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Bacteriological Assessment of African Catfish (*Clarias gariepinus*) Isolated from Earthen and Concrete Fish Pond

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Authors' contributions

This work was carried out in collaboration among all authors. Authors OTO, AAM and KTM. Author OTO designed the materials and methods used in the course of the research work. Authors OTO and KTM designed the antimicrobial assay procedure. Author AAM designed the fish model used during the course of research work. Authors OTO wrote the first and the final draft of the manuscript. All authors read and approved the final manuscript.

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ABSTRACT

The purpose of this research work is to evaluates, isolate, identify, characterized and compare the bacteria load in African Catfish (*Clarias gariepinus*) from Earthen and Concrete Fish Pond. African Catfish (*Clarias gariepinus*) is a choice culture fish and an African delicacy to African consumers. Concrete pond and Earthen pond are the two types of ponds used in fish farming in West Africa. An earthen pond is a water body that is enclosed by earth while a concrete pond are pond constructed

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with bricks (plastered) or tanks. *Clarias gariepinus* were obtained from the earthen and concrete ponds from Adekunle Ajasin University, Akungba–Akoko, Ondo state, Nigeria. The fishes were harvested and eviscerated and different organs of the fish were collected for the purpose of this research work. Isolation of bacteria was done using the streaking method of cultural media. Preliminary characterization of bacterial isolates were based on Gram staining, morphological and cultural characteristics. Further characterization was carried out with various biochemical tests (Catalase, Citrate, Indole, Oxidase test, Starch hydrolysis, Urease and Sugar fermentation) and Bergey's manual Microbiology. In concrete pond, it was observed that *Bacillus subtilis* was the most percentage frequently distributed bacteria isolate in *Clarias gariepinus* with (8%) *Staphylococcus aureus* (9.5%), *Alcaligenes xylosidans* (4.7%), *Alcaligenes paradoxus* (4.7%), *Acinetobacter calcoa ceticus* (4.7%), *Pseudomonas putida* (4.7%), *Bacillus cereus* (23.8%), *Citrobacter amalonaticus* (9.5%), *Acinetobacter baumanni*(4.7%), *Listeria grayi*(9.5%) and *Listeria monocytogenes* (4.7%) while In earthen pond *Enterococcus gallinarum* (4.0%), *Streptococcus ub eris* (8.0%) and *Micrococcus luteus*(4.0%) was the most frequently distributed bacteria isolate in *Clarias gariepinus* earthen pond, *Marinococcus halophilus*(4.0%),*Enterobacter aerogenes* (4.0%), *Micrococcus lylae* (4.0%), *Alcalige nes faecalis*(4.0%), Entero*coccus molodoratus* (4.0%), *Enterococcus gallinarum* (8.0%), *Bacillus pumilus* (4.0%), *Citrobacter freundii* (4.0%), *Sporosa rcina inulinus* (4.0%), *Deinococcus radiodurans* (4.0%), *Vibrio marinus*(4.0%), *Listeria murrayi* (4.0%), *Deinobacter grandis*(4.0%), *Deinococcus proteolyticus* (4.0%), *Bacillus lautus*(4.0%) *and Micrococcus halobius* (4.0%). Highest viable colony counts (5.6×10^4 for *C. gariepinus* were found in the concrete pond and (6.3×10^4) from the earthen pond respectively. Alimentary canal of fish in the concrete pond has the highest value of 4.73 ± 0.81^a and fish body has the lowest values (3.53 ± 0.99^a).Fish water has the highest value (4.33 ± 1.15^a) and lowest value ($2.20\pm1.2^{a)}$) were found in earthen pond. It can be concluded that this organisms isolated from *C. gariepinus* in this study has the potential of becoming pathogenic and dangerous health risk and constitute severe economic loss to fish farmers and general populate especially those that consume catfish, therefore the Catfish should be raised in an hygienic and properly processed methods before consumption.

Keywords: Bacterial load; Clarias gariepinus; pathogen; nutrient agar.

1. INTRODUCTION

Bacterial agents are among the highly encountered causes of diseases in stressed warm water aquaculture [1]. Aquatic micro organisms not only influence the water quality but are known to be closely associated with the physiological status of the fish and the postharvest quality of fish [2]. The health of the fish and it yields are therefore dependent on the quality of the water from which it was produced and harvested. Fish acts as an important food vehicle for some zoonotic pathogens such as *Salmonella typhi* and *Vibrio cholerae.* Contamin ation of fish with pathogens is a major public health concerns. However, consumption of fish may also cause disease due to food infection or food intoxication.

Some of these diseases have been specifically associated with pathogen which are resistant to antibiotics [3] and this poses a great risk to human health. Although only a few infectious agents in fish are able to infect humans some exceptions exist that may result in fatalities. Several species of bacteria were found to be associated with fish diseases which are caused by the presence of pathogenic microbial flora, leading to reduced fish production and affecting the normal physiology of fish, if left un-curtailed, can result in mass mortalities of fish, or in some cases, transmits infections of man and other vertebrates that consume them [4].

The African Catfish genus can be defined as displaying an eel shape, having an elongated cylindrical body with dorsal and anal fins being extremely long (nearly reaching or reaching the caudal fin) both fins containing only soft fin rays. The outer pectoral ray is in the form of a spine and the pelvic fin normally has six soft trays. The head is flattened, highly ossified, the skull bones (above and on the sides) forming a casque and the body is covered with a smooth scale less skin. The skin is generally darkly pigmented on the dorsal and lateral parts of the body. The colour is uniform marbled and changes from grayish olive to blackish according to the substrate. On exposure to light, skin the colour generally becomes lighter [5]. They have four

pairs of un-branched barbels, one nasal, one maxilla (longest and most mobile) and two mandibles(inner and outer) on the jaw. Tooth plates are present on the jaws as well as on the vomer. The major function of the barbels is prey detection [6]. *Clarias* species inhabit calm waters from lakes, streams, rivers, swamps to floodplains, some of which are subject to seasonal drying. The most common habitats frequented are floodplain swamps and pools in which the catfish can survive during the dry seasons due to the presence of the accessory air breathing organs [7].

Concrete ponds are used for intensive fish farming; concrete walls eliminate erosion due to currents caused by mechanical aeration, waves generated by the wind and fish activity (notably nesting behaviour). This type of pond is more expensive to build and, therefore, should be made profitable by a higher production per volume utilized. Conversely, the firmer walling reduces maintenance and re-building costs that will be necessary after a few years of operation. This type of pond is smaller than earthen ponds and should not exceed 1,000 m^2 surface area [8]. The bottom can also be in concrete but for reasons of construction costs, only if the pond size does not exceed 200 m^2. Brick or stone walls must have strong foundations and, if they are built with bricks or blocks, they must be plastered, in order to avoid the effects of erosion [9].

Plate 1. Earthen pond

Plate 2. Concrete pond

2. MATERIALS AND METHODS

2.1 Sample Collection of Fish Sample

A total of 48 samples were collected from both concrete pond and earthen pond. The African Catfish (*Clarias gariepinus*) used for this study, *C, gariepinus* were obtained from concrete and earthen ponds, Department of Animal and Environmental Biology (Fisheries Units), Adekunle Ajasin University Akungba-Akoko, Ondo State. The fish samples were transported alive in plastic containers (covered with net) to the Research Laboratory of the Department of Microbiology, Adekunle Ajasin University, Akungba-Akoko, Ondo State after which they were eviscerated with a sterile knife and different organs were swabbed with sterile swab sticks soaked in sterile peptone water. Sterile sample bottles were used for the collection of pond water [10].

