

# ASCORBIC ACID, VITAMIN E, VITAMIN A, AND TRACE ELEMENTS IN SERUM OF ZOO CROCODILIANS

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## Abstract

A potential clinical case of ulcerative gingivitis in a male gharial (*Gavialis gangeticus*) initiated an investigation to determine if there was adequate ascorbic acid in the diet of crocodilians at the San Diego Zoo (SDZ), and San Diego Zoo Safari Park (SP). Reptiles can synthesize ascorbic acid and classic deficiency is rarely seen. The objective of this summary was to compare serum trace-mineral and vitamin concentrations of zoo-housed crocodilians to wild and farmed American alligators.<sup>2-6</sup> For the 20 individuals from four species (American alligators, *Alligator mississippiensis*; Chinese alligators, *Alligator sinensis*; Johnston's crocodiles, *Crocodylus johnsoni*; and gharials), serum nutrient concentrations averaged 11.06 mg Ca/dl, 91.66 µg Cu/dl, 36.88 µg Fe/dl, 2.86 mg Mg/dl, 4.03 mg P/dl, 3.97 mEq K/L, 153.88 mEq Ma/L, 41.43 mg Zn/dl, 0.50 µg vitamin A/dl, 46.70 µg vitamin E /dl, and 0.66 mg ascorbic acid /dl. No additional deficiencies or toxicities have been observed, although some values were above and below those of wild and farmed American alligators.

## Introduction

A lack of reference information exists regarding the serum nutrient concentrations of crocodilians. The current report was initiated after a potential case of ulcerative gingivitis in a gharial (*Gavialis gangeticus*) at the San Diego Zoo (SDZ), and a subsequently-discovered lack of serum ascorbic acid reference ranges in crocodilians. In crocodilian species, cases of ulcerative gingivitis have been associated with ascorbic acid deficiency.<sup>4</sup> Reptiles have the capacity to synthesize ascorbic acid and classic deficiency is rarely seen.<sup>2</sup> However, individuals can become deficient with continuous stress associated with captivity or sickness.<sup>1,3</sup> During times of combined stress and infection, the demand for ascorbic acid is increased and endogenous production cannot keep up with demand.<sup>3</sup> To prevent deficiencies it is common to supplement the diet of farmed crocodilians with ascorbic acid (1000 mg/kg DM).<sup>4</sup> The goal of the current report is to start developing serum nutrient reference ranges for zoo crocodilians.

## Methods

Upon arrival into quarantine or during examinations at the SDZ or San Diego Zoo Safari Park (SP), blood samples were collected from American alligators (*Alligator mississippiensis*, 0.0.2, 10-April-2013), Chinese alligators (*Alligator sinensis*, 0.2, 2-July-2013), Johnston's crocodiles (*Crocodylus johnsoni*, 0.5, 10-May-2012) and gharials (*Gavialis gangeticus*), 8.2.1, 23-

February-2011 and 16-May-2012). Animals were fed frozen, thawed whole prey (mice or fish, with the fish supplemented with thiamin and vitamin E), and in the case of the American alligators, were also fed Mazuri Fish Analog 50/10 Mix (PMI Nutrition Interational LLC Saint Louis, MO 63108).

Blood was collected by venipuncture into an acid-washed blood collection tubes and centrifuged. Serum was harvested and shipped cool overnight, for analysis of trace mineral concentrations (Ca, Cu, Fe, Mg, P, K, Na, Zn), vitamin A, and vitamin E at California Animal Health and Food Safety Laboratory System (University of California, Davis, Davis, CA 95617), and ascorbic acid at Diagnostic Center for Population and Animal Health (Michigan State University, Lansing, MI 48910-8104). Individual animals with multiple samples were averaged. Serum nutrients were compared to reference values from wild and farmed American alligators.<sup>2-6</sup> Because no crocodylian reference range for serum ascorbic acid was found in the literature, concentrations were compared among the three species in this report and poultry.<sup>7</sup>

## Results

American alligator (Table 1) serum copper and sodium concentrations were similar to wild and farmed American alligators. Serum calcium, iron, magnesium, phosphorus, potassium, vitamin A and vitamin E concentrations were below the range of wild and farmed American alligators. Serum zinc concentration and the calcium to phosphorus ratio were above the corresponding ranges for wild or farmed American alligators.

Chinese alligator (Table 2) serum magnesium and potassium concentrations were similar to those in American alligators. Serum calcium, iron, phosphorus, sodium, zinc, and vitamin E concentrations were below the range of wild and farmed American alligators. Serum copper concentration was above the range of wild or farmed American alligators.

Johnston's crocodile (Table 3) serum calcium, magnesium and potassium concentrations were similar to those of American alligators. Serum iron, phosphorus, zinc, and vitamin E concentrations were below the range of wild and farmed American alligators. Serum copper and sodium concentrations were above the range of wild and farmed American alligators. The calcium to phosphorus ratio was substantially greater for Johnston's crocodiles than for wild or farmed American alligators.<sup>6</sup>

Gharial (Table 4) serum iron, magnesium, potassium, and zinc concentrations were similar to those of American alligators. Serum calcium and phosphorus concentrations were slightly below the range. Serum copper and sodium concentrations were above the range. Calcium to phosphorus ratio was greater than that of wild or farmed American alligators.

