

## PARASITIC PROFILE OF FIVE SPECIES OF TERRESTRIAL *Achatina* SNAIL IN CROSS RIVER STATE, NIGERIA: PUBLIC HEALTH IMPLICATIONS

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### AUTHORS' CONTRIBUTIONS

This work was carried out in collaboration among all authors. Author JOM designed the study and wrote the first draft. Author EEI performed the statistical analysis and managed the literature searches. Authors COA and EEI managed the analyses of the study. All authors read and approved the final manuscript.

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### ABSTRACT

A total of 760 snails of the genera *Achatina*, belonging to five species (*Achatina achatina*, *Achatina belteata*, *Achatina degneri*, *Achatina fulica* and *Achatina marginata*) were sampled between January and August 2021, from six communities located in six Local Government Areas (LGAs) in the Central Senatorial District, Cross River State. *A. achatina* was the most abundant species collected (32.89%) while *A. degneri* was the least collected (11.19%). A greater number of *Achatina* snails were collected in the wet season than in the dry season with no observed dominance of any of the species. Overall, 319 (42%) snails were infected with parasites. *A. fulica* had the highest prevalence of parasitic infection (50.50%) while *A. marginata* had the least parasitic infection (28%). Snail species sampled in Boki LGA had the highest prevalence of parasitic infection (56.25%), while Obubra LGA recorded the least prevalence (21.28%). Mean intensity of *Angiostrongylus* spp. in *A. achatina* was 4.780 (4.56 – 5.00; 95% CI), while *Strongyloides* spp. was 4.667 (4.11-5.22; 95%CI). Testing parasite species diversity in the snail species assessed using diversity indices, *A. belteata* recorded the highest values for Shannon-Wiener (1.653) and Margalef's indices (1.995), and also for species dominance using the Simpson index (0.22). Public health education and provision of adequate toilet facilities are recommended for control of snail-borne parasites.

**Keywords:** Snail; *Achatina* spp; *angiostrongylus*; Cross River State; Nigeria.

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## 1. INTRODUCTION

Giant African land snails belonging to the gastropod family Achatinidae, are natives of Africa and represents about 200 species in 13 genera [1]. They can be found throughout Sub-Saharan Africa. Snails could be grouped based on their habitats into two groups- terrestrial and aquatic snails [2]. Snails live in habitats that are often moist and *Achatina* species are usually restricted to humid forest areas where they exist in large numbers. In as much as they have different habitats, snails are host to many parasites [3].

The Giant African land snail is an intermediate host for several parasites including *Angiostrongylus cantonensis*, *Schistosoma mansoni*, *Hymenolepis* spp. and *Fasciola hepatica* [4], which cause severe diseases in humans.

Specifically, the Giant African land snail is the main gastropod responsible for worldwide spread of *Angiostrongylus cantonensis* that cause human eosinophilic meningoencephalitis which result in severe neurological disorder [2]. Despite harbouring disease causing organisms, land snail meat has become an important delicacy for many Africans [5]. *Achatina fulica* is sometimes processed and sold to consumers as escargot [2]. In many parts of Africa and West Africa, *Achatina achatina* is served as a delicacy. For example, in Cameroon, it is a delicacy called 'nyamangoro', and in Northern Morocco, small snails are eaten as snacks in spicy soup [2].

Snail meat consumption is both nutritional and medicinal. The mineral composition of *Achatina* species has been reported to include zinc, iron, magnesium, calcium, sodium, potassium, and phosphorus [6]. In some parts of Nigeria, *Achatina marginata* haemolymph is used for oral rehydration therapy and also administered orally for the treatment of diarrhea and vomiting [7]. Despite these beneficial attributes of the Giant African land snail, its consumption could be a route to human infection with parasitic diseases, particularly when eaten raw or undercooked. A recent study has shown the susceptibility of the Giant African land snail to rat lungworm parasite [8], with the increase risk of transmission to man and animal [9].

Parasitic infections of *Achatina* species are enormous and cannot be overemphasized. Therefore, this study investigates the helminth parasite status of some

edible land snails in the Central Senatorial District of Cross River State.