2.1.1 Sample preservation

Samples were preserved by refrigeration at 4°C, thereby slowing down metabolic activity of microorganisms, to enhance good result when further used [10].

2.1.2 Sterilization and disinfection of materials used

An autoclave was used for sterilization of the media at 121℃ for 15 minutes. Hot air oven was used for the sterilization of various glass wares at 160℃ for 2 hours. Red hot flame from sprit lamp was used to sterilize wire loops. Surface of benches used were sterilized by swabbing with cotton wool soaked in 70% ethanol [11].

2.2 Isolation of Bacteria Isolates

2.2.1 Purification and isolation of bacteria Isolates

Distinct colonies observed from the growth of mixed culture colonies after 24 hrs incubation of the isolates are sub cultured in a new agar to obtain pure colony; this is done by streaking plate method. After incubation, growth of the bacteria sub-cultured colonies, the pure isolates obtained were stored on slants of Nutrient Agar (NA) in the refrigerator at 4ºC. Inoculums from these sources were used for the study [12]. Isolated colonies were counted using a colony counter and documented.

2.2.2 Macroscopic identification Bacteria Isolates

Morphological examination were observed and recorded on the growth isolates in the plate (Macroscopically), the parameter used are as follows colonial appearances, shape, edge, colour, and opacity.

2.3 Identification and Characterization of Bacteria Isolates

2.3.1 Gram staining technique of the bacteria Isolates

Working solution of reagents used for the Gram staining technique was prepared according to manufacturer's instruction. Staining was carried out by emulsifying approximately one isolated 18- 24hours old colony in a drop of water placed at the centre of a clean grease free slide until a thin smear was made. The smear was air heat fixed by passing the slide through a Bunsen burner flame and then air dried. The heat fixed smear was flooded with a basic aniline dye (crystal violet) for 60 seconds. This was flooded with Lugol's iodine and allowed to remain for 60 seconds. This was then rinsed off with running tap water. The smear was decolorized with 70% ethanol which was immediately washed out to avoid total decolorization. The smear was counter stained with safranin for 60seconds, washed off with running tap water and blot-dried. The slide was then examined under oil immersion objective microscope. Organisms that retained the purple colour of crystal violet- iodine complex (CV-1 complex) were recorded as Gram- positive, while those that appeared pink were Gram- negative [13].

2.4 Biochemical Tests of the Isolated Organisms

2.4.1 Catalase test

This test detects the presence of catalase enzyme when present in a bacterium, it catalyse the breaking down of hydrogen peroxide with the release of oxygen as bubble. With a wire loop, a colony was picked from the pure culture and was transferred to the centre of a glass slide. 1- 2 drops of 3% hydrogen peroxide was added to the bacterial isolates. Immediate production of bubbles indicated positive result and if no bubble indicated negative [12].

$2H_2O_2 \rightarrow 2H_2O + O_2$

2.4.2 Oxidase test

The isolated organisms were inoculated and grown in Nutrient broth for 24 hrs at 37°C. After 24 hrs Oxidase strip was dipped into the broth and colour change was observed. Bacteria Isolates were oxidase positive when the colour changes to purple within 15 seconds to 30 seconds and oxidase negative when the colour did not change at al. [12].

2.4.3 Indole test

This test demonstrates the ability of certain bacteria to decompose the amino acid tryptophan to indole which then accumulates in the medium for indole production. Bacterial isolates were inoculated not peptone water medium contained in a sterile test .tubes then incubated at 37°C for 48 hours. After the incubation period about 3 drops of kovac's indole reagent was added to the peptone water culture. The bottles were shaken thoroughly and allowed to stand and observed for colour development. A red colour ring at the interface of the medium denotes a positive result. And if the isolate is negative, the reagent layer will remain yellow or slightly cloud [12].

2.4.4 Urease test

The Urease test is used to identify those organisms that are capable of hydrolyzing urea to produce ammonia and carbon dioxide. In this test each isolate was inoculated into test tubes containing sterilized urea agar medium and incubated at 37°C. The medium was observed for a colour change at 24 hrs and everyday up to 6 days. Urease production was indicated by a bright pink colour throughout the medium [12].

2.4.5 Simmon's citrate test

The citrate test screens bacterial isolates for the ability to utilize citrate as its carbon and energy source. Citrate agar was prepared and homogenized on a magnetic stirrer after which it was dispensed into test tubes and sterilized in the autoclave and slants were prepared. The slants were inoculated with the test organisms and incubated at 37°C for 24hrs. Slant culture was observed for the growth and coloration of the medium, positive with blue colour and negative with green colour [10].

2.5 Sugar Fermentation of the Bacteria Isolates

This test shows the ability of microorganisms to ferment certain sugars. Five sugars were used; mannitol, sucrose, maltose, galactose and fructose using [14].

Mannitol: 3 g of peptone powder was dissolved in 180 ml of distilled water in appropriately labeled conical flask and 0.5 g of phenol red was added. 1 g of Mannitol sugar was added into the conical flask and shaken thoroughly. The solution was dispensed in 5 ml amounts into test tubes with inverted Durham's tubes and autoclaved for 15 minutes. The test tubes were then inoculated with loop full of test organisms and incubated at 37°C for maximum of 48 hours. The test was observed for acid production leading to colour change (red to yellow) as well as gas production that causes the displacement of the liquid in the inverted Durham's tubes which indicates a positive test [14].

Sucrose: 3 g of peptone powder was dissolved in 180 ml of distilled water in appropriately labeled conical flask and 0.5 g of phenol red was added. 1 g of Sucrose sugar was added into the conical flask and shaken thoroughly. The solution was dispensed in 5 ml amounts into test tubes with inverted Durham's tubes and autoclaved for 15 minutes. The test tubes were then inoculated with loop full of test organisms and incubated at 37°C for maximum of 48 hours. The test was observed for acid production leading to colour change (red to yellow) as well as gas production that causes the displacement of the liquid in the inverted Durham's tubes which indicates a positive test [14].

Maltose: 3 g of peptone powder was dissolved in 180 ml of distilled water in appropriately labelled conical flask and 0.5 g of phenol red was added. 1 g of Maltose sugar was added into the conical flask and shaken thoroughly. The solution was dispensed in 5ml amounts into test tubes with inverted Durham's tubes and autoclaved for 15 minutes. The test tubes were then inoculated with loop full of test organisms and incubated at 37°C for maximum of 48 hours.. The test was observed for acid production leading to colour change (red to yellow) as well as gas production that causes the displacement of the liquid in the inverted Durham's tubes which indicates a positive test [14].

Galactose: 3 g of peptone powder was dissolved in 180 ml of distilled water in appropriately labeled conical flask and 0.5 g of phenol red was added. 1 g of Galactose sugar was added into the conical flask and shaken thoroughly. The solution was dispensed in 5 ml amounts into test

tubes with inverted Durham's tubes and autoclaved for 15 minutes. The test tubes were then inoculated with loop full of test organisms and incubated at 37°C for maximum of 48 hours. The test was observed for acid production leading to colour change (red to yellow) as well as gas production that causes the displacement of the liquid in the inverted Durham's tubes which indicates a positive test [13].

