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Blood Mineral Concentrations in the Endangered Huemul Deer (*Hippocamelus bisulcus*) from Chilean Patagonia

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ABSTRACT: Concentrations of calcium, magnesium, copper, zinc, and selenium were measured in plasma from 11 huemul (*Hippocamelus bisulcus*) from Chilean Patagonia. Except for zinc and copper, concentrations of these minerals were similar to those of other deer species.

Balance alterations of one or more minerals in mammals, especially deficiencies, affect fertility, immune response, and survival of young (Wilson and Grace 2001). In grazing animals, mineral deficiencies are attributed to low food availability and to soils containing insufficient minerals that result in lower forage mineral concentrations (Ellison 2002). In endangered species, the importance of mineral deficiencies in dynamics and persistence of populations has not been addressed in depth. This information could be crucial for effective conservation actions to support population recovery of ruminant ungulates.

The huemul (*Hippocamelus bisulcus*), endemic to southern Chile and Argentina, is the most endangered Neotropical deer. Huemul currently inhabit mountainous ecosystems dominated by southern beech (*Nothofagus* spp.) forests (Corti et al. 2010). Those environments are formed on volcanic soils with pH tending to acidity (Veblen and Schlegel 1982), and forages are commonly deficient in selenium (Se), failing to meet livestock requirements (Gissel-Nielsen et al. 1984). Currently, the huemul population consists of <2,000 individuals, distributed in fragmented groups throughout the remaining 50% of its original distribution (Corti et al. 2011). It is unknown whether the environments within their current distribution meet optimal nutritional requirements for huemul, particularly regarding forage mineral levels. There is evidence that in some huemul populations, survival of offspring is low, resulting from high mortality from mostly unknown causes (Corti et al. 2010). The lack of trace minerals, including zinc (Zn), could reduce an animal's immune response, increasing vulnerability to diseases (Rosa et al. 2008). We measured plasma concentrations of calcium (Ca), magnesium (Mg), copper (Cu), Zn, and Se in huemul and compared values with those reported for other deer species.

The study was conducted in the Cochrane Lake area (47°12′S, 72°30′W), Chilean Patagonia. Blood samples were obtained by jugular venipuncture (Corti et al. 2010) from eight adult females and three adult males captured during autumn and winter 2009. Blood samples were placed into tubes with heparin. Plasma was obtained by centrifugation at 1,100 × G for 15 min. in a portable centrifuge (Mobilespine, Vulcan Technologies, Grandview, Missouri, USA) and stored at −10 C until analysis.

Determination of Ca, Mg, Cu, Zn, and Se was performed using an atomic absorption spectrophotometer (Series AA Solaar, Thermo Fisher Scientific Inc., Waltham, Massachusetts, USA) at the Universidad Austral de Chile. The measurement of Ca and Mg was carried out at 422.7 nm and 285.2 nm, respectively, after plasma was diluted in 1:10 lanthanum chloride (LaCl) solution. Measurements of Cu and Zn were performed at 324.8 nm and 213.9 nm respectively, following dilution of plasma samples, 1:5 for Cu, and 1:10

TABLE 1. Mean±SD, range, median, and interval calculated (mean±2 SD) of plasma concentrations of Ca, Mg, Cu, Zn, and Se in 11 adult huemul (*Hippocamelus bisulcus*) at Lago Cochrane area, Chilean Patagonia. Autumn and winter 2009.

Mineral	Mean±SD	Range	Median	Interval calculated
Ca (mmol/L)	2.16±0.32	1.52–2.52	2.23	1.52–2.80
Mg (mmol/L)	1.50±0.19	1.24–1.73	1.48	1.12–1.88
Cu (μmol/L)	22.10±8.81	14.0–37.0	19.0	4.48–39.72
Zn (μmol/L)	6.27±2.24	2.0–9.0	7.0	1.79–10.75
Se (nmol/L)	680.3±468.4	215.2–1355.1	633.2	0.00–1617.1

for Zn, in deionized distilled water. Se determination was carried out at 196 nm with a continuous vapor generator device (Hydride Vapour System Model VP100, Thermo Fisher Scientific), after acid digestion of the sample following Muñiz-Naveiro et al. (2005). Because not all authors reported data ranges, we calculated a reference interval for each mineral level (mean±2 SD) to standardize the information to be compared with huemul data.

