

Vectors of *Paragonimus Uterobilateralis* a Causative Fluke for Paragonimiasis in Cross River State-Nigeria (Pp. 414-423)

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Abstract

Investigation into suggested vectors of Paragonimus uterobilateralis a causative agent of Paragonimiasis was carried out. The investigation was informed by the need to ascertain vectors of the lung fluke-paragonimus to enhance health education of the inhabitant of the affected village for control purpose. Dissected Pseudanautes Africanus, Astacus sp, Potedema freethii and Onchomelina sp were examined for intermediate stages of Paragonimus. Samples of crustaceans and gastropods examined for larvae of Paragonimus showed Pseudanautes Africanus, Astacus sp, Potedema freethii and Onchomelina sp as carriers of larvae of Paragonimus. Ninety three (93%) of P. africanus were incriminated with 30.5% of all isolated eggs, 33.9% of cercaria and 39.7% of rediae. Ninety six (96%) of Astacus were incriminated with 39.4% of all eggs, 66.1% cercaria and 60.3% of Rediae. Sixty (60) percent of P. freethii were incriminated with 11% of all eggs and 67% of miracidium and sporocysts while eighty eight (88%) of Onchomelina sp were incriminated with 19.2% of all eggs and 33% of miracidium. The hosts were wide spread along Cross River tributaries suggesting the possibility of infection along the course of the river. The examined crustaceans and

gastropods are vectors of P. uterobilateralis in Cross River state of Nigeria. Since metacercaria was isolated only from P. africanus, infections was suggested to be mainly through consumption of this crab host.

Introduction

Following earlier report by Ikpeme (1991) and recent positive report of the presence or occurrence of *Paragonimasis* in Cross River State, Nigeria, investigation with suspected vectors was encouraged.

Paragonimasis is a lung fluke infection (Hunter, 1976) which is caused by *Paragonimus*. Various species of *Paragonimus* have been reported in many parts of the world. Volker and Vogel (1965) reported *P. africanus* in West Africa, *P. Westermani inchuensis* in China, *P. phillippinensis* in Philippines and so on. The species reported in Nigeria is *P. africanus* and *P. uterobilateralis* by Udensi (1987).

One common feature about *Paragonimus* is that infection is only possible where intermediates hosts, fresh water crab (*Pseudanautes africanus*) or crayfish (*Astacus*) are eaten raw or in poorly cooked meals. Udensi (1987) reported infection in Igun river basin of Imo State-Nigeria to be through ingestion of raw crabs by children who erroneously believed that raw crabs have more nutritional value.

Until this report vectors of *Paragonimus* in Cross River State was not investigated. Though Nwokolo (1964, 1972) suggested *Pseudanautes africanus* as carriers of *Paragonimus* larvae, the case of Cross River had to be determined.

Materials and Methods

One hundred (100) each of *Pseudanautes Africanus* (Crab), *Astacus sp* (Crayfish), *Potadema freethii* (Periwinkle) and *Onchomelina sp* (water snail) were fished from streams in the infected village and neighbouring two villages.

1. Dissection of Periwinkle and Water Snail

Snails and periwinkle were carefully cracked with stones. Forceps were inserted into the cracked lines and gently lifted to separate the carapace from the visceral hump. Thin sections of muscles of these gastropods were cut and examined on slide under compound microscope.

Intestinal contents were separated and small quantity per time was smeared directly on slides with 2 drops of saturated saline. A cover slip was then gently lowered on the smeared specimen and viewed on compound microscope.

2. Dissection of Crayfish and Crab hosts

These carapaces of crayfish and crabs placed on a dissecting board were carefully and forcefully opened with the help of forceps and spatula. Thin sections of muscles were cut and prepared for examination on microscope as was done with snails and periwinkles. Intestinal contents were also prepared for examination as was done with snail and periwinkle.

3. Saturated saline floatation technique

The gills of crabs and crayfish were rinsed with saturated saline and solution of it taken in test tubes. Also intestinal contents of each of the hosts were dissolve and a solution of it taken in separate test tube. The test tubes were placed in a centrifuge and centrifuged at 1500 r.p.m. for 5 minutes. This helps to concentrate any ova or oocysts presents in solutions. The supernatant was decanted off and precipitate extracted. The precipitate was then made into another solution by adding sodium hydroxide (NaOH) solution as solvent. These were again taken in test tubes and cover slips placed over the tubes. The tubes were then left to stand for 45 minutes. The cover slips were quickly transferred on to clean micro slides held over the open ends of the tubes and viewed under compound microscope. Any ova or oocyst observed were identified using identification keys according to Soulby (1968) and Chandler and Reed (1961).

