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Original article

Growth and reproductive performances of African giant snail (*Archachatina marginata*) as affected by dietary calcium levels

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A B S T R A C T

This study was designed to evaluate the effect of dietary calcium level on biological performances of African giant snail *Archachatina marginata*. The snails were fed on 4 diets containing graded calcium content (12%, 14%, 16% and 18%) for thirty-two weeks. The main results revealed that, growth performances were higher ($p<0.05$) with the highest calcium content in feed (18%) followed by 16%. The cumulative mortality rate was higher (24.64%) in snails fed on diet containing the smallest amount of calcium (12%), while snails fed on the highest calcium level (18%) recorded the highest survival rate (89.41%). Except for the body weight at the first laying, which varied with the variation of the calcium in the ration, no significant difference was recorded between treatment groups for reproductive performances. The highest ($p<0.05$) number of eggs laid were recorded with 16% (27 eggs) and 18% (26 eggs) of calcium. The lowest reproductive traits were recorded with the lowest calcium level. It was concluded that, *A. marginata* performed better with 18% of calcium in the ration with regard to growth and reproduction performances.

1. Introduction

The giant African snail is an important source of animal protein and income for many households in the Western and Central African forest zone (Brescia et al., 2002; Kouassi et al., 2008; Sika et al., 2014). Its flesh contains almost all the essential amino acids needed by the human body with a crude protein content varied between 81-82% (Okon et al., 2016). It is also an important source of energy (Fagbuaro et al., 2006; Diomandé et al., 2008; Otchoumou et al., 2010), and macro-elements such as iron, calcium, phosphorus, magnesium and vitamins (Brescia et al., 2002; Okon et al., 2016). Fat content of snail's flesh varied between 0.05 and 0.8% (Narku, 2013) with the major fatty acids being oleic and stearic acids (Malaisse, 1997). Its shell is an important source of calcium in animal nutrition especially pigs, broilers and layers (Houndounougbo et al., 2013). However, wild snail populations have declined considerably as a result of human activities such as deforestation, pesticide use, bush fire, agriculture and the increasingly excessive collection of immature snails (Sika et al., 2014). The intensive harvest and exploitation of forest resources by local populations raises questions about the continued availability of wildlife (Bouye et al., 2017).

The success of breeding non-conventional animals requires an adequate feed formulation. Several studies have been carried out to determine the dietary calcium content necessary to shorten the growth and reproduction cycle of giant snails (Otchoumou et al., 2004; Karamoko et al., 2015). However, no detailed study has been conducted on the dietary calcium level required for better growth and reproduction of *Archachatina marginata.* A sustainable effort in finding a balanced diet to improve the growth and reproductive performances of this snail became a necessary. This study was designed to determine the growth and reproductive calcium need of African giant snail *Archachatina marginata*.

2. Materials and methods

2.1. Study area

This study took place at the Teaching and Research Farm of the University of Dschang between March 2018 and October 2018. The breeding took place in bins arranged along the walls inside a building. The average temperature and the relative humidity in the building were 19.7°C and 62.4% respectively, while the temperature of the rearing substrate and its humidity were 16°C and 84% respectively. The photoperiod was natural.

2.2. Animals

One hundred and eighty (180) young snails (*Archachatina marginata*) with average weight 10±1 g and 33.33±1 mm in length and 24.25±1 mm in width were used in this study. They were collected in cocoa plantations in the Moungo area (Njombé). The snails used in the study were selected based on their morphology and behavior: same live weight, well-formed shell, no breakage, individuals free from all visible and active traumas.

2.3. Substrate and breeding

The snails were raised in rectangular plastic tanks with 45 cm, 30 cm and 25 cm respectively for length, width and depth. An anti-leak device consisting of a fine mesh (2 mm mesh) was placed above each tank to prevent snails from escaping. Feeders and drinkers were made of small kitchen dishes.

2.4. Experimental diets

Table 1 summarises the composition of experimental diets containing respectively 12% (R1); 14% (R2); 16% (R3) and 18% (R4) calcium. The rations were formulated by varying the content of ingredients including seashell as main source of calcium. To avoid the simultaneous variation of the nutrients content due to the variation of the ingredients in the different rations, agar-agar powder was used to obtain experimental rations with variable level of calcium with other nutrients constant.

