.3%). Ascarids from 15 of the 17 animals were identified morphologically, and prevalence of Toxocara canis and Toxascaris leonina was found to be 33.3% (5/15) and 80.0% (12/15), respectively. Two individuals were co-infected with both *Toxocara canis* and Toxascaris leonina. All cestodes were of the genus Taenia. Preliminary THg results showed concentrations in pooled, homogenized cestodes ranging from 2.95 to

75.03 ppb (ww). Nematode THg concentrations have ranged from 3.34 to 6.32 ppb ww, possibly suggesting a greater potential for uptake by the cestodes. Initial results confirm that these parasites are capable of mercury uptake, and that THg concentrations in these parasites lie within a detectable range. These data will be of critical importance as we move forward in addressing the role of macroparasites in mercury distribution and biotransformation within the host.

• XIIth International Congress of Parasitology (ICOPA), Melbourne, Australia, August 15th to August 20th, 2010 Poster presentation: Toxicant-Parasite Interactions in Alaskan Grey Wolves: The Role Of Intestinal Macroparasites in the Uptake & Biotransformation Of Mercury (Hg)

A Linton: Department of Microbiology, Immunology, and Pathology, College of Veterinary Medicine and Biomedical Sciences, Colorado State University, Fort Collins, CO

KB Beckmen: Alaska Department of Fish & Game, Fairbanks, Alaska TM O'Hara: The Wildlife Toxicology Lab, at Department of Biology and Wildlife and Institute of Arctic Biology, University of Alaska Fairbanks, Fairbanks, Alaska M Salman: Department of Clinical Sciences, Colorado State University, College of Veterinary Medicine and Biomedical Sciences, Fort Collins, CO LR Ballweber: Department of Microbiology, Immunology, and Pathology, College of Veterinary Medicine and Biomedical Sciences, Colorado State University, Fort Collins, CO

Abstract: Certain intestinal macroparasites (e.g. cestodes, acanthocephalans) have been shown to bioaccumulate heavy metals at significantly higher concentrations than that of their fish host. Mercury is a heavy metal that is increasing in the environment, worldwide, and has the potential to become methylated and enter the foodchain. The objective of this ongoing study is to assess the ability of gastrointestinal macroparasites to bioaccumulate mercury, and to determine their role in mercury distribution and biotransformation within the host. We hypothesize that toxicant-parasite interactions within the host intestine have a positive effect on overall host-health. Macroparasites were removed and weighed, and nematodes were enumerated from the intestinal tracts of Alaska Gray wolves (*Canis lupus*). Additionally, host luminal contents and various tissue samples were collected for total mercury (THg) analysis. Prevalence of cestodes and ascarids in the 89 intestinal tracts examined was 61.8% (55/89) and 19.1% (17/89), respectively. Nine wolves contained both cestodes and ascarids, out of 63 parasitized animals (14.3%). Ascarids from 15 of the 17 animals were identified morphologically, and prevalence of *Toxocara canis* and *Toxascaris leonina* was found to be 33.3% (5/15) and 80.0% (12/15), respectively. Two individuals were co-infected with both Toxocara canis and Toxascaris leonina. All cestodes were of the genus Taenia. Preliminary THg results showed concentrations in pooled, homogenized cestodes ranging from 2.95 to 75.03 ppb (ww), with a median of 12.98 ppb (ww). Nematode THg concentrations ranged from 3.34 to 6.32 ppb ww, with a median of 4.72 ppb (ww), possibly suggesting a greater potential for uptake by the cestodes. Initial results confirm that these parasites are capable of heavy metal uptake, and that THg concentrations in these parasites lie within a detectable range. These data will be fundamental as we move forward in addressing the role of macroparasites in mercury distribution and biotransformation.

• Annual Meeting of the Alaska Chapter of The Wildlife Society, Juneau, AK, April 5-7, 2011