Fructose: 3 g of peptone powder was dissolved in 180 ml of distilled water in appropriately labeled conical flask and 0.5 g of phenol red was added. 1 g of Fructose sugar was added into the conical flask and shaken thoroughly. The solution was dispensed in 5 ml amounts into test tubes with inverted Durham's tubes and autoclaved for 15 minutes. The test tubes were then inoculated with loop full of test organisms and incubated at 37°C for maximum of 48 hours. The test was observed for acid production leading to colour change (red to yellow) as well as gas production that causes the displacement of the liquid in the inverted Durham's tubes which indicates a positive test [15].

2.6 Starch Hydrolysis Test of Bacteria Isolates

Nutrient agar was prepared and the isolates were inoculated onto the plates with sterile inoculating loop using streak method. The plates were incubated at 37°C for 24 hrs, after incubation the plates were flooded with Gram's iodine. Plates were observing for clear zone around the test organisms [13].

3. RESULTS AND DISCUSSION

3.1 The Result of this Research Work is Represented in Table 1-6 and Fig. 1a and 1b

Table 1. The total bacterial colony count CFU (colony forming unit) of isolates obtained from *C. gariepinus* in concrete pond and earthen pond. Bacteria colonies after 24 hrs of incubation were subjected to counting and were expressed in Colony Forming Unit per ml (Cfu/ml). In Table 1, the total bacteria count of isolates in African Catfish (*Clarias gariepinus*) raised in concrete and earthen fish ponds. It was observed that 22 (twenty two) samples were collected from the concrete pond while 27 (Twenty seven) sample were collected from the earthen pond. The sample CPAAC (Concrete pond A fish alimentary canal) has the highest bacteria count value of $5.6x10^4$, while CPBB(Concrete pond B fish body) has the lowest bacteria count value of $2.4\ x10^4$ from concrete pond. EPCAC (Earthen pond C alimentary canal) has $6.3\ x\ 10^4$ highest bacteria count value of $6.3\ x\ 10^4$ and EPBK (Earthen pond B kidney) has the lowest bacteria count value of $1.0\ x10^4$ in earthen pond.

Table 2. The morphological and microscopic characterization of isolates obtained from *C, gariepinus* in concrete pond and earthen pond. The bacteria colonies were examined for surface appearance, color, shape, edge and appearance in light after 24 hours of incubation. Table 2a and 2b, Morphological, macroscopic and cultural characteristics of bacterial isolate in African Catfish (*Clarias gariepinus*) from Concrete and earthen ponds. This table depicts the Surface, Colour, Shape, Edge, and Appearance In Light. Surface (Smooth, Rough, Dull and Glistering). Colour (Milk, Greenish, Cream, White, Greenish blue), Edge (Tenate, Lobate, Fimbrate, Round and Filamentous), Shape (Irregular, Filamentous, Circular and Spindle), while Appearance in light (Transparent, Opaque and Transparent).

Table 3 The microscopic examination of all the isolates obtained from *C. gariepinus* in concrete pond and earthen pond. Using a pure culture, the isolates were Gram stained and viewed under the microscope using X100 lens. The results were recorded as Gram positive rod, Gram negative rod, Gram positive cocci and Gram negative cocci. Microscopic examination of bacterial isolates in African Catfish (*Clarias gariepinus*) raised from concrete and earthen ponds. In this table, the observable features are as follows GPC (Gram positive cocci), GPR (Gram positive rod), GNR (Gram negative rod) and GNC (Gram Negative cocci). In the concrete pond, 7 (Seven) sample were Gram negative rod, 14(fourteen) samples were Gram positive rod and 1(One) sample was Gram positive cocci. In the earthen pond, 15 (Fifteen) sample were Gram positive cocci, 5(Five) sample were Gram negative rod, 2(two) were Gram positive rod and 3 (three) samples were Gram Negative cocci.

Table 4a and 4b, Biochemical characteristics and sugar fermentation of bacteria isolates in African Catfish (*Clarias gariepinus*) fr*om* concrete and earthen ponds. This table depicts several biochemical characteristics which include the following CAT(Catalase test), URE (Urease test), SH (Starch hydrolysis) and MAN (Mannitol test),

the sugar fermentation include the following SUC (Sucrose test), IND (Indole test), MOT (Motility test), OXI (Oxidase test), CIT (Citrate test), MAL (Maltose), GAL (Galactose), NM (Non Motile), FRU (Fructose), GP (Gas producti on), and NG (No gas).

Table 5 Probable or suspected bacteria isolated from *C. gariepinus* in concrete and earthen pond, after identification with biochemical test, sugar fermentation test, Macroscopic and microscopy examination of the isolates and identification with Bergey's manual systematic microbiology. Probable organisms found in African catfish (*Clarias gariepinus*) from *concrete* and earthen ponds. Te probable organisms were *Staphyloco ccus aureus, Alcaligenes xylosidan, Alcaligenes paradoxus, Acinetobacter calcoaceticus, Bacillus subtilis, Pseudomonas putida, Bacillu cereus, Citrobacter amalonaticus, Citrobacter amalonat Icus, Acinetobacter baumanni, Listeria grayi and Listeria monocytogenes* from concrete pond. In earthen pond, this are the probable organisms, *Marinococcus halophilus, Enterococcus gallinar um, Bacillus cereus, Enterobacter aerogenes, Micrococcus lylae, Staphylococcus aureus, Stre ptococcus uberis, Enterococcus gallinarum, Alc aligenes feacalis, Micrococcus luteus, Listeria grayi, Enterococcus molodoratus, Sporosarcina inulinus, Enterococcus molodoratus, Bacillus pumilus, Deinococcus radiodurans, Vibrio marinus, Citrobacterfreundii, Deinococcus proteolyticus, Listeria murrayi, Micrococcus luteus, Bacillus lautus, Micrococcus halobius* and *Deinobacter grandis.*

Table 6 The Statistical analysis of bacteria colony count of isolates obtained from *C. gariepinus* in Concrete and Earthen pond. Across the row, Concrete pond water (4.33 ± 1.15^{a}) is not significantly different from Earthen pond water (4.00 ± 0.00^{a}), concrete pond alimentary canal (4.73 ± 0.81^{a}) is not significantly different from earthen pond alimentary canal (5.50 ± 0.72^{a}), concrete pond liver (3.27 ± 0.64^{a}) is not significantly different from earthen pond liver (2.20 ± 1.2^{a}), concrete pond kidney (3.67 ± 0.58^{a}) shows no significance difference from earthen pond kidney (2.95 ± 1.56^{a}), concrete pond mouth (3.93 ± 0.31^{a})shows no significance difference from earthen pond mouth (3.25 ± 1.38^{a}), concrete pond body (3.53 ± 0.99^{a}) shows no significance difference from earthen pond body (3.43 ± 1.17^{a}) while concrete pond gills (4.00 ± 0.69^{b}) shows significant difference from earthen pond gills (2.33 ± 1.1^{a}). And down the column water, alimentary canal, liver, kidney, mouth and body were significantly different from gills. The analysis were significantly different at *P<0.05.*

Fig. 1a &1b The percentage frequency distributl on of isolated microorganisms in African catfish (*Clarias gariepinus*) from concrete and earthen pond.

