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HEMATOLOGIC VALUES IN BRAZILIAN *Boa constrictor* SNAKE

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Abstract: *Boa constrictor* snakes are among the most commercialized unconventional domestic animals in Brazil, following a global trend. Veterinary clinics are undergoing a transformation, requiring more specialized knowledge in specific areas, such as snake cardiology. For this reason, this study aimed to study the blood profile of this species, as well as characterize its electrocardiographic profile. Forty healthy adult snakes (*Boa constrictor*) were selected, distributed into two groups, males, and females and blood count was measured. The test results were described as mean and standard deviation, generating reference values for the *Boa constrictor* species.
Keywords: reptiles, Boidae, hematology.

INTRODUCTION

Boa constrictors (family Boidae) are non--venomous, viviparous snakes with aglyphous dentition and are among the largest snakes in the world (Pyron et al., 2014). They are crepuscular, nocturnal, and semi-arboreal animals with a life expectancy of around 25 to 30 years, and can reach 40 years in captivity (Francisco et al., 2001). They are currently divided into 11 subspecies distributed from northern Argentina to northern Mexico. Two of these, occurs in Brazil: *Boa constrictor amarali* and *Boa constrictor constrictor* (Card et al., 2016). The *Boa constrictor amarali* specie has a predominantly gray or brown coloration of varying intensities. In general, they are smaller than *Boa constrictor constrictor* specie, and rarely reach 2.4 meters (O'Malley, 2005). These exotic animals are currently recognized as one of the "new companion animals". The breeding of pet reptiles is already one of the largest pet industries in the United States (Reed, 2005) and in Europe, highlighting the urgent need to establish reference parameters in veterinary clinics to improve the medical care dedicated to these animals.

The determination of health status in reptiles is based on physical examination, as well as hematologic values. A CBC (complete blood count) can detect conditions such as anemia, inflammatory diseases, parasitemia, hematopoietic disturbance, and hemostatic alterations. In addition, the CBC is an important tool to evaluate response to disease and subsequent treatment. Reptiles are ectothermic animals, relying exclusively on the environment for proper temperature maintenance.As their activity depends on environmental variations, cellular responses in reptiles are less predictable than in endothermic animals.

Changes in the blood constituents that have been associated with seasonal influence has been reported in some species. A significant increase in heterophil, eosinophil, basophil, and lymphocyte concentration was observed in reptiles during the summer, followed by a reduction in cell counts during the winter. A significant variation in lymphocyte counts was also reported in *Crotalus durissus terrificus* subspecies, with higher values during the summer.16 However, no significant effects on packed cell volume values that were caused by en vironmental temperature alterations were observed in turtles belonging to the *Elseya novaeguineae* species. Hemoglobin polymorphism has been identified in several domestic species; however, similar data have not been reported in snakes or reptiles. Studies have attempted to correlate the hemoglobin type with reproductive status and disease resistance. In humans, there is evidence that hemoglobin C and S renders protection against malarial infection. The aim of this study was to determine the influence of season on hematologic values and the electrophoretic profile of hemoglobin of the *Boa constrictor amarali* subspecies.

In this context, the aim of this study was to study the hematological profile of healthy boa constrictors, raised in the same environment and under the same conditions.

MATERIALS AND METHODS

This research was approved by the Ethics Committee on the Use of Animals (114/2022 – CEUA/UFMG) and complies with the ARRIVE (Animal Research: Reporting of In Vivo Experiments) guidelines.

ANIMALS

Forty boa constrictors (*Boa constrictor*) were used, 20 males (average of 1.80 m) and 20 females (average of 2.10 m), adults, ranging from 5.2 kg to 12.6 kg, in the preprandial period, 21 days of fasting, from the "Jibóias Brasil" Breeding Facility.

EXPERIMENTAL PROTOCOL

The inclusion criteria established for this study were boa constrictors of both sexes and ages, clinically healthy, with adequate handling conditions and a balanced diet. The animals were handled in compliance with all biosafety and exotic animal handling protocols. All animals underwent the same experimental protocol to avoid different stress changes.

Blood samples were collected by intervertebral venipuncture using a 10 mL syringe and a 25 x 7 mm needle (BD Solomed). The blood was stored in tubes with 10% tripotassium ethylenediaminetetraacetic acid (K3EDTA) for hematological profiling; 10% sodium citrate and no anticoagulant, which were centrifuged (3000 rotations per minute for 10 minutes) to obtain plasma and serum, respectively. The samples were identified and stored at -20ºC until processing.

The blood count was performed on an automatic hematological analyzer (specific for reptiles) – IDEEX®, and blood smears were prepared on glass slides, stained with panoptic, for differential leukocyte count (Thrall et al. 2022). The packed cell volume was determined using the microhematocrit method.

STATISTICAL ANALYSIS

Data processing and analysis were performed using SPSS © IBM version 20 and STATA version MP™ software. The mean and standard deviation were calculated for descriptive statistics purposes. To study whether there was a significant difference between groups, mixed model regression analysis was used at a significance level of $p<0.05$.