PREVENTION OF REMOTE DART-DELIVERY SYSTEM AND NET-GUN CAPTURE RELATED MORBIDITY AND MORTALITY

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Abstract

Use of remote dart-delivery systems to live capture free-ranging wildlife is critical for research and management objectives of wildlife biologists. However, unacceptable mortality and morbidity can be associated with explosive dart and high-powered projectors historically used by our department since its invention compared to modern systems now available. Despite higher mortality rates, net-gun capture has often been viewed as a 'better alternative' for reasons such as speed and elimination of drug residues. However, these perceived advantages are mostly erroneous when the higher risks to the animal's health, welfare and survival are considered. Recent increased documentation of mortality for animal welfare concerns has identified previously unrecognized morbidity and many potentially preventable injuries such as septicemia from explosive dart infections and muscle ruptures and rib fractures occurring net-gun restraint. A review of capture records of over 2400 animals of 11 mammal species documents a range of mortality rates from 0% up to 16.7%. Explosive dart systems used on 1846 ungulates and large carnivores had an overall mortality rate of 1.4% whereas net-gun captures of 4 species had a mortality rate of 2.3%. Non-explosive darting systems (Dan-inject) used on ~8000 captures of similar species in Scandinavia had a significantly lower mortality rate of 0.06%. Frequent post-capture monitoring with necropsy of mortalities, refinement of capture techniques including elimination of explosive darting systems and recognition of the morbidity risks/consequences associated with our capture techniques are paramount in preventing unnecessary pain and death of the wildlife we aim to study.

MASS MORTALITY EVENT OF MOOSE FROM CONSUMPTION OF TOXIC ORNAMENTALS

Kimberlee Beckmen, Stephanie Sell, Jessy Coltrane, Kathy Burek Huntington, and Merl Raisbeck

ABSTRACT

From January 2 to Feb 7 2011, three moose calves were found dead in a residential neighborhood of Anchorage. Just prior to death, apparently healthy moose were observed browsing on ornamental shrubs, staggered away and fell over dead. A carcass that was necropsied within a few hours of death had an overpowering odor of bitter almond classically associated with cyanide. Toxicologic analysis of liver, muscle, rumen content

and plants that had been browsed determined that all three calves had died from cyanide toxicosis from the consumption of *Prunus spp*. (European bird cherry, Mayday or chokecherry). In addition, the third calf had consumed an extremely toxic *Taxus sp*. (Yew) with fatal concentrations of taxine B in the rumen. These three plus an single case in 2006 in the same area are the only documented cases of cyanide toxicosis from chokecherry consumption in moose. The only other documented yew poisoning in moose is reported from Norway in 2008. Typically, freezing causes cyanogenic glycosides to accumulate in the buds of chokecherry trees. The toxin builds up immediately after a freeze, but then dissipates within days. When the buds are chewed and swallowed, they react with chemicals in the rumen to release cyanide gas which can kill in minutes. A midwinter poisoning at this latitude is extremely unexpected. Yew, an evergreen, are toxic year round do both ruminants and monogastrics. Urban sprawl, invasive *Prunus sp.*, climate change and increased awareness are expected to increase the number of cases detected.

REMOTELY-DELIVERED CHEMICAL IMMOBILIZATION OF ADULT FEMALE STELLER SEA LIONS (*EUMETOPIAS JUBATUS*) FOR PHYSIOLOGICAL SAMPLING AND SATELLITE TELEMETRY ATTACHMENT

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Understanding foraging behavior and body condition of adult female Steller sea lions (*Eumetopias jubatus*) is a critical data need, but also the most difficult to obtain due to the high risk of capture mortality. A previous study using zolazepam-tiletamine resulted in high mortality rates from drowning. During November 2010 we immobilized six adult females in Alaska using a combination of medetomidine-butorphanol-midazolam (dosage range approximately 0.038-0.044, 0.13-0.15, and 0.19-0.22 mg/kg respectively) via remotely delivered dart (DaninjectTM) at ~12-20 meters. Three animals (206–238 kg) were approached ~12 minutes after darting and were found on the haul-out at a deep plane of anesthesia. After intubation animals were supplemented with isoflurane (range 0.5-1.5%) to maintain anesthesia for \leq 130 minutes. Heart rate, respiratory rate, body temperature, oxygen saturation and blood gases were monitored. Sedation was reversed with naltrexone (30 mg) and atipamezole (45 mg) IM. Full recovered occurred in 9-12 minutes. Behavior continues to be monitored using Mk10 Argos Fast-GPS transmitters.

Three additional darted animals entered the water after darting. One hauled-out again and was monitored for 85 minutes until recovered. Two other animals were found deeply sedated but spontaneously breathing while floating in the water. The first was followed for 145 minutes. She was deeply sedated but alert enough to avoid capture. The second was darted with the reversal agent at ~63 minutes. This study resulted in successful reversible immobilization of adult animals on haul-outs. Equally important we demonstrated that animals entering the water after darting survived with the use of this combination.