In concrete pond, it was observed that *Bacillus subtilis* was the most percentage frequently distributed bacteria isolate from *Clarias gariepinus* with (8%) *Staphylococcus aureus* (9.5%), *Alcaligenes xylosidans*(4.7%), *Alcaligenes paradoxus* (4.7%), *Acinetobacter calcoa ceticus* (4.7%), *Pseudomonas putida* (4.7%), *Bacillus cereus*(23.8%), *Citrobacter amalonaticus* (9.5%), *Acinetobacter baumanni* (4.7%), *Listeria grayi* (9.5%) and *Listeria monocytogenes* (4.7%).

In earthen pond *Enterococcus gallinarum* (4.0%), *Streptococcus uberis*(8.0%) and *Micrococcus luteus* (4.0%) was the most percentage frequentl y distributed bacteria isolate in *Clarias gariepinus* earthen pond, *Marinococcus halophilus* (4.0%), *Enterobacter aerogenes* (4.0%), *Micrococcus lylae* (4.0%), *Alcaligenes faecalis* (4.0%), Entero *coccus molodoratus* (4.0%), *Enterococcus galli narum* (8.0%), *Bacillus pumilus* (4.0%), *Citro bacter freundii* (4.0%),*Sporosarcina inulinus* (4.0%), *Deinococcus radiodurans* (4.0%), *Vibrio marinus* (4.0%), *Listeria murrayi* (4.0%), *Deino bacter grandis* (4.0%),*Deinococcus proteo lyticus* (4.0%), *Bacillus lautus* (4.0) *and Micrococcus halobius* (4.0%).

The purpose of this research work is to evaluate, isolate, identify, characterized and compare the bacteria load in African Catfish (*C. gariepinus*) found Earthen and Concrete ponds, Different bacteria were isolated from seven samples of *C. gariepinus*. The different species of bacteria isolated from both ponds were *Staphylococcus aureus, Alcaligene sxylosidans, Alcaligenes paradoxus, Acinetobacter calcoaceticus, Bacillus subtilis, Pseudomonas putida, Bacillus cereus, Citrobacter amalonaticus, Acinetobacter bauma nni, Listeria grayi, Listeria monocytogenes, Marinococcus halophilus, Enterobacter aero genes, Micrococcus lylae, Alcaligenes faecalis, Enterococcus molodoratus, Enterococcus gallin arum, Enterococcus, Bacillus pumilus, Citro bacterfreundii, Sporosarcina inulinus, Deino co ccus radiodurans, Vibrio marinus, Listeria murrayi, Deinobacter grandis, Deinococcus proteolyticus, Bacillus lautus and Micrococcus halobius.*

Table 1. Total bacterial counts of isolates in African catfish (*Clarias gariepinus*) raised from Concrete and Earthen Ponds

Isolates	Total bacterial count(cfu/ml) Concrete fish pond	Isolates	Total bacterial count (cfu/ml 10^{-3}) Earthen fish pond
CPAPW	5.0x 10^3	EPW	4.0 x 10^3
CPAAC	5.6 x 10^4	EPAAC	5.0 x 10^4
CPAL	4.0 x 10^3	EPAL	1.6 x 10^4
CPAK	3.0 x 10^4	EPAK	3.2 x 10^4
CPAM	4.2 x 10^4	EPAM	4.4 x 10^4
CPAB	4.0 x 10^4	EPAB	4.8 x 10^4
CPAG	3.6 x 10^4	EPAG	I.6 x 10^4
CPBPW	3.0 x10^3	EPBAC	4.8 x 10^4
CPBAC	4.0 x 10^4	EPBL	1.6 x 10^4
CPBL	2.8 x 10^4	EPBK	1.0 x 10^4
CPBK	4.0 x 10^4	EPBM	4.2 x 10^4
CPBM	4.0 x 10^4	EPBB	4.0 x 10^4
CPBB	2.4 x 10^4	EPBG	1.2 x 10^4
CPBG	4.8 x 10^4	EPCAC	6.3 x 10^4
CPCPW	5.0 x 10^3	EPCL	1.6 x10^4
CPCAC	4.6 x 10^4	EPCK	2.8 x 10^4
CPCL	3.0 x 10^4	EPCM	1.4 x 10^4
CPCK	4.0 x 10^4	EPCB	2.4 x 10^4
CPCM	3.6 x 10^4	EPCG	3.5 x10^4
CPCB	4.2 x 10^4	EPDAC	5.9 x 10^4
CPCG	3.6 x 10^4	EPDL	4.0 x 10^4
-	-	EPDK	4.8 x 10^4
-	-	EPDM	3.0 x 10^4
-	-	EPDB	2.5 x 10^4
-	-	EPDG	3.0 x 10^4

Keys: CPAPW= concrete pond A pond water, CPAAC= concrete pond A alimentary canal, CPAL= concrete pond A liver, CPAK= concrete pond A kidney, CPAM= concrete pond A mouth, CPAB= concrete pond A body, CPAG= concrete pond A gills, CPBPW= concrete pond B pond water, CPBAC= concrete pond B alimentary canal, CPBL= concrete pondBliver, CPBK= concrete pond B kidney, CPBM= concrete pond B mouth, CPBB= concrete pond B body, CPBG= concrete pond B gills, CPCPW= concrete pond C pond water, CPCAC= concrete pond Calimentary canal, CPCL= concrete pond C liver, CPCK= concrete pond C kidney, CPCM= concrete pond C mouth, CPCB= concrete pond C body and CPCG= concrete pond C gills
Keys: EPW= earthen pond water, EPAAC= earthen pond A alimentary canal, EPAL= earthen pond A liver, EPAK= earthen pond A kidney, EPAM= earthen pond A mouth, EPAB= earthen pond A body, EPAG= earthen pond A gills, EPBAC= earthen pond B alimentary canal, EPBL= earthen pond B liver, EPBK= earthen pond B kidney, EPBM= earthen pond B mouth, EPBB= earthen pond B body, EPBG= earthen pond B gills, EPCAC= earthen pond C alimentary canal, EPCL= earthen pond C liver, EPCK= earthen pond C kidney, EPCM= earthen pond C mouth, EPCB= earthen pond C body, EPCG= earthen pond C gills, EPDAC= earthen pond D alimentary canal, EPDL= earthen pond D liver, EPDK= earthen pond D kidney, EPDM= earthen pond D mouth, EPDB= earthen pond D body, EPDG= earthen pond D gills

These organisms were variously present on the different organs of *C. gariepinus* from the earthen and concrete ponds [16]. The higher bacteria load in concrete pond may be due to improper hygiene of the fish pond, how the pond was used and how frequently the pond water is changed. The loads of bacteria associated with *C. gariepinus* from earthen pond may be due to contamination as a result of indiscriminate deposition of waste materials into the ponds through runoffs, animal excreta and other environmental wastes, free roaming animals and pets such as dogs also contribute to fecal contamination of the fish pond.

Variations in the bacterial load of gills, alimentary canals, kidney, liver, pond water and body of the fish samples existed, being highest in the

alimentary canals of the fish samples from both ponds but lowest in the pond water. The highest bacterial load encountered in the alimentary canal of the fish samples compared to the other organs could be due large surface area provided by the alimentary canal and availability of different stages of digested food particles present in the environment.

