Studies on Microbial and Helminths Associated with *Parachanna obscura* in Eleyele Reservoir and Ajilete-Yewa River, South-Western, Nigeria

Adeleke, M.T.[1]*, O.N. Adekunle[1], T.O. Adesetan[2], R.Y. Oladunjoye[1], H.O. Mogaji[3], F.M. Oyeyipo[2] and O.A. Lawal[1]

[1]Department of Zoology and Environmental Biology, Olabisi Onabanjo University, Ago-Iwoye, Ogun State, Nigeria.
[2]Department of Microbiology, Olabisi Onabanjo University, Ago-Iwoye, Ogun State, Nigeria.
[3]Department of Animal and Environmental Biology, Federal University Oye-Ekiti, P.M.B.373, Ekiti State, Nigeria

*Corresponding author e-mail: *adeleke.mistura@oouagoiwoye.edu.ng;* Phone: *+2348076518030*

Abstract

This research aimed to increase our understanding of the microbial and parasitic composition of Parachanna obscura (African Snakehead) which is a commercially important freshwater fish species in Nigeria. The study was conducted in two locations, the Eleyele Reservoir in Oyo State, and the Ajilete-Yewa River in Ogun State, both in the southwestern, Nigeria. A total of 130 healthy P. obscura was collected from June to December, 2022 and examined for microbial and parasitic presence using standard methods. The prevalence of parasites was calculated, and the results were analyzed using one sample T test with $p \leq 0.05$. Out of the 130 fishes examined, 94 (74.31%) were found to have parasites. The prevalence of parasites was higher in the Ajilete-Yewa River (75.9%) than in the Eleyele Reservoir (69.7%). Seven types of parasites were identified, including Procamallanus, Camallanus, P. africanus, C. metacercaria, Genarchopsis, Pallisentis and Trichodina. The female P. obscura had a higher parasite intensity of 50.36% than males (49.63%). Additionally, bacterial analysis conducted, reveal the presence of Bacillus spp, Escherichia coli (13.79 x 104CFU/g), Staphylococcus aureus (9.59 x 104CFU/g), Listeria monocytogenes (7.46 x 105CFU/g), and Salmonella spp(21.10 x 105CFU/g),. The findings highlight the need for public education on the potential dangers associated with consuming P. obscura and imply that the fish may contain variable degrees of parasitic and bacterial infections, some of which may be higher than the allowable amount.

Keywords: *Escherichia coli*; fish helminth; *Listeria monocytogenes*; Nigeria; *Parachanna obscura*.

Introduction

Fish is an important source of protein and plays a significant part in medicine since it replaces the body's supply of vitamins A and D, amino acids, calcium, lysine, phosphorus, and sulphur (Ohene-Adjei *et al.,* 2007). Most of tropical Africa, the Middle East, and Asia are home to snakeheads of the family Channidae. They can be found in a wide range of freshwater settings, including hills, streams, and swamps (Ajah *et al.,* 2020).

Presently, the family Channidae consists of twenty-six genuine species, with three species within the genus Parachanna from Africa and 23 species from South-east Asia (Adamson & Britz, 2018; Praveenraj *et al.,* 2018). The African Snakehead *Parachanna obscura* an Osteichthyes belongs to the Channidae Family. The anal, caudal, and dorsal fins of this species are iridescent green or blue with an orange edge, while the body colour ranges from grey to greyish brown. The ventral region is white or dirty white. Two huge cycloid scales are located on either side of the lower jaw, and the pectoral fins have different numbers of dark and light semi-circular bands (Vishwanath & Geetakumari, 2009). They are known to be carnivorous predators in nature and the younger ones are guided by a large adult (Gosse, 1963).

Because *P. obscura* provides so many

advantages, it is imperative that it be used for commercial purposes to reap the substantial financial rewards. Fishery resources have decreased because of ecological changes brought on by prolonged wet seasons and overexploitation by people, together with declining flows within the studied areas. Also, helminthic infections of fishes are known to cause several degrees of fish harm, from reproductive issues brought on by Eustrongylides to lamellar destruction by monogenetic trematodes. which in turn endangers the human consumer (Ogawa, 2015). Additionally, helminths can serve as metal sinks for the fish host (Bamidele & Kuton, 2016).

Due to the emphasis on pisciculture and growing knowledge of the contamination of natural freshwater resources in the tropics, accumulation of chemicals and microbial load on fish has taken greater significance. Such research is typically employed as a sensitive and effective indicator to track physiologic and genetic changes in fish. This was reflected by Akinsanya *et al.* (2020) who assessed the bioaccumulation of Organochlorine Pesticides and the associated parasitological and microbial susceptibility in *P. obscura* to assess the potential environmental effects of the chemical in a Lagos Lagoon Nigeria.

Nigeria is blessed with abundance of natural aquatic resources in marine, estuarine and freshwater environments, with over 270 fish species (Adewumi *et al.,* 2017). Croaker, catfish, tilapia, threadfin, and clupeids are a few of the most significant species that make up 90% of Nigeria's fishery (Federal Department of Fisheries, 2008). In Nigeria, studies on *P. obscura* so far include the biology of the species reported by Odo *et al.* (2012) and Olasunkanmi and Ipinmoroti, (2014).

The haematological profile of the species from several rivers in Nigeria was studied by Ajah *et al.* (2020). Olutimehin *et al.* (2019) and Osho *et al.* (2020) have provided information on the microbes associated with *P. obscura* from Eleyele Dam and Ogun River. *Procamallanus* spp., *Contracaecum* sp., *Clinostomum metacercariae*, *Cucullamus* sp., *Camallanus* sp., *Wenyonia* sp. and *Spirocamallanus* sp. are parasites that have been associated with *P. obscura* (Bamidele & Kuton, 2016; Osho, 2017; Adegbehingbe & Umezurike, 2018). This study aims to expand our knowledge of the parasites and microbes associated with *P. obscura* in the study areas. It also addresses the lack of research on how these biological factors impact *P. obscura* populations in Nigeria.

