

## BLOOD CHEMISTRY AND TRACE MINERAL REFERENCE VALUES FOR CALIFORNIA BIGHORN SHEEP IN OREGON

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*Abstract:* Many agencies, including the Oregon Department of Fish and Wildlife, routinely collect blood samples from animals captured during management, research, and transplant operations. Resulting blood chemistry and mineral data are often used as indices of animal health. However, in most cases data are compared to livestock standards because baseline values are not well established for most wildlife species. We report on analysis of over 400 California bighorn sheep blood samples taken from 6 populations over a 13 year period. Where repeated sampling events have occurred (n=3), we attempt to relate blood parameters to population performance. Relative value of this information as baseline for California bighorns is discussed.

*Key words:* blood chemistry, California bighorn sheep, *Ovis canadensis californiana*, Oregon, serum trace mineral.

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Bighorn sheep (*Ovis canadensis*) are an extremely popular wildlife species in much of western North America. For many agencies, bighorn sheep programs are high profile in nature and may involve intensive management using trap and transplant operations, frequent population monitoring, and biological sampling to assess overall health of individual animals and populations.

Evaluation of blood chemistry parameters and mineral levels can be used to assess animal health (Puls 1995) and as a diagnostic tool for identifying disease in individual animals (Chin 1997). Difficulties arise, however, because limited information exists documenting normal blood chemistry values for wild species, and ranges reported for domestic breeds may not be representative of wild species' profiles (Hebert 1978). Additionally, stress, capture, nutritional variability, and captivity may alter blood chemistry profiles (Franzmann and Thorne 1970, Hebert 1978, Kock et al. 1987a, Kock et al. 1987b).

Limited information is available documenting blood chemistry profiles for wild sheep species. We found 2 reports of blood chemistry profiles for thinhorn sheep (*O. dalli dalli* and *O. d. Stonei*, Foreyt et al. 1983, Franzmann 1971b). For bighorn sheep, most reports describe blood chemistry profiles for Rocky Mountain bighorn sheep (*O. c. canadensis*, Bottrell et al. 1978, Davies 1976, Franzmann 1971a, Franzmann and Thorne 1970, Hebert 1978, Wolf and Kradel 1969) or desert bighorn sheep (*O. c. nelsoni*; Borjesson et al. 2000, McDonald et al. 1981). Only 1 report specifically describes blood chemistry profiles for California bighorn sheep (*O. c. californiana*; Bottrell et al. 1978). Throughout all reports we found for all species or subspecies of wild sheep, data were not consistent with regard to which parameters were specifically reported and with few exceptions, sample sizes were generally limited with regard to development of normal values and ranges.

We analyzed results of over 400 blood

samples collected from 6 California bighorn sheep populations over a 13-year period.

Our objectives for this analysis were:

- 1) To evaluate the effects of age and sex on mean serum chemistry values;
- 2) Compare mean serum chemistry values across time within populations with repeated sampling events;
- 3) Compare mean serum chemistry values between populations within sampling years where multiple populations were sampled;
- 4) Explore the possibility of relationships between blood chemistry and population growth; and
- 5) Develop normal values and reference ranges for blood chemistry of California bighorn sheep.

## STUDY AREAS

California bighorn sheep were native to the fault-block mountains and high plains of southeastern Oregon but were extirpated by about 1915 (Oregon Department of Fish and Wildlife 1992). In 1954, California bighorns from British Columbia were reintroduced to Hart Mountain and since establishment have been captured and used for transplants to suitable habitat throughout most of central and southeastern Oregon. A spring 1992 population estimate indicated Oregon had about 1,950 California bighorns (Oregon Department of Fish and Wildlife 1992). By 1999, populations had increased to over 2,700 animals (Oregon Department of Fish and Wildlife 1999).

Hart Mountain is a major fault-block mountain range that has its escarpment facing west. It is within the Hart Mountain National Antelope Refuge, and reaches an upper elevation of 8,065 feet. The escarpment drops 3,600 feet to the Warner Valley at the base of the west slope. Bighorns are associated with the rugged cliffs and adjacent sagebrush ridges.

Steens Mountain also is a fault-block

mountain, but its escarpment faces east. Elevation drops from 9,730 to 4,200 in about 5 lateral miles. Bighorns primarily occupy moist, green, hanging valleys and cirques carved by glaciation. Precipitous cliffs provide excellent lambing habitat. Bighorns were reintroduced to Steens Mountain in 1960 from the established population on Hart Mountain.

Leslie Gulch is a rimrock canyon associated with the rugged Owyhee River Canyon of Malheur County, Oregon. It lies east of Owyhee Reservoir and is formed by highly dissected canyons within ancient lava flows. Elevations range from about 6,000 feet on Mahogany Mountain to about 2,500 feet at the water's edge of Owyhee Reservoir. Bighorns were established in Leslie Gulch in 1965 from the Hart Mountain population.

Both the John Day River and Deschutes River herds occupy major river canyon habitats. Elevations are lower (600 – 3,000 feet) than most other Oregon California bighorn ranges. The John Day River herd was established with a 1989 release from the Hart Mountain herd and a 1990 release of British Columbia California bighorns. The Deschutes River herd was established during 1995 with a transplant from the established population on Steens Mountain. The John Day River also has received additional transplants from British Columbia and the Deschutes River.

The Santa Rosa Mountains of northern Nevada are similar to several mountain ranges of southeastern Oregon, but are not uplifted fault-block mountains. The 16 bighorns sampled in 2000 were captured on Sawtooth Mountain near Oroville, and in 8-Mile Canyon south of McDermitt. These bighorns were released onto Steens Mountain as part of a genetics research study recently initiated by Oregon. The Santa Rosa herd was originally established

using bighorns from Penticton, British Columbia.

