Assessment of faecal contamination in selected concrete and earthen ponds stocked with *Clarias gariepinus*

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Research Article

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Abstract

Background

This study was carried out to monitor the levels of faecal pollution markers in catfish (*Clarias gariepinus*) and their growing waters in selected earthen and concrete ponds. Water and catfish samples were collected weekly in the months of February, March, April, May, June and July, 2019. The concentrations of enteric bacteria in the water and catfish samples were determined using membrane filtration and pour plate methods, respectively. The rate of bioaccumulation of faecal indicator bacteria was obtained by dividing the log concentration of each organism in catfish by the corresponding log concentration in the growing waters.

Result

The concentration of faecal coliforms in catfish samples from concrete and earthen ponds ranged from 1.41 to 2.28 $\log_{10}$ CFU/100 ml and 1.3 to 2.47 $\log_{10}$ CFU/100 ml respectively and in growing waters from the concrete and earthen ponds; 1.43 to 2.41 $\log_{10}$ CFU/100 ml and 1.50 to 2.80 $\log_{10}$ CFU/100 ml respectively. Faecal coliforms exhibited positive relationships with alkalinity in water samples from the earthen (r = 0.61) and concrete ponds (r = 0.62). *Salmonella* and faecal coliforms had the highest and least bioaccumulation in catfish raised in earthen pond, respectively, whereas *Salmonella* and enterococci had the highest and least bioaccumulation in catfish raised in concrete pond, respectively. Faecal coliforms and *E. coli* had the highest and least counts in water samples from the earthen pond during the dry and wet months, *Salmonella* and *E. coli* had the highest and least counts in water samples from the concrete pond during the dry and wet months.

Conclusion

There were high levels of bacterial faecal pollution markers in water and *C. gariepinus* from the earthen and concrete ponds. Physicochemical characteristics of the water and seasonality played major roles in the rate of bioaccumulation of the faecal pollution markers in *C. gariepinus* raised in the earthen and concrete ponds.

Highlights

- The concentration of faecal indicator bacteria (FIB) in *Clarias gariepinus* and their growing waters
- Distribution pattern of enteric bacteria in *Clarias gariepinus* and their growing waters from the earthen and concrete ponds
- The physicochemical characteristics of the growing waters (concrete and earthen ponds)
- The relationship between FIB and the physicochemical characteristics of the growing waters

- Rates of bioaccumulation of FIB in *Clarias gariepinus*
- Effect of different seasons on the bioaccumulation rates

Background

Clarias gariepinus is one of the most studied tropical catfish within the genus *Clarias* (Okaeme, 2006). Aquaculture has gained much attention as a fast growing sector of global food production and source of animal protein in the world today. Microorganisms contribute a significant fraction of importance in the aquatic ecosystem and they have been observed to be among the factors that can cause the emergence of infectious diseases in aquacultural practices (Ikpi and Offem, 2011; Noga, 2000). The prevalence of infectious diseases has been observed to depend on the interaction between fish pathogens and the aquatic environment (Noga, 2000). Hence, the need to quantify and monitor microbial population in this sector. The African catfish has been introduced in at least 37 countries of Africa, Europe, Asia and America mainly for aquaculture with economic impacts on freshwater and brackish ecosystems (Krishnakumar et al., 2011; Weyl et al., 2016). In the Caribbean, the species has been introduced for aquaculture in Cuba (Kubota et al., 2012). Members of the genus *Clarias* are among the world's worst invasive species (Lowe, 2004) and *C. gariepinus* has developed important adaptations for surviving in unsuitable environments with low oxygen levels and long period of desiccation (Bruton, 1979; Donnelly, 1973; Opasola et al., 2013).

The bacteriological profile of *Clarias gariepinus* reported by Ajayi (2012) and Omeji et al. (2013) indicated that, water contaminated with bacteria (faecal coliforms, faecal streptococci and *Salmonella* spp.) might infect fish in ponds. Studies have shown that fish or the aquatic environments may serve as sources of pathogenic microorganisms that may be transmitted to humans exposed to the untreated water or undercooked fish (Novotny *et al.*, 2018). In earthen ponds, bacteria may be found on the surface of different types of decaying material such as leaves, metallic objects, rocks or wood. The distribution of heterotrophic bacteria and total aquatic bacteria vary with the water layers in the earthen pond. There are different factors affecting the circulation of bacteria in fish pond which comprises predatory protozoa present in water (Ganesh et al., 2010). Bacteriological analysis of fish pond water is very important in aquaculture as this gives insight to the likely hazards that may occur to the fishes, the farmer and the consumer (Ganesh et al., 2010). Agricultural practices such as allowing livestock to graze near water bodies, spreading manure as fertilizer on fields during wet periods, using sewage sludge bio-solids and allowing livestock watering in streams may result to faecal contamination in ponds (Hinks et al., 2016). The prevention of excessive faecal contamination of ponds is difficult since most of the contaminations are from non-point sources and many extended sources (Taiwo, 2011). In some integrated ponds, faeces from poultry birds are directly released into ponds or stored in a tank to allow the development of maggots which are then discharged into the ponds.

Studies have revealed that fish and their aquatic environment can harbor microorganisms, especially members of the coliform group. Moreover, faecally-contaminated water from ponds when discharged into other water bodies poses a significant risk to human health (Pinfold, 2009; WHO, 2005). This study set out to monitor the levels of faecal pollution markers in catfish (*Clarias gariepinus*) and their growing waters in selected earthen and concrete ponds. The objectives of the study were to determine the concentration of faecal indicator bacteria (FIB) in the catfish and their growing waters; assess the physicochemical characteristics of the growing waters; examine the relationship between FIB and the physicochemical characteristics of the growing waters; investigate the rates of bioaccumulation of FIB in the catfish and examine the effect of different seasons on the bioaccumulation rates.

Methods

Study design

This is an experimental study conducted between February to July, 2019.

Study area

The study area is Federal University of Technology, Akure (FUTA) Teaching and Research Fish Farm in Obakekere campus. It is located between latitude 7° and 8° N and longitude 5° and 18° E with elevation of approximately 30.5 metres (100 feet) above sea level (**Fig. 1**). It contains characteristic swamp, water ways, vast plains and rain forest. The farm combine fish production and livestock production, they also operate integrated fish production having different concrete and earthen ponds stocked with tilapia, catfish and other species of fish. The earthen and concrete ponds were selected as the sampling site in this study because of the frequent usage of the ponds. The ponds were also prone to faecal contamination from farm animals grazing around the pond and also the landscape encourages direct flow of erosion into the earthen pond during wet season.

Collection of catfish and their growing water samples from earthen and concrete pond

Water and catfish samples were collected weekly from February to July, 2019. On each sampling occasion, water and catfish samples were collected from the earthen and concrete ponds. The water samples were collected aseptically with sterile 800 ml screw-capped bottles labeled appropriately and the catfish samples were collected with conventional scoop net and thereafter placed in a sterile polythene bag with appropriate labeling. In total, 80 grabs of water and 120 catfish samples were collected over the study period. All samples were transported to the laboratory within 1 hr.

Enumeration of enteric bacteria in catfish and their growing water samples

The concentrations of *Escherichia coli*, faecal coliforms, *Salmonella, Shigella* and intestinal enterococci in the catfish and their growing waters were determined using standard microbiological methods as described by Maheux *et al.* (2009). Preparation of the catfish samples was carried out by dissecting the intestinal tract of the fish using a sterile knife and measuring 1 g into a sterile mortar. This was macerated with about 4 ml of sterile distilled water and 1 ml aliquot was taken into a sterile test tube containing 9 ml of sterile distilled water resulting into 1:10 dilution. Serial dilution was further carried out until the fifth dilution. Using membrane filters (0.45 µm), the concentrations of the bacteria were determined by placing the filters on freshly prepared selective media: Membrane lauryl sulphate agar (MLSA), Eosin methylene blue (EMB), Membrane faecal coliform agar (*m*-FC), Membrane intestinal enterococci agar (*m*-EA) and *Salmonella-Shigella* agar (SSA). Agar plates were incubated at 37 °C for 24 hours (MLSA, EMB and SSA), 44 °C for 24 hours (*m*-FC) and 37 °C for 48 hours (*m*-EA). Colonies were counted recorded and expressed as colony forming unit (CFU) per 100 ml of water or CFU per 100 g of catfish utilizing a colony counter (J-2 PEC MEDICAL, New Jersey, USA).

