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Comparative Study and Zoonotic Implications of Endoparasitic Fauna in Chrysichthys nigrodigitatus from Great Kwa River and Calabar River, Cross River State, Nigeria

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Abstract
A total of two hundred *Chrysichthys nigrodigitatus* samples bought from artisanal fishermen in Nsidung beach of Calabar River and Obufa Esuk Beach of Great Kwa River and subjected to parasitological investigation, 67 (33.5%) were infected and 97 parasites recovered. The endoparasites recovered from the organs of *Chrysichthys nigrodigitatus* were *Diphyllobothrium latum, Capillaria philippinensis, Schistocephalus solidus*, and *Neoechinorynchus rutili.* Sex related infection was observed to be higher in female fish 15 (13.6%) than the male fish 10 (9.1%) from Great Kwa River, and 24 (26.7%) in females than 18 (20.0%) in males from Calabar River. There was no statistical significant difference ($p > 0.05$) between increased endoparasitic infection of female fish and male fish. Generally, more parasites 55 (56.7%) were recovered from Calabar River than Great Kwa River 42 (43.3%). This study revealed that larger fish from Great Kwa River and Calabar River had highest parasitic infection (32.0%) and (38.0%) respectively in length range 51-60 cm. However, the highest weight related infection (28.6%) and (26.2%) in *C. nigrodigitatus* were recorded in weight range 201-300g from Great Kwa River and Calabar River respectively. Apart from the cheap protein supplies of this fish to humans, when eaten raw their parasites cause zoonotic diseases like Diphyllobothriosis and intestinal capillariasis.
Keywords: Endoparaites, *Chrysichthys nigrodigitatus*, zoonotic implications, Great kwa, Calabar River.

Introduction
Silver catfish or *Chrysichthys nigrodigitatus* is indigenous to Nigeria and most West African countries. Freshwater fish in developing countries of Asia and Africa are more at risk of extinction than in other developing countries (Craig, 2000). It has been established that one factor responsible for such decline is the negative impact of parasites on host-growth and survival as demonstrated in several parasite-fish system in both aquaculture and natural population (Yanong, 2000). Fish parasites have serious pathological changes, retard growth, lowers fish quality and subsequent death, leading to economic losses in fish industry (Klinger and Francies Floyd, 2002). The most common parasites of fish are Protozoa (Uneke , et al., 2015). Ray Sahelian (2012), reported that more than 70 species of Protozoa and helminth parasites get to humans through consumption of sea food like fishes, molluscs, frogs, tadpoles, camarons and crayfish. *C. nigrodigitatus* has an omnivorous feeding habit which exposes it to a variety of parasites and negatively impact on its health (Sidney et al, 2014). But due to its role in the food chain of man, zoonotic infection arising from ingesting raw fish, knowingly or unknowingly, could cause serious problems (Ikomi, 2012; Nelson, 2006). Infected fish species manifest numerous symptoms including anaemia, lesions, ulcers, skin and fin rot, distended stomach, blockage of the intestine, pop-eyes, immune suppression, retard growth, loss of appetite, depressed reproduction, emaciation and loss of organ (Ajayi, 1983; FAO, 2014; Paperma, 2014, Woo, 2011). Parasites also compete for food, thereby depriving fish of essential nutrients and inhibiting growth leading to morbidity and mortality with consequent economic losses (Khalil and Polling, 1997). Accoding to Omeji et al. (2013), fish is the most parasitize of all vertebrate and the importance of parasitic infection on fish production has largely remained an issue of concern to fish farming industry. Some parasites have been discovered to have zoonotic potential in mammalian host including man thereby making them of public health importance. In Nigeria, a large scale of fish supply comes from river systems where fish are landed at fishing jetties and purchased for distribution by large and small scale fish dealers (Ekanem et al., 2011). The increased demand for *Chrysichthys nigrodigitatus* as a delicacy and a cheap source of protein to people in Calabar necessitate comparism of its endoparasitic fauna and zoonotic implications from Great kwa River and Calabar River in this study.