## 2. METHODOLOGY

### 2.1 Study Area

The study was carried out in six communities in the Central Senatorial District, Cross River State, Nigeria. These communities are located in six Local Government Areas (LGAs), namely, Abi, Boki, Etung, Obubra, Ikom, and Yakurr. One community was selected from each of the six LGAs. Cross River State is characterized by tropical rainforest vegetation, with two distinct seasons – the dry and wet seasons. The dry season range from November to March while the wet season, April to October. Cross River State is located on latitude 4°34'59.99"N and longitude 8°24'59.99"E. Deep sea fishing and farming is the main occupation in the state. These activities are encouraged by the availability of water bodies that surround the state.

### 2.2 Sample Collection

This study was conducted between January and August 2021. Seven hundred and sixty (760) samples of snails were randomly collected from different quarters of the six communities selected in the Central Senatorial District of Cross River State, Nigeria. They were handpicked from different niches such as undersides of logs and leaves, farmlands, bushes, dumpsites, buttresses of large trees and base of houses. The snails were sorted according to size, identified and afterwards examined for endoparasites. Identification of snail species was carried out using keys provided by Herbert & Kilburn [10].

### 2.3 Identification

Snails were identified according to their shape, size, markings, color, spire angle, sculpture and aperture form as described by Raut & Baker [1] and Herbert & Kilburn [10].

### 2.4 Isolation and Parasitological Examination

Isolation of parasites was carried out according to methods described by Cheesebrough [11] and Onyishi et al. [2]. The method described by Onyishi et al. [2] involves dipping the snail in a water container and exposing it under electric light for two hours. Thereafter, transferring the content to a petri dish and examining it for parasites. The shell of each snail was broken to expose the body of the snail. The snail species were further dissected to expose the entire

viscera, including the stomach and intestine. These were separately dissected and teased in petri dishes containing normal saline, and examined using wet preparation techniques. The contents of the alimentary canal were further processed using the formal ether concentration technique. The tissues of the snails were also digested within 1hr at 37°C, as modified from Wallace and Rosen [12]. The isolated parasites were identified using temporary mounts following identification keys of Cruz and Mills [13] and Yamaguti [14].

## 2.5 Data Analysis

Data obtained were analyzed using Statistical Package for Social Sciences (SPSS) version 20 and Microsoft Office Excel 2007. Data obtained were presented using descriptive statistics. Differences in prevalence of parasitic infection among the different species of land snails were tested using the Chi-square test. Mean intensity calculations and analysis for prevalence of infection were done using SPSS. Shannon Wiener index was used to determine the distribution of the snail species during sampling. Significant levels were set at  $P \leq 0.05$ .

## 3. RESULTS

Seven hundred and sixty (760) snails belonging to one genus - *Achatina* (Lamarck 1799), were collected and examined for helminth parasite infection. The snail species collected includes, *Achatina achatina*

(Linnaeus 1958), *Achatina fulica*, *Achatina marginata*, *Achatina belteata* (Reeve 1849) and *Achatina degneri* (Bequaert and Clench 1936) (Table 1). *A. achatina* (32.89%) was the most abundant in the Central Senatorial District with the highest number (N=60) collected in August, followed by *A. fulica* (26.31%); the least abundant was *A. degneri* (11.18%) (Table 1) (Fig. 1). There was no complete dominance of any of the snail species across the months ( $D = \{pi^2 = 0.1436\}$ ). Similarly, the Shannon-Wiener index value ( $H = \{piln(pi) = 2.0061\}$ ) indicated that there was almost equal abundance of snails from the six species across the months sampled (Table 1).

Out of the 760 snails examined, 319 (41.97%) were infected (Table 2). *A. fulica* had the highest prevalence of infection (50.50%) followed by *A. achatina* (42.80%) and *A. belteata* (42.40%). The least (28.00%) was observed in *A. marginata*. The disparity in prevalence of infection showed no significant variation ( $P = .18$ ).

Based on the location, Ikom LGA (n=215) recorded the highest number of snails collected. This was followed by Boki LGA (n=160) and the least was collected in Yakurr LGA (n=66). Snails collected in Boki LGA recorded the highest infection (56.25%) while snails collected in Obubra LGA recorded the least infection (21.28%) (Table 3). Prevalence of infection according to location showed significant variation ( $\chi^2 = 17.253$ ,  $df = 5$ ,  $P = .004$ ).

