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# Endo-Helminth Fauna of the Rainbow Lizard (Agama Agama)

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**Abstract**

*This parasitological study was carried out between February and May 2019 to determine the prevalence and intensity of helminthiasis in the rainbow lizard (Agama agama) in Okrika, Rivers State, Nigeria. A total of one hundred and fifty-one (151) specimens made up of 93 males and 75 females were caught by a local netting system during the day and anaesthesized with chloroform. Samples were collected from two stations (Ogoloma-Ama and Oba-Ama). After dissection, the esophagus, stomach, small intestine, rectum, liver, lungs, urinary bladder, heart and body cavity were searched for helminths using conventional methods. Five species of helminths belonging to Nematoda - Strongyluris brevicaudata, Parapharyngodon awokoyai, encysted Ascaridida larva, Cestoda - Oochoristica sp. and Trematoda - Mesocoelium spp - were recovered from infected lizards. Helminths infected one hundred and twenty-three (123) (82%) of the lizards. In Oba-Ama, forty-five (45) (76.3%) out of 59 and in Ogoloma-Ama, seventy-eight (78) (84.8%) out of 92 were infected with helminths. By abundance, in both locations, the males were more infected than their female counterparts with a prevalence of 51(93%) and 72(75%) (P<0.05), respectively. This study has revealed the helminth parasites infecting the agamid lizard of Rivers State, Nigeria. It has also shown some unidentified species of Mesocoelium and Oochoristica sp. Additionally the trapping system used was also found to be effective and efficient.*

## Introduction

Lizards are seen in most warm places around the world as a result of their nature as poikilotherms (Omonona et al., 2011). They are about the most numerous extant species of reptiles, with over 4300 members (Benneth, 1998). They vary in their sizes, morphologies and colors. They exhibit various means of movement and defense. In Africa, commonly found lizards are the geckos, chameleons, monitor lizards, skinks, alligators, crocodiles and the Agama lizards (Benneth, 1998). The Agama is a typical member of the Agamidae family seen abundantly in Africa, Asia, Australia, and sighted in Southeastern Europe by Benneth (1998).

The agamid lizard is diurnal in nature residing in crevices at night close to habitations (Harris, 1964) though it has been reported just recently to have become active at night feeding off on insects and resting on warm walls having electric panels reasons for which



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could be increased predation due to rapid expansion of the urban environment and thermoregulation at night (Amadi et al., 2020).

Helminths are mostly parasites of vertebrates but could also be found in invertebrates, specifically arthropods and mollusks, which serve as intermediate hosts (Smyth, 1994). Parasites are very important causes of high death rates in wild animal populations (Anderson and Gordon, 2009).

In Nigeria, the investigations of Babero & Okpala (1962) and Wekhe & Olayinka (1999) revealed the parasite Lecudina sp., a gregarinid protozoan which is pathogenic and poses human health risks and uses the Rainbow lizard as its intermediate host. According to Ameh & Ajayi (1996), man could also be exposed to Lecudina infection by termite consumption due to the presence of this protozoan in the gut of termites. Another helminth parasite, Raillietiella sp. (a pentastomid) is also frequently isolated from lizards (Nash, 2005).

The aim of this study is to identify the helminth endoparasites of Agama agama in Ogoloma-Ama and Oba-Ama of Okrika, Rivers State, and to determine the intensity and prevalence of the helminth endoparasites of A. agama in both locations while also investigating any relationship between the snout-vent length (SVL) and body weight (WT) of the lizards, and parasite burden.

## Methods

### *Study Area*

Okrika is an island between Latitudes 4° 44' 31.74" N and Longitude 7° 05' 1.25"E while Ogoloma Ama and Oba Ama are settlements (Fig 1) on the island between latitudes 4° 73' 38.76" N, 4° 77' 35.71"N and Longitude 7° 08' 11.03"E and 7° 07' 82.41."E respectively, in Rivers State, Eastern Niger Delta, Nigeria, it is a tropical region with the river system been an estuary with tidal movements. the rainy season is from April to September with a break in August though it lasted longer till about December in 2018-2019. Okrika has temperatures ranging from 26oC to 34°C.