Materials and methods

Description of study sites

This was a cross-sectional study conducted in two study sites: Eleyele Reservoir (Oyo State) and Ajilete -Yewa River (Ogun State) all in South-western Nigeria. Eleyele Reservoir lies between the latitude of 7°25’25N and longitude of 3°52’22E, is situated in Ibadan North-West Local Government Area. The area is surrounded by Eleyele neighbourhood to the south, Apete to the east, and Awotan to the north. Ajilete-Yewa River is a ward in Ogun State, Nigeria's Yewa South Local Government Area, which borders the Republic of Benin. The Yewa River, located at latitude 6°44’57N and longitude 3°12’52E, is a transboundary river between Nigeria and the Republic of Benin. It flows along the coast of Benin and crosses the border into Nigeria at one point. Ajilete has diversity of activities which includes fishing, selling of smoked fish, laundry, welding and farming of crops.

Collection of water samples

Water samples were collected for the physiochemical parameters of temperature, pH, dissolved oxygen (DO), total dissolved solids (TDS), and transparency for six months in the morning between 6:00am and 10:00am at both sampling locations using APHA (2005) standard protocols from June to December, 2022.

Collection of P. obscura samples

A total of 76 and 54 live fishes were acquired from fisherfolk within the period of 6:00am to 10:00am at Eleyele Reservoir and Ajilete-Yewa River correspondingly, spanning from the month of June to December in the year 2022. All the fishes appeared healthy and were transported to the Zoology Laboratory of Olabisi Onabanjo University in transparent containers filled with 25 litres of water to keep them alive for further laboratory analysis.

Sex determination, Length and Weight measurements

Using a digital weighing balance (VIBRA SJ), the weight of each sample of fish was determined and tagged with paper tape identifying their location and number. Using a calibrated meter rule to the nearest centimetre (cm), the total length of each fish was measured from the tip of the snout to the extreme end of the caudal fin. According to Lagrue *et al.* (2011), the urogenital papillae of each fish which are behind the anus and are long or enlarged in males while being round and reddish in mature females were examined to ascertain its sex. Afterwards, this was verified through dissection by examining the testicles in males and the ovaries in females.

Parasitological identification

The abdominal cavity of each fish sample was dissected from the anal opening to the lower jaw, and the gastrointestinal part (gut, stomach, and intestines) was removed and washed in the appropriate labelled petri dishes containing physiological saline solution for sedimentation and floatation as described by Akinsanya *et al.* (2020).

Observation of physical nematodes was done under a microscope (XSZ-07) with the aid of a slide and identified to specie level using the key Systema Helminthum by Hoffman (1999); Ajala and Fawole (2014), Kawe *et al.* (2016). Parasite inside each sample were counted and noted. Using the formula below, the prevalence and intensity were computed:

$$\text{Prevalence}(\%) = \frac{Number\ of\ fish\ infected}{Number\ of\ fish\ examined} \times 100$$

$$\text{Intensity} = \frac{Number\ of\ parasite}{Number\ of\ fish\ infected}$$

Bacteriological Analysis

A part of the Gills and intestine from each fish were weighed individually at one gram each in sterile Petri dishes and homogenized with mortar and pestle aseptically. In order to generate a 10^{-1} dilution, the fish tissues (skin and intestine) were carefully inserted into two distinct test tubes, each containing 9 ml of sterile distilled water.

According to Willey *et al.* (2008), Serial dilutions of the stock solutions up to 10^{-3} was carried out. Pour plate method was employed; sterilized Nutrient agar was aseptically poured on 1 ml inoculum in a petri dish, swirled gently, allowed to set, followed by a 24-hour incubation period at 37ºC. To determine the total viable count (TVC) in CFU/g., colonies were enumerated and multiplied by the dilution factor. To create pure culture, distinct colonies were subcultured and kept in the refrigerator in a McCarthney bottles for further analysis.

For characterization of the bacterial isolates, the procedures for Gram staining and biochemical assays were as outlined by Chessbrough (2006).

Statistical analysis

Parasite prevalence was calculated using Excel 2020 spreadsheet. The significant difference at 5% between infection levels in the two environments was determined using one sample T test of Statistical Package for the Social Science (SPSS version 25).

Results

Physicochemical Parameters of Eleyele Reservoir and Ajilete-Yewa River

The physicochemical parameter: temperature, Dissolved Oxygen (D.O), transparency and Total Dissolved Solids (TDS) for Eleyele Reservoir and Ajilete-Yewa river ranged between 24 - 27.1ºC, 4.4 – 4.8, 39 – 48.4 cm, 0.37 - 0.42 and 22.6 - 30 ºC, 3.9 - 4.46, 44-58.7 cm and 0.47 - 0.63. respectively. This shows that the two environments are productive water bodies that supports for growth of *Parachanna obscura*.

Sex determination, Length and Weight measurements

Eleyele Reservoir had the highest population of fish samples, with males (59.2%) dominating the sample while Ajilete-Yewa had higher percentage of females (48.1%) per total sample from the river. In both study sites, there are more males (56.2%) than females (43.8%) (Table 1). The results revealed minimum and maximum total length of *P. obscura* as 18.4 cm, 49.7 cm and 16.2 cm, 44 cm from Eleyele Reservoir and Ajilete-Yewa river respectively with fishes from Ajilete-Yewa river having more

Table 1: Sex and Length ratio of *P. obscura* in Eleyele and Yewa Rivers

SEX	RIVER		TOTAL	Length (cm)	River A		River B	
	A	B			No of fish	Mean	No of fish	Mean
MALE	45(59.2%)	28(51.9%)	73(56.2%)	11-20	6	17.75	2	17.45
				21-30	43	25.28	46	24.94
FEMALE	31(40.8%)	26(48.1%)	57(43.8%)	31-40	21	34.8	5	32.88
				41-50	6	45.07	1	44
TOTAL	**76**	**54**	**130**	**TOTAL**	**76**		**54**	

fishes that are between the range of 11 – 30 cm. Whereas the fish body weight varies between 70 - 600 g with the mean as 225 g and 247 g in Eleyele Reservoir and Ajilete-Yewa river respectively (Fig. 1).

