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Changes in Haemolymph Biochemical values during different growth phases in African Giant Land Snail (*Archachatina Marginata*) Swainson

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Abstract

The impact of growth phases on the haemolymph biochemical value of African giant land snail Archachatina marginata was studied. The growth phases were: snailet, juvenile and Adult based on the number of whorls on the shell and weight of the snails. Highest concentration of glucose and lipids were recorded at the juvenile phase (40.20mg/dl and 48.60mg/dl)., while least values for glucose (20.00mg/dl) and lipids (37.80mg/dl) were recorded during snailet and adult phases respectively. A progressive increase in the concentration of protein was observed as the snails moved from snailet phase to adult phase, hence snails in adult phase had the highest concentration of protein in their haemolymph. Growth phases significantly ($P<0.05$) affected the haemolymph mineral composition of the snails. Juvenile phase had the highest values in Ca^{2+}, PO_4^{3-} and Na^+, while the adult phase recorded the least concentration in Ca^{2+}, Na^+ and Cl^-. Juvenile growth phase of A. marginata thus has better haemolymph biochemical values than both snailet and adult growth phases.

Key words: *growth phase, biochemical value, haemolympmph, A marginata*

Introduction

The African giant land snail, *Archachatina marginata* (Swainson) occurs commonly in the high forest and fringing forest of the derived savanna regions of West Africa (Yoloye, 1994). It is a nocturnal animal becoming very active at night, but can eat at any time of the day if the environment is cool (Akinnusi, 2002). Research has shown that exposure of snail to continuous light at night increase their activity and rate of food consumption and also promotes rapid growth (Hodasi, 1984). Before ingesting their food, snails explore their food with their tongue using their gustatory sense (Ademolu *et al*, 2004).

The snail haemolymph is blue in color indicating the presence of haemocyanin, a blue copper-containing pigment in the solution. Studies have revealed that the haemolymph is influenced by several factors, like diets and stocking density (Ademolu *et al*., 2006) suggesting that the physiological condition of land snail is evidenced in its haemolymph properties (Akinloye and Olorode, 2000).

South (1992) described the 3 growth phases during the development of snail: infantile phase, juvenile phase and mature (adult) phase. While infantile and juvenile phases are marked with rapid growth, little or no growth is experienced during the mature phase.

Young snail have been reported to lay fewer number of eggs than the old ones (Adeleye *et al*, 2001) while older ones did not lay eggs. Hodasi (1984) similarly discovered that aged snails (24-30 month old) having reached full size failed to lay eggs. The objective of this study was to investigate the impact of growth phases on the biochemical value of the haemolymph of A. *marginata*.

Materials and Methods

Sixty (60) African giant snails. A. *marginata* of different sizes were obtained from the snail pen of Forestry and Wildlife Management Department in the University of Agriculture, Abeokuta (UNAAB), Ogun state, Nigeria Grouping of the snails into different growth phases (Snailet, juvenile and Adult) was done based on the number of complete shell whorls (4.0, 6.0, and 7.0), weight of the snail (100.5g, 133.2g and 211.7g), the shell length (8.4cm, 11.0cm, and 15.8cm) and shell circumference (8.5cm, 17.0cm and 19.0cm) as described by Ebokogi and Osinowo (1998).

Biochemical analysis of the haemolymph

The methods described by Akinloye and Olorode (1986) and modified by Ademolu *et al* (2006) was used in collecting the haemolymph after removing the apex of the shell.

The protein concentration was determined by biuret method as described by Henry *et al*, (1974). The glucose content was estimated by colorimetric method of Baumniger (1974). The lipid assay was done following the method of Grant (1987). The mineral content (Na^+, K^+, Ca^{2+}, PO_4^{3-}, Cl^-) of the haemolmph was determined by the methods of A.O.A.C (1990). The analysis was replicated twice. The data obtained were subjected to one-way analysis of variance (ANOVA) using Minitab (1988) version package.

Results

The morphologicval features of the experimental snails are shown in Table 1 below. The weight ranged from 109.7g-216-85g, while the number of shell whorls varied from 4.0.-7.0.

The chemical analysis of the haemolymph revealed the presence of proteins, lipids and glucose in varying concentrations. (Table 2). Growth phases significantly ($P<0.05$) influenced the concentration of the three organic substances. The haemolymph of juvenile had the highest values of glucose (40.20mg/dl) and lipids (48.60mg/dl) followed by the adult phase and snailet phase for glucose and lipid respectively. However, highest concentration of protein was recorded by adult stage followed by the juvenile stage.