## METHODS

All but 16 of 423 bighorns sampled for this study were captured using Coda Netgun technology (Barrett et al. 1982). The remaining 16 were captured using a linear drive net and a helicopter (Beasom et al. 1980). Early netgun operations utilized department personnel but captures occurring after 1990 were performed by contractors with specific experience with bighorn sheep. Contractors provided aircraft, pilots, gunners, muggers and appropriate equipment for capture and transport of bighorns to base camp. Department personnel managed base camp operations including health evaluation, veterinary care, biological sampling, vaccinations, marking (ear tag, telemetry), and vehicle transport if required. A veterinarian was present for most capture operations and animal handling protocols were consistent with care and use guidelines of the American Society of Mammalogists (1987).

All blood samples were collected by a designated capture crew member or the veterinarian. A 50 ml sample was collected by jugular venipuncture from each bighorn using a 1.5 inch 16–20 gauge needle and a 60 cc syringe. Samples were immediately placed in sterile glass tubes without anticoagulant. All records and samples were cross-referenced with capture data sheets and included animal ID, age, sex, health and condition data, inoculations received, samples collected, tag and transmitter information, and pertinent comments.

Blood samples were maintained at ambient temperature (or in a warm pickup cab) for about 2-hr prior to being centrifuged for harvest of serum. Serum was decanted into cryotubes, labeled, and frozen for 1-3 weeks until capture operations

were complete for a year, or until entire groups of samples could be sent in a single shipment to appropriate labs for analyses. A sub-sample (3–5 ml) was retained from each bighorn and currently is maintained frozen in a serum bank by the department.

The majority of samples for this study were analyzed by the Veterinary Diagnostic Laboratory at Oregon State University in Corvallis, Oregon. Samples collected in 1988 were analyzed at the Central Oregon Laboratory in Bend, Oregon, and those done in 1989 were analyzed at the Treasure Valley Laboratory, Inc. in Boise, Idaho. Serum was analyzed for the following parameters: blood urea nitrogen (BUN, *mg/dl*), total protein (*g/dl*), albumin (*g/dl*), globulin (*mg/dl*), creatinine (*mg/dl*), creatine kinase (CK, *IU/l*), glucose (*mg/dl*), cholesterol (*mg/dl*), total bilirubin (*mg/dl*), direct bilirubin (*mg/dl*), indirect bilirubin (*mg/dl*), gamma-glutamyltransferase ( $\gamma$ GT, *IU/l*), lactate dehydrogenase (LDH, *IU/l*), alanine aminotransferase (ALT, *mg/l*), aspartate aminotransferase (AST, *IU/l*), sorbitol dehydrogenase (SDH, *IU/l*), uric acid (*mg/dl*), calcium (Ca, *mg/dl*), phosphorous (P, *mg/dl*), sodium (Na, *Meq/l*), chloride (Cl, *Meq/l*), potassium (K, *Meq/l*), magnesium (Mg, *mg/l*), iron (Fe, *mCg/dl*), and carbon dioxide (CO<sub>2</sub>, *Meq/l*). Ratios of albumin:globulin, BUN:creatinine also were calculated from lab results. Abbreviations and measurement units are consistent with those reported by Puls (1995).

Individual parameter values were first compared using Analysis of Variance (AOV). Data comparing mean values between male and female sheep were compared using *t*-tests. Where 3 or more means were compared (e.g. age classes, across multiple years within a population, across multiple populations within a year) an *F*-test was used to determine if individual mean parameter values differed. Tests

between individual means were conducted using Least Significant Difference (LSD) tests but were only conducted when the *F*-test was significant. All mean comparisons were considered significant when  $P \leq 0.05$ . The central 90<sup>th</sup> percentile was used to determine the normal range for each parameter. The median, mean ( $\bar{x}$ ), standard error (SE), and coefficient of variation (CV) for each parameter also were determined for comparisons in this study and comparison with other reported studies. No assumption of normality was made, even though many parameter distributions approximated normality. Using 4 parameters consistently found across 12 sampling events (blood urea nitrogen [BUN], creatinine, BUN:creatinine ratio, and aspartate aminotransferase [AST]) we used stepwise linear regression to determine their relationship with net population growth rate ( $R_0 = N_{t+1}/N_t$ ).

## RESULTS

Over 400 blood samples were collected from 6 distinct California bighorn sheep populations over a 13-year period (Table 1). Three populations (Hart Mountain, Steens Mountain, John Day River) had 3 or more sampling events allowing comparison within populations through time. In 3 years (1994, 1995, and 2000) 3 distinct populations were

sampled allowing comparisons across populations within a year. Because most capture operations were conducted for transplant operations, females (n=314) dominated the samples where sex was identified (n=399) and adults (n=264) dominated samples where age was identified (n=367).

Few differences in mean blood chemistry parameters were found between males and females (Table 2). Of those that differed, most were generally associated with nutrition. Mean blood urea nitrogen ( $P = 0.001$ ) and mean BUN:Creatinine ratio ( $P = 0.002$ ) were higher for males. Mean total protein ( $P = 0.036$ ), albumin ( $P = 0.026$ ), and creatinine ( $P = 0.043$ ) values were higher in females. Only 1 diagnostic parameter, mean total CO<sub>2</sub>, differed between the sexes and was higher in males.