Determination of the physicochemical characteristics of the growing waters in the earthen and concrete ponds

The temperature of the growing water was determined on-site during sample collection using mercury-in-glass thermometer (ACCU-SAFE Thomas Scientific, New Jersey, US). The pH, electrical conductivity, salinity, total dissolved solids, turbidity and dissolved oxygen of the water samples were determined using a multi-parameter analyzer (HI98194, PH/ORP/EC/DO). The biological oxygen demand (BOD) was determined via the Winkler's method. The chemical oxygen demand (COD) of the water samples was determined via the method described by Kolb *et al.* (2017).

Bioaccumulation of enteric bacteria in catfish samples from the earthen and concrete ponds

The rate of bioaccumulation of faecal indicator bacteria was obtained by dividing the log concentration of each organism in catfish by the corresponding log concentration in the growing waters at the same point in time.

Statistical analysis

Data were transformed to $\log_{10}$, and then examined using general descriptive statistics. The normality and distribution pattern of enteric bacteria in the catfish and their growing waters in concrete and earthen ponds were determined using Kolmogorov-Smirnov and Shapiro-Wilk statistic. Further analyses were carried out via one-way analysis of variance (ANOVA) with significance at $p>0.05$ using GraphPad Prism version 5.0 (mean +/- standard error) and (mean +/- standard deviation) for concentration of faecal indicator bacteria and physicochemical characteristics of growing water in concrete and earthen ponds respectively. The relationships between the concentrations of enteric bacteria and those of the physicochemical properties of the growing waters from the earthen and concrete ponds were analyzed using Pearson's correlation coefficient at $p \leq 0.05$ level of significance.

Results

Concentration of enteric bacteria in catfish samples and their growing waters in the earthen and concrete pond

Result revealed that the concentration of faecal coliforms in catfish samples from the concrete and earthen ponds ranged from 1.41 to 2.28 $\log_{10}$ CFU/100 ml and 1.3 to 2.47 $\log_{10}$ CFU/100 ml respectively, while in the growing water samples from the concrete and earthen ponds, it ranged from 1.43 to 2.41 $\log_{10}$ CFU/100 ml and 1.50 to 2.80 $\log_{10}$ CFU/100 ml respectively (Fig. 2). The concentration of *Salmonella* in catfish samples from the concrete and earthen ponds ranged from 1.52 to 2.56 $\log_{10}$ CFU/100 ml and 1.60 to 2.70 $\log_{10}$ CFU/100 ml respectively, while in the growing water samples from the concrete and earthen ponds, it ranged from 1.51 to 2.56 $\log_{10}$ CFU/100 ml and 1.60 to 2.70 $\log_{10}$ CFU/100 ml respectively, (Fig. 3). The concentration of *Escherichia coli* in catfish samples from the concrete and earthen ponds ranged from 1.23 to 2.44 $\log_{10}$ CFU/100 ml and 1.30 to 2.30 $\log_{10}$ CFU/100 ml respectively, while the concentration of *Escherichia coli* in the growing water samples from the concrete and earthen ponds ranged from 1.30 to 2.1 $\log_{10}$ CFU/100 ml and 1.20 to 1.53 $\log_{10}$ CFU/100 ml respectively (Fig. 4). The concentration of *Shigella* in catfish samples from the concrete and earthen ponds ranged from 1.0 to 1.2 $\log_{10}$ CFU/100 ml and 1.40 to 1.50 $\log_{10}$ CFU/100 ml respectively, while the concentration of *Shigella* in the growing water samples from the concrete and earthen ponds ranged from 1.46 to 1.60 $\log_{10}$ CFU/100 ml and 1.40 to 1.50 $\log_{10}$ CFU/100 ml respectively (Fig. 5). The concentration of intestinal enterococci in catfish samples from the concrete and earthen ponds ranged from 1.30 to 1.40 $\log_{10}$ CFU/100 ml and 1.30 to 1.70 $\log_{10}$ CFU/100 ml respectively while in the growing water samples in the concrete and earthen ponds, it ranged from 1.40 to 1.50 $\log_{10}$ CFU/100 ml and 1.32 to 1.82 $\log_{10}$ CFU/100 ml respectively (Fig. 6).

Distribution pattern of enteric bacteria in catfish and their growing waters from the earthen and concrete ponds

Results revealed that faecal coliforms (Sig. = 0.757) and *Salmonella* (Sig. = 0.843) in catfish samples from the earthen pond; were normally distributed while enterococci (Sig. = 0.001), *Shigella* (Sig. = 0.029) and *E. coli* (Sig. = 0.028) were not normally distributed. Furthermore, *Salmonella* (Sig. = 0.761) and *Shigella* (Sig. = 0.761) in water samples from the earthen pond, were normally distributed while faecal coliforms (Sig. = 0.000), enterococci (Sig. = 0.038) and *E. coli* (Sig. = 0.038) were not normally distributed (Table 1).

Table 1
Normality and distribution pattern of enteric bacteria in *Clarias gariepinus* and their growing water in earthen pond

Tests of Normality						
	Kolmogorov-Smirnov[a]			Shapiro-Wilk		
CATFISH SAMPLES	Statistic	Df	Sig.	Statistic	Df	Sig.
Faecal coliforms	0.130	20	0.200*	0.970	20	0.757
Salmonella	0.120	20	0.200*	0.974	20	0.843
Enterococci	0.258	20	0.001	0.795	20	0.001
Shigella	0.197	20	0.040	0.891	20	0.029
E. coli	0.141	20	0.200*	0.891	20	0.028
WATER SAMPLES Faecal coliforms	0.345	20	0	0.717	20	0
Salmonella	0.096	20	0.200*	0.970	20	0.761
Enterococci	0.208	20	0.024	0.898	20	0.038
Shigella	0.096	20	0.200*	0.970	20	0.761
E. coli	0.208	20	0.024	0.898	20	0.038
Keys: Df-Difference; Sig.-Significance						

On the other hand, faecal coliforms (Sig. = 0.527) and *Salmonella* (Sig. = 0.643) in catfish samples from the concrete pond were normally distributed while enterococci (Sig. = 0.011), *Shigella* (Sig. = 0.029) and *E. coli* (Sig. = 0.018) significantly deviated from normal distribution. In addition, *Salmonella* (Sig. = 0.461) and *Shigella* (Sig. = 0.751) in water samples from the concrete pond were normally distributed while faecal coliforms (Sig. = 0.000), enterococci (Sig. = 0.038) and *E. coli* (Sig. = 0.028) significantly deviated from normal distribution (Table 2).

Table 2
Normality and distribution pattern of enteric bacteria in *Clarias gariepinus* and their growing water in concrete pond

Tests of Normality						
	Kolmogorov-Smirnov[a]			Shapiro-Wilk		
CATFISH SAMPLES	Statistic	Df	Sig.	Statistic	Df	Sig.
Faecal coliforms	0.130	20	0.200*	0.970	20	0.757
Salmonella	0.120	20	0.200*	0.974	20	0.843
Enterococci	0.258	20	0.001	0.795	20	0.001
Shigella	0.197	20	0.040	0.891	20	0.029
E. coli	0.141	20	0.200*	0.891	20	0.028
WATER SAMPLES Faecal coliforms	0.345	20	0	0.717	20	0
Salmonella	0.096	20	0.200*	0.970	20	0.761
Enterococci	0.208	20	0.024	0.898	20	0.038
Shigella	0.096	20	0.200*	0.970	20	0.761
E. coli	0.208	20	0.024	0.898	20	0.038
Keys: Df-Difference; Sig.-Significance						

Physicochemical characteristics of the growing waters in the earthen and concrete ponds

The mean values of temperature of the growing waters in the earthen and concrete ponds were 26.7±0.28°C and 29.3±0.35°C respectively (Table 3) while those of pH of the growing waters in the earthen and concrete ponds were 7.0±0.19 and 6.95±0.07 respectively (Table 3). Similarly, the mean values of electrical conductivity of the growing waters in the concrete ponds were 20.8±1.06 and 26.8±0.35 µS/cm respectively (Table 3). The mean values of salinity of the growing waters in the earthen and concrete ponds were 149.3±1.06 and 148.0±0.71 PSU (practical salinity unit) respectively. Additionally, the mean values of turbidity of the growing waters in the earthen and concrete ponds were 43.5±0.25 and 44.3±0.29 (NTU) (Nephelometric turbidity unit) respectively whereas those of total dissolved solids of the growing waters in the earthen and concrete ponds were 23.5±0.38 and 22.3±0.35 mg/L respectively. The mean values of alkalinity levels of the growing waters in the earthen and concrete ponds were 109.8±2.89 and 107.8±3.89 mEg/L respectively. Similarly, dissolved oxygen (DO) in the growing waters of the earthen and concrete ponds was 11.95±2.13 and 4.6±6.29 mg/l respectively. The mean values of the biological oxygen demand (BOD) of the growing waters in the earthen and concrete ponds were 2.53±1.23 and 1.53±0.03 mg/L respectively, whereas that of chemical oxygen demand (COD) was 199.6±2.55 and 202.5±3.54 mg/L respectively (Table 3).

