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Short Communication

SEASONAL AND SEX VARIATION IN THE BLOOD PARAMETERS OF THE COMMON AFRICAN TOAD *BUFO REGULARIS*

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ABSTRACT

The study investigated the effect of rainy and dry seasons on the Packed Cell Volume (PCV), Red Blood Cell count (RBC count), and Hemoglobin concentration (Hb conc.) of the Common African Toad (*Bufo regularis*). Adult toads of both sexes weighing between 70-100g were randomly selected and used in the study. Each toad was fasted for 24hr and anesthetized with sodium pentobarbitone (3mg/100g i.p). Blood sample was taken from truncus arteriosus to determine the PCV, RBC counts, and Hb conc. using standard laboratory techniques. The results showed the normal fasting PCV, RBC counts, and Hb conc. during rainy season was significantly higher ($p<0.01$) than the fasting PCV, RBC count, and Hb conc. during the dry season. Also, the PCV, RBC count, and Hb conc. of the female toad was significantly ($p< 0.01$) lower compared with the male toad during the rainy season while there was no significant difference in the PCV, RBC count, and Hb concentration of male and female toads during the dry season. Therefore, seasonal changes have significant effect on the PCV, RBC count, and Hb conc. of the Common African Toad (*Bufo regularis*).

Keywords: *Bufo regularis*, packed cell volume, red blood cell count, hemoglobin, concentration, seasons.

INTRODUCTION

Several factors have been reported to affect blood parameters in amphibians. These factors include sex, season, age, and nutrition (Jungreis and Hooper, 1970; Meints and Carver, 1972; Sinha, 1983). In 1964, Foxon reported an increase in hemopoietic activity following hibernation that is associated with higher thyroid activity and the nutritional state (Meisner, 1962). There is also report of individual variation in the blood parameters of various species of amphibians. For instance, PCV is 13-39 ±2.04%, RBC counts of 120,000-470,000/mm^3, and Hb concentration of 2.4-9.6±0.45g/dl in *Rana pipiens* (Rouf, 1969), while in tropical frog *Leptodactylu fallax*, RBC counts is 600,000-744,000/mm^3, Hb concentration is 10.9% (Gattens and Brooks, 1969). And, RBC counts is 252,000/mm^3 in *Rana catesbeiana* (Hutchinson and Szarki, 1965), RBC counts of 480,000/ mm^3 and Hb content of 8.9g/dl in *Rana esculenta* (Sinha,1983). The PCV is 34% in *Bufo paracnemis* (Johansen and Ditadi, 1966), 13% in *Bufo marinus* (Andersen and Wang, 2002), 30.4 ±5.3% in *Bufo Woodhousei* (Burggren and Vitalis, 2005).

While there are many reports on the blood parameters of frogs, there are very few studies on toads. The common African toad, B*ufo regularis* is commonly found in Nigeria especially during the rainy season. There is no study on the influence of seasons on blood parameters of the common African toad. This study investigated the effect of rainy and dry seasons on the blood parameters such as PCV, RBC counts, and Hb conc. of the Common African toad *Bufo regularis*.

MATERIALS AND METHODS

Adult toads of both sexes weighing between 70-100g were randomly selected during rainy and dry seasons for the study. The toads were collected from the Botanical garden of the University of Ibadan, and its environment. The toads were randomly picked as found at night and brought into laboratory after capture. They were then kept in plastic wire-gauged cage containing water and free from insect. The cage was kept in the dark room till the following day. Each animal was fasted 24hr before the start of the experiment and anesthetized with sodium pentobarbitone (3mg/100g i. p). The animal was secured on its back on a dissecting board. The thorax was opened and the truncus arteriosus was dissected free from the surrounding connective tissue.

Each toad was then allowed thirty minutes to stabilize. After stabilization period, blood sample was taken from the truncus arteriosus to determine blood parameters: Packed Cell Volume (PCV), Red Blood Cell counts (RBC counts) and Hemoglobin Concentration (Hb conc.). Due to the small size of the toad, blood sample of just 1ml was obtained from each animal on a once for all basis. Hence,

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Table 1. PCV, RBC count, and Hemoglobin Concentration during rainy and dry seasons.

Seasons	Total number of animals	PCV (%)	RBC Count $x10^{12}$/l	Hb Concentration (g/dl)
Rainy season	60	*34.8 ± 0.7	*5.2 ± 0.1	*11.1 ± 0.4
Dry Season	60	28.2 ± 1.4	3.3 ± 0.3	8.8 ± 0.4

Students t-test: Significant *($p<0.01$)

a quanta response design was used in the study. A total of one hundred and twenty toads were collected and used for this study during two rainy seasons (May-October) and two dry seasons (November-April). For all blood samples, the values were determined immediately after blood sample was collected. The PCV, RBC counts, and Hb conc. were determined using standard laboratory techniques. The ambient temperature was measured throughout the period of the study, the ambient temperature was 28°C (26-28°C) during the rainy season and 32°C (30-32°C) during dry season.