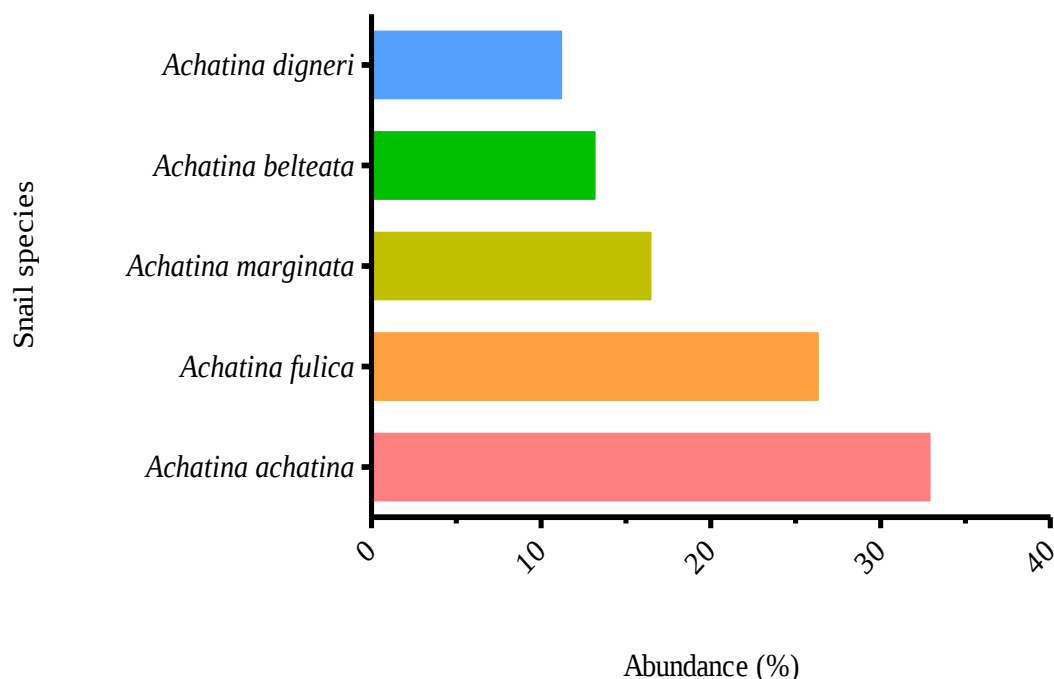


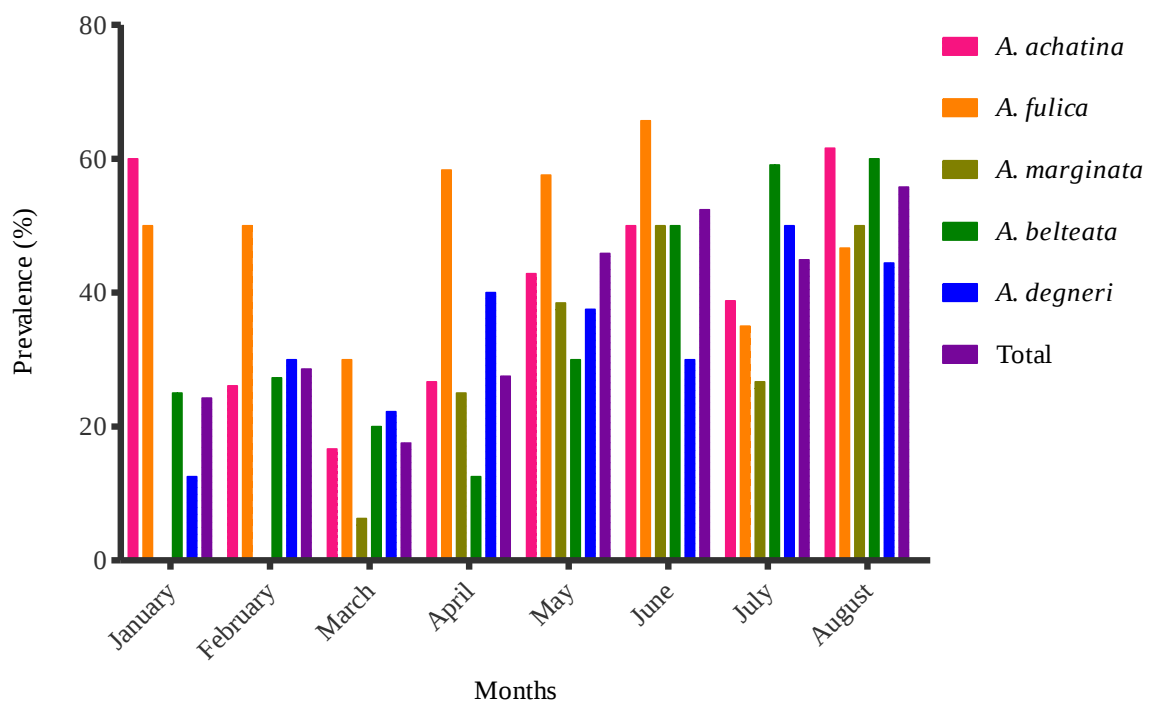
Fig. 1. Abundance of snail species in the Central Senatorial District, Cross River State

