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Parasite Prevalence and Bioaccumulation of Polycyclic Aromatic Hydrocarbons as Stressors in the Silver Catfish, *Chrysichthys nigrodigitatus* (Siluriformes: Claroteidae)

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ABSTRACT

The experiment explored the impacts of polycyclic aromatic hydrocarbons (PAHs) and the prevalence of *Aspidogastrea africanus* (endoparasite) in *Chrysichthys nigrodigitatus* (host). Host-parasite allotment of PAHs, histopathological analysis, and the oxidative status of parasite and host were investigated. Oxidative status of fish and endoparasites were determined by assessing the levels of Glutathion-S-Transferase (GST), Sodium Oxide Dismutase (SOD), and Catalase (CAT). PAH concentrations were determined in the tissues of the host and parasite using gas chromatograph coupled to flame ionization detector (GC-FID). Physico-chemical parameters of water and sediment were assessed using a handheld multi-parameter probe (Horiba Water Checker Model U-10). The parasitic prevalence in the examined fish was proportional to length and weight of fish individuals. The parasites were more predominant among the length and weight cohorts of the female fishes than the males. Higher induction of oxidative stress enzymes in the intestine of the male *C. nigrodigitatus* than in the female, and the parasite can be attributed to the higher levels PAH and partly absence of parasites to depurate the fish. *A. africanus* shared the toxic burdens of chrysene, benzo(b)fluoranthene, dibenz(a,h)anthracene, benzo(g,h,i)perylene, dibenzo(a,l)pyrene, dibenzo(a,i)pyrene and particularly indo(1.2,3-cd)pyrene from the intestine of both sexes. In return, the endoparasite contributed the oxidative stress in the intestine of the fish. Synergistic and antagonistic interactions between PAH congeners and *A. africanus* on silver catfish, *C.nigrodigitatus* is evident in the current study. We suggest mitigation of PAH-releasing anthropogenic activities around Lekki lagoon for the protection of *C. nigrodigitatus*.

Keywords: polycyclic aromatic hydrocarbons; silver catfish; parasite intensity; oxidative stress; bioaccumulation factor

1. Introduction

Polycyclic aromatic hydrocarbons (PAHs) are a group of persistent, bioaccumulative and toxic compounds formed by two or more fused aromatic rings which comprise of carbon and hydrogen atoms (IARC, 2016). PAH are carcinogenic and mutagenic compounds that emanate from coal and tar deposits, combustion of engines, incinerations and forest fires (Cébron *et al.*, 2013).

Rise in population and urbanization are accompanied by increase in use of petroleum products, hence release of PAHs into the environment (Bouloubassi *et al.*, 2012; Bragato *et al.*, 2012). PAH was earlier reported in Lekki Logoon, Lagos State, Nigeria (Akinsanya *et al.*, 2014). This may hamper the productivity of the aquatic ecosystem thereby threaten the ecological and economic values (Costanza *et al.*, 1997; Akinsanya *et al.*, 2015).

Polycyclic aromatic hydrocarbons (PAHs) may adsorb on particulates and get precipitated to the bottom of sediment, thereby constituting future re-pollution of the overlying water (Peng *et al.*, 2014) thereby posing threat to aquatic organisms such as fish and shellfish (Chau, *et al.*, 2005; Cachot *et al.*, 2006; Valentín *et al.*, 2006; Tobiszewski and Namieśnik, 2012; Lindgren *et al.*, 2014). Deleterious impacts of PAHs on fish has been widely reported (Guzzella *et al*., 2005, Culotta *et al*., 2006; Commendatore *et al*., 2012; Aoki *et al*., 2014; Li et al., 2014; Soliman *et al*., 2012; Yuan *et al.*, 2014). Previous studies have also shown levels of PAHs in *C. nigrodigitatus* in Lekki lagoon (Akinsanya *et al.*, 2007, Saliu 2008, Olarinmoye *et al.*, 2009). Incidences of PAHs in other aquatic organisms in Lekki lagoon has previously been reported (Enuneku and Ilegomah, 2015; Akinsanya *et al.*, 2015; Doherty and Otitoloju, 2016). Sogbamu *et al.* (2016) also detected PAHs in the sediment and zebra fish captured from Lekki lagoon.

Chrysichthys nigrodigitatus is a highly demanded delicacy was in Nigeria. Reduced abundance of the fish species in Nigerian aquatic systems has been documented broadly reported (Idodo-Umeh, 2003, Ajah *et al.*, 2006, Akinsanya *et al.*, 2007; Offem *et al.*, 2008). This may be linked to toxic, carcinogenic and mutagenic potentials of PAHs (Callén *et al.*, 2013; Cébron *et al.*, 2013) in conjunction with their extended half-life which can range from years to

decades (Cachot *et al.*, 2006; Lindgren *et al.*, 2014; Siddall *et al.*, 1994; Tobiszewski and Namieśnik, 2012; Vane *et al.*, 2013).