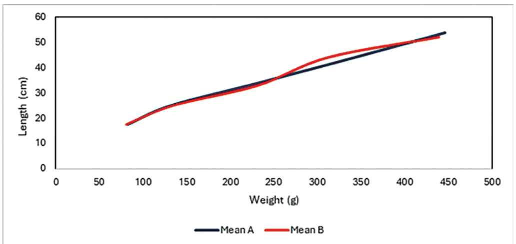

Figure 1: Mean length-weight distribution of *P. obscura* in the rivers

Prevalence and intensity of parasites in the study environments and in relation to sex of *P. obscura*

The outcome shown in Table 2 revealed that a total of 130 samples of *P. obscura* were examined from the two study sites. Out of the 130 fish examined, 94 (74.31 %) carried various parasites, with Ajilete-Yewa river having the highest incidence (75.9 %) than in Eleyele Reservoir despite the lower quantity of fish caught. The parasitic load on infected *P. obscura* was 693 with 69.7% of the parasites found in Eleyele Reservoir which is lower than Ajilete-Yewa river with 75.9%. However, the intensity was more in Eleyele Reservoir (8.28) than in Ajilete-Yewa river (6.2).

Moreso, the prevalence of parasites in both males and females of *P. obscura* found in both study sites showed higher prevalence in females (80.70 %) in both waterbodies than in males (65.75 %). The intensity was highest in females from Eleyele Reservoir (8.92) and lowest in females from Ajilete-Yewa river (6.14). However, there was no significance at ($p < 0.05$) in prevalent rates between the sexes.

Taxonomic Classification of Parasites

The total 693 parasites found in this study were classified into 7 species and four taxa namely Nematoda, Trematoda, Acanthocephalan and Protozoa (Table 3). The parasites include three (3) Nematoda (*Procamallanus, Camallanus, Philometroides africanus*), two (2) Trematoda (*Clinostomum metacercaria*, *Genarchopsis*), one (1) *Acanthocephala (Pallisentis)*, and one (1) Protozoa (*Trichodina* spp). Of all the species, *Procamallanus* had the highest occurrence in both Eleyele Reservoir and Ajilete-Yewa river

Table 2: Prevalence of parasites in relation to the sex of *P. obscura* in Eleyele and Yewa Rivers

RIVER	Sex	No of Fishes	No of Infected fishes	% Prevalence	Parasite Count	Intensity
A	MALE	45	29	64.44	225	7.76
	FEMALE	31	24	77.42	214	8.92
	SUB TOTAL	76	53	69.74	439	8.28
B	MALE	28	19	67.86	119	6.26
	FEMALE	26	22	84.62	135	6.14
	SUB TOTAL	54	41	75.93	254	6.2
GROUPED	MALE	73	48	65.75		
	FEMALE	57	46	80.70		
	TOTAL	**130**	**94**	**72.31**	**693**	**7.37**

Table 3: Parasite occurrence in *P. obscura* in Eleyele and Yewa Rivers

TAXA	PARASITE	RIVER A			RIVER B		
		No of Infected Fish	Species Prevalence (%)	Parasite Recovered	No of Infected Fish	Species Prevalence (%)	Parasite Recovered
Nematoda	*Procamallanus*	15	28.00	104	14	34.00	75
	Camallanus	10	19.00	67	9	22.00	63
	P. africanus	3	6.00	52	2	5.00	22
Trematode	*C. metacercaria*	9	17.00	55	4	10.00	17
	Genarchopsis	3	6.00	48	1	2.00	26
Acanthocephala	*Pallisentis*	9	17.00	56	10	24.00	37
Protozoa	*Trichodina*	4	8.00	57	1	2.00	14

(104, 28% and 75, 34%) respectively. While the Genarchopsis and *Trichodina* had the least from Ajilete-Yewa river (2%) with just 1 fish being infected each, although the parasite counted were 14 and 22 respectively (Table 3). The *Camallanus* also had high representative in both waters with counts of 67 (19 %) and 63 (22 %) from Eleyele Reservoir and Ajilete-Yewa river respectively.

Parasite Infection to Bodyweight

The result revealed that fishes within the range of 101-200 g and 201-300 g had the highest prevalence of 83.87 % and 83.33 % in Eleyele Reservoir and Ajilete-Yewa river respectively, while fishes between 301-400 g and 401-500 g had 100% and 60% prevalence respectively in Ajilete-Yewa river. It was observed that there are no fishes weighing between 501-600 g in Ajilete-Yewa river (Table 4).

Distribution in relation to site of infection

The distribution in relations to site of infections is presented in Table 5. It was generally observed that the intestine was more infested with parasites (n=424) than the stomach (n=269) from both waterbodies. Of the 7 identified species, *P. africanus* had the least presence in the intestine (n=24) while *C. metacercaria* had the least in the stomach (n=26).

Microbial Load in fishes from both rivers

From the results obtained, the total viable count of bacterial isolates from the stomach and intestine of the sampled fishes are presented in Table 6. The range of the total viable count from the two sites analysed were between 2.10 x 104 - 11.69 x 104 $CFUg^{-1}$ from the skin and 7.2 x 105 – 13.9 x 105 $CFUg^{-1}$ from the intestine in Eleyele Reservoir while it is 1.7 x 104 - 8.2 x 104 $CFUg^{-1}$ and 3.46 x 105 – 9.2 x 105 $CFUg^{-1}$ from the skin and intestine for fishes in Ajilete-Yewa river respectively. The mean count computed for each fish part revealed that the skin of fishes in Ajilete-Yewa river had the least count of 5.8 x 104 $CFUg^{-1}$ and intestines of fishes in Eleyele Reservoir had the greatest number of 13.9 x 105