2.5. Experimentation and data collection

One hundred and eighty (180) young *Archachatina marginata* with equal average weight (10±1g) and length of shells (33.33±1 mm) were randomly distributed in 20 breeding tanks with a density of 75 snails/m² with 9 snails/tank. Inside each tank, 10 cm of sawdust was introduced as breeding substrate. The moisture content of the substrate was maintained above 85% by watering every day each tank with the same amount of water (120 ml).

Table 1
Proximate composition of the experimental diet.

Ingredients	Experimental diets			
	R1 (12%Ca)	**R2 (14%Ca)**	**R3 (16%Ca)**	**R4 (18%Ca)**
Maize	15	15	18	20
Soybean cake 49	20	20	20	13.5
Cotton cake	14.75	14.75	9	0
Palm kernel cake	6	0	0	0
Fish meal	0	0	5.5	17
Wheat bran	12	5	0	0
Seashell	31	32.5	37.5	40.5
Palm oil	0.5	0.5	0.5	0.5
Salt	0.25	0.25	0.25	0.25
Premix 0.5%	0.5	0.5	0.5	0.5
Agar-agar	0	11.5	6.25	7.75
Total (kg)	100	100	100	100
Analysed chemical composition				
Crude protein (%)	18.86	18.93	19.13	19.21
Crude energy (Kcal)	4193.00	4361.14	4315.82	4331.17
Fat (%)	1.12	2.23	1.48	2.28
Calcium (%)	11.85	13.57	16.01	17.76
Phosphorous (%)	0.28	0.26	0.21	0.23
Potassium (%)	0.17	0.20	0.30	0.20
Magnesium (%)	0.56	1.02	1.04	1.04
Natrium (%)	0.10	0.07	0.10	0.07
Iron (%)	0.22	0.25	0.23	0.25

2.5.1. Growth performances

The feed was weighed and fed to the snails, and every two days the leftover was weighed. Troughs and feeders were properly cleaned before being reused. Faeces were also removed after every two days and once a week the breeding substrate was stirred and mortalities of snails recorded. The snails were weighed every two weeks for a period of eight months using a 1 g precision scale. Length of the shells was measured with electronic callipers to assess monthly shell growth. Feed intake, weight gain, shell growth, and mortality rate were calculated as proceeded by Otchoumou et al. (2003).

- Feed intake (FI)

Feed intake was calculated as the amount of feed that a snail consumed per day per unit of live weight, using the following formula:

FI (g/d/g live weight) = Q / Pm

With Q = average daily feed intake; Pm = average live body weight (g),

Knowing that: Q (g/d) = (qi - qf)/t, with qi: initial quantity of feed (g); qf: final quantity of feed (g); t: duration of feeding trial.

- Live weight

Live weight is used to assess changes in body mass, snails were weighed at the beginning of the trial and every 14 days thereafter using a 1 g precision scale. The weight gain was calculated by making the difference in weight between two consecutive weighing. The length and diameter of the shells were measured with electronic callipers in order to evaluate shell growth.

- Weight gain (Wg)

The daily gain in live weight was expressed using the following formula:

Wg (g/d) = (P2-P1) / (T2-T1)
With Wg = weight gain, P1 (g) = initial weight; P2 (g) = final weight; T1 (d) = day 1; T2 (d) = final day.

- Shell Growth (Sg)

The shell growth expresses the growth in length and width of the shell:
Sg (mm/d) = (L2-L1) / (T2-T1)
With L1 (mm) = initial length of the shell; L2 (mm) = final length of the shell; T1 (d) = day 1; T2 (d) = final day

- Feed conversion ratio (FCR)

The feed conversion ratio was expressed by the following formula:
FCR = (Amount of feed consumed (g) / (Weight gain (g)

- Cumulative mortality rate (CMr)

The mortality rate was calculated according to the following formula:
CMr (%) = Nm x 100 / Ei
With Nm = Total number of dead snails; Ei = initial number of snails.