Parameters indicative of nutrition also tended to differ between age class (lamb, yearling, adult) of bighorns (Table 3). Mean values for adults and yearlings tended to be similar whereas lambs tended to differ from yearlings, or yearlings and adults. Mean blood urea nitrogen ( $P < 0.01$ ) and BUN:Creatinine ratio ( $P < 0.01$ ) were higher for lambs than for either yearling or adults which did not differ from each other. Total

Table 1. Minimum sample size, sample population, and sample year for blood parameters of California bighorn sheep in Oregon, 1988-2000.

Herd Sampled	Year								
	1988	1989	1990	1994	1995	1996	1999	2000	Total
Hart Mountain		17	52	39	32	29			169
Steens Mountain	23			20	18			17	78
John Day River			13			13	20	28	74
Leslie Gulch				20	38				58
Deschutes River							28		28
Santa Rosa Mt, NV								16	16
Total	23	17	65	79	88	42	48	61	423
Age/Sex	Male	Female	Lamb	Yearling	Adult				
N	85	314	47	56	264				

Table 2. Mean blood serum chemistry values for male and female California bighorn sheep in Oregon, 1988-2000.

Parameter	Male		Female		<i>T</i>	<i>P</i> <  <i>T</i>
	<i>n</i>	$\bar{x}$	<i>n</i>	$\bar{x}$		
Blood Urea Nitrogen (BUN, <i>mg/dl</i> )	85	18.5	314	15.3	-3.35	0.001
Total Protein ( <i>g/dl</i> )	69	6.8	251	6.9	2.11	0.036
Albumin ( <i>g/dl</i> )	69	3.8	251	4.0	2.26	0.026
Globulin ( <i>mg/dl</i> )	23	2.8	86	2.7	-0.77	0.438
Albumin:Globulin Ratio	23	1.4	86	1.5	1.44	0.153
Creatinine ( <i>mg/dl</i> )	64	2.0	247	2.2	2.04	0.043
BUN:Creatinine Ratio	64	10.3	246	7.7	-3.22	0.002
Creatine Kinase ( <i>IU/l</i> )	41	1319	161	1031	-0.98	0.330
Glucose ( <i>mg/dl</i> )	64	152	247	149	-0.46	0.577
Cholesterol ( <i>mg/dl</i> )	23	58.6	86	57.2	-0.63	0.464
Total Bilirubin ( <i>mg/dl</i> )	48	0.2	184	0.2	0.19	0.847
Direct Bilirubin ( <i>mg/dl</i> )	23	0.0	86	0.0	0.01	0.996
Indirect Bilirubin ( <i>mg/dl</i> )	23	0.1	86	0.1	0.14	0.890
$\gamma$ GT <sup>1</sup> ( <i>IU/l</i> )	56	64.3	183	56.7	-1.43	0.155
LDH <sup>1</sup> ( <i>IU/l</i> )	29	766	131	791	0.59	0.556
ALT <sup>1</sup> ( <i>mg/l</i> )	10	42.7	18	38.6	-0.87	0.393
AST <sup>1</sup> ( <i>IU/l</i> )	85	277	314	234	-1.80	0.075
SDH ( <i>IU/l</i> )	25	37.5	98	40.8	0.32	0.753
Uric Acid ( <i>mg/dl</i> )	13	0.2	68	0.2	0.21	0.836
Calcium ( <i>mg/dl</i> )	69	10.5	251	10.6	0.39	0.700
Phosphorous ( <i>mg/dl</i> )	69	6.7	251	6.7	-0.16	0.877
Sodium ( <i>Meq/l</i> )	69	153	251	154	1.45	0.147
Chloride ( <i>Meq/l</i> )	69	97.0	251	98.1	1.15	0.254
Potassium ( <i>Meq/l</i> )	69	5.0	251	5.3	1.44	0.151
Magnesium ( <i>mg/l</i> )	46	3.1	165	3.2	1.14	0.257
Total Carbon Dioxide ( <i>Meq/l</i> )	25	10.0	98	5.5	-2.29	0.031
Iron ( <i>mCg/dl</i> )	13	209	68	199	-0.69	0.490

<sup>1</sup>  $\gamma$ GT=Gamma-glutamyltransferase; LDH=Lactate dehydrogenase; ALT=Alanine aminotransferase; AST=Aspartate aminotransferase; SDH=Sorbitol dehydrogenase.

protein differed ( $P < 0.01$ ) across all age classes with lambs having the lowest and adults having the highest mean value. Mean globulin value was lowest in lambs and greatest in yearlings, with no difference between lambs and adults or between adults and yearlings ( $P = 0.01$ ). Mean creatinine values were lowest in lambs ( $P < 0.01$ ).

Direct bilirubin and total CO<sub>2</sub> differed ( $P < 0.01$ ) with direct bilirubin lower in lambs, and CO<sub>2</sub> higher in lambs than for yearlings and adults. Sodium and chloride were the only electrolytes that differed ( $P < 0.01$ ) and tended to be lower in lambs.

Considerable variation existed across populations analyzed within years (Table 4).

Table 3. Mean serum chemistry values for lamb, yearling, and adult California bighorn sheep in Oregon, 1988-2000. Means within a row with the same letter are not different (LSD,  $P = 0.05$ ).

