BLOOD PROFILES AND ASSOCIATED BIRTH CHARACTERISTICS OF FREE-RANGING MOOSE (*ALCES ALCES*) NEONATES IN A DECLINING POPULATION IN NORTHEASTERN MINNESOTA

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ABSTRACT: Sources of natural variability of blood analytes related to physiological development pose both challenges and opportunities to deriving and interpreting the most useful nutritional and health-related information from blood profiles of free-ranging animals. Preliminary evidence suggests accurate interpretation of blood profiles may be particularly important relative to newborns given their high probability of death. Our goal was to establish hematological and serum reference values for free-ranging moose (*Alces alces*) neonates. Sixteen neonates (8 females, 8 males) were captured and blood was sampled during 8–12 May 2013. Mean age was 2.9 days old (range = 1.4–6.0); mean body mass and hind foot length were 16.8 kg (13.8–20.5) and 46.8 cm (45.0–49.0). We present mean, 95% confidence interval and range of values for 15 hematological and 24 serum characteristics, including metabolites, chemistries, electrolytes, enzymes, and metabolic and stress hormones. We observed significant (r^2 = 0.423–0.747, $P \leq 0.016$) positive relationships between body mass and red blood cell and white blood cell counts, hemoglobin, and packed cell volume. Hind foot length was positively related (r^2 = 0.369, P = 0.028) only to red blood cell counts. No serum constituents were affected by body size metrics, but sex influenced ($P \leq 0.052$) several whole blood and serum characteristics. At the individual level, blood profiles facilitated discrimination of one individual neonate in poor nutritional condition that was not evident in the original physical examination at capture. As wildlife researchers and veterinarians increasingly assess the nutritional and health status of free-ranging moose and other species by clinical biochemistry and laboratory methods, cumulative banks of blood reference values will aid in data interpretation.

ALCES VOL. 52: 85–99 (2016)

Key words: *Alces alces*, blood profiles, blood reference values, hematology, moose neonates, serum profiles

The effective use of blood profiles to assess the health of domestic animals and their metabolic, nutritional, and reproductive status has relied on a long history of biochemical and physiological research and the establishment of reference values of quantifiable constituents (Davidsohn and Henry 1969, Cole 1980, Benjamin 1981, Swenson 1984, Kaneko 1989). This work supports similar, more recent efforts of wildlife researchers and veterinarians investigating the influence of intrinsic and extrinsic factors on hematological and serum characteristics in captive and free-ranging wild animals, most commonly for adult cervids (Kitchen and Pritchard 1962, Johnson et al. 1968, Seal and Erickson 1969, Thurley and McNatty 1973, White and Cook 1974, Seal et al. 1981, DelGiudice et al.

1990a, 1990b, 1990c, 1994); however, blood reference values for juveniles are limited for most Cervidae species (Tumbleson et al. 1970, Franzmann and LeResche 1978, Rawson et al. 1992, Kunkel and Mech 1994, Sams et al. 1995, 1996, Carstensen Powell and DelGiudice 2005, Rostal et al. 2012).

Sources of natural variability of measured blood analytes related to physiological development must be understood to best distinguish that variability from the influence of altered nutrition or health issues on blood profiles of free-ranging animals. This understanding is important beginning with rapidly developing newborns. For example, quantifiable relationships between age and rapid growth, increasing energy requirements, and physiological development reflected by changes in hematology have been reported for white-tailed deer from birth to 90 days of age (Rawson et al. 1992). Additionally, chronic maternal nutritional restriction during gestation may subsequently compromise immunocompetence of neonates, affect hematological and serum profiles, and predispose young to mortality by various agents (Sams et al. 1995, 1996). The probability of mortality is greatest for northern free-ranging ungulates within the first weeks of life (DelGiudice et al. 2006, Carstensen et al. 2009, Lenarz et al. 2010, Keech et al. 2011, Severud et al. 2015a, 2015b).

With the assistance of global positioning system (GPS) collar technology, we recently had the opportunity to capture and handle 49 free-ranging moose (*Alces alces*) neonates (≤ 6 days old) in northeastern Minnesota to study survival and cause-specific mortality (Severud et al. 2015a). The long-term persistence of this population is in jeopardy; moose numbers have declined an estimated 55% from 8,840 to 4,020 from 2006 to 2016 (DelGiudice 2016). However, pregnancy rates indicate that fertility is comparable to moose across North America (84%, Boer 1992, Severud and DelGiudice, unpublished data). Our goal in sampling blood from a portion of these neonates was to establish reference values and increase our understanding of the potential of hematology and serum profiles in assessing the nutritional status and overall health of newborns. Specifically, our objectives were to quantify the relationship and potential influence of 1) age at capture, 2) metrics of body size (i.e., mass, hind foot length), and 3) sex of moose neonates on values of hematological and serum characteristics, and 4) to highlight any particularly informative findings (i.e., values) at the individual level.

