Original Research Article

Growth status and Parasitic Fauna in Intestines, Gills and Skins of *Clariasgariepinus*Collected from Ogbese River and Owena River, South-West Nigeria

ABSTRACT

The study aimed to determine condition status and identify parasitic fauna in intestine, gills and skins of *Clariasgariepinus* collected from two natural waters:Ogbese River (River A)(Longitude 5°26'E' and Latitude 6°43'N), andOwena River (River B)(Longitude 5.03E and Latitude 7.03N)in Ondo state, Nigeria respectively. A total of 120 live *C. gariepinus* African Mud Catfish were collected by the assistance of fishermen using cast net during wet season duringApril to July, 2016 from the two natural water bodies.The fish were transported live to the laboratory for examinations. Length (cm) and weight (g) measurement of fish were determined. Condition factor (K), isometric value (b) and regression coefficient were determined. Fish samples were examined using electronic Microscope (x 40 Mag.) by dissecting fish to remove organs (Intestines, gills and skins) for parasites occurrence (s). Descriptive and analytical statistics were used to analyse the data obtained. The condition factor for all *C.gariepinus* samples collected from both Rivers were less than one (<1), which indicated that health status of the fish is biased, and environment is not conducive. Parasitic examination carried out revealed that seventy-eight (65%)*C. gariepinus* fish samples were infested; while 42 (35 %) of fish samples showed no parasite infestation.A total of Ninety-six (96) individual parasites were recovered from River A while a total of two hundred and twelve (212) individual parasites were recovered from River B. A total of eight (8) different parasites specieswere recovered while their percentage of occurrence were recorded. These include *Ambiphryaspp*(4.17%), *Camallanusspp*(6.25%; 2.83%), C*apillariaspp*(16.98%), *Chilodonellaspp*(14.58%), *Dactylogyrusspp*(64.58%; 5.66%), *Diphyllobothrium latum* (10.42%; 4.72%), *Gyrodactylusspp*(61.32%) and *Protoopalinasymphysodonis*(8.49%). The water bodies need to be protected against further pollutants to prevent disease condition for benefit of aquatic organisms and public health.

Keywords: Parasitic Occurrences, Growth status, *Clariasgariepinus*organs, Natural waters.

1. INTRODUCTION

Fish is one of the most important food and is valued for its nutritional qualities (Onyia*et. al.,* 2013). It is one of the important sources of protein for humans and other

animals in the tropics (Biu and Akorede, 2013). Fish is a good source of high quality and easily digestible protein containing essential amino acids and other beneficial nutrients (Onyia*et. al.,* 2013) required for good health: it provides a good source of vitamins and minerals (Onyia*et. al.,* 2013). Fish serve as a good source of animal protein for man and his livestock (Bichi and Yelwa, 2010). Fish not only provides food for immediate consumption but people rely on fishing for economic gains and jobs (Biu and Akorede, 2013). A well-processed fish product from the tropics has a ready market in developed countries and is therefore a good foreign earner (Imam and Dewu, 2010). The most common fish available in Nigeria are the catfish species (e.g. *Clarias spp.*). The sharp mouth catfish, *Clariasgariepinus*(Burchell, 1822) occurs mainly in quiet waters, lakes and pools but may also occur in fast flowing rivers (Ayanda, 2009). It is highly priced in Nigeria either as smoked, dried or fresh (Imam and Dewu, 2010).

The study of parasites of freshwater fishes in Africa is sporadic and inadequate considering the fish health of the continent (Ajala and Fawole, 2014). Studies in Africa vary considerably from area to area and the parasites are mostly mentioned, as part of the fulfilment of the biology of the host fish species (Ajala and Fawole, 2014).Parasites are a major concern to freshwater and marine fishes all over the world, and of particular importance in the tropics (Bichi and Dawaki, 2010; Ekanem*et. al*.,2011). The effects of parasites on fish include nutrient devaluation (Hassan *et. al*.,2010); lowering of immune capability, induction of blindness and mechanical injuries depending on the parasite species and load (Echi*et. al*.,2009 a, b). Parasites may induce a shift in fish species densities, size, composition and affect commercially relevant stocks. Parasites are also good indicators of environmental contaminants and stress (Palm, 2011).

