

BACTERIOLOGICAL AND PHYSICOCHEMICAL ASSESSMENT OF FISH POND WATERS COLLECTED FROM IDOGBO COMMUNITY, EDO STATE, NIGERIA



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Abstract: The physicochemical and bacteriological profiles of water obtained from fish ponds stocked with the African catfish (*Clarias gariepinus*) in Idogbo community were evaluated using standard procedures. A grand total of 30 water samples were collected from 10 different fishponds (15 samples each from concrete and plastic ponds, respectively) between May and July 2018. The recorded mean physicochemical values for pH, temperature, electrical conductivity (EC), total dissolved solid (TDS), dissolved oxygen (DO), biological oxygen demand (BOD₅), nitrate and phosphate were within Federal Environmental Protection Agency(FEPA) guidelines for water quality. The DO of the fish pond water negatively correlated with BOD₅ (r=-0.310, p<0.01), and PO⁻⁴ (r=-0.230) at p<0.01 level. The mean heterotrophic bacterial counts and total coliform counts ranged from 26.11×10³±12.42 to 124.44×10³±59.24 cfu/ml and 12.11±1.27 to 33.78±6.61 MPN/100ml, respectively. The bacterial isolates identified were; *Pseudomonas aureginosa, Klebsiella pneumoniae, Proteus vulgaris, Bacillus sp., Staphylococcus aureus, Escherichia coli* and *Serratia sp.* PO⁻⁴₄, EC and TDS correlated positively with total bacterial count (r=

0.532), (r=0.429) and (r=0.309), respectively atp< 0.01 significant level. The results showed that the sampled fish pondwater contains water borne pathogenic bacteria of public health significance. There is therefore need for proper monitoring of the ponds and education of the farmers on effect of the use of organic manure as food additives in fish ponds.

Keywords: Fishponds, bacteriological, water, physicochemical, Idogbo community

Introduction

World fish center (2003) described fish as a dependable protein source to various global population groups. Increased daily need for fish and its products has resulted in more fish production by both public and private sectors in Nigeria (Obire and Ariye, 2015).

The success of rearing fish in ponds has been known to be dependent on the physicochemical and biological characteristics of the water utilized for fish culturing. Aquaculture is the rearing of the natural produce of water (fish, shellfish, algae and other aquatic organisms). It is different from fishing because it requires the input of humans in growing the number of organisms involved, unlike removing them from the wild (Kinsey, 2006). Fish ponds of various kinds which include concrete or earthen vats, wooden or fiber glass and plastics are used for the cultivation of fishes in different environment (Osawe, 2000). Ezenwa (2006) reported that among the various culture systems of fish rearing, concrete and earthen ponds are mostly employed. There is a low prevalence of disease when fishes are reared under natural conditions. This has been attributed to the large area of water and lower risk of interaction between the fish and the parasites (Khan, 2012). The incessant application of antibiotics in aquaculture has led to the problem of antibiotic resistance (Nwogu et al., 2011). The utilization of animal manure to augment traditional fish feed as a result of increased price of fish feed is also a problem in fish farming especially in countries like Nigeria. The use of organic manure as feed supplement in fish production could result to the buildup of opportunistic and pathogenic microorganisms of public health significance in ponds (Erondu and Ayanwu, 2005).

Good fish management begins with an understanding of the physical, chemical and biological characteristics of the ponds. The synergy of the distinct chemical composition of ponds determines the pleasant of water in fish ponds (Roy and Chavhan, 2017). Environmental stresses which consist of low dissolved oxygen, high temperature, and excessive ammonia content have an effect on the capacity of organisms to modify and conserve its internal environment (Ezra and Nwankwo, 2001).

Carbon dioxide, pH, alkalinity, hardness, oxygen availability and the toxicity of ammonia as well as the presence of heavy metals are interrelated and may have been suggested to have resultant effect on efficiency of ponds, the level of strain and fish fitness (ICAR, 2006). In view of this, tracking of the microbiological and physicochemical quality of fish ponds water as a way to determine the favourable, sub-lethal and lethal values of suitable conditions for aquaculture should be practiced. The aim of the present study therefore was to examine the physicochemical and microbiological characteristics of the ponds with a view to determining the quality of pond water as utilized for fish production.