2. Materials and Methods
2.1. The Study Area
Great Kwa River and Calabar River are the major tributaries of the Cross River Estuary. Both Rivers take their rise from the Oban hills and drain through heavily forested landscapes of South Eastern Nigeria, to Calabar city (Fig. 1). The city stands $4^0$57' N of the Equator and $8^0$20' E. Calabar is situated on a peninsular stretching between Great Kwa River on the eastern boundary and Calabar River on the western boundary (Olu Sule, 1987). Calabar River joins the main Cross River at about 8 km^2 to the south, while the Great Kwa River discharges into

the Cross River Estuary around latitude $4^{\theta}45'$ N and longitude $8^{0}20'$E (Akpan, 2000). The river system which is part of Cross River and Great Kwa River including other tributaries form extensive flood plain and wetlands that empty into the Cross River Estuary. The major fish exploited by the artisanal fishermen in Great Kwa River and Calabar River include croakers, clupeids Shrimps and catfish, our study specimen.

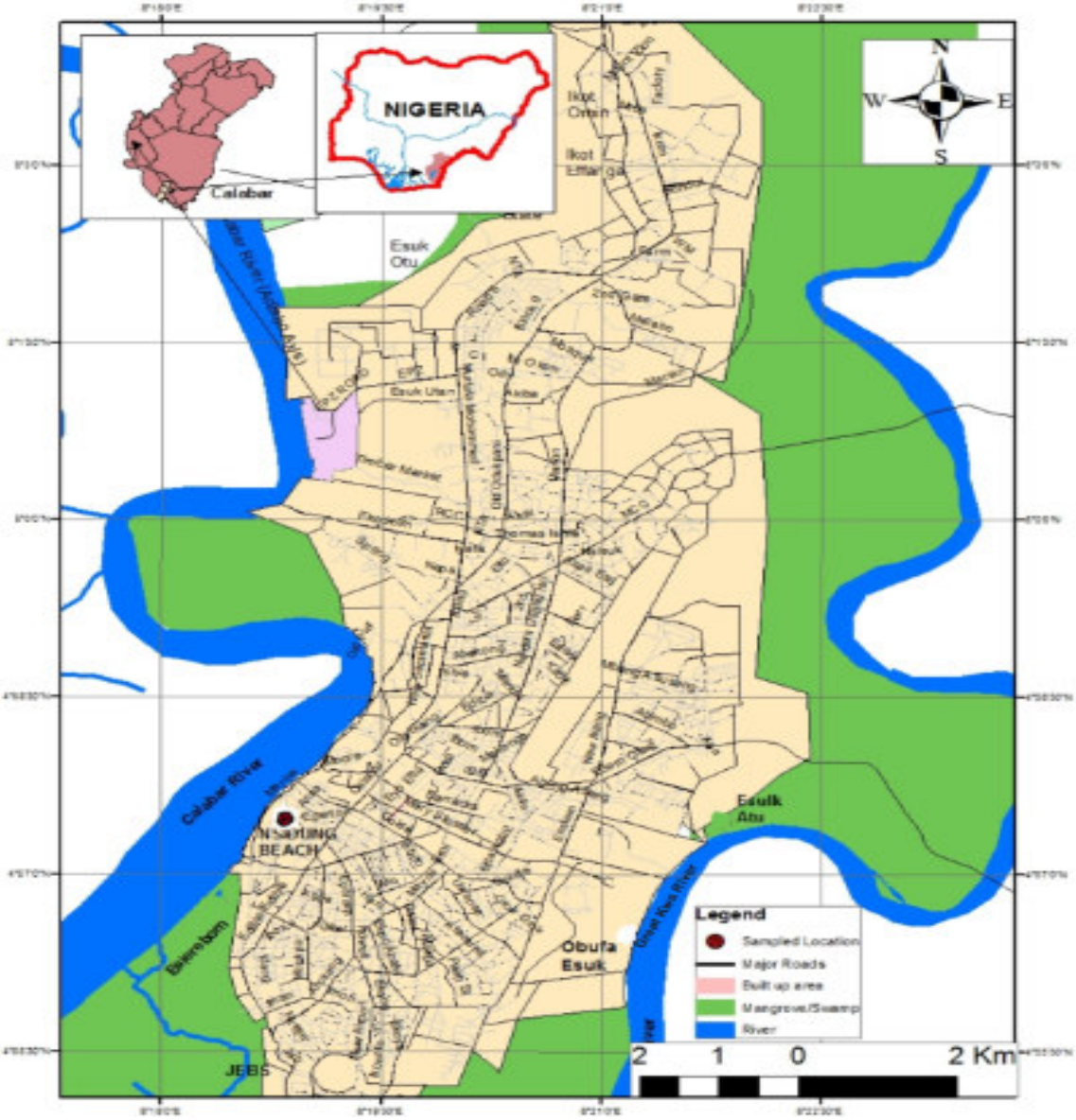


Fig.1: Map showing the study area, Calabar and Great Kwa Rivers

2.2. Collection of Fish Specimen

The fishes used for this study were bought from artisanal fishermen at Nsidung beach along Calabar River and Obufa Esuk beach along Great Kwa River. The methods of fishing in these rivers include hook and line, cast nets and set lines. The fish caught is sold to both far and near local market women. A total of 200 fishes were bought between August and October 2015, and transported to the Biological Science Laboratory of Cross River University of Technology, Calabar, for parasitological investigation. Purchases were made thrice monthly.

2.3. Weighing and Measurement of Fish Specimen.

The weights and total lengths of the fishes were recorded. The weight was taken with the aid of a standard top loading Denward balance, while the lengths of the fishes were measured with a metre rule. The length in this case was considered as from the beginning of the snout to the end of the caudal fin.

2.4. Dissection and identification of fish parasites

The fish was placed on the dissecting board and the abdomen cut open from the anal aperture to the operculum