STUDY AREA

We captured calves within a 6,068-km^2 study area located between 47° 06'N and 47° 58'N latitude and 90° 04'W and 92° 17'W longitude in northeastern Minnesota. This region is described as the Northern Superior Upland (Minnesota Department of Natural Resources [MNDNR] 2015) and includes bogs, swamps, lakes, and streams, with lowland stands of northern white cedar (*Thuja occidentalis*), black spruce (*Picea mariana*), and tamarack (*Larix laricina*), and upland balsam fir (*Abies balsamea*), jack pine (*Pinus banksiana*), white pine (*P. strobus*), and red pine (*P. resinosa*). Conifers are frequently intermixed with trembling aspen (*Populus tremuloides*) and white birch (*Betula papyrifera*).

Wolves (*Canis lupus*) and American black bears (*Ursus americanus*) are predators of moose (Fritts and Mech 1981, Patterson et al. 2013, Severud et al. 2015a) with recent densities estimated at 3.4 wolves and 23 bears/100 km^2 (Erb and Sampson 2013, Garshelis and Noyce 2015). White-tailed deer (*Odocoileus virginianus*) are managed at pre-fawning densities of < 4 deer/km^2, and are primary prey of wolves in most of northern Minnesota (Nelson and Mech 1986, Kunkel and Mech 1994, DelGiudice et al. 2006,

Carstensen et al. 2009, Grund 2014). Maximum daily temperatures have been generally increasing since at least 1960 (Lenarz et al. 2010). Mean daily minimum and maximum temperatures ranged from −5.2 °C to 13.3 °C and 3.3 °C to 24.6 °C, respectively, during April to July 2013 at Ely, Minnesota (Midwestern Regional Climate Center 2015).

METHODS

Neonate Capture and Handling

We began monitoring 73 GPS-collared cows on 1 May and captured neonates during 8–17 May 2013; cow collars were programmed to obtain hourly fixes in May and to transmit 4 times daily (Severud et al. 2015a). We used 3 different and complementary approaches for computer-monitoring the hourly locations and movements of dams and their GPS-collared neonates: a base station computer, a web-mapping service, and automated reports (Severud et al. 2015a). Our primary monitoring objective was to record when and where individual pregnant females increased activity reflected by a "calving movement," a variable atypical, long distance move that ends with localization for 1–15 days (Bogomolova et al. 1992, Poole et al. 2007, DeMars et al. 2013, Severud et al. 2015a). We assumed that once a female localized, the birthing process had begun, and birth occurred within 12 h (Hydbring et al. 1999, Bogomolova et al. 1992, Asher et al. 2014). We then allowed an additional 24 h for dam-calf bonding, whereupon calves were designated "eligible" for capture. Actual bonding time was calculated as that 24 h plus the elapsed time prior to capture, which depended on the daily schedule and logistical constraints. Additional details of our computer-monitoring approaches are provided in Severud et al. (2015a).

The capture team (Quicksilver Air, Inc., Fairbanks, Alaska, USA) located designated (eligible) dams from their most recent GPS coordinates, and captured and collared calves as time and conditions allowed on a daily basis. They located the target dam from the air and then landed some distance (≥100 m) away to allow handlers to approach the calves on foot. Neonates were not netted or chemically immobilized; handlers could simply walk up to them with most moving <10 m from where first observed and subsequently captured and handled (DelGiudice et al. 2015); twins were captured, handled, and released together. The handling protocol included fitting a 420 g-GPS collar (GPS PLUS VERTEX Survey-1 GLOBALSTAR with expandable belt, Vectronic Aerospace GmbH, Berlin, Germany); fixing ear tags; collecting 25 mL of blood by syringe from the jugular vein into a 5-mL ethylenediamine tetraacetic acid (EDTA) tube for hematology and into two 10-mL serum tubes for chemistry and hormone assays; measuring body mass (BM, ± 0.5 kg) by spring-scale, hind foot length (± 1 cm; HFL), and rectal temperature (± 0.1 °F) by digital thermometer; and a physical examination to record injuries or abnormalities. Blood samples were stored on ice and allowed to clot for 1.7 ± 0.2 h before separation by a portable centrifuge.

Overall handling time averaged 12.9 minutes (± 1.14 [SE], range = 7–18 min, n = 9). By 12 May 2013, 2 of 11 (18.2%) dams had abandoned 2 of 17 (11.8%) neonates in apparent response to capture and handling (DelGiudice et al. 2015). Consequently, we removed blood-sampling, presumably the most invasive handling technique, from our protocol. Ultimately, despite excluding blood-sampling and other components of the protocol, capture-induced abandonment continued intermittently, during 13–17 May 2013. Our analyses indicated that abandonments reflected more of a disturbance to the dams than the neonates (DelGiudice et al. 2015). All captures and handling protocols adhered to requirements of the Institutional Animal Care and Use

Committee for the University of Minnesota (Protocol 1302-30328A) and followed guidelines of the American Society of Mammalogists (Sikes et al. 2011).