**Table 1. Species-Specific distribution of monthly collected snail species in selected communities in the Central Senatorial District, Cross River State**

| Month    | <i>A. achatina</i> | <i>A. fulica</i> | <i>A. marginata</i> | <i>A. belteata</i> | <i>A. degneri</i> | Total | D (pi) | D (pi <sup>2</sup> ) | H (pi ln pi) |
|----------|--------------------|------------------|---------------------|--------------------|-------------------|-------|--------|----------------------|--------------|
| January  | 20                 | 10               | 5                   | 12                 | 8                 | 55    | 0.0724 | 0.0052               | -0.1901      |
| February | 23                 | 18               | 9                   | 11                 | 10                | 71    | 0.0934 | 0.0087               | -0.2214      |
| March    | 12                 | 10               | 16                  | 10                 | 9                 | 57    | 0.0750 | 0.0056               | -0.1943      |
| April    | 15                 | 24               | 12                  | 8                  | 10                | 69    | 0.0908 | 0.0082               | -0.2179      |
| May      | 21                 | 33               | 13                  | 10                 | 8                 | 85    | 0.1118 | 0.0125               | -0.2450      |
| June     | 50                 | 35               | 20                  | 32                 | 10                | 147   | 0.1934 | 0.0374               | -0.3178      |
| July     | 49                 | 40               | 15                  | 22                 | 12                | 138   | 0.1816 | 0.0330               | -0.3098      |
| August   | 60                 | 30               | 10                  | 20                 | 18                | 138   | 0.1816 | 0.0330               | -0.3098      |
| Total    | 250                | 200              | 100                 | 125                | 85                | 760   | 1.0000 | 0.1436               | 2.0061       |

**Table 2. Prevalence of helminth parasite in snail species**

| Samples             | No. Examined | No. Infected | % Infection | $\chi^2$ | P value |
|---------------------|--------------|--------------|-------------|----------|---------|
| <i>A. achatina</i>  | 250          | 107          | 42.80%      | 7.639    | .18     |
| <i>A. fulica</i>    | 200          | 101          | 50.50%      |          |         |
| <i>A. marginata</i> | 100          | 28           | 28.00%      |          |         |
| <i>A. belteata</i>  | 125          | 53           | 42.40%      |          |         |
| <i>A. degneri</i>   | 85           | 30           | 35.29%      |          |         |
| Total               | 760          | 319          | 41.97%      |          |         |