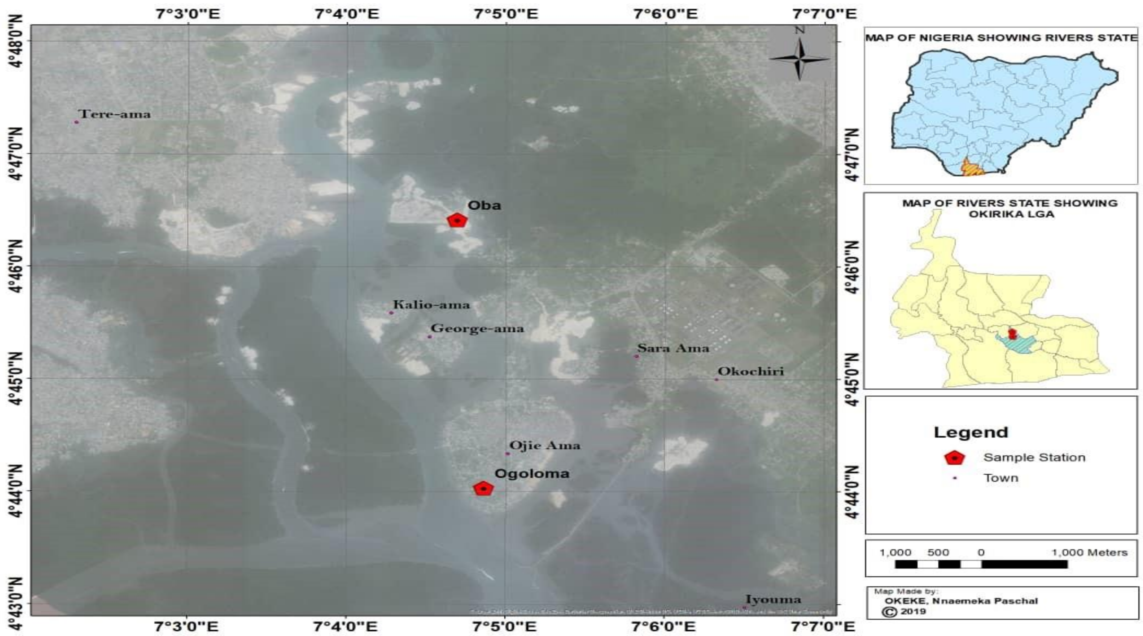


*Figure 1. Map of study area showing sample stations*



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*Sample collection*

One hundred and fifty-one (151) *Agama agama* were randomly captured by trapping during the day near residential buildings (between 7am and 4pm) from February to May, 2019. The trap used was made up of four wooden pecks of 24 cm x 2 cm set in alternate positions in the ground forming a square around residential buildings where lizards were sighted. The pegs were covered with a nylon net of mesh size 2 cm / 20 mm as shown in Fig 2 and Plate 1. A 100 g weight was used to keep the edges in place. A local bait made from grinded and dried cassava (*Manihot esculenta*) and palm oil was placed in the middle of the trap. The bait attracted insects which attracted the lizards. Lizards were trapped when they entered the net to feed on the insects. Specimens were removed from the trap and transported to the Entomology and Parasitology Laboratory, Rivers State University, Port Harcourt, in buckets covered with a net to allow ventilation. Specimens were anaesthesized with chloroform and dissected.

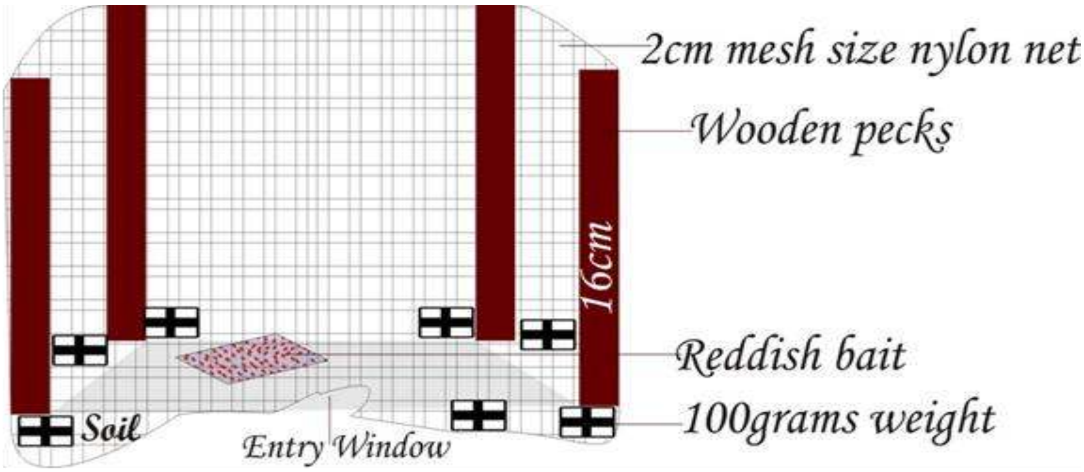


*Figure 2. Outlook of the local netting for lizard sampling*

*Figure 3. A trap showing a successful catch at Oba Ama, Okrika LGA, Rivers State, Nigeria*