Laboratory and Statistical Analyses

Hematological analyses were performed using an Advia 2120 Hematology analyzer (Siemens Healthcare Diagnostics, Inc., Tarrytown, New York, USA) at the University of Minnesota's Clinical Pathology Laboratory (St. Paul, Minnesota, USA). Additionally, a peripheral blood film evaluation was conducted to determine a manual 5-part leukocyte differential and to assess cell morphology. Total plasma protein (TPP) and fibrinogen concentrations were determined by refractometry and by the heat precipitation method (Stockham and Scott 2008: 369–413). Serum biochemical profiles were determined on an AU480 chemistry analyzer (Beckman Coulter, Inc., Brea, California, USA). Serum total thyroxine (TT_4, Clinical Assays™ M Total T_4 ^{125}I RIA Kit), free thyroxine (FT_4, GammaCoat™ Free T_4 [Two-Step] ^{125}I, RIA Kit), and free triiodothyronine (FT_3, Clinical Assays™ GammaCoat™ Free T_3 ^{125}I RIA Kit) were assayed with kits from DiaSorin Inc. (Stillwater, Minnesota, USA) at the Diagnostic Center for Population and Animal Health (DCPAH, Michigan State University, East Lansing, Michigan, USA). Serum total triiodothyronine (TT_3) concentrations were determined at DCPAH; the assay procedure and a subsequent modification are described by Refsal et al. (1984) and Panciera et al. (1990), respectively. DCPAH also conducted serum cortisol assays (Coat-a-Count Cortisol radioimmunoassay, Siemens Medical Solutions Diagnostics, Los Angeles, California, USA).

We examined potential relationships between physical development (BM, HFL) and values of hematological and serum characteristics with linear regression (Ott 1984: 245–250, Microsoft Excel 2010). We examined the influence of sex on physical and blood characteristics of neonates with *t*-tests assuming equal variances (Ott 1984: 140–142, Microsoft Excel 2010). We report data as means and 95% confidence intervals (CI) or ± SE.

RESULTS

We collected blood from 16 moose neonates; EDTA tubes for hematology from 13 (6 males, 7 females) and serum tubes to examine chemistries, metabolites, electrolytes, and hormones from all 16 (8 males, 8 females). Mean age was the same for both groups (Tables 1 and 2). There was no significant ($P \geq 0.401$) relationship between age at capture and BM or HFL; however, HFL was related ($r^2 = 0.530$, $P = 0.001$) to BM (Fig. 1). We also observed significant ($r^2 = 0.423$–0.747, $P \leq 0.016$) positive relationships between BM and red blood cell (RBC) and white blood cell (WBC) counts, hemoglobin (Hgb), and packed cell volume (PCV, Fig. 2). HFL was positively related ($r^2 = 0.369$, $P = 0.028$) to RBC counts (Fig. 3), but only marginally to WBC counts ($r^2 = 0.248$, $P = 0.083$) and Hgb ($r^2 = 0.245$, $P = 0.085$). Neither BM nor HFL was related to mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), platelets, differential WBCs, TPP, or fibrinogen (Table 1).

There were no significant relationships between BM or HFL at capture and any serum characteristics presented in Table 2. However, age at capture was positively related to FT_3 ($r^2 = 0.260$, $P = 0.044$) and FT_4 ($r^2 = 0.381$, $P = 0.011$, Fig. 4). Blood characteristics that differed by sex included WBC ($t_{11} = 2.20$, $P = 0.040$), monocytes ($t_{11} = 2.20$, $P < 0.001$), fibrinogen ($t_{11} = 2.20$, $P = 0.052$), and serum calcium (Ca, $t_{14} = 2.14$, $P = 0.024$), total protein (TP, $t_{14} = 2.14$, $P = 0.021$), globulin ($t_{14} =$

Table 1. Mean, 95% confidence interval (CI), coefficient of variation, and range of age, physical characteristics, and values of hematological characteristics of moose neonates (n = 13) at capture, northeastern Minnesota, 8–12 May 2013.[a]

Characteristic[b]	Mean	95% CI	Coefficient of variation	Range
Age at capture (days)	2.9	2.1–3.7	0.52	1.4–6.0
Body mass (kg)	16.8	15.5–18.1	0.14	13.8–20.5
Hind foot length (cm)	46.8	46.0–47.5	0.03	45.0–49.0
RBC (10^6/μL)	6.1	5.7–6.4	0.10	5.1–6.8
WBC (10^3/μL)	5.4	4.7–6.2	0.25	3.9–7.8
Neutrophils segs (% [10^3/μL])	74.4	69.4–79.2	0.12	52.1–84.1
Lymphocytes (% [10^3/μL])	20.0	15.5–24.6	0.42	6.9–36.0
Monocytes (% [10^3/μL])	3.4	1.9–5.0	0.85	0.0–9.1
Eosinophils (% [10^3/μL])	2.0	1.0–2.9	0.89	0.0–5.9
Basophils (% [10^3/μL])	0.1	-0.05–0.34	2.45	0.0–1.0
Hgb (g/dL)	9.6	9.0–10.3	0.12	8.2–11.8
PCV (%)	31.4	29.3–33.5	0.12	25.7–38.9
MCV (fL)	51.8	49.5–54.1	0.08	45.9–57.2
MCH (pg)	15.9	15.2–16.5	0.08	14.1–17.6
MCHC (g/dL)	30.7	30.2–31.2	0.03	28.7–31.8
Platelets (10^3/μL)	550	436–665	0.38	357–1,065
TPP (g/dL)	5.8	5.5–6.2	0.11	4.8–7.5
Fibrinogen (g/dL)	0.5	0.39–0.53	0.27	0.3–0.7