Due to low fat composition, fish parasites are unable to bioconcentrate lipophilic substances above the levels of the host (Akinsanya *et al.*, 2014; Akinsanya *et al.*, 2015). Trade-offs between host-parasite allotments of PAHs, in conjunction with debilitating effects of parasite on host fish may provide useful insight in understanding the survival chances of the fish when simultaneously faced with parasitism and PAHs toxicity (Costa *et al.*, 2009; Au, 2004; Isibor, 2017).

The study was aimed at evaluating the bioaccumulation of PAHs in silver catfish, *Chrysichthys nigrodigitatus* and the net host-parasite (*Aspidogastrea africanus*) allotment of PAH.

2. Materials and methods

2.1 Description of study area

Lekki Lagoon is situated between latitudes 3°50' - 4°10' N and longitudes 5°30' - 5°40' E. It has a surface area of more than 243km² and it is flanked by the Epe axis of the lagoon (freshwater) in the east and Lagos lagoon (brackish water) in the west. We collected water and sediment samples from 14 random locations at the lagoon (Figure 1).

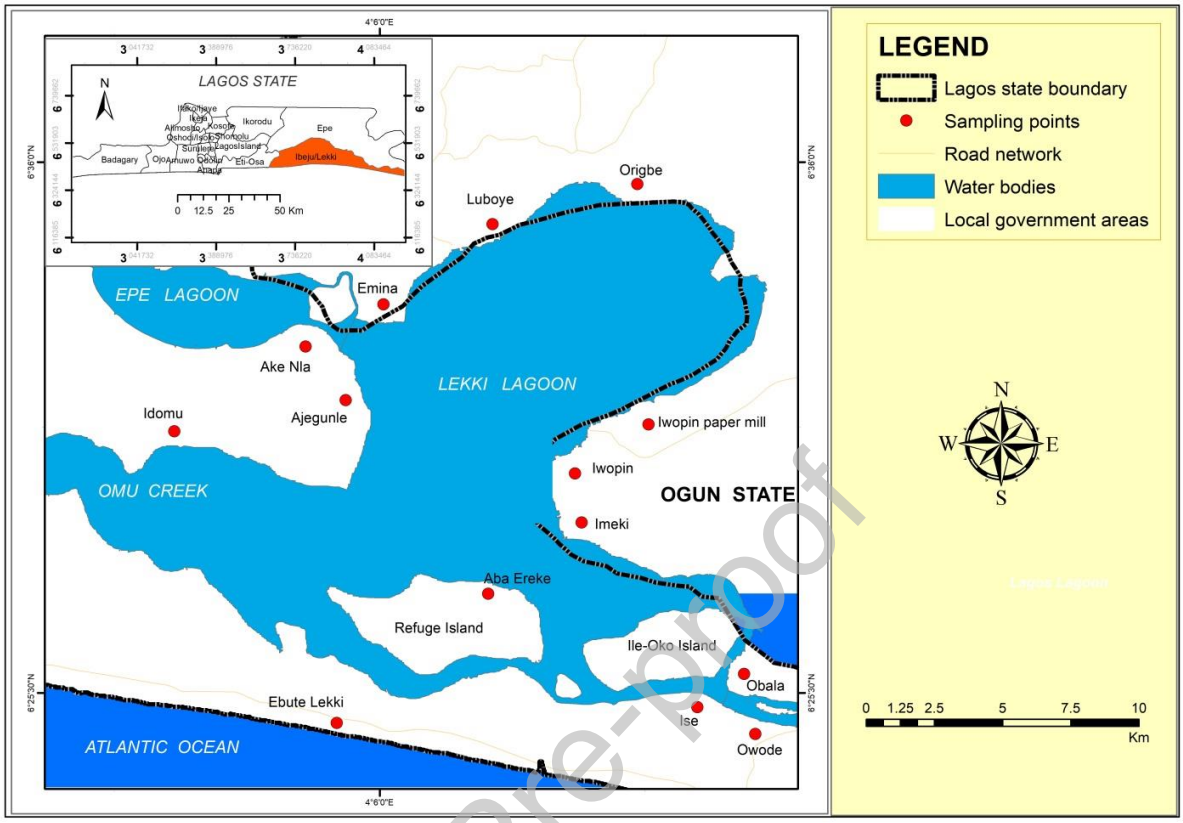

Fig. 1: Map of study area showing sampled locations

2.2 Collection and analysis of fish

2.2.2 Fish morphometrics

A total of 120 juvenile *C. nigrodigitatus* (126.11±54.7 g) were purchased from fishermen at Oluwo market, Epe, Lagos, Nigeria between March, 2017 and August, 2017. They were identified using procedures prescribed by Paugy *et al.* (2015). The standard length of each fish was measured using a transparent meter rule and recorded to the nearest 0.5 centimeter (cm), while the weight (w) was obtained using a Standard loading Denward balance and recorded to the nearest 0.1 g. Nine regression models which include: linear, logarithm, growth, exponential, logistics, quadratic, inverse, cubic, and power was used to analyze the length-weight relationship.