Table 3
Physicochemical characteristics of growing water samples in earthen and concrete ponds

Parameters	Earthen pond	Concrete pond
Temperature (°C)	26.7±0.28	29.3±0.35
pH	7.0±0.19	6.95±0.07
EC (μs/cm)	20.8±1.06	26.8±0.35
Salinity (PSU)	149.3±1.06	148.0±0.71
Turbidity (NTU)	43.5±0.25	44.3±0.29
TDS (mg/L)	23.5±0.38	22.3±0.35
Alkalinity (mEg/L)	109.8±2.89	107.8±3.89
DO (mg/L)	11.95±2.13	14.6±6.29
BOD (mg/L)	2.53±1.23	1.53±0.03
COD (mg/L)	199.6±2.55	202.5±3.54

Values presented are expressed as mean values ± standard deviation (n=6). Temp – Temperature; EC – Electrical conductivity; TDS – Total dissolved solids; DO –Dissolved oxygen; BOD-Biological oxygen demand; COD-Chemical oxygen demand.

Relationship between enteric bacteria in catfish samples and physicochemical characteristics of the growing waters in the earthen and concrete ponds

In the earthen pond, alkalinity exhibited positive relationships with enterococci (r = 0.600, $p < 0.05$), faecal coliforms (r = 0.610, $p < 0.05$) and *E. coli* (r = 0.650, $p < 0.01$). Total dissolved solids had a positive correlation with *E. coli* (r = 0.51, $p < 0.05$) and turbidity showed a positive relationship with faecal coliforms (r = 0.54, $p < 0.05$). Biological oxygen demand had positive correlations with enterococci (r = 0.52, $p < 0.05$) and *E. coli* (r = 0.75, $p < 0.01$). Chemical oxygen demand also demonstrated positive relationships with enterococci (r = 0.500, $p < 0.05$), faecal coliforms (r = 0.530, $p < 0.05$) and *E. coli* (r = 0.750, $p < 0.01$) (Table 4).

Table 4
Correlation between enteric bacteria and physicochemical characteristics of growing waters in earthen pond

Physicochemical parameters	**Enterococci**	**Faecal coliforms**	***Escherichia coli***	***Salmonella***	***Shigella***
Temp (°C)	0.03	0.04	0.08	-0.10	-0.05
pH	-0.32	0.08	0.05	-0.28	-0.29
EC (µs/cm)	0.05	0.43	0.08	0.48	0.49
Turbidity (NTU)	0.10	**0.54***	0.34	0.29	0.22
Alkalinity (mEg/L)	**0.60**	**0.61**	**0.65**	-0.18	-0.08
DO (mg/l)	-0.10	-0.13	0.29	-0.02	-0.01
Salinity (PSU)	0.31	-0.03	0.30	0.16	0.16
TDS(mg/l)	0.34	0.11	**0.51***	-0.14	-0.14
BOD (mg/L)	**0.52***	0.44	**0.75**	-0.18	-0.18
COD (mg/L)	**0.50***	**0.53***	**0.73**	-0.14	-0.14
*Correlation is significant at the 0.05 level (2-tailed)**					
*Correlation is significant at the 0.01 level (2-tailed) ***					
Key: Temp (Temperature); EC (Electrical conductivity); DO (Dissolved oxygen); TDS (Total dissolved solids); BOD (Biological oxygen demand); COD (Chemical oxygen demand). Values in bold figures indicate significant correlation					

In the concrete pond, alkalinity exhibited positive relationships with faecal coliforms ($r = 0.620$, $p < 0.01$) and *E. coli* ($r = 0.600$, $p < 0.01$). Total dissolved solids had a positive correlation with *E. coli* ($r = 0.650$, $p < 0.05$). Biological oxygen demand had positive correlations with enterococci ($r = 0.580$, $p < 0.05$) and *E. coli* ($r = 0.820$, $p < 0.01$). Chemical oxygen demand exhibited positive correlations with enterococci ($r = 0.540$, $p < 0.05$) and faecal coliforms ($r = 0.580$, $p < 0.05$). Salinity showed positive correlations with enterococci ($r = 0.51$, $p < 0.05$) and *E. coli* ($r = 0.50$, $p < 0.05$) (Table 5).

Table 5
Correlation between enteric bacteria and physicochemical characteristics of growing waters in concrete pond

Physicochemical parameters	Enterococci	Faecal coliforms	*Escherichia coli*	*Salmonella*	*Shigella*
Temp (^oC)	-0.11	-0.20	-0.22	-0.11	-0.12
pH	-0.42	-0.08	-0.15	-0.25	-0.25
EC (µs/cm)	-0.05	0.33	0.04	0.29	0.29
Turbidity (NTU)	0.10	0.37	0.21	0.19	0.19
Alkalinity (mEg/L)	0.45	**0.62**	**0.60**	-0.28	-0.28
DO (mg/l)	-0.16	-0.23	0.19	-0.04	-0.04
Salinity (PSU)	**0.51***	0.03	**0.50***	0.36	0.26
TDS (mg/l)	0.44	0.21	**0.65**	0.18	-0.10
BOD (mg/l)	**0.58***	0.49	**0.82**	0.19	-0.12
COD (mg/l)	**0.54***	**0.53***	**0.77**	0.04	0.04
*Correlation is significant at the 0.05 level (2-tailed)**					
*Correlation is significant at the 0.01 level (2-tailed) ***					
Key: Temp (Temperature); EC (Electrical conductivity); DO (Dissolved oxygen); TDS (Total dissolved solids); BOD (Biological oxygen demand); COD (Chemical oxygen demand). Values in bold figures indicate significant correlation					

Bioaccumulation of enteric bacteria in catfish samples from the earthen and concrete ponds

The bioaccumulation of enterococci in catfish from their growing waters in earthen and concrete ponds ranged from 0.90 to 2.20 log10 CFU/100 ml and 0.92 to 2.19 log10 CFU/100 ml respectively, moreover, the bioaccumulation of faecal coliforms in catfish from their growing waters in earthen and concrete ponds ranged from 0.79 to 2.00 log10 CFU/100 ml and 0.70 to 2.40 log10 CFU/100 ml respectively. Similarly, the bioaccumulation of *E. coli* in catfish from their growing waters in earthen and concrete ponds ranged from 0.94 to 2.65 log10 CFU/100 ml and 0.00 to 1.89 log10 CFU/100 ml respectively, whereas the bioaccumulation of *Salmonella* in catfish from their growing waters in earthen and concrete ponds ranged from 0.97 to 2.70 log10 CFU/100 ml and 0.95 to 1.75 log10 CFU/100 ml respectively. Additionally, the bioaccumulation of *Shigella* in catfish from their growing waters in earthen and concrete ponds ranged from 0.97 to 1.89 log10 CFU/100 ml and 0.95 to 1.86 log10 CFU/100 ml respectively (Fig. 7).