STATISTICAL ANALYSIS

The mean ±S.E.M of all measurements were computed. Significance was assessed by student's t-test for two means of independent variables. P values of 0.05 or less was taken as statistically significant.

RESULTS AND DISCUSSION

The measured normal fasting levels of blood parameters of PCV, RBC count, and Hb concentration of common African toad *Bufo regularis* during rainy and dry seasons are presented in table 1. While tables 2 and 3 show the normal fasting levels of PCV, RBC count, and Hb conc. for different sexes of the toad during the rainy and dry seasons respectively.

Table 2. PCV, RBC count and Hemoglobin Concentration of male and female toads - rainy season.

Sex	PCV (%)	RBC count $x10^{12}$/I	Hb (g/dl)
Male	34.3 ±1.1	5.2 ± 0.5	11.2 ± 0.3
Female	*28.2 ± 2.0	*3.6 ± 0.8	*9.0 ± 0.6

Students t-test: Significant *($p<0.05$)

Table 3. PCV, RBC count and Hemoglobin Concentration of male and female toads - dry season.

Sex	PCV (%)	RBC count $x10^{12}$/I	Hb (g/dl)
Male	31 ± 1.5	3.9 ± 0.4	10.1 ± 0.6
Female	33.2 ± 2.0	4.2 ± 0.5	10.4 ± 0.9

The fasting levels of blood parameters, PCV, RBC count, and Hb concentration of *Bufo regularis* during rainy and dry seasons observed in this study were higher compared with those reported for various frog species (Kaplan, 1951; Rouf, 1969; Sinha, 1983), except for *Rana pipiens* (Meints and Carver, 1972), Indian frog (*Rana tigrina*) (Singh,1977), and toad species, *Bufo paracnemis* (Johansen and Ditadi, 1966), and *Bufo Woodhousei* (Burggren and Vitalis, 2005) in which the PCVs reported are within the range observed in the present study. The difference in the fasting levels of blood parameters, PCV, RBC counts, and Hb concentration of *Bufo regularis* and those of frogs may be due to species variation and differences in habitat. This is consistent with the findings in frogs (Schermer, 1954; Hutchinson and Szarski, 1965). Among poikilotherms, the blood parameters are reported to correlate with habitat and activity (Gaumer and Goodnight, 1957). The terrestrial amphibians are reported to have higher number of RBC counts and Hb values than semi-terrestrial and aquatic species (Leftwich and Burke, 1964; Hutchinson and Szarski, 1965). Since erythrocyte is the most important carrier of oxygen and carbon dioxide, oxygen carrying capacity of the animal is proportional to the amount of RBC counts or Hb concentration (Prosser, 1973). The higher fasting RBC counts of *Bufo regularis* observed in the present study probably suggest that the toad has a higher metabolic activity than frogs. The blood values in frogs are related to the general metabolic activities (Meints and Carver, 1972; Sinha, 1983). The terrestrial species have higher blood oxygen carrying capacity than aquatic species (Johansen and Ditadi, 1966).

The differences in the fasting levels of blood parameters of *Bufo regularis* and those of the frogs might also be due to differences in geographical location. This is consistent with findings in frogs and reptiles (Hutchinson and Szarski, 1965; Rouf, 1969).

The differences in the fasting levels of blood parameters PCV, RBC counts and Hb concentration of *Bufo regularis* during rainy and dry seasons observed in the present study can be explained by the different seasonal conditions, activities of the toad, and availability of food during the two seasons. The result agrees with the findings in frogs (Meisner, 1962; Foxon, 1964; Meints and Carver, 1972; Busk *et al*., 2000a). In frogs, the

increase in hemopoietic activity following hibernation correlates with higher thyroid activity and nutritional state (Meisner, 1962; Foxon, 1964). While the rate of nitrogen anabolism was reported to decrease with the length of starvation in wintering *Rana pipiens*, the metabolic and hematopoietic activity increased with feeding. Similar observation was also made in mammals (Kurata *et al*., 1993).

The observation of the present study in which the female blood parameters PCV, RBC count, and Hb conc. was lower compared with that of the male during rainy season may due to the different reproductive activities engaged by the two sexes. This agrees with the study of (Leftwich, 1958; Foxon, 1964). However, absence of sexual difference has been reported for some amphibians (Foxon, 1964; Hutchison and Szarski, 1965).

In conclusion, the result of this study has shown that seasonal and sex variations exist in the PCV, RBC count and Hb conc. of the common African toad *Bufo regularis*. The results also show that the fasting levels of blood parameters PCV, RBC count, and Hb concentration of *Bufo regularis* are higher than those of frogs.

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