Parameter	Lamb		Yearling		Adult		<i>F</i>	<i>P&gt;F</i>
	N	$\bar{x}$	n	$\bar{x}$	N	$\bar{x}$		
BUN (mg/dl)	47	19.3a	56	16.1b	264	15.7b	5.33	<0.01
Σ Protein (g/dl)	45	6.3a	42	6.7b	201	7.0c	17.49	<0.01
Albumin (A, g/dl)	45	3.7	42	3.9	201	3.9	2.44	0.09
Globulin (G, mg/dl)	21	2.6a	8	3.1b	62	2.8ab	4.47	0.01
A:G Ratio	21	1.6	8	1.4	62	1.5	1.22	0.30
Creatinine (C, mg/dl)	37	1.9a	45	2.1b	197	2.2b	8.18	<0.01
BUN:C Ratio	36	11.6a	45	8.6b	197	7.9b	8.73	<0.01
Creatine Kinase (IU/l)	16	1769	37	890	135	1078	2.01	0.14
Glucose (mg/dl)	37	142	45	152	197	149	0.98	0.38
Cholesterol (mg/dl)	21	54.0	8	59.6	62	57.9	1.90	0.16
Σ Bilirubin (mg/dl)	35	0.1	31	0.2	134	0.1	0.19	0.85
Dir. Bilirubin (mg/dl)	21	0.0a	8	0.1b	62	0.0a	8.72	<0.01
Ind. Bilirubin (mg/dl)	21	0.1	8	0.1	62	0.1	0.10	0.89
γGT <sup>1</sup> (IU/l)	32	47.9	34	66.0	159	59.4	2.32	0.10
LDH <sup>1</sup> (IU/l)	15	803	22	862	105	154	0.96	0.38
ALT <sup>1</sup> (Mg/l)	8	40.6			20	39.8	0.02	0.88
AST <sup>1</sup> (IU/l)	47	267	56	235	264	240	0.62	0.54
SDH <sup>1</sup> (IU/l)	14	33.0	23	40.0	72	34.0	0.21	0.81
Uric Acid (mg/dl)	13	0.2	8	0.1	42	0.2	0.86	0.43
Ca (mg/dl)	45	10.1	42	10.3	201	10.8	0.60	0.55
P (mg/dl)	45	4.8	42	5.2	201	5.2	1.64	0.20
Na (Meq/l)	45	149a	42	153ab	201	155b	7.07	<0.01
Chl (Meq/l)	45	94.5a	42	95.5a	201	99.0b	5.81	<0.01
K (Meq/l)	45	4.8	42	5.2	201	5.2	1.64	0.20
Mg (mg/dl)	24	2.9	34	3.0	139	3.2	2.66	0.07
Σ CO <sub>2</sub> (Meq/l)	14	14.4a	23	6.6b	72	5.3b	19.94	<0.01
Fe (mCg/dl)	13	216	8	180	42	210	2.16	0.12

<sup>1</sup> γGT=Gamma-glutamyltransferase; LDH=Lactate dehydrogenase; ALT=Alanine aminotransferase; AST=Aspartate aminotransferase; SDH=Sorbitol dehydrogenase.

Mean BUN values were higher ( $P < 0.01$ ) in Leslie Gulch compared to Hart Mountain and Steens Mountain during both 1994 and 1995. Creatinine and BUN:Creatinine ratios differed across populations in 1994.

Aspartate aminotransferase differed ( $P < 0.01$ ) during 1994 but not during 1995 ( $P = 0.66$ ). Four minerals differed (Ca, P, Na, and Mg,  $P < 0.01$ ) between populations

during 1995. All nutritional indices analyzed except for glucose and mean total bilirubin, calcium, phosphorous, and potassium differed ( $P < 0.01$ ) between Steens Mountain, John Day River, and Santa Rosa Mountains (Nevada) populations during 2000.

Considerable variation also existed within populations through time (Table 5).

Table 4. Mean blood serum chemistry values within years for California bighorn sheep in Oregon. Means within a row with the same letter are not different (LSD,  $P = 0.05$ ).

Year Parameter	Population			<i>F</i>	<i>P&gt;F</i>
	Hart	Steens	Leslie		
1994					
BUN (mg/dl)	9.4a	13.9b	17.4c	27.76	<0.01
Creatinine (mg/dl)	2.6a	2.7a	2.3b	14.53	<0.01
BUN:Creat.	3.7a	5.2b	7.7c	43.13	<0.01
CK <sup>1</sup> (UI/l)	745	864	698	0.40	0.67
Glucose (mg/dl)	156	152	162	0.74	0.48
LDH <sup>1</sup> (UI/l)	628	681	687	2.71	0.03
AST <sup>1</sup> (UI/l)	183a	182a	432b	14.16	<0.01
1995					
BUN (mg/dl)	11.1a	9.2a	22.9b	104.20	<0.01
Σ Protein (g/dl)	6.4a	7.0b	6.8b	6.82	<0.01
Albumin (g/dl)	3.3	3.3	3.4	1.02	0.36
γGT <sup>1</sup> (UI/l)	62.4	70.5	55.6	1.40	0.25
AST <sup>1</sup> (UI/l)	196	214	216	0.42	0.66
Ca (mg/dl)	10.1a	10.3a	11.5b	27.06	<0.01
P (mg/dl)	6.0a	5.4a	6.8b	6.17	<0.01
Na (Meq/l)	154a		158b	14.01	<0.01
Chl (Meq/l)	100		100	0.10	0.90
K (Meq/l)	4.8a		5.1b	4.27	0.02
Mg (mg/dl)	2.6a	2.9b	3.1b	16.26	<0.01
2000					
	Steens	John Day	Nevada		
BUN (mg/dl)	18.3a	23.6b	11.7c	39.21	<0.01
Σ Protein (g/dl)	8.4a	6.7b	6.2b	21.97	<0.01
Albumin (g/dl)	5.2a	4.0b	4.1b	18.23	<0.01
Creatinine (mg/dl)	2.2a	1.6b	2.0a	14.70	<0.01
BUN:Creat.	8.4a	14.9b	5.9c	61.73	<0.01
Glucose (mg/dl)	150	138	107	1.57	0.216
Σ Bilirubin (mg/dl)	0.3a	0.1b	0.1b	28.88	<0.01
γGT <sup>1</sup> (UI/l)	52.5	74.5	50.1	2.17	0.12
AST <sup>1</sup> (UI/l)	207	263	200	1.99	0.15
Ca (mg/dl)	11.8a	10.0b	9.7b	9.56	<0.01
P (mg/dl)	7.8a	6.9a	5.1b	6.84	<0.01
Na (Meq/l)	162	152	147	2.20	0.12
Chl (Meq/l)	100a	89.8b	91.6b	3.58	0.03
K (Meq/l)	7.8a	4.8b	4.3b	7.63	<0.01