Effect of seasonality on the bioaccumulation of enteric bacteria in catfish from their growing waters in earthen and concrete ponds

In the earthen pond, the mean values of bioaccumulation of enterococci in catfish samples in dry and wet periods were 0.99 $\log_{10}$ CFU/100 ml and 0.62 $\log_{10}$ CFU/100 ml respectively, while the mean values of bioaccumulation of faecal coliforms in catfish samples in dry and wet periods were 0.50 log10 CFU/100 ml and 0.51 log10 CFU/100 ml respectively. The mean values of bioaccumulation of *E. coli* in catfish samples in dry and wet periods were 1.02 log10 CFU/100ml and 0.89 log10 CFU/100ml respectively. The mean values of bioaccumulation of *Salmonella* in catfish samples in dry and wet periods were 0.60 log10 CFU/100ml and 0.45 log10 CFU/100ml respectively, while the mean values of bioaccumulation of *Shigella* in catfish samples in dry and wet periods were 0.55 log10 CFU/100ml and 0.54 log10 CFU/100ml respectively. *E. coli* had the highest mean value of bioaccumulation during the dry period, while *Shigella* had the highest mean value of bioaccumulation during the wet period (Fig. 8).

In the concrete pond, the mean values of bioaccumulation of enterococci in catfish samples in dry and wet periods were 0.5 log10 CFU/100ml and 0.93 log10 CFU/100ml respectively, while the mean values of bioaccumulation of faecal coliforms in catfish samples in dry and wet periods were 0.85 log10 CFU/100ml and 1.06 log10 CFU/100ml respectively. The mean values of bioaccumulation of *E. coli* in catfish samples in dry and wet periods were 0.21 log10 CFU/100ml and 1.07 log10 CFU/100ml respectively. The mean values of bioaccumulation of *Salmonella* in catfish samples in dry and wet periods were 0.52 log10 CFU/100ml and 0.80 log10 CFU/100ml respectively, while the mean values of bioaccumulation of *Shigella* in catfish samples in dry and wet periods were 1.77 log10 CFU/100ml and 1.07 log10 CFU/100ml respectively (Fig. 9).

Discussion

The assessment of faecal contamination in selected concrete and earthen ponds stocked with *Clarias gariepinus* in a fish farm was investigated in this study. The load of enteric bacteria in water and the catfish samples were in agreement with Njoku, (2015) who observed that load of heterotrophic bacteria in the pond water fluctuated between 0.01 and 8.7×10^5 CFU/ml. The level of faecal contamination in the earthen pond was higher than the concrete pond and this may likely be as a result of faecal coliforms generally consisting part of *Enterobacteriaceae* group that thrive at temperature ranges of between 26 ºC and 29 ºC observed in earthen and concrete ponds respectively in this study. Furthermore, increased rainfall and subsequent pollution from both point and diffuse sources may have also contributed to the high levels of faecal coliforms observed in the earthen pond (Wilkes et al., 2013). This is in agreement with the studies conducted by Ajayi and Okoh, (2014) where the authors established that microbial load is usually higher in earthen pond as a result of the natural nutrients in soils which enhances microbial growth. It was noted that the load of faecal coliforms in catfish samples and their growing waters from the earthen and concrete pond were high and this may be because of increased temperature (26-30°C) of the growing water and increased dissolved nutrient in the growing water after feeding of the catfish. This is similar to the findings of Wyatt et al. (2006) in which the increased temperature and dissolved nutrients in catfish growing waters influenced the load of faecal coliforms in the catfish samples and their growing waters. In addition, the load of faecal coliforms in catfish samples and their growing waters that were higher in earthen pond compared to concrete pond may be attributed to the similarity of the earthen pond to the natural habitat of catfish thereby providing vital nutrients and minerals that might support microbial growth better than the concrete pond. This is in conformity with the findings of Njoku et al. (2015) where the authors attributed high load of faecal coliforms in earthen pond growing water samples to its natural capability to support microbial growth.

The load of intestinal enterococci in catfish samples in the earthen and concrete ponds that were high may be as a result of the bacteriological profile of *Clarias gariepinus* reported by Ajayi, (2012) and Omeji et al. (2013) where the authors reported that water contaminated with faecal streptococci have the tendency to infect fish in ponds. The mean counts of *E. coli* in catfish samples and their growing waters that appeared to be high may be due to the poultry manure fed into the pond to serve as food for the *Clarias gariepinus.* The load of *Salmonella* in catfish samples and their growing waters from the earthen and concrete pond was also high as observed in our study. This is related to the outcome of Hudson et al. (2005) and Olalemi, (2018) who both inferred that the indigenous and non-indigenous bacterial pathogens may be associated with fresh fish or their habitat Additionally, *Salmonella* is not a recognized typical bacterial flora of catfish, and the occurrence of this pathogen is normally linked to its breeding, as well as due to inept hygiene practices and inadequate handling (Fernandes *et al.*, 2015) who also affirmed that *Salmonella* can survive in the gastrointestinal tract in fish in a transitory form. The load of *Shigella* in catfish samples and their growing waters from the earthen and concrete pond was also high as observed in our study. The higher load is possibly sustained by amplified nutrient load as a consequence of flood and runoffs into the ponds as supported by Salome and Faith, (2016).

The normal distribution of faecal coliforms and *Salmonella* in catfish samples of earthen and concrete ponds may be as a result of the use of maggots from chicken dungs as feed and these may serve as reservoir for the bacteria, while the deviation of enterococci, *Shigella* and *E. coli* from normal distribution in catfish samples in earthen and concrete ponds may be as a result of change in varying environmental conditions. The outcome of the normal distribution pattern of faecal indicator bacteria in earthen and concrete ponds conducted in this study is in alliance with the observation of Ganesh et al. (2010) who reported the distribution of heterotrophic bacteria and total aquatic bacteria divergence within the water layers which could be linked to various factors affecting the distribution of bacteria in fish pond.

The mean temperature values obtained in this study ranged from 25 – 30°C and this is very similar to the observation of Ntegwu and Edema, (2008). Weather condition during the sampling could have influenced the reading of the water temperature (Pedrazzi et al., 2013). The values of pH were also in agreement with Ehiagbonare and Ogunrinde, (2010) emphasizing that pH between 6 and 9 are basic requirement for increased fish production. Stone and Thomforde, (2003) had also reported that the desirable range for pond pH is 6.5 - 9.5 and acceptable range is 5.5 - 10.0 in order to maintain good pond productivity and fish health. It is interesting to note that the findings from this study is in conformity with Ntengwe and Edema, (2008) where the authors examined the physicochemical and microbiological characteristics of water for fish production using small ponds. Pedrazzi et al. (2013) recorded higher pH values during the wet season, similar with the present study. Climatic condition such as rainfall that occurred during the sampling period influenced the electrical conductivity of the pond water as surface runoff that contained large nutrients during the wet season was probably one of the factors that increased the pond water conductivity (Terra et al., 2010). However, the electrical conductivity values were still within the normal range for fish rearing (10 – 1000 μS/cm) (Offem et al., 2011). Dissolved oxygen condenses as a result of boost in water temperature, respiration and organic matter breakdown by aerobic aquatic organisms (Eze and Ogbaran, 2010). Although, the least amount of dissolved oxygen for tropical fish should be 5 mg/l (Saloom and Duncan 2005), the high levels of dissolved oxygen obtained in this study may be as a result of photosynthetic activities of primary producers that comprise an elevated bio-assortment of plants especially in the earthen pond. The mean alkalinity values of earthen and concrete ponds were 109 and 107 mEg/L within the permissible limit of 200 mEg/L (WHO, 1993). Wurts and Durborow, (1992); Rana and Jain, (2017) reported alkalinity between 75 to 200 mEg/L, but not less than 20 mEg/L is ideal in an aquaculture pond. Swann, (1997); Rana and Jain, (2017) recommended total alkalinity values of at least 20 mEg/L for catfish production and for good pond productivity. The considerably high mean alkalinity of pond water in our study indicates that a minute amount of acid will not elicit any alteration in pH. The mean total dissolved solids (TDS) in this study were below the standard permissible limit of 1000 mg/L (Rana and Jain, 2017). The high BOD diminishes the oxygen level to grave conditions thus indicative of the pollution status of waters. According to Bhatnagar and Singh, (2010); Rana and Jain (2017) only BOD levels of between 3.0-6.0 mg/L is most favorable for usual activities of catfishes as 6.0-12.0 mg/L is toxic to catfishes and >12.0 mg/L could effectively cause their death via suffocation. The present study revealed that the mean values of chemical oxygen demand in the ponds were more than the standard permissible limit of 10 mg/L (WHO 1993; Rana and Jain, 2017).