2.2.3 Bioaccumulation

Bioaccumulaton Factor (BAF) was determined thus:

$$BAF = \frac{Concentration\ of\ PAH\ in\ tissue}{Concentration\ of\ PAH\ in\ water\ sample}$$

Biota-Sediment Accumulaton Factor (BSAF) was determined thus:

$$BSAF = \frac{Concentration\ of\ PAH\ in\ tissue}{Concentration\ of\ PAH\ in\ sediment\ sample}$$

2.2.4 Antioxidant enzymes analysis

We determined lipid peroxidation using the method of Jiang *et al.* (1992). Fish tissue was dried in oven at 40^{o} C for 48 hours. It was then grinded into powdery form using porcelain grinder. We then homogenized 10 g of dried sample using 0.5 mL Tris-Hcl buffer (pH= 7.5). The homogenate produced was kept for all subsequent analysis in the research. We then treated 0.1 mL of tissue homogenate with 2 mL of TBA-TCA-HCI reagent (thiobarbituric acid 0.37%, 0.25N HCI and 15% TCA) at ratio 1:1:1 ratio. We then placed the mixture in water bath for 15 minutes. It was allowed to cool to room temperature and centrifuged it at room temperature for 10 min at 3,000rpm. We then measured the absorbance of clear supernatant against reference blank at 535 nm.

Reduced glutathione (GSH) was determined by the method of Ellman (1959). 10% TCA was added to the homogenate centrifuge. 1.0 ml of supernatant was treated with 0.5 ml of Ellmans reagent (19.8 mg of 5, 5'-dithiobisnitro benzoic acid (DTNB) in 100 ml of 0.1% sodium nitrate) and 3.0 ml of phosphate buffer (0.2M, pH 8.0). The absorbance was read at 412 nm.

Catalase (CAT) was assayed calorimetrically at 620nm and expressed as moles of hydrogen peroxide (H_2O_2) consumed/min/mg protein as described by Quinlan *et al.*, (1994). The reaction mixture (1.5ml) contained 1.0ml of 0.01M pH 7.0 phosphate buffer, 0.1ml of Plasma and 0.4ml of 2M H_2O_2. We discontinued the reaction and added 2.0 mL of dichromate-acetic acid reagent (5% potassium dichromate and glacial acetic acid were mixed in 1:3 ratio).

We estimated the total SOD activity in tissue using the ability of SOD to inhibit the autoxidation of pyrogallol. We then mixed 970 µL of buffer (100 m MTris-HCl, 1 mM EDTA, pH 8.2), 10 µL

of homogenates and 20µL pyrogallol 13 mM. We performed in thermostated cuvettes at 25°C and were recorded. Changes of absorption were recorded by a spectrophotometer (Spectronic 20D) at 480nm.

2.2.5 Determination of PAH

We determined PAH in the intestinal tissues of the fish using KOH refluxing/ vortex extraction and EPA Method 3611C. We weighed 50g (wet weight) of tissue sample and homogenized it with 0.5 mL Tris-Hcl buffer, at pH 7.5. 15 mL of 6N KOH was added to the homogenate and transferred into a sealed tube and incubated for 18hours in a 35ºC water bath, while we centrifuged sample for 30seconds at intervals of 30 minutes for 4hours. Afterwards, 15 mL of methylene was added to the centrifuge tube at 2000rpm for 5 minutes to allow mixture separate into phases. The upper aliquot layer was removed using a pasteur pipette into a 250 mL round-bottom flask.

GC-FID determination of polycyclic aromatic hydrocarbons (PAHs) was done using Agilent 7890B gas chromatograph coupled to flame ionization detector (FID). The stationary phase of compound separation used was HP-5 capillary column coated with 5% Phenyl Methyl Siloxane (30m length ×0.32mm diameter ×0.25µm film thickness) (Agilent technologies). 1µm of the samples was injected in split less mode at an injection temperature of 300ºC, at a pressure of 13.74psi and a total flow of 21.364ml/min, purge flow to split vent was set at 15ml/ min at 0.75min.oven was initially programmed at 40ºC (1min) then ramped at 12ºC/min to 300ºC (10min). FID temperature was 300ºC with hydrocarbon: Air flow at 30ml/min: 300ml/min while nitrogen was used as makeup gas at a flow of 22ml/min. After calibration the samples were analyzed and corresponding PAHs concentration obtained.