Parasitic diseases of fish are most frequently caused by small microscopic organisms called protozoa which live in the aquatic environment. There are varieties of protozoans which infest the gills and skin of fish causing irritation, weight loss, and eventually death. Most protozoan infections are relatively easy to control using standard fishery chemicals such as copper sulphate, formalin, or potassium permanganate etc. Protozoans are the most commonly encountered fish parasites (Klinger and Floyd, 2013). Protozoans are single-celled organisms, many of which are free-living in the aquatic environment (Klinger and Floyd, 2013). They typically have a direct life cycle, that is, no intermediate host is required for the parasites to reproduce (Klinger and Floyd, 2013).

Fish like any other valuable natural resources, require careful management.Inspite of the interest in the freshwater ichthyofauna of Nigeria, little or no attempt is made to identify and manage or control parasites. As at present, the paucity of research in fish diseases in Africa is not seen as a factor that will have negative impact on fisheries development and as such is not a target research area. However, occurrences of helminth parasites in fishes have been studied extensively in various water bodies in Nigeria, with most of the work done primarily from the morphologic and morphometric descriptions, but factors that may limit the ability of parasites to co-exist in multiple infections in a host fish species had in most studies been neglected (Ajala*et. al*., 2014).

In Nigeria, the emanating need to culture fishes for protein consumption for the rapidly growing populations have made it necessary to intensify studies on the parasitic fauna of the African freshwater fishes (*Clariasgariepinus*); and if these parasites are left uncurtailed, they may lead to mass mortality of fish, or in some cases, emergence of zoonotic species (Ajala and Fawole, 2014). The study of parasites in fishery resource management is of paramount importance. Hence, there is need to provide a deeper appreciation for the role of parasites in fish health assessments using *Clariasgariepinus*collected from two different natural water bodies. Therefore, this study was designed to investigate and identify the parasitic fauna in the intestine, on the gills and skin of adult *Clariasgariepinus* from two natural waters in Ondo State, Nigeria.

2. MATERIALS AND METHODS

2.1 Study area

This study was conducted inOgbese River (A) which lies between Longitude 5^{o}26'E' and Latitude 6^{o}43'N; and Owena River (B) which lies between Latitude 7.03N Longitude 5.03E. Ogbese River is one of the major perennial rivers in South-Western Nigeria and it took its source from Awo-Ekiti in Ekiti State. Owena Riveris also perennial in nature, and is used asa major source of domestic water supply to the people of Ondo and Akure townships. It has a surface area of about $15Km^2$.

2.2 Sample collection

A total of one hundred and twenty (120) live *Clariasgariepinus* fishes were collected by the assistance of fishermen from Ogbese and Owena Rivers in Ondo state during April to July, 2016. Fish samples were transported during the early hours (9:00hours-10:00hours) of the day in sanitized plastic container (25 litres) with water from respected River sourceto Fisheries laboratory, Federal University of Technology, Akure, where growth assessments and parasitological examination were carried out.

2.2.1 Growth Parameters Assessment

- Measurement of standard length (cm) was taken using graduated meter rule while weight (g) of fish were taken using electronic scale (Mettler Toledo electronic weighing balance (PB8001)).
- Condition factor (K) of the fish were determined toevaluate the health status of the fish in relation to its environmentusing:

$K = 100W/ L^3$……..(Abowei, 2009).

Where:

K = The Condition factor

W = Weight of fish in grams (g)
L = Standard length of fish in centimetres (cm)

- Regression analysis was carried out to assesses the relationship between increase in length with weight gain of the fish using:
  $W=aL^b$…… Equation 1 (Leonard *et. al.,* 2012)
  Where:
  W=Weight of fish in grams (g)
  L= Total Length (TL) of fish in centimetres
  a= Scaling Constant
  b= Allometric growth coefficient
  The "a" and "b" values were obtained from a linear regression of the length and weight of fish.
  Transformed equation into linear regression:
  Log W = Log a + b Log L …… Equation 2 (Dan-Kishiya, 2013)
  The regression coefficient (R^2) correlation coefficient of the fish were determined.