[a]Handlers were able to approach and handle neonates with minimal excitement, no nets or chemicals (Severud et al. 2015a).
[b]Characteristics include RBC = red blood cells, WBC = white blood cells, Hgb = hemoglobin, PCV = packed cell volume, MCV = mean corpuscular volume, MCH = mean corpuscular hemoglobin, MCHC = mean corpuscular hemoglobin concentration, and TPP = total plasma protein.

2.14, $P = 0.011$), alkaline phosphatase (ALP, $t_{14} = 2.14$, $P = 0.010$), and gamma-glutamyl transferase (GGT, $t_{14} = 2.14$, $P = 0.002$; Table 3).

DISCUSSION

A total of 49 moose neonates of 31 dams were captured and handled during 8–17 May 2013 (Severud et al. 2015a). Based on physical examination, measurements and their behavior, all appeared to be calm and in good overall health. Mean body size of the 16 blood-sampled neonates was comparable to that of neonates of the typically larger tundra moose (*A. americanus gigas*) in south-central and western Interior Alaska (Ballard et al. 1996, Keech et al. 2011). The range of neonatal body size captured during our 5-day operation indicated that skeletal development accounted for just over half of the variability in BM, suggesting the remaining 50% was associated with water, protein, and fat (i.e., brown fat) content (Schoonderwoerd et al. 1986, DelGiudice et al. 1990c, Watkins et al. 1991). A similar association ($r^2 = 0.85$) was observed for free-ranging deer neonates (Carstensen Powell and DelGiudice 2005). This relationship has value for interpreting blood profiles, particularly hematology, given our findings (Figs. 2 and 3). Rawson et al. (1992) showed that increases in age, BM, and resting metabolic rate contributed to a concomitant steady

Table 2. Mean, 95% confidence interval (CI), coefficient of variation, and range of age, physical characteristics, and values of serum constituents of moose neonates (n = 16) at capture, northeastern Minnesota, 8–12 May 2013.[a]

Characteristic[b]	Mean	95% CI	Coefficient of variation	Range
Age at capture (days)	2.9	2.2–3.5	0.47	1.4–6.0
Body mass (kg)	16.8	15.8–17.9	0.13	13.8–20.5
Hind foot length (cm)	46.9	46.3–47.5	0.03	45.0–49.0
Ca (mg/dL)	10.1	9.8–10.5	0.07	8.8–11.4
P (mg/dL)	8.9	8.3–9.5	0.13	5.9–11.0
Na (mEq/L)	141	139–142	0.02	137–150
K (mEq/L)	5.1	4.8–5.3	0.08	4.4–5.9
Cl (mEq/L)	95.3	94–96	0.02	92–100
Mg (mg/dL)	1.7	1.7–1.8	0.08	1.5–2.0
Bicarbonate (mEq/L)	22.3	20.3–24.3	0.19	15.2–32.6
SUN (mg/dL)	17.9	14.1–21.8	0.44	10.0–44.0
Creatinine (mg/dL)	0.7	0.6–0.8	0.27	0.5–1.2
Total bilirubin (mg/dL)	0.4	0.34–0.44	0.25	0.3–0.7
Glucose (mg/dL)	108	90.5–124.7	0.32	8.0–145
SUN:C (mg:mg)	24.2	20.8–27.6	0.29	14.0–36.0
Albumin (g/dL)	2.1	2.0–2.3	0.12	1.7–2.6
TP (g/dL)	4.2	3.9–4.5	0.16	3.3–5.9
Globulin (g/dL)	2.1	1.7–2.4	0.32	1.0–3.7
TT_4 (μg/dL)	7.5	6.5–8.5	0.26	5.0–12.1
Free T_4 (ng/dL)	1.5	1.3–1.7	0.24	0.9–2.1
TT_3 (ng/dL)	272	251–293	0.16	195–352
Free T_3 (pg/ml)	6.9	5.4–8.3	0.42	3.5–13.8
Cortisol (μg/dL)	6.2	1.9–10.5	1.41	1.5–38.5
ALP (U/L)	294	252–335	0.29	170–436
GGT (U/L)	52.0	39.6–64.4	0.49	21.0–101
AST (U/L)	63.0	53.7–72.3	0.30	42.0–110
CK (U/L)	135	91–178	0.66	52.0–399

[a]Handlers were able to approach and handle neonates with minimal excitement, no nets or chemicals (Severud et al. 2015a).