MICROFILARIA PREVALENCE RATES IN MOOSE CALVES: FIRST DETECTION OF CO-INFECTIONS WITH A CRYPTIC PARASITE

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Abstract

Setaria yehi is a filarid nematode identified in the abdominal cavity of Alaskan moose (Alces alces) and caribou (Rangifer tarandus). The infective L1 microfilaria are found in the bloodstream and transmitted by a biting insect vector. Between December 2004 and March 2005, nine moose calves in the Fairbanks area (GMU20A) died with secondary bacterial fibrinous peritonitis. The lesions were associated with migrating S. yehi and microscopic evidence of thrombi and severe inflammatory reactions to the massive numbers of microfilaria. This study was undertaken to determine the prevalence and distribution of this parasite by examining blood samples from moose across various regions of Alaska. In the course of the study, a species of a cryptic nematode, Rumenfilaria sp. previously known in North America from a single case report, was detected. We found that the prevalence rates of both Setaria and Rumenfilaria were significantly lower in GMU 22C than the three other GMUs sampled (Setaria: p=0.00002; GMU19D: 80% prevalence, 20A: 70%, 20B: 100%, 22C: 0%; Rumenfilaria *sp.*: *p*=0.013; GMU19D: 70% prevalence, 20A: 40%, 20B: 50%, 22C: 0%). We also found that moose infected with *Rumenfilaria sp.* had a significantly higher probability of being infected with Setaria, but that moose infected with Setaria did not have a significantly higher probability of being infected with Rumenfilaria sp. We speculate that the absence of either parasite in GMU22C, is either due to: the low density of susceptible moose or environmental factors affecting the arthropod vector(s).

- 13th Northern Furbearer Conference in Whitehorse, Yukon, Canada, 12–14 April 2011. Management of Dog Louse Infestation of Wolves in Interior Alaska. Craig Gardner, Kimberlee Beckmen, Patty Delvecchio, Nate Pamparin. Presented by Craig Gardner, Abstract not available
- Joint Annual Conferences of the American Association Of Zoo Veterinarians and American Association of Wildlife Veterinarians, South Padre Island, Texas, October 23-29, 2010.

CERULOPLASMIN AND COPPER STATUS IN FREE-RANGING ALASKAN CARIBOU (Rangifer tarandus tarandus)

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Abstract

The copper status of caribou (*Rangifer tarandus*) in Alaska was evaluated by liver copper concentration [liver Cu] (*n*=70), serum copper concentration [serum Cu]

(n=341), and the concentration of ceruloplasmin (n=31), a copper carrying protein found in the serum. We compared these results with respect to age class, gender, season, herd, region, and pregnancy status. Age class was defined as: fetus, neonate (<1month), calf (>1 month & <10 months), or adult (>10 months). We categorized two seasons in our analysis: 1) a combination of winter and spring samples, and 2) a combination of summer and fall samples. Two arctic (Western Arctic and Teshekpuk) and two southern herds (Northern Alaska Peninsula and Mulchatna) were pooled to establish the northern and southern regions, respectively. An Analysis of Variance procedure was used to test for differences between covariates, where $\alpha =$ 0.05 (SAS, PROC ANOVA). No serum samples from neonates or pregnant females, or liver samples from calves, were available so we were unable to test for differences within all categories of each variable. Age class was significant for both [serum Cu] (p = 0.0001) and [liver Cu] (p = 0.0001). [Serum Cu] from neonates (mean = 0.959, SE = 0.145) were significantly greater than calves (mean = 0.580, SE = 0.036) and adults (mean =0.544, SE = 0.01). [Liver Cu] from fetuses (mean = 224.2, SE = 78.6) and neonates (mean = 123.7, SE = 12.3) were significantly greater than adults (mean = 16.9, SE = 12.4). This was not unexpected since in domestic ungulates, fetal and neonatal liver storage is high to meet the Cu requirements prior to weaning. Herd was significant for [serum Cu] (p = 0.0001). [Serum Cu] from Western Arctic (mean = 0.692, SE= 0.031) were significantly greater than other herds. [Serum Cu] from Teshekpuk (mean= 0.576, SE = 0.018) were significantly greater than Northern Alaska Peninsula (mean = 0.515, SE = 0.013). Seasonal differences of [serum Cu] within a herd were only significant within the Northern Alaska Peninsula herd (p < p0.0001). [Serum Cu] winter/spring (mean = 0.445, SE = 0.014) was significantly less than summer/fall (mean = 0.578, SE = 0.018). This is consistent with this herd's poor health status and heavy parasite loads. These results lead to additional studies to elucidate the significance of the nutritional – parasite interaction on caribou herd health. A significant difference was found between genders for [serum Cu], particularly during the summer/fall season (p = 0.0014). [Serum Cu] for males (mean = 0.648, SE = 0.016) were significantly greater than females (mean = 0.577, SE =0.041). While region wasn't statistically significant, the northern herds had a lower mean [liver Cu] (mean = 47.4, SE = 13.3) than the southern herds (mean = 64.9, SE= 10.9), despite all the high fetal liver copper included in the northern herds. Nonpregnant adults (mean = 18.4, SE = 2.0) and pregnant female (mean = 8.2, SE = 1.7) were significantly different for [liver Cu] (p < 0.021). Our observation of high fetal [liver Cu] and low pregnant female [liver Cu] supports the conclusion that there is mobilization of copper stores from the cow to the fetus. In addition to these analyses, we also examined the correlations between [serum Cu], ceruloplasmin, and [liver Cu] (SAS, PROC CORR, Pearson). No correlation was detected among any combination of copper measurement, likely limited by the small sample size. Thus, until additional paired serum and liver samples can be evaluated, neither ceruloplasmin or [serum Cu] can be substituted as a direct measure of copper status in live-captured caribou. We were limited by opportunistic sampling but in the future, evaluation of more representative samples with respect to age class, gender, season, and herd, will allow us to better evaluate the variation in copper status and its implications for herd health in caribou.