2.3 Sex grouping

*Clariasgariepinus*samples collected from Ogbese River and Owena River were sepereted into male and female respectively.

2.4 Parasitological study

*Clariasgariepinus*fish samples were dissected, and the body cavities were opened with the aid of a dissecting set. The fish wer examined for endoparasites and ectoparasites using microscopic technique (direct wet mounts using Giesma staining method).

The skin, intestine and gills of the fish samples were dissected and a gram specimen of each organ were cut to make a squash with a mixture of 1 gram Nacl and 10 ml distilled water. A drop of this was placed on the cavity slide with a syringe and viewed under Olympus trinocular microscope (CX 40) mounted with microphotograph (Scope image). The parasites observed were counted, identified and recorded. Degree of parasitic infection in intestine, gills and skin of *Clariasgariepinus* collected from the rivers were observed and recorded.

2.5 Statistical analysis

Data were subjected to statistical analysis using Software Package Social Sciences (SPSS Version 6.0). Analytical and descriptive statistics were engaged to analyse data collected. Furtheranalysis was carried out using Duncan Multiple Range Test. mean and standard deviation (Mean ± Standard Deviation) of data were determined. Regression analysis were carried out and correlation (r) for respective data on growth were determined.

The condition factor (K) was calculated using the appropriate statistical formula given below:

$$K = \frac{W \times 100}{L^3}$$

K= The Condition factor

W= Weight (g) of fish

L= Total Length (cm) of fish

3. RESULTS

3.1 Growth Parameters Determinations

3.1.1 Length and Weight Measurements

A total of 120 *Clariasgariepinus*collected from Ogbese River and Owena River indicated length range from 22.90 – 34.40 cm and weight range 133.5 - 332.4 g. Table 1 shows mean and standard deviation of standard length (cm) and weight (g) of fish samples collected over four months.

Table 1: Mean and standard deviation Length (cm) and Weight (g) *Clariasgariepinus*collected fromOgbeseRiverandOwenaRiver

	Weight (g)	Standard length (cm)
OgbeseRiver		
April	201.00 ± 16.72^{c}	27.89 ± 2.58^{a}
May	232.99 ± 31.92^{a}	28.08 ± 1.73^{a}
June	219.53 ± 48.25^{b}	27.29 ± 3.64^{a}
July	228.35 ± 26.17^{a}	27.73 ± 2.56^{a}
OwenaRiver		
April	208.00 ± 57.17^{c}	28.01 ± 2.10^{a}
May	234.68 ± 58.19^{a}	27.96 ± 2.65^{a}
June	155.36 ± 20.20^{d}	27.06 ± 1.90^{a}
July	212.47 ± 31.22^{b}	26.84 ± 2.14^{a}

Means with different alphabet superscriptrepresent significant level at P is ≥5% within the column.n = 120.

3.1.2 Regression Analysis

The regression analysis of the length (cm) and weight (g) of fish from the two Rivers were revealed in Figure 1 and 2. Frequency of occurrence of fish, mean and standard deviation on standard length (cm) and weight (g) of all fish samples collected; Condition Factor (K), regression coefficient (R^2), coefficient of determination (r), and isometric values (b) of fish were determined, (Table 2).

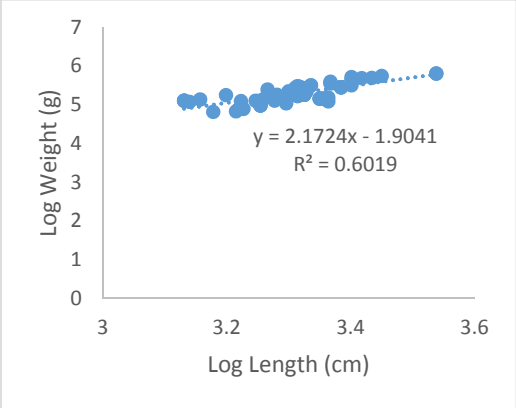

Figure 1. Regression of Clariasgariepinus collected from Ogbese River.