2.5.2. Reproductive performance

During the breeding period, the eggs collected each morning were gently weighed individually using a 0.001g precision electronic scale while the large and small diameter of each egg was recorded using a calliper. The eggs were immediately incubated by spawning cluster in plastic pots with a white sawdust substrate. Each pot was watered every 14 days with the same amount of water (100 ml) to maintain humidity. From 35 days of incubation, the pots were gently checked every two days for the evaluation of eggs hatching. After hatching the spat was weighed and the length of shells measured. At the end of the breeding phase, incubation time and hatching rates were calculated and the following parameters were evaluated:

- ✓ Weight at first laying
- ✓ The number of eggs per spawning cluster
- ✓ Eggs weight
- ✓ Length and width of eggs
- ✓ Spat weights (Sw)
- ✓ Incubation time

2.6. Statistical analysis

The data collected on the different parameters were submitted to one way analysis of the variance test by General Linear Model procedure of statistical package for Social Science Software (SPSS 20.0). In case of significant difference between treatment groups, the means were separated by the Duncan Multiple Range test at 5% threshold.

3. Results and discussion

3.1. Growth performances

The growth performances of *Archachatina marginata* as affected by dietary calcium level is summarised in Table 2. Growth parameters studied were higher ($p<0.05$) in snails fed on the highest calcium level (18%) followed by those fed on 16% calcium. Snails fed on 12 and 14% calcium diets had almost the same growth performances. Rations containing respectively 12 and 14% calcium; 16 and 18% calcium contents were comparable for shell growth in length and width. Daily feed intake/g of live weight and feed conversion ratio were not significantly ($p>0.05$) affected by the graded level of calcium in the rations. However, the cumulative mortality rate was higher in snails fed on ration containing 12% calcium while the highest survival rate was recorded with the highest calcium content (18%).

Table 2
Effects of calcium level in diet on growth performance of *Archachatina marginata*.

Growth characteristics	Experimental diets				p
	R1 (12%Ca)	R2 (14%Ca)	R3 (16%Ca)	R4 (18%Ca)	
Initial wheight (g)	10±1	10±1	10±1	10±1	
Final wheight (g)	42.97±1.92^{a}	42.93±2.21^{a}	47.57±3.70^{b}	51.68±1.55^{c}	0.00
Total wheight gain (g)	32.97±1.92^{a}	32.93±2.21^{a}	37.57±3.70^{b}	41.68±1.55^{c}	0.00
Feed intake (g/d)	0.34±0.082^{ab}	0.26±0.15^{a}	0.36±0.034^{ab}	0.40±0.048^{b}	0.159
Daily feed intake (g/d/g LW)	0.0078±0.002	0.0079±0.001	0.0075±0.0003	0.0077±0.001	0.964
Shell growth in lengh (mm/d)	0.24±0.006^{a}	0.24±0.006^{a}	0.28±0.006^{b}	0.28±0.00^{b}	0.00
Shell growth in width (mm/d)	0.12±0.006^{a}	0.13±0.00^{a}	0.15±0.006^{b}	0.15±0.00^{b}	0.00
Daily wheight gain (g/j)	0.15±0.0087^{a}	0.15±0.010^{a}	0.17±0.016^{b}	0.19±0.007^{c}	0.00
Feed conversion ratio	2.33±0.63	2.30±0.44	2.12±0.23	2.22±0.40	0.831
Mortality rate (%)	24.64±0.77^{c}	15.85±1.03^{b}	13.68±1.01^{b}	10.59±2.63^{a}	0.00

a, b, c: averages bearing the same letter on the same line are not significantly different (P>0.05); R1: diet containing 12% calcium; R2: diet containing 14% calcium; R3: diet containing 16% calcium; R4: diet containing 18% calcium.

The curve of the biweekly feed intake as affected by the graded level of calcium has the same profile and the same trend from the beginning to the end of the trial (figure 1). However, the feed intake of snails fed on 12% calcium remained below that of the other three groups regardless of the period of study. During the entire study period, the feed intake of snails fed on diet containing the highest calcium level (18%) was higher compared to the other three treatments.