<sup>1</sup> γGT=Gamma-glutamyltransferase; LDH=Lactate dehydrogenase; ALT=Alanine aminotransferase; AP=Alkaline phosphatase; AST=Aspartate aminotransferase; SDH=Sorbitol dehydrogenase; CK=Creatine Kinase.

Only mean glucose, calcium, sodium, and potassium levels did not differ ( $P > 0.05$ ) between 5 sampling events that occurred between 1989 and 2000 in the Hart Mountain population. The John Day River population, sampled 4 times between 1990 and 2000, differed ( $P < 0.01$ ) in mean BUN, creatinine, BUN:creatinine ratio, total bilirubin, and calcium. Steens Mountain

also differed ( $P < 0.01$ ) in mean values for most nutritional indices (BUN, total protein, albumin, creatinine) as well as Ca and Na in 4 sampling events between 1988 and 2000. Although nutritional indices again tended to have most of the observed differences, no particular parameter tended to have higher or lower values for any given year.

Table 5. Selected parameter means through time for 3 California bighorn sheep in Oregon, 1988-2000. Means within a row with the same letter are not different (LSD,  $P = 0.05$ ).

Population Parameter	Years Sampled					<i>F</i>	<i>P&gt;F</i>
	1989	1990	1994	1995	1996		
<b>Hart Mountain</b>							
BUN (B, <i>mg/dl</i> )	9.1a	12.7c	9.4a	10.4a	11.8bc	6.21	<0.01
Σ Protein ( <i>g/dl</i> )	6.6a	7.0b		6.6a	7.2b	6.45	<0.01
Albumin ( <i>g/dl</i> )	4.0a	4.1a		3.3b	4.4c	55.64	<0.01
Creatinine ( <i>mg/dl</i> )	2.3a	2.5ab	2.6b		2.0c	29.76	<0.01
B:Creat.	4.0a	5.1b	3.7a		6.0c	12.82	<0.01
Glucose ( <i>mg/dl</i> )	153ab	163a	156a		137b	3.99	0.01
AST <sup>1</sup> ( <i>IU/l</i> )	256a	324a	183ab	203ab	157c	9.88	<0.01
Ca ( <i>mg/dl</i> )	9.7	10.4		10.2	12.7	1.64	0.18
P ( <i>mg/dl</i> )	5.1a	7.6c		5.7ab	6.4b	9.00	<0.01
Na ( <i>Meq/l</i> )	150	154		153	152	1.67	0.18
Chl ( <i>Meq/l</i> )	100ab	102a		100ab	96b	11.83	<0.01
K ( <i>Meq/l</i> )	4.8	5.4		5.0	5.3	0.65	0.58
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<b>John Day River</b>	1990	1996	1999	2000		<i>F</i>	<i>P&gt;F</i>
BUN (B, <i>mg/dl</i> )	14.4a	24.6b	24.7b	23.6b		30.39	<0.01
Σ Protein ( <i>g/dl</i> )	6.8	6.9	6.8	6.7		0.20	0.90
Albumin ( <i>g/dl</i> )	4.2	4.2	4.1	4.0		1.88	0.14
Creatinine ( <i>mg/dl</i> )	2.2a	1.7b	1.7b	1.6b		21.99	<0.01
B:Creat.	6.7a	15.0b	14.4b	14.9b		33.08	<0.01
Glucose ( <i>mg/dl</i> )	157	147	140	138		0.87	0.46
γGT <sup>1</sup> ( <i>IU/l</i> )		60.8	55.9	74.5		1.10	0.34
AST <sup>1</sup> ( <i>IU/l</i> )	386	190	311	263		2.40	0.08
Ca ( <i>mg/dl</i> )	10.7a	10.0b	10.1b	10.0b		4.96	<0.01
P ( <i>mg/dl</i> )	8.0	7.8	6.9	6.9		1.25	0.03
Na ( <i>Meq/l</i> )	151a	157b	152a	152a		4.15	0.01
Chl ( <i>Meq/l</i> )	98.2a	89.9b	97.4a	89.8b		3.35	0.02
K ( <i>Meq/l</i> )	6.0	5.7	5.1	4.8		2.44	0.07
<hr/>							
<b>Steens Mountain</b>	1988	1994	1995	2000		<i>F</i>	<i>P&gt;F</i>
BUN (B, <i>mg/dl</i> )	12.0a	13.9a	9.2b	18.3c		24.44	<0.01
Σ Protein ( <i>g/dl</i> )	7.4a		7.0b	8.4c		24.93	<0.01
Albumin ( <i>g/dl</i> )	3.3a		3.3a	5.2b		208.46	<0.01
Creatinine ( <i>mg/dl</i> )	2.2a	2.7b		2.2a		17.72	<0.01
Glucose ( <i>mg/dl</i> )	144	152		150		0.08	0.92
γGT <sup>1</sup> ( <i>IU/l</i> )	46.3		70.5	52.5		2.49	0.09
AST <sup>1</sup> ( <i>IU/l</i> )	208	182	214	207		0.36	0.79
Ca ( <i>mg/dl</i> )	11.1a		10.3b	11.8c		11.26	<0.01
P ( <i>mg/dl</i> )	6.2ab		5.4b	7.8a		4.34	0.02
Na ( <i>Meq/l</i> )	151a		152a	162b		31.08	<0.01
K ( <i>Meq/l</i> )	6.6		5.4	7.8		2.97	0.06

