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Assessment of Ectoparasites and Heavy Metal Concentrations in Hemichromis Fasciatus, Liza Falcipinnis and Sarotherodon Galileus from Abalama and Ilelema Waterfronts, Buguma Creek, Rivers State, Nigeria

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Ectoparasites and heavy metal pollution pose health risks to both wild and aquaculture fish species. Here, we examine the ectoparasites of fish species (Hemichromis fasciatus, Liza falcipinnis and Sarotherodon galileus) from Abalama and Ilelema locations of the Buguma Creek, Rivers State, Nigeria, over a period of six months (February to July, 2020). Fish tissues (muscles, gills and gut) were also examined for heavy metals (Cr, Cd and Pb). Parasitological investigations were done following standard procedures, as well as the heavy metal analysis which was done using atomic absorption spectrophotometer after dry ashing-acid digestion. Two ectoparasites were isolated: Zeylanicobdella arugamensis from L. falcipinnis at both locations and Cymothoa exigua from H. fasciatus at Ilelema. In surface water, Cr concentration was <0.003µg/l throughout the study period; Cd concentrations ranged between $0.06\mu g/l$ and $0.09\mu g/l$ at both locations, while Pb values were 0.29µg/l - 0.64µg/l at Abalama, and $0.56\mu g/l - 0.71\mu g/l$ at Ilelema. These values were within international acceptable standards. In fish tissues, however, Cr values were $<0.003\mu g/g$ in both locations throughout the study period. At Abalama, Cd ranged between 7.18 μ g/g and 8.15 μ g/g, and Pb between 72.83 μ g/g and 92.23µg/g. At Ilelema, Cd concentrations were between 7.56µg/g and 8.67 μ g/g while Pb values ranged between 78.32 μ g/g and 92.88µg/g. Differences in heavy metal concentrations in both water and fish tissues between both locations were not statistically significant (p>0.05). All fish species and tissues bioaccumulated appreciable concentrations of the heavy metals. Cr in fish tissues was within permissible limits, but all Cd and Pb values exceeded international permissible limits. These fishes would, as such, pose health hazards to consumers. It is therefore recommended that local authorities would regulate the harvest and trading of these fishes for human consumption, and tackle illegal refinery activities at both study locations to reduce heavy metal pollution.

Introduction

Ectoparasites of fish in southern Nigeria include monogeneans (Eyo et al., 2015), parasitic isopods and mites (Ugbomeh and Nwosu, 2016; Okere et al., 2019). These have been reported from several fish species, especially cichlid and Clarid species and Pomadasys. Ectoparasites are reported to be causative agents of diseases (Finley et al., 2003; Boxshall, 2005; Wall, 2007) in both wild and cultured fish populations. Overstreet et al. (2009) reported that crustacean ectoparasites were vectors of viral diseases such as Taura syndrome virus, white spot syndrome virus and yellowhead virus.

Heavy metals such as cadmium, chromium and lead can be bio-accumulated in fish tissues and are hazardous to man even at low concentrations (Oehlenschlager, 2010; Anacletus et al., 2014). According to Ryder et al. (2014), the concentration of heavy metals, such as Cd and Pb, in fish muscle (fillets) is usually of a low level, but high in organs such as the liver and kidney. This however, does not exclude the hazard associated with consuming fish from metal polluted habitats (Monroy et al., 2014).

In this study, we examined the fishes, Hemichromis fasciatus, Liza falcipinnis and Sarotherodon galileus, for ectoparasites and tissue heavy metal concentrations. The aim was to report on the ectoparasites as previous studies in same location examined helminth endoparasites (Ogbeibu et al., 2014) and heavy metal concentration in fish from Buguma Creek was poorly studied.

Methods

Study location: Buguma Creek is a brackish water system located in Asari-Toru Local Government Area, Rivers State, Nigeria. Samples were taken from two locations along the Buguma Creek: Abalama (latitude 4o 76' 30.2", longitude 6o 83' 84.5"), and Ilelema (latitude 4o 79' 76.5",longitude 6o 78' 69.7"). Economic activities of residents at both locations included fuel wood production, periwinkle farming, fishing, and illegal refinery of petroleum products.

Sample collection: Water samples were collected in plastic bottles of capacity 1 to 2 liter. These samples were collected from approximately 15-20 cm below the water surface. Triplicate samples was taken and analyzed. Before the collection of samples, all the sample containers were washed, dried and labeled. Analyses of samples for heavy metals was conducted at the Institute of Pollution Studies (IPS), Rivers State University, Port Harcourt, Nigeria, following the protocols of (APHA, 1975).

