Journal of Zoo and Wildlife Medicine 36(2): 188–191, 2005

VITAMIN D STATUS OF WILD RICORD'S IGUANAS (*CYCLURA RICORDII*) AND CAPTIVE AND WILD RHINOCEROS IGUANAS (*CYCLURA CORNUTA CORNUTA*) IN THE DOMINICAN REPUBLIC

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Abstract: Calcidiol (25-hydroxyvitamin D) values are reported for 22 wild Ricord's iguanas (*Cyclura ricordii*) and seven wild rhinoceros iguanas (*Cyclura cornuta cornuta*). Calcitriol (1,25-hydroxyvitamin D) values are reported for 12 wild Ricord's iguanas and seven wild rhinoceros iguanas. These animals were captured as part of a larger health assessment study being conducted on Ricord's iguanas in Isla Cabritos National Park, Dominican Republic. A total of 13 captive rhinoceros iguanas held outdoors at Parque Zoológico Nacional were also sampled for comparison. Mean concentrations of 25-hydroxyvitamin D were 554 nmol/L (222 ng/ml) with a range of 250–1,118 nmol/L (100–448 ng/ml) for wild Ricord's iguanas, 332 nmol/L (133 ng/ml) with a range of 260–369 nmol/L (104–148 ng/ml) for wild rhinoceros iguanas, and 317 nmol/L (127 ng/ml) with a range of 220–519 nmol/L (88–208 ng/ml) for captive rhinoceros iguanas. On the basis of these results, serum concentrations of at least 325 nmol/L (130 ng/ml) for 25-hydroxyvitamin D should be considered normal for healthy Ricord's and rhinoceros iguanas.

Key words: Vitamin D, Ricord's iguana, rhinoceros iguana, *Cyclura*.

INTRODUCTION

Hypocalcemia and hypovitaminosis D are commonly encountered medical problems in captive animals, especially in herbivorous reptiles, which do not have access to sunlight.[8,10–12] The role of sunlight in Vitamin D synthesis is well described. Vitamin D_3 (cholecalciferol) is produced in the skin during exposure to sunlight, specifically ultraviolet (UV) light in the range of 290–315 nm (UVB), in a temperature-dependent isomerization.[8] Circulating Vitamin D_3 is hydroxylated in the liver to 25-hydroxyvitamin D (calcidiol).[7,12] This metabolite is typically very plentiful and has a half-life of 3–4 wk in humans. 25-Hydroxyvitamin D is hydroxylated in the kidney to 1,25-hydroxyvitamin D (calcitriol) and is the active form of the hormone. This step is typically very tightly regulated.[7,12] The half-life of 1,25-hydroxyvitamin D is measured in hours in humans (*Homo sapiens*)[12] but has not been characterized in reptiles. Vitamin D status in animals is usually characterized by determining the levels of 25-hydroxyvitamin D because of the relatively higher levels and longer half-life of this form of the vitamin.[12] Levels of 1,25-hydroxyvitamin D are not typically used to determine vitamin D deficiencies in animals because of the relatively short half-life and tight regulation of the form. However, some animals are known to maintain very high circulating levels of 1,25-hydroxyvitamin D, presumed to be related to different sensitivity of receptors in target organs, so knowledge of 1,25-hydroxyvitamin D levels may also be helpful as more is learned about vitamin D metabolism in these animals.[6]

Studies regarding UVB and dietary calcium–vitamin D have been conducted in several reptile species and have shown that there is considerable variation in ability to use dietary vitamin D and in the need for UVB.[2,8,10–12] Although many animals can use dietary cholecalciferol or ergocalciferol (vitamin D_2, the plant form of calciferol or vitamin D), some species of birds, reptiles, and New World primates cannot use this form of the vitamin.[6,9,12] Artificial UVB has been shown to increase circulating vitamin D levels in several animals, but there are many variables affecting the efficiency of artificial lights, including distance, temperature, and time spent basking.[2,3,6,7,11,12] Ultimately, sunlight appears to be the best way to ensure adequate levels of vitamin D in most animals, but unfortunately there are very few reported baseline vitamin D levels for free ranging animals in their native habitat with which captive animal values can be compared.