Table 4: Prevalence of parasites in relation to the weight of *P. obscura* in Eleyele and Yewa Rivers

Weight (g)	RIVER A			RIVER B		
	No of fish Examined	No Infected	Prevalence (%)	No of fish Examined	No Infected	Prevalence (%)
1-100	18	11	61.11	8	6	75
101-200	31	26	83.87	38	28	73.68
201-300	11	7	63.64	6	5	83.33
301-400	5	3	60	1	1	100
401-500	7	4	57.14	1	1	100
501-600	4	2	50	0	0	0
TOTAL	76	53	69.74	54	41	75.93

Table 5: Distribution of parasites in relation to the site of infection on infected *P. obscura* in Eleyele and Yewa Rivers

Taxa	Parasite	No of Infected Fish	Stomach	Intestine	No of Parasite Recovered	Prevalence (%)	Intensity
Nematoda	*Procamallanus*	29	58	121	179	22.31	6.17
	Camallanus	19	43	87	130	14.62	6.80
	P. africanus	5	50	24	74	3.85	14.80
Trematode	*C. metacercaria*	13	26	46	72	10.00	5.54
	Genarchopsis	4	28	46	74	3.08	18.50
Acanthocephala	*Pallisentis*	19	36	57	93	14.62	4.90
Protozoa	*Trichodina*	5	28	43	71	3.85	14.20
TOTAL	**94**	**269**	**424**	**693**	**72.31**	**7.37**	

CFUg^{-1}.

Six (6) bacteria isolates were obtained from *P. obscura* from the two waterbodies. These includes 2 Gram-negative rod bacteria (*Escherichia coli* and *Salmonella* sp.), 1 Gram-positive rod bacteria (*Bacillus* sp.) and 3 Gram-positive cocci bacteria (*Listeria monocytogenes, Staphylococcus aureus* and *Streptococcus* sp). Species of bacteria that have been isolated from *P. obscura* from both sites revealed that all observed bacteria were present in Eleyele Reservoir except *E. coli* while *Salmonella* and *Bacillus* were not observed in Ajilete-Yewa river (Table 6).

Discussion

The dark brown African Snakehead fish *P. obscura* from two different waterbodies were studied for growth, parasite, and microbial investigations. It was revealed that the physicochemical parameters in these waterbodies were favourable for growth and the distribution of length and weight showed a positive connection amongst the fishes from both rivers. This outcome was consistent with previous research using *P. obscura* and other fish species from several water basins in southwest Nigeria (Adeleke *et al.,* 2019; Osho & Usman, 2019).

For the period of study, the Male populations outnumbered female populations in both waterbodies (1.4♂: 1♀). This is in line with research on *P. obscura* by Komolafe and Arawomo (2011), Bamidele and Kurton (2016),

Table 6: Total bacterial loads and Biochemical Characterization of Isolates from *P. obscura* in Eleyele and Yewa Rivers

Total bacterial loads			Occurrence							KIA							Probable organism
	River A	River B	River A	River B	Gram stain/ Shape	Catalase	Citrate	Oxidase	Indole	Urease	H2S	Gas	Lactose	Mannitol	Glucose	Sucrose	
TCs CFU/ gx 10^4	13.79	9.9	-	+	- R	+	-	-	+	-	-	+	+	+	+	-	*Escherichia coli*
RAs (CFU/g x10^4)	9.59	6.5	+	+	+ C	+	+	-	-	+	-	-	+	+	+	+	*Staphylococcus aureus*
M±SD (CFU/gx 10^4 (skin)	6.40±0.20	5.8±0.47	+	-	+ R	+	+	-	-	-	-	-	-	-	+	+	*Bacillus* sp.
TCi (CFU/gx 10^5)	21.1	13.66	+	-	- R	+	-	-	-	-	+	+	-	+	+	-	*Salmonella* sp.`
RAi (CFU/g x10^5)	6.7	5.74	+	+	+ C	-	-	-	-	-	-	-	+	+	+	+	*Streptococcus* sp.
M± SD (CFU/g x10^5 (intestine)	5.8± 0.47	7.46±1.59	+	+	+ C	+	+	-	-	-	-	-	+	-	+	+	*Listeria* sp.

TCs - Total count in Skin; RAs – range in Skin; TCi- Total count in Intestine; RAi - range in Intestine; R=Rod; C=Cocci; + = Positive; - = Negative; KIA= Klingler's Iron Agar and Osho and Usman (2019).

This study finding is in contrasts with the results from Adeleke *et al.* (2019) and Kareem *et al.* (2019), which found a predominance of female *P. obscura* in two lagoons in Lagos and Eleyele Reservoir respectively.

The results are consistent with Osho *et al.,* (2014) whose findings indicates that males of many tropical species develop more quickly and have larger standard sizes than females, perhaps as a result of the anabolism-enhancing androgens even if the b values for the two sexes were not statistically different.

According to Garg *et al.* (2009) various tests have been conducted on fishes to demonstrate their reliability as markers of contamination in the aquatic environment. It has been demonstrated that biotic and abiotic changes in the environment influence fish parasites (Dzika and Wylic, 2009). The results of this study showed a higher prevalence of parasites (75.93%) in Ajilete-Yewa river fishes, which may be caused by domestic activity-related pollution, the presence of intermediate hosts that harbour infective larval stages and the size of the water body (Kawe *et al.,* 2016; Osho, 2017). Meanwhile, the lower incidence found in Eleyele Reservoir may be related to controlled household activities and it might harbour some of these parasites and disperse parasite eggs throughout the water body (Aliyu & Solomon, 2012).

The significant incidence of *P. obscura* fish parasites observed in this study is consistent with findings from Bamidele and Kuton (2016); and Osho (2017). According to Kawe *et al.* (2016) and Afolabi *et al.* (2020a), there was a high frequency of gastrointestinal helminths parasites of 67.5 and 75%, respectively in *C. gariepinus* in Nigerian waters.