Fish samples (Hemichromis fasciatus, Liza falcipinnis and Sarotherodon galileus) were purchased monthly directly from fishers at both locations for six months, February to July, 2020. A total of 288 fish samples- comprised of 96 samples of each species- were examined. Sixteen samples of each fish species were examined per month from each location for six months. The sample size was determined following the formula of Cochran (1963). Samples were transported in moist aerated containers to the Parasitology Laboratory of the Department of Animal and Environmental Biology, Rivers State University, Port Harcourt, Nigeria.

Examination of fish species for parasites: The fishes were euthanized by pithing. Samples were examined for ectoparasites in the gills and mouth and on the body surface. Parasites recovered were fixed in 70% alcohol and identified using keys by Kabata (1985). Prevalence and mean intensity of infection were computed after Bush et al. (1997).

Determination of heavy metal concentrations in water and fish tissues: One liter of water sample was placed in a Kyeldahi flask. Twenty milliliters (20ml) of concentrated nitric acid (HNO3) was added to the sample which was predigested by heating gently for 20 mins under a bunsen burner. Thereafter, more acid was added for 30-40mins. When a clear digest of the solution was obtained, the digestion process was stopped and the flask was allowed to cool. The sample was then transferred into a 56 ml volumetric flask and made up to 50 ml with distilled water. The resulting solution was analyzed for lead, chromium and cadmium concentration in an Atomic Absorption Spectrophotometer (model 210 VGP, UK). The instrument was first calibrated using the standard solutions of lead, chromium and cadmium.

The AAS absorbed the digest and displayed the concentration of the metals present in the sample.

The muscles, gills and guts of Hemichromis fasciatus, Sarotherodon galileus and Liza falcipinnis were removed and placed in separate foils, labeled, and dried in a thermostat oven (model DHG-9053A) at 105°C. Thereafter, they were ground and 2g was placed in a Kyeldahi flask, digested using concentrated nitric acid and passed through AAS, as described above. Statistical analysis: Student t-tests were used to test for significance differences in the heavy metal concentrations between both locations, using SPSS Ver. 25.

Results and Discussion

Ectoparasites Recovered from Examined Fish Species

Two parasite species were recovered from the fishes. These were the metazoan parasite, Zeylanicobdella arugamensis from Liza falcipinnis and the isopod, Cymothoa exigua from Hemichromis fasciatus (Plate 1-2). Parasite prevalence and intensity were very low. The metazoan parasite was recovered from the mouth and intestines of L. falcipinnis from both study locations at a mean intensity of one parasite per infected host and prevalence of 10.4% per location (that is, 10 infected hosts at each location). The isopod, however, was recovered from the gill region of two H. fasciatus specimens from Ilelema at a prevalence of 2.1% and mean intensity of one parasite/infected host.



Plate 1. Zeylanicobdella arugamensis from Liza falcipinnis from Abalama and Ilelema, Buguma Creek, Rivers State, Nigeria (Scale: 0.5mm)



Plate 2. Isopod (Cymothoa exigua) collected from Hemichromis fasciatus, Ilelema, Buguma Creek, Rivers State, Nigeria (Scale: 1mm)

Heavy Metal Concentrations in Surface Water and Fish Tissues

In surface water samples, chromium values were below $<0.003\mu g/L$ at both locations throughout the study period. Cadmium and lead values were similar at both locations. Mean concentrations of Cd ranged between $0.06\mu g/l$ and $0.09\mu g/l$ at both locations. At Abalama, mean Pb concentrations ranged from $0.29\mu g/l$ to $0.64\mu g/l$ and at Ilelema, the values were between $0.56\mu g/l$ and $0.71\mu g/l$. There was no significant difference in heavy metal concentrations of surface water between both locations (p>0.05).

Concentrations of heavy metals in fish tissues were significantly higher than those in surface water (p<0.001). Chromium values were <0.003µg/g in all fish tissues throughout the study period. In *L. falcipinnis* from Abalama, mean Cd concentrations were 7.93µg/g, 8.15 µg/g and 7.45 µg/g in the muscle, gills and guts, respectively. The order of bioaccumulation of Cd in *L. falcipinnis* was Gut<Muscle<Gill. Mean Pb concentrations in *L. falcipinnis* from Abalama were 95.30µg/g, 90.17µg/g and 89.43µg/g in the muscle, gills and guts, respectively. In *L. falcipinnis* from Ilelema, mean Cd concentrations were 7.58µg/g, 8.63µg/g and 7.87µg/g, in the muscle, gills and guts, respectively. On the other hand, mean Pb concentrations in *L. falcipinnis* from Ilelema were 85.82µg/g, 92.88µg/g and 78.32µg/g in the muscle, gills and guts, respectively. There was no significant difference in heavy metal concentrations between both locations (p=0.865).

