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Reproductive performance of *Archachatina marginata* bred on substrates amended with oyster shell powder and substrates amended with pig bone powder

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Abstract

In order to determine a rearing substrate suitable for optimising the reproductive performance of *Archachatina marginata*, substrates were prepared by amending the compost at different rates (0%, 10%, 20%, 30%, 40%) with either oyster shell powder or pig bone powder. Spat were reared on these substrates until they reached sexual maturity. The age of first oviposition, the number of eggs per oviposition, the number of oviposition per snail per year, the weight and size of the eggs then the hatching rate of these eggs as a function of the calcium source and the substrate amendment rate were recorded and compared. The results showed that increasing the substrate amendment rate in oyster shell powder from 0% to 40% was favourable to reducing the sexual maturity of *A. marginata* (from 10.17 months to 9.03 months), optimising the number of egg-layings (from 7.04 to 12.04) and the number of eggs per laying (from 7.27 eggs to 8.18 eggs). Also, the largest eggs (1.54 ± 0.12 cm in large diameter and 1.33 ± 0.93 cm in small diameter) and the heaviest (1.63 ± 0.19 g) were laid on the substrate amended to 30% oyster shell powder. The best reproductive performances recorded on substrates amended with bone powder were not as good as those recorded on substrates amended with oyster shell powder. This study therefore showed that amending the compost with oyster shells optimised the reproductive performance of *Archachatina marginata*.

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Introduction

The giant African snail is an animal species with great zootechnical potential (Awohouédji, 2010). This mollusc is often found under plant debris, in certain agricultural areas, and especially just about everywhere in the tropics during the rainy seasons (Sodjinou *et al.*, 2003). Its flesh is appreciated in both urban and rural environments in many African countries, including Côte d'Ivoire (Kauassi *et al.*, 2008). It is a genuine and very valuable source of animal protein (Ajayi *et al.*, 1978).

The snail carries out most of its shell enlargement before reaching sexual maturity. The process of shell enlargement relies on a growth organ located in the parietal part of the mantle ridge straddling the labrum (Stievenart, 1996). Soft tissue development only occurs in the production of shell material results in shell enlargement (Otchoumou *et al.*, 2011). The enlargement of the shell therefore conditions the development of the snail's soft tissues (Stievenart, 1996). The shell of this mollusc is essentially made up of calcium carbonate crystals taken from the soil and food (Ajayi *et al.*, 1978). The snail uses calcium to build its shell and to calcify its eggs. This mineral is therefore essential not only in the preparation of diets, but also in breeding substrates (Kouassi *et al.*, 2007; Kouassi *et al.*, 2014; Aman *et al.*, 2018). In fact, this mollusc draws around 40% of its nutrients from the soil in which it lives, transcutaneously via its pedal sole (Jess, 1989). This is why the population densities and abundance of these molluscs are strongly correlated with the calcium content of soils colonised in nature (Hotopp, 2002 ; Memel *et al.*, 2011). However, not all soils are rich in calcium. In some cases, for the soil to be more suitable for rearing giant African snails, it needs to be amended with calcium from naturally available sources (Aman *et al.*, 2018 ; Bouyé *et al.*, 2017). Moreover, the rate and bioavailability of calcium vary from one source to another. Furthermore, the rate and bioavailability of calcium vary from one source to another. This is why, the aim of this study is to verify the bioavailability of calcium in oyster shell and in pig bones for Archachatina marginata through its reproductive performance.

Material and methods

Study site

The experimental farm at the University of Abobo Adjamé (now NANGUI ABROGOUA University) was used as the study site.

Animal

Archachatina marginata spat with an average live weight of 1.44 ± 0.23 g and an average shell length of 19.1 ± 1.9 mm (Fig. 1) were used for this study. These snails were born from breeders that were themselves born at the University's experimental farm.

Fig. 1. *Archachatina marginata* spat (Source : Aman *et al.*, 2011)

Breeding enclosure

Parallelepiped wooden tanks with a base area of 0.6 m^2 and a volume of 0.6 m^3 (Fig. 2) were used as snail breeding enclosures.

Fig. 2. Breeding enclosure

Calcium source

Two biological sources of calcium were used to amend the soil used as a substrate for snail rearing. The choice of these calcium sources that are, the oyster shell and the pig bones, was based on their availability in nature.

Feed

A concentrated feed based on cereal flour and agri-food by-products, enriched with vitamins and calcium (Fig. 3), was used to feed the snails on a daily basis.

