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HEMATOLOGIC VALUES FOR TULE ELK (*CERVUS ELAPHUS NANNODES*)

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ABSTRACT: Hematologic values for 99 tule elk (*Cervus elaphus nannodes*) from California (USA) are presented. These were obtained from individuals from three captures at Tomales Point (Point Reyes National Seashore, California) from 1997–98. Differences between capture groups were assessed. Greatest differences were detected between yearling bulls and cows in December 1998 which may be a reflection of age and reproductive status.

Key words: Hematology, *Cervus elaphus nannodes,* reference values, tule elk.

INTRODUCTION

Physiologic reference values can be useful for evaluation of the health of populations of free-ranging elk (*Cervus elaphus*). Deviations from established standard values may be the result of variation in the sample population, may reflect nutritional status, or changes in the presence of disease. There are published hematologic values for Roosevelt and Rocky Mountain elk (*C. elaphus roosevelti, C. elaphus nelsoni*) (Herin, 1968; Knight, 1969; Boyd, 1970; Follis, 1972; Vaughan et al. 1973; Weber and Bliss, 1972; Pedersen and Pedersen, 1975) but little has been published on hematology of tule elk (*C. elaphus nannodes*), a subspecies of North American elk found only in California (USA). Tule elk were hunted to near extinction in the late 1800s and early 1900s and have recovered to numbers approaching 4,000 through the efforts of the California Department of Fish and Game. The purpose of this report is to present hematologic values for free-ranging tule elk at Point Reyes National Seashore (California).

MATERIALS AND METHODS

Point Reyes National Seashore (PORE; 38°07′30″N, 122°45′00″W) is approximately 256 km^2 and includes most of the 48 km long peninsula north of San Francisco that lies between the Pacific Ocean on the west and the Coast Range on the east. It is partially separated from the mainland by Tomales Bay that marks the San Andreas Fault and its rift zone. The tule elk are located in a wilderness area within the seashore on Tomales Point. This area was formerly a mixed dairy and beef cattle ranch. Elk are restricted to a 1,053 ha range by a tall fence running from the ocean to the bay.

Elk were captured by the staff of PORE, veterinarians from the University of California School of Veterinary Medicine (Davis, California) and Humboldt State University (Arcata, California), and PORE authorized volunteers from the United States Geologic Survey—Biological Resource Division (Fort Cronkhite, Sausalito, California) and the Golden Gate National Recreation Area (Fort Mason, San Francisco, California). Blood samples were collected under a National Park Service Collection Permit (96-04, Identification Number 248).

Tule elk sampled in this study were captured and radio collared as subjects of an immunocontraception trial and translocation effort within the park. In addition, elk were ear tagged during 1998. In August 1997 and June 1998 elk were captured and processed using physical restraint (Stoops et al., 1999). Elk captured in December 1998 were transported by helicopter to the processing area. All elk cows received one of several formulations of an intramuscular injection of porcine zona pellucida (pZP) for reversible contraception (Shideler, 2000).

The first set of blood samples ($n=25$) was collected from elk cows over a 2 day capture-and-release effort in August of 1997 (August 1997 cows). A second set ($n=28$) was collected over 2 days in June 1998 (June 1998 cows). The third sample set ($n=46$) was collected from five yearling bulls (December 1998 males) and 41

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elk cows (December 1998 cows) over 2 days in December 1998.

Blood was collected by jugular venipuncture using either an 18 gauge needle on a 20 ml syringe or a 20 gauge needle coupled to an evacuated blood collection tube. Blood was placed in 5.0 ml tubes containing ethylenediaminetetracetic acid (EDTA). Filled blood tubes were kept cool on crushed ice. In December 1998 blood smears were made in the field. The Hematology Laboratory (University of California, Veterinary Medical Teaching Hospital [VMTH], Davis, California) conducted the hematology. The August 1997 blood samples were delivered to the laboratory within 24–36 hr, the June 1998 blood samples were delivered within 36–48 hr, and the December 1998 samples were kept in the field 24–60 hr prior to being taken to the laboratory.