2.3 Examination of gastrointestinal parasites

The abdominal cavity of each fish was dissected using a sterile blade and the gastrointestinal part was eviscerated, segmented and placed in petri dishes containing physiological saline. The intestines were further opened and explored for endo-parasites. The parasites discovered were identified as *Aspidogastrea africanus* using identification guidelines of Akinsanya *et al.* (2014). Parasites were counted, fixed in 70% alcohol and recorded accordingly.

Parasites prevalence, load and intensity were estimated thus:

$$Parasite\ Prevalence = \frac{\text{Number of infected fish}}{\text{Total number of fish examined}}$$

$$Bioload = \frac{\text{Number of collected parasites}}{\text{Number of infected fish}}$$

$$Percentage\ parasite\ Intensity = \frac{\text{Number of collected parasites}}{\text{Number of fish examined}}\ X\ 100$$

All estimations such as mean values and standard deviations were conducted at confidence interval of 95%.

2.4 Collection and analysis of water and sediment samples

Water and sediment samples were collected from 14 random stations of varied anthropogenic activities liable to produce PAHs. Water samples were collected in 1 L sterile glass bottles, while sediment samples were collected using a Van Veen grab sampler (15x15x12 cm) and preserved in sterile aluminum foil pretreated with 10% nitric acid. Preserved samples were transported to the Laboratory of Department of Marine Sciences of the University of Lagos where they were held at 4±1 °C for two weeks prior to laboratory analysis.

2.4.1 Analysis of water samples

We conducted in-situ measurement of physiochemical parameters of water such as temperature, using a mercury-in-glass thermometer. Salinity, dissolved oxygen, pH, Turbidity, total suspended solids (TSS), total dissolved solids (TDS) and conductivity were measured using a handheld multi-parameter probe (Horiba Water Checker Model U-10).

PAH in water samples was determined by adding 5 mL of 6N KOH to 50 mL of water sample. The mixture was transferred into a sealed tube and incubated for 18hours in a 35ºC water bath, while we centrifuged sample for 30seconds at intervals of 30 minutes for 4hours. Afterwards, 15 mL of methylene was added to the centrifuge tube and then centrifuged again at 2000rpm for 5

minutes to allow mixture separate into phases. The upper aliquot layer was removed using a Pasteur pipette into a 250 mL round-bottom flask. PAH in water was then determined by GC-FID using Agilent 7890B gas chromatograph coupled to flame ionization detector (FID).

2.4.2 Analysis of sediment samples

For determination of PAHs in sediment, we air-dried soil samples for 48 hours to remove moisture. 2.5 grams of the air dried soil sample was then dissolved in 10ml of hexane and shaken for 10 minutes using a mechanical shaker. The solution was filtered using a Whatman filter paper No. 42. Afterwards, 15 mL of methylene was added to the filtrate and centrifuge at 2000rpm for 5 minutes. The upper aliquot layer was removed using a Pasteur pipette into a 250 mL round-bottom flask. PAH in water was then determined by GC-FID using Agilent 7890B gas chromatograph coupled to flame ionization detector (FID).

2.5 Statistical Analysis

The descriptive statistics (mean±SE) of PAH in *A. africanus* and in the intestinal tissues of *C. nigrodigitatus* samples, concentrations of antioxidant enzymes in the *C. nigrodigitatus,* and physicochemical parameters of the water and sediment samples were subjected to analysis of variance (ANOVA). The outcomes were further subjected to the Duncan Multiple Range test (DMR) in order to ascertain the actual locations of significant differences using the 2007 Excel and SPSS 2010 version tool packages.

3. Results

The pH (7.45) of water sample from the lagoon was within the range specified by WHO (2009). However, electrical conductivity (EC) and total dissolved solids (TDS) of the water were very much significantly higher ($p< 0.01$) than the set standard limits of WHO. The dissolved oxygen concentration (8.39 mg/L) was also significantly higher ($p< 0.05$) than the set regulatory standard limit (Table 1).

Table 1: Mean values of physico-chemical parameters of water samples from 14 locations at Lekki Lagoon

Parameter	Concentration	WHO (2009)	p Value
pH	7.45±0.23	6-8	>0.05
EC (µS/cm)	**28305**±43.2	400	<0.01
TDS (mg/L)	**14125**±38.3	2000	<0.01
Salinity (ppt)	17.33±2.23	-	-
Dissolved oxygen(mg/L)	**8.93**±0.13	7.5	<0.05
Chloride(mg/L)	9571.50±23.2	-	-
Sulfate(mg/L)	138.60±7.81	500	>0.05
TSS (mg/L)	28.50±1.23	30	>0.05
Turbidity(NTU)	35.00±2.21	-	-
Ammonia (mg/L)	8.949±0.43	-	-
Nitrate (mg/L)	0.1202±0.00	20	>0.05
Nitrite (mg/L)	0.0730±0.00	-	-
Acidity (mg/L)	7.6±0.63	-	-
Alkalinity (mg/L)	78±4.63	-	-
Bicarbonate (mg/L)	95±2.28	-	-
Phosphorus (mg/L)	0.1426±0.00	<5	>0.05
THCs (mg/L)	2.12±0.01	10	>0.05
Chemical oxygen demand (mg/L)	32±3.23	80	>0.05