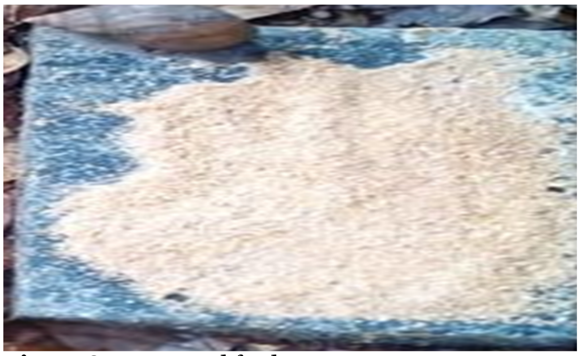


Fig. 3. Concentrated feed

Confection of breeding substratum

Pig bones were collected from delicatessens and oyster shells from lagoon sand. After collection, these calcium sources were properly cleaned to remove any animal tissue residues that might encourage microbial growth. They were then oven-dried at 90°C for 24 hours before being crushed using a sledgehammer. These crushed elements were then ground and sieved using a 2 mm diameter mesh sieve. The potting soil taken from under the university forest was heated with charcoal for 20 minutes in a large aluminum basin. Heating eliminates insects and insect eggs (*Alluaudihella flavicornis*, *Fusarium* fungus) that can attack snails. This soil was then sieved to remove pieces of wood and stones that could break the snails' shell edges. A substrate consisting solely of potting soil was used as a control (So = 100% potting soil). Amending the potting soil with oyster shell powder at different rates (5%; 10%; 20%; 30% and 40%) made it possible to make 5 types of rearing substrate (Sh5; Sh10; Sh20; Sh30; Sh40). Bone powder was also used to make 5 types of substrate (Sos5; Sos10; Sos20; Sos30; Sos40) (Aman *et al.*, 2011).

Breeding techniques

Four hundred and ninety-five *Archachatina marginata* spat were randomly distributed at a density of 25 snails/m² on the 11 types of substrate, with three replicates per substrate type. The snails were fed ad libitum every two days. At the end of the two days, the feeders were properly washed before being reused. Substrates are regularly cleaned of food rejects and animal faeces to prevent the development of pathogens. All these hygiene rules help to limit mortalities. The enclosures were watered daily, morning and evening, at a rate of 0.25 litres/substrate/watering, to maintain a relatively constant humidity (Aman *et al.*, 2011).

Determination of reproductive performance

The snails were bred on the various substrates until they laid their eggs. During the breeding periods, the substrates were regularly visited to check for the presence of any eggs. The substrates were prospected manually every 4 days. Eggs found simultaneously in the same place (buried or on the substratum of breeding) were considered to constitute a clutch. After harvesting, the eggs were counted per clutch and their dimensions (large and small diameters) were measured to the nearest millimetre using a caliper. They were then individually weighed before being incubated.

After 24 days, incubated eggs were regularly checked for hatchlings. The average incubation time of eggs from each clutch was taken as the incubation time of that clutch. Hatched were counted and hatching rates determined. The reproductive performance of the snails on each substrate type was estimated on the basis of age at first laying, number of clutches per breeder, number of eggs per clutch, weight, egg diameter and incubation time.

Total number of eggs laid per breeder (NP/R) = NP / NE

Hatching rate (%) (TE) = Necl X 100 / NŒ

NP: Total number of eggs laid

NE: Total number of snails

Necl: Total number of hatchlings

NŒ: Total number of eggs laid

Statistical analysis

STATISTICA software version 7.1 was used to carry out the various statistical treatments. The physical characteristics of the eggs (weight and diameter), the annual number of ovipositions per snail and the number of eggs per oviposition, determined on the different substrates according to the calcium source used, were compared by an analysis of variance (ANOVA) using TUKEY's HSD test at a confidence level of 5%. The hatching rates recorded as a function of calcium source and substrate amendment rate were also compared using the non-parametric one-factor Anova test (Kruskal-Wallis test).

Results

Evolution of the age at first oviposition

Fig. 4 shows the evolution of the age at first oviposition of snails reared on substrates amended at different rates with oyster shell powder and bone powder. The age at first oviposition decreased as the rate of amendment of the substrate with oyster shell powder increased. On the other hand, on substrates amended with bone meal, the age at which the snails first laid eggs remained practically constant and equal to that recorded on the control substrate, which consisted mainly of potting soil. Also, analysis of the graph shows that the age of first oviposition of snails on substrates amended with oyster shell powder was earlier than that of snails reared on substrates amended with bone powder. Oyster shell powder in the substrate therefore reduces the age of sexual maturity of the snails. Amending the substrate with bone powder, on the other hand, had no effect on the age of sexual maturity of *Archachatina marginata*.