Due to deterioration of some samples, white blood cell (WBC) differential counts were not performed on the June 1998 samples or on individual samples from other groups if they were clotted. Thus, sample sizes for some parameters within groups were variable. Red and WBC counts, platelet counts, and hemoglobin values were determined using a Baker System 9000 Counter (Serono Baker Diagnostics, Allentown, Pennsylvania, USA) according to manufacturer's recommendations. Blood smears for differential counts were stained with a Wescor Aerospray Slide Stainer (Ogden, Utah, USA) (Veterinary Medical Teaching Hospital, 2001). Packed cell volume was determined on an Adams Micro-Hematocrit Reader (Clay Adams, Becton Dickinson and Co., Franklin Lakes, New Jersey, USA). Red blood cell parameters were derived from calculations based on the red blood cell counts and the packed cell volume. Serum protein content was determined on a refractometer (TS* Meter, American Optical Instrument Company, Buffalo, New York, USA); fibrinogen by heat precipitation (Veterinary Medical Teaching Hospital, 1999), and icterus index by visual comparison of plasma to a potassium dichromate standard (Sigma Chemical Co., St. Louis, Missouri, USA). Pregnancy status was determined by measurements of fecal ovarian steroid metabolites (Stoops et al., 1999) which was later confirmed by counts of live and dead calves and fetuses found during a cull.

Standard descriptive statistics were derived for hematologic values for each capture group and those results were tested for differences. Normality and equal variance of capture group distributions were tested by one-way analysis of variance. If passed, differences between the means of capture groups were compared by pairwise multiple comparison procedures (Bonferroni's Method, $\alpha=0.05$). When the test for normality failed, the Kruskal-Wallis one-way analysis of variance on ranks was used to test differences in median values among capture groups, followed by pairwise multiple comparison procedures (Dunn's Method) to isolate differences. The Student's *t*-test (two sample sets assuming equal variances) was performed as an additional test of differences between two sample groups. In comparisons using Student's *t*-test where the test for normality failed, the Mann-Whitney rank sum test was used to test differences between group medians. All descriptive and comparison statistics were done using SigmaStat® (SPSS Science, Chicago, Illinois, USA).

RESULTS

Pair-wise multiple comparisons between capture groups showed significant differences ($P<0.05$) between means of August 1997 and June 1998 cows and between means of June 1998 and December 1998 cows for mean red blood cell counts (RBC, Table 1). Difference between means of the December 1998 cows and the December 1998 males also was significant (Table 1). Mean RBC counts were highest in June 1998 cows and lowest in December 1998 males (Table 2).

Mean hemoglobin (HGB) concentrations differed significantly between August 1997 and June 1998 cows, June 1998 and December 1998 cows, and December 1998 cows and December 1998 males (Table 1). The lowest mean HGB concentration was in December 1998 males (Table 2). Hemoglobin values of December 1998 males were lower than those of December 1998 nonpregnant cows ($P<0.01$) whose values were lower than those of pregnant cows ($P<0.01$).

Mean hematocrit (HCT) values differed significantly between August 1997 and June 1998 cows ($P<0.05$), June 1998 and December 1998 cows, and December 1998 cows and December 1998 males (Table 1). December 1998 male HCT mean values were lower than those of nonpregnant and pregnant December 1998 cows

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TABLE 1. Summary of differences between capture groups of tule elk at Point Reyes National Seashore, California.