Because only BUN was represented in all sampling events, data were only marginally sufficient for conducting stepwise linear regression to determine the usefulness of blood chemistry as a predictor of population growth. However, 2 parameters did meet minimum entry level requirements of  $P = 0.11$  with BUN entering ( $P = 0.11$ ) the first

step and Total Protein entering ( $P = 0.12$ ).

The final model,

$$R_0 = 5.083 + 0.026(\text{BUN}) - 0.659(\Sigma\text{Protein}) + \varepsilon$$

explained 81% of the variation in population growth ( $P = 0.08$ ) with 52% explained by mean BUN value.

California bighorn sheep reference values



Table 6. Normal blood serum chemistry values for California bighorn sheep populations in Oregon, 1988-2000.

Parameter	n	Normal Range	Med.	$\bar{x}$	SE	CV
BUN (mg/dl)	423	7 - 28	14	15.8	0.34	44.2
$\Sigma$ Protein (g/dl)	344	5.8 - 8.3	6.9	6.9	0.05	12.1
Albumin (g/dl)	344	3.0 - 4.9	3.9	3.9	0.04	17.0
Globulin (mg/dl)	133	2.1 - 4.2	2.9	3.0	0.06	23.3
Albumin:Globulin	133	0.8 - 1.9	1.5	1.4	0.03	25.6
Creatinine (mg/dl)	335	1.5 - 2.8	2.2	2.1	0.02	20.6
BUN:Creatinine	334	2.7 - 17.3	6.3	8.1	0.30	59.9
CK <sup>1</sup> (IU/l)	225	235 - 2532	713	1124	99.40	133
Glucose (mg/dl)	335	89 - 199	152	149	2.40	29.6
Cholesterol (mg/dl)	110	42 - 71	58	57.6	0.90	16.1
Total Bilirubin (mg/dl)	256	0 - 0.4	0.2	0.2	0.01	69.8
Direct Bilirubin (mg/dl)	133	0 - 0.3	0.0	0.1	0.01	175
Indirect Bilirubin (mg/dl)	133	0 - 0.2	0.1	0.1	0.01	71.7
$\gamma$ GT <sup>1</sup> (IU/l)	262	23 - 123	46.0	57.4	2.10	59.2
LDH <sup>1</sup> (IU/l)	161	534 - 1160	726	801	29.50	46.7
ALT <sup>1</sup> (IU/l)	28	26 - 60	37.5	40.0	2.30	30.1
AP <sup>1</sup> (IU/l)	133	82 - 1050	221	345	25.20	84.1
AST <sup>1</sup> (IU/l)	423	117 - 545	193	243	8.30	70.4
SDH <sup>1</sup> (IU/l)	123	7.1 - 122	24.0	40.1	5.70	157
Uric Acid (mg/dl)	82	0.1 - 0.6	0.2	0.2	0.02	87.6
Triglycerides (mg/dl)	82	71 - 422	212	212	11.10	47.4
Calcium (mg/dl)	344	8.9 - 12.4	10.4	10.6	0.20	35.9
Phosphorous (mg/dl)	344	4.0 - 9.6	6.5	6.7	0.10	32.8
Sodium (Meq/l)	344	146 - 164	154	153	0.60	6.7
Chloride (Meq/l)	344	88 - 105	99	98	0.50	9.1
Potassium (Meq/l)	344	4.0 - 7.2	5.0	5.4	0.10	38.0
Magnesium (mg/dl)	211	2.3 - 4.5	3.2	3.2	0.05	21.5
Iron (mCg/dl)	82	142 - 263	201	200	4.90	22.2
$\Sigma$ CO <sub>2</sub> (Meq/l)	123	1.6 - 14.2	5.0	6.4	0.50	86.9

<sup>1</sup> CK= Creatine Kinase;  $\gamma$ GT=Gamma-glutamyltransferase; LDH=Lactate dehydrogenase; ALT=Alanine aminotransferase; AP=Alkaline phosphatase; AST=Aspartate aminotransferase; SDH=Sorbitol dehydrogenase.

were completed for 29 parameters commonly evaluated with standard serum blood chemistry analyses (Table 6). Reference values are based on sample sizes ranging from 28 to 423 as a result of variability in specific parameters reported by different labs. However, most (79%) are based on > 100 samples with 55% of the reference values based on > 200 individual samples. Overall variability estimates (CV) for parameter values ranged from a low of 6.7% (very little variation across all samples) for sodium to a high of 175% (highly variable across samples) for direct

bilirubin. Generally, diagnostic indices appeared to be more variable than either nutritional indices or mineral values.

## DISCUSSION

Blood characteristics have been explored as potential indices to nutrition in wildlife for a number of years. Blood urea nitrogen (BUN) is most widely used in ruminants because it appears to be directly related to protein intake and is relatively unaffected by capture and handling stress (Harder and Kirkpatrick 1996). However, BUN may have diurnal cycles, may be affected by

energy intake (Harder and Kirkpatrick 1996), and is affected by catabolic processes during prolonged periods of protein intake below that required for maintenance and seasonal fluctuation in diet (Hebert 1978, Harder and Kirkpatrick 1996). Regardless, values of blood chemistry constituents, primarily BUN, protein, cholesterol, and alkaline phosphatase have been suggested as indicative of relative nutritional differences in bighorns (Franzmann and Thorne 1970, Franzmann 1971a, Hebert 1978, Borjesson et al. 2000).

