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The effect of different incubation media of eggs of *Archachatina marginata* **(Swainson, 1821) ovum in the University of Calabar, Nigeria**

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ABSTRACT

Hatchability of snail eggs in a cage is normally impaired as they are often unearthed by other snails looking for laying sites. This study evaluated the effects of different incubation media on egg hatchability of the giant African land snail (*Archachatina marginata* ovum) over a seven-week period. The experiment was conducted at the Department of Forestry and Wildlife Resources Management Teaching and Research Farm, University of Calabar, Calabar, Nigeria. One hundred eggs were collected from 2 boxes stocked with adult snails. The eggs were incubated in 4 media comprising river sand, top soil, sawdust, mixture of river sand, top soil and saw dust. Seventy-seven eggs were randomly assigned to each medium having six replicates of three eggs each incubated in a box measuring top diameter 11. 5 cm, bottom diameter 11 cm and height 9.5 cm. Data were collected on daily temperature (morning, and evening), length of incubation and % hatchability and subjected to one way analysis of variance in a completely randomized design. Duncan's Multiple Range Test was used to separate the means. Mean daily temperature ranged from 25.86-26.03 which was not significantly (P>0.05) different between incubation media. Incubation period ranged from 22 -30 days and was significantly (P<0.05) different between media. Eggs incubated in sawdust hatched earlier (25 days), followed by those in a mixture of river sand and sawdust, topsoil (26 days), river sand (27 days), and top soil gave the longest incubation period (28 days). Percentage Egg hatchability differed (P<0.05) significantly between incubation media. Sawdust gave 66.67 % hatchability, 50% was recorded in a mixture containing river sand and top soil and sawdust, river sand has 41.67% while topsoil gave the least hatchability of 25 %. It was thus

recommended that saw dust could be adopted as the ideal incubating medium for eggs of *Archachatina marginata* Saturalis.

*Keywords***:** incubation media, incubation period, % hatchability, temperature, *Archachatina marginata*

1. INTRODUCTION

The increasing focus on breeding and rearing non-traditional livestock species, such as the Giant African Land Snail (*Archachatina marginata* ovum), underscores the potential these species have in addressing food security, biodiversity conservation, and economic diversification. Indigenous to tropical regions of Africa, including Nigeria, these snails offer potential benefits in terms of protein sources, traditional medicine, and economic opportunities (Bode & Olorunshola, 2020). Research and commercial interest have surged as efforts are made to exploit their reproductive potential sustainably. The reproductive cycle of Archachatina marginata Ovum involves intricate stages, with egg incubation being particularly critical. This phase requires precise environmental management to ensure successful hatching and healthy juvenile development, impacting overall reproductive success and viability (Igwe, Ndubuaku, Ezea, & Eze, 2021).

The African giant land snail, *Archachatina marginata* (Swainson) is a pulmonate gastropod mollusc that inhabits the dense forest and the fringe forest of savanna where the conditions are favourable. This animal is rare in brackish areas and absent in arid and desert zones. The Giant African Land Snails are nocturnal, hermaphroditic gastropods of the Family Achatinidae. They are indigenous to Africa and are distributed in sub-Saharan Africa, ranging from the Gambia in the West to the Lake Chad region in the East. Нeir distribution extends southwards to the Orange River in South Africa (Hodasi, 1998). African giant land snail (*Archachatina marginata*) is one of the largest known land snails in West Africa. Most land snails dwell naturally in the forest litters of dense tropical high forest to fringing riparian forests of the derived guinea savannah in Nigeria. Land snails are non-timber products of forest that thrive under captive setting in humid environment. Many people use snail meat to complement their conventional and regular sources of animal protein. Nigeria is known to be richly endowed with different species of snails which vary in sizes, colors, adaptability and performance (Ibom, 2020). These land snail species include *Archachatina marginata, Achatina achatina, Achatina fulica, Limcolaria* spp*,* and other garden snails.