with scissors. The gastrointestinal tract was carefully extracted and placed on a Petri dish containing normal saline. The fish sex was determined by noting the presence or absence of ovaries. The different parts, stomach, small and large intestine, heart and liver were further slit open to aid the emergence of parasitic worms. The content was allowed to mix with the normal saline and finally processed for microscopic examination using the formal-ether concentration method (Zdenek, 1977; Sikoki et al., 2013; Al Mofarreh et al., 2000). The parasites were identified using keys provided by Pouder et al. (2005).

Results

Two hundred fishes were subjected to parasitological investigation. Of the one hundred and ten (110) fish samples examined from Great Kwa River, 25 (12.5%) were found to be infected and 42 parasites recovered from them. Out of 90 fishes examined from Calabar River 42 (21.0%) were infected and 55 (56.7%) parasites extracted from these fishes. A total of 67 fishes were infected in this study giving an overall infective rate of 33.5% in fishes investigated, and 97 parasites recovered (Table 1).

Table 2 shows the prevalence of endoparasitic fauna according to the site of infection in C. nigrodigitatus from both rivers. The Parasites recovered in this study are *Diphyllobothrium latum, Capillaria philippinensis, Schistocephalus solidus* and *Neoechinorhynchus rutili* (Plate 1). From Great Kwa River, 7 fish samples harboured 10 (23.8%) *Diphyllobothrium latum*, while 5 fish from Calabar River were infected with 8 (14.5%) *D. latum. Diphyllobothrium latum* species were extracted in the liver of *C. nigrodigitatus* from both rivers. From Calabar river, 25 (45.5%) *Capillaria philippinensis* were found in the intestine of 18 fish samples, although 18 (42.9%) Capillaria philippinensis were harboured by 9 fish specimens from Great Kwa River. Table 2 also revealed that 8 (19.0%) *Schistocephalus solidus* were encountered in the intestine of 4 fish samples from Great Kwa River, as compared to 12 (21.8%) *S. solidus* recovered in 9 fishes from Calabar River. Five fishes from Great Kwa River revealed the presence of 6 (14.3%) *Neoechinorhynchus rutili* ova in their heart, in contrasts with the extraction of 10 (18.3%) *N. rutili* in 10 fish samples from Calabar River. However, the fish intestine had the highest total parasite occurrence of 63 (64.9%), followed by the heart 10 (18.2%) and finally the liver with 8 (14.5%) (Table2).