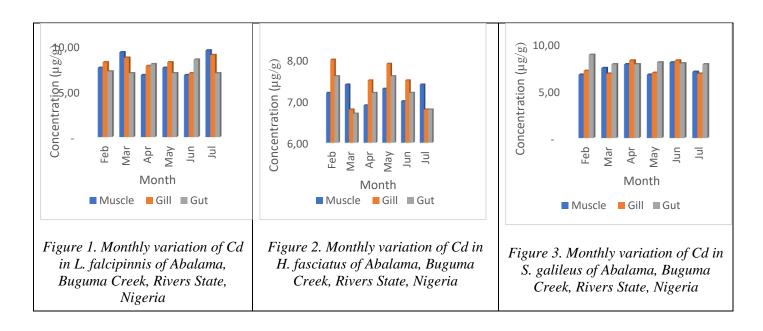
Monthly variation of the metals did not exhibit any seasonal trend (Figure 1-6). Graphical presentations of fish species bioaccumulation of heavy metals (Figure 7-18) showed that higher concentrations of Pb were accumulated over Cd. *Sarotherodon galileus* bioaccumulated the highest concentration of Pb ($105.9\mu g/g$) in the gut in February, 2020.

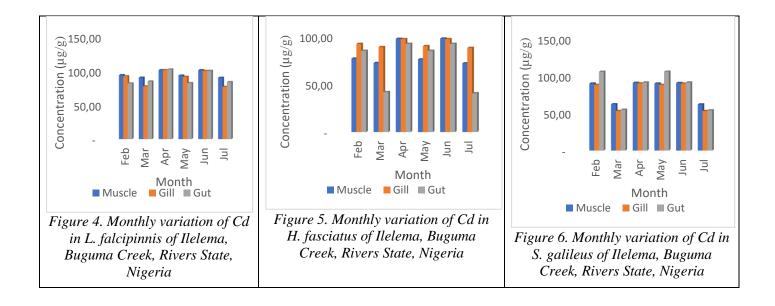
Table 1. Mean concentrations (µg/g) of heavy metals in fish samples from Abalama and Ilelema, Buguma Creek, Rivers State, Nigeria

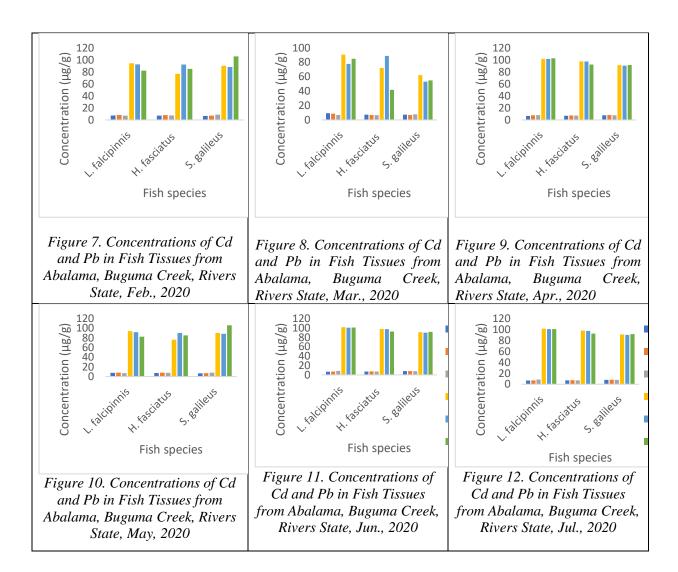
Location/Heavy metals	Fish Species		
	H. fasciatus	L. falcipinnis	S. galileus
Abalama			
Cd			
Muscle	7.20±0.21	7.93±1.09	7.27±0.56

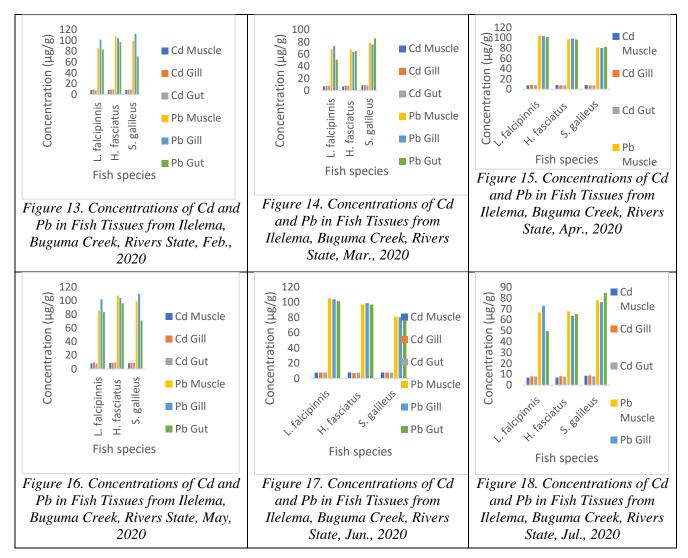
Gill	7.42±0.52	8.15±0.96	7.33±0.68
Gut	7.18±0.38	7.45 ± 1.04	8.02±0.39
Pb			
Muscle	82.03±12.26	95.30±5.08	81.03±14.67
Gill	92.23±4.09	90.17±5.08	77.17±18.66
Gut	72.83±24.72	89.43±9.50	83.97±23.76
Ilelema			
Cd			
Muscle	7.85±0.70	7.56±0.63	8.42±0.35
Gill	8.15±0.80	8.63±0.85	8.67±0.80
Gut	8.33±1.00	7.87±0.10	8.28±0.77
Pb			
Muscle	90.80±18.56	85.82±16.76	86.13±10.18
Gill	88.88±19.66	92.88±15.39	89.22±17.16
Gut	86.1±16.27	78.32±23.19	79.38±6.81