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***Dissection of Specimens***

The lizards were anaesthesized by exposure to chloroform vapor in an airtight container. Each individual lizard was weighed with Adams electronic weighing balance (model AQP 1600). The Snout vent length (SVL) was taken by the aid of a measuring board and a transparent meter rule. The ventral surface was longitudinally cut open by making an incision from the vent to the throat of each lizard. The gastrointestinal tract (GIT) was recovered by cutting the anterior of the oesophagus and the posterior end of the rectum. The body cavity and internal organs (liver, lungs, urinary bladder and heart) were properly examined separately in Petri dishes containing 0.72% saline solution for the presence of motile and adhering helminth parasites with the aid of a dissecting microscope. The skin was carefully removed exposing the subcutaneous layers which were examined for the presence of microfilaria. Recovered parasites were washed in the 0.72% saline solution to remove debris and mucus and ensure proper preservation.

***Fixing and Staining of Helminth Parasites***

Nematodes recovered were stretched in hot 70% alcohol and later fixed in fresh cold 70% alcohol. Few drops of glycerol were added to reduce total dehydration of the nematode. Nematodes were cleared in lactophenol before microscopy. Cestodes and trematodes were flattened in 5% formol saline by placing them between two microscope slides for about 15 minutes. Thereafter, they were washed in several changes of distilled water to remove the excess fixative used, stained overnight in acetocarmine and dehydrated within 24 h in series of graded alcohol (30%, 50%, 70% and 100%) at intervals of 1 h. They were then cleared in a solution of 50/50% alcohol and xylene and in absolute xylene, mounted in Canada balsam for permanent slides, and examined under a Compound microscope using a x10 objective.

Helminths were identified according to protocols of Yamaguti (1958) and Schmidt (1986). The number of helminth parasites recovered was recorded per location in the gut. Photomicrographs of representative helminth species were made using a Nikon Digital Camera attached to the eye piece of the light microscope.

***Statistical Analyses***

Student t-test was used to check for significant difference in the prevalence of parasite infection between sexes and locations. Chi-square was used to test for associations between parasite prevalence by sexes and locations. Analysis of variance was used to test for significant difference between parts of the GIT and Tukey-Kramer test was used for mean separation. Pearson correlations were used to test for relationships between selected variables. These Analyses were done using JMP-SAS package.

## Results and Discussion

***Abundance and Total Prevalence of Helminth Parasites in both Locations***

One hundred and fifty-one (151) lizards were examined in the course of the research; fifty-nine (59) were from Oba-Ama and ninety-two (92) from Ogoloma-Ama. The hosts from Oba-Ama were comprised of twenty-one (21) males and thirty-eight (38) females, while those from Ogoloma-Ama were thirty-four (34) males and fifty-eight (58) females. Overall, one hundred and twenty-three (123) lizards were infected accounting for a total prevalence of 82%. By location, out of 59 hosts, 45 were infected at Oba-Ama with a prevalence of 76.3%, while 78 out of the 92 hosts from Ogoloma-Ama were also infected accounting for a prevalence of 84.8%. Total parasites recovered from these hosts numbered 2,446: 788 from



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Oba-Ama and 1, 658 from Ogoloma-Ama. There was, statistically, no difference between the parasite load of both locations ($t_1$=2.7682, p=0.0983).

The parasites recovered included cestodes (*Oochoristica sp*.), trematodes (*Mesocoelium spp*) and nematodes (encysted *Ascaridida* larva, *Parapharyngodon awokoyai* and *Strongyluris brevicaudata*) (Plates 2-5). The prevalence and mean intensity of infection by these parasites are presented in Table 1.

Table 1. Prevalence (P%) and mean intensity (MI) of helminth parasite infection in Agama agama, Okrika, Rivers State, Nigeria

| **Parasite** | **Location** | | | |
|---|---|---|---|---|
| | **Oba-Ama** | | **Ogoloma-Ama** | |
| | P% | MI±SD | P% | MI±SD |
| **Cestoda** | | | | |
| *Oochoristica sp* | 5 | 2.64±1.8 | 4 | 1.92±1.3 |
| **Trematoda** | | | | |
| *Mesocoelium sp.* | 14 | 27.75±13.1 | 6 | 15.00±9.2 |
| **Nematoda** | | | | |
| Encysted ascaridida larva | 1 | 1.00±0.0 | 2 | 1.88±0.9 |
| *Parapharyngodon awokoyai* | 31 | 7.09±4.6 | 36 | 8.88±6.9 |
| *Strongyluris brevicaudata* | 50 | 8.59±7.7 | 51 | 11.28±9.3 |

***Gender influence on Prevalence of Parasites***

Nematodes were more prevalent than both cestodes and trematodes and higher values were obtained in male hosts than in their female counterparts (Table 2). For instance, in hosts from Ogoloma Ama, nematodes infected 94% of the male hosts and 79% of the females. Similarly, higher prevalence of infection was recorded in hosts from Ogoloma-Ama. Similarly, nematodes infected 91% of the male hosts and 71% of the females in Oba-Ama. However, student t-tests showed the differences were not statistically significant ($t_1$= 2.768, p=0.098). Generally, in Okrika, the male hosts were more infected. Student-t tests showed that there was a statistical difference between the male and female hosts ($t_1$= 26.25, p=0.0001).