Physical characteristics of eggs

The average weight of eggs laid by snails reared on the non-calcium-amended substrate was 1.49 ±0.14 g (Table 1). Eggs from snails reared on the substrates amended with oyster shell powder ranged from 1.5 ± 0.13 (SH5) to 1.63 ± 0.19 g (SH30). On substrates amended with bone powder, on the other hand, egg weights ranged from 1.49 ± 0.14 g to 1.52 ± 0.14 g. No significant difference was observed between the mean egg weight (1.49 ± 0.14 g) of snails reared on the control substrate and the mean egg weights of animals reared on substrates amended with bone powder. In contrast, the mean egg weights of snails on substrates amended 20% (1.6 ± 0.18 g) and 30% (1.63 ± 0.19 g) with oyster shell powder were significantly higher than the weights of eggs laid on the control substrate and those laid on substrates amended with bone powder. Also, there was no difference between the average weight of eggs collected on substrates amended with bone powder and that of eggs laid on the substrates amended with 5%, 10% and 40% oyster shell powder. The small and large diameters of eggs laid by snails reared on the control substrate were 1.16 ± 0.13 cm and 1.44 ± 1 cm respectively. On substrates amended with bone powder, the snails laid eggs with large and small diameters varying respectively between 1.44 ± 0.1 mm and 1.47 ± 0.11 mm and between 1.16 ± 0.13 cm and 1.2±0.14 cm. As for the eggs laid on substrates amended with oyster shell powder, their large diameters varied between 1.46 ± 0.11 cm and 1.54 ± 0.12 cm and their small diameters between 1.17±0.13 cm and 1.33 ± 0.93 cm. Statistical analysis showed no significant difference between the large and small diameters of eggs laid on the control substrate and those laid on the substrates amended with bone powder. Also, the dimensions of eggs laid on substrates amended with 5%, 10% and 40% oyster shell powder and those amended with bone powder were statistically identical.

Number of egg-laying

The number of eggs laid per snail per year on substrates amended with bone meal varied between 5.88 and 7.24 (Fig. 5). Analysis of the results shows that the number of eggs laid per snail decreased as the rate of amendment of the substrate with bone powder increased. The number of eggs laid per animal varied between 8.96 and 12.4 on substrates amended with oyster shell powder. The increase in oyster shell powder content led to an increase in the annual number of egg-laying per snail. Whatever the rate of amendment, the number of egg-laying using oyster shell powder remained higher than that using bone powder.

Number of eggs per laying

The highest number of eggs recorded per spawning on substrates amended with oyster shell powder was 8.18 and the lowest number was 7.61 (Fig. 6).

Table 1. Physical characteristics of eggs as a function of calcium source and substrate amendment rate

Source of amendment	Substrate amendment rate					
	0%	5%	10%	20%	30%	40%
Average weight of eggs laid						
Oyster shell powder	1.49 ± 0.14^b	1.5 ± 0.13^b	1.52 ± 0.15^b	1.6 ± 0.18^a	1.63 ± 0.19^a	1.53 ± 0.15^b
Bone powder	1.49 ± 0.14^b	1.5 ± 0.14^b	1.52 ± 0.14^b	1.49 ± 0.14^b	1.51 ± 0.15^b	1.5 ± 0.14^b
Large egg diameter						
Oyster shell powder	1.44 ± 1^b	1.46 ± 0.11^b	1.47 ± 0.11^b	1.52 ± 0.11^a	1.54 ± 0.12^a	1.48 ± 0.11^b
Bone powder	1.44 ± 0.1^b	1.46 ± 0.11^b	$1.48 \pm 0{,}1^b$	$1.44 \pm 0{,}1^b$	1.47 ± 0.11^b	1.45 ± 0.1^b
Small diameter						
Oyster shell powder	1.16 ± 0.13^b	1.17 ± 0.13^b	1.18 ± 0.14^b	1.33 ± 0.93^a	1.33 ± 0.92^a	1.19 ± 0.39^b
Bone powder	1.16 ± 0.13^b	1.18 ± 0.13^b	1.2 ± 0.14^b	1.17 ± 0.13^b	1.16 ± 0.13^b	1.18 ± 0.14^b

NB: Values in the same column and row indexed by the same letters are not statistically different in the TUKEY HSD test at a confidence level of 50%.