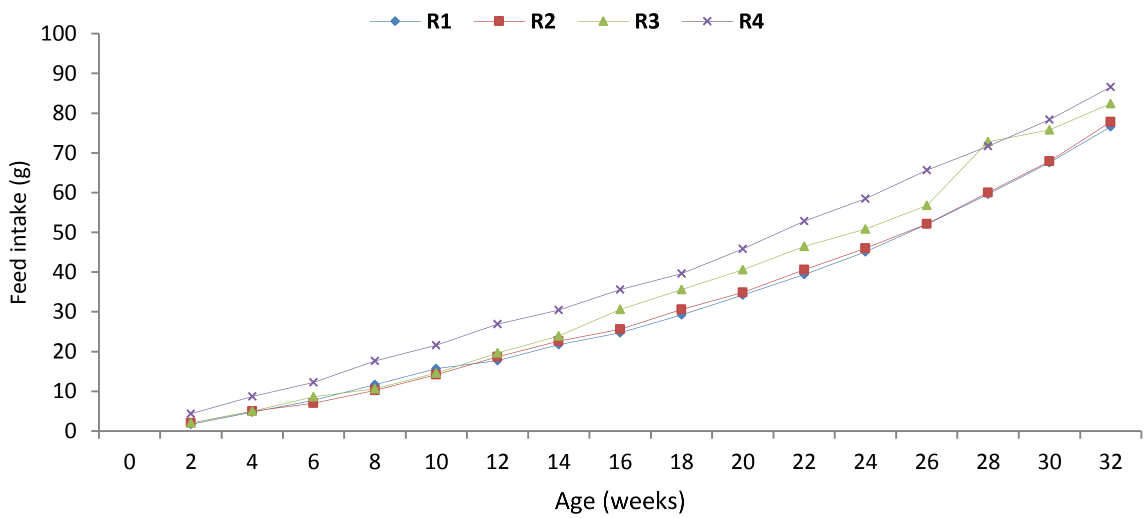

Fig. 1. Bi-weekly evolution of feed intake of *Archachatina marginata* as affected by the calcium level of the diet.

The biweekly change in live weight of snails as affected by the calcium level of the ration is presented in figure 2. From the second week till the end of the rearing period, live weight of snails fed on 16 and 18% calcium was significantly higher ($p<0.05$) than the weight of snails fed on 12 and 14% calcium. When compared the treatments with the highest calcium contents (16 and 18%), it is noticed that from the 18th week, the live weight of the snails fed on 18% calcium took over that of the snails fed on 16% calcium.

3.1.1. Shell growth

From figure 3 showing the shell growth in length, it appears that the curves have the same profile and the same trend from the beginning until the end of the experiment. However, the length of the shell decreased with the lowest calcium level during the entire study period. From week 4, the shell length of the snails increased proportionally with the calcium content of the ration.

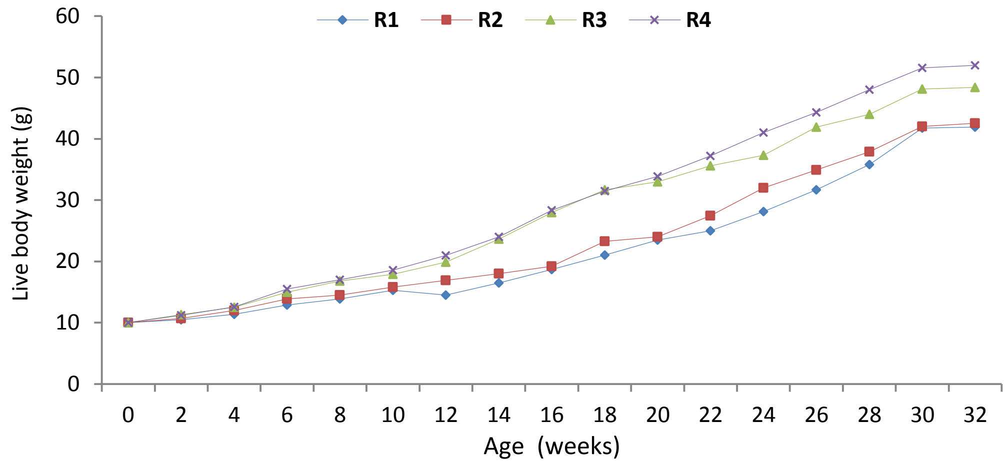

Fig. 2. Bi-weekly evolution of live body weight of *Archachatina marginata* as affected by the calcium level of the ration.

