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Gut Helminth Parasites of *Citharinus citharus* in Anambra River Flood System, Southeastern Nigeria

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Abstract
A total of forty (40) *Citharinus citharus* (Geoffroy Saint-Hilaire, 1809), order (Characiformes) and family (Citharinidae) were purchased randomly from local fishermen who fished in Anambra River from May to October 2012 and examined for the gut helminth parasites. Macroscopic and microscopic examinations were used for parasite isolation and identification. Ten (25.0%) of the fish examined were infected with the two species of helminth parasites *Procamallanus laeviconchus* and *Chariniella citharini*. The parasites were isolated which were restricted to the intestine, except for *P. laeviconchus*, which was also found in the oesophagus and stomach. *P. laeviconchus* was identified in the majority of the fish with a prevalence rate of 15.0% while *C. citharini* was identified in the minority with an infection rate of 10.0%. The highest prevalence (30.0%) of the helminth infection was recorded in fish with total lengths ranging from 35cm-39.9cm while those with lengths ranging from 20cm-24.9cm recorded the lowest prevalence (11.7%). Fish specimens with lengths ranging from 15cm-19cm were not infected. Fish weighing between 250g-349.9g and 350g-449.9g recorded 100% prevalence. Those with weights ranging from 50g-149.9g and 150g-249.9g recorded low prevalence of 13.6% and 31.3% respectively. No juveniles were encountered in this study; only adults and sub-adults were encountered with infection higher in adults (31.0%) than sub-adults (9.1%).

1. Introduction

By virtue of their economic importance to man, especially with the rapidly increasing human population and consequent increase in protein demand, information on the parasites of fish becomes particularly important as these parasites may affect fisheries production (Onusiriuka, 2001 and Oniye *et al.,* 2004). *Citharinus* species are among the major exploitable fish species of the Anambra River. The genus *Citharinus* belongs to the family citharinidae with two species *Citharinus citharus* (Geoffery Saint Hillary, 1809) and *Citharinus latus* (Muller and Troschel, 1845) found in Anambra river. They are common in swamps and rivers throughout Africa they are highly priced and often in smoked-dried form because they die-off immediately, when removed from their habitat. Their deep and flattened bodies earn them the popular name 'moon fish. They feed chiefly on insect larvae, aquatic plants and plankton (Olaosebikan and Raji, 1998). Various parasites are associated with *C. citharus* in the wild and cultured environment. In spite of the premium placed on this fish, existing information on the parasites are very

limited. Virtually every species of free-living organism has parasites (Williams and Jones 1994). Thus parasites contribute significantly to biodiversity simply in terms of the number and variety of species in existence. Unlike in many developed countries where fishing in inland waters has considerable sport and leisure components, the majority of people who fish in Africa, including Nigeria, depend on fish for their livelihood. Thus any large scale occurrence of parasites has not only a negative influence or impact on the fisheries production but would tend to destabilize the means of subsistence of many people in the hinterland especially Nigeria (Onwuliri and Mgbemena 1987). In natural populations, a complex dynamic equilibrium exists between organisms and their environment, both biotic and abiotic. Besides serving .as food, the effect of worms as fish parasites is noteworthy. Among these helminthes, the digenetic, trematodes are considered less dangerous to health of fish compared with the monogenetic trematodes, but they should not be under-estimated especially as some of them may cause unpleasant sickness to humans who eat fish infected with parasite of this group (Onusiriuka, 2001). Helminth infections have been shown to disfigure fish thereby leading to loss of market value for such fish. The palatability of fish is also reduced by helminth infections (Yakubu *et al*., 2002). There is a rich helminth parasite fauna among fresh water fish in Nigeria (Yakubu *et al*., 2002 and Oniye *et al*., 2004) but only a few known to us are specifically directed to address the helminth parasites of *C. citarus*. Under natural conditions, most of the parasites in the environment are cost in the expanse of water, or eaten by other animals, before they can infect fish. Under crowded conditions or inadequate water and oxygen supply, fish may become heavily parasitized; such conditions lead to heavy loss of fish (Onwuliri and Mgbemena 1987). Most fishermen have little or no idea of this, as a result, when they clean their catch; they transfer frequently parasites from infected to uninfected fish. This is considered a serious problem as some of these parasites can survive even when the fish is outside the water rendering a large number of fish unacceptable for human consumption and reducing their market value. A better understanding of fish diseases particularly those caused by animal parasites are essential to a complete fish management programme. Thus the essence of this work is to provide the much needed information on the gut helminth parasites of *C. citarus*.