[b]Characteristics include SUN = serum urea nitrogen, SUN:C = serum urea nitrogen:creatinine, TP = total protein, Ca = calcium, P = phosphorous, Na = sodium, K = potassium, Cl = chloride, Mg = magnesium, ALP = alkaline phosphatase, GGT = gamma-glutamyl transferase, AST = aspartate aminotransferase, CK = creatine kinase, TT_4 = total thyroxine, Free T_4 = free thyroxine, TT_3 = total triiodothyronine, and Free T_3 = free triiodothyronine.

increase in the total daily metabolic energy requirement for developing deer fawns. The positive relationships between BM and RBC, PCV, and Hgb, also observed in deer fawns, may maximize the O_2-carrying capacity of blood of growing juveniles to meet their increasing O_2 requirements, and might have positive implications for escaping predators (Rawson et al. 1992).

Hematologic characteristics may reflect changes in nutrition, hydration, and physical exertion, and frequently respond to

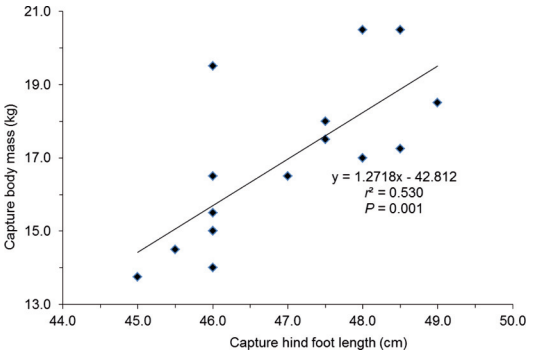

Fig. 1. Relationship of hind foot length (cm) to body mass (kg) at capture of moose neonates (1.4–6.0 days old), northeastern Minnesota, 8–12 May 2013.

infectious disease (Seal et al. 1978, Coles 1980, Benjamin 1981, DelGiudice et al. 1990b, Sams et al. 1995). Consequently, though hematological findings have inherent value in assessing the overall well-being of neonates, interpretation of findings is most accurate when the influence of physical and natural physiological development are understood. Unfortunately, established reference values for blood constituents of free-ranging, North American ungulate neonates are rare (Kunkel and Mech 1994, Ballard et al. 1996, Carstensen Powell and DelGiudice 2005). The earliest published study of blood profiles of free-ranging moose that included juveniles (newborns to 5 months old pooled), and attempted to assess condition of the group, did not account for changes in hematology associated with natural physiological development and growth (Franzmann and LeResche 1978), which potentially confounds interpretation of condition assessments. Not surprisingly, mean values of PCV and Hgb from that study are more reflective of the physiological development that occurs by 3 months of age (Rawson et al. 1992), 36 and 78% higher, respectively, than those observed in our moose neonates (Table 1). These elevated values may also reflect greater capture and handling stress in the older calves (Franzmann and LeResche 1978, Seal et al. 1981). Ballard et al. (1996) reported