Average mineral plasma values (Table 1) are the first reported for huemul. The methodology used to measure Se in plasma did not allow quantifying concentration in four animals, where results were <0.011 nmol/L. The average value for calcemia was similar to those reported for other deer, falling within intervals calculated for free-ranging red deer (*Cervus elaphus*) from Norway and fallow deer (*Dama dama*) from Slovenia (Table 2). Similarity of calcemia values in these three deer species may also be attributed to strong hormonal regulation of Ca (Suttle 2010).

Mean magnesemia found in huemul exceeded values reported for captive and free-ranging red deer and for fallow deer (Table 2). Mg metabolic balance has no hormonal regulation, and magnesemia constitutes a reflection of what an animal is able to ingest and absorb on a daily basis.

Mean huemul cupremia was greater than the interval calculated for free-living and captive red deer (Table 2). In domestic ruminants, Cu metabolism can be negatively affected by the presence of other minerals, such as molybdenum and sulphur, and this seems to be the case in free-living deer reported to date (Frank 2004).

Average plasma Zn concentration of huemul was within the reference intervals calculated for free-living and captive red deer but lower than those reported for red deer (Table 2). There are no exclusively recognized body reserves for this mineral; therefore, tissues are not capable of maintaining normal plasma Zn concentrations for long periods under conditions of sustained nutritional deficiency (Suttle 2010). With the exception of four huemul, plasma Se values fell within the interval reported for white-tailed deer (*Odocoileus virginianus*; Table 2). The high data dispersion observed suggested a heterogeneous Se intake, not discounting Se deficiencies.

Overall, plasma concentrations of minerals obtained from huemul were within reference intervals calculated for other deer, except for Cu and Mg. While voluntary intake and selectivity of huemul are important factors, quality and quantity of available food fluctuate seasonally throughout the year. Consequently, it is important to know if current habitat restrictions are also relevant in limiting some critical nutritional mineral requirements of huemul. The low plasma Zn concentration we found might be associated with high fawn mortality reported in this population, where a high percentage of causes for mortality are unknown (Corti et al. 2010). Presence of bovine viral diarrhea virus in this population (Corti et al. 2013) is relevant because low Zn

TABLE 2. Mean±SD, and calculated interval (mean±2 SD) of plasma concentration of Ca, Mg, Cu, Zn, and Se from three species of deer.

Mineral	Free-ranging red deer (*Cervus elaphus*)[a] (*n*=183)	Captive red deer[b] (*n*=30)	Farmed fallow deer (*Dama dama*) (*n*=4)	White-tailed deer (*Odocoileus virginianus*)[d] (*n*=174)
Ca (mmol/L)	1.94±0.37	2.12±0.26	1.80±0.20	—
	1.20–2.68	1.60–2.64	1.40–2.20	
Mg (mmol/L)	0.48±0.11	0.91±0.20	0.90±0.10	—
	0.26–0.7	0.51–1.31	0.70–1.10	
Cu (μmol/L)	13.0±3.60	9.86±1.64	—	—
	6.10–20.20	6.58–13.14		
Zn (μmol/L)	8.23±1.79	16.97±9.05	—	—
	4.65–11.81	0.00–35.07		
Se (nmol/L)	—	—	—	645.8±667.4
				0.00–1980.6

[a] Rosef et al. 2004.

[b] Padilla et al. 2000.

[c] Vengušt et al. 2006.

[d] McDowell et al. 1995.

concentrations could weaken the immune system. We recommend expanding this database to other huemul populations, including mineral levels in forages along with seasonal samples, to develop reference intervals for huemul mineral plasma concentrations across their range.

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