Emboldened figures are significantly higher than regulatory limits at $p< 0.05$= significant, $p<0.01$= very much significantly higher, $p> 0.05$= not significant. Sample size, N= 14. EC= electrical conductivity, TDS= total dissolved solids, TSS= total suspended solids, THCs= total hydrocarbons

We compared the mean values of parameters investigated in the sediment of Lekki Lagoon for 6 months with reference values obtained from other locations in previous studies. The electrical conductivity of the sediment of Lekki was far higher than the WHO limit (Table 2). The total nitrogen in the current study area was higher than the level observed at Mada River in Nassarawa State, Nigeria by Tukura *et al.* (2012).

Table 2: Mean values of selected physico-chemical parameters of sediment samples from 14 locations at Lekki Lagoon

Parameter	Concentration	Reference limits
pH	7±0.23	6.5-8.5 (WHO, 2009)
Electrical Conductivity (µS/cm)	**7439**±82.23	100 (WHO, 2009)
SO_4^{2-}(ppm)	3528.49±30.55	-
Total organic compound (%)	0.351±0.03	16.87 (Avramidis *et al.*, 2012)
Cl^- (ppm)	2378.50±44.53	-
Total Nitrogen (%)	**2.058**±0.13	1.88 (Tukura *et al.*, 2012)
Total Phosphorus (%)	0.005±0.0	0.45 (Avramidis *et al.*, 2012)
Exchangeable acidity (cMol/Kg)	0.10±0.0	-
Total hydrocarbons (ppm)	2.822±0.13	-

Emboldened figures are significantly higher than reference limits at $p< 0.05$. Sample size, N= 14.

A total of 120 samples of juvenile *C. nigrodigitatus* (126.11±54.7 g) were collected from Lekki lagoon between the period of May and August, 2017. The population comprised of 77 (64.20%) males and 43 (35.80%) females (Table 3). Only 10 (8.33%) individuals (all females) were infected with parasitic cestode- *Aspidogastrea* africanus (3.30%).

Table 3: Prevalence of *Aspidogastrea africanus in* Lekki Lagoon.

Status	Male	Female	Total Examined
No Examined	77 (64.20%)	43 (35.80%)	120 (100.00%)
Infected Individuals	0 (0.00%)	10(8.33%)	10 (8.33%)
Non-Infected Individuals	77 (64.20%)	38 (32.50%)	115 (96.70%)

We further explored the parasitic prevalence and intensity of infection in *C. nigrodigitatus* individuals in relation to their standard length and weight. Result showed remarkable correlation between fish length and parasitic susceptibility. The various length groups showed variation in prevalence, intensity and parasite load. The highest prevalence, intensity and load occurred in the length cohort 33cm- 42.9cm (Table 4). Parasite prevalence was directly proportional to the standard length. Fish in the highest weight cohort (190 – 219 g) exhibited the highest parasite intensity (Table 5). A directly proportional relationship occurred between weights and parasite intensities of the female fish, while no infection was detected in the males.

Table 4: Prevalence of *Aspidogastrea africanus* infections in *Chrysicthys nigrodigitatus* relative to standard length of the fish

Standard Length (cm)	Fish Examined	Fish Infected	Parasite Prevalence	Number of parasistes	Bioload	Parasite Intensity (%)
Female						
13.0 – 22.9	30	2	0.06	120	60	4
23.0 – 32.9	7	3	0.43	135	45	19.3
33.0 – 42.9	6	5	0.83	156	31.2	26
Male						
13.0 – 22.9	64	0	0	0	0	0
23.0 – 32.9	10	0	0	0	0	0
33.0 – 42.9	3	0	0	0	0	0

Table 5: Prevalence of Gastrointestinal *Aspidogastrea* Infections in *Chrysichthys nigrodigitatus* relative to their body weight

Weight (g)	Fish Examined	Fish Infected	Parasite Prevalence	Number of parasites	Bioload	Parasite Intensity (%)
Female						
40 – 69	9	0	0	0	0	0
70 – 99	7	0	0	0	0	0
100 – 129	5	1	0.20	14	14	280
130 – 159	5	2	0.40	14	7	280
160 – 189	9	3	0.11	6	2	66.7
190 – 219	7	4	0.14	28	7	400
Male						
40 – 69	22	0	0	0	0	0
70 – 99	12	0	0	0	0	0
100 – 129	5	0	0	0	0	0
130 – 159	14	0	0	0	0	0
160 – 189	16	0	0	0	0	0
190 – 219	9	0	0	0	0	0