Table 2. Total prevalence (P%) and mean intensity (MI) of classes of helminth parasites in male and female lizards (*A. agama*) of Ogoloma-Ama and Oba-Ama, Okrika LGA, Rivers State, Nigeria

| **Parasite** | **Ogoloma-Ama** | | **Oba-Ama** | |
|---|---|---|---|---|
| | **Male** | **Female** | **Male** | **Female** |



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| | P% | MI±SD | P% | MI±SD | P% | MI±SD | P% | MI±SD |
|---|---|---|---|---|---|---|---|---|
| Nematode | 94 | 24.6±18.6 | 79 | 12±11 | 91 | 20.8±16.7 | 71 | 8.8±12.5 |
| Cestode | 62 | 2.2±1.5 | 28 | 1.5±0.6 | 48 | 1.4±1.9 | 11 | 0.2±0.7 |
| Trematode | 12 | 15.5±10.1 | 5 | 14.3±7.6 | 19 | 1.9±3.9 | 0 | 0.0±0.0 |

***Site Preference of Parasitic Helminth Species of Agama agama***

The rectum and the small intestine had the highest abundance and infection rates, respectively (Tables 3- 4). In hosts from Oba-Ama, a total of 395 individulas of *Strongyluris brevicaudata* were recovered from the specimens- 359 (91%) and 36 (9%) from the rectum and small intestine, respectively. 241(100%) *Parapharyngodon awokoyai* were only found in the rectum. 71(64%) and 40(36%) of *Mesoceolium spp* were recovered from the rectum and small intestine, respectively. 37(100%) *Oochoristica sp.* was recovered from the small intestine and 4(100%) *Ascaridida* larva were encysted on the body cavity. In Ogoloma-Ama, the rectum and the small intestines were the most infected sites too (Table 4). 808 (96%) *Strongyluris brevicaudata* were recovered from the rectum, 28(3%) from the small intestine and 10(1%) from the body cavity. 604 (100%) *Parapharyngodon awokoyai* were only recovered from the rectum. (100%) *Mesocoelium spp* were recovered from the rectum, 71(100%) *Oochoristica sp.* from the small intestine and 32(100%) Ascaridida larva were seen encysted in the body cavity.

Table 3. Site preference of helminth endoparasites in *A. agama* from Oba-Ama, Okrika, Rivers State, Nigeria

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| Parasite | Parasite Abundance | Distribution | | |
|---|---|---|---|---|
| | | (BC) | (RC) | (SI) |
| *S. brevicaudata* | 395 | 0 (0%) | 359 (91%) | 36 (9%) |
| *P. awokoyai* | 241 | 0 (0%) | 241 (100%) | 0 (0%) |
| *Mesoceolium spp* | 111 | 0 (0%) | 71 (64%) | 40 (36%) |
| *Oochoristica sp.* | 37 | 0 (0%) | 0 (0%) | 37 (100%) |
| *Encysted Ascaridida larva* | 4 | 4 (100%) | 0 (0%) | 0 (0%) |

Key: RC= Rectum; SI= Small Intestine; BC= Body Cavity

Table 4. Site preference of helminth endoparasites in *A. agama* from Ogoloma-Ama, Okrika, Rivers State, Nigeria

| Parasite Ogoloma | Abundance | Distribution |
|---|---|---|



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| | | BC | RC | SI |
|---|---|---|---|---|
| *S. brevicaudata* | 846 | 10 (1%) | 808 (96%) | 28 (3%) |
| *P. awokoyai* | 604 | 0 (0%) | 604 (100%) | 0 (0%) |
| *Mesocoelium spp.* | 105 | 0 (0%) | 105 (100%) | 0 (0%) |
| *Oochoristica sp* | 71 | 0 (0%) | 0 (0%) | 71 (100%) |
| *Encysted Ascaridida* larva | 32 | 100% | 0 (0%) | 0 (0%) |