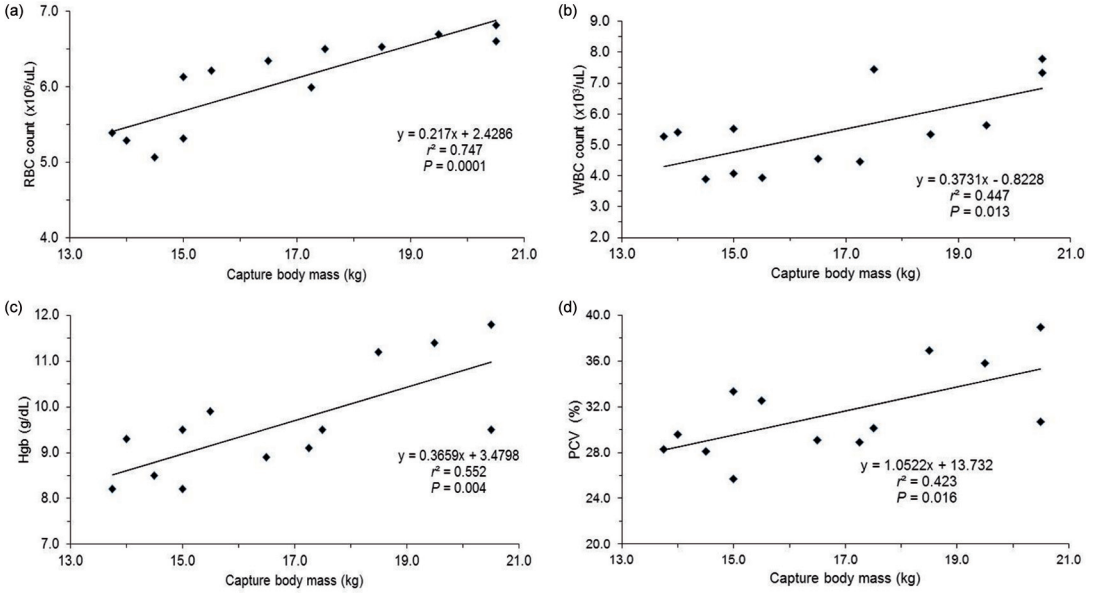

Fig. 2. Relationships (top to bottom) of body mass (kg) at capture to red blood cell (RBC) and white blood cell (WBC) counts, hemoglobin (Hgb) concentrations, and packed cell volume (PCV) of moose neonates (1.4–6.0 days old), northeastern Minnesota, 8–12 May 2013.

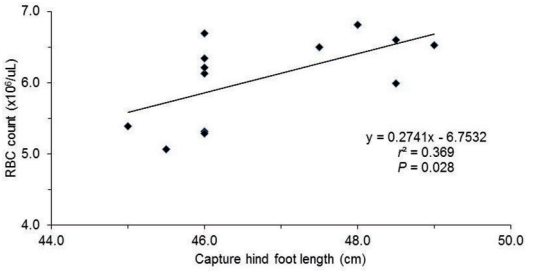

Fig. 3. Relationship of hind foot length (cm) to red blood cell (RBC) counts at capture of moose neonates (1.4–6.0 days old), northeastern Minnesota, 8–12 May 2013.

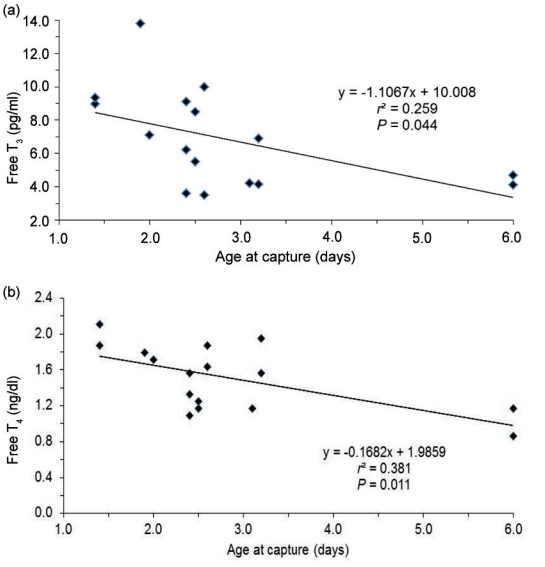

Fig. 4. Relationships of age at capture to serum free triiodothyronine (T_3, top) and free thyroxine (T_4, bottom) of moose neonates (1.4–6.0 days old), northeastern Minnesota, 8–12 May 2013.

reference values for blood characteristics of free-ranging moose neonates, but the hematological profiles were limited to Hgb and PCV. Hematological profiles of similarly aged, free-ranging white-tailed deer neonates were generally comparable to those of our much larger moose neonates (Carstensen Powell and DelGiudice 2005), emphasizing the importance of physiological development associated with BM within species, not related to size alone. Mean WBC counts of our free-ranging moose neonates fall within the range of variability of leukocyte counts of free-ranging and captive white-tailed deer neonates, and young calves of domestic cattle (Benjamin 1981: 77, Rawson et al. 1992, Sams et al. 1995, 1996, Carstensen Powell and DelGiudice 2005). Differences between values of differential leukocyte counts of free-ranging moose neonates (Table 1) and older calves in Norway (Rostal et al. 2012) are difficult to interpret due to the potential for physiologic leukocytosis from capture pursuit and chemical immobilization of the older calves.

The absence of relationships between body size (i.e., BM or HFL) of neonates and values of serum constituents simplifies data interpretation relative to assessments of health and nutritional status. As expected in generally healthy newborns, the variability of homeostatically-maintained serum electrolytes (cations and anions) was notably more limited (mean coefficient of variation [cv] = 8.6 ± 2.22%) than for proteins (20.4 ± 6.3%), metabolites (32.0 ± 4.2%), enzymes (43.5 ± 8.7%), and metabolic and stress hormones (49.7 ± 23.0%). We included these other serum analytes in the neonate serum profile primarily for their diagnostic value in assessing nutritional intake (e.g., crude protein, digestible energy) and status (i.e., condition), hydration, responses to excitement or exertion, or a combination of these measures (Benjamin 1981, Seal et al. 1981, Warren et al. 1982, Kaneko 1989, DelGiudice et al. 1990b, 1990c, 1994, Watkins et al. 1991). Whereas the values of some of these constituents exhibit wide ranges (e.g., SUN, glucose, enzymes, thyroid hormones), most of them exhibit relatively narrow CIs (Table 2). Specifically, characteristics such as SUN, glucose, TP, albumin, and thyroid hormones (e.g., TT_4 and TT_3) may respond (e.g., increasing,