Table 3 illustrates that the highest endoparasitic fauna infection 32.0% and 38.0% of fishes were recorded in length range 51-60 cm from Great Kwa River and Calabar River respectively. The least infection of 8.0% was shown in fish length range 0-20 and 21-30 cm from Great Kwa River and 4.8% in fish length range 0-20 cm from Calabar River. Infection rate of fishes increases from smaller size fishes to the larger ones. The highest parasites recovered 26.2% and 45.5% were also seen at the same fish length range 51-60 of both rivers.
In table 4, the highest infection rate of endoparasitic fauna of 28.6% and 26.2% occurred in fish weight range 201-300g of the two rivers. However, the highest parasites 28.6% recovered from Great Kwa River and 29.1% from Calabar River were found at fish weight range 201-300g in both rivers.

Table 1. Prevalence of endoparasitic fauna according to the river sampled

River	No of fish examined	No. of fish infected	Percentage infected (%)	Parasites (%) recovered
Great Kwa River	110	25	12.5	42 (43.3)
Calabar River	90	42	21.0	55 (56.7)
Total	200	67	33.5	97 (100)

Table 2. Prevalence of endoparasitic fauna according to site of infection in *C. nigrodigitatus* from Great Kwa River and Calabar River.

		Great Kwa River		Calabar River		
Parasites	Site of infection	No of fish infected	Percentage(%) parasites recovered	No of fish infected	Percentage (%) of fish recovered	Total (%) parasites recoverd
Diphyllobothrium Latum (Cestode)	Liver	7	10 (23.8)	5	8 (14.5)	12 (17.9)
Cpillaria philippinensis (Nematode)	Intestine	9	18 (42.9)	18	25 (45.5)	27 (40.3)
Schistocephalas solidus (Cestode)	Intestine	4	8 (19.0)	9	12 (21.8)	13 (19.4)
Neoechinorhynchus rutili ova (Acanthocephalan)	Heart	5	6 (14.3)	10	10 (18.2)	15 (22.4)
Total		25	42 (100)	42	55 (100)	67 (100)

Table 3. Prevalence of endoparasitic fauna according to length (cm) of *C. nigrodigitatus* from Great Kwa River and Calabar River

Length (cm)	Great KwaRiver			Calabar River		
	No of fish examined	No and % of fish infected	No and % Parasites recovered	No of fish examined	No. and % of fish infected	No. and % Parasites recovered
0-20	10	2 (8.0)	4 (9.5)	8	2 (4.8)	4 (7.3)
21-30	16	2 (8.0)	6 (14.3)	12	4 (9.5)	5 (9.1)
31-40	35	4 (16.0)	8 (19.0)	13	6 (14.3)	7 (12.7)
41-50	20	5 (20.0)	10 (23.8)	14	7 (16.7)	8 (14.5)
51-60	12	8 (32.0)	11 (26.2)	27	16 (38.0)	25 (45.5)
61-70	17	4 (16.0)	3 (7.1)	16	7(16.7)	6 (10.9)
Tatal	110	25 (100.0)	42 (99.9)	90	42 (100.0)	55 (100.0)

Table 4 Prevalence of endoparasitic fauna in *Chrysichthys nigrodigitatus* according to weight range from Great Kwa River and Calabar River

weght (g)	Great Kwa River			Calabar River		
	No. of fish examined	No. and % of fish infected	No. and % parasites recovered	No. of fish examined	No. and % of fish infected	No and % parasites recovered
0-100	10	2 (8.0)	4 (9.5)	8	3 (7.1)	7 (12.7)
101-200	16	3 (12.0)	6 (14.3)	12	5 (11.9)	8 (14.5)
201-300	35	8 (32.0)	12 (28.6)	27	11 (26.2)	16 (29.1)
301-400	20	5 (20.0)	10 (23.8)	14	9 (21.4)	12 (21.8)
401-500	12	4 (16.0)	7 (16.7)	13	8 (19.0)	7 (12.7)
501-600	17	3 (12.0)	3 (7.1)	16	6 (14.3)	5 (9.1)
Total	110	25 (100.0)	42 (100.0)	90	42 (99.9)	55 (99.9)