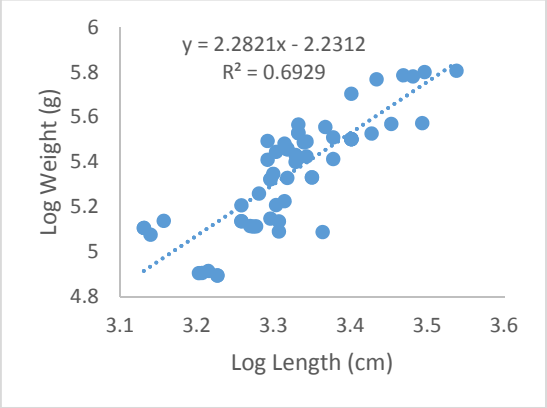

Figure 2. Regression of Clariasgariepinus collected from Owena River.

Table 2. Growth Parameters Determined on *Clariasgariepinus* Collected from Ogbese River and Owena River.

FreshWater Environments→ Growth Parameters↓	**Ogbese River**	**Owena River**
Frequency of Occurrence	60	60
Mean Standard length (cm)± standard deviation	27.58± 0.32	27.86± 0.68
Mean Weight (g) ± standard deviation	205.34± 2.24	217.26± 2.74
Condition Factor (K)	0.98	1.00
Regression Coefficient (R^2)	0.60	0.69
Coefficient of Determination (r)	0.78	0.83
Isometric Value (b)	2.17	2.28

3.2 Parasite Occurrence in *Clariasgariepinus* Samples Collected

Parasites highest occurrence(64.58 %) inOgbeseRiver occurred in *Dactylogyrus specie*with 232.49 prevalence;Gyrodactylus species ranked highest (61.32) in occurrence and 220.75 prevalence in Owena River. Tables 3 and 4 showed frequency and prevalence of parasites ocuurence on *C. gariepinus* from the two environments.Figure 3 showed prevalence of parasites in male and female samples of *C. gariepinus*in both environments over the experimental period.

Table 3: Frequency, Percentage Occurrence and Prevalence of Parasitic fauna in *Clariasgariepinus* from Ogbese River and OwenaRiver

Parasites	Ogbese River			Owena River		
	Frequency	% Occurrence	Prevalence	Frequency	% occurence	Prevalence
Ambiphrya spp.	4	4.17	15.01	0	0.00	0.00
Camallanus spp.	6	6.25	22.50	6	2.83	10.19
Capillaria spp.	0	0.00	0.00	36	16.98	61.13
Chilodonella spp.	14	14.58	52.49	0	0.00	0.00
Dactylogyrus spp.	62	64.58	232.49	12	5.66	20.38
D. latum	10	10.42	37.69	10	4.72	16.99
Gyrodactylus spp.	0	0.00	0.00	130	61.32	220.75
P. symphysodonis	0	0.00	0.00	18	8.49	30.56
Total	96	100.00	360.00	212	100.00	360.00

Table 4: Monthly Frequency of Occurrence and Percentage Occurrence of Parasites Infestation in *Clariasgariepinus* from Ogbese River and Owena River

Month	Frequency of Occurrence of Parasites in OgbeseRiver	Percentage Occurrence in Ogbese (%)	Frequency of Occurrence of Parasites in OwenaRiver	Percentage Occurrence in Owena (%)
April	30	31.25	74	34.91
May	24	25	65	30.66
June	24	25	40	18.87
July	18	18.75	33	15.56
Total	96	100	212	100