According to this study, females were somewhat more likely to have parasites than males in both waterbodies; this could be explained by the physiological differences between the sexes such as hormonal fluctuations or differences in immune response. However, there were no appreciable sex-specific differences in infection rates ($p>0.05$). The intensity in Ajilete-Yewa river was lowest in the female fish, indicating that the sex of the fish had no bearing on the rate of infection. This study is in line with other research on catfish by Bamidele and Kuton (2016) and Ogonna *et al.* (2017), which reported higher parasite helminth levels in female *P. obscura* from Ogun River. These authors noted that females were more likely to have parasitic illnesses than males. However, female *P. obscura* samples from the lower Cross River system and Lekki Lagoon, Lagos, showed much lower parasite burdens than the male samples as reported by Akinsanya and Otubanjo (2006); and Oden *et al.* (2015).

As observed by Omeji *et al.* (2013), female fish require more food to meet their dietary needs for egg production, which may have exposed the female fishes to more parasite interaction and consequently raised their risk of infection. In general, fishes in Eleyele Reservoir had more parasites than those found in Ajilete-Yewa river. This might be caused by a lot of domestic activity and the existence of an intermediate host in Eleyele Reservoir rather Ajilete-Yewa river. However, according to Ogonna *et al.* (2017) and Osho (2017), the primary cause of the changes in parasite burden with sex is physiological rather than sexes, it is connected to their struggle for survival and differential eating on food amount or quality.

Nematodes and protozoans were the most and least common types of helminth parasites found in the current study respectively. The Procamallanus species and Camallanus spp. were the two parasite species that were most common in the fish samples. The results of Oden *et al.* (2015), Osho (2017) and Adegbehingbe and Umezurike (2018) in *P. obscura* are consistent with this. According to Onwuliri and Mgbemena (1987), the availability of an appropriate intermediate host could explain why fish in Ajilete-Yewa river had more nematodes and acanthocephalans while fish in Eleyele reservoir had more trematodes and protozoans.

Previous research by Eyo and Iyaji (2014) and Okoye *et al.* (2014) has documented the presence of these parasites in various Nigerian freshwater fish species. They suggested that the high cestode parasite infection of *C. gariepinus* may be caused by eating copepods, molluscs, and eggs, which serve as intermediary hosts for the cestode parasites' larva.

In both waterbodies, the prevalence of parasite infection in *P. obscura* fishes was insignificant. However, the results showed that medium-sized fish were more heavily infested than very large fish. This might be due to the fact that they were more active and required more food to support their development, which exposed them to more parasite infestation. This result is consistent with Osho (2017), who found that the size of the fish had an impact on the quantity of parasites for most of the organisms. In contrary, Oden *et al.* (2015) discovered that fish size has no effect on nematode infection in the *P. obscura* population from the lower Cross River system of Nigeria.

The results of this study concurred with those of Omeji *et al.* (2013), Abdel-Gaber *et al.* (2015), Ani *et al.* (2017) and Afolabi *et al.* (2020a) who found that larger catfish had higher percentages of parasitic infection, indicating that parasitism increases with size. In the work of Bichi and Dawaki (2010), the prevalence was shown to increase with fish size, which could be explained by the prolonged exposure to the environment due to body size. Furthermore, the ratio of parasite infection to sex was about the same in the two waterbodies studied, with the percentage change being largely related to chance and quantity.

Other researchers have previously documented the parasites collected from the colon and stomach of *P. obscura* employed in this work (Eyo *et al.,* 2014; Oden *et al.,* 2015; Uruku &Adikwu, 2017). The intestine contained the highest number of nematodes overall, indicating that the parasites prefer this area to other potential attachment sites. This preference may be due to the food supply in the intestine. The results of this study are consistent with those of Ibiwoye *et al.* (2004); and Oden *et al.* (2015), who stated that worms favour areas of the fish alimentary canal where partially digested meals are present and where easy habitation attachment to the absorbent gut is possible.

In comparison to Ajilete-Yewa river, Eleyele Reservoir had considerably higher bacterial counts (TBC). This can be due to the Reservoir's sweltering temperature, which is near optimal for many mesophilic bacteria, the bacteria's improved microbial environment adaptation and capacity to form symbiotic relationships within fish digestive tracts (Shinkafi & Ukwaja, 2010; Jimoh *et al.,* 2014). The result of bacteria population (10^4 - 10^5 $CFUg^{-1}$) from this study conforms to the finding of Osho *et al.* (2020) on young and adult *P. obscura* in Ogun River as well as Afolabi *et al.* (2020b) who recorded counts of 10^4 $CFUg^{-1}$ for *C. gariepinus* in a natural and artificial habitat study. Additionally, Osungbemiro *et al.* (2014) reported the same microbial load on *C. gariepinus* in Ondo State's fresh and brackish water habitats. Musefiu *et al.* (2011) noted a decreased bacterial population in Ibadan, nevertheless. Eze *et al.* (2011) state that no fish should be consumed by humans if it contains more than 10^6 $CFUg^{-1}$ bacteria per gram but the bacterial load recorded from the two waterbodies in this investigation was less than 10^6 $CFUg^{-1}$.

Additionally, it was discovered that the bacteria load in the intestines was substantially higher ($P \leq 0.05$) than in the skin. This difference could be explained by the fact that food only passes through the gills, whereas the gut contains both pathogenic and non-pathogenic organisms, making the intestine more habitable than the skin (Musefiu, *et al.,* 2011). The sediment, water, and food that fish consume contain bacteria, fungus, and other microorganisms that they pick up from their surroundings to put in their digestive tracts (Saha *et al.,* 2006). This result is congruent with research by Olugbojo and Ayoola (2015 and Osho *et al.* (2020) which found that the guts of adult *P. obscura* and three other species of fish collected in Lagos State had larger bacterial populations than their gills and skin. Similar to this, elevated bacterial loads in the colon were also found by Ajani *et al.* (2016) and Albert *et al.* (2016). However, it was at odds with research by Afolabi *et al.* (2020b), who found that fresh Tilapia fish (*Oreochromis niloticus*) and catfish skin had greater bacterial loads than the intestines and gills, respectively.