Serum ascorbic acid concentrations were compared among the crocodylian species sampled and poultry due to a lack of crocodylian data from the literature.<sup>7</sup> American alligators had the lowest ascorbic acid concentration, followed by gharials, then Johnston's crocodiles (Tables 1, 3, 4). American alligators, gharials, and Johnston's crocodiles had lower serum ascorbic acid concentration than poultry.<sup>7</sup> Ascorbic acid was not analyzed on serum collected from Chinese alligators.

## Conclusions

No further deficiencies or toxicities have been observed despite some mineral and vitamin concentrations being above or below the referenced ranges. Continuing monitoring of health and feed will allow for any diet revisions as new data is obtained. The data provides preliminary data to use as a benchmark for future diet evaluations.

## Literature cited

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**Table 1.** Serum mineral and vitamin concentrations of zoo American alligators (*Alligator mississippiensis*).

Nutrient	n <sup>1</sup>	Mean	SD	Range		Reference values	
				Low	High	Wild	Farmed
Ca, mg/dl	2	10.75	3.54	10.50	11.00	12.51 <sup>6</sup>	11.50 <sup>6</sup>
Cu, µg/dl	2	63.25	0.01	62.50	64.00	76.00 <sup>6</sup>	60.00 <sup>6</sup>
Fe, µg/dl	1	24.00	NA <sup>2</sup>	24.00	24.00	53.00 <sup>6</sup>	54.00 <sup>6</sup>
Mg, mg/dl	2	2.55	2.12	2.40	2.70	3.21 <sup>6</sup>	2.58 <sup>6</sup>
P, mg/dl	2	3.70	4.24	3.40	4.00	6.17 <sup>5</sup>	5.73 <sup>5</sup>
K, mEq/L	2	3.53	0.11	3.45	3.60	5.42 <sup>5</sup>	3.71 <sup>5</sup>
Na, mEq/L	2	152.50	10.61	145.00	160.00	154.62 <sup>4</sup>	144.20 <sup>5</sup>
Z, mg/dl	2	53.00	0.37	27.00	79.00	44.00 <sup>6</sup>	42.00 <sup>6</sup>
Ca:P ratio	2	2.92	0.24	2.75	3.09	2.03 <sup>4,6</sup>	2.01 <sup>5,6</sup>
Vit E, µg/dl	1	45.50	NA	45.50	45.50	53.60 <sup>6</sup>	52.50 <sup>6</sup>
Vit. A, µg/dl	2	0.50	NA	0.50	0.50	0.90 <sup>7</sup>	0.85 <sup>7</sup>
Ascorbic Acid, mg/dl	2	0.60	0.12	0.51	0.68	NA	1—2 <sup>3</sup>

<sup>1</sup>Two animals sampled for serum minerals, vitamin E, vitamin A, and ascorbic acid, collected in 2013. Multiple samples from any given animal were averaged.

<sup>2</sup>Not available

<sup>3</sup>Compared to poultry serum concentrations (Puls, R. 1994. Vitamin levels in animal health. Sherpa International. Pp. 78).

<sup>4</sup>Honeyfield, D.C., J.P. Ross, D.A. Carbonneau, S.P. Terrel, A.R. Woodward, T.R. Schoeb, H.F. Perceval, and J.P. Hinterkopf. 2008. Pathology, physiologic parameters, tissue contaminants, and tissue thiamine in morbid and healthy central Florida adult American alligators (*Alligator mississippiensis*). J. Wild. Dis. 44:280-294.

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**Table 2.** Serum mineral and vitamin concentrations of zoo Chinese alligators (*Alligator sinensis*).

Nutrient	n <sup>1</sup>	Mean	Reference values	
			Wild	Farmed
Ca, mg/dl	1	10.00	12.51 <sup>4</sup>	11.50 <sup>4</sup>
Cu, µg/dl	1	120.00	76.00 <sup>4</sup>	60.00 <sup>4</sup>
Fe, µg/dl	1	29.00	53.00 <sup>4</sup>	54.00 <sup>4</sup>
Mg, mg/dl	1	2.80	3.21 <sup>4</sup>	2.58 <sup>4</sup>
P, mg/dl	1	4.20	6.17 <sup>2</sup>	5.73 <sup>3</sup>
K, mEq/L	1	3.80	5.42 <sup>2</sup>	3.71 <sup>3</sup>
Na, mEq/L	1	140.00	154.62 <sup>2</sup>	144.20 <sup>3</sup>
Z, mg/dl	1	34.00	44.00 <sup>4</sup>	42.00 <sup>4</sup>
Ca:P ratio	1	2.38	2.03 <sup>2,4</sup>	2.01 <sup>3,4</sup>
Vit E, µg/dl	1	52.00	53.60 <sup>4</sup>	52.50 <sup>4</sup>

<sup>1</sup>Two animals total were sampled: samples from one animal were analyzed for serum minerals; samples from the other animal were analyzed for vitamin E; collected in 2012.