Snail farming, or heliciculture, has emerged as a viable agricultural practice with the potential to address issues of protein deficiency and income generation in diverse regions of the world (Hart & Moreno, 2020). The cultivation of edible snails presents an opportunity to bridge the gap between protein demand and supply, especially in regions where access to traditional livestock is limited. *Archachatina marginata* Ovum, commonly referred to as the Giant African Land Snail, has gained prominence as a key species for snail farming due to its unique attributes and economic significance.

Archachatina marginata ovum stands out among the plethora of snail species suitable for farming due to its remarkable adaptability to a range of environmental conditions. Its ability to thrive in various ecological settings, including tropical forests and cultivated areas, makes it a suitable candidate for both subsistence and commercial farming (Okoli et al., 2021). The species' adaptability contributes to its relatively higher survival rates in captivity, making it an

attractive choice for snail farmers seeking consistent production. Furthermore, the relatively large size of *Archachatina marginata* ovum distinguishes it from other snail species, enhancing its appeal from a consumption perspective. The substantial meat content of the snail, coupled with its low fat and cholesterol levels, positions it as a nutritious protein source. This dietary profile aligns with modern dietary preferences, as consumers seek healthier protein alternatives to diversify their nutritional intake.

Beyond its dietary attributes, *Archachatina marginata* ovum holds cultural and economic significance. Indigenous communities in West Africa have long recognized the snail's value in traditional medicine, using its extracts for various therapeutic purposes (Saliu et al., 2020). This utilization further underscores the multifaceted potential of the species.

Incubation of Giant African Land Snail eggs necessitates a careful balance of temperature, humidity, substrate composition, and ventilation. Variations from optimal conditions can lead to poor hatching rates, developmental issues, and reduced hatchling survival (Ofojekwu et al. 2020). The environment experienced during incubation has long-term effects on the snails' growth and resilience. Effective incubation practices are crucial for promoting robust growth, ensuring strong juvenile snails, and supporting the sustainability of breeding programs. Understanding and optimizing these conditions are essential for enhancing the success of snail farming initiatives, which could contribute significantly to local economies and biodiversity conservation efforts (Oke & Agbaje 2020).

Egg incubation represents a pivotal and intricate phase within the life cycle of snails, wielding a profound influence over the fate of the ensuing generation. This critical period, characterized by the transformation of embryonic structures and the accumulation of vital resources, is a cornerstone of successful reproduction in snails. A multitude of interwoven factors, including temperature, humidity, and substrate composition, converge to orchestrate the delicate dance of embryogenesis, ultimately determining the prospects of hatchling survival and subsequent growth.

Temperature, perhaps one of the most influential environmental determinants, exerts a paramount impact on the developmental trajectory of snail embryos. As elucidated by Eze and Oke (2020), temperature regimes directly impinge upon the rate of embryonic development, with higher temperatures often accelerating metabolic processes and subsequently shortening the incubation period. Conversely, cooler temperatures can extend the developmental timeline, potentially bestowing the embryo with additional time for resource acquisition and structural refinement. The pivotal role of temperature in snail egg incubation has been demonstrated through empirical studies such as those by Ikeobi et al. (2021), wherein varying temperature conditions were shown to induce disparities in hatching success rates and developmental timing.

Humidity, an oft-overlooked yet equally critical factor, plays an integral role in egg incubation by influencing moisture levels within the egg capsule and regulating gas exchange. The delicate balance between water retention and evaporation profoundly affects the embryonic microenvironment, influencing cell division, gas diffusion, and waste elimination (Akinneye et al., 2019). Perturbations in humidity levels can lead to desiccation or suffocation of developing embryos, underscoring the necessity of maintaining optimal humidity conditions to ensure successful hatching (Aina et al., 2022).

Substrate composition, encompassing both the physical structure and chemical composition of the incubation medium, represents another facet of paramount importance. The composition of the substrate can influence water availability, gas permeability, and nutrient

accessibility, all of which bear direct consequences on embryo development. Studies by Ofojekwu et al. (2020) have highlighted the significance of substrate composition in *Archachatina marginata* ovum incubation, revealing that specific substrate formulations can engender variations in hatching success rates and hatchling vitality.