Five blood constituents commonly associated with nutrition (BUN,  $\Sigma$  protein, albumin, creatinine, and BUN:creatinine ratio: Harder and Kirkpatrick 1996, Chin 1997) differed between male and female California bighorns in Oregon. Of these, BUN and BUN:creatinine ratios were higher for males than females (Table 2). Borjesson et al. (2000) also report some differences with males having higher BUN, alkaline phosphatase, and glucose values than females. It is well documented that male and female bighorns typically segregate for most of the year (Wishart 1978, Bleich et al. 1997). Bleich et al. (1997) demonstrated that sexual segregation in desert bighorns led to dietary differences that resulted in differences in available nutrition between rams and ewes. Sexual segregation has been observed in California bighorn sheep in Oregon as well (Kornet 1978, Payer 1992). Thus, differences we observed between rams and ewes likely represent slight differences in nutrition available to the sexes. However, the large sample size available for our study resulted in high statistical power for this comparison. Although we feel these differences represent real nutritional differences, we also believe the observed differences between rams and ewes were not sufficient to result in differential impacts to the ram and ewe segments in populations we

studied.

Similar to the comparison between sexes, observed differences between age class (lamb, yearling, adult) of California bighorns in Oregon tended to occur for constituents commonly used to index nutrition (BUN, protein, globulin, creatinine, BUN:creatinine, Na, Chl: Table 3). Further, lambs tended to differ from either yearlings or adults. Similar to Borjesson et al. (2000) and Kock et al. (1987b), we found lambs had lower total protein and lower globulin than either yearlings or adults. Kock et al. (1987b) also found lower BUN in lambs as well, whereas we found higher BUN levels in lambs compared to adults and yearlings. Lower total globulin levels in lambs are primarily a result of low immunoglobulin levels resulting from a developing immune system in lambs Borjesson et al. (2000). Total protein and BUN are correlated with diet (Hebert 1978, Harder and Kirkpatrick 1996). BUN also is affected by environmental factors, temporal, and diurnal cycles (Franzmann 1972, Hebert 1978, Harder and Kirkpatrick 1996). Although we found higher mean BUN values in lambs (19.3) than adults (15.7) and yearlings (16.1), we also found mean BUN values much higher than many other reports for wild bighorns (Franzmann and Thorne 1970, Davies 1976, Borjesson et al. 2000). It is possible that the higher BUN values we observed in bighorn lambs are a result of better diets on ewe/lamb ranges in Oregon compared to most other studies. Because of our large sample size, we were able to compare values in yearlings as a separate age class. Where differences occurred between age classes, the trend was for similarity in yearlings and adults. Thus we conclude that yearlings are not too different from adults physiologically as measured by blood chemistry.

Our unusually large and diverse sample

size (n=423) allowed us to compare blood constituents across distinct populations within several sampling years. In addition to finding differences in indices of diet, we also found differences in minerals. During 1994 and 1995, most nutritional indices were generally higher in the Leslie Gulch population compared to the Hart Mountain and Steens Mountain populations. During 2000, most of the same indices were higher in the John Day River population compared to either the Steens Mountain, or Nevada populations. Of those studies we reviewed, few report comparisons between distinct populations or within a population through time (Franzmann and Thorne 1970, Franzmann 1971a, Franzmann 1971c, Hebert 1978). However, many authors indicate that ungulate diet quality is reflected in blood chemistry parameters (Franzmann and Thorne 1970, Franzmann 1971c, Franzmann 1972, Hebert 1978, McDonald et al. 1981, Harder and Kirkpatrick 1996). It is likely that differences we observed reflect the relative differences in dietary quality available to the populations (Hebert 1978). The trend for populations associated with river corridors (Leslie Gulch and John Day River) suggests these habitats may provide better nutrition to California bighorns in Oregon.

The duration of our sampling effort also allowed us to compare changes in blood serum chemistry within 3 populations across years. Comparisons indicated significant variation through time, primarily in parameters used as indices of nutrition. Although there were exceptions (1990 on Hart Mountain, 1994 on Steens Mountain), most differences suggest nutrition was likely better for populations sampled during recent years (1995-2000) compared to earlier sampling efforts (1988-1994). As available nutrition improves, indices measurable in bighorn sheep blood serum chemistry also

improve (Franzmann and Thorne 1970, Hebert 1978). Thus, differences we observed through time in this analysis likely reflect the temporal dynamics of nutrition available to the populations we sampled. Additionally, stepwise linear regression suggested the possibility of a relationship between BUN and total protein with population growth. Sample size was limited for this analysis, however, and even though the model explained 81% of the variation, it was not significant ( $P > 0.10$ ). Further development of this data set through time may increase the predictive strength of this relationship.

Normal values and ranges for most parameters we measured are similar to other published reports (Table 7) for Rocky Mountain, California, and desert bighorn sheep (Davies 1976, Bottrell et al. 1978, Puls 1994, Borjesson et al. 2000), thimhorn sheep (Bottrell et al. 1978, Foreyt et al. 1983) and domestic sheep (Puls 1994). The greatest variation in central measures (mean or median) across studies we found was for BUN with glucose and cholesterol also varying considerably. However, our study mean of 15.8 for BUN, was well within the range reported by other authors (8.7 – 37.0). We feel the wide range of values reported for BUN, as well as glucose and cholesterol reflects the wide range of habitats and nutritional planes available to bighorn sheep.