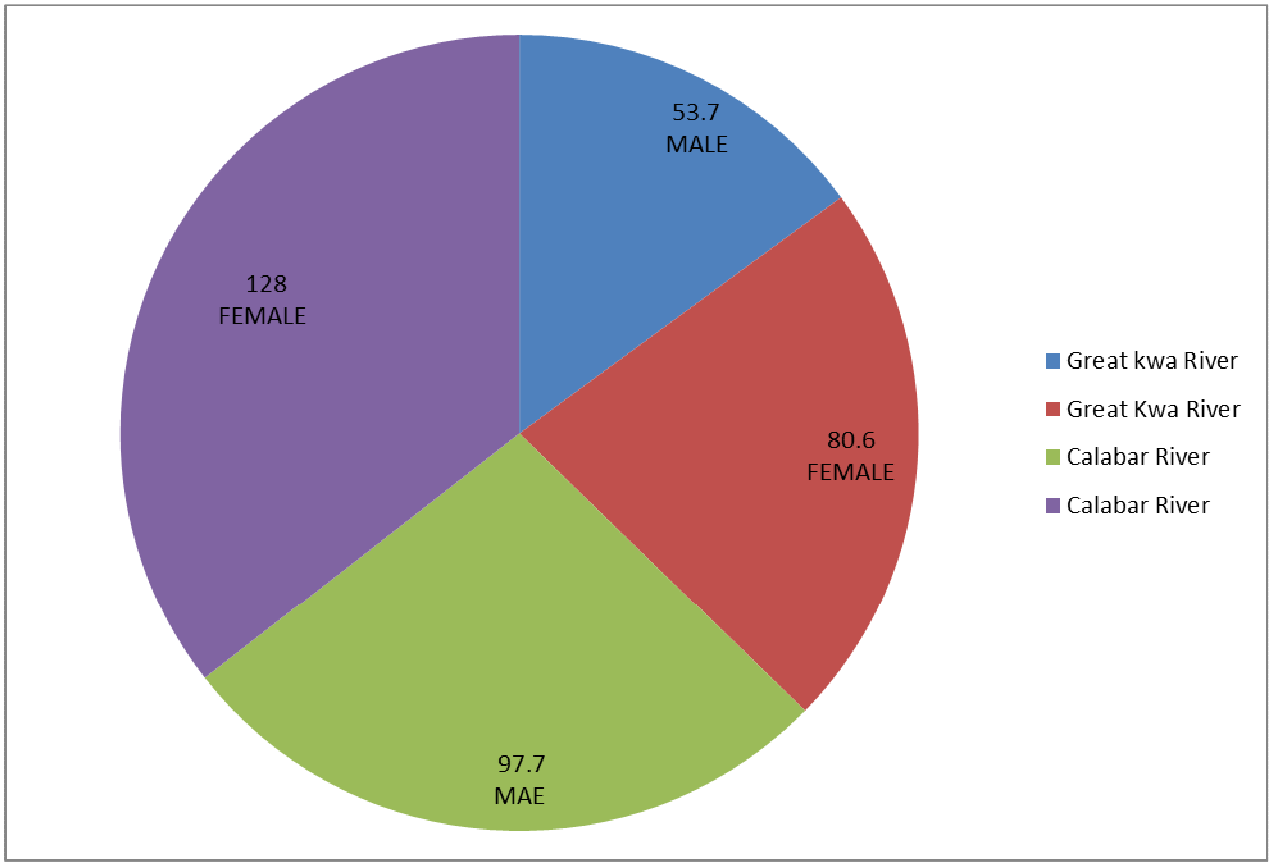
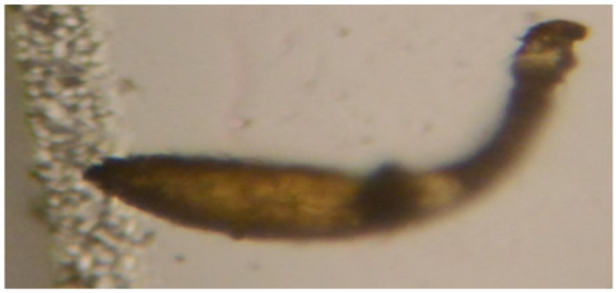
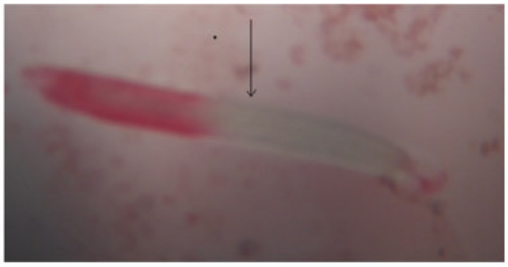