Parameter[a]	Statistical test	Group 1	Group 2	*P* Values
	1[b]	August 97 cows	June 98 cows	−<0.05
	1	June 98 cows	December 98 cows	+<0.05
RBC	2[c]	December 98 cows	December 98 males	+0.0077
	1	August 97 cows	June 98 cows	−<0.05
	1	December 98 cows	December 98 males	+<0.0001
	3[d]	December 98 males	December 98 nonpregnant cows	−<0.0001
HGB	4[e]	December 98 pregnant cows	December 98 nonpregnant cows	+<0.0001
	1	August 97 cows	June 98 cows	−<0.05
	1	June 98 cows	December 98 cows	+<0.05
	3	December 98 cows	December 98 males	+<0.0001
HCT	4	December 98 nonpregnant cows	December 98 pregnant cows	−<0.0001
	3	December 98 cows	December 98 males	+0.0328
	1	December 98 nonpregnant cows	December 98 pregnant cows	−0.01
MCV	1	December 98 males	December 98 pregnant cows	<0.05
	4	June 98 cows	December 98 cows	−<0.05
	3	December 98 cows	December 98 males	+0.0040
MCH	4	December 98 nonpregnant cows	December 98 pregnant cows	−0.007
	4	August 97 cows	June 98 cows	+<0.05
MCHC	4	June 98 cows	December 98 cows	−<0.05
Plasma protein	1	August 97 cows	December 98 cows	−<0.05
Fibrinogen	2	December 98 cows	December 98 males	−0.0197
	3	December 98 cows	December 98 males	−0.0001
Platelets	1	December 98 nonpregnant cows	December 98 pregnant cows	+<0.003
	1	August 97 cows	June 98 cows	+0.0001
WBC	1	June 98 cows	December 98 cows	−0.0001
Neutrophils	3	December 98 cows	August 97 cows	+0.0001
	3	August 97 cows	December 98 cows	+0.0011
Lymphocytes	3	December 98 males	December 98 cows	−0.0366
Monocytes	2	December 98 cows	December 98 males	−0.0045
Icterus index	3	August 97 cows	June 98 cows	+0.0001

[a] See text for abbreviations for tests.
[b] 1 = Pairwise multiple comparisons procedures (Bonferroni's Method).
[c] 2 = Mann-Whitney rank sum test.
[d] 3 = Students *t*-test.
[e] 4 = Pairwise Multiple comparisons procedures (Dunn's Method).

($P<0.01$) and levels were lower in nonpregnant compared to pregnant December 1998 cows.

While differences in the values of mean corpuscular volume (MCV) of cow capture groups were not significant, differences between December 1998 cows and December 1998 males were significant (Table 1). December 1998 male MCV values were lower than those of nonpregnant or pregnant December 1998 cows ($P<0.01$). Mean corpuscular hemoglobin (MCH) values varied significantly between June 1998 and December 1998 cows ($P<0.05$), as well as between mean values of December 1998 cows and December 1998 males ($P<0.01$, Table 1). December 1998 nonpregnant cows had lower HCT, MCV, and

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TABLE 2. Hematology parameters of tule Elk at Point Reyes National Seashore, California.

CBC parameters[a]		Cow elk August 97	Cow elk June 98	Cow elk Dec 98
RBC	*n*	25	28	41
$\times 10^6$ µl	Mean	9.90	11.00	10–12
	Range	7.29–12.49	8.66–13.98	7.80–11.86
	Std. deviation	1.19	1.26	0.70
HGB	*n*	25	28	41
g/dl	Mean	16.95	18.05	17.32
	Range	15.40–19.10	15.80–20.00	14.30–20.10
	Std. deviation	0.98	1.09	1.39
HCT	*n*	25	28	41
%	Mean	50.16	54.97	51.10
	Range	44.60–57.70	47.50–60.60	42.50–58.50
	Std. deviation	3.05	3.24	3.61
MCV	*n*	25	28	41
Fl	Mean	51.17	50.45	50.71
	Range	42.40–62.60	39.50–63.30	45.0—59.70
	Std. deviation	4.85	5.03	2.65
MCH	*n*	25	28	41
Pg	Mean	17.30	16.60	17.17
	Range	14.40–21.40	12.40–21.30	14.70–20.80
	Std. deviation	1.67	1.77	1.03
MCHC	*n*	25	28	41
g/dl	Mean	33.81	32.10	33.87
	Range	31.40–36.10	13.00–34.20	32.20–35.20
	Std. deviation	1.04	3.79	0.67
Icterus	*n*	20	23	40
index	Mean	3.67	1.74	2.08
	Range	2–5	2–5	2–5
	Std. deviation	3.67	1.00	0.47
Plasma	*n*	25	28	41
protein	Mean	7.43	7.38	7.79
g/dl	Range	7.00–8.70	6.70–8.60	7.10–8.40
	Std. deviation	0.40	1.50	0.30
Fibrinogen	*n*	25	28	41
mg/dl	Mean	342	333	322
	Range	100–600	100–600	200–600
	Std. deviation	128.83	121.72	92.43
Platelets	*n*	25	28	40
$\times 1000/\mu l$	Mean	2.56E[b]+05	2.85E+05	2.66E+05
	Range	8.00E+04–3.71E+05	4.30E+04–4.20E+05	1.05E+05–4.44E+05
	Std. deviation	5.93E+04	8.98E+04	7.58E+0r
WBC/µl	*n*	25	28	41
	Mean	7.43E+03	1.07E+04	8.12E+03
	Range	4.40E+03–1.11E+04	5.50E+03–1.59E+04	5.40E+03–1.17E+04
	Std. deviation	1.48E+03	2.04E+03	1.27E+03
Neutrophils	*n*	25	ND[c]	41
/µl %	Mean	23		38
	Range	8–44		12–77
	Std. deviation	9		13
abs	Mean	1.70E+03		3.12E+03
	Range	552–4092		1164–6006
	Std. deviation	8.20E+02		1.14E+03