The values we report for California bighorns provide an additional diagnostic tool for management and research. Blood chemistry results can be used when designing monitoring schedules for radio-marked sheep. Individuals with measured values outside normal ranges for specific diagnostic parameters may warrant additional monitoring efforts. Additionally, serum blood chemistry profiles may be helpful in determining cause of death, or

Table 7. Normal blood serum chemistry values from Oregon California bighorn sheep and other published reports.

Parameter	This Study		Bottrell et al. 1978 ( $\bar{x}$ )		Davies 1976 Rocky		Borjesson et al. 2000 Desert		Puls 1994	
	Range	Med	Rocky	Calif.	$\bar{x}$	Range	Range	Med	Bighorn	Domestic
BUN (mg/dl)	7.0-28.0	14.0	37.0	33.8	14.0	10-19	5.0-28.0	14.0	8.0-25.0	8.0-20.0
$\Sigma$ Protein (g/dl)	5.8-8.3	6.9	7.1	6.2	6.5	5.6-7.7	6.0-9.3	7.4	5.0-7.0	6.0-7.9
Albumin (g/dl)	3.0-4.9	3.9	3.8	3.5	---	---	2.8-3.7	3.3	---	2.4-3.0
Globulin (mg/dl)	2.1-4.2	2.9	3.3	2.7	2.9	2.2-3.8	2.8-6.1	4.0	---	---
Albumin:Globulin	0.8-1.9	1.5	---	---	---	---	0.5-1.2	0.9	---	---
Creatinine (mg/dl)	1.5-2.8	2.2	---	---	2.6	2.1-3.1	1.6-2.6	2.0	---	1.0-2.9
BUN:Creatinine	2.7-17.3	6.3	---	---	---	---	2.5-14.8	7.0	---	---
CK (IU/l)	235-2532	713	---	---	135	49-385	175-2300	392	74-114	8-13
Glucose (mg/dl)	89-199	152	141	129	140	65-240	95-185	151	80-150	50-80
Cholesterol (mg/dl)	42-71	58	52	38	70	51-85	---	---	35-90	52-76
$\Sigma$ Biliruben (mg/dl)	0.0-0.4	0.2	---	---	0.5	0.4-0.8	0.0-0.1	0.1	---	0.1-0.4
D. Biliruben (mg/dl)	0.0-0.3	0.0	---	---	---	---	0.0-0.0	0.0	---	---
I. Biliruben (mg/dl)	0.0-0.2	0.1	---	---	---	---	0.0-0.1	0.1	---	---
$\gamma$ GT <sup>1</sup> (IU/l)	23.0-123	46.0	---	---	---	---	20.0-130	36.0	---	18.0-40.0
LDH <sup>1</sup> (IU/l)	534-1160	726	---	---	344	200-555	409-788	534	268-593	60-111
ALT <sup>1</sup> (mg/l)	26-60	37.5	---	---	110	74-145	---	---	109-141	10.0-12.0
AP <sup>1</sup> (IU/l)	82-1050	221	276	356	82	33-185	73-575	166	---	70-263
AST <sup>1</sup> (IU/l)	117-545	193	---	---	212	115-525	78-312	137	130-250	68-200
SDH <sup>1</sup> (IU/l)	7.2-122	24.0	---	---	---	---	---	---	---	6.0-30.0
Calcium (mg/dl)	8.9-12.4	10.4	10.1	9.8	11.5	10.2-14.0	9.3-11.5	10.3	8.0-10.0	11.0-13.0
Phosphorous (mg/dl)	4.0-9.6	6.5	7.8	7.4	5.9	2.9-8.4	4.0-9.3	6.5	---	5.0-7.0
Sodium (Meq/l)	146-164	154	153	151	143	141-148	145-160	153	---	139-152
Chloride (Meq/l)	88-105	99	---	---	---	---	89-107	99	---	98-110
Potassium (Meq/l)	4.0-7.2	5.0	5.6	5.3	4.9	3.7-6.7	3.8-6.3	4.7	---	3.9-5.4
Magnesium (mg/dl)	2.3-4.5	3.2	---	---	---	---	---	---	0.8-3.0	2.0-3.5
Iron (mCg/dl)	142-263	201	---	---	---	---	---	---	---	166-222

<sup>1</sup> CK=Creatine Kinase;  $\gamma$ GT=Gamma-glutamyltransferase; LDH=Lactate dehydrogenase; ALT=Alanine aminotransferase; AP=Alkaline phosphatase; AST=Aspirate aminotransferase; SDH=Sorbitol dehydrogenase.

predisposition to outside mortality factors. For example, adult ewe #00-06 captured in Nevada during winter 2000 died during transport to Oregon. Blood chemistry results revealed that values for BUN,  $\lambda$ GT, AST, and SDH were above the normal range we observed. Subsequent necropsy revealed she died of severe cancer to the liver.

The data we report here are not without limitations. Over the duration of our sampling effort, results were obtained from 4 different laboratories without blind samples to quantify potential variation. Although most labs utilize comparable techniques and equipment, variation between labs may produce bias, thus decreasing the power (Probability of a Type II error, or  $\beta$ ) of our comparisons. However,

we feel that differences we observed between groups (age, sex, population, or temporally) were real due to the magnitude of the differences and the extremely low statistical probabilities observed for the comparisons. Our inference also is constrained by the observational nature of the data set. This is especially true for the normal values and ranges we report which do not describe critical levels. Further experimental work is required to determine these critical values, and to determine the effects of being outside the range of critical values for both individuals and populations.

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