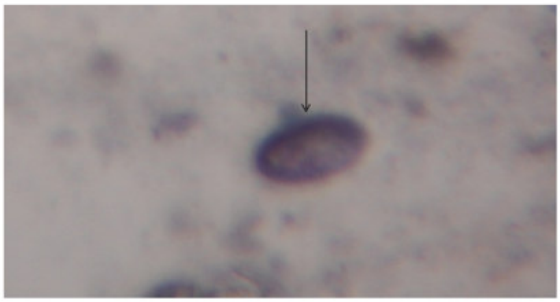
Fig. 2. Pie Chart showing sex related infection in *C. nigrodigitatus* from both rivers



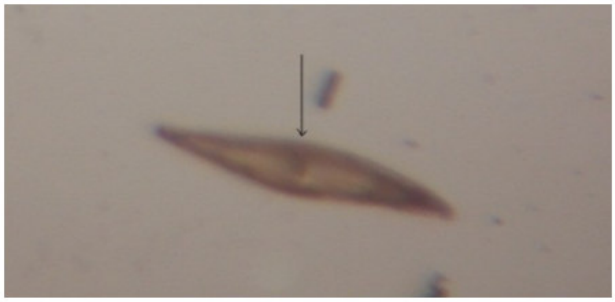
Diphyllobothrium latum X40



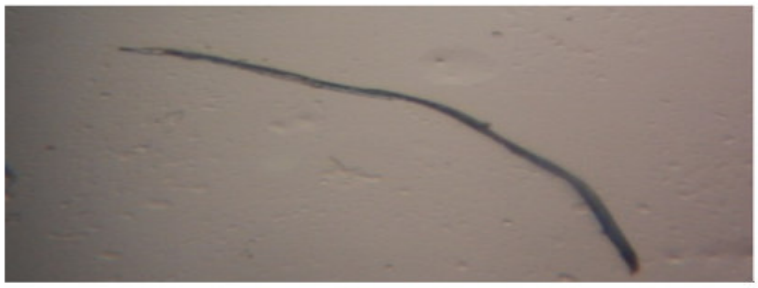
Schistocephalus solidus X40



Capillaria philippinensis ova X40



Neoechinorhynchus rutili ova X40



Capillaria philippinensis adult X40

Plate 1: Pictorial representation of parasites recovered in *C. nigrodigitatus* from both rivers.

Discussion

The investigation results of this study revealed an overall infection rate of 33.5% in *Chrysichthys nigrodigitatus* examined from Great Kwa River and Calabar River. This finding is higher than 12.7 reported by Akinsanya et al,. (2007) at Lekki, Lagos State; 3.33% by Ekanem et al., (2011) at Great Kwa River, Calabar, Cross River State; 20.0% by Sidney et al., (2014) at new Calabar, Rivers State. In this study, three genera of parasites namely, Cestoda, Nematoda and Acanthocephalans were extracted from *C. nigrodigitatus* organs. The cestodes include *Diphyllobothrium latum* and *Schistocephalus solidus*, the nematodes were *Capillaria philippinensis* ova and adults and the acanthocephalans were *Neoechinorhynchus rutili.* The nematodes were the highest occurring parasites, followed by the cestodes and finally the acanthocephalans. It was observed that the intestine harboured the highest number of parasites as earlier reported by Akinsanya et al., (2007), Ekanem e al., (2011) and Sidney et al., (2014), who held that most digestive activities occur in the intestine, resulting in the presence of absorbable food nutrient on which nematodes depend.