2. Materials and Methods

2.1. The Study Area

The Anambra river flood system (Fig. 1) is located between latitude 5° 55 and 7° 40 'N and longitude 6° 40' and 7 ° 42'E, it is situated east of the River Niger into which it empties south of the confluence of the Niger and Benue. The river has a total length of approximately 207, 4km and has its source from Ankpa highlands in Kogi state of Nigeria (Azugo, 1978). The Anambra River has a few major tributaries among which are; river Ofu, Okulu, Adada, and Otuocha. (Ita, 1993). The river basin is located in the rain forest zone with adequate rainfall. The period of rainy season begins in April and lasts for seven months up to October, with a short break between late July and early August. Many variations have been known to occur specially in time of the second rains. During the rains, water level increases drastically in the main river channel. The rise in the water levels of the river channels is brought about by direct precipitation within the catchments area as well as by inflow from the Niger flood plains. The inundated soils are composed of clay with good water retention capacity and hence several flood plains are left behind after the flood season. The period November to March is usually that of the dry season. The water level is restricted to the main river channel and to some flood plain ponds. This season is associated. With maximum zooplankton and phytoplankton increase, this has a marked correlation with productivity. The River normally has its bank covered by such plants like *Echinocloa colonum*, *Salvinia* etc. (Nwani, 1998).

2.2. Sampling of Fish

A total of forty fish samples of *C.citharus* were collected from May to October 2012 for helminth parasites. A variety of fishing techniques (Castnets, dragnets, fish traps) were used to capture the fish but the most common method was the use of cast nets. The fish were preserved in formalin and transported in plastic containers to the laboratory of the department of Applied Biology, Ebonyi State University, Abakaliki, Nigeria. In the laboratory, identification of fish samples was done using the method of Olaosebikan and Raji (1998). Using a meter rule and a digital electronic balance the total length (TL) of the fish and the weight of the fish samples were determined and recorded respectively. The age of the fish were determined using their length, convincingly assuming this to be a reasonable indication of their relative ages (Holden and Reed, 1972), thus based on the measurement of the total, they were categorized into juveniles (5-10cm) sub adults (11-20cm) and adults (21cm above) (Ugwuzor, 1987).