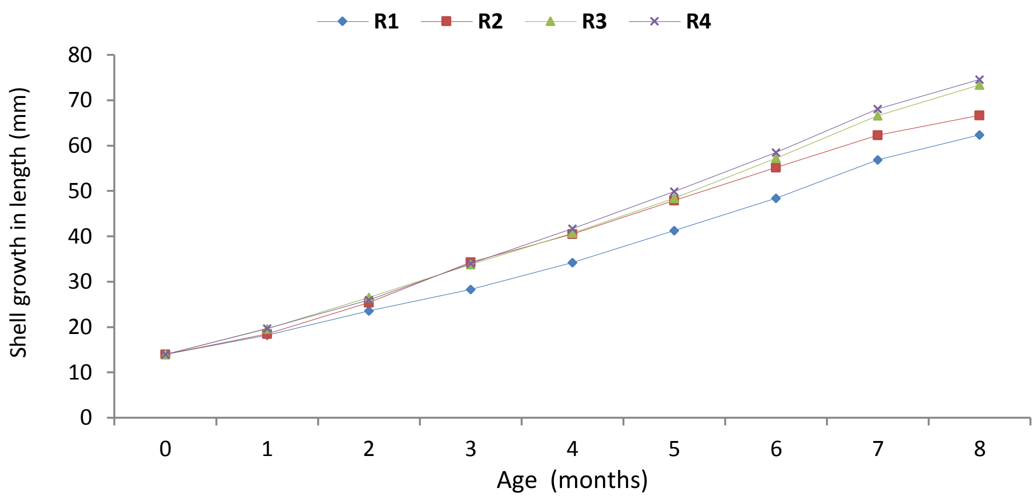

Fig. 3. Monthly evolution of shell growth in length of *Archachatina marginata* as affected by the dietary calcium level.

Table 3
Effects of dietary calcium levels on the reproductive performances of *Archachatina marginata*.

Reproductive characteristics	Experimental diets				
	R1 (12%Ca)	R2 (14%Ca)	R3 (16%Ca)	R4 (18%Ca)	*p*
Weight at first laying (g)	45.19±1.31[a]	47.37±1.61[a]	56.15±3.39[b]	57.27±1.81[b]	0.00
Number of egg-laying	1[a]	3[b]	4[c]	4[c]	0.00
Number of eggs per spawning cluster	5±0.00[a]	5.67±1.55[a]	7±1[b]	6.67±1.55[a]	0.11
Total number of egg laid	5[a]	17[b]	27[c]	26[c]	0.00
Weight of the eggs (g)	1.13±0.057	1.26±0.11	1.26±0.15	1.3±0.10	0.33
Lengh of egg (mm)	14.38±0.076	14.43±0.17	14.46±0.14	14.41±0.36	0.97
Diameter off egg (mm)	11.33±0.33	11.26±0.28	11.30±0.30	11.36±0.33	0.97
Incubation period (days)	47.68±2.51	49.33±1.15	49.33±1.52	50±2.00	0.5
Total hatching rate (%)	100	100	100	100	-
Weight of snailet (g)	0.93±0.060	1.02±0.064	1.01±0.076	0.99±0.10	0.47

a, b, c: averages bearing the same letters on the same line are not significantly different (P>0.05); R1: diet containing 12% of calcium; R2: diet containing 14% of calcium; R3: diet containing 16% calcium; R4: diet containing 18% of calcium.