The bioaccumulation of enteric bacteria excluding *Salmonella* and *Shigella* in *Clarias gariepinus* raised in the concrete pond showed a positive relationship with alkalinity, total dissolved solids; biological oxygen demand, chemical oxygen demand and salinity and a negative relationship with temperature, pH, dissolved oxygen and turbidity. Comparably, in earthen ponds, alkalinity, total dissolved solids, turbidity, biological oxygen demand and chemical oxygen demand exhibited positive relationships with enteric bacteria excluding *Salmonella* and *Shigella* in *C. gariepinus* while a negative relationship was observed with salinity, temperature, pH, electrical conductivity and dissolved oxygen. The positive relationship observed between *E. coli* and total dissolved solids is contrary to the outcome of Olalemi and Oluyemi, (2018) who both divulged the upshot, incidence and existence of faecal pollution markers in an earthen fish pond in Akure, Nigeria. The positive correlation levels exhibited between turbidity and faecal coliforms is parallel to the result of Olalemi and Oluyemi, (2018). *Salmonella* showing a negative relationship with physicochemical parameters in this study was similarly reported by Olalemi and Oluyemi, (2018). Previous studies have revealed that the bioaccumulation progression of microbes in marine animals like *C. gariepinus* is influenced by an array of ecological factors as evident in this study (Ringo and Strom, 1994; Rajiv et al., 2012). The outcome of this study revealed that seasonality played major roles in the rate of bioaccumulation of enteric bacteria in catfish samples from the earthen and concrete ponds.

Conclusions

The findings of this study revealed high levels of faecal pollution in water and *Clarias gariepinus* from the earthen and concrete ponds. Physicochemical characteristics of the pond water and seasonality influenced the rate of bioaccumulation of enteric bacteria in *C. gariepinus* raised in the earthen and concrete ponds.

Declarations

Ethics approval and consent to participate: Membrane filtration techniques conducted in this study were as approved by International Standard Organization (ISO) methods.

Consent for publication: Not applicable

Availability of data and material: There's availability of data and materials in supplementary data files

Competing interests:

Authors declare that no competing financial interests or personal relationship exist.

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Authors' contribution

"AOO designed and supervised the study. OMO developed the methodology, literature, conducted the study, acquired, analyzed and interpreted the data obtained. MTB wrote the first draft. AOO previewed and fine-tuned the draft. All authors read and approved the manuscript".