Staphylococcus aureus, *Listeria monocytogenes* and *Streptococcus* sp. were the most frequently discovered bacteria on *P. obscura*'s skin and stomach from the two water environments while *E. coli*, *Salmonella* sp and *Bacillus* sp were the least common.

The presence of *Staphylococcus* sp is an indication of human contamination during the fish harvesting and handling process which was seen in the two water bodies. This observation is consistent with the findings of Eze *et al.* (2011); and Afolabi *et al.* (2020b). It is very helpful to isolate enteric organisms like *E. coli* as a marker for faecal pollution (Rodrigues and Cunha, 2017), and our analysis revealed that Ajilete-Yewa river contains *E. coli* that could result from faecal pollution in the water body brought on by humans and other environmental waste. This is comparable to the observation made by Osungbemiro *et al.* (2014) and Afolabi *et al.* (2020b). Because of the bacteria present, consumers' health who rely on freshwater fish as a source of protein are at risk although being in an acceptable range. Humans have been known to contract food-borne bacterial illnesses from fish and fish products (Efuntoye *et al.,* 2012).

Conclusion

The study revealed that both water bodies examined were contaminated with parasites, with Nematoda, Trematoda, Acanthocephalans and Protozoan species identified. The bacterial populations were composed of both Gram-positive and Gram-negative strains, and the pollution levels were higher in Eleyele Reservoir compared to Ajilete-Yewa river. The infection rate was higher in medium-sized fish than in smaller ones, and the intestine had a higher concentration of bacteria than the skin. While bacterial counts in fish samples were within permissible limits, the presence of pathogenic bacteria poses a risk to public health.

Fish farmers and consumers in this study areas need education on bacterial and parasitic infection risks and proper cooking methods. Human encroachment into fish habitats must be avoided to prevent contamination.

Further research is required to genetically characterize the gastrointestinal helminths and investigate the intermediate hosts for the parasitic helminths. It is crucial to examine the correlation between pollution, bioaccumulation, and the frequency of helminth parasites in fish from these water bodies. Additionally, the antibiotic sensitivity of bacteria isolated from fish samples should also be examined.

Acknowledgments

The authors acknowledged the contribution of the fishermen in Eleyele and Yewa Rivers in capturing the snakeheads used for this research. The authors are also grateful to the Management of Olabisi Onabanjo University and the Department of Zoology and Environmental Biology for the opportunities given to use their facilities for this study.

Conflicts of Interest

The authors declare that they have no conflict of interest in publication of this work because this research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

References

Abdel-Gaber, R., El Garhy, M., and Morsy, K. (2015). Prevalence and intensity of helminth parasites of African catfish *Clarias gariepinus* in Lake Manzala, Egypt. *Advances in Bioscience and Biotechnology,* 6(07): 464 – 469.

Adamson, E.A. and Britz, R. (2018). The snakehead fish *Channa aurolineatus* a valid species (*Teleostei: Channidae*) distinct from *Channa marulius*. *Zootaxa*, 4514(4): 542- 552.

Adegbehingbe K.O., and Umezurike, E. (2018). Gastrointestinal Helminth Parasites of the *Parachanna obscura* from Epe Lagoon, Lagos, Nigeria. *Open Access Journal of Public Health* 2(1):1-3.

Adeleke, M.T., Adekunle, O.N., Lawal, O.A., and Adeyemi, F.M. (2019). Length-Weight Relationship and condition factor of African snakehead *Parachanna africana* (Steindachner 1879) from Ijede and Agbowa Lagoons, Lagos State, Nigeria. *African Journal of Science and Nature* 9: 35-44.

Adewumi, A.A., Idowu, E.O., Edward, J.B., Oso, J.A., and Fadiya, O.O. (2017). The Ichthyofauna of Elemi River, Nigeria, A Preliminary Pre-Impoundment Survey. *FUW Trends in Science and Technology Journal,* 2(18): 506 – 509.

Afolabi, O.J., Oladele, O.O., and Olususi, F.C. (2020b). Assessment of bacterial loads of Clarias gariepinus (Burchell, 1822) obtained from cultured and natural habitats. *The Journal of Basic and Applied Zoology,* 81(1): 1-7.

Afolabi, O.J., Olususi, F.C., and Odeyemi, O.O. (2020a). Comparative study of African catfish parasites from cultured and natural habitats. *Bulletin of the National Research Centre,* 44(1): 1-9.

Ajah, P.O., Ita, E.O.B., and Allison, N.L. (2020). Characterization of Nematode Infestation on *Parachanna obscura* (Gunther, 1861) (Channidae) and Infection in the Blood. Aquaculture Studies, 20(1): 55-64.

Ajala, O.O., and Fawole, O.O. (2014). Multiple infections of helminths in the alimentary system of *Clarias gariepinus* (Burchell, 1822) in a tropical reservoir. *International Journal of Fisheries and Aquaculture,* 6(6): 62-70.

Ajani, E.K., Orisasona, O., and Omitoyin, B.O. (2016). Comparison of bacterial flora and frequency of occurrence in water and *Clarias gariepinus* raised in ponds fertilized with raw poultry manure. *Journal of Fisheries Sciences,* 10(1): 016– 021.

Akinsanya, B., and Otubanjo, O.A. (2006). Helminth Parasites of *Clarias gariepinus* (Clariidae) in Lekki Lagoon, Lagos, Nigeria. Revista de biología tropical, 54(1): 93-99.

Akinsanya, B., Olaleru, F., Samuel, O.B., Akeredolu, E., Isibor, P.O., Adeniran, O.S., Saliu, J.K. and Akhiromen, D.I. (2020). Bioaccumulation of organochlorine pesticides, *Procamallanus* sp. (Baylis, 1923) infections, and microbial colonization in African snakehead fish sampled from Lekki Lagoon, Lagos, Nigeria. *Brazilian Journal of Biology*, 81: 1095-1105.