Results

The following results were observed

A. *Pseudanautes africanus*

Ninety three (93) percent of crabs were incriminated. Examination of muscles of crabs showed neither eggs nor any intermediate stages of *Paragonimus*. However, many eggs were isolated from the gills and mature second generation rediae containing metacercariae were isolated from intestinal contents. Mean (\bar{x}) number of eggs per smear of specimen from gills and intestinal contents were 2 and 5 respectively. Altogether 30.5% of total eggs, 33.9% cercaria and 39.7% of redial were recovered from *Pseudanautes africanus*.

B. *Astacus sp*

Ninety six (96%) of crayfish were incriminated. This represented the highest value of all percentage of respective host sampled. Eggs of *Paragonimus uterobilateralis* were recovered from swimmerets and intestine. Cercariae and second generation rediae were recovered only from intestine. Mean (\bar{x}) number of eggs from the swimmerets and intestine were 1 and 4 per smear respectively. Altogether 39.4% of total eggs, 66.1% of cercaria and 60.3% of rediae were isolated from *Astacus*.

C. *Potadema freethii* and *Onchomelina sp*

Sixty (60%) of *P. freethii* were incriminated. Eggs and miracidium were isolated from intestinal contents only. At least each smear prepared show 2 eggs and 1 or no miracidium. 11% of all eggs and 67% of miracidium were isolated from *P. freethii*. Altogether, *Onchomelina* contributed 19.2% of all eggs and 33% of miracidium recovered.

Isolated *Paragonimus* eggs were cylindrical in shape and golden brown in colour. Each egg was blunt at the anterior end and with a neck-like construction at the posterior end plate 4a.

Miracidium had green pigmented background and more or less spherical in shape. They wriggled in a spiral forward and slow motion plate 4b.

The cercariae were sperm-like but with a forked tail. Opening and closing of the tail muscles provided the propelling force for the fast moving cercariae.

Rediae were cylindrical in shape with a curved anterior and posterior end. Both ends were separated by a more bulging middle part. One could observe the non motile metacercaria in this cyst (plate 5a and b). The overall result is presented in table 1 below:

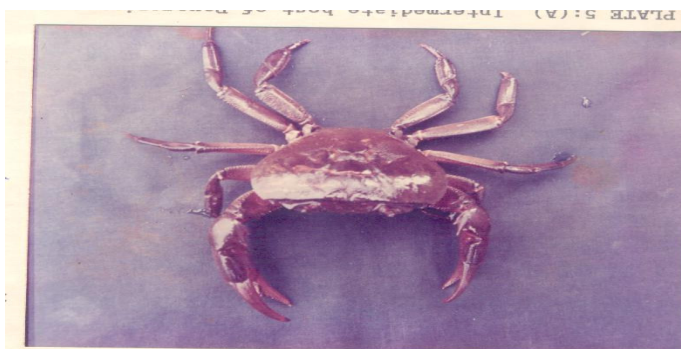


PLATE 1: VENTRAL AND DORSAL VIEWS OF *Pseudanantes africanus* (crab)

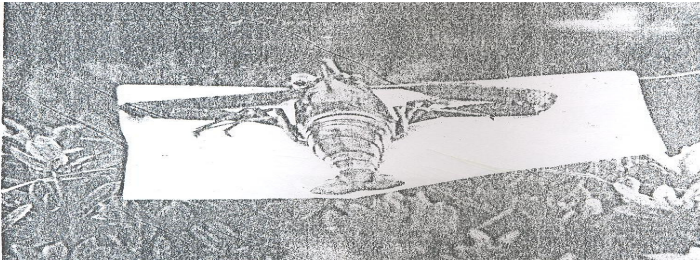


PLATE 2: *Astacus* (crayfish)