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Figures

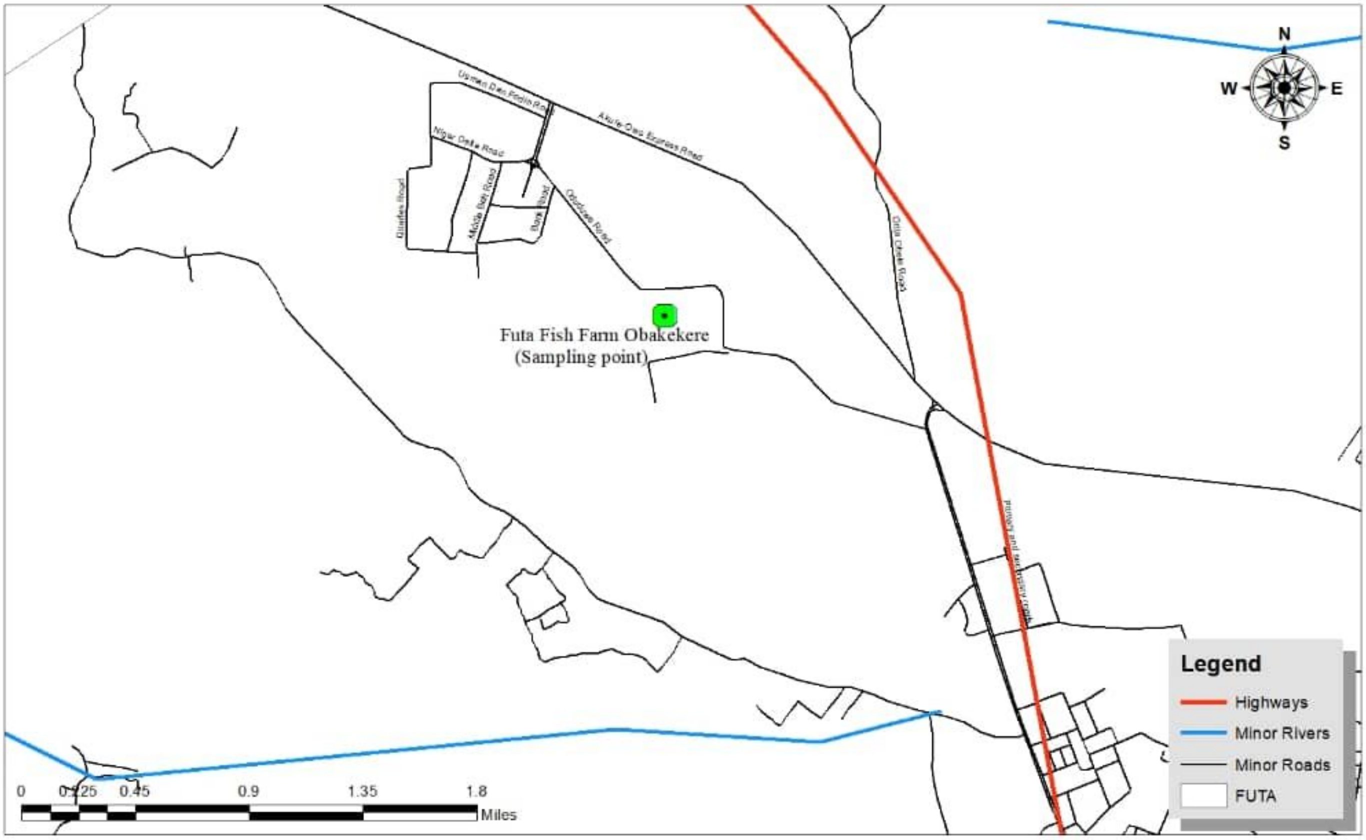

Figure 1

Locality map of FUTA Teaching and Research Fish Farm in Obakekere campus

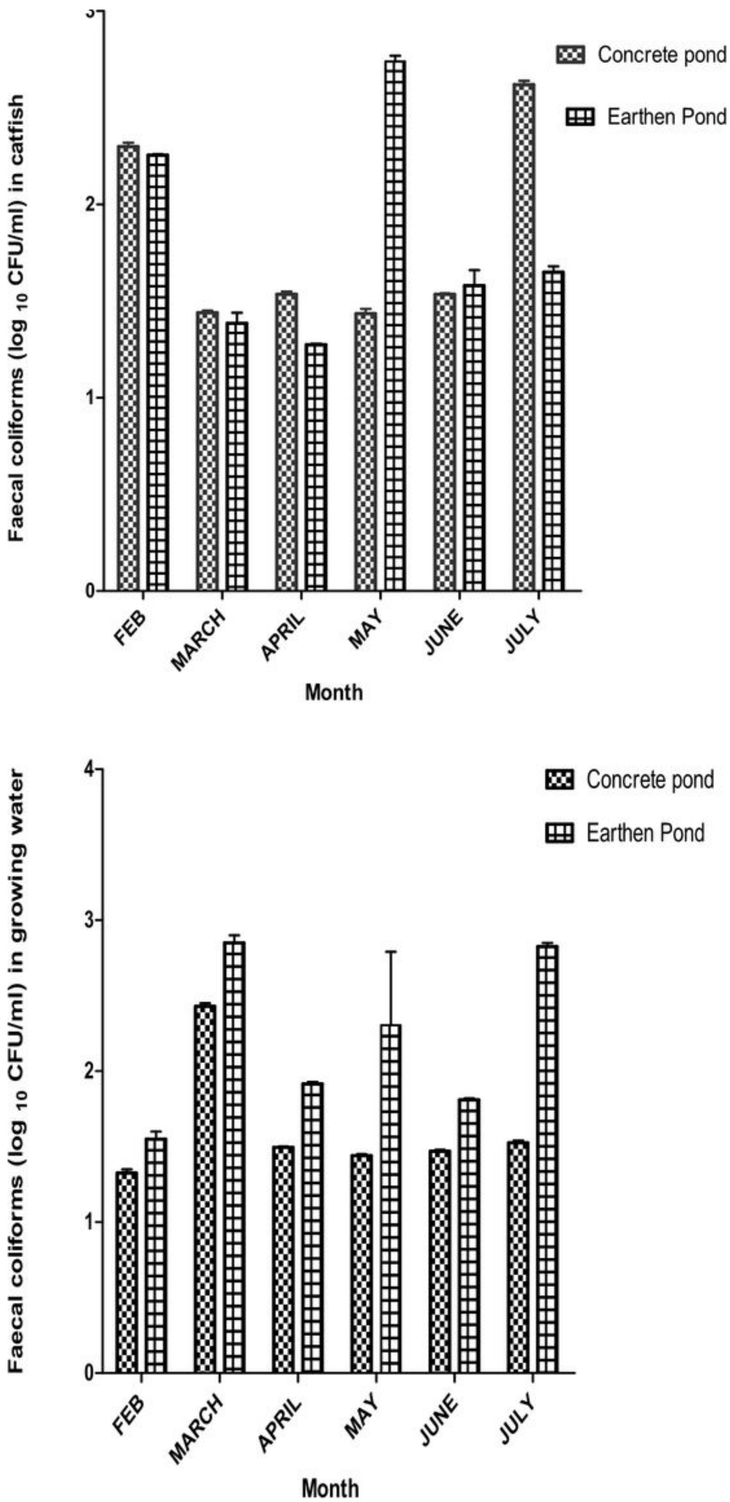

Figure 2

Mean concentration of faecal coliforms in catfish and their growing water samples in concrete and earthen pond. Values are expressed as the (log_{10} CFU/100 ml) mean ± Standard error of mean (SEM)

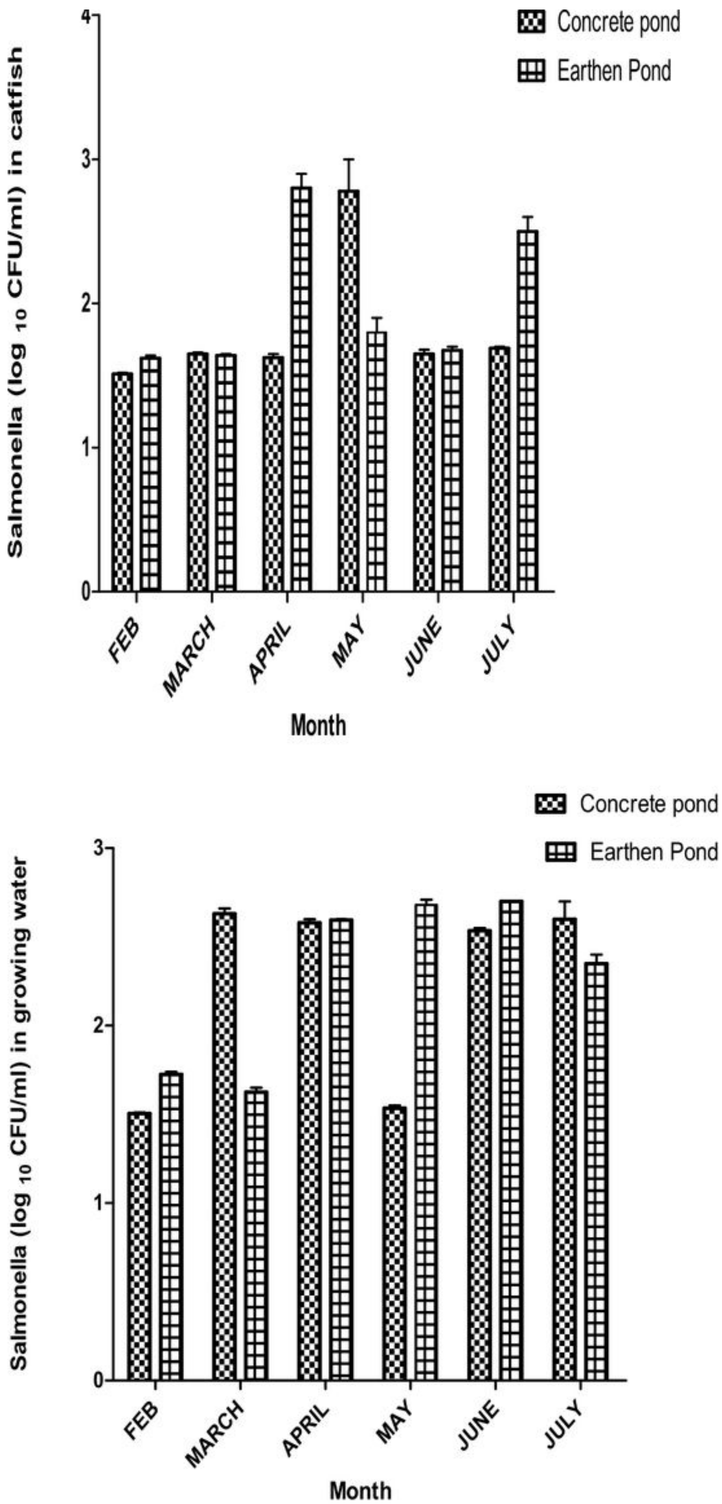

Figure 3

Mean concentration of *Salmonella* in catfish and their growing water samples in concrete and earthen pond. Values are expressed as the (log_{10} CFU/100 ml) mean ± Standard error of mean (SEM)

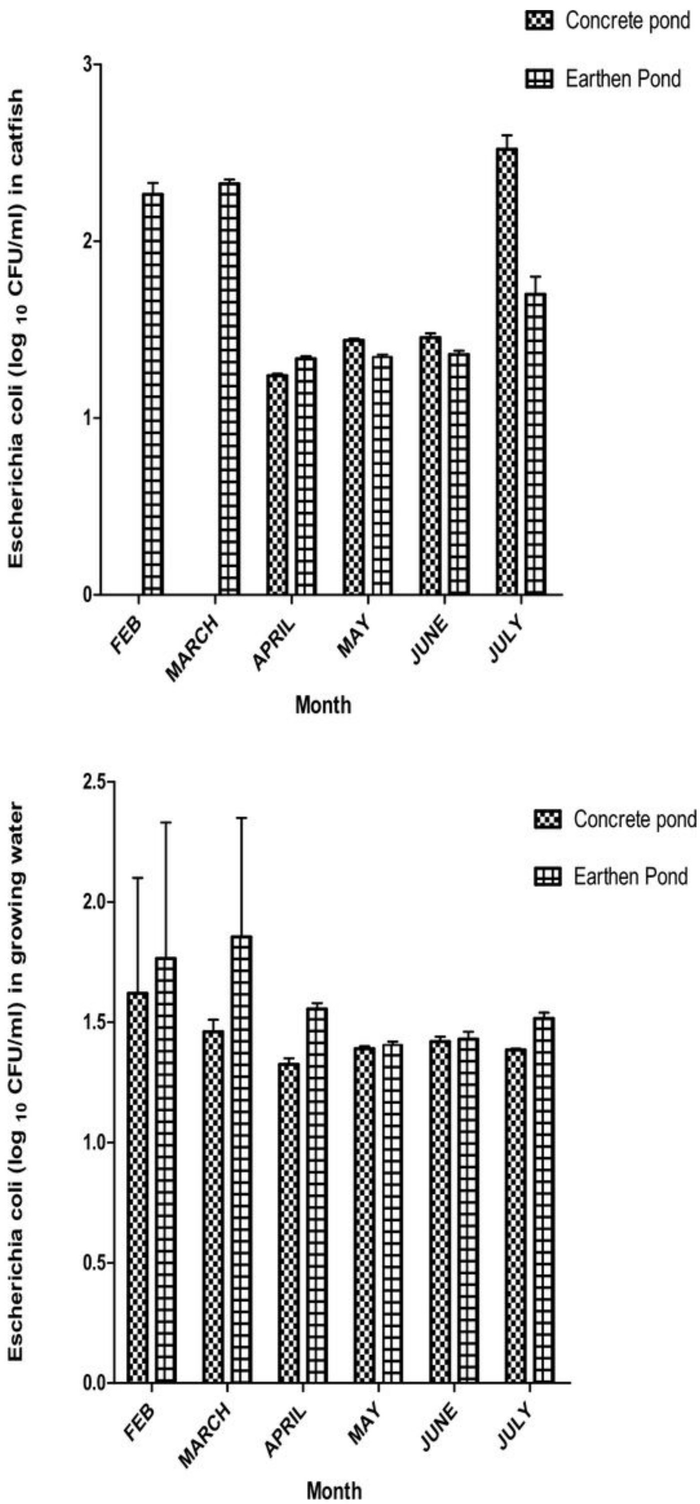

Figure 4

Mean concentration of *E. coli* in catfish and their growing water samples in concrete and earthen pond. Values are expressed as the (log_{10} CFU/100 ml) mean ± Standard error of mean (SEM)