3.1.2. Reproductive performances

The reproduction characteristics of *Archachatina marginata* as affected by the level of dietary calcium is summarised in Table 3. With the exception of the weight at first laying and the number of eggs laid which varied with the calcium level of the ration, no significant difference was recorded among the treatment groups for the reproductive characteristics. With regard to the total number of eggs laid, the highest values ($p<0.05$) were recorded with the highest calcium levels (16 and 18%). Snails fed on 16% calcium recorded the highest number of eggs per spawning cluster. The lowest values ($p<0.05$) of all the reproductive parameters were recorded with the lowest calcium level (12%).

Successful snail breeding requires a modernisation of breeding techniques including the establishment of nutrients requirements. Many authors reported that feeding snails with a compound feed balanced in calcium and protein is very important for young snails (Sika et al., 2014; Karamoko et al., 2015; Sika et al., 2015). In addition to its physiological role, calcium is involved in shell growth and therefore the whole individual. The present study revealed that growth performances were better with the highest calcium level (18%). Snails fed on feed containing 12 and 14% calcium recorded the lowest performances. The same trend was recorded for shell growth in length and width. Indeed, the increase in the calcium content in feed induced an increase in the speed of weight and shell growth that results in significant weight gains (Bouye et al., 2017). The intake of nutrients such as calcium contributes enormously in improving growth performances of snails in captivity (Codja et al., 2002; Otchoumou, 2005; Kouassi et al., 2007; Karamoko et al., 2015). According to Jess (1989) and Bonnet et al. (1990), the snail use calcium to elaborate its shell and organs involve in growth. However, this mineral is not the only nutrient responsible for the growth performances achieved. It is essential to note the synergistic action of all the mineral elements and also the organic matter such as proteins in the metabolism of growth and even on reproduction (Cobbinah et al., 2008). Mineral such as magnesium is essential for the assimilation of vitamin B2 (Thianin), essential for protein synthesis, carbohydrate metabolism and would also facilitate the absorption of calcium and phosphorus (Parigi, 1986). Regnier (1976) emphasised the role of proteins in calcium absorption by forming easily absorbable complexes with calcium and phosphorus.

The results of the present study revealed that the cumulative mortality rate was higher in snails fed on diet containing 12% calcium while the highest survival rate was recorded with 18% calcium. In fact, insufficient intake of calcium led to a slowdown in growth and the production of a thin shell (Thompson and Cheney, 2004; Karamoko et al., 2015; Bouye et al., 2017) exposing snails to various traumas, leading to the death of animals. Analysis of data on the reproductive characteristics of *Archachatina marginata* revealed that with the exception of weight at first laying and the total number of eggs lay which varied with the calcium level of the diet, no significant difference was recorded. Indeed, stunted snails reach sexual maturity with low weight and in this condition snails exhibit poor reproductive performances (Thompson and Cheney, 2004). With regard to the total number of eggs laid, the highest performances were recorded with the highest calcium levels (16 and 18%). Similarly, snails fed on diet containing 16% and 18% calcium produced the highest number of eggs. This result can be explained by the calcium content of feed and also by the synergistic action of all the mineral elements and organic matter involved in the metabolism of growth and reproduction (Cobbinah et al., 2008).

The reproductive performances of *Archachatina marginata* recorded in this study was lower than that obtained by Awah et al. (2001), Brescia et al. (2002), Malaisse (1997) and Stiévenart (1997). This could be explained by the potentially different genetic characteristics of the subspecies of snails used by each of the authors. Indeed, there are several subspecies not yet identified (Hardouin et al., 1995). The present poor performances compared to those of the above could be also due to the study conditions. The results of Stiévenart (1997) were obtained in the laboratory under controlled conditions whereas the snails in this study were exported from their natural environment (hot and humid climate) and raised in artificial conditions in an area with very different climatic conditions (cold and dry climate). Then, in the present study, acclimation stress in an environment very different from their natural biotope could justify the poor performances. Any abrupt variation of the environmental condition constitutes a traumatic factor for the snails. In such situations, snails exhibit low growth and poor reproductive performances. Thus, such disorders are manifested by very small reproductive sizes, or even true dwarfism (Hardouin *et al.,* 1995) which leads to poor growth (Stievenart, 1997).

4. Conclusion

The present results showed that *A. marginata* performed better with 18% calcium in the ration with regard to growth rates and reproductive traits.

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