STRESS LEVELS DURING CHEMICAL VS. MANUAL RESTRAINT IN WOOD BISON (*Bison bison athabascae*) AS INDICATED BY BLOOD LACTATE AND GLUCOSE

Stephanie G. Crawford, BS¹ and Kimberlee B. Beckmen, MS, DVM, PhD¹* ¹Alaska Department of Fish and Game, Fairbanks, AK 99701 USA Abstract

Blood lactate (mmol/L) and glucose (mg/dL) concentrations were measured in captive wood bison (*Bison bison athabascae*) restrained for disease testing and health, pending reintroduction into Alaska. The majority of the herd (n = 70) were restrained in a hydraulic squeeze chute, though the larger, more aggressive animals (n = 9) were chemically immobilized via darting using a combination of thiafentanil and xylazine. A General Linear Model was used to test for differences in blood [lactate] and [glucose] among eight categorical and three continuous variables related to demographics, restraint technique, handling times, and factors assumed to cause additive stress, where α = 0.05 (SAS, PROC GLM). Additive stressors were documented during the handling and include things such as the use of an electric prod, an injury occurring during handling, and observed aggressive conspecific interactions. We also tested for a correlation between [lactate] and [glucose] (SAS, PROC CORR, Pearson).

Consistent with our expectations, a highly significant difference (p < 0.0001) was found in [lactate] among the three categories of restraint techniques; this relationship was not demonstrated for [glucose]. Darted animals exhibited the lowest [lactate] (\bar{x} = 0.9, SE = 0.15), while animals handled through the chute system had the highest [lactate] ($\bar{x} = 11.7$, SE = 0.53) and bison unsuccessfully moved through the chute system and subsequently darted fell in between ($\bar{x} = 4.2$, SE = 0.90). No correlation was observed between [lactate] and [glucose] (R = 0.075). For chute-handled bison, no significant differences were found for [lactate] or [glucose] with respect to the day of handling, gender, age, order through the chutes, or additive stressors. Animals with shorter handling times were shown to have significantly higher [lactate] (p = 0.0347). Restraint time, the amount of time in the hydraulic squeeze, did not affect [lactate]. However, animals with shorter restraint times did have significantly higher [glucose] (p = 0.0293), such that each minute increase in handling time resulted in a $\sim 1\%$ decrease in the median [glucose]. The total handling time did not have a significant affect on [glucose]. While we expected that chemical restraint would be less stressful to the bison than other techniques, we did not anticipate that shorter handling times would yield higher [lactate]. We propose that wood bison, as herd animals, likely recover quickly from individual stressful events. However, swift processing through a chute system does not allow time for this lactate recovery, as there is little to no rest period between segments of the chute system. In brief handlings (< 18 minutes), the point in time where we collected our blood sample allowed us to actually measure cumulative stress, whereas in the longer handlings (> 38 minutes) our blood sample was more representative of an acute response to the most recent stressor(s).