Table 3. Mean, 95% confidence interval (CI), and range of age, physical characteristics, and values of blood constituents of moose neonates at capture that differ between males and females, northeastern Minnesota, 8–12 May 2013.[a]

	Males				Females			
Characteristic[b]	n	Mean	95% CI	Range	n	Mean	95% CI	Range
Body mass (kg)	8	16.9	15.2–18.7	14.0–20.5	8	16.8	5.4–18.1	13.8–19.5
HFL (cm)	8	47.1	46.3–47.8	45.5–48.5	8	46.8	45.8–47.7	45.0–49.0
WBC ($x10^3/\mu L$)	6	6.2	5.0–7.5	3.9–7.8	7	4.8	4.3–5.2	3.9–5.6
Monocytes (% [$x10^3/\mu L$])	6	4.8	2.5–7.1	0.9–9.1	7	2.3	0.34–4.19	0.0–7.9
Fibrinogen (g/dL)	6	0.53	0.45–0.62	0.4–0.7	7	0.4	0.31–0.49	0.3–0.6
Ca (mg/dL)	8	9.7	9.3–10.2	8.8–11.1	8	10.5	10.1–10.9	9.8–11.4
TP (g/dL)	8	3.8	3.6–4.0	3.3–4.1	8	4.6	4.0–5.1	3.7–5.9
Globulin (g/dL)	8	1.64	1.37–1.90	1.0–2.1	8	2.5	1.98–2.95	1.3–3.7
Bicarbonate (mEq/L)	8	24.4	21.7–27.1	20.4–32.6	8	20.3	17.9–22.6	15.2–24.0
ALP (U/L)	8	243	204–282	170–350	8	345	290–399	211–436
GGT (U/L)	8	34.8	26.9–42.6	21–51	8	69.3	52.8–85.7	35–101

[a]Handlers were able to approach and handle neonates with minimal excitement, no nets or chemicals (Severud et al. 2015a).
[b]Characteristics include HFL = hind foot length, WBC = white blood cells, Ca = calcium, TP = total protein, ALP = alkaline phosphatase, and GGT = gamma-glutamyl transferase.

decreasing) readily but somewhat moderately to changes in recent nutritional intake, but more extreme values may reflect dehydration, low energy status, or poor condition (e.g., accelerated net catabolism of endogenous protein) relative to nutritional restriction (Bahnak et al. 1981, Benjamin 1981, Warren et al. 1982, DelGiudice et al. 1990c, 1994, Watkins et al. 1991).

At the study-cohort level, concentrations of most serum constituents (Table 2) were similar to values reported for captive and free-ranging white-tailed deer neonates (Sams et al. 1995, Carstensen Powell and DelGiudice 2005), as well as elsewhere for free-ranging moose neonates and various domestic livestock (Benjamin 1981, Kaneko 1989: 886–891, Ballard et al. 1996). However, serum Ca and phosphorous (P) concentrations tended to be higher than in adult white-tailed and desert mule deer (*Odocoileus hemionus*) (DelGiudice et al. 1990b, 1990c), and highly variable cortisol values, on average, were higher than in most domestic livestock species and adult white-tailed deer, but were similar to values of white-tailed deer neonates (Kaneko 1989: 886–891, DelGiudice et al. 1990c, Sams et al. 1995, Carstensen Powell and DelGiudice 2005). Elevated Ca and P concentrations, associated with increased ALP values, are normal in young animals and reflective of the increased osteoblastic activity of early skeletal development and postprandial effects of periodic nursing (Jacobson and McGilliard 1984). The variable cortisol concentrations are suggestive of varied excitement levels associated with handling, but also may be influenced by reduced energy intake and time since nursing (Thurley and McNatty 1973, Seal et al. 1981, DelGiudice et al. 1990b). Serum creatinine concentrations are directly related to muscle mass, and along with albumin, TP, and globulin, are typically lower in the young of domestic livestock and

wild deer species (summarized by Benjamin 1981: 111–112, Finco 1989, DelGiudice et al. 1990c). Among the serum enzymes, ALP, GGT, and CK in particular were higher than in several domestic livestock species (Kaneko 1989: 886–891). Concentrations of serum enzymes can be highly variable. Because these enzymes are synthesized intracellularly, relative to their primary source(s), moderate to extreme elevations in serum may be indicative of physiological functions or events ranging from normal bone turnover (e.g., ALP), particularly in juveniles, to cellular damage associated with the heart and skeletal muscle (e.g., CK), liver, kidneys, and other organs from a variety of pathologies or extreme physical exertion (Zimmerman and Henry 1969, Benjamin 1981: 229– 232, Seal et al. 1981, Kramer 1989).