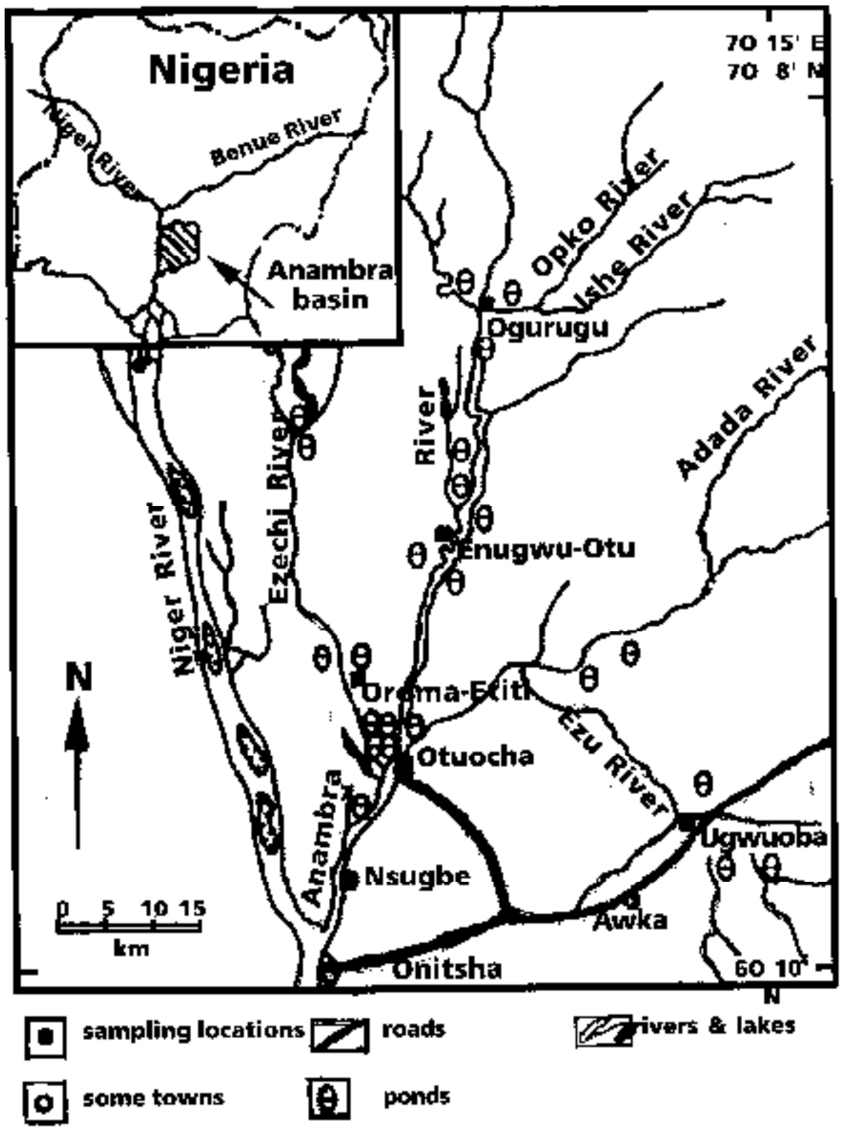

Fig. 1. Map of Anambra river flood system showing the sampling locations

2.3. Isolation, Collection and Identification of Parasites

The fish were examined individually for parasites according to the methods described by Roberts (1978). The fish were dissected as a mid-ventral cut was made on fish from the body wall was cut open at both sides, enabling a thorough exposure and examination of the alimentary canal. The alimentary canal was isolated and the various sections of the gut (Oesophagus, Stomach and intestine), were placed separately on Petri dishes containing normal physiological saline. Each section was slit longitudinally with a dissecting knife and carefully and thoroughly examined for endoparasitic helminth using a magnifying hand lens. The numbers of parasites recovered per fish were isolated and their locations/positions of attachment recorded. With the aid of a pin, forceps and camel hairbrush, the parasites isolated were collected after rinsing the sections of the gut in the saline solution in the Petri dishes great care was taken in collection of parasites in order not to damage parts of their bodies. The helminth parasites recovered were put in vials and duly labeled, indicating the name of host; location in the fish from which the parasites were recovered. The nematodes were placed on clean glass sides a drop of water was added and the specimen covered with cover slip and examined under the microscope. The parasites were cleaned overnight in warm iron-haematoxylin according to the methods described by Roberts (1978). Parasites recovered were identified with the aid of a microscope using the keys of Yamaguti (1957) and Khalil (1971). Analysis of data was done using the infection statistics (Mangolis *et al*., 1982).

3. Results

Of the forty (40) fish samples examined 10 (25%) were infected. A total of eighty (80) nematodes, belonging to two species of two families. Of the two species of nematode recovered, *Procamallanus laeviconchus* (Wedl, 1862), order (Camallanida) and family (Camallanidae) occurred in the majority (6) of the fish examined with an infection rate of 15%

and mean intensity of 9.83 while *Chathariniella citharini* (Khalil, 1964), order (Oxyurida) and family (Pharyngodonidae) occurred in the minority (4) of the fish examined with an infection rate of 10% and mean intensity of 5.25 (Table 1). Analysis of infection rate data above indicate that with the small number of fish samples examined the rate of intensity of infections was generally high.