The highly infected part of the examined° *C. gariepinus* used for bacteriological studies was the skin compared to the intestine, this may be due to the fact that the skin is always in contact with the surrounding water and also, the skin may get contaminated with bacteria during handling. This observation agrees with the work of [17] who recorded high bacterial loads on the skin of fresh Tilapia fish (*Oreochromis niloticus*) when compared with the intestines and gills; Adebayo – Tayo et al. [18] also made similar observation but this observation was in contrary to the work of Olugbojo et al. [19] who reported high population of bacteria in the gut of three fish species sampled in Lagos State; likewise [20] and [21], also reported high bacterial loads in the intestine part of African Catfish.

The bacteria isolated included facultative pathogens which under stress, could give rise to disease of fish, and subsequently, to humans. *Staphylococcus sp.* Has been implicated in fish-borne diseases [22]. Staphylococcus aureus frequently causes *septicemia, osteomyelitis, bacteremia and otitis* [23]. *Pseudomonas sp* could cause general inflammation and sepsis in critical body organs such as lungs, kidneys, urinary tract, which can be fatal because it thrives in most surfaces [23]. *Enterococcus molodoratus* is a causative agent of dental infection and scarlet fever and has been implicated in human infections like pharyngitis, scarlet fever and pneumonia [24].

Table 2a. Morphological, Macroscopic and Cultural Characteristics of bacterial isolates in African catfish (*Clarias gariepinus*) from Concrete Pond

Isolates	Surface	Colour	Edge	Shape	Appearance in light
CPAPW	Smooth	Milk	Tenate	Irregular	Transparent
CPAAC	Rough	Greenish	Lobate	Filamentous	Transparent
CPAL	Dull	Cream	Fimbrate	Irregular	Opaque
CPAK	Smooth	Milk	Tenate	Irregular	Transparent
CPAM	Smooth	Cream	Round	Irregular	Opaque
CPAB	Rough	Cream	Entire	Circular	Opaque
CPAG	Smooth	Greenish Blue	Lobate	Filamentous	Transparent
CPBPW	Dull	Cream	Round	Irregular	Opaque
CPBAC	Glistering	White	Round	Irregular	Transparent
CPBL	Rough	Cream	Fimbrate	Circular	Opaque
CPBK	Rough	Cream	Round	Circular	Opaque
CPBM	Dull	Cream	Round	Irregular	Opaque
CPBB	Smooth	Greenish Blue	Fimbrate	Irregular	Translucent
CPBG	Smooth	Cream	Lobate	Irregular	Transluscent
CPCPW	Dull	Yellow	Crenate	Irregular	Opaque
CPCAC	Glistering	White	Round	Irregular	Transparent
CPCL	Dull	Milk	Filamentous	Irregular	Translucent
CPCK	Smooth	Milk	Crenate	Irregular	Transluscent
CPCM	Smooth	Green	Round	Spindle	Opaque
CPCB	Smooth	Yellow	Crenate	Rhizoid	Opaque
CPCG	Rough	White	Round	Circular	Transparent

Keys: CPAPW= concrete pond A pond water, CPAAC= concrete pond A alimentary canal, CPAL= concrete pond A liver, CPAK= concrete pond A kidney, CPAM= concrete pond A mouth, CPAB= concrete pond A body, CPAG= concrete pond A gills, CPBPW= concrete pond B pond water, CPBAC= concrete pond B alimentary canal, CPBL= concrete pond B liver, CPBK= concrete pond B kidney, CPBM= concrete pond B mouth, CPBB= concrete pond B body, CPBG= concrete pond B gills, CPCPW= concrete pond C pond water, CPCAC= concrete pond C alimentary canal, CPCL= concrete pond C liver, CPCK= concrete pond C kidney, CPCM= concrete pond C mouth, CPCB= concrete pond C body and CPCG= concrete pond C gills

Table 2b. Morphological, Macroscopic and Cultural Characteristics of bacterial isolates in African catfish (*Clarias gariepinus*) from Earthen Pond

Isolates	Surface	Colour	Edge	Shape	Appearance in light
EPW	Glistering	Yellow	Crenate	Irregular	Translucent
EPAAC	Smooth	Milk	Rhizoid	Puntiform	Translucent
EPAL	Glistering	Cream	Crenate	Irregular	Translucent
EPAK	Dull	Milk	Lobate	Circular	Translucent
EPAM	Dull	Cream	Lobate	Puntiform	Translucent
EPAB	Rough	Milk	Fimbrate	Filamentous	Opaque
EPAG	Rough	Milk	Entire	Irregular	Translucent
EPBAC	Dull	Cream	Fimbrate	Irregular	Translucent
EPBL	Smooth	Yellow	Crenate	Circular	Opaque
EPBK	Smooth	Cream	Lobate	Irregular	Translucent
EPBM	Glistering	Cream	Lobate	Filamentous	Translucent
EPBB	Rough	Milk	Entire	Rhizoid	Opaque
EPBG	Rough	Milk	Tenate	Irregular	Translucent
EPCAC	Rough	Milk	Tenate	Circular	Translucent
EPCL	Smooth	Milk	Entire	Irregular	Opaque
EPCK	Dull	Milk	Lobate	Filamentous	Translucent
EPCM	Dull	Cream	Crenate	Puntiform	Translucent
EPCB	Glistering	Pink	Fimbrate	Irregular	Translucent
EPCG	Glistering	Milk	Circular	Irregular	Transparent
EPDAC	Smooth	Milk	Tenate	Circular	Translucent
EPDL	Rough	Pink	Lobate	Spindle	Translucent
EPDK	Dull	Pink	Entire	Circular	Translucent
EPDM	Glistering	Pink	Fimbrate	Spindle	Translucent
EPDB	Dull	Yellow	Circular	Puntiform	Opaque
EPDG	Dull	Milk	Lobate	Irregular	Translucent

Keys: EPW= earthen pond water, EPAAC= earthen pond A alimentary canal, EPAL= earthen pond A liver, EPAK= earthen pond A kidney, EPAM= earthen pond A mouth, EPAB= earthen pond A body, EPAG= earthen pond A gills, EPBAC= earthen pond B alimentary canal, EPBL= earthen pond B liver, EPBK= earthen pond B kidney, EPBM= earthen pond B mouth, EPBB= earthen pond B body, EPBG= earthen pond B gills, EPCAC= earthen pond C alimentary canal, EPCL= earthen pond C liver, EPCK= earthen pond C kidney, EPCM= earthen pond C mouth, EPCB= earthen pond C body, EPCG= earthen pond C gills,EPDAC= earthen pond D alimentary canal, EPDL= earthen pond D liver, EPDK= earthen pond D kidney, EPDM= earthen pond D mouth, EPDB= earthen pond D body, EPDG= earthen pond D gills