Our ability to observe and recognize most of the "… circumstantial, behavioral, and physical conditions …" influencing the vulnerability of prey species relative to the forces of nature is limited (Mech 1970: 247–248). The unique value of blood profiles at the *individual level* is demonstrated clearly in our assessment of the nutritional and health status of neonate number 520. This 2.6-day old, GPS-collared neonate died within 4.5 h of capture and release, and was one of the largest calves at 20.5 kg (HFL = 48.0 cm, Table 1). It appeared healthy and calm during handling, and the apparently attentive dam remained nearby during capture and handling, when the team departed, and when they returned to recover the dead calf. Initially, the mortality was classified as "capture-related." However, a detailed necropsy at the University of Minnesota's Veterinary Diagnostic Laboratory, including extensive macroscopic and microscopic examinations, was inconclusive, except for an empty abomasum (devoid of curdled milk), suggestive of hypoglycemia. Subsequent analyses of the neonate's blood samples generated a hematological profile with the highest RBC, Hgb, and PCV values, and the lowest TPP of the sampled cohort (Table 1), indicative of hemoconcentration and dehydration concomitant with prolonged nutritional deprivation (Coles 1980: 116– 117, Benjamin 1981: 73, 146–147, DelGiudice et al. 1994). This neonate also exhibited the cohort's highest SUN, creatinine, and SUN:creatinine values, indicative of accelerated net catabolism of endogenous protein, and the cohort's lowest Ca, TP, globulin, glucose, and FT_3 concentrations, reflecting nutritional deprivation and condition deterioration (Table 2; Coles 1980: 246, Benjamin 1981: 175, 178, DelGiudice et al. 1990c, 1994, Watkins et al. 1991). Additionally, this neonate exhibited the highest AST and CK concentrations, indicative of cellular damage associated with skeletal muscle (Benjamin 1981: 231), as well as the most elevated serum cortisol concentration (38.4 µg/dL, Table 2). Elevated serum cortisol often accompanies severe nutritional restriction and increased protein catabolism, and, associated with stress, induces a physiologic leukocytosis (Coles 1980: 50– 51, 280–281, DelGiudice et al. 1990c), indicated in this neonate by the sampled cohort's highest WBC count (7.8 x 10^3/µL) and neutrophilia (84.1% [x 103/µL], Table 1).

The blood profile of neonate 520 provided a plethora of evidence that this individual was in very poor condition, likely moribund prior to capture, an assessment and prognosis not possible simply from physical examination in the field or during laboratory necropsy. It is unclear whether this neonate's poor condition was due to a problem with its ability to nurse or the dam's inability to provide nutrition. However, many of its hematological and serum analyte measurements were starkly different from those of 2 other similarly large neonates, numbers 519 (19.5 kg) and 503 (18.5 kg). Their RBC counts and Hgb concentrations were similar to those of neonate 520, but not

due to hemoconcentration and dehydration. Their RBC and Hgb values, associated with a near average PCV, TPP, SUN, creatinine, Ca, and TP values, higher globulin, glucose, and FT_3 concentrations, and markedly lower cortisol were reflective of calves adequately nourished and in good nutritional condition. Indeed, neonate 519 was the only calf that yielded several analyte values (TPP, TP, globulin, and free T_3) higher than 2 standard deviations (SD) above the sampled cohort's respective mean values (95% portion of the distributions). This calf slipped its collar at 13 days old, whereas neonate 503 survived until at least 279 days old, when we removed its collar.

Reproductive success, defined as producing a calf that survives to 1 year of age (i.e., recruitment), is an important driver of population performance (Gaillard et al. 2000, Raithel et al. 2007). Since 2012, the study population has experienced a 5-year interval of relative stability at about 4,000 moose (DelGiudice 2016). Recruitment has been low, but pregnancy rates indicate that fertility is not limiting reproductive success. Neonatal blood profiles and morphological characteristics indicate that physical and physiological development were relatively robust and did not reflect any specific vulnerabilities, except in the case of neonate 520. Comparing this case to the overall data set of reference values demonstrates the key role that blood profiles can play in improving understanding of the nutritional and health status of cervid neonates when mortality pressures are greatest. Taking advantage of every research means possible becomes increasingly important in attempting to identify and best comprehend factors impacting a steadily declining moose population.

ACKNOWLEDGEMENTS

Thanks to T. Enright, K. Foshay, T. Obermoller, J. Lodel, B. Betterly, A. Jones, K. Miedtke, B. Smith, E. Hildebrand, D. Pauly, M. Carstensen, M. Dexter, L. Cornicelli, M. Larson, J. Forester, A. Wünschmann, A. Armien, B. Patterson, R. Moen, A. McGraw, J. Terry, and M. Schrage for varying combinations of technical and cooperative support during the planning and operational phases of this study. We are appreciative for the flying, capture, and calf-handling skills of R. Swisher and M. Keech (Quicksilver Air, Inc., Fairbanks, Alaska) and the fixed-wing aircraft assistance of A. Buchert and L. Ettl. This project was funded in part by the Minnesota Department of Natural Resources Section of Wildlife and the Wildlife Restoration (Pittman-Robertson) Program. We thank the Minnesota Deer Hunters Association for supplemental funding.

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