Albert, A.C., Felix, E.C., Solomon, O.E.,and Emeh, C.C. (2016). Effect of habitat locations on the bacteriological and physicochemical assessment of aquaculture freshwater catfish (*Clarias gariepinus*) using small scale depuration system. *Journal of Advances in Microbiology,* 1(1): 1-9.

Aliyu, M.D., and Solomon, J.R. (2012). The intestinal parasite of *Clarias gariepinus* found at lower Usman Dam, Abuja. Researcher, 4(9): 38-44.

American Public Health Association (APHA), (2005). Standard methods for the examination of water and waste analysis (21st ed.). American water works association/water environment Federation, Washington D.C.1288pp.

Ani, O.C., Nnamonu, E.I., and Ejiogu, C. (2017). Prevalence of intestinal parasites of fish farmed and harvested in Abakiliki, Nigeria: A pointer to the level of their vulnerability. *International Journal of Research in Pharmacy and Biosciences,* 4(9): 7-10.

Bamidele, A., and Kuton, M.P. (2016). Parasitic diseases and heavy metal analysis in *Parachanna obscura* (Gunther 1861) and *Clarias gariepinus* (Burchell 1901) from Epe Lagoon, Lagos, Nigeria. Asian Pacific *Journal of Tropical Disease,* 6(9): 685-690.

Bichi, A., and Dawaki, S. (2010). A survey of ectoparasites on the gills, skin and fins of Oreochromis niloticus at Bagauda fish farm, Kano, Nigeria. *Bayero Journal of Pure and Applied Sciences*, 3(1): 83–86.

Cheesbrough, M. (2006). District laboratory practice in tropical countries, part 2. Cambridge University Press, 434pp.

Dzika, E., and Wyzlic, I. (2009). Fish parasites as quality indicators of aquatic environment. *Zoologica Poloniae*, 54(1-4): 59-65.

Efuntoye, M.O., Olurin, K.B., and Jegede, G.C. (2012). Bacterial flora from healthy *Clarias gariepinus* and their antimicrobial resistance pattern. *Advance Journal of food science and technology,* 4(3): 121-125.

Eyo, J.E., and Iyaji, F.O. (2014). Parasites of *Clarotes laticeps* (Ruppell, 1832, Siluriformes, Bagridae) at Rivers Niger-Benue Confluence, Lokoja, Nigeria. *Journal of Fisheries and Aquatic Science,* 9(3): 125-133.

Eyo, V.O., Ekanem, A.P., and Jimmy, U.I.U. (2014). A comparative study of the gonado-somatic index (GSI) and gonad gross morphology of African catfish (*Clarias gariepinus*) fed unical aqua feed and coppens commercial feed. *Croatian Journal of Fisheries*, Ribarstvo, 72(2): 63-69.

Eze, E.I., Echezona, B.C., and Uzodinma, E.C. (2011). Isolation and identification of pathogenic bacteria associated with frozen mackerel fish (*Scomber scombrus*) in a humid tropical environment. *African Journal of Agricultural Research,* 6(8): 1947–1951.

Federal Department of Fisheries, (2008). Fisheries Statistics of Nigeria, 4th ed. Federal Department of Fisheries, Abuja.49pp.

Garg, S., Gupta, R.K., and Jain, K.L. (2009). Sublethal effects of heavy metals on biochemical composition and their recovery in Indian major carps. *Journal of Hazardous Materials*, 163(2-3): 1369-1384.

Gosse, J.P. (1963). Le milieu aquatique et l'écologie des poissons dans la région de Yangambi. Musée royal de l'Afrique centrale 116:113–249.

Hoffman, G.L. (1999). Parasites of North American freshwater fishes. Cornell University Press, Poland.527pp.

Ibiwoye, T.I.I., Balogun, A. M., Ogunsusi, R.A., and Agbontale, J.J. (2004). Determination of the infection densities of mudfish Eustrongylides in *Clarias gariepinus* and *C. anguillaris* from Bida floodplain of Nigeria. *Journal of Applied Sciences and Environmental Management*, 8(2): 39-44.

Jimoh, W.A., Oladele-Bukola, M.O., Adebayo, M.D., Yusuff, A.A., Azeez, F.A. and Salami, O.O. (2014). Microbial flora of the gastro-intestinal tract of *Clarias gariepinus* caught from river Dandaru Ibadan, Nigeria. *Sokoto Journal of Veterinary Sciences*, 12(2): 19-24.

Kareem, O.K., Olanrewaju, A.N., and Orisasona, O. (2019). Aspect of reproductive biology of *Parachanna obscura*, Gunther 1861 in a southwestern Nigerian reservoir. *International Journal of Environmental Impacts*, 2(3): 272-282.

Kawe, S.M., God'spower, R.O., Balarabe, M.R., and Akaniru, R.I. (2016). Prevalence of gastrointestinal helminth parasites of *Clarias gariepinus* in Abuja, Nigeria. *Sokoto Journal of Veterinary Sciences*, 14(2): 26-33.

Komolafe, O.O., and Arawomo, G.A.O. (2011). Observations on the composition, physiological condition and fisheries in Erinle Lake, Osun State, Nigeria. *West African Journal of Applied Ecology,* 18: 71-78.

Lagrue, C., Kelly, D.W., Hicks, A., and Poulin, R. (2011). Factors influencing infection patterns of trophically transmitted parasites among a fish community: host diet, host–parasite compatibility or both? *Journal of Fish Biology*, 79(2): 466-485.

Musefiu, T.A., Obuko, E.B., and Bolarinwa, A.O. (2011). Isolation and identification of aerobic bacteria flora of the skin and stomach of wild and cultured *Clarias gariepinus* and *Oreochromis niloticus* from Ibadan, Southwest Nigeria. *Journal of Applied Sciences Research*, 7(7): 1047–1051.