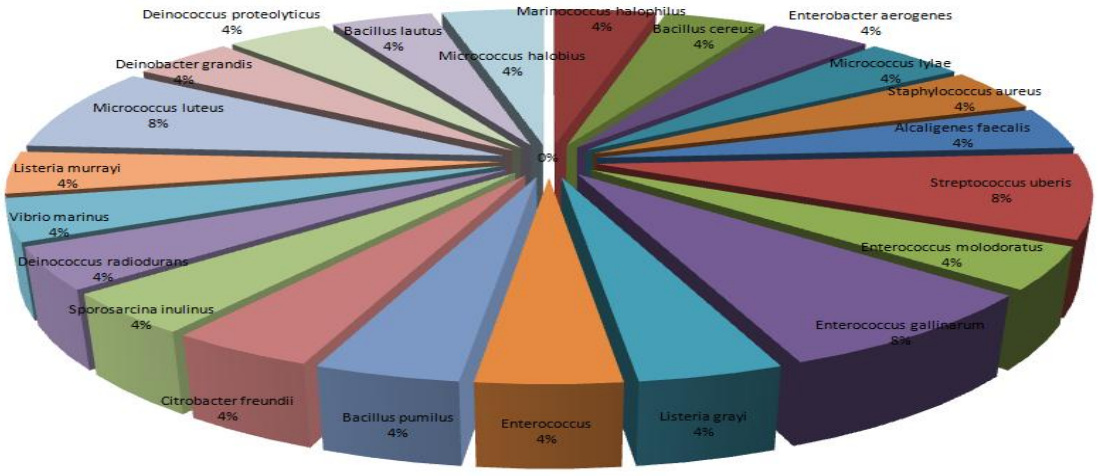

Fig. 1a. Percentage Frequency Distribution isolates in African catfish (*Clarias gariepinus*) from Earthen Pond

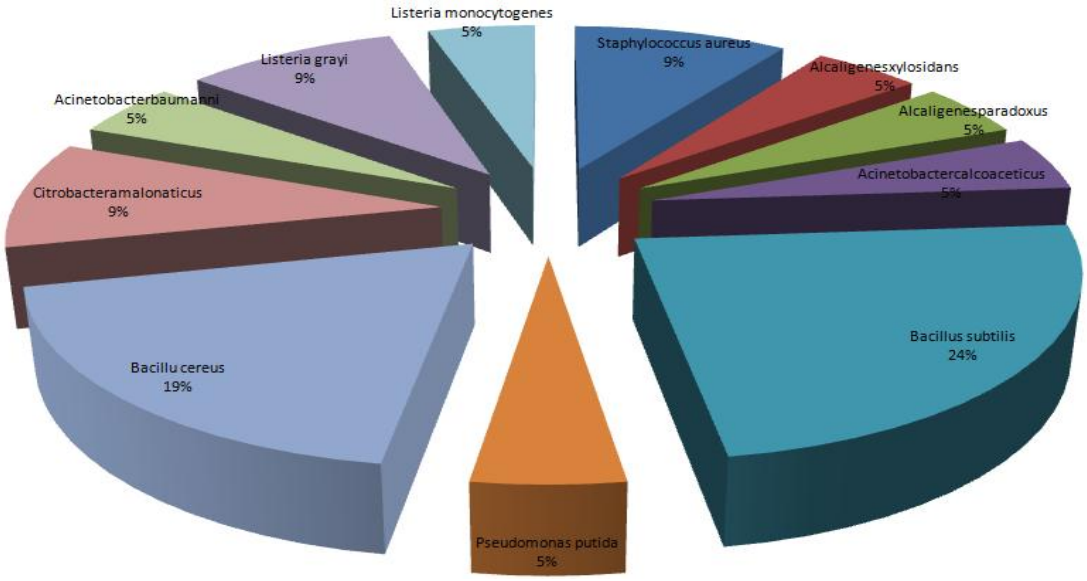

Fig. 1b. Percentage Frequency Distribution of isolates in African catfish (*Clarias gariepinus*) from Concrete Pond

The fish microflora contains substantial and complex collection of microorganisms forming a biologically pivotal component of the host body. This microflora are composed of different species of microorganisms which interact with each other. This microflora exerts properties which are potentially damaging or health promoting for the host [25].

Most bacteria species identified in this study that were present in *Clarias gariepinus*, include both pathogenic and normal flora. These bacteria species found in the tissues of *C. gariepinus* in this study were similar to the isolated isolates in cultured *Clarias gariepinus* by [26] and [27]. The occurrence of these bacteria species in different organs of fish, could be indication of presence of certain predisposing factors such as handling of the fish feeding, water changing as at when necessary and also cleaning and disinfecting the ponds.

Some normal floral of humans such as *Staphylococcus and Streptococcus* spp were found predominantly in the fish organs from the different ponds. Some of the isolates were potential spoilage organisms. Live healthy fish can be colonized by bacteria while the flesh remains sterile. After death, incorrect or inadequate handling can introduce bacteria to the flesh resulting in spoilage. The natural flora of fish that play a predominant role in spoilage include the genera *Pseudomonas, Vibrio* and *Micrococcus*, while *Pseudomonas sp.* are among the major spoilage bacteria at near freezing temperatures [24].

The occurrence of *Staphylococcous aureus*. observed from the two habitats (earthen and concrete ponds) is an indication of contamination of C. *gariepinus* by man during fish harvesting and handling process; this observation agrees with the work of [28]. The isolation of enteric organisms such as *E. coli* is particularly useful as an indicator of faecal contamination [29] and from this study, *E. coli* was found to occur in river A and river B which are both natural habitat and they are both large unprotected river. Therefore, the presence of *E. coli* may be due to the presence of faecal pollution caused by human and other environmental wastes in the water bodies from which the *C. gariepinus* was obtained, similar observation was also made by Osungbemiro et al. [30].

It should be mention here that Water management in earthen fish pond is a lot easier than in concrete fish pond due to the fact that, most wastes from the fish and the fish feeds will be absorbed by the soil in earthen fish ponds while they will be flushed out in concrete ponds, this is associated with the number of microbial load grater in the earthen pond compared with the concrete pond as demonstrated in Fig. 1a and 1b [31]. It should be mentioned that the

phytoplanktons and zooplanktons are available as additional sources of feed in earthen pond while artificial feeds are the only source in concrete ponds. There is no need to clean earthen ponds as it is done for concrete pond [32].

Furthermore, it is easier to ensure adequate security with concrete pond than with earthen ponds due to natural predators and most concrete ponds are usually built in and enclosure. Also sorting, numbering and harvesting are easily done in concrete ponds than in earthen ponds. Diseases, cannibalism and death is also easily observed in concrete tanks, meanwhile how well the fishes are consuming and absorbing their feeds is easily known in concrete ponds than in earthen ponds and more importantly, there is no need of a water-logged (swampy) area before embarking in concrete pond construction [32].