Table 1. *Infection rate data of C. citharus*

Parasite species	No. of fish infected	No. of parasites recovered	Prevalence (%)	Mean intensity	Abundance
P. laeviconchus	6	59	15	9.83	1.48
C. citharini	4	21	10	5.25	0.53

Data concerning the attachment of the nematodes in different regions of the gut showed great, affinity for-the intestine. The intestine recorded the highest prevalence of 22.5% and mean intensity of 6.44. The oesophagus recorded the lowest prevalence of parasites (5%) but with a mean intensity of 4.5, which is considerably higher than that of the stomach (Table 2). *P. laeviconchus* occurred in the three regions of the gut (Oesophagus, stomach and intestine)'at infection rates of 5%, 10% and 12.5% respectively, while *C. Citharini occurred* only in the intestine at an infection of rate 10% (Table 2). It can also be deduced from the table that percentage occurrence of *P. laeviconchus* was highest (12.5%) in the intestine, and least (5%) on the Oesophagus.

Table 2. *Distribution of P. laeviconchus and C. citharini in different regions of the gut of C. citharus*

Attachment site	Parasite	No. of host infected	No. of parasites found	Prevalence (%)	Mean intensity	Abundance
	A+B	2	9	5	4.5	0.23
Oesophagus	A	2	9	5	4.5	0.23
	[B]	[-]	[-]	[-]	[-]	[-]
	A+B	4	13	10	3.25	0.33
Stomach	A	4	13	10	3.25	0.33
	[B]	[-]	[-]	[-]	[-]	[-]
	A+B	9	58	22.5	6.44	1.45
Intestine	A	5	[37]	12.5	7.40	0.93
	[B]	[4]	[21]	[10]	[5.25]	[0.60]

A = P. laeviconchus
[B] = C. citharini
A+B refers to data obtained from pooled total of the two species of the parasites

Table 3 shows the prevalence, intensity and abundance of parasites in the gut of different length classes of *C. citharus*. The mean intensity of the parasites was highest (11) in fish of total lengths 35-39.9cm and lowest in fish of total length 30-34.9cm. Only one fish was examined within the total length range of 35-39.9cm and had 100% prevalence rate, which was the highest prevalence, recorded. The lowest prevalence of 11.77% was recorded in fish of total length 20-24.9cm. Fish with total lengths of 15-19.9cm did not harbor parasites. It was observed that the prevalence of each parasite species increased with increase in length of host. Size of fish therefore, has a marked relationship with parasitism. Fish of length ranging from 20-24.9 recorded the lowest prevalence of *P. laeviconchus* and *C. citharini*. *C. citharini* did not infect fish with total length range of 35.39.9cm.

Table 3. *Infestation rate data of P. laeviconchus and C. citharini in the gut of different length ranges of C. citharus.*

Length range (cm)	No. (%) of fish examined	No. of host infected	No. of parasites found	Prevalence (%)	Mean intensity	Abundance
		A+B 0	0	0	0	0
15-19.9	6(15)	A 0	0	0	0	0
		B [0]	[0]	[0]	[0]	[0]
		A+B 2	16	11.77	8	0.94
20-24.9	17(42.5)	A 1	13	5.88	13	0.77
		B [1]	[3]	[5.88]	[3]	[0.18]
		A+B 4	32	33.33	8	2.67
25-29.9	12(30)	A 2	19	16.67	9.5	1.58
		B [2]	[13]	[16.67]	[6.5]	[1.08]
		A+B 3	21	75	7	5.25
30-34.9	4(10)	A 2	16	50	8	4
		B [1]	[5]	[25]	[5]	[1.20]
		A+B 1	11	100	11	11
35-39.9	1(2.5)	A 1	11	100	11	11
		B [0]	[0]	[0]	[0]	[0]

A refers to P. laeviconchus
B refers to C. citharini
A+B refers to data obtained from pooled total of the two species of the parasites