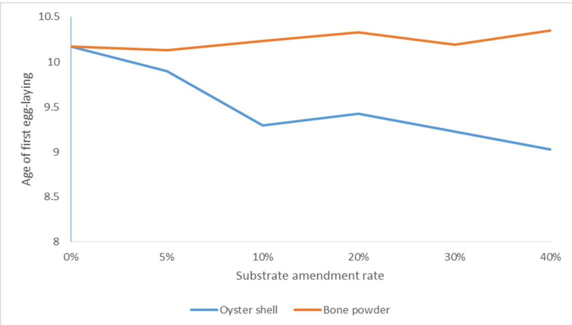
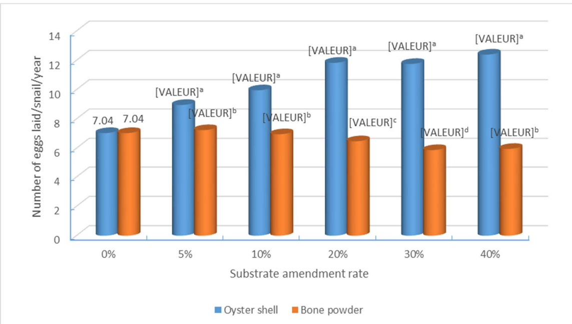


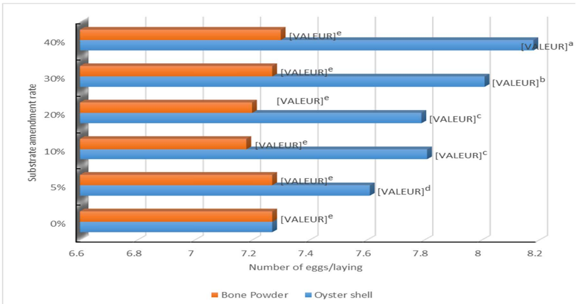
Fig. 4. Age of first snail egg-laying as a function of substrate source and amendment rate



NB: Values indexed by the same letters are not statistically different in the TURKEY HSD test at a confidence level of 5%.

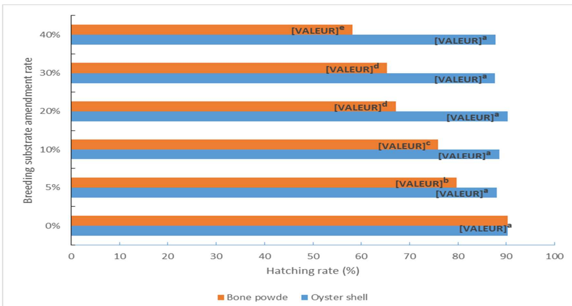
Fig. 5. Number of egg-laying events as a function of calcium source and substrate amendment rate

The increase in the content of oyster shell powder in the rearing substrate led to an increase in the number of eggs per clutch. The number of eggs per clutch on substrates amended with bone powder varied between 7.18 and 7.3. On the control substrate, there was an average of 7.27 eggs per clutch. No significant difference was observed between the number of eggs per oviposition recorded on the control substrate and those obtained on the substrates supplemented with bone powder. On the other hand, the number of eggs counted per oviposition on substrates amended with oyster shell powder was significantly better than those recorded on the control substrate and those recorded on substrates amended with bone powder.



NB: Values indexed by the same letters are not statistically different in the TURKEY HSD test at a confidence level of 5%.

Fig. 6. Number of eggs as a function of calcium source and substrate amendment rate



NB: Values indexed by the same letters are not statistically different in the Kruskal-Wallis test at a confidence level of 5%.

Fig. 7. Hatching rate as a function of calcium source and substrate amendment rate

Hatching rates

Fig. 7 shows the variation in egg hatching rate as a function of calcium source and substrate amendment rate. Eggs harvested from the substrate consisting solely of compost had an average hatching rate of 90.27%. The hatching rate of eggs laid on substrates amended with bone powder ranged from 58.21% to 79.66%. Thus, the hatching rate of eggs laid on the control substrate was better than that of eggs laid on substrates amended with bone powder. The hatching rate of eggs laid by snails reared on substrates amended with oyster shell powder varied between 90.26% and 93.43%. The hatching rates of eggs harvested from these substrates were higher than those of eggs from substrates amended with bone powder.