High bioaccumulation factor of benzo(c)phenanthrene occurred concurrently in the intestine of *C. nigrodigitatus* and in the parasite (Table 6). PAHs with significant BAFs in the intestine of fish quite corresponded with those in the parasite. The total PAHs in female *C. nigrodigitatus* constituted 23.6% of the total PAHs in the entire fish samples collected while the male constituted 76.4%. This implies that the male *C. nigrodigitatus* had 52.8% higher PAHs than the female counterparts.

Total IM = 48.861, Total IF = 15.056 (Table 6), Overall Total = 63.917

Percentage of IM = $\frac{48.861}{63.917} X\ 100 = 76.444\ \%$

Percentage of IF = $\frac{15.056}{63.917} X\ 100 = 23.556\ \%$

Where IF= concentration of PAH in female fish and IM= concentration in male

The trend of total PAHs in the environmental media (Table 6) was male fish (48.861) > water (19.33) > female fish (15.056) > parasite (7.103). Although higher levels of PAHs were detected in the intestine of the male fish than the female, more PAHs were bioaccumulated into the parasites from the female fish that the male fish (Table 6). High levels of PAH in the male fish corroborate the outstandingly high bioaccumulation factor of PAH from the water medium into the intestine of the male. The parasites showed great ability to mop up indo(1.2,3-cd)pyrene from the intestine of both sexes with the high bioaccumulation factors of 5.9 and 6.01 recorded in the male and female fish respectively.

Table 6: Bioaccumulation factors of PAHs in parasite, *A. africaus* and host, *C. nigrodigitatus*

Components (ppm)	Water	IM	IF	Parasite	BCFim/w	BCFif/w	BCFp/im	BCFp/if
Naphthalene	0.23	2.331*	1.667	BDL	**10**	**7**	-	-
Acenaphthylene	0.12	1.223	0.255	BDL	**10**	**2**	-	-
Acenaphthene	0.18	3.112*	0.298	0.145	**17**	**2**	0.05	0.5
Fluorene	0.52	1.221*	0.549	0.194	**2**	**1.**	0.16	0.35
Anthracene	0.18	5.661*	0.368	0.123	**31**	**2**	0.02	0.33
Phenanthrene	0.17	1.223*	0.601	0.262	**7**	**4**	0.21	0.41
Fluoranthene	0.24	7.322*	0.214	BDL	**31**	**1**	-	-
Pyrene	0.55	1.344	0.120	BDL	**2**	0	-	-
Benzo(c)phenanthrene	0.01	0.000	4.327*	0.047	**0**	**432**	-	0.01
Benz(a)anthracene	0.46	3.112*	0.289	BDL	**6.8**	**1**	-	-
Chrysene	0.49	5.211*	0.238	0.315	**10.6**	0	0.06	**1.32**
Benzo(e)pyrene	12.38	0.755	0.668	0.352	0	0	0.47	0.51
Benzo(b)fluoranthene	0.47	0.822	0.474	0.464	**2**	**1**	0.56	**1**
Benzo(j)fluoranthene	0.00	0.622	0.437	BDL	-	-	-	-
Benzo(k)fluoranthene	0.38	0.521	0.511	BDL	**1**	**1**	-	-
Benzo(a)pyrene	0.20	0.566	0.317	0.236	**3**	**2**	0.42	0.74
7,12-Dimethylbenz(a)anthracene	0.69	3.211*	0.882	0.382	**5**	**1**	0.12	0.43
3-Methylcholanthrene	0.38	0.000	0.326	0.238	0	0	-	0.73
Indo(1.2,3-cd)pyrene	0.30	0.321	0.315	1.894	**1**	**1**	**5.9**	**6.01**
Dibenz(a,h)anthracene	0.52	0.521	0.651	0.829	**1**	**1.**	**1.59**	**1.27**
Benzo(g,h,i)perylene	0.49	8.422*	0.294	0.692	**17**	**1**	0.08	**2.35**
Dibenzo(a,l)pyrene	0.37	0.000	0.415	0.400	0	**1**	-	**1**
Dibenzo(a,i)pyrene	0.00	0.433	0.355	0.393	-	-	0.9	**1.1**
Dibenzo(a,h)pyrene	0.00	1.228	0.485	0.137	-	-	0.11	0.28
TOTAL PAH IN MEDIA	**19.33**	**48.861**	**15.056**	**7.103**				

Asterisked figures= significant difference among concentrations in media. Emboldened figures= significant bioaccumulation factors. IM- conc. of PAH in intestine of male fish, IF- conc. of PAH in intestine of female fish. BCFim/w- bioaccumulation factor of PAH from water to intestine of male fish= conc. in intestine ÷ conc. in water. BCFif/w- bioaccumulation factor of PAH from water to intestine of female fish= conc. in intestine ÷ conc. in water. BCFp/i- bioaccumulation factor of PAH from intestine to parasite= conc. in parasite ÷ conc. in intestine.