Table 3. Microscopic examination of bacterial isolates in African catfish (*Clarias gariepinus*) raised from Concrete and Earthen Ponds

Isolates	Gram staining (Concrete Fish Pond)	Isolates	Gram Staining (Earthen Fish Pond)
CPAPW	GPC	EPW	GPC
CPAAC	GNR	EPAAC	GPC
CPAL	GNR	EPAL	GNR
CPAK	GPR	EPAK	GPC
CPAM	GNR	EPAM	GPC
CPAB	GPR	EPAB	GPC
CPAG	GNR	EPAG	GNR
CPBPW	GPR	EPBAC	GNC
CPBAC	GPR	EPBL	GPC
CPBL	GNR	EPBK	GPC
CPBK	GPR	EPBM	GPC
CPBM	GNR	EPBB	GPC
CPBB	GNR	EPBG	GPR
CPBG	GPR	EPCAC	GPC
CPCPW	GPR	EPCL	GPR
CPCAC	GPR	EPCK	GNR
CPCL	GPR	EPCM	GPC
CPCK	GPR	EPCB	GPC
CPCM	GPR	EPCG	GNC
CPCB	GPR	EPDAC	GPR
CPCG	GPR	EPDL	GPC
		EPDK	GPC
		EPDM	GPC
		EPDB	GPR
		EPDG	GPC

Keys: CPAPW= concrete pond A pond water, CPAAC= concrete pond A alimentary canal, CPAL= concrete pond A liver, CPAK= concrete pond A kidney, CPAM= concrete pond A mouth, CPAB= concrete pond A body, CPAG= concrete pond A gills, CPBPW= concrete pond B pond water, CPBAC= concrete pond B alimentary canal, CPBL= concrete pond B liver, CPBK= concrete pond B kidney, CPBM= concrete pond B mouth, CPBB= concrete pond B body, CPBG= concrete pond B gills, CPCPW= concrete pond C pond water, CPCAC= concrete pond C alimentary canal, CPCL= concrete pond C liver, CPCK= concrete pond C kidney, CPCM= concrete pond C mouth, CPCB= concrete pond C body and CPCG= concrete pond C gills. GPC= Gram positive cocci, GPR= Gram positive rod and GNR= Gram negative rod. Keys: EPW= earthen pond water, EPAAC= earthen pond A alimentary canal, EPAL= earthen pond A liver, EPAK= earthen pond A kidney, EPAM= earthen pond A mouth, EPAB= earthen pond A body, EPAG= earthen pond A gills, EPBAC= earthen pond B alimentary canal, EPBL= earthen pond B liver, EPBK= earthen pond B kidney, EPBM= earthen pond B mouth, EPBB= earthen pond B body, EPBG= earthen pond B gills, EPCAC= earthen pond C alimentary canal, EPCL= earthen pond C liver, EPCK= earthen pond C kidney, EPCM= earthen pond C mouth, EPCB= earthen pond C body, EPCG= earthen pond C gills, EPDAC= earthen pond D alimentary canal, EPDL= earthen pond D liver, EPDK= earthen pond D kidney, EPDM= earthen pond Dmouth, EPDB= earthen pond D body, EPDG= earthen pond D gills .GPC= Gram positive cocci, GPR= Gram positive rod, GNC= Gram negative cocci and GNR= Gram negative rod

Table 4a. Biochemical characteristics and sugar fermentation of bacteria isolates in African catfish (*Clarias gariepinus*) from Concrete Pond

Isolates	CAT	IND	OXI	URE	MOT	SH	CIT	MAN	SUC	FRU	MAL	GAL
CPAPW	+	+	-	-	NM	-	+	+ GP	+GP	+ GP	+ GP	+ GP
CPAAC	+	+	+	+	NM	+	+	+ GP	+NG	+NG	+ GP	-
CPAL	+	+	+	+	NM	+	+	+ GP	+GP	+ GP	+ GP	+ GP
CPAK	+	+	-	-	NM	+	+	+ GP	+GP	+ GP	+ GP	+NG
CPAM	+	+	-	-	NM	+	+	+ GP	+GP	+ GP	+ GP	+ GP
CPAB	+	+	-	-	M	+	+	+ GP	+GP	+ GP	+ GP	-
CPAG	+	+	-	-	NM	+	+	+ GP	+GP	+ GP	+ GP	+ GP
CPBPW	+	+	-	-	M	+	+	+ GP	+NG	+ GP	+ GP	-
CPBAC	+	+	-	-	M	+	+	+ GP	+GP	+ GP	+ GP	-
CPBL	+	+	-	-	NM	+	+	+ GP	+NG	+NG	+ GP	+ GP
CPBK	+	+	-	-	M	+	+	+GP	+NG	+ GP	+ GP	-
CPBM	+	+	-	-	NM	+	+	+ GP	+NG	+ GP	+ NG	-
CPBB	+	+	-	+	NM	+	+	+ GP	+GP	+ GP	+GP	-
CPBG	+	+	-	-	M	+	+	+ GP	+GP	+GP	+GP	-
CPCPW	+	+	-	+	NM	-	+	+ GP	+NG	+ GP	+ GP	-
CPCAC	+	+	+	+	NM	+	+	+ GP	+NG	+ GP	+ GP	-
CPCL	+	+	-	-	M	-	+	-	+NG	+NG	+ NG	-
CPCK	+	+	-	-	M	+	+	+ GP	+GP	+ GP	+ GP	+ GP
CPCM	+	+	-	+	M	+	+	+ GP	+GP	+ GP	+ GP	+ GP
CPCB	+	+	-	-	M	+	+	+ GP	+GP	+ GP	+ GP	+NG
CPCG	+	+	-		NM	-	+	+NG	-	+NG	+ GP	-

Keys: CAT= Catalase test URE= Urease test SH= Starch hydrolysis MAN= Mannitol test SUC= Sucrose test IND= Indole test MOT= Motility test OXI= Oxidase test CIT= Citrate test MAL= Maltose GAL= Galactose NM= Non Motile FRU= Fructose GP= Gas production NG= No gas

Table 4b. Biochemical characteristics sugar fermentation of bacteria isolates in African catfish (*Clarias gariepinus*) from Earthen Pond

Isolates	CAT	IND	OXI	URE	MOT	SH	CIT	MAN	SUC	FRU	MALT	GAL
EPW	+	-	-	-	NM	+	+	+ GP	+ GP	+ GP	+GP	+GP
EPAAC	+	+	-	-	NM	+	+	+ GP	+ NG	+ NG	+ GP	+ GP
EPAL	+	+	-	-	NM	+	+	+ GP	+ GP	+ GP	+ GP	+ GP
EPAK	+	+	-	-	NM	+	+	+ GP	+ GP	+ GP	+ GP	+ GP
EPAM	+	-	+	-	NM	+	+	+ GP	+ GP	+ GP	+ GP	+ GP
EPAB	+	+	+	-	NM	+	+	+ GP	+ GP	+ GP	+ GP	+ GP
EPAG	+	+	-	-	NM	+	+	+ GP	+ GP	+ GP	+ GP	+ GP
EPBAC	+	+	+	-	NM	+	+	+ GP	+ GP	+ GP	+ GP	+ GP
EPBL	+	+	-	-	NM	+	+	+ GP	+ GP	+ GP	+ GP	+ GP
EPBK	+	-	-	-	NM	+	+	+ GP	+ GP	+ NG	+ GP	+ GP
EPBM	+	-	-	-	NM	+	+	+ GP	+ GP	+ GP	+ GP	+ GP
EPBB	+	+	+	-	NM	+	+	+ GP	+ GP	+ GP	+ GP	+ GP
EPBG	+	+	-	-	NM	+	+	+ GP	+ GP	+ GP	+ GP	+ GP
EPCAC	+	-	-	-	NM	+	+	+ GP	+ GP	+ GP	-	+ GP
EPCL	+	-	-	-	NM	+	+	+ GP	+ GP	+ GP	-	-
EPCK	+	-	+	-	M	+	+	+ NG	-	+ GP	-	+ GP
EPCM	+	-	-	-	NM	+	+	+ GP	-	-	-	-
EPCB	+	-	+	-	NM	+	+	-	+ GP	+ GP	-	-
EPCG	+	-	-	-	NM	+	+	+ GP	+ GP	+ GP	-	+ GP
EPDAC	+	-	+	-	NM	+	+	+ GP	+ GP	+ NG	-	+NG
EPDL	+	-	-	-	NM	+	+	+ NG	+ GP	+ GP	-	+ GP
EPDK	+	-	-	-	NM	+	+	+ NG	+ GP	+ GP	-	+NG
EPDM	+	-	-	-	NM	+	+	+ GP	+ NG	+ NG	-	+NG
EPDB	+	-	+	-	NM	+	+	+ GP	+ GP	+ GP	-	+ GP
EPDG	+	-	-	-	NM	+	+	+ NG	+ GP	+ GP	-	+ GP