Oden, E.M., Daniel, A.A., and Chris, N. (2015). Incidence of nematode parasites in snakehead, *Parachanna obscura* of the lower Cross river system, Nigeria. *International Journal of Fisheries and Aquatic Studies,* 2(4): 331-336.

Odo, G.E., Onoja, S.U., and Onyishi, G.C. (2012). The biology of *Parachanna obscura* (*Osteichthyes: Channidae*) in Anambra River, Nigeria. *International Journal of Fisheries and Aquaculture*, 4(8):154-169.

Ogawa, K. (2015). Diseases of cultured marine fishes caused by Platyhelminthes (*Monogenea, Digenea, Cestoda*). Parasitology, 142(1): 178-195.

Ogonna, C.A., Emmanuel, I.N., and Michael, D.A. (2017). Survey of ectoparasites of cultured fish from selected farms in Ebonyi State: potential for food and nutrient security. *International Journal of Research in Pharmaceutical and Biosciences,* 4(7): 1-6.

Ohene-Adjei, S., Teather, R.M., Ivan, M., and Forster, R.J. (2007). Postinoculation protozoan establishment and association patterns of methanogenic archaea in the ovine rumen. *Applied and Environmental Microbiology,* 73(14): 4609-4618.

Okoye, I.C., Abu, J.S., Obiezue, N.N., and Ofoezie, E.I. (2014). Prevalence and seasonality of parasites of fish in Agulu Lake, Southeast, Nigeria. *African Journal of Biotechnology,* 13(3): 502-508.

Olasunkanmi, J.B., and Ipinmoroti, M.O. (2014). Food of the African snake head (*Parachanna obscura*) in a protected area.

International Journal of Development and Sustainability, 3(1): 177-183.

Olugbojo, J.A., and Ayoola, S.O. (2015). Comparative studies of bacteria load in fish species of commercial importance at the aquaculture unit and lagoon front of the University of Lagos. *International Journal of Fisheries and Aquaculture,* 7(4): 36–46.

Olutimehin, I.O., Balogun, O.B., and Omoniyi, I.S.(2019). Qualitative and microbial characteristics of *Parachanna obscura* under cold storage temperature. *International Journal of Biology Research,* 4 (3): 01-09.

Omeji, S., Solomon, S.G., and Uloko, C. (2013). Comparative study on the Endo-parasitic infestation in *Clarias gariepinus* collected from earthen and concrete ponds in Makurdi, Benue State, Nigeria. *Journal of Agriculture and Veterinary Science*, 2(1): 45-49.

Onwuliri, C.O.E., and Mgbemena, M.O. (1987). The parasitic fauna of some freshwater fish from Jos Plateau, Nigeria. Nigerian *Journal of Applied Fisheries and Hydrobiology,* 2: 23-37.

Osho, E.F., Ajani, E.K., Omitoyin, B.O., and Aniebo, V. (2014). Physical masculization and growth performance of *Oreochromis niloticus* placed on 17α-methyltestosterone treated diet. *Journal of Environmental Extension,* 12: 41-45.

Osho, F.E. (2017). Parasitic helminth fauna of *Parachanna obscura* in River Ogun, southwest Nigeria. *African Journal of Fisheries and Aquatic Resources Management,* 2:79-85.

Osho, F.E., and Usman, R.A. (2019). Length-weight relationship, condition factor and fecundity of African snakehead *Parachanna obscura* from the Anambra River, South East Nigeria. Croatian. *Journal of Fisheries,* 77(2): 99-105.

Osho, F.E., Jenyo-Oni, A., Ajani, E.K., Okpokpo, A.N., and Okeke, E.O. (2020). Skin and gut microbiota of the African snakehead, *Parachanna obscura*, Günther, 1861 from River Ogun, Southwest Nigeria. *Ife Journal of Agriculture* 32(2): 67-78.

Osungbemiro, N.R., Sanni, R.O., Olaniyan, R.F., and Olajuyigbe, A.O. (2014). Bacteria flora in the gut and respiratory organs of *Clarias gariepinus* in fresh and brackish water habitats of Ondo State, South/West Nigeria. *International Journal of Animal and Veterinary Sciences*, 8(6): 558-561.

Praveenraj, J., Uma, A., Knight, J.D.M., Moulitharan, N., Balasubramanian, S., Bineesh, K.K. and Bleher, H. (2018). *Channa quinquefasciata*, a new species of snakehead (*Teleostei: Channidae*) from Torsa River, North Bengal, India. Aqua *International Journal of Ichthyology*, 24(4): 141-152.

Rodrigues, C., and Cunha, M.Â. (2017). Assessment of the microbiological quality of recreational waters: indicators and methods. *Euro-Mediterranean Journal for Environmental Integration*, 2: 1-18.

Saha, S., Roy, R.N., Sen, S.K., and Ray, A.K. (2006). Characterization of cellulase-producing bacteria from the digestive tract of tilapia, *Oreochromis mossambica* (Peters) and grass carp, *Ctenopharyngodon idella* (*Valenciennes*). Aquaculture Research, 37(4): 380-388.

Shinkafi, S.A., and Ukwaja, V.C. (2010). Bacteria Associated with fresh Tilapia fish (*Oreochromis niloticus*) Sold at Sokoto Central Market in Sokoto, Nigeria. *Nigerian Journal of Basic and Applied Sciences*, 18(2): 217-221.

Uruku, M.N., and Adikwu, I.A. (2017). Seasonal prevalence of parasites of Clariids fishes from the lower Benue River, Nigeria. *Nigerian Journal of Fisheries and Aquaculture*, 5(2):11–19.

Vishwanath, W., and Geetakumari, K. (2009). Diagnosis and interrelationships of fishes of the genus *Channa Scopoli* (*Teleostei: Channidae*) of northeastern India. *Journal of Threatened Taxa*, 1(2): 97-105.

Willey, J. M., Sherwood, L. M., and Woolverton, C. (2008). Prescott, Harley and Klein's microbiology. McGraw Hill Higher Education, New York.1088pp.