Figure 5

Mean concentration of *Shigella* in catfish and their growing water samples in concrete and earthen pond. Values are expressed as the (log_{10} CFU/100 ml) mean ± Standard error of mean (SEM)

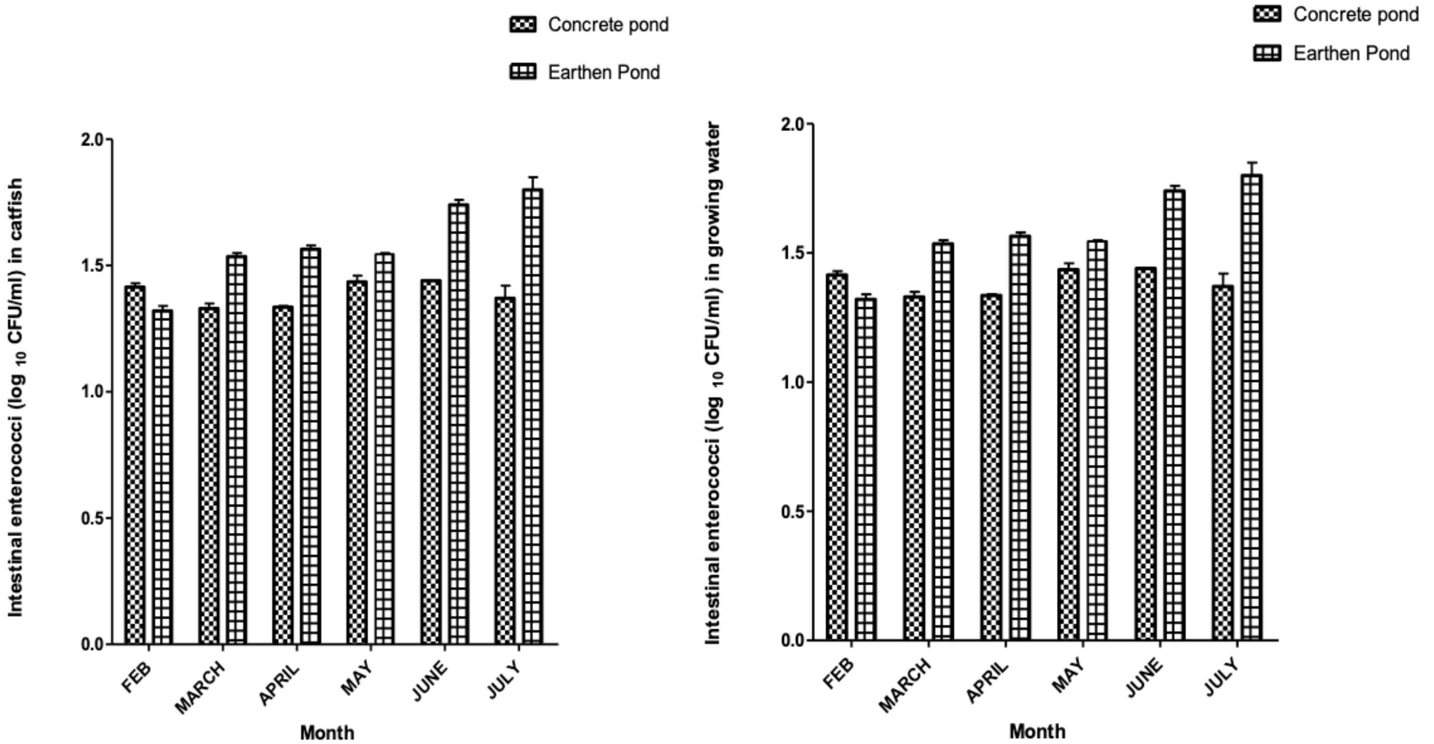

Figure 6

Mean concentration of intestinal enterococci in catfish and their growing water samples in concrete and earthen pond. Values are expressed as the (log_{10} CFU/100 ml) mean ± Standard error of mean (SEM)

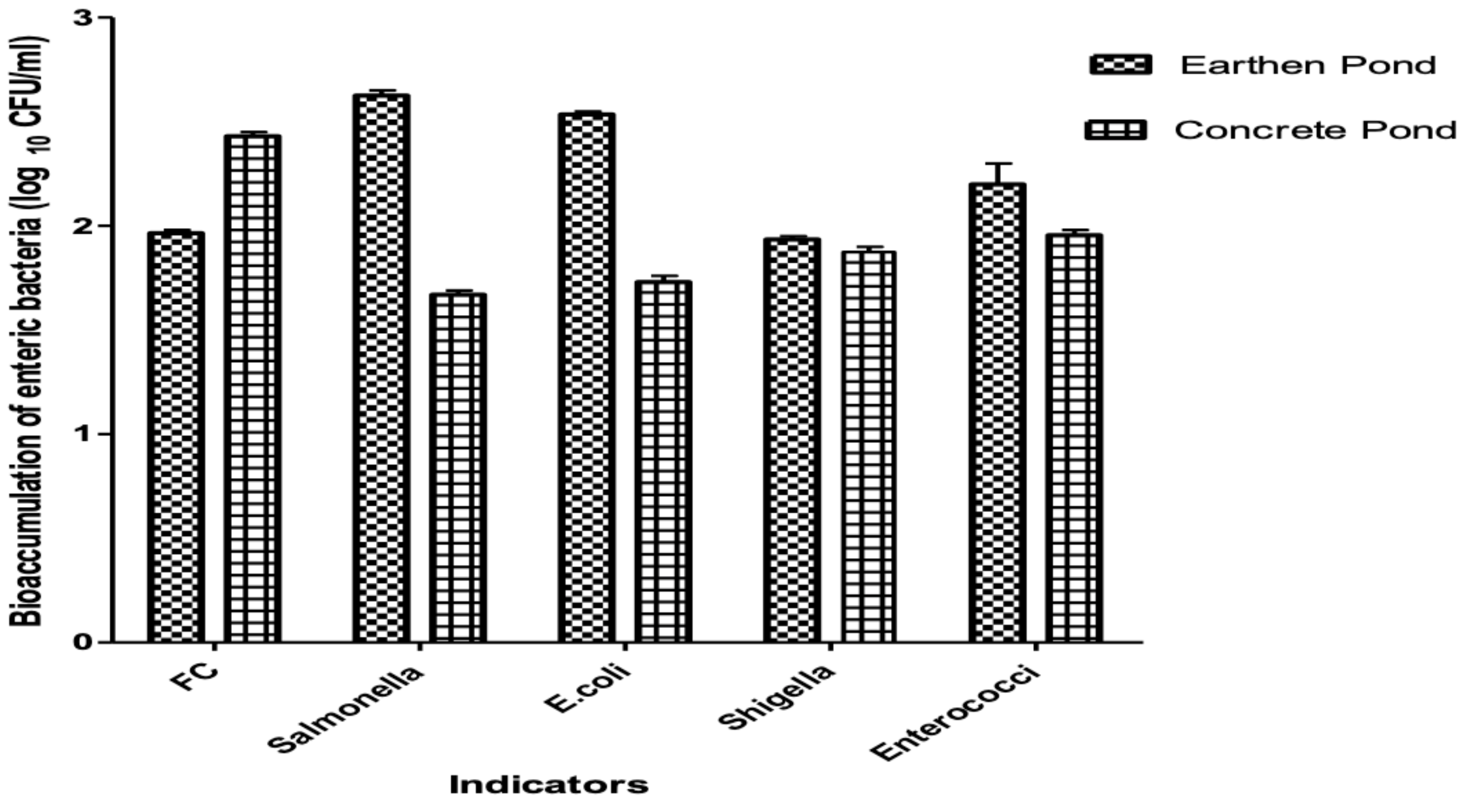

Figure 7

The bioaccumulation of enteric bacteria in catfish from their growing waters in earthen and concrete ponds