Key: RC= Rectum; SI= Small Intestine; BC= Body Cavity

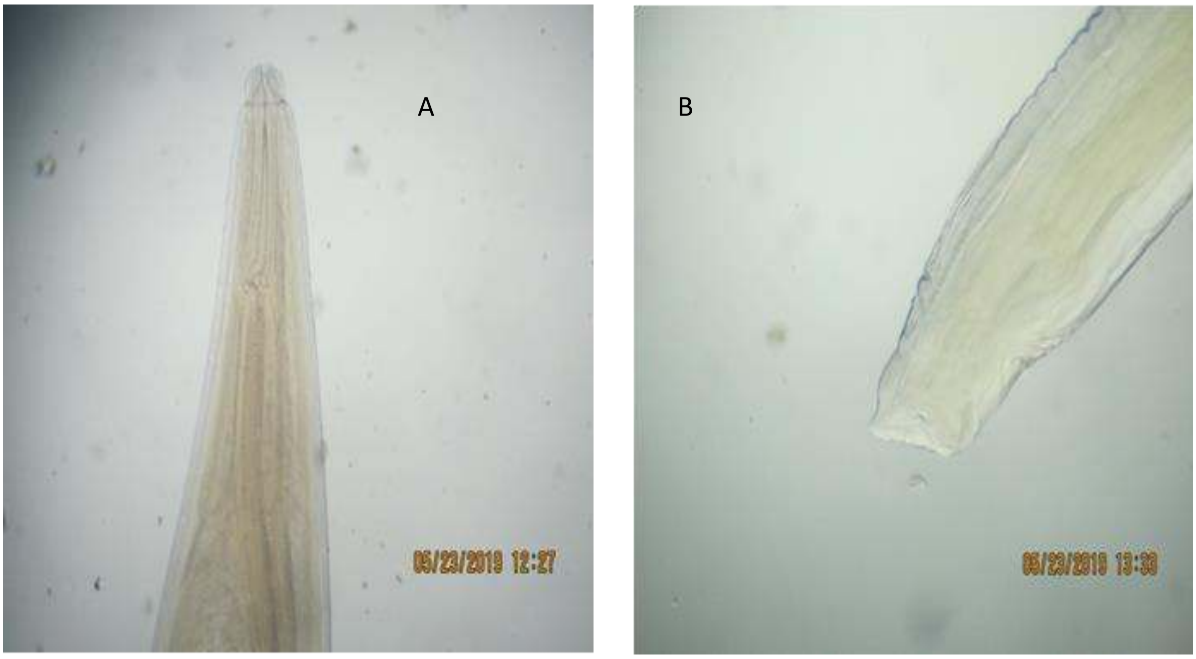


*Figure 4. Photomicrographs of Parapharyngodon awokoyai recovered from Agama agama in Okrika, Rivers State, Nigeria. A(anterior) and B(posterior). Scale: 0.5mm*



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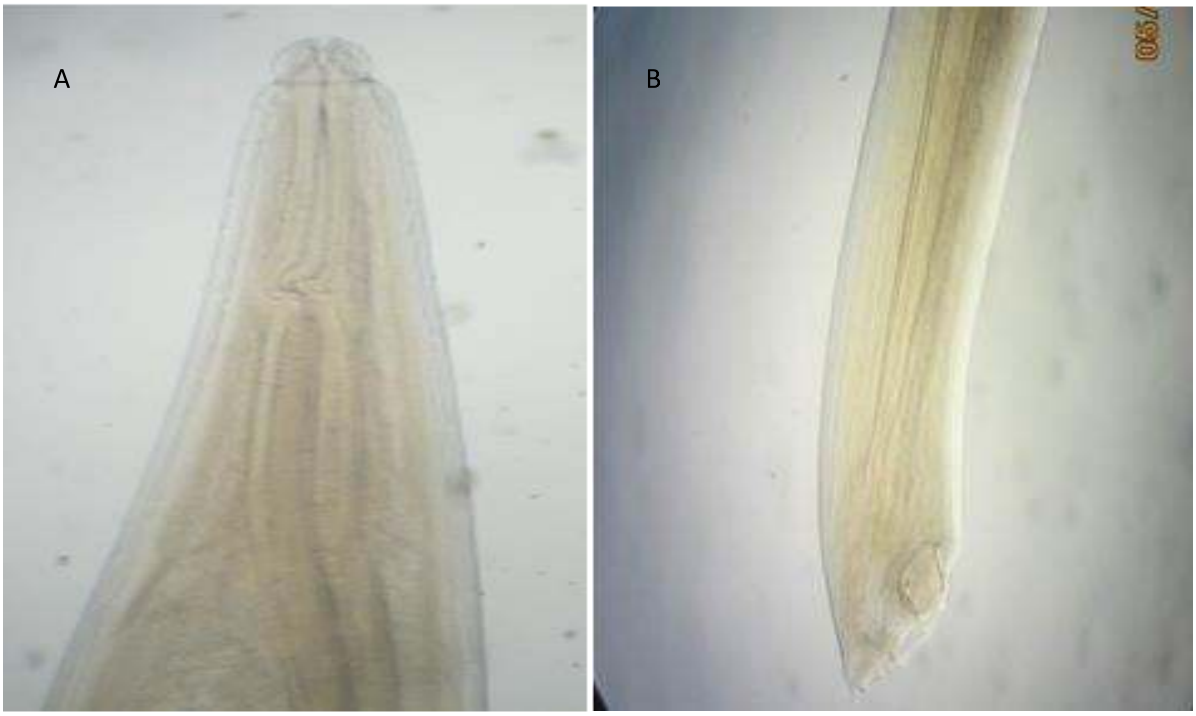