Materials and Methods

Study area

The area selected for the study was Idogbo community which is the administrative headquarters of Ikpoba Okha Local Government area in Edo state. Its geographical coordinates are 6° 16' 00" N and 5° 43' 00" E. Residents of Idogbo are predominantly farmers, artisans and traders, while some are civil servants. Fish farming is a common practice in the community. The majority of the farmers rear fishes in either concrete or plastic ponds with ground water as the main source of water supply.

Sample collection

Water samples were collected from private owned fish ponds stocked with the African catfish (*Clarias gariepinus*) sited at different locations in Idogbo, Ikpoba Okha Local Government Area, Edo State, Nigeria. A total of 30 samples were collected from 10 different fishponds (5 concrete and 5 plastic ponds) between May and July 2018. The samples were collected using sterile screw-capped labeled bottles. The water samples were transported to the laboratory in an ice-packed container for bacteriological and physiochemical analyses between 2-3 h after collection.

Bacteriological analysis

The heterotrophic bacterial plate counts of the respective water samples was evaluated using serial dilution and pour plate techniques as described by Harley and Prescott (2002). Commercially available Nutrient agar (NA) was utilized in the heterotrophic bacterial enumeration whilst the total coliform flora of the respective samples was evaluated using the multiple tube dilution method as described by Cheesebrough (2006). The inoculated NA plates were incubated at 35°C for 48 h while the presumptive seeded test tubes for TCC evaluation were also incubated at 35°C for 48 h under aseptic conditions.

After incubation, resultant bacterial colonies from the NA plates were picked and purified by sub-culturing. The subcultured isolates were subjected to an assortment of relevant physiological, biochemical and sugar fermentation tests and the respective results were collated and compared to relevant identification schemes described by Cullimore (2000). The presumptive tubes which showed visible color change were documented and the MPN value was ascertained. Further appropriate confirmatory and completed tests were conducted on the presumptive positive tubes to determine the tentative identity of the coliforms present in the sample.

Physicochemical analysis

Several physicochemical parameters which include; pH, temperature, Electrical Conductivity (EC), Total Dissolved Solids (TDS), Dissolved Oxygen (DO), Biological Oxygen Demand (BOD), nitrate and phosphate were determined for the pond water samples. Temperature, pH, EC and DO were determined onsite using the relevant meters (Hanna instruments, version HI9828). TDS, BOD₅ phosphate and nitrate were determined using methods described by APHA (1998).

Statistical analysis

All parameters tested in this study were reported as means \pm standard deviations. The statistical package for social sciences (SPPSS version 20) was used to determine the analysis of variance (ANOVA), paired T- test and Duncan's multiple range tests were used to test for significance and mean separation respectively at 5% level of confidence. Pearson correlation was employed to examine and establish the association between the physicochemical and the bacteriological parameters of the pond water samples.

Results and Discussion

The results of the physicochemical parameters of the fish pond water samples collected in Idogbo are presented in Table 1. Temperature is an essential parameter in water. It affects the chemical and physicochemical characteristics of the environment as well as the organisms in the pond. The temperature profile of the fishpond water samples assessed during the study ranged from 27.21 ±1.60 to $28.70\pm1.05^{\circ}$ C. The temperature values of water in concrete and plastic ponds and across the study sites were not statistically different (p < 0.05). This trend might indicate that the pond types did not affect the water temperature in the ponds. The temperature

values were within the FEPA recommended range of 20-35°C. The findings in this study are similar to the reports of Ntegwu and Mojisola (2008) and Fafioye (2011) who reported 28 to 29°C and 27 to 28°C, respectively. The water pH is vital because it affects the solubility and availability of nutrients, and how they can be utilized by aquatic organisms (Osman and Werner, 2010). The pH levels of the collected fish pond water in this study were between 5.52±0.30 and 6.95±0.46°C. This is within FEPA regulatory limit of 5.0 to 9.0°C for optimum fish production and is in agreement with the study of Ntengwe and Mojisola (2008). There was a significant difference between the pH values across the sampled sites (p < 0.05). The result also revealed that the pH of water samples from the plastic fish ponds (6.52±0.60°C) were significantly higher than the pH of samples in concrete ponds $(5.96\pm0.58^{\circ}C)$ at p < 0.05. Table 4 showed that the pH was positively correlated with EC (r=0.721); TDS (r=0.528), BOD₅ (r=0.689), PO⁻⁴ (r= 0.531) at (p < 0.01) but negatively correlated with DO (r=-0.392) at p < 0.01. The sampled water pH weakly correlated positively with the total heterotrophic bacterial count (THBC) (r=0.202, p<0.05).