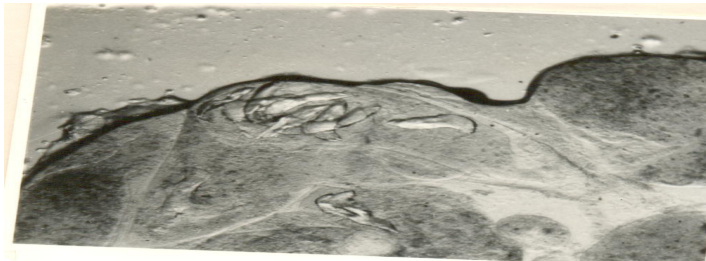


PLATE 3: Mature second generation rediae and sporocysts of *P. Uterobilateralis* from *P. africanus* and *P. freethii*





PLATE 4: (A) *Potedema freethii* (periwinkle) (B) And *Onchomelina* (Water snail)



Eggs



Miracidium

PLATE 5: Egg and Miracidium and CerCariae of *P. Uterobilateralis* from *P. freethii*, *Ochomelina* sp and *Astacus*

Discussion

Examination of suspected vectors of *Paragonimus Uterobilateralis* in Cross River State of Nigeria revealed larvae of the lung fluke. Larvae isolated along with eggs included miracidium, cercariae, sporocysts and mature second generation rediae containing metercariae. *Pseudosanautes africanus* (crab), *Astacus* sp (Crayfish), *Potedema freethii* (Periwinkle), *Onchomelina* sp

(water snail) were incriminated. These vectors were earlier suspected by Nwokolo (1972) and Ikpeme, (1991).

Crab and crayfish contributed the largest percentage of isolated eggs (30.5% and 39.4% respectively). This may be explained by the fast migratory tendencies of these organisms. Such migration was earlier mentioned by Udensi (1987) and it enables the organisms to feed in a variety of niches. Moreover, eggs are freely suspended in water and could be taken faster, during filter feeding, than having to be crushed with leaves in meals by other vectors. The host active enzymes or environmental specificity by parasites and their larvae may not have allowed the hatching of eggs into miracidium in this host. Thus, absence of miracidium in crab and crayfish. 33.9% and 66.1% cercariae, 39.7% and 60.3% rediae were recovered from crab and crayfish respectively. Physiological provision of the body of crab and crayfish could enhance development of ingested cercariae into metacercariae and subsequent encystment into redia.

The occurrence of more cercariae and redia in crayfish than crab may be by chance. Also it is not certain whether the eggs and miracidium invade periwinkle and water snail from water or the eggs hatch into miracidium in these hosts. However, it is here suggested that eggs hatch in water snail into miracidium and are released into water during excurrent processes of the snail. This suggestion is based on the fact that some degenerated eggs were recovered in the intestine of water snail.

Eggs and miracidia were isolated from periwinkle and water snail. This showed that eggs could hatch in these hosts into miracidium. Absence of cercaria and redia containing metacercaria in periwinkle and water snail suggested inadequacy of the environment in these hosts for such development. Parasites are hosts specific for development as earlier mentioned (Klinger and Francis- Floyd, 2002).

The life cycle of *P. Uterobilateralis* as it occurs in Cross River State can be explained as follows: Eggs from sputum and faeces of infected person released into water hatched either in water or periwinkle or water snail into miracidium. Miracidium invade crustacean (crab or crayfish) host where they developed into cercariae and metacercariae. Human definite host become infected through poorly cooked or roasted crab or crayfish meal.

Acknowledgements

We are grateful to Ntufam Ogar Ita Agor, the village head of Old Netim 1, the affected village for granting access to the village and the villagers. The pupils of the community primary school are hereby acknowledged for fishing of crab and snail hosts. We are also grateful to the staff of Biological Science Laboratory of Cross River State University of Technology who assisted in various aspects of equipment monitoring and cleaning.

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Table 1: Distribution of egg larvae of *Paragonimus uterobilateralis* in different vectors

Vector	Eggs Isolated		Miracidium Isolated		Cercariae Isolated		Rediae Isolated	
	Count	Percentage	Count	Percentage	Count	Percentage	Count	Percentage
<i>Pseudanautes Africanus</i> (Crab)	327	30.5%	-	-	20	33.9%	18	39.7%
<i>Astacus</i> sp (Crayfish)	422	39.4%	-	-	39	66.1%	123	60.3%
<i>Potadema freethii</i> (Periwinkle)	116	11.0%	55	67.0%	-	-	-	-
<i>Onchomelina</i> sp(water snail)	206	19.2%	27	33.0%	-	-	-	-
Total	1071		82		59		204	