The result of mineral analysis indicated the presence of all the ions tested in the snail haemolymph. Juvenile stage recorded highest values in Ca^{2+}, PO_4^{3-}, Na^+ (Table 3). It is noteworthy that the concentration of Na^+ ranked

Table 1: the morphological features of the experimental snail

	Snailet	Juvenile	Adult
Av. Shell length (cm)	8.8±0.08	11.75±0.03	15.10±0.05
Av. Shell Circumference (cm)	12.65±0.01	17.00±0.25	18.25±0.02
Av. Weight (g)	109.7±0.03	136±0.01	216.85±0.21
No of Shell whorls	4.0±0.01	6.0±0.01	7.0±0.02

Table 2: Mean concentration of the substances in the haemolymph of different growth phases of anails (*A. marginata*)

	SNAILET	JUVENILE	ADULT
Glucose (mg/dl)	20.00 ± 0.02^{c}	40.20 ± 0.05^{a}	26.70 ± 0.02^{b}
Protein (g/l)	36.50 ± 0.12^{b}	39.10 ± 0.10^{b}	49.60 ± 0.20^{a}
Lipids (mg.dl)	43.20 ± 0.04^{b}	48.60 ± 0.23^{a}	37.80 ± 0.30^{c}

** mean values in each row with the same superscripts are not significantly different ($P<0.05$).

highest followed by Cl^{-} while PO_4^{3-} ranked least in the haemolymph of the experimental sails.

Discussion

The result of the present study revealed that growth phases significantly affect the morphological features of the snail, that is, the higher the age. The bigger that snails. The number of shell whorls also varied with the age, Akinnusi (2002) had earlier observed that snail age can be determined by the number of whorls on its shell.

Juvenile stage haemolymph had high concentrations of lipids and glucose than both adult and snailet phases. Lipids and glucose are energy sources which supply energy to the cells

Table 3: Mean concentration (mg/ml) of minerals in the haemolymph of different growth phase of snails (*A marginata*)

	SNAILET	JUVENILE	ADULT
Ca^{2+}	2.7 ± 0.07^{b}	3.10 ± 0.05^{a}	2.60 ± 0.1^{b}
PO_4^{2}	0.06 ± 0.02^{b}	1.00 ± 0.25^{a}	0.09 ± 0.11^{a}
Na^{+}	80.00 ± 0.21^{b}	87.20 ± 0.31^{a}	78.00 ± 0.6^{b}
K^{+}	3.4 ± 0.12^{b}	3.20 ± 0.01^{b}	4.20 ± 0.2^{a}
Cl^{-}	50.10 ± 0.03^{a}	43.20±0.2b	34.20 ± 0.74^{c}

** mean values in each row with the same superscripts are not significantly different (P<0.05).

when oxidized through metabolic pathways (Odiete, 1999). The higher concentration of these energy sources in juvenile phase might be due to the fact that juvenile phase is the most active phase in snail life hence requiring higher energy resources to support and sustain their activities. On the hand, adult phase because of its weight is not very active thus requiring less concentration of energy substrates.

Protein concentration was observed to be building up as the snails' ages, from 36.50 (snailet) to 49.60 (Adult). Proteins are regulators of physiological processes such as growth and especially reproduction which is the main activity during adult stage (South, 1992).

During the adult stage, protein is significantly higher (P<0.05) than the two other substrates (lipids and glucose). This is in agreement with the results of previous study (Ademolu *et al.* 2004). Snails are known to be a good protein source which can be consumed to ameliorate the problem of protein deficiency in diets, a common phenomenon in Nigeria (Imevbore and Ademosun, 1988).

The ionic analysis of the haemolymph showed that the highest values of Ca^{2+}, PO_4^{3-} and Na^{+} were recorded by the juvenile stage. The higher concentration of Ca^{2+} at this stage might probably be as a result of rapid increase in shell size and the replacement of the lost radula.

Phosphate plays an important role in the formation of ATP, a high energy form. Its higher concentration in the juvenile stage is not unexpected as this is the most active phase.

The concentration of $Na^{+}$and Cl^{-}were more than other minerals analyzed. This result contradict the report of Akinnusi (2002) and Ogunsanmi *et al.* (2003) that snails must not be given salt. Na^{+} and Cl^{-} contribute significantly to the osmotic pressure of the animal tissues and

are needed for nervous communication in animals.

Results from this study runs in agreement with findings of other researchers. Recently, Abiona *et al* (2007) reported that medium size snails (juvenile) produce highest number of spermatozoa and ova, suggesting that they are more sexually active than other live weight groups. In related experiment Idokogi and Osinowo (1998) observed that in smallsize snails reproductive structure are rudimentary, showing that they were not sexually mature at this stage. It can be concluded that the juvenile stage of snails has a better physiological status than both adult and snailet stages and thus should be recruited for breeding purposes.

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