The Electrical conductivity (EC) of the fish pond water ranged from 109.62±62.84 to 554.39±52.93 µS/cm.EC of the plastic ponds (447.14 ±182.98) was significantly higher than that of concrete pond (200.67± 91.74) at p < 0.05 level. The EC values correlated positively with TDS(r=0.706); BOD₅ (r=0.687), PO₄⁻ (r= 0.855) at p < 0.01 level. The pond water

EC was positively but weakly correlated with THBC (r=0.429) at p < 0.01 level. There was a negative correlation between EC and DO (r=-0.385) at p < 0.01 level as shown in Table 4. The EC values were within the regulatory limit of 20-1500 µS/cm thus suitable for fish production. The findings from this study differs from those of Ehiagbonare and Ogunrinde (2010) who reported conductivity value of 0.012 -0.017 μ S/cm. The TDS in this study ranged from 77.04 \pm 27.32 to 299.66±29.31 mg/L. The TDS values differs significantly in their pond types with the plastic ponds values higher than the values of concrete ponds. The TDS values correlated positively with BOD₅ (r=0.487), PO_4^- (r= 0.616) and THBC (r=0.309) at p < 0.01 level and associated with NO⁻³(r=0.213, p < 0.05). TDS was negatively correlated with DO (r=-0.324) at p < 0.01 level (Table 4). The TDS values were within the regulatory limit of 500 mg/L (FEPA, 1999). This is could be attributed to regular partial water changes in the sampled fish ponds that help to dilute wastes and algae-promoting nutrients.

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sample code	Temp	pН	EC	TDS	DO	BOD ₅	NO $_3^-$	\mathbf{PO}_4^+
	° C		μS/cm	mg/L	mg/L	mg/L	mg/L	mg/L
CP1	27.21 ± 1.60^{b}	6.13±0.53°	167.38±63.73 ^b	84.82±32.06 ^{ab}	6.22 ± 0.56^{b}	2.17±0.69 ^a	0.75 ± 0.51^{a}	0.43 ± 0.19^{a}
CP2	27.33±1.63 ^{bc}	5.94 ± 0.87^{bc}	150.66±51.32 ^b	77.04±27.32 ^{ab}	6.18±0.63 ^b	3.34 ± 1.48^{bc}	0.59 ± 0.18^{a}	0.28±0.10a
CP3	27.96±0.76 ^{bcd}	5.81 ± 0.44^{abc}	265.69±23.66°	137.96±10.20bc	6.14 ± 0.83^{b}	2.90 ± 0.89^{ab}	2.70 ± 1.44^{b}	2.29±0.63 ^{bc}
CP4	28.70±1.05 ^d	6.09±0.60°	310.03±59.07°	156.27±30.43°	6.08 ± 0.58^{ab}	4.47 ± 0.35^{d}	0.57 ± 0.13^{a}	2.13±0.40 ^b
CP5	27.56±1.08 ^{bcd}	5.82 ± 0.44^{abc}	109.62±62.84bc	159.99±145.79°	6.02 ± 0.70^{ab}	2.52 ± 1.25^{ab}	1.47 ± 0.61^{a}	0.75 ± 0.54^{a}
PP1	28.01±0.89 ^{bcd}	6.68 ± 0.19^{d}	528.73±133.36 ^{ab}	299.66±29.31 ^d	6.40 ± 0.46^{bc}	4.66 ± 0.57^{d}	2.86 ± 1.18^{b}	3.78 ± 1.44^{d}
PP2	27.25±0.67 ^b	6.78±0.33 ^d	542.78±47.37d	272.01±20.36 ^d	6.16 ± 0.78^{b}	4.53±0.65 ^d	0.75 ± 0.14^{a}	4.64±0.77 ^e
PP3	27.80±0.53 ^{bcd}	5.52 ± 0.30^{ab}	118.93±62.72bc	162.56±144.94°	6.39 ± 0.60^{bc}	2.73±0.71 ^{ab}	3.79±2.39°	$0.92{\pm}1.09^{a}$
PP4	28.32±1.25 ^{bcd}	6.65 ± 0.30^{d}	554.39±52.93d	275.40±28.37 ^d	5.51 ± 0.25^{a}	4.10 ± 0.14^{cd}	1.16 ± 0.60^{a}	3.13±1.42 ^{cd}
PP5	28.44±0.38 ^{cd}	6.95 ± 0.46^{d}	490.89±57.62d	258.73±48.13 ^d	5.51 ± 0.26^{a}	3.82 ± 0.58^{cd}	1.23 ± 0.18^{a}	2.70 ± 1.02^{bc}
FEPA limits	20.00-35.00	5.00-9.00	20.00-1500.00	500.00	>2.00	10.00-20.00	30.00	0.01-3.0