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TABLE 2. Extended.

Cow elk, nonpregnant Dec 98	Cow elk, pregnant Dec 98	Spike bulls Dec 98	All elk total
10 (of 41)	31 (of 41)	5	99
9.82	10.19	8.28	9.83
8.85–11.34	7.80–11.60	6.57–9.90	6.57–13.98
0.71	0.68	1.43	0.99
10	31	5	99
15.80	17.70	12.82	16.29
14.30–17.80	16.10–20.10	9.70–14.40	9.70–20.10
1.31	1.15	1.80	1.29
10	31	5	
47.85	52.20	37.94	48.10
42.50–54.40	46.60–58.50	28.00–43.20	28.0–60.60
3.72	2.97	5.96	3.67
10	31	5	99
48.99	51.30	45.86	49.55
45.00–51.50	45.60–59.70	42.60–52.80	39.50–63.30
2.58	2.47	3.70	3.55
10	31	5	99
16.40	17.40	15.56	16.61
14.70–17.90	15.40–20.80	13.90–18.20	12.40–21.40
1.03	0.95	1.48	1.32
10	31	5	99
33.60	34.00	33.90	33.42
32.20–34.90	32.70–35.20	32.50–34.90	13.00–36.10
0.88	0.60	1.04	1.34
10	30	5	88
2	2.10	2.0	2.58
2	2–5	2.0	2–5
0	0.55	0	1.18
10	31	5	99
7.56	7.85	6.86	7.37
7.10–8.20	7.40–8.40	5.8–7.50	5.80–8.70
0.35	0.26	0.63	0.57
10	31	5	99
311	325	460	364.25
200–400	200–600	300–600	100–600
78.20	98.40	114.00	105.60
10	31	5	98
3.30E+05	2.47E+05	4.31E+05	3.10E+05
2.07E+05–4.44E+05	1.05E+05–4.00E+05	3.19E+05–5.81E+05	4.30E+04–5.81E+05
5.74E+04	7.19E+04	1.07E+05	7.68E+04
10 (of 41)	31 (of 41)	5	99
8.23E+03	8.09E+03	8.62E+03	8.07E+03
5.40E+03–1.03E+04	6.10E+03–1.17E+04	5.70E+03–1.09E+04	4.40E+03–1.59E+04
1.55E+03	1.23E+03	1.90E+03	1.58E+03
10	31	5	71
40	38	44	37
30–53	12–77	30–59	8–77
7	14	13	11
3.24E+03	3.09E+03	3.82E+03	2.96E+03
2160–4876	1164–6006	2520–6431	552–6431
8.16E+02	1.24E+03	1.67E+03	1.14E+03

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TABLE 2. Continued.