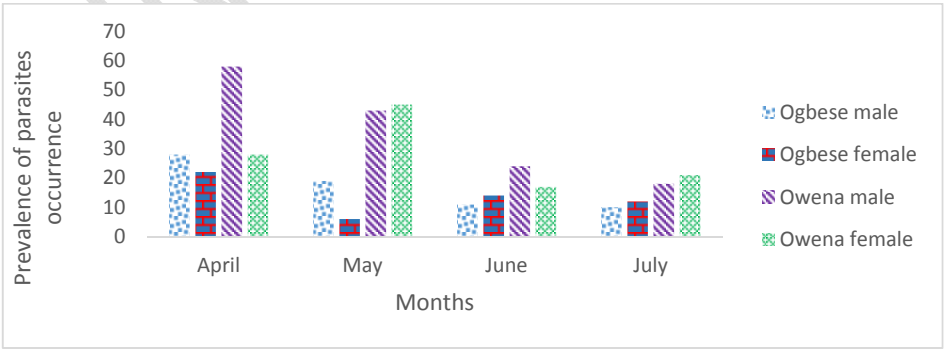

Figure 3: Prevalence of parasites in Male and Female *Clariasgariepinus* from Ogbese River and Owena River in

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206 Prevalence (%) and comparative parasitic fauna recoveredof parasite in fish organs revealed
207 parasites occurred most in the gills and intestines, and least in skins of *C.gariepinus*fish
208 samples fromOgbese River and Owena River(Tables 5 and 6).

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210 **Table 5:Prevalence (%) of Parasites in Intestines, Gills and Skins of *Clariasgariepinus***

Parasite	Ogbese River			Owena River			Total
	Intestine	Gills	Skin	Intestine	Gills	Skin	
Ambiphrya spp.	0.00	4.17	0.00	0.00	0.00	0.00	4.17
Camallanus spp.	6.25	0.00	0.00	2.83	0.00	0.00	9.08
Capillaria spp.	0.00	0.00	0.00	16.98	0.00	0.00	16.98
Chilodonella spp.	0.00	0.00	14.58	0.00	0.00	0.00	14.58
Dactylogyrus spp.	0.00	64.58	0.00	0.00	5.66	0.00	70.24
D. latum	10.42	0.00	0.00	4.72	0.00	0.00	15.14
Gyrodactylus spp.	0.00	0.00	0.00	61.32	0.00	0.00	61.32
P. symphysodonis	0.00	0.00	0.00	8.49	0.00	0.00	8.49
Total	16.67	68.75	14.58	94.34	5.66	0.00	200

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213 **Table 6: Comparative Parasitic Fauna Recovered in Organs (intestine, gills and skin)**
214 **of*Clariasgariepinus*in Ogbese River and Owena River**

Parasitic species	**River**		**Part/Location**		
	Ogbese	Owena	Intestine	Gills	Skin
Ambiphrya spp.	+	-	-	+	-
Camallanus spp.	+	+	+	-	-
Capillaria spp.	-	+	+	-	-
Chilodonella spp.	+	-	-	-	+
Dactylogyrus spp.	+	+	-	+	-
Diphyllobothrium spp.	+	+	+	-	-
Gyrodactylus spp.	-	+	-	+	-
Protoopalina spp.	-	+	+	-	-

215 *spp*: Species; + Present; - Absent

216

217 Figures 4 and 5 showed percentage infestation of parasites on *C. gariepinus* from Ogbese and
218 Owena Rivers. Dactylogyrus spp. ranked highest in Ogbese River, while Gyrodactylus spp.
219 ranked highest in Owena River.