Sex related infection (Fig. 2) shows that female fish had more overall infection (58.2%) compared with their male counterparts (41.8%). This observation is in conformity with the reported work of Obiekezie and Enyenihi (1988); Ekanem et al., (2011) and Uneke et al., (2015). There was no statistical significant difference ($p > 0.005$) in the infection of females than males in this study as earlier observed by Sikoki et al., (2013). However, this finding is at variance with the work of Awharitoma and Okaka (2000), and Omeji et al., (2013), who reported more infection in males than females. Generally, low parasitic infection was observed in fish from Great Kwa River than Calabar River. The low infestation rate of fish from Great Kwa River could be due to its sanitary condition, located away from residential buildings and low visitation of people for laundry, and defecation, which is the reverse in Calabar River and thus higher infestation of fish. This observation corroborate the work of Ekanem et al., (2011) in Calabar, Cross River State. Prevalence of parasitic infection according to

the size of *C. nigrodigitatus* revealed an increased infection in bigger fishes than smaller ones. The recorded highest infection rates of (32.0%) and (38.0%) in bigger fishes of length range 51-60 cm from Great Kwa River and Calabar River respectively , is in agreement with the universal pattern of parasitic infection in fishes (Bello-Olusoji et al., 2011; Ekanem et al., 2011; Chowdhury and Hossain, 2015). The increase in parasitic infection as fish age increases, is attributable to wide range search for food which predisposes them to infection. This finding is in sharp contrast with the reported work of Ekanem et al., (2011) who revealed highest infection rate in lower length class. However, the recorded highest parasitic infection rate (32.0%) and (26.2%) in weight range 201-300 g of Great Kwa River and Calabar River could be due to their great search for food and survival needs. It was observed that larger weight range had more parasites than smaller weight range. These results agree with the reported works of Omeji et al., (2011) and Uneke et al., (2015). The high parasitic infection observed in this study is of public health concern.

Worthy of mention in this study is the recovery of Diphyllobothrium latum, Capillaria philippinensis and *Neoechinorhynchus rutili* (an acanthocephalan) from *C. nigrodigitatus* and their zoonotic implications. *C. nigrodigitatus* is a delicacy and widely used by people in Calabar and beyond. Apart from its cheap protein supplies to humans, it has zoonotic disease consequences. Diphyllobothriosis is a well known disease of humans especially in Europe, where infections caused by 3 species of *Diphyllobothrium* have recently been reported. *D. latum* isolated in this study has been reported as the principal species infecting persons in Europe (Kuchta et al., 2014). World-wide, at least 13 species of Diphyllobothrium have been reported from humans, with infections by D. dendriticum being the most prevalent Adams et al. (1997). D. latum has been of interest for many years because the cestode causes pernicious anaemia, probably due to competition between the worm and the host for vitamin B_{12} (Adams et al. (1997). *Capillaria philippinensis* has been incriminated for the deaths of people in the Philippine and Thailand (John, 1992). According to John (1992), populations in which intestinal capillariasis occurs have unique eating habits in that they prefer to eat fish raw. However, Heinz (2008) reported that two species of archiacanthocepalans and three species of Palcanthocephalans have been found in humans in parts of Asia, with associated symptoms of weight loss, intermittent fever, bulging abdomen, diarrhea and severe pains. Often Acanthoephalans in human species occupies extra-intestinal positions, and the migration of these perforating acanthocephalans through the gut wall causes severe pains (Heinz, 2008).

In conclusion, caution must be exercised by people clamouring for the cheap protein in *C. nigrodigitatus* from Great Kwa River and Calabar River. The processing of this particular fish before getting to the final consumer must be given proper attention. Eating of raw fish should be avoided, because cooking the fish for a short time would be sufficient to kill larvae in the intestine (John, 1992).

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