*Figure 5. Photomicrographs of Strongyluris brevicaudata recovered from Agama agama in Okrika, Rivers State, Nigeria. A(anterior) and B(posterior). Scale: 0.5mm*

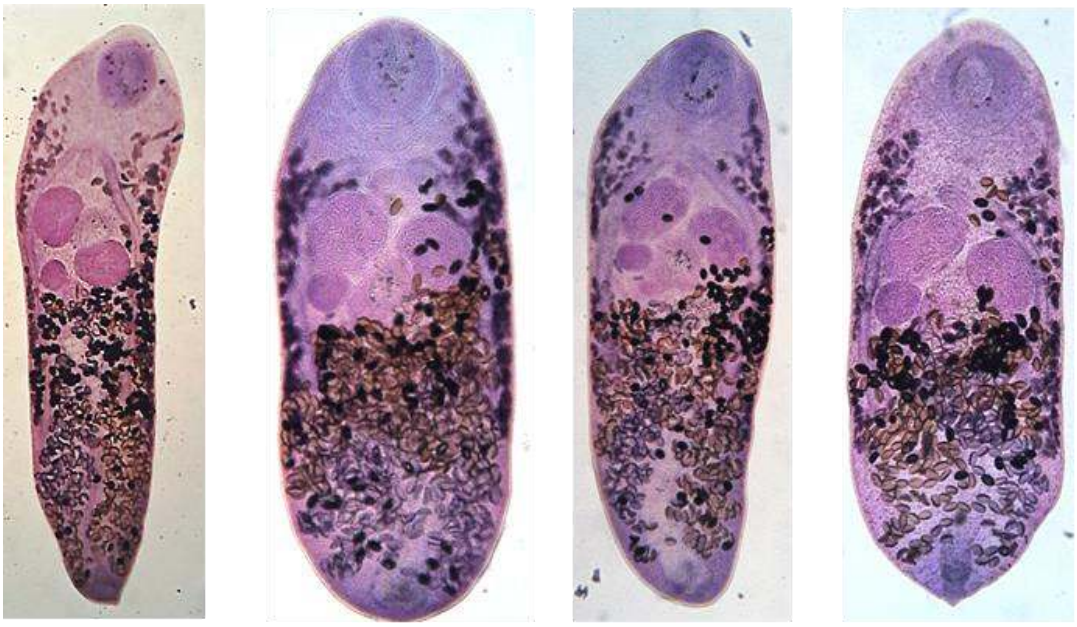

*Figure 6. Photomicrographs of Mesoceolium spp recovered from Agama agama in Okrika, Rivers State, Nigeria*