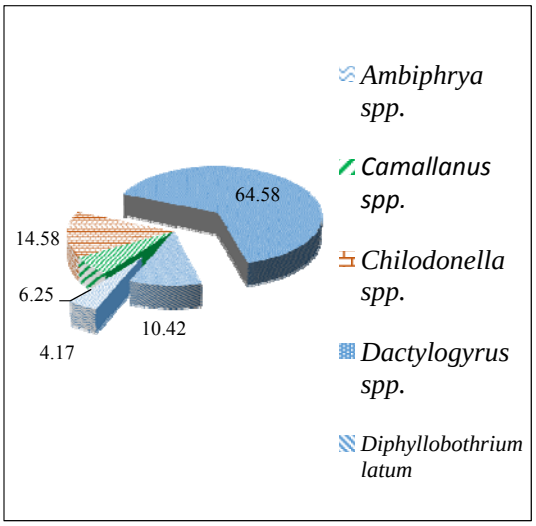

Figure 4: Percentage Infestation in Clariasgariepinusfrom Ogbese River

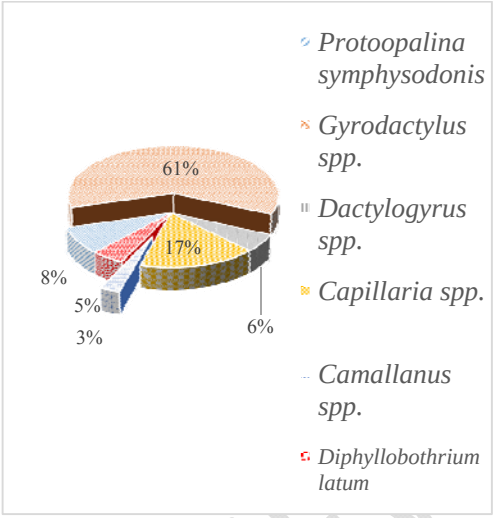

Figure 5: Percentage Infestation in Clariasgariepinusfrom Owena River

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223 Taxonomy and classification with site of recovery of parasitic fauna in C. gariepinus is
224 indicated in Table 7; and plates 1 – 8 showed the parasitic fauna pictorially.

Table 7: Taxonomical Classifications and Sites of Recovery of Parasitic Fauna Recovered in *Clariasgariepinus*Fish Samples

Parasites	Taxonomical group or classification							Site of Recovery	Type of parasite
	Kingdom	Phylum	Class	Order	Family	Genus	Species		
Ambiphrya	Animalia	Protozoa	-	Sessilida	Ambiphridae	*Ambiphrya*	-	Gills	Ectoparasite
Camallanus	Animalia	Nematoda (roundworms)	Secernentea	Camallanida	Camallanidae	*Camallanus*	*lacusris, truncatus*	Intestine	Endoparasite
Capillaria	Animalia	Nematoda	Adenophrea	Trichurida	Capillaridae	*Capillaria*	Multiple *spp.* e.g. *hepatica*	Intestine	Endoparasite
Chilodonella	Protista	Ciliophora	Phyllopharyngea	Cyrtophorida	Chilodonellidae	*Chilodonella*	*Uncinata*	Skin	Ectoparasite
Dactylogyrus	Animalia	Trematoda (Platyhelminthes)	Monogenea	Monopisthocotylea	Dactylogyridae	*Dactylogyrus*	*extensus*	Gills	Ectoparasite
Diphyllobothrium	Animalia	Platyhelminthes	Cestoidea	Pseudophyllidea	Diphyllobothriidae	*Diphyllobothrium*	*latum*	Intestine	Endoparasite
Gyrodactylus	Animalia	Trematoda (Platyhelminthes)	Monogenea	Monopisthocotylea	Gyrodactylidae	*Gyrodactylus*	*salaris*	Gills	Ectoparasite
Protoopalina	Chromista	Heterokontophyta	Opalinea	Opalinida	Opalinidae	*Protoopalina*	*symphysodonis*	Intestine	Endoparasite

PLATES SHOWING RECOVERED PARASITES IN *Clariasgariepinus* FROM OGBESE RIVER AND OWENA RIVER

A total of eight (8) parasites recovered in the intestine, on the gills and skin of *Clariasgariepinus*comprised of two ectoparasitic protozoans (*Ambiphrya spp.* and *Chilodonella spp.*), one endoparasitic protozoan (*Protoopalinasymphysodonis*), two monogenean trematodes (*Dactylogyrus spp.* and *Gyrodactylus spp.*), two nematodes (*Camallanus spp.* and *Capillaria spp.*) and cestode (*Diphyllobothrium latum*).

Theparasites recovered in *Clariasgariepinus*catfish samplesfrom Ogbese River and Owena River are as shown below, (Plates 1 - 8).