There was no significant difference in the activities of GSH between both sexes of fish and the parasite (Table 7). The activity of MDA was in the order of male fish > female fish > parasite, while the CAT in both sexes of fish were not significantly different and were higher than concentrations detected in the parasite. Activity of SOD was in the order of female fish > male fish > parasite. The level of SOD in the female fish was significantly higher than the levels in the male and parasites.

Table 7: Summarized anti-oxidative responses in host, *C. nigrodigitatu* and parasite, *A. africanus*

	GSH	SOD	CAT	MDA
Female *C.nigrodigitatus*	0.03±0.00	232.34±12.33^{a}	1.38±0.27^{a}	18.89±3.55^{b}
Male *C.nigrodigitatus*	0.05±0.01	217.84±52.33^{b}	1.04±0.37^{a}	23.59±9.35^{a}
A. africanus	0.04±0.00	207.90±26.07	0.76±0.32^{b}	12.60±3.25

Values with different superscripts are significantly different at $p<0.05$. N= 120. SI UNITS= SOD (U/mg prot.), CAT (nmoles/ min/mg prot.), MDA (µmol MDA/g tissue).

A highly significant correlation relationship (0.929) occurred between bioaccumulation factor of parasite and antioxidant enzymes in female fish (Table 8). This further buttressed the link between accumulation capacity of the enteric parasite and the stress level detected in the female fish.

Table 8: Correlation analysis between bioconcentration factors of PAHs in parasite and antioxidant enzymes in female fish

	GSH	SOD	CAT	MDA	BCF
GSH	1				
SOD	0.089	1			
CAT	0.041	0.113	1		
MDA	0.329	0.056	0.029	1	
BCF	**$0.529^{0.6}$**	**$0.929^{0.6}$**	**$0.629^{0.6}$**	0.429	1

Emboldened values are significant = $p<0.05$

Taking a cue from the significant difference that occurred in the antioxidant enzymes and the highly significant correlation between the BAF of enteric parasite and SOD in the intestine of the

female fish. We further employed regression analysis of SOD in the intestine of the female fish on the BAF of the parasite in order to ascertain the actual relationship.

The calculated p value of 0.01 was less than the tabulated value of 0.05, indicating a significant regression of oxidative stress on the bioaccumulation factor of PAH into the tissues of the parasite from the intestine of the female fish (Figure 2). Furthermore a correlation analysis of GSH, SOD, CAT, and MDA in the intestine of female fish with the bioaccumulation factors of PAHs in the enteric parasite showed a highly significant correlation between

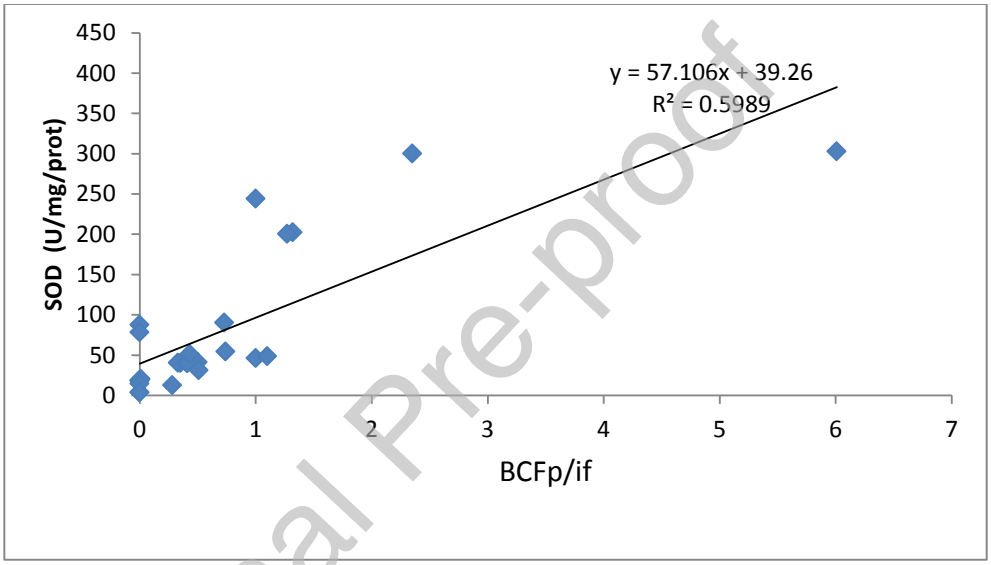

Figure 2: Regression of SOD on the bioaccumulation factor of PAHs from female intestine to enteric parasite