Discussion

The rearing substrate is the support on which the snail lives, grows and reproduces. Its chemical composition has a considerable effect on the biological performance of these animals (Stievenart and Hardouin, 1990; Memel *et al.*, 2011; Kouassi *et al.*, 2008). In fact, these animals draw almost 40% of their nutrients from the soil transcutaneously using their pedal sole (Jess, 1989). Analysis of the results of this study showed that amending the substrate with oyster shell powder reduced the time taken for Archachatina marginata to reach sexual maturity. At a substrate amendment rate of 40%, this calcium source reduced the age at sexual maturity of these molluscs by around 1 month compared with animals reared on unamended substrate (10.17 months). The earlier sexual maturity of the snails is thought to be the result of an increase in their growth rate with the addition of bioavailable calcium to their substrate (Aman *et al.*, 20 11). The results also showed that an increase in the level of oyster shell in the substrate led to an increase in the annual number of eggs laid per breeder, the number of eggs per clutch and the physical characteristics of the eggs. However, amending the rearing substrate with pig bone powder had no significant effect on the age of sexual maturity or the physical characteristics (weight and diameter) of A. marginata eggs. This suggests that the calcium in pig bone powder is not very bioavailable to snails. The hatching rate of eggs collected on substrates amended with bone powder fell as the calcium content of the rearing substrate increased. This drop in the hatching rate of eggs from breeders reared on substrates with a high level of bone meal is thought to be linked to the poor quality of the eggs laid, due to a lack of bioavailable calcium in the substrate and in the feed. The eggs laid on these substrates had very thin shells. These eggs, with their thin and very fragile shells, had undergone excessive dehydration, which is thought to be the cause of their low hatching rate. In fact, the calcium drawn by the snail from the food and the soil is precipitated in the shell, in the calcium connective cells that are widely distributed throughout the connective tissue, in the epithelial cells of the digestive gland, in the glandular cells of the skin and in the collar of the mantle. During the egg-laying period, a flow of calcium occurs through the epithelium of the reproductive tract to supply the egg shell and fluid. This calcium, precipitated in the egg fluid, is then recovered by the embryo (Fournié and Chetail, 1984).

Conclusion

The addition of oyster shell powder to the substrate helps to improve the reproductive performance of *Archachatina marginata*. It shortens the time to sexual maturity for this species of snail to around 9 months instead of 10 months. It induces a high number of egg-layings (12.04) per animal per year and a high number of eggs (8.18) per laying. This source of calcium in the substrate also improved the physical characteristics and hatching rate of Archachatina marginata eggs. Bone meal, on the other hand, although an important source of calcium, is not suitable for calcium supplementation of the rearing substrate with a view to improving the reproductive performance of *A. marginata*.

References

Ajayi SS, Tewe OO, Moriarty C, Awesu MO. 1978. Observations on the biology and nutritive value of the giant African snail *Archachatina marginata*. African Journal of Ecology **16**, 85-95. https://doi.org/10.1111/j.1365-2028.1978.tb00430.x

Aman JB, Kouassi KD, Karamoko M, Bouyé RT, Otchoumou A. 2011. Effet de la teneur en poudre de coquilles d'huîtres dans le substrat d'élevage sur la croissance d'*Archachatina marginata*. Journal of Applied Biosciences **47**, 3205–3213. https://www.m.elewa.org/JABS/2011/47/2.pdf

Aman JB, Memel JD, Kouassi KD, Otchoumou A. 2018. Fiche technique de l'impact de l'amendement du substrat en poudre de coquille d'œuf de poule sur la reproduction de *Archachatina marginata* (Swainson, 1821). European Scientific Journal **14** (**33**), 239-250. https://doi.org/10.19044/esj.2018.v14n33p239

Bouyé TR, Ocho-Anin AAL, Memel JD, Otchoumou A. 2017. Effet de l'amendement au carbonate de calcium (mikhart) de substrat d'élevage sur les performances de reproduction de l'escargot *Achatina achatina* (Linné, 1758). Journal of Applied Biosciences **109** (**1**), 10662-10662. https://doi.org/10.4314/jab.v109i1.10

Bouyé TR, Sika A, Memel JD, Karamoko M, Otchoumou A. 2013. Effets de la teneur en poudre de coquilles de bivalves (*Corbula trigona*) du substrat sur les paramètres de croissance d'*Achatina achatina* (Linné, 1758) en élevage hors-sol. Afrique Science **09**(**2**), 142 – 153. https://www.ajol.info/index.php/afsci/article/view/112091

Awohouédji DYG. 2010**.** Influence du substrat sur les performances des juvéniles et le niveau de parasitisme des géniteurs d'*Archachatina marginata* en élevage contrôlé. Mémoire de Diplôme d'Études Approfondies, Faculté des Sciences Agronomiques de l'Université d'Abomey-Calavi (UAC). **57**p.