CBC parameters[a]			Cow elk August 97	Cow elk June 98	Cow elk Dec 98
Lymphocytes		*n*	25	ND	41
/μl	%	Mean	60		44
		Range	44–74		18–73
		Std. deviation	8		11
	abs	Mean	4.47E+03		3.61E+03
		Range	2860–7104		1404–7081
		Std. deviation	1.00E+03		1.11E+03
Monocytes		*n*	25	ND	41
/μl	%	Mean	4		3
		Range	1–9		1–10
		Std. deviation	2		2
	abs	Mean	272		282
		Range	44–999		67–1030
		Std. deviation	220		184
Eosinophils		*n*	25	ND	41
/μl	%	Mean	13		13
		Range	3–34		4–26
		Std. deviation	6		5
	abs	Mean	935		1034
		Range	279–2584		312–2470
		Std. deviation	472		462
Basophils		*n*	25	ND	41
/μl	%	Mean	0.7		1.1
		Range	0–3		0–5
		Std. deviation	1		1
	abs	Mean	46		89
		Range	0–186		0–450
		Std. deviation	60		96

[a] See text for abbreviation of parameters.
[b] E = exponent.
[c] ND = not determined.

MCH values than December 1998 pregnant cows ($P<0.01$, $P=0.01$, and $P<0.01$, respectively). The index of the average hemoglobin content of RBCs or the mean corpuscular hemoglobin concentration (MCHC) differed significantly between June 1998 and December 1998 cows as well as between August 1997 and June 1998 cows (Table 1).

Mean plasma protein levels varied significantly between August 1997 and December 1998 cows ($P<.05$). A difference also was found between the December 1998 cows and the December 1998 males (Table 1). Mean plasma protein levels were highest in December 1998 cows and lowest in December 1998 males (Table 2). There was also a statistically significant difference in fibrinogen levels between December 1998 cows and December 1998 males (Table 1), with levels being higher in the bulls. No significant differences were found between platelet counts of any of the cow capture groups but there was a difference between December 1998 cows and December 1998 males ($P<0.01$), Mann-Whitney rank sum test, (Table 1). The platelet counts in the bulls were almost twice that in the cows. In addition, higher platelet counts were found in the December 1998 nonpregnant cows compared to the pregnant cows (Table 1). There was a significant difference between the icterus index of the August 1997 and

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TABLE 2. Extended Continued.

Cow elk, nonpregnant Dec 98	Cow elk, pregnant Dec 98	Spike bulls Dec 98	All elk total
10	31	5	71
42	45	29	45
33–53	18–73	5–45	5–74
7	12	16	11
3.63E+03	3.64E+03	2.45E+03	3.55E+03
2430–5459	1404–7081	435–3780	435–7104
8.78E+02	1.18E+03	1.13E+03	1.06E+03
10	31	5	71
3	3	7	5
2–10	1–9	5–8	1–10
3	2	1	2
319.10	278	597	384
162–1030	67–702	285–763	44–1030
270.58	157	164	201
10	31	5	71
13	12	9	12
5–26	4–24	3–14	3–34
6	5	4	5
1102	997	725	898
515–2470	312–2040	171–1176	171–2584
581	439	335	458
10	31	5	71
1.1	1.0	1.8	1.2
0–3	0–5	0–3	0–5
1	1	1	1
97	86	166	100
0.225	0–450	0–252	0–450
97	99	87	88

June 1998 cows ($P<0.01$, Mann-Whitney rank sum test, Table 1).

Variations in mean WBC counts were significant between August 1997 and June 1998 cows and between June 1998 and December 1998 cows ($P<0.01$, Table 1). The highest mean population WBC count was observed in June 1998 cows and the lowest in August 1997 cows (Table 2).

Analysis of WBC counts demonstrated differences among groups only in neutrophil, lymphocyte, and monocyte counts. Mean absolute neutrophil counts were significantly higher in December 1998 cows than in August 1997 cows ($P<0.01$, Table 1). Mean absolute lymphocyte counts were significantly higher among individuals August 1997 cows than in December 1998 cows or males ($P<0.01$, and 0.04, respectively, Table 1). Absolute monocyte counts also were significantly higher among December 1998 males as compared to December 1998 cows ($P<0.01$, Table 1).

The August 1997 cows were sampled at the end of their calving season. With an estimated calving rate of 90% for the 1996–97 year (Stoops et al., 1999) and the start of the rut in late August, the majority of elk cows from this capture group were lactating and had begun their seasonal ovarian cycling. Most blood parameters appeared similar to those observed in other capture groups with the exception of neutrophils counts, which had a lower mean and wider range (Table 2).