**Fig. 2. Prevalence of parasitic infection in snails according to seasons**

**Table 3. Prevalence of infection in snails collected according to location**

| Location | No Examined | Total no. Infected (%) | <i>S. stercoralis</i> | <i>A. cantonensis</i> | <i>F. gigantica</i> | <i>D. dendriticum</i> | <i>S. mansoni</i> | <i>A. lumbricoides</i> |
|----------|-------------|------------------------|-----------------------|-----------------------|---------------------|-----------------------|-------------------|------------------------|
| Abi      | 105         | 40 (38.10%)            | 40 (100%)             | 7 (17.50)             | 2 (5.00)            | 5 (12.50)             | 2 (5.00)          | ---                    |
| Boki     | 160         | 90 (56.25%)            | 90 (100%)             | 12 (13.33)            | 5 (5.56)            | 10 (11.11)            | 4 (04.44)         | ---                    |
| Etung    | 120         | 44 (36.67%)            | 44 (100%)             | 3 (6.82)              | 6 (13.64)           | --                    | 6 (13.64)         | ---                    |
| Ikom     | 215         | 100 (46.51%)           | 100 (100%)            | 15 (15.00)            | ---                 | ---                   | 5 (5.00)          | 2 (2.00)               |
| Obubra   | 94          | 20 (21.28%)            | 20 (100%)             | ---                   | ---                 | ---                   | ---               | 3 (15.00)              |
| Yakurr   | 66          | 25 (37.99%)            | 25 (100%)             | 3 (12.00)             | ---                 | 7 (28.00)             | ---               | 5 (20.00)              |
| Total    | 760         | 319 (41.97)            | 319 (41.97)           | 40 (5.26)             | 13 (1.71)           | 22 (2.89)             | 17 (2.24)         | 10 (1.32)              |

**Table 4. Mean intensity of parasites isolated from snails examined**

| Snail species       | Total no. of parasites recovered | Parasite species infected | No. of parasites recovered | Mean Intensity |           |
|---------------------|----------------------------------|---------------------------|----------------------------|----------------|-----------|
|                     |                                  |                           |                            | Mean           | 95%CI     |
| <i>A. achatina</i>  | 250                              | <i>S. stercoralis</i>     | 112                        | 4.308          | 4.02-4.59 |
|                     |                                  | <i>A. cantonensis</i>     | 160                        | 4.000          | 4.56-5.00 |
|                     |                                  | <i>F. gigantica</i>       | 22                         | 2.750          | 2.75-2.75 |
|                     |                                  | <i>D. dendriticum</i>     | 63                         | 3.938          | 3.51-4.37 |
|                     |                                  | <i>S. mansoni</i>         | 34                         | 2.615          | 2.28-2.95 |
|                     |                                  | <i>A. lumbricoides</i>    | 12                         | 3.000          | 2.43-3.57 |
| <i>A. fulica</i>    | 200                              | <i>S. stercoralis</i>     | 130                        | 3.333          | 3.08-3.59 |
|                     |                                  | <i>A. cantonensis</i>     | 155                        | 4.079          | 3.84-4.32 |
|                     |                                  | <i>F. gigantica</i>       | 5                          | 2.500          | 2.06-2.94 |
|                     |                                  | <i>D. dendriticum</i>     | 102                        | 3.778          | 3.48-4.07 |
|                     |                                  | <i>S. mansoni</i>         | 8                          | 2.667          | 2.34-2.99 |
|                     |                                  | <i>A. lumbricoides</i>    | 33                         | 2.750          | 2.38-3.12 |
| <i>A. marginata</i> | 100                              | <i>S. stercoralis</i>     | 28                         | 4.667          | 4.11-5.22 |
|                     |                                  | <i>A. cantonensis</i>     | 39                         | 3.900          | 3.49-4.31 |
|                     |                                  | <i>F. gigantica</i>       | 16                         | 3.200          | 2.83-3.55 |
|                     |                                  | <i>D. dendriticum</i>     | 7                          | 3.500          | 3.13-3.87 |
|                     |                                  | <i>S. mansoni</i>         | 3                          | 3.000          | -----     |
|                     |                                  | <i>A. lumbricoides</i>    | 10                         | 2.500          | 2.19-2.81 |