West Indian iguanas of the genus *Cyclura* are the largest land vertebrates endemic to the Caribbean islands and as a group are considered to be among the most endangered lizards in the world.[1] The island of Hispañiola is home to the only two sympatric species of *Cyclura*, the critically endangered

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Table 1. 25-Hydroxyvitamin D levels in Dominican Republic iguanas, nmol/L (ng/ml), from samples collected in August 2000, November 2000, November 2001, and February 2002.

Species	Mean ± SD	Range	*n*
Wild Ricord's iguana	554 ± 275	250–1,118	22
(*Cyclura ricordii*)	(222 ± 110)	(100–448)	
Wild rhinoceros iguana	332 ± 47	260–369	7
(*Cyclura cornuta cornuta*)	(133 ± 19)	(104–148)	
Captive rhinoceros iguana	317 ± 81	220–519	13
(*Cyclura cornuta cornuta*)	(127 ± 33)	(88–208)	

Table 2. 25-Hydroxyvitamin D levels in wild Ricord's iguanas (*Cyclura ricordii*) based on month of collection, nmol/L (ng/ml).

Month	Mean ± SD	Range	*n*
Aug 2000	329 ± 52	260–389	4
	(132 ± 21)	(104–156)	
Nov 2000	383 ± 73	250–470	10
	(153 ± 29)	(100–188)	
Nov 2001	750	720–780	2
	(300)	(288–312)	
Feb 2002	929 ± 147	759–1,118	6
	(372 ± 59)	(304–448)	

Ricord's iguana (*Cyclura ricordii*) and the rhinoceros iguana (*Cyclura cornuta cornuta*), which is considered threatened in its natural environment. Little is published regarding the general biology or physiology of these species in the wild or captivity. Elucidation of vitamin D status of these animals in their natural habitat will provide a basis for determinations of normal to be used in captive management and clinical diagnosis environments for these animals.

MATERIALS AND METHODS

A total of 22 wild Ricord's iguanas and seven wild rhinoceros iguanas were captured on Isla Cabritos National Park, Dominican Republic, during the course of three trips during August and November 2000, November 2001, and February 2002. Animals were captured either using a pole snare or by live trapping with baited traps and were manually restrained for all procedures. A complete physical examination was conducted. Blood was collected from the ventral tail vein into lithium heparin or serum separator blood tubes, or both. Animals were then released at the capture site. Blood tubes were properly stored in an ice-filled cooler until they were centrifuged, and the serum or plasma decanted within 12 hr of collection. Decanted serum or plasma was frozen at −20°C for days to weeks, then was transported back to Indianapolis and stored at −70°C until analysis.

A total of 13 captive rhinoceros iguanas from Parque Zoológico Nacional, Dominican Republic, were also sampled in August and November 2000, using techniques described above. These animals are housed outside and fed a diet of vegetables, fruit, and commercial dog food. Animals chosen for this study were determined to be clinically healthy on the basis of physical examination and normal complete blood counts and serum chemistry values. Plasma 25-hydroxyvitamin D and 1,25-hydroxyvitamin D levels for all samples were assayed at Boston University using previously described methods.[4,5] Data were analyzed using a two-tailed *t*-test for unequal variance.

RESULTS

Table 1 shows the mean 25-hydroxyvitamin D levels for wild Ricord's iguanas, wild rhinoceros iguanas, and captive rhinoceros iguanas in the Dominican Republic. Values for the mean 25-hydroxyvitamin D levels for Ricord's iguanas are significantly higher than those for wild or captive rhinoceros iguana group ($P < 0.05$ for each). There was no significant difference found between the wild and the captive rhinoceros iguana groups ($P = 0.7$).

Table 2 shows the mean 25-hydroxyvitamin D levels for wild Ricord's iguanas on the basis of month and year of collection. It is interesting to note that the mean 25-hydroxyvitamin D level in February 2002 is significantly higher than those in November or August 2000 ($P < 0.05$ in each case), but there is no significant difference between the November 2000 and August 2000 means ($P > 0.05$). The average of the two samples from November 2001 is significantly higher than the November 2000 mean ($P < 0.05$). When the wild and captive rhinoceros iguana values from Table 1, which were all collected in November and August, are compared with the November and August wild Ricord's values, there is no significant difference ($P = 0.26$ and 0.1, respectively).