Calving was still in progress during the June 1998 capture. The estimated calving rate for the 1997–98 year was 77% (Shi-

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deler, 2000). Thus, the majority of adult female cows were either in late pregnancy or lactating; some were both, because it is not unusual for yearlings to nurse until birth of a new calf. None had begun cycling. This capture group had the highest mean RBC and WBC counts, HGB values, and percent HCT of all capture groups which may have been related to their reproductive condition.

Elk cows for translocation during December 1998 were selected on the basis of age, health, and reproductive status; young females in early pregnancy were preferred. Thirty-one cows were in this category. Young males (yearling bulls in first velvet) were selected for translocation in order to avoid the danger to personnel and other animals of moving unsedated, fully-grown bulls with large antlers.

The December 98 cows had higher RBC counts, HCT, HGB, MCV, MCHC, protein, and fibrinogen values than the yearling bulls from the same capture. While yearling males did not have higher WBC counts, the proportion of their WBC components differed from those of the cows, with spikes having higher absolute platelet, neutrophil, and monocyte counts. When pregnant and nonpregnant cows from this capture were analyzed as separate groups, the nonpregnant cows had lower RBC, HGB, HCT, MCV, MCH, MCHC, icterus index, plasma protein, fibrinogen, and absolute lymphocyte measurements and higher platelet, WBC, and absolute neutrophil and basophil counts. The yearling bulls had the highest platelet, fibrinogen, neutrophil, monocyte, and basophil values and the lowest RBC, HGB, HCT, MCV, MCH, and protein values of all groups.

DISCUSSION

Yearling males in this study were weaned by mothers with new calves at approximately 1 yr old and were excluded from the safety of their natal groups 3 mo later by the rutting bulls. The young males in this study were growing their first antlers. The stresses of being on their own, living as new additions to bachelor groups, as well as the bioenergetic costs of growing antler, may be reflected in their hematologic values.

The December 1998 cows were primarily young, pregnant females. The remaining females in this group were either not pregnant at capture or had suffered fetal losses. These young female tule elk may remain at the side of their mothers after a new calf is born but they are frequently herded away by bulls in rut or driven off by other adult females in the group with calves. This in contrast to tule elk cows that do not have new calves and which have been seen grooming and intermittently nursing young bull or cow offspring for up to 3 yr.

There are many changes that accompany pregnancy and some of these changes may account for differences between June 1998 elk cows, December 1998 elk cows, and other capture groups. Maternal oxygen levels, cardiac output, erythrocyte numbers, and efficiency of intermediary metabolism of amino acids increase during pregnancy (Johnson and Everitt, 1990). Blood volume and fibrinogen increase while HCT decreases (Greenspan and Forsham, 1983). Increased RBC numbers account for the increase in HCT in December 1998 pregnant cows. Fibrinogen was higher in pregnant compared to nonpregnant cows but was lower than in the yearling bulls. Some of these changes may be due to increased production of specific serum binding proteins or decreased serum albumin levels (Greenspan and Forsham, 1983). Others may be mediated by differences in clearance rates, perhaps due to increased glomerular filtration, decreased hepatic excretion of anions, or metabolic clearance of steroid and protein hormones by the placenta (Greenspan and Forsham, 1983). It is possible that the differences in hematologic values between 1998 December males and 1998 December pregnant cows reflect differences in

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age and/or physiologic and reproductive status.

Certain hematologic measurements such as WBC differential counts were not conducted for some groups or individuals due to the length of time the samples were in the field prior to analysis. No special statistical tests were performed to determine if the delay between collection and analyses affected results in other sample groups.

Hematologic values reported here are for cows in estrus, lactating, early pregnancy, and late pregnancy and yearling bulls in first velvet. The results do not represent resting values. While their body temperatures were not allowed to get as high as those reached in Herin (1968) study (41.6–42.4 C), the elk were stressed and agitated by chase and capture, and, in December 1998, helicopter transport to the processing area which could have affected these hematologic parameters.

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