<sup>2</sup>Honeyfield, D.C., J.P. Ross, D.A. Carbonneau, S.P. Terrel, A.R. Woodward, T.R. Schoeb, H.F. Perceval, and J.P. Hinterkopf. 2008. Pathology, physiologic parameters, tissue contaminants, and tissue thiamine in morbid and healthy central Florida adult American alligators (*Alligator mississippiensis*). J. Wild. Dis. 44:280-294.

<sup>3</sup>Huchzermeyer, F.W. 2003 Crocodiles Biology, Husbandry and Diseases. CABI Publishing, Wallingford, UK.

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**Table 3.** Serum mineral and vitamin concentrations of zoo Johnston's crocodiles (*Crocodylus johnsoni*).

Nutrient	n <sup>1</sup>	Mean	SD	Range		Reference values	
				Low	High	Wild	Farmed
Ca, mg/dl	5	12.20	13.04	11.00	14.00	12.51 <sup>6</sup>	11.50 <sup>6</sup>
Cu, µg/dl	5	86.80	0.10	73.00	98.00	76.00 <sup>6</sup>	60.00 <sup>6</sup>
Fe, µg/dl	5	36.00	0.06	26.00	40.00	53.00 <sup>6</sup>	54.00 <sup>6</sup>
Mg, mg/dl	5	2.68	2.49	2.50	3.10	3.21 <sup>6</sup>	2.58 <sup>6</sup>
P, mg/dl	5	3.84	10.50	2.80	5.10	6.17 <sup>4</sup>	5.73 <sup>5</sup>
K, mEq/L	5	4.22	0.35	3.60	4.40	5.42 <sup>4</sup>	3.71 <sup>5</sup>
Na, mEq/L	5	162.00	4.47	160.00	170.00	154.6 <sup>4</sup>	144.20 <sup>5</sup>
Z, mg/dl	5	34.00	0.15	21.00	57.00	44.00 <sup>4</sup>	42.00 <sup>6</sup>
Ca:P ratio	5	3.34	0.87	2.35	4.64	2.03 <sup>4,6</sup>	2.01 <sup>5,6</sup>
Vit E, µg/dl	5	42.60	1.28	32.00	64.00	53.60 <sup>6</sup>	52.50 <sup>6</sup>
Ascorbic Acid, mg/d	5	0.78	0.04	0.73	0.84	NA <sup>2</sup>	1—2 <sup>3</sup>

<sup>1</sup>Blood samples were collected from five animals once in 2012 for analysis of minerals, vitamin E, and ascorbic acid.

<sup>2</sup>Not available.

<sup>3</sup>Compared to poultry serum concentrations (Puls, R. 1994. Vitamin levels in animal health. Sherpa International. Pp. 78).

<sup>4</sup>Honeyfield, D.C., J.P. Ross, D.A. Carbonneau, S.P. Terrel, A.R. Woodward, T.R. Schoeb, H.F. Perceval, and J.P. Hinterkopf. 2008. Pathology, physiologic parameters, tissue contaminants, and tissue thiamine in morbid and healthy central Florida adult American alligators (*Alligator mississippiensis*). J. Wild. Dis. 44:280-294.

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**Table 4.** Serum mineral and vitamin concentrations of zoo gharials (*Gavialis gangeticus*).

Nutrient	n <sup>1</sup>	Mean	SD	Range		Reference values	
				Low	High	Wild	Farmed
Ca, mg/dl	11	11.28	9.81	9.80	13.00	12.51 <sup>6</sup>	11.50 <sup>6</sup>
Cu, µg/dl	11	96.60	0.15	73.00	120.00	76.00 <sup>6</sup>	60.00 <sup>6</sup>
Fe, µg/dl	11	58.50	0.24	15.00	81.00	53.00 <sup>6</sup>	54.00 <sup>6</sup>
Mg, mg/dl	11	3.40	4.64	2.70	4.00	3.21 <sup>6</sup>	2.58 <sup>6</sup>
P, mg/dl	11	4.36	14.19	2.40	6.50	6.17 <sup>5</sup>	5.73 <sup>5</sup>
K, mEq/L	11	4.32	0.30	3.80	4.80	5.42 <sup>5</sup>	3.71 <sup>5</sup>
Na, mEq/L	11	161.00	5.68	150.00	170.00	154.62 <sup>5</sup>	144.20 <sup>5</sup>
Z, mg/dl	11	44.70	0.17	32.00	84.00	44.00 <sup>6</sup>	42.00 <sup>6</sup>
Ca:P ratio	11	2.93	1.00	1.69	4.62	2.03 <sup>4,6</sup>	2.01 <sup>5,6</sup>
Ascorbic Acid, mg/dl	11	0.61	0.07	0.49	0.71	NA <sup>2</sup>	1—2 <sup>3</sup>

<sup>1</sup>Blood samples were collected from five animals once in 2012 for analysis of minerals, vitamin E, and ascorbic acid.

<sup>2</sup>Not available.

<sup>3</sup>Compared to poultry serum concentrations (Puls, R. 1994. Vitamin levels in animal health. Sherpa International. Pp. 78).

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