The profound impact of incubation methods on hatchling survival rates, growth rates, and overall breeding success cannot be overstated. Suboptimal incubation conditions can yield diminished hatchling vigor, compromised immune systems, and heightened susceptibility to environmental stressors (Nnadi & Igboeli, 2021). Conversely, meticulous management of incubation parameters can furnish hatchlings with a robust physiological foundation, enhancing their ability to cope with post-hatching challenges and fostering a higher likelihood of successful establishment in subsequent life stages (Oke & Agbaje, 2020).

In the realm of snail breeding, recent years have witnessed a significant paradigm shift with the emergence of modern incubation techniques (Eze & Oke, 2020). These innovative approaches are underpinned by advancements in technology and a deeper understanding of the physiological requirements of snail embryogenesis. Central to these contemporary methods is the utilization of controlled incubation systems, commonly known as incubators, designed to offer a meticulously regulated environment that optimizes the conditions necessary for successful egg development. Through precise control over factors such as temperature, humidity, and ventilation, modern incubation techniques aim to enhance hatchling rates, promote uniformity in growth, and ultimately amplify breeding success.

Central to the effectiveness of modern incubation techniques is the incorporation of specialized incubators. These devices represent the pinnacle of technology-driven snail breeding, facilitating precise control over the incubation environment. Temperature, a pivotal factor that profoundly influences the developmental trajectory of embryos, can be precisely maintained within the optimal range conducive to embryonic growth (Eze & Oke, 2020). The capacity to manipulate humidity levels offers a fine-tuned approach to thwarting desiccation or over-humidification, critical for the prevention of developmental anomalies (Capinera, 2017). Ventilation, often overlooked in traditional methods, is managed to ensure adequate gas exchange, preventing the accumulation of detrimental metabolic byproducts.

Modern incubation techniques have demonstrated their efficacy in generating higher hatchling success rates and fostering consistency in hatchling quality. Studies, such as those conducted by Amoah et al. (2020), have illuminated the advantages of controlled incubation methods. These techniques not only yield greater hatchling survival but also ensure a higher degree of synchronicity in hatching, leading to more uniform developmental stages. Additionally, the precision of modern incubation techniques minimizes the risk of developmental aberrations, contributing to robust hatchlings with enhanced physiological resilience.

The adoption of modern incubation techniques extends beyond commercial snail farming, carrying implications for conservation efforts. Given the vulnerability of wild populations to overharvesting and habitat degradation, controlled breeding through advanced incubation methods can mitigate pressures on natural populations. This conservation-oriented perspective underscores the broader impact of modern techniques in preserving genetic diversity and supporting the long-term sustainability of snail species.

The theoretical framework for studying the different incubation methods of *Archachatina marginata* ovum eggs at the University of Calabar draws upon Ecological Systems Theory, introduced by Urie Bronfenbrenner in 1979. This theory provides a multi-layered perspective

on how various environmental and contextual factors influence the development and outcomes of snail egg incubation. It highlights the significance of the microsystem (immediate incubation conditions), mesosystem (interactions between incubation factors), exosystem (external influences such as management practices), macrosystem (broader cultural and economic contexts), and chronosystem (temporal changes) in shaping the effectiveness of incubation methods. This comprehensive framework aids in understanding the complex dynamics between different ecological layers and their impact on snail reproductive success.

The empirical reviews of various studies offer insightful perspectives on the impact of different incubation methods on the development and outcomes of *Archachatina marginata* ovum eggs. Ikeobi et al. (2021) elucidate the critical role of temperature regulation during incubation, highlighting its influence on successful hatching and post-hatching growth. The study further revealed that maintaining a stable and ideal temperature during incubation is essential for promoting successful hatching and ensuring subsequent growth. The researchers demonstrated that controlled temperature conditions led to higher rates of hatching success, emphasizing the critical role of temperature in shaping the transition from embryos to hatchling. Oyewole and Sowemimo (2020) investigate the economic potential of snail farming in southwestern Nigeria.