| Snail species      | Total no. of parasites recovered | Parasite species infected | No. of parasites recovered | Mean Intensity |           |
|--------------------|----------------------------------|---------------------------|----------------------------|----------------|-----------|
|                    |                                  |                           |                            | Mean           | 95%CI     |
| <i>A. belteata</i> | 125                              | <i>S. stercoralis</i>     | 55                         | 3.056          | 2.66-3.45 |
|                    |                                  | <i>A. cantonensis</i>     | 25                         | 2.778          | 2.45-3.14 |
|                    |                                  | <i>F. gigantica</i>       | 12                         | 2.400          | 2.12-2.68 |
|                    |                                  | <i>D. dendriticum</i>     | 18                         | 2.250          | 1.94-2.56 |
|                    |                                  | <i>S. mansoni</i>         | 27                         | 4.500          | 4.14-4.86 |
| <i>A. degneri</i>  | 85                               | <i>A. lumbricoides</i>    | 15                         | 2.143          | 1.82-2.47 |
|                    |                                  | <i>S. stercoralis</i>     | 22                         | 3.225          | 2.60-3.85 |
|                    |                                  | <i>A. cantonensis</i>     | 15                         | 3.750          | 3.09-4.41 |
|                    |                                  | <i>F. gigantica</i>       | 28                         | 2.800          | 2.40-3.20 |
|                    |                                  | <i>D. dendriticum</i>     | 11                         | 2.200          | 1.76-2.64 |
|                    |                                  | <i>S. mansoni</i>         | 8                          | 2.667          | 2.34-2.99 |
|                    |                                  | <i>A. lumbricoides</i>    | 5                          | 2.500          | 2.06-2.94 |

Table 5. Diversity characteristics of parasites community found in different species of *Achatina*

| Diversity Index                          | <i>A. achatina</i>    | <i>A. fulica</i>      | <i>A. marginata</i>   | <i>A. belteata</i>    | <i>A. degneri</i>     |
|------------------------------------------|-----------------------|-----------------------|-----------------------|-----------------------|-----------------------|
| Species diversity (Shannon-Wiener index) | 1.441                 | 1.391                 | 1.523                 | 1.653                 | 1.646                 |
| Species richness (Margalef index)        | 0.823                 | 0.824                 | 1.079                 | 0.995                 | 1.114                 |
| Species evenness (Pielou evenness index) | 0.804                 | 0.776                 | 0.850                 | 0.923                 | 1.919                 |
| Species dominance (Simpson index)        | 0.291                 | 0.280                 | 0.256                 | 0.220                 | 0.215                 |
| Dominant species                         | <i>A. cantonensis</i> | <i>A. cantonensis</i> | <i>A. cantonensis</i> | <i>S. stercoralis</i> | <i>S. stercoralis</i> |

Larvae of *Strongyloides stercoralis* was seen in all snail samples examined. *Angiostrongylus cantonensis* was only recovered in 5.26% of snails examined. The least encountered was the ova of *Ascaris lumbricoides* (1.32%) (Table 3).

Fig. 2 shows the seasonal prevalence of infection among the five *Achatina* species. All snail species were infected with different species of parasites. *Achatina fulica* recorded the highest prevalence of infection during the late dry season (February and March) and onset of the wet season (April, May, and June), while *Achatina achatina* recorded highest prevalence of infection in January and August. *Achatina marginata* recorded the highest prevalence in July. Overall prevalence of infection among snails was generally higher in the wet season compared with the dry season.

The highest number of parasitic helminths was recovered from *A. achatina*. The mean intensity of *S. stercoralis* in *A. marginata* was highest, 4.667 (4.11-5.22 (95% CI)) followed by *S. mansoni* intensity in *A. belteata* at 4.500 (4.14-4.86 (95% CI)). *A. belteata* recorded the least intensity for *A. lumbricoides* at an intensity of 2.143 (1.82-2.47 (95% CI)). The intensity of *A. cantonensis* was highest in *A. fulica* at an intensity of 4.079 (3.84-4.32 (95% CI)) (Table 4).

In the land snails the diversity of parasite species assessed using diversity indices accorded *A. balteata* the highest, 1.653 (Shannon-Wiener index). For dominant species *A. achatina*, *A. fulica* and *A. marginata* recorded similar results (Table 5). *A. degneri* recorded the highest value for species richness (Margalef's index = 1.114). *A. degneri* also had the second highest parasite diversity ( $H=1.646$ ) but recorded the highest value for parasite richness (1.919). The snail species with the most restricted parasite species infection was *A. fulica* ( $H=1.391$ , Margalef's index=0.776 and species richness=0.824), although *A. marginata* recorded a slightly lower value for species richness ( $H=0.823$ ) (Table 5).