Keys: CAT= Catalase test URE= Urease test SH= Starch hydrolysis MAN= Mannitol test SUC= Sucrose test IND= Indole test MOT= Motility test OXI= Oxidase test CIT= Citrate test MAL= Maltose GAL= Galactose NM= Non Motile FRU= Fructose GP=Gas production NG= No gas

Table 5. Probable organisms found in African catfish (*Clarias gariepinus*) from Concrete and Earthen Ponds

Isolates	Probable Organisms (Concrete Fish Pond)	Isolates	Probable organisms (Earthen Fish Pond)
CPAPW	*Staphylococcus aureus*	EPW	*Marinococcus halophilus*
CPAAC	*Alcaligenes xylosidans*	EPAAC	*Enterococcus gallinarum*
CPAL	*Alcaligenes paradoxus*	EPAL	*Bacillus cereus*
CPAK	*Acinetobacter calcoaceticus*	EPAK	*Enterobacter aerogenes*
CPAM	*Staphylococcus aureus*	EPAM	*Micrococcus lylae*
CPAB	*Bacillus subtilis*	EPAB	*Staphylococcus aureus*
CPAG	*Pseudomonas putida*	EPAG	*Streptococcus uberis*
CPBPW	*Bacillus subtilis*	EPBAC	*Enterococcus gallinarum*
CPBAC	*Bacillu cereus*	EPBL	*Alcaligenes feacalis*
CPBL	*Citrobacter amalonaticus*	EPBK	*Streptococcus uberis*
CPBK	*Bacillus subtilis*	EPBM	*Micrococcus luteus*
CPBM	*Citrobacter amalonaticus*	EPBB	*Listeria grayi*
CPBB	*Acinetobacter baumanni*	EPBG	*Enterococcus molodoratus*
CPBG	*Bacillus subtilis*	EPCAC	*Sporosarcina inulinus*
CPCPW	*Bacillus subtilis*	EPCL	*Enterococcus molodoratus*
CPCAC	*Listeria grayi*	EPCK	*Bacillus pumilus*
CPCL	*Listeria monocytogenes*	EPCM	*Deinococcus radiodurans*
CPCK	*Bacillus cereus*	EPCB	*Vibrio marinus*
CPCM	*Bacillus cereus*	EPCG	*Citrobacterfreundii*
CPCB	*Bacillus cereus*	EPDAC	*Deinococcus proteolyticus*
CPCG	*Listeria grayi*	EPDL	*Listeria murrayi*
‘	‘	EPDK	*Micrococcus luteus*
‘	‘	EPDM	*Bacillus lautus*
‘	‘	EPDB	*Micrococcus halobius*
‘	‘	EPDG	*Deinobacter grandis*

Keys: CPAPW= concrete pond A pond water, CPAAC= concrete pond A alimentary canal, CPAL= concrete pond A liver, CPAK= concrete pond A kidney, CPAM= concrete pond A mouth, CPAB= concrete pond A body, CPAG= concrete pond A gills, CPBPW= concrete pond B pond water, CPBAC= concrete pond B alimentary canal, CPBL= concrete pond B liver, CPBK= concrete pond B kidney, CPBM= concrete pond B mouth, CPBB= concrete pond B body, CPBG= concrete pond B gills, CPCPW= concrete pond C pond water, CPCAC= concrete pond C alimentary canal, CPCL= concrete pond C liver, CPCK= concrete pond C kidney, CPCM= concrete pond C mouth, CPCB= concrete pond C body and CPCG= concrete pond C gills. GPC= Gram positive cocci, GPR= Gram positive rod and GNR= Gram negative rod.

Keys: EPW= earthen pond water, EPAAC= earthen pond A alimentary canal, EPAL= earthen pond A liver, EPAK= earthen pond A kidney, EPAM= earthen pond A mouth, EPAB= earthen pond A body, EPAG= earthen pond A gills, EPBAC= earthen pond B alimentary canal, EPBL= earthen pond B liver, EPBK= earthen pond B kidney, EPBM= earthen pond B mouth, EPBB= earthen pond B body, EPBG= earthen pond B gills, EPCAC= earthen pond C alimentary canal, EPCL= earthen pond C liver, EPCK= earthen pond C kidney, EPCM= earthen pond C mouth, EPCB= earthen pond C body, EPCG= earthen pond C gills, EPDAC= earthen pond D alimentary canal, EPDL= earthen pond D liver, EPDK= earthen pond D kidney, EPDM= earthen pond D mouth, EPDB= earthen pond D body, EPDG= earthen pond D gills .GPC= Gram positive cocci, GPR= Gram positive rod, GNC= Gram negative cocci and GNR= Gram negative rod

Table 6. Statistical analysis of bacteria colony count of isolates obtained in African catfish (*Clarias gariepinus*) from Concrete and Earthen Ponds

Eviscerated Fish Organ	Concrete Pond	Earthen Pond
Fish Water ($x10^3$)	4.33 ± 1.15^a	4.00 ± 0.00^a
Alimentary canal($x10^3$)	4.73 ± 0.81^a	5.50 ± 0.72^a
Liver ($x10^3$)	3.27 ± 0.64^a	2.20 ± 1.2^a
Kidney ($x10^3$)	3.67 ± 0.58^a	2.95 ± 1.56^a
Mouth ($x10^3$)	3.93 ± 0.31^a	3.25 ± 1.38^a
Fish Body ($x10^3$)	3.53 ± 0.99^a	3.43 ± 1.17^a
Gill ($x10^3$)	4.00 ± 0.69^b	2.33 ± 1.1^a

4. CONCLUSION

This study have shown that fish samples from both earthen and concrete ponds were all colonized with most bacteria species which include normal flora as well as the pathogenic forms of bacteria. The isolation of these organisms from the organs of *C. gariepinus* is problematic, due to their potential risk factors in causing ill-health to human. It is assumed that these organisms might be introduced into the ponds by human healthy carriers through handling. The presence of microorganisms in *C. gariepinus* may constitute a public health risk, due to improperly handle. The sanitary conditions under which fishes are raised or cultured should be improved by following standards or good practices such as good quality water, use of feeds with high microbial quality, regular draining of pond water after specific period of time and closure of ponds to the public.

5. RECOMMENDATION

Farmers should embrace standard operating practices as applicable in fish farming and the workers should be educated on good hygienic practices. They should be provided with necessary working and safety equipment. The need for proper processing and adequate cooking of *Clarias gariepinus,* the African catfish is advocated since it is in high demand at fast food joints popularly known as “point and kill” in most parts of Nigeria. It is also recommended good water quality such as well or borehole should be used in fish pond other than water from questionable sources such as river, also, microbiological analysis and physicochemical examination of pond water for signs of possible contaminants should be conducted on a regular basis and water in the fish pond should be changed regularly.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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