Their study underscores that snail farming provided a source of income for both rural and urban communities, supporting livelihoods and economic development. The snail's contribution to income generation was particularly pronounced among small-scale farmers, demonstrating its role in poverty alleviation and empowerment. Eze and Oke (2020), explored the impact of various incubation temperatures on the development and hatchability of *Archachatina marginata* eggs. The study highlighted the significance of specific temperature ranges in relation to enhanced hatchability rates and the emergence of healthier hatchlings. It also pinpointed out the critical role that temperature plays in the developmental processes of these eggs by identifying optimal temperature ranges, the study provided insights into creating more favorable conditions for successful hatching and the subsequent growth of *Archachatina marginata.* Ofojekwu et al. (2020), assessed the impact of substrate composition on the hatchability of snail eggs.

The study demonstrated that the choice of substrate significantly affected the success rates of hatching by carefully examining various substrate formulations, the study highlighted the pivotal role of substrate composition in the incubation process. The study underscore the importance of selecting appropriate substrates to create optimal conditions for the successful development and hatching of snail eggs and the interplay between substrate choice and hatching outcomes, providing valuable insights for improving incubation practices and promoting better overall reproductive success in snails.

Oke and Agbaje (2020), explored the effects of different incubation temperatures on the growth and survival of hatchling snails. The study highlighted the crucial connection between incubation conditions and the long-term well-being of the snails after hatching by investigating the effects of different temperature settings, the study emphasized the significance of the incubation environment in shaping the subsequent growth trajectories and overall survival rates of the snails and the enduring influence of early incubation conditions on the snails' posthatching outcomes, contributing to a deeper understanding of the complex interplay between environmental factors and the development of these organisms.

Amoah et al. (2020) delve into the intricate relationship between incubation temperature and hatching success of *Archachatina marginata*. By meticulously analyzing controlled incubation scenarios, they unveil a direct correlation between optimized temperature conditions and enhanced hatching success rates. These empirical reviews collectively address various dimensions of snail egg incubation, spanning temperature, humidity, substrate composition, economic implications, and post-hatching growth. Each study contributes to a deeper understanding of the factors influencing snail development and outcomes, thus this study different incubation methods of eggs of arch Achatina marginata ovum in the University of Calabar, Nigeria is to advance the field of snail breeding, conservation, and economic enhancement.

This study aims to address the gaps in current knowledge by evaluating different incubation media, their impact on hatchability rates, incubation periods, and temperature variations. By scientifically assessing and comparing these methods, the study seeks to develop evidence-based protocols that can improve the efficiency and effectiveness of snail breeding practices. The results of this research will not only benefit local farmers and the broader community but also contribute to the scientific understanding of snail reproduction and sustainable agricultural practices.

2. MATERIALS AND METHODS

The study will be carried out at the snail unit of the Department of Forestry and Wildlife Resources, University of Calabar. The University of Calabar is situated in Cross River State, South-south part of Nigeria. It is one of the Nigeria's second-generation Universities, and was a campus of University of Nigeria Nsukka until 1975. Calabar lies between latitude 4° 57ʹ 40″and 4° 57ʹ 39″N and longitude 8° 18ʹ 28″and 8° 18ʹ 27.8″E, with elevation of 32m (Mfam, 2022).

The wet season starts from Mid-March to early November, while the dry season spans from Mid-November to early March. The annual rainfall ranged between 2,500 mm and 3000 mm (Table 1), while the relative humidity varies from 60 percent to 70 percent in January and 70 percent to 80 percent in July, with a mean monthly temperature of 27 °C. The extreme low temperature in July is attributed to cloud effect and high humidity. Usually, strong wind normally accompanied the on-set of dry seasons which is caused by North East wind.

Parameters	> anuar ➡	February	March	April	May	June	July	August	September	October	November	December
Average Temperature $(^{\circ}C)$	24	25	26	25	25	24	23	23	23	24	24	25
Average Rainfall (days)	6	9	16	12	18	29	31	31	30	28	15	3
Average R. Humidity $(\%)$	69	74	76	77	81	85	87	87	87	84	79	70
Average Sun Hours (hours)	360	305	335	330	325	275	230	200	260	310	330	360

Table 1. Climatic Data for Calabar from 2012 to 2021.