The number of examined host within weight range of 50-149.9g was highest (22) with prevalence of 13.63% which is the lowest prevalence rate recorded. Maximum infection of 100% each was recorded in fish with weight range between 250-349.9g and 350-449.9g (Table 4). *P. laeviconchus* occurred in the whole weight ranges of fish countered, having its highest prevalence of 100% each in fish with weight ranging from 250-349.9g and 350-449.9g; *C. citharini* did not occur in fish of this weight ranges. *C. citharini* infection occurred in fish with weight range between 50-149.9g and 150-249.9g only, having its highest prevalence rate of 12.5% in fish of weight ranging from 150-249.9g and its lowest prevalence rate of 9.09% in fish of weight range between 50-149.9g. Prevalence rate of each parasite species increased with increase body weight of fish.

Table 4. *Infestation rate data P. laeviconchus and C. citharini in the gut of different weight ranges of C. citharus.*

Weight range (g)	No. (%) of fish examined	No. of host infected	No. of parasites found	Prevalence (%)	Mean intensity	Abundance
		A+B 3	28	13.63	9.33	1.28
50-149.9	22(55)	A 2	25	9.09	12.5	1.14
		[B] [1]	[3]	[4.54]	[3]	[0.14]
		A+B 5	36	31.25	7.20	2.26
150-249.9	16(40)	A 2	18	12.5	9	1.13
		[B] [3]	[18]	[18.75]	[6]	[1.13]
		A+B 1	4	100	4	4
250-349.9	1(2.5)	A 1	4	100	4	4
		[B] [0]	[0]	[0]	[0]	[0]
		A+B 1	11	100	11	11
350-449.9	1(2.5)	A 1	11	100	11	11
		[B] [0]	[0]	[0]	[0]	[0]

A refers to P. laeviconchus
[B] refers to C. citharini
A+B refers to data obtained from pooled total of the two species of the parasites

Only sub-adult and adult fish were collected and examined for helminth infections (Table 5) No juveniles were encountered. Prevalence of infection increased with increase in age of fish host, being more in adults (31.03%) than in sub-adults (9.1%) (Table 5). It is also observed that *P. laeviconchus* had the highest prevalence rate of 17.2% in adults with *C. citharini* recording the lowest prevalence rate of 13.79% in adults. *C. citharini* occurred only in fish of this age group.

Table 5. *Relationship between host age and parasitization with P. laeviconchus and C. citharini.*

Age group (cm)	No. (%) of fish examined	No. of host infected	No. of parasites found	Prevalence (%)	Mean intensity	Abundance
Juveniles (5-10)	-	-	-	-	-	-
		A+B 1	13	9.1	13	1.18
Sub-adults (11-20)	11(27.6)	A 1	13	9.1	13	1.18
		[B] [0]	[0]	[0]	[0]	[0]
		A+B 9	67	31.03	7.44	2.31
Adults (21-40)	29(12.5)	A 5	46	17.20	8.20	1.59
		[B] [4]	[21]	[13.79]	[5.25]	[0.72]

A refers to P. laeviconchus
[B] refers to C. citharini
A+B refers to data obtained from pooled total of the two species of the parasites

4. Discussion

The nematodes recovered from *C. citharus* of Anambra river flood system were *P. laeviconchus* and *C. citharini.* Infection *of C. citharus* with nematodes is probably acquired through eating fish harbouring viable adult worms. Khalil (1969) pointed out that the susceptibility of fish to infection with parasitic helminths depends on various factors including the feeding habit of the host. The nematodes recorded in this present study are among those check listed by Khalil (1971), Paperna (1980) and recovered by Azugo (1978) among several authors of freshwater fish parasites in Africa.

The absence of cestodes, acanthocephalans and trematodes in this study is worthy of note. The absence of these groups of helminth may be due to the absence of suitable intermediate host required for the transmission of these groups of helminthes in the aquatic environment (Paperna, 1980 and Ugwuzor, 1987). The absence of acanthocephalans in this study however, contrasts the findings of Balarin (1979) where species of acanthocephalan were reported to be the commonest parasite of freshwater fish in African sub-region. Conversely, Paperna (1980) on data of infection levels in Africa by acanthocephalans showed that even though fish were found heavily infected in Lake Chad, they were not in the same group of fish in the low Niger River. This implies that the incidence of parasitic infection differs from one

location to another due to the interplay of ecological factors of the environment.