Fournie´ J, Chétail M. 1984. Calcium dynamics in land gastropods. American Zoologist **24** (**4**), 857–870. https://doi.org/10.1093/icb/24.4.857

Hotopp KP. 2002. Land snail and soil calcium in central Appalachian mountain forest. Southeastern Naturalist **1** (**1**), 27- 44. https://doi.org/10.1656/1528-7092(2002)001[0027:LSASCI]2.0.CO;2

Johannessen LE, Solhoy T. 2001. Effects of experimental increased calcium levels in the litter on terrestrial snail populations. Pedobiologia **45** (**3**), 234-242. http://dx.doi.org/10.1078/0031-4056-00082

Memel JD, Karamoko M, Otchoumou A, Kouassi DK. 2011. Abondance, taille et mortalité des escargots terrestres du Parc National du Banco (Côte d'Ivoire): effets de la composition granulométrique et chimique du sol. Livestock Research for Rural Developpement **23** (**9**). https://www.lrrd.org/lrrd23/9/meme23195.htm

Sodjinou E, Biaou G, Codjia JTC. 2003. Commercialisation des escargots géants africains (achatines) dans les départements de l'Atlantique et du littoral au sud-Bénin. Annales des Sciences Agronomiques **4** (**2**), 161-188. http://dx.doi.org/10.4314/asab.v4i2.43374

Stievenart C, Hardouin J. 1990. Manuel des escargots géants africains sous les tropiques. Centre technique de Coopération Agricole et Rural, Pays-Bas, **35**p. https://www.researchgate.net/publication/39017439

Stievenart C. 1996. Morphologie coquillière, Croissance, Reproduction et Estivation chez les escargots géants africains : *Archachatina marginata suturalis*, *Achatina achatina* et *Achatina fulica*. Thèse de doctorat en philosophie (PH.D.) en Production Animale Tropicale. Institut de médecine Tropicale Prince Léopold, Antwerpen, Belgique, **204**p.

Jess MRJ. 1989. The interaction of the died and substrate on the growth of *Helix aspersa* (Müller) variety *maxima*. Slues and snail in word agriculture Henderson, I Edition.**1**, 311-317.

Kouassi KD, Otchoumou A, Dosso H. 2007. Les escargots comestibles de Côte d'Ivoire: influence de substrats d'élevage sur les paramètres de croissance de *Archachatina ventricosa* (Gould, 1850) en élevage hors sol. *Tropicultura* **25** (**1**), 16-20. http://www.tropicultura.org/text/v25n1/16.pdf

Kouassi KD, Otchoumou A, Gnakri D. 2008. Le commerce des escargots (*Achatina achatina*), une activité lucrative en Côte d'Ivoire. Livestock Research for Rural Development **20** (**4**).
http://www.lrrd.org/lrrd20/4/koua20058.htm.

Kouassi KD, Aman JB, Karamoko M. 2014. Growth performance of *Archachatina marginata* bred on the substrate amended with industrial calcium. International Journal of Science and Research **5**(**1**), 582-586.
https://www.ijsr.net/archive/v5i1/NOV152703.pdf

Stievenart C. 1996. Morphologie coquillière, Croissance, Reproduction et Estivation chez les escargots géants africains: *Archachatina marginata suturalis*, *Achatina achatina* et *Achatina fulica*. Thèse de Doctorat en Production Animale Tropicale. Institut de médecine Tropicale Prince Léopold, Antwerpen, Belgique, **204**p.

Otchoumou A, Dupont-Nivet M, Dosso H. 2005. Les escargots comestibles d'Afrique : Effet de la teneur en calcium alimentaire sur les performances de croissance d'*Achatina achatina* (Linné, 1758). Agronomie Africaine, **17** **(2),** 81-90.
https://doi.org/10.4314/aga.v17i2.1660

Otchoumou A, Dupont-Nivet M, Dosso H. 2011. Effects of diet quality and dietary calcium on reproductive performance in *Archachatina ventricosa* (Gould 1850), Achatinidae, under indoor rearing conditions. Invertebrate Reproduction & Development **56**(1), 14-20.
http://dx.doi.org/10.1080/07924259.2011.588013