Source: Worldwideweatheronline.com-Calabar

The harmattan, which significantly influences weather in West Africa, is noticeably less pronounced in the city. As equatorial climate, sun shine hours is relatively high especially toward the end and the beginning of the year (Table 1).

Figure 1. Map of University of Calabar, showing study area. Source: Mfam, (2022)

2. 1. Vegetation of Calabar

The vegetation lies within the lowland rainforest vegetation. The structure and physiognomy of the vegetation is highly stratified with a heterogenous floristic composition. It exhibits a three layer (namely: upper storey, middle storey and under storey) of heavy branched trees of about 35m high in area of little or no disturbance. Many plant species may be recognized within this area including;Spear grass (*Imperata cylindrica*), Siam weed (*Chromolaena odorata*), Nut grass (*Cyperus rotundus*), Milk weed (*Euphorbia heterophylla*), Nuke-Noh (*Tridax procumbens*), and Witchweed (*Striga genus*) (Inyang *et al.,* 2021)

2. 2. Experiment

Plastic jar with (top diameter 11.5 cm, bottom diameter 11 cm and height 9.5 cm) with perforated lids and bottoms for aeration and to prevent water logging, were constructed for the study, each plastic jar was filled up with different incubation substrate, sawdust, topsoil, river sand and mixture of river sand, topsoil, sawdust. All the substrate were sterilized before being use in the experiment according to the method highlighted by Ogogo et al (2023). Before the eggs were assigned to incubating medium, to destroy any potential microbes in the soil. Hundred (100) snails were purchased from Ikom local government area of cross river state, Nigeria. They were managed intensively; feed and water were provided adlibitum for one month prior to collection of experiment eggs. The snails were reared with a mixture feed comprising of carica papaya (unripe pawpaw fruit and leaves) and concentrated feed recommended by Amuode and Ogogo (1995). Eggs were embedded in each medium to a depth of 3 to 5 cm and lightly covered with the incubation substrate. Seventy-seven (77) freshly laid snail eggs were collected from the breeder snails were randomly distributed in four (4) plastic jar each with a different substrate. (River sand, topsoil, sawdust and mixture of River sand, topsoil, sawdust). the substrate was sprinkled with water once a day to prevent dehydration. the temperature in each medium was measured daily in the morning (7.00 hr), and in the evening (18.00 hr) throughout the experiment using digital thermometer. The average daily temperature is 25 ℃, while the wind speed/direction is 8.1 km/hr west Google Earth (2022).

2. 3. Preparation of site

During the preparation of site various condition were considered to ensure the success of incubation process. Four (4) suitable plastic jars with top diameter 11.5cm, bottom diameter 11cm and height 9.5cm that is capable to accommodate the number of eggs were made available. Suitable substrate; T1: River sand was obtained from the river, the river sand was sterilized using the local method, to destroy any potential microbes in the soil, the river sand was allowed to cool before been put into the plastic jar. T2: Topsoil was obtained from the upper layer of the soil, the topsoil was sterilized using the local method, to destroy any potential microbes in the soil, the topsoil was allowed to cool before been put in to the plastic jar. T3: Sawdust was obtained from the timber market; the sawdust was not sterilized. T4: Mixture of river sand +topsoil+ sawdust.

2. 4. Material used

The research material used in the experimental work Include: The snails used for the experiment (*Archachatina marginata* ovum) were gotten from Ikom local government area in cross river state. They were healthy mature snail 100 in number. The plastic jar used for the experiment was divided in to four to cover for replication in the treatment administered. The size of the plastic jar was top diameter 11.5 cm, bottom diameter 11 cm and height 9.5 cm that is capable to accommodate the number of eggs were made available.A clean spoon was used in carrying the egg from the compartment to incubating plastic jar.

2. 5. Experimental layout

T1: RIVER SAND T2: TOP SOIL

T3: SAWDUST

T4: MIXTURE OF RIVER SAND +TOP SOIL+SAWDUST

Table 2. Experimental layout

2. 6. Data collection and statistical analysis

Data collected include: incubation period (days) and total number of Eggs hatched per medium, Daily monitoring of temperature in each incubation setup. Hatch eggs were counted visually and estimated as a percentage of total number of eggs per medium.