#### 4. DISCUSSION

A total of 760 *Achatina* snail species were collected in this study. The species of *Achatina* recovered in this study (*A. achatina*, *A. fulica*, *A. marginata*, *A. belteata* and *A. degneri*) have been reported in other studies conducted in Nigeria [2,5,15]. *A. achatina* was the most abundant species while *A. degneri* was the least abundant. This finding agrees with reports by Elom and Opara-Elom [5]. However, other studies have reported *A. marginata* to be the least abundant [5,16,17] compared to *A. degneri* reported in this study.

More snails were collected in the wet season than in the dry season which is in line with the normal biology of snails. The highest number of snails collected in May is in line with the report of Hodasi [18], who observed that *Achatina* breeds mainly in the wet season from April to July. Onyishi et al. [2] reported that weather conditions in the wet season in Nigeria is favourable for terrestrial snail populations to blossom. Snails hibernate under prolonged dry conditions and aestivate under prolonged hot conditions. The period of the year with extreme weather conditions are spent in dormancy, a behavioural survival strategy that sustains the different species. During hibernation and aestivation, only a few of the snails are seen foraging on the ground surfaces. Hence, the fewer number collected in the dry season and more in the wet season. It has been reported that Nigerian snails aestivate in the dry weather and when the phenomenon of aestivation is in progress, the aperture is usually temporarily closed by epiphragm and when rains begin to fall, the epiphragm opens and releases the snail to forage [19,20].

All snail species were infected with different species of parasites and the overall prevalence of infection among snails was generally higher in the wet season compared with the dry season. Elom and Okpara-Elom [5] also reported the prevalence of parasites in snails to be high during the wet season. For example, the larvae of *Angiostrongylus* species under normal conditions should survive better in the rainy season and should be able to penetrate the snail intermediate hosts better in the rainy season than in the dry season. In the dry season, the phenomena of aestivation and hibernation reduce snail exposure to the parasites because only very few snails forage during that period.

In the wet season, the soil-transmitted helminths (*A. lumbricoides* and *S. stercoralis*) infected almost all the snail species with higher prevalence than in the dry season. Most of the farmlands and bushes that the snails inhabited were contaminated with human faeces as a result of inadequate toilet facilities that encourage open defecation by the people in the study area. As the wild snails forage, they become more exposed to the soil-transmitted helminths ova deposited in the soil. These have been reported to survive and develop in climatic conditions of optimum temperature and humidity as observed in the wet season months [21].

An overall prevalence of 41.97% was recorded for parasitic infection in snails in this study. This is lesser than results reported by Onyishi et al. [2] in Ugwueme agricultural zone in Enugu State, Nigeria. This is of public health significance especially to individuals

who consume these snails for its protein content. The meat quality of the infected snails is also affected in terms of quality, depriving the consumers of valuable nutrients.

The highest number of parasitic helminths was recovered from *A. fulica* and this observation is in agreement with previous reports that some snail species are more susceptible to certain parasites than others [21]. Elom and Okpara-Elom [5] also reported *A. fulica* to be more infected by parasites than other snail species examined.

*A. lumbricoides* infected all the snail intermediate hosts in both seasons, with less prevalence in the dry season. The versatility in infectivity of *A. lumbricoides* with respect to seasons and hosts as observed in this study has been reported and attributed to the resilience of its ova which has been observed to withstand extreme environmental conditions [22].

In this study, the intensity of *A. cantonensis* was highest in *A. fulica* at an intensity of 4.079 and it has been reported that *A. fulica* is the major snail host of *Angiostrongylus* species all over the world [23] and that observation has been attributed to its ubiquity and high level of susceptibility to the parasite [24].

## 5. CONCLUSION

The results of this study indicate the prevalence of different parasitic species of public health importance. Infection of the snails which are endemic in the study area with the recovered parasites, majority of which are zoonotic could be attributed to poor environmental sanitation and open defecation. Therefore, health education, provision of adequate toilet systems and creation of public health awareness are required for sustainable control and interfering with the transmission of the parasites.

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## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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