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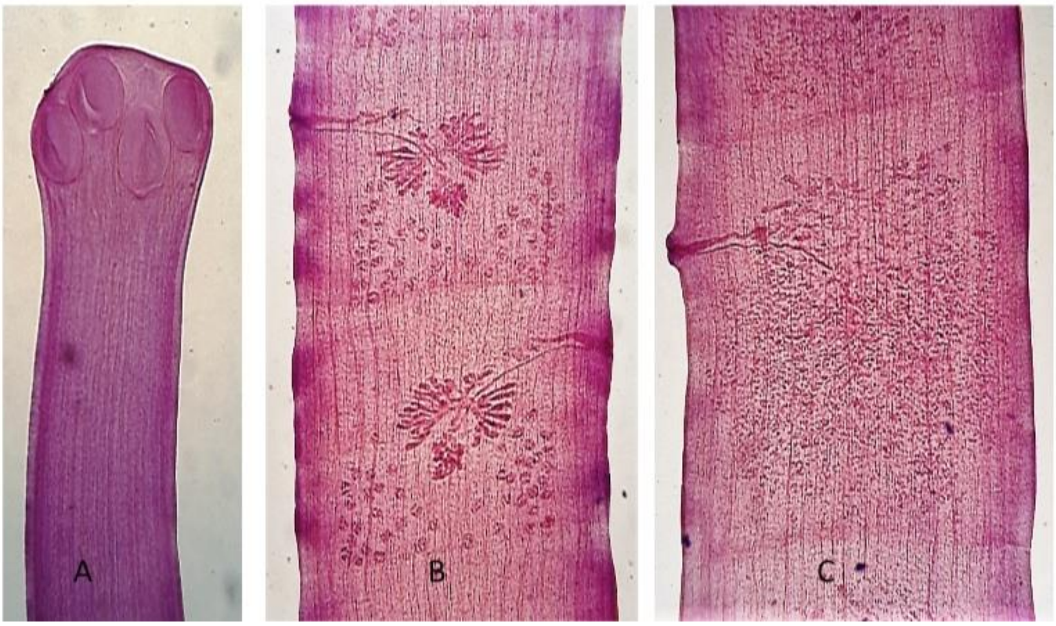


*Figure 7. Photomicrographs of Oochoristica sp. recovered from Agama agama of Okrika, Rivers State, Nigeria*

Five helminth species were isolated from the rainbow lizard (*Agama agama*) in this study and they included *Strongyluris brevicaudata, Parapharyngodon awokoyai,* encysted Ascaridida larva, *Mesocoelium spp and Oochoristica sp.* Some of these helminths observed have been reported previously from similar studies in Nigeria. Babero & Okpala (1962) and Adeoye & Ogunbanwo (2007) reported *Strongyluris brevicaudata, Parapharyngodon awokoyai, Capillaria sp, Oxyuris sp, Oochoristica agamae, Mesocoelium monas* and *Railiettiella sp.* from Lagos and Osun States, respectively. In their own research, Omonona *et al.* (2011) recovered *Strongyluris brevicaudata* and *Thelandros annulatus* from same host in Lagos. These reports show that these helminths have a wide geographical distribution. The environmental factors of all the locations could be said to be alike therefore equally supporting the existence of these parasites.

The overall prevalence (82%) recorded in this investigation is same with the 82% recorded by Zazoo (2013) from Ogoni Kingdom in Rivers State also but is lower than 100% reported by Sowemimo & Oluwafemi (2017) in Osun State from same animal hosts. This observation could be attributed to existing environmental and ecological factors between these locations.

This investigation has shown that of the two communities (Oba-Ama and Ogoloma-Ama) in Okrika, Ogoloma-Ama had the highest prevalenve and mean infection rate with an average of 18.02±17.2 parasites per lizard and a prevalence of 84.8% while Oba Ama had a mean infection of 13.36±16.2 per lizard and a prevalence of 76.3%. This could be due to the settlement types; Ogoloma-Ama is a more urbanized area with high anthropogenic activities in addition with the release of so much petroleum pollutants. Pollutants pose some sub lethal physiological stress to hosts thereby reducing their capacity to withstand parasite invasion which results into increasing infection levels (Koprivnikar *et al*., 2007) as against Oba-Ama which still maintains its pure vegetation with less human activities around.

Nematodes had a prevalence of 67.1%, cestodes 26.7% and trematodes 6.2%. This shows that the most prevalent helminth taxa recovered from the lizard *A. agama* examined were the



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nematodes, especially *S. brevicaudata*. Similar findings have previously been reported in Nigeria and other parts of the world. Four of the seven helminths recovered from *A. agama* in Lagos State, Nigeria (Adeoye & Ogunbanwo, 2007) and 3 of the 4 helminthes parasites reported from Nsugbe, Anambra State, Nigeria (Nwadike & Ilozumba, 2010) were nematodes with *S. brevicaudata* the most abundant. The rich diversity of nematodes in the agamid lizard and other previous reports could be due to the fact that they exhibit a direct life cycle (Al-barwari & Saeed, 2007). On the other hand, it also shows that *S. Brevicaudata* is a major parasite of the Agamid lizards.

Trematodes in this study had a low prevalence of 6.2% which is higher than the prevalence of trematodes from other studies in Nigeria such as Adeoye & Ogunbanwo (2007) who recorded a prevalence of 1.6%, while Sowemimo and Oluwafemi (2017) had 1.5%. Generally, the low prevalence of trematodes in this and other studies in Nigeria and other countries (Glalaktionov & Dobrovolsky, 1998) could be as a result of the inability of the trematode larval stages to locate their intermediate hosts, and could also be due to the diet preference of the agamid lizard which may not include some trematode intermediate hosts. The few reported trematodes in agamids are basically members of the Genus *Mesocoelium* (*M. monas* and *M. monodi*). This study recovered different unidentified species of *Mesocoelium* which could not be seen as the conventional species due to morphological differences such shape, positions of the testes and lenght of the body.