 $\label{eq:Values} Values are Mean \pm SD \ of triplicates. Different superscripts in the same column indicate significant differences at p < 0.05 according to Duncan Multiple Range Test (State 1) and (State 2) according to Duncan Multiple Range Test (State 2) and (State 2) according to Duncan Multiple Range Test (State 2) and (State 2) according to Duncan Multiple Range Test (State 2) accordin$

(DMRT) Temp: Temperature; EC electrical conductivity; TDS total dissolve solids; DO: Dissolved oxygen; BOD₅ biological oxygen demand; NO $\frac{1}{3}$: nitrate; PO $\frac{1}{4}$ Phosphate.; FEPA: Federal environmental protection agency, 1999. CP=concrete pond; PP: plastic pond One of the most important parameter for controlling the activities of living organisms is dissolved oxygen (DO). It is used as a good index for water quality as it has strong association with several other parameters (Ravindra *et al.*, 2003; El-Nemaki *et al.*, 2008). The dissolved oxygen of the fish pond water negatively correlated with BOD₅ (r= -0.310, p< 0.01), and PO⁻⁴ (r= -0.230) at p< 0.01 level (Table 4). The DO values ranged from 5.51±0.26 to 6.39±0.60 mg/L across the sapling points and were within the acceptable limit (FEPA, 1999). The dissolved oxygen of the fish pond water negatively correlated with BOD₅ (r= -0.310, p< 0.01), and PO

 $\frac{1}{4}$ (r= -0.230) at p< 0.01. DO concentrations were not significantly different across the various sampling points and in the concrete and plastic ponds. This is most likely due to rainfall that resulted in high water volumes in the sampled fish ponds. The findings of this study were similar to those of Onome and Ebinimi (2010) but contrasted with values of Njoku et al. (2015) who reported DO readings below 5 mgL. Biological Oxygen Demand (BOD5)values in this study varied from 2.17 \pm 0.69 to 4.66 \pm 0.57 mg/L. BOD₅ values were within the permissible limit (10-20 mg/L) but differs significantly between the samples drawn from the concrete and plastic ponds (p < 0.05). BOD₅ was significantly higher in plastic ponds (3.08±1.26 mg/L) than concrete ponds (3.97±0.88 mg/L) and the parameter correlated positively with PO_4 (r = -0.230) at p < 0.01. The findings are similar to Ehiagbonare and Ogunrinde (2010) who reported BOD₅ of 2.2, 2.36 mg/L. In this study, Nitrate and phosphate values ranged from 0.75±0.14 to 3.79±2.39 mg/L and 0.28±0.10 to 4.64±0.77 respectively. These values were lower than those of Ehiagbonare and Ogunrinde (2010) who reported 2.21 - 4.91 and 1.40 - 4.81 mg/L. The values fell within the recommended standards (FEPA, 1999). Phosphate correlated positively with total heterotrophic bacterial count (r=0.532) at p < 0.01. There were significant difference between the

nitrate and phosphate levels in concrete ponds and plastic ponds at (p < 0.05).

Higher nitrates and phosphates in plastic ponds could be attributed to high protein content in feed accumulation of the decomposed feeds.

 Table 2: Mean counts of the microbiological parameters of water from fish ponds in Idogbo community

Sample	THBC	TCC
code	(×10 ³ cfu/g)	(MPN/100ml)
CP1	57.11±26.70 ^a	17.11±4.20 ^{cd}
CP2	38.44±20.501 ^a	12.11±1.27 ^b
CP3	26.11±12.42 ^a	16.89±2.67 ^{cd}
CP4	40.22±12.16 ^a	24.89±3.06e
CP5	41.22±23.62 ^a	32.56 ± 1.42^{fg}
PP1	53.00±26.08 ^a	32.78±5.21 ^{fg}
PP2	105.44±44.75 ^b	28.56±6.71 ^{ef}
PP3	97.11±37.38 ^b	33.78±6.61 ^g
PP4	124.44±59.24 ^b	20.33±8.66 ^d
PP5	94.78±2.59 ^b	13.00±2.40 ^{bc}