Plate 1: Protoopalinasymphysodonisin the intestine of Clariasgariepinus (Mg. 400X)

Plate 2: Diphyllobothrium latum in the intestine of Clariasgariepinus(Mg. 400X)

Plate 3: Gyrodactylus spp. on the gills of Clariasgariepinus(Mg. 400X)

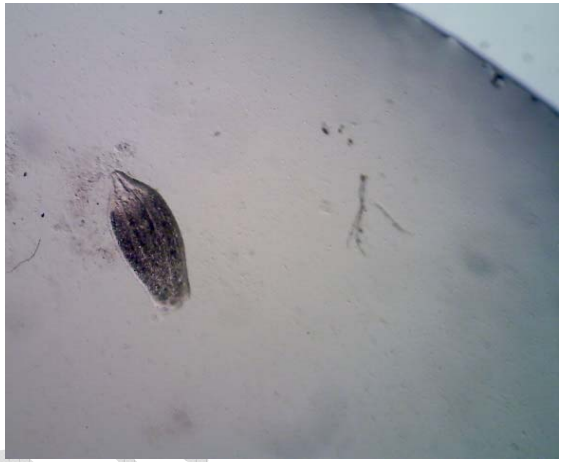

Plate 4: Dactylogyrus spp. on the gills of Clariasgariepinus(Mg. 400X)

Plate 5: Capillaria spp. in the intestine of Clariasgariepinus(Mg. 400X)

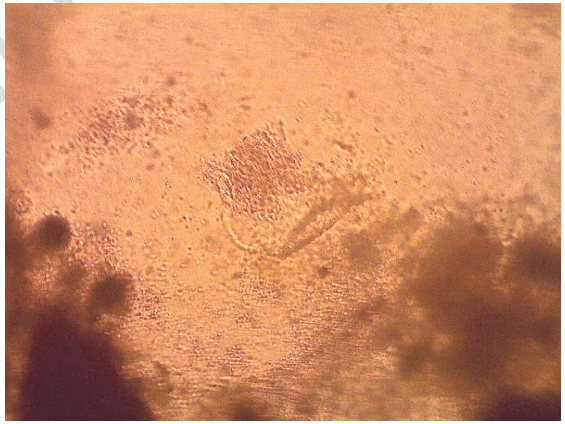

Plate 6:Ambiphrya spp. on the gills of Clariasgariepinus(Mg. 400X)

Plate 7: Chilodonella spp. on the skin of Clariasgariepinus(Mg. 400X)

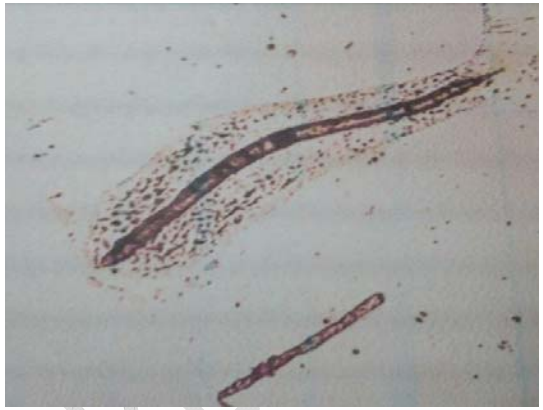

Plate 8Camallanusspp in the intestine of Clariasgariepinus(Mg. 400 400X)

4. DISCUSSION

4.1 Parasites Recovered

The condition factor for all the fish samples (*Clariasgariepinus*) collected from both Rivers were less than one which indicated that the living aquatic environment for the fishes were not conducive. Parasitic fauna in and on wild *Clariasgariepinus is* made up of myriads of parasitic and pathogenic organisms. These organisms are in their own individual ways of more or less economic and health importance for the fish and humans.

A total of eight (8) parasites recovered in the intestine, on the gills and skin of *Clariasgariepinus*belong to different *phyla*; Protozoa, Nematoda, Ciliophora, Trematoda and Heterokontophyta. The parasites comprised of two ectoparasitic protozoans (*Ambiphrya spp.* and *Chilodonella spp.*), one endoparasitic protozoan (*Protoopalinasymphysodonis*), two monogenean trematodes (*Dactylogyrus spp.* and *Gyrodactylus spp.*), two nematodes (*Camallanus spp.* and *Capillaria spp.*) and one cestode (*Diphyllobothrium latum*).