4. Discussion

The parasitic prevalence in the examined fish was proportional to length and weight of fish individuals. The parasites were more predominant among the length and weight cohorts of the female fishes than the males. The relatively higher prevalence of parasites in the female fish explains the higher bioaccumulation rates of the PAHs from the intestine of the female into the parasite than the male fish. Observation of higher parasitic prevalence in females than in males is at variance with the observations of Akinsanya *et al.* (2014), Ekanem *et al.* (2011), Esiest (2013), and Idris *et al.* (2013). However, the current observation of proportionate increase in parasitic

prevalence with weight and length of fish conforms to the findings of Paraskevi and Konstant (2012).

Results showed host-parasite toxicant transfer is implicated in the difference between PAH levels in the intestines of male and female *C. nigrodigitatus.* This is attributable to the ability of *A. africanus* to absorb significant amount of PAHs from the female fish. *A. africanus* which relieved female *C. nigrodigitatus* of PAH burden was absent in the male counterparts, thereby resulting in the male fish having 52.9% higher PAHs than the female. This conforms to the findings of Heinonein *et al.* (2000) who pointed out that clams (*Pisidiium amnium*) infected with trematodes had 12% less 2,4,5-trichlorophenol, and 40% less benzo(c)pyrene when exposed. The proportion of the total PAH congeners available for uptake by the intestine of *Chrysichthys nigrodigitatus* is mainly a function of the bioavailability of the contaminants, organism's physiology and behavior (Isibor, 2017). The physiochemical properties of an aquatic environment also affect the fate of the constituent contaminants (Isibor *et al.,* 2016). Higher induction of oxidative stress enzymes in the intestine of the male *C. nigrodigitatus* than in the female, and the parasite can be attributed to the higher levels PAH and partly absence of parasites to depurate the fish (Akinsanya et al., 2019). This effect was sequestered in the female host by the endoparasite, *A. africanus* as the parasite took up substantial amount of PAHs from the female fish. Homoestatically, the antioxidant defense released oxidative stress enzymes to scavenge for reactive oxygen species produced in the process of lipid peroxidation in order to abate oxidative stress. However, in extreme cases the enzymes may be used up and the concentration may no longer commensurate with stress levels. Combined effects of multiple stressors on fish host could have a significant impact.

The bioload of endoparasites in the female fish favoured reduced PAH concentration, however, the parasite intensity required for a substantial PAH withdrawal from the host is liable to result in serious disease conditions in the fish, which was evident in its oxidative stress level which did not commensurate with the level of PAH. The stress level in the female fish must have been elevated due to the presence of the parasites *A. africanus*.

Outstandingly high bioavailability of benzo(c)phenanthrene in the water may have contributed to its high bioaccumulation factor in the intestine of female *C. nigrodigitatus.* Result therefore

showed the ability of *A. africanus* to detoxify its host. Result further shows that parasites have a considerable share of host's toxicant load and may attain same stress levels as the host in extreme cases. The observation conforms to the findings of Akinsanya *et al.* (2014).

This study has provided size-based susceptibility to parasitic infections which may be triggered by immunosuppression, following exposure to PAH. Organ-specificity of PAH bioaccumulation in *C. nigrodigitatus* was earlier also reported by Ikue *et al.* (2016). These suggest that multiple factor determine toxicodynamics of PAHs in the fish.

Protection of the coastal ecosystem is imperative as the most susceptible cohorts in the fish population are the vital components. Protection of the silver catfish is feasible in the lagoon through abatement of human discharge of petroleum by-products from petrogenic source, use of fuel and other organic substances from pyrogenic sources.

4.1 Conclusion

Lekki Lagoon is considerably perturbed by the predominant anthropogenic activities to which we advise stringent mitigations. The study demonstrated the ability of *A. africanus* to share the toxic burdens of chrysene, benzo(b)fluoranthene, dibenz(a,h)anthracene, benzo(g,h,i)perylene, dibenzo(a,l)pyrene, dibenzo(a,i)pyrene and particularly indo(1.2,3-cd)pyrene from the intestine of both sexes. In return, the endoparasite contributed the oxidative stress in the intestine of the fish. Synergistic and antagonistic interactions between PAH congeners and *A. africanus* on silver catfish, *C.nigrodigitatus* is evident in the current study. We suggest mitigation of PAH-releasing anthropogenic activities around Lekki lagoon for the protection of *C. nigrodigitatus*..

DECLARATION STATEMENT

Authors declare no conflict of interest

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