Key: FC- Faecal coliforms

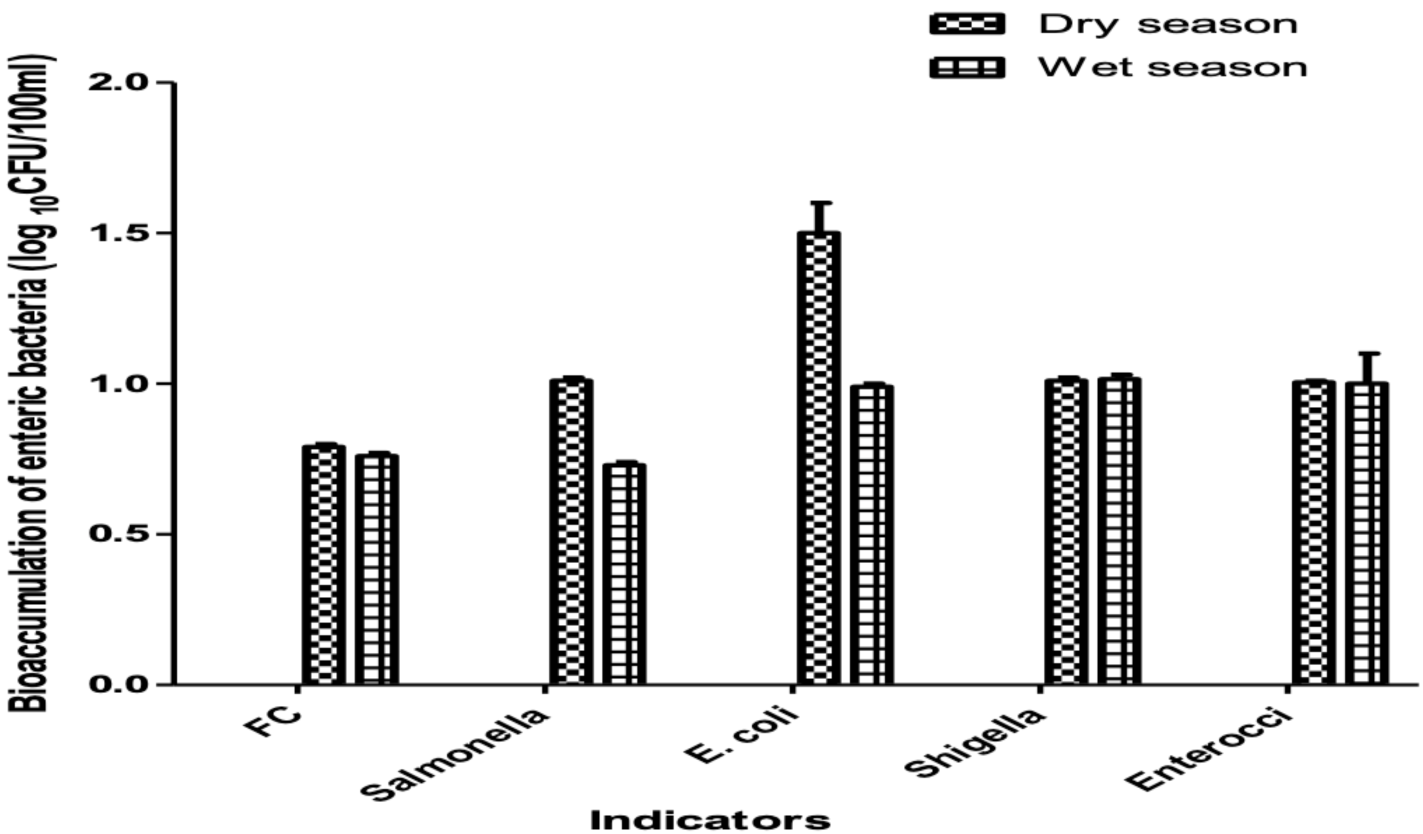

Figure 8

The rate of bioaccumulation of enteric bacteria in the catfish samples from the earthen pond during wet and dry periods

Key: FC- Faecal coliforms

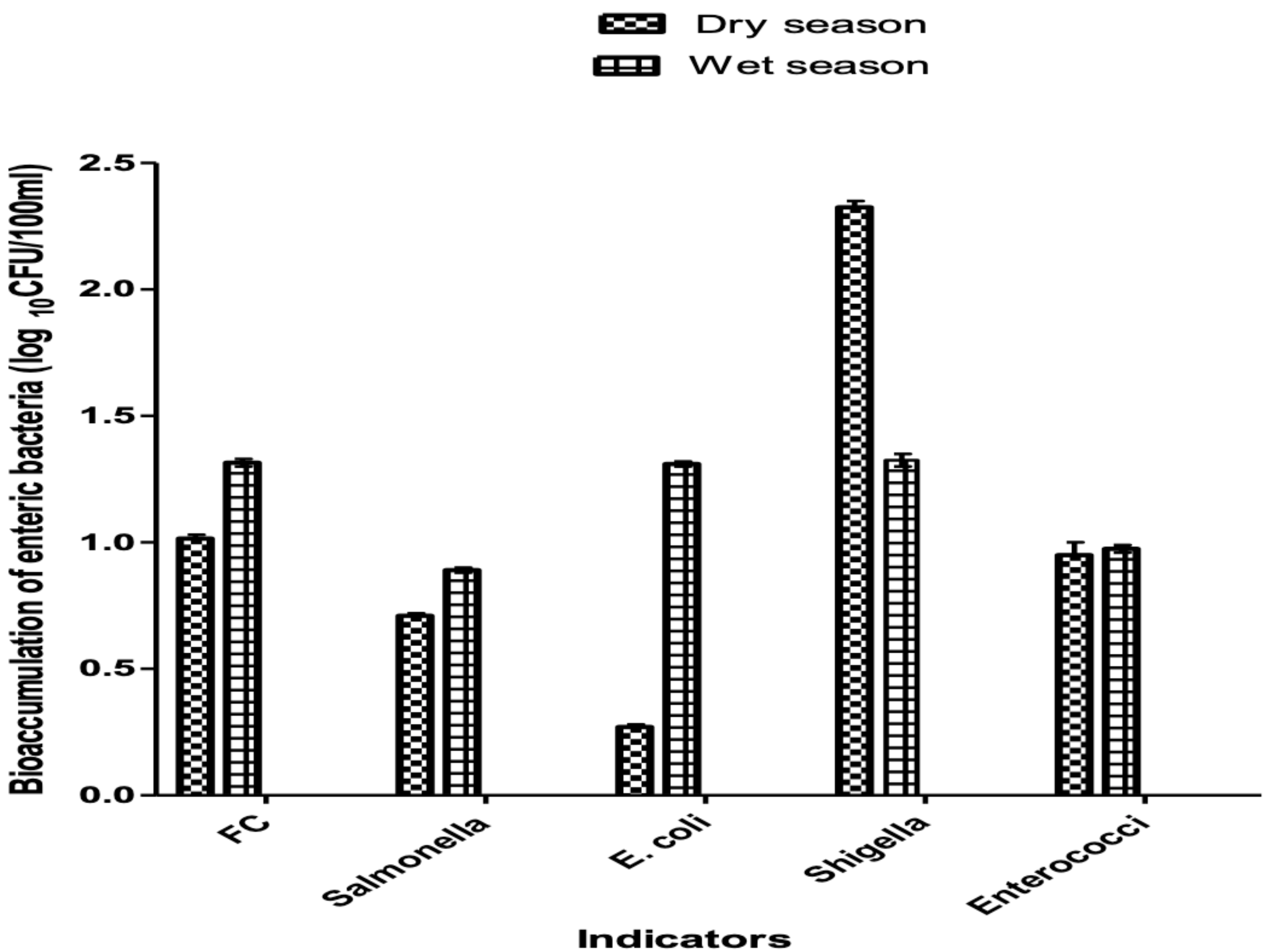

Figure 9

The rate of bioaccumulation of enteric bacteria in the catfish samples from the concrete pond during wet and dry periods

Key: FC- Faecal coliforms

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