The occurrence of nematodes only in ten (10) of the forty (40) fish examined agrees with earlier findings that nematode infection is common in fish. This may be as a result of the size and food preferences, which may affect the abundance of parasites; as reported by Ugwuzor (1987). This is similar to the observations of Okaka (1991) who isolated three nematodes form *Oreochromis niloticus and T. zilli,* from Asa River, Onwuliri and Mgbemena (1987) who reported that nematode infection was common among freshwater fish from Jos and Ugwuzor (1987) who recorded five nematodes out of the seven helminths recovered in Imo River. The observed differences in prevalence of nematode infections in present study and these previous studies could be due to variations in ecological factors of the various aquatic habitats (Awaritoma and Okaka, 1999).

In all these previous reports, high infection rate of *Cammalanus* species and *Procamallanus* species were recorded, showing that these two nematodes are probably the most frequent parasites of fish in Nigeria. In this present study, although no *Cammalanus* species was recorded, the prevalence and occurrence of *P. laeviconchus* was highest, this agrees with the previous reports. This could be attributed to the probably relative abundance of the intermediate host of *P. laeviconchus*, which according to Paperna (1980) is Cyclopoid copepods.

The relative abundance and prevalence of the 2 nematodes varied form one anatomical site of recovery to another. The intestine stomach and oesophagus were observed to be infected. This regional localization could be attributed to several factors like the nutrient level, pH, Osmotic tension and oxygen tension operating in the gut as well as food reserves (Ugwuzor, 1987 and Onusiriuka, 2001). According to Smyth (1979), with a few exceptions, the food materials of adult nematodes appear to be solid or semi-solid digested food and debris. This could account for the predilection sites of majority of the nematodes found in the intestine (72.5%) in contrast with 16.25% and 11.25% that were found in the stomach and oesophagus respectively. The observation that the preferred site of parasitic infection in the gut is the intestine agrees with the findings of Khalil (1971), Ugwuzor (1987), Okaka (1991) and Oniye *et al*., (2004). Another feature of considerable interest in the attachment sites of the nematodes is that the two nematodes were restricted to the intestine except for *P. laeiconchus* that was also found in the stomach as well as in the oesophagus. Bushmann and Lindenstrom (2002) suggested that parasites have an in-built molecular disguise to avoid the host hostile secretions present in its microhabitats. This explains the reason for the nematodes attachment to specific organs of the gut.

An increase in size is "a reflection of increase in length and weight, which is hereby considered as the measure of age (Holden and Reed, 1972). From this study, it was observed that the length and weight of fish affects the rate of parasitic infection, as the rate of infection with each parasite species increased with increase in length and weight of fish host, no juveniles were encountered. Infection rate and parasite burden varied among the age groups, being higher in adults than sub-adults. However, the intensity of infection varied according to other factors related to size (length, weight and age) of host. Such factors might be related to the behaviours of the host, breeding and other physiological characteristics of various sizes of *C. citharus.*

Another plausible explanation for the pattern of infection observed could be due to certain factors emanating from the feeding differences or change of diet between the various age groups. Similar observation of infected being higher in the adults was also reported by Roberts (1978), Ugwuzor (1987), Anosike et al (1992) and Oniye et al, (2004). Roberts (1978) noted that large fish provided greater surface for infection than smaller fish. Conversely Awaritoma and Okaka (1999) and Okaka and Omoigberale (2002) recorded higher infection in smaller fish than adult fish.

In conclusion, the findings of this study suggest that *C. citharus* can harbor high worm load of helminth parasites. There is therefore need for future studies that will assess the impact of infection with these parasites on the survival of *C. citharus* in tropical aquatic habitat.

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