The effects of parasites on fish hosts in the wild may be difficult to quantify because the aquatic environment is constantly polluted from different sources (Mastan*et. al*., 2009). *Ambiphryaspp* and *Protoopalinasymphysodonis* occurred in very small percentages when compared to total parasitic percentage; this may indicated possibility of the parasites naturally existing at a negligible level in wild *Clariasgariepinus*. *Camallanusspp* nematode has negative health effect on fish with high infestation. *Dactylogyrusspp* and *Gyrodactylusspp*had high prevalence while *Diphyllobothrium latum*(broadfish tapeworm)had negative health implications on fish and humans (the end-users of fish and fish products), causes human *Diphyllobothriosis*(Scholz *et.al.,* 2009).

A total of one hundred and twenty (120) live fish samples (*Clariasgariepinus*) were examined, and seventh - eight (78) fish samples were infested with parasites, giving a prevalence of 65%. The frequency of parasite infestation including the percentage intensity in *Clariasgariepinus*from the two natural water bodies. Table 4 revealed higher parasite prevalence in OwenaRiver than OgbeseRiver. And more parasites were recovered in fish samples from Owena River thanOgbese River.Occurrence of intestinal parasites *Diphyllobothrium latum*corroborated Biu and Akorede, (2013) who reported helminth infections as quite common in wild fish. Infestation rates vary greatly from one area to another. Previously work by Bichi and Yelwa, (2010) is in line with the findings as he reported such infestation in Northern Nigeria.Overall infestation rate (65%)obtained depicted high infestation when compared to 16.6% reported from Asa River at Ilorin. This may be due to the fact that definitive host amongst others determine to a large extent the rate of infection (Obano*et.al.,* 2010).

Rate of parasites infestation differed with sex of fish in the study, male fish had higher parasites occurrence than female. This may be as a result of differential feeding either by quantity or quality of food or as a result of different degrees of resistance to infestation. However, this contradicts Biu and Akorede, (2013) who reported that variations in parasitic infestation among the sexes of fish studied were not significant implying that higher infestation rates in either male or female were simply by chance. In addition, the occurrence of parasites in *Clariasgariepinus* may be indicative of similar diets, feeding habits and patterns among the freshwater fishes. The pathological effects of helminths recovered are as a result of the mechanical damage caused by the attachment organs.

Owena River revealed higher frequency and percentage prevalence parasite infestation on *C.gariepinus*fishsamples than Ogbese river samplesover experimental months (Figures 1 and 2).The parasites recovered were found majorly in intestine and on gills but to a lesser extent on

skin. Ectoparasites recovered include *Ambiphryaspp*, *Chilodonellaspp*, *Dactylogyrusspp* and *Gyrodactylus spp*. Endoparasites recovered include *Protoopalinasymphysodonis*, *Diphyllobothrium latum*, *Capillaria spp.* and *Camallanus spp.* Of the endoparasites identified in the course of this research work, *Capillariaspp* and *Diphyllobothrium latum* were very common.*Ambiphryaspp* and *Protoopalinasymphysodonis* only occurred in very small percentages (Table 7) when compared to the whole. *Camallanusspp* nematode a serious negative health effect on fish but only in the case of high infestation.

5. CONCLUSION

Fish parasites cause commercial losses in both the fisheries and aquaculture industries. Different parasite species affect fisheriesby decreasing the yield, reducing the quality of fish or rendering them aesthetically unacceptable. Hence, affecting human health and socio-economic implication.

Inferencesfrom this study revealedendo parasites and ecto-parasite fauna identified in wild *Clariasgariepinus*consistedof pathogenic and non-pathogenic organisms. These organisms are in their own individual of more or less economic and health importance for the fish, other organisms and humans. However, parasite occurrence should not be neglected because its increasing population in fish environment will be problematic and crease public health menace.

Therefore, control of parasites should be looked upon as a major aspect of management in fish production.Proper processing and culinary methods should also be put in place to reduce transmission of parasites within aquatic environment and for public health purposes.

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