Values are Mean \pm SD of triplicates. Different superscripts in the same column indicate

Significant differences at p < 0.05 according to Duncan Multiple Range Test (DMRT)

THBC: total heterotrophic bacteria count and TCC: Total coliform count; CP=concrete pond; PP: plastic pond

The result of the bacteriological evaluation of water sampled from the respective fish ponds is shown in Table 2 and 3. The mean heterotrophic bacterial counts ranged from 26.11 ± 12.42 to 124.44 ± 59.24 (×10³ cfu/g). There were significant difference between the total bacterial counts in the plastic ponds and concrete ponds (P < 0.05). The total bacterial count observed in this study was high and this trend is similar to that of Ahmed and Naim (2003) who reported a range of 10^3 to 10^4 cfu/ml.

Table 3: Bacterial genera isolated from the fish pond water samples

Bacterial isolates	Description
Pseudomonas aureginosa	Gram negative, aerobic, motile, curved single rod, non-lactose fermenter, catalase and oxidase positive.
Klebsiella pneumonia	Gram negative, facultative anaerobic rod shaped lactose fermenter but oxidase and indole negative.
Proteus vulgaris	Gram negative, facultative anaerobes, highly motile and urease positive.
Bacillus sp.	Gram positive, catalase positive, aerobic spore-forming (as indicated by sliminess) rods
Staphylococcus aureus	Spherical, gram-positive, aerobic bacteria in clusters, oxidase and indole negative.
Escherichia coli	Gram negative, aerobic rod, heavy lactose fermenter, does not utilize citrate, positive for indole test
Serratia sp.	Gram negative, non-motile, citrate positive, oxidase and indole negative

	Table 4: Association among soil	hysicochemical and microbial	parameters in po	ond water from Idogbo community
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Parameter	pН	EC	TDS	DO	BOD ₅	NO ⁻ 3	PO ⁻ 4	THBC
pH	1.000							
EC	0.721**	1.000						
TDS	0.528^{**}	0.706^{**}	1.000					
DO	-0.392**	-0.385**	-0.324**	1.000				
BOD	0.689^{**}	0.687^{**}	0.487^{**}	-0.310**	1.000			
NO ⁻ 3	-0.174	-0.065	0.213*	0.125	-0.067	1.000		
PO ⁻ 4	0.531**	0.855**	0.616**	-0.230*	0.587^{**}	-0.019	1.000	
THBC	0.202^{*}	0.429**	0.309**	-0.116	0.153	-0.117	0.532**	1.000

**. Correlation is significant at the 0.01 level (2-tailed).

*. Correlation is significant at the 0.05 level (2-tailed).Temp: Temperature; EC electrical conductivity; TDS total dissolve solids; DO: Dissolved

oxygen; BOD₅ biological oxygen demand; NO $\frac{1}{3}$: nitrate; PO $\frac{1}{4}$ Phosphate, THBC: Total heterotrophic bacterial count

The high bacterial counts could be attributed to the presence of organic matter remains from the decomposition of the feed within the pond water. It could also be due to the optimum temperature conditions of the pond which can aid the growth of bacterial commensals and pathogens (Eze and Ogbaran, 2010). The bacterial isolates cultured from the pond water samples include; Pseudomonas aureginosa, Klebsiella pneumonia, Proteus vulgaris, Bacillus sp., Staphylococcus aureus Escherichia coli and Serratia sp. (Table 3). Majority of these bacteria have been reported in related studies by Ahmed and Naim (2006); Torimiro et al. (2014) and Dabbor (2008). Total coliform counts ranged from 12.11±1.27 to 33.78±6.61 MPN/100 ml. The isolation of coliforms in this study was an indication of fecal contamination of the fish pond water. This could be as a result of augmentation of the ponds with animal manure discharged directly into the fish ponds or through runoff (Kay et al., 2008).

Conclusion

The physicochemical parameters assessed in this current study were all within regulatory standards. This observation would indicate that the pond water were good for fish production. The mean physicochemical values of the pond waters were significantly higher in the plastic ponds than those of concrete ponds except temperature which was not significantly different for both pond types. The study also revealed the presence of high microbial load enhanced by the optimum growth conditions and the fertilization of the ponds with animal manure.

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Conflict of Interest

Authors declare that there is no conflict of interest related to this study.

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