*Oochoristica sp*. was the only cestode encountered in this study. This cestode differs morphologically from the previously reported *Oochoristica* species (Zazoo, 2013; Adeoye & Ogunbanwo, 2007; Sowemimo & Oluwafemi, 2017; Nwandike & Ilozumba, 2017; Goldberg & Bursey, 2001) . This species was previously encountered by Aisien & Igetei (2018) in their study on some anurans from southern Nigeria and shows same features such as sizes of mature proglottids, shape and arrangements of the testes and presence of a neck which is in direct contrast with the previously reported *Oochoristica* species. This observation does not only shows increase in the *Oochoristica* species and wide geographical ranges but also indicate its presence as not accidental (Aisien & Igetei, 2018). Moreso,the presence of members of the genus *Oochoristica* in anurans and agamids suggest there could be a relationship in diet selections of both host groups.

In this study, males were more associated with parasitic helminth infections than the females. This is in agreement with the reports of Adeoye & Ogunbanwo (2007). Sulieman *et al*. (2019) also recoreded a higher prevalence of helminths in the male white-spotted geckos in Sudan. This trend could be because the males are more active and cover more grounds during foraging which exposes them to more parasites. In contrast, Omonona *et al*. (2011) reported female agamids to be more infected. Nwadike & Ilozumba (2010) and Sowemimo & Oluwafemi (2017) recorded no significant differences in prevalence of infections between gender. Amo *et al.* (2005) observed that females and males showed similar susceptibility to parasite infections. These authors attributed this to the similar diet composition of both sexes exposing them to equal chances of infection. Some other studies on lizards and other organisms have shown male hosts to be more prone to parasite infections (Uller & Olsson, 2003) which they linked to the immune suppressive effects of the male hormone (testosterone) during reproductive periods (Roberts *et al.,* 2004). Secondly, in our own view, another reason for sex dependent parasitism of helminths could be due to size. Morphologically, a large body mass provides the parasite an abundance of desired nutrients, resources needed for colonization and survival i.e space to feed and reproduce (Aho, 1990; Van sluys *et al.*, 1994; Poulin, 1997). Robert *et al*., (2020) in a correlation study on the



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agamid lizards in Rivers State , Nigeria reported a significant positive correlation between parasite prevalence and some morphometric measurements.This could be the case of the rainbow lizard (*A. Agama*) as the adult males are larger than the females (Harris, 1963).

The rectum harboured two nematodes (*Strongyluris brevicaudata* and *Parapharyngodon awokoyai*) and a trematode (*Mesoceolium spp*). This showed the rectum to be the most preffered microhabitat for parasite infections from both locations. This agrees with those of Adeoye & Ogunbanwo (2007), Nwadike & Ilozumba (2010) and Sowemimo & Oluwafemi (2017) where the rectum was also reported to be the predilection site for *Strongyluris brevicaudata* and *Parapharyngodon awokoyai.* Helminths seek out places in their hosts that could provide máximum nutritional and survivability. The rectum in the *A. agama* has a larger lumen than most parts of the gut, it contains enormous amount of undigested food which could be beneficial to these nematodes who have functional digestive systems. Also the rectum devoid of the high peristaltic movement seems safer for nematodes who are slow in movement and lack serious adhering structures. The small intestine hosted a single cestode (*Oochoristica sp.*) and some *Strongyluris brevicaudata.* Cestodes generally lack digestive systems and will always inhabit the small intestine of vertebrates which is a region rich with soluble nutrients and good for absorption (Jennings, 1997). Encysted Ascaridean larva was only recovered from the body cavity of the agamid lizard which serves as its paratenic host. The presence of this larval stage shows that there is a feeding relationship between members of the herpetofauna. Some anurans also serve as paratenic hosts to this larvae whose definitive hosts are snakes (Imasuen *et al*., 2012).

## Conclusion

In all, the present study has established the endohelminth fauna of A. agama in Ogoloma-Ama and Oba-Ama of Okrika, Rivers State, Nigeria. The helminths included Strongyluris brevicaudata, Parapharyngodon awokoyai, encysted Ascaridida larva (Nematoda); Oochoristica sp (Cestoda) and Mesoceolium spp. (Trematoda). More interesting is the incidence of different species of Mesocoelium and a single species Oochoristica which was quite different from the ones previously reported in A. agama by other authors in Nigeria and other parts of the world.

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