Table 3 shows the mean 1,25-hydroxyvitamin D levels for wild Ricord's iguanas, wild rhinoceros iguanas, and captive rhinoceros iguanas in the Dominican Republic. 1,25-Hydroxyvitamin D levels were not evaluated for the Ricord's iguana samples collected in February, and there was no significant difference between those collected in August and November ($P = 0.1$). Values for wild Ricord's iguanas are significantly higher than those for captive rhinoceros iguanas ($P < 0.05$ in each case). Values

Table 3. 1,25-Hydroxyvitamin D levels in Dominican Republic iguanas, nmol/L (pg/ml), from samples collected in August 2000, November 2000, and November 2001.

Species	Mean ± SD	Range	*n*
Wild Ricord's iguana	504 ± 174	185–785	12
(*Cyclura ricordii*)	(194 ± 67)	(71–302)	
Wild rhinoceros iguana	195 ± 62	112–307	7
(*Cyclura cornuta cornuta*)	(75 ± 24)	(43–118)	
Captive rhinoceros iguana	317 ± 99	78–432	12
(*Cyclura cornuta cornuta*)	(122 ± 38)	(30–166)	

for captive rhinoceros iguanas are significantly higher than those for wild rhinoceros iguanas ($P < 0.05$).

DISCUSSION

Very few baseline vitamin D values are reported for wild reptiles or for those in captivity housed in natural sunlight. Captive green iguanas (*Iguana iguana*) housed in natural sunlight are reported to have mean circulating 25-hydroxyvitamin D values of 374 nmol/L (150 ng/ml).[2,12] A captive adult female Komodo dragon (*Varanus komodoensis*) had more than 225 nmol/L (90 ng/ml) of this metabolite after 3 mo in natural sunlight.[2]

In general, the mean circulating levels of 25-hydroxyvitamin D observed in these wild Ricord's iguanas (554 nmol/L; 222 ng/ml) and rhinoceros iguanas (332 nmol/L; 133 ng/ml) are similar to the 374 nmol/L (150 ng/ml) reported for green iguanas in direct sunlight. It is curious that there are significantly higher circulating values of 25-hydroxyvitamin D in samples collected from wild Ricord's iguanas in February 2002 than in samples collected in August or November 2000. The higher average for the two samples collected in November 2001 when compared with November 2000 is also curious. These differences in circulating levels of 25-hydroxyvitamin D are unlikely to be because of lab or handling error and could represent behavioral or physiologic changes based on season. Temperatures on Isla Cabritos in February are generally several degrees cooler than in spring, summer, and the fall months, and field notes show that November 2001 was generally 2–3 degrees cooler than November 2000. Perhaps, cooler temperatures encourage longer basking times and therefore more exposure to UVB. Further investigations into seasonal- and temperature-related variations in vitamin D levels including ambient and body temperature measurements are merited. The significantly higher mean circulating levels of 25-hydroxyvitamin D in wild Ricord's iguanas than in wild rhinoceros iguanas is also interesting and may also represent a behavioral, physiologic, dietary, or ecological difference between the two species.

The significance of the 1,25-hydroxyvitamin D levels is not known but reported in this study as potential baseline data for future studies and comparison with other animals. The significantly higher values for wild Ricord's iguanas when compared with wild and captive rhinoceros iguanas correlates well with the same result for 25-hydroxyvitamin D levels.

On the basis of this study, serum concentrations of at least 325 nmol/L (130 ng/ml) for 25-hydroxyvitamin D should be considered normal for healthy Ricord's and rhinoceros iguanas. This information will be useful in evaluating the health of captive animals, especially in the northern hemisphere where animals must spend part of the year indoors and require artificial UVB during those periods. The majority of the animals collected in this study were adult male, so statistical comparison between gender and age class was not possible at this time but will be considered for future investigations.

Acknowledgments: We acknowledge the Indianapolis Zoo Conservation and Research Department, The Toledo Zoo Conservation and Research Department, the Riverbanks Zoo Conservation Support Fund, the Indianapolis Chapter of the American Association of Zoo Keepers, and NIH grant NO. RO1-AR-36963 to MFH for financial support of this work.

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Received for publication 28 September 2004