2. 7. Data analysis

The data collected were subjected to a one- way analysis of variance in a completely randomized design, using GENSTAT for windows (version 8.0) computer software. Means were separated using Duncan's Multiple Range Test procedures (Duncan, 1955) and values were accepted at 5%probability.

3. RESULTS AND DISCUSSION

Table 1 shows the effects of different incubation media on egg hatchability traits of African giant and snails. Mean Temperature in the incubating media ranged from 25.86 - 26.03 °C which did not differ significantly ($P > 0.05$) between media.

The highest mean temperature was recorded in river sand followed by saw dust, mixture of river sand plus top soil, top soil with the least temperature in mixture of river sand and saw dust. These values were higher than media, saw dust was suitable for growth of temperature $(10 - 23 \degree C)$ reported for optimum snail performance (FAO, 1989) but within the values (22.6-31.5 °C) reported by Mobo et al., (2013). Temperature at the different periods of the day (morning and evening) were not also different between incubating media. Temperature in the morning was lower than those observed in the evening in line with sunlight intensity.

There were significant $(P < 0.05)$ differences in the incubation period between incubating media. Eggs on saw dust hatched earlier (25days) followed by those on mixture of rivers and, saw dust, top soil (26), river sand (27 days), and top soil (28 days) being the longest as shown in Figure 3 above. This result agrees with previous reports that saw dust encourages earlier hatching of snail eggs (Bamimore, 2020).

Egg percentage hatchability was highest ($P < 0.05$) in saw dust medium than in mixtures of river sand and least in top soil medium as illustrated in figure 2. Higher hatchability in saw dust medium could be due to better aeration and higher moisture retention in the medium compared to other incubating media. Furthermore, it was high because as sawdust decay it generates heat, which promote hatchability of the eggs. High incubation rate correlates with high moisture content (Agbelusi and Adeparusi (1999).

Hatchability of 64.4% and 52 - 76% have been reported by Amubode and Ogogo (1995) and Ukwu *et al.,* (2011), respectively in snails comparable to values obtained in saw dust and soil mixtures of media. This result contradicts the report of Amata (2021) who reported that whereas top soil and river sand were preferred hatching media, saw dust was suitable for growth of hatchings.

Variations reported could be due to the differences in soil texture, geographical location of study and methodology adopted. Significantly lower hatchability was recorded in top soil medium whereas a 100% hatchability had been previously reported. (Bamimore,2020; Amata, 2021) in *Archachatina marginata* snails.

Table 3. Effects of incubating media on egg hatchability traits.

a, b. c, d: Mean with different superscripts indicate significant (P<0.05) difference between incubating media

Figure 2. Numbers of hatched eggs.

Figure 3. Incubation period.

The relatively lower hatchability recorded in the present result could be due to the high temperature in the media and the period of experiment. Top soil contains decomposing plant material that generates heat. High temperature results in a high number of undeveloped eggs (Agbelusi and Adeparusi, 1999). For improved hatchability, optimum temperature (20-25 °C, (www.stevegreaves.com, 2016) and climatic condition in the incubating medium should be encouraged.

Mateo (2021) noted that the percentage of unhatched eggs on sawdust, plantation soil, virgin forest soil, coconut husks and absorbent cotton could be because they are parasitized, desiccated or not fertilized. In this study sterilization was practiced to reduce parasitic contamination of incubating media. Dedi *et al.* (2021) also opined that increased temperature or moisture can cause appearance of small maggots which attack the seed oysters and eggs; that leads to the putrefaction of the substrates with a considerable reduction of the rate of hatching.

4. CONCLUSION

The study on *Archachatina marginata* ovum reveals that the choice of incubation media significantly affects the hatchability and survivability of snail eggs. It was found that sawdust is the most effective medium for hatching, yielding better results than sole river sand or topsoil. Additionally, a mixture of river sand and topsoil is a viable alternative when sawdust is unavailable. The research suggests that hatchability improves further at lower temperatures when using sawdust. Therefore, it is recommended to use sawdust for incubating snail eggs, or alternatively, a mixture of river sand and topsoil if sawdust is not available.

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