±Standar Deviation









Cymothoid ectoparasites have been reported in previous studies in Rivers State, Nigeria (Ugbomeh and Nwosu, 2016; Okere *et al.*, 2019). These parasites are commonly recovered from *Pomadasys* and *Tilapia* spp. as well as other fish species such as *Elops lacerta*, *Eucinostomus (Gerres) melanopterus, Pseudotholitus elongatus* and *Pseudotholitus senegalensis*. We recovered *Cymothoa exigua* from *Hemichromis fasciatus* from Ilelema. These parasites did not appear to cause deleterious effects. However, Binning *et al.* (2014) reported that the large cymothoid isopod, *Anilocra nemipteri*, parasitized the bridled monocle bream, *Scolopsis bilineata*, causing reduced host size, increased energy costs and decreased endurance.

Zeylanicobdella species are rarely reported in scientific research from Nigeria. In this research, we recovered them from *Liza falcipinnis* at both Abalama and Ilelema locations. Rueckert *et al.* (2008) also reported *Z. arugamensis* from *Mugil cephalus, Scatophagus argus* and *Lutjanus johnii* in Segara Anakan lagoon, Indonesia. *Zeylanicobdella arugamensis* is a hirudinean which commonly infects herbivorous fish and has a wide host range including freshwater, estuarine and marine fishes (Cruz-Lacierda *et al.*, 2000).

Heavy metal concentrations in water were below the standard levels for drinking water of $2000\mu g/l$, $3.0\mu g/l$ and $50.0\mu g/l$, for Cr, Cd and Pb, respectively (WHO, 2017). This could be attributed to higher accumulation in sediments and biota (Abdel-Baki *et al.*, 2011). Low concentrations of heavy metals in surface water were also reported by Uzairu *et al.* (2014) in Kano, Nigeria.

Chromium concentrations were $<0.03\mu g/g$ in all fish tissues. This was below the higher permissible limit of $0.07\mu g/g$ (WHO, 1995). In Abalama, Cd concentrations ranged between 7.18µg/g and 8.15µg/g, in *H. fasciatus* gut and *L. falcipinnis* gills, respectively. Lead (Pb) concentrations in this location ranged between 72.83 µg/g and 92.23 µg/g, in *H. fasciatus* gut and *H. fasciatus* gill respectively. At Ilelema, Cd concentrations ranged between 7.56 µg/g and 8.67 µg/g, respectively in *L. falcipinnis* muscle and *S. galileus* gills. Lead (Pb) values in fish tissues in this locations ranged between 78.32 µg/g and 92.88 µg/g, in *L. falcipinnis* gut and *L. falcipinnis* gills, respectively. All values for Cd and Pb exceeded the WHO (1995) permissible limits of $0.05\mu g/g$ and $0.01\mu g/g$, for Cd and Pb, respectively. This is contrary to previous reports that heavy metal concentrations were generally low in fish muscles (Ryder *et al.*, 2014).

Lead concentrations were generally higher than Cd concentrations in both surface water and fish tissues. This could be attributed to illegal refinery of petroleum products (Uzairu *et al.*, 2014), an activity which was very common at both locations. This could also be due to a higher affinity for Pb over Cd in the fish tissues examined (Winter *et al.*, 2012).

Conclusion

This research revealed two ectoparasites recovered from Liza falcipinnis and Hemichromis fasciatus. Zeylanicobdella arugamensis was isolated from Liza falcipinnis at both study locations while Cymothoa exigua was isolated from H. fasciatus at Ilelema, howbeit at low prevalence and mean intensities. Concentrations of the heavy metals (Cr, Cd and Pb) in surface water were below international permissible limits, but Cd and Pb concentrations in all fish tissues exceeded the standard limits. As such, the fish species examined in this research pose a health risk to consumers and should therefore, not be consumed.

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