RESULTS AND DISCUSSION

The hematological study of snakes has become of great importance in boas, since the techniques for keeping these snakes in captivity and at home have been improving and increasing their life expectancy. Few studies have worked with automated analysis of snake blood. However, blood smears are essential to determine the differential leukocyte count, estimate the thrombocyte mass, evaluate cell morphology, and investigate the presence of parasites and abnormal cells (Schnelle, 2022).

There was no difference ($p>0.05$) between the erythrogram values between males and females (Table 1), making it clear that the gender of the species does not influence red blood cells.

The presence of hemoparasites was also not found in the blood samples collected from these captive boas, attesting to excellent sanitary and prophylactic management.

Table 2 shows the results of the total and differential leukocyte counts with identification of heterophils, eosinophils, basophils, lymphocytes, azurophils, and monocytes. There was no difference ($p>0.05$) in the leukogram values between males and females (Table 2). Thus, the gender of the species does not influence the absolute and relative leukogram values.

It is known that the environment, temperature, humidity, handling, feeding frequency, and diseases in general influence hematological changes in reptiles (Campbell, 2006). Machado et al. (2006) studied the seasonal influence (winter and summer) on the hemato-

	Males	Females
Erythrocytes (x 10^6/µL)	0.44 ± 0.18	0.41 ±0.20
Hemoglobin (g/dL)	7.8 ± 1.70	7.5 ± 0.21
Globular volume (%)	21 ± 3.1	20 ± 2.18
VCM (fL)	430 ± 120	455 ± 135
HCM (pg)	160 ± 24	165 ± 32
CHCM (g/dL)	32.10±0.60	32.62±1.28
Thrombocytes (x10^3/µL)	195,080 ± 236	212,020 ± 755

TABLE 1 Mean values and standard deviation of the number of erythrocytes, hemoglobin, globular volume, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), and thrombocytes of male and female boa constrictors (*Boa constrictor*). Mixed model regression analysis at significance level p<0.05.

Parameters	**Males**		**Females**	
Leukocytes/µL		9,2000 ± 2.100		8,800 ± 3,2000
	Relatives	Absolutes	Relatives	Absolutes
Heterophils	40 ± 1.7 %	3,680 ±156.4/µL	42 ± 1.2 %	3,696 ± 106.5/µL
Eosinophils	1 ± 0.8 %	92 ± 7.2/µL	1 ± 0.9 %	88 ± 80.9/µL
Basophils	0 ± 0.2 %	0 ± 18.4/µL	0 ± 0.2%	0 ± 17.6/µL
Lymphocyte	22 ± 1.4 %	2,024 ± 128.8/µL	20 ± 3.2 %	1,760 ± 281.6/µL
Azurophils	32 ± 0.6 %	2,944 ± 55.2/µL	34 ± 1.3%	2,992 ± 114.4/µL
Monocytes	2 ± 1.1 %	184 ± 101.2/µL	2 ± 0.6%%	176± 52.8/µL

TABLE 2 Mean values and standard deviation of the total number of leukocytes and the relative and absolute values of heterophils, eosinophils, basophils, lymphocytes, azurophils, and monocytes of male and female boa constrictors (*Boa constrictor*). Mixed model regression analysis at significance level p<0.05.

logical profile of 25 adult *Boa constrictor amarali* snakes kept in captivity. The authors found hematocrit values of 23.2 and 21.7% in winter and summer, respectively. It is worth noting the similarity of the mean value found in summer with our results (Table 1). However, only the erythrocyte values evaluated in winter (0.404 x 106 /µL) were similar to those found in this study (0.44 and 0.41 x 106 /µL) for male and female boas, respectively. Regarding hemoglobin, the present results of 7.8 and 7.5 g/dL in males and females, respectively, were similar to those reported by Machado et al., both in winter (7.4 g/dL) and summer (7.2 g/dL).

The differential count (especially the heterophil/lymphocyte ratio) and cell morphology in the reptile leukogram are more important than the total leukocyte count. Arguedas et al. (2018), studying 15 healthy snakes of the species *Bothrops asper* raised in captivity and distributed in three experimental groups, reported basal values for leukocytes of 13,711.00 ± 5,939.02/µL; 6,532.50 ± 2,596.52/µL, and 8,614.29 ± 3,283.60/µL. The authors also reported that stress after handling animals can cause relative monocytosis and lymphopenia, results not observed in this study.

CONCLUSIONS

The determination of hematological and biochemical profile parameters of healthy boa constrictors was very similar to that of other snakes.

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CONFLICT OF INTERESTS STATEMENT

The authors declare that they have no conflict of interest.

DATA AVAILABILITY STATEMENT

Data will be made available on request.

ETHICS STATEMENT

This project was approved by the animal care and use committees at *Universidade Federal de Minas Gerais*: Ethics Committee on the Use of Animals (114/2022 – CEUA/UFMG).

CREDIT AUTHORSHIP CONTRIBUTION STATEMENT

Marthin R. Lempek: Writing – review & editing, Formal analysis, Data curation, Methodology, Investigation, Conceptualization. **Renato M. F. Yabiku**: Investigation. **Paula B. U. Fernandes**: Writing – review & editing, Investigation. **Marilia M. Melo**: Writing – review & editing, Visualization, Validation, Supervision, Resources, Project administration, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization.

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