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# Research Article
# Bacterial Flora Associated with *Clarias gariepinus* and *Oreochromis niloticus* in a Natural Habitat

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## Abstract

**Background and Objective:** Fish is a highly desirable foodstuff as it possesses excellent nutritional components, it is man's most important source of high-quality protein. Fish supplies, however, are dwindling due mainly to pollution of the aquatic environment. Toxicants enter fish, they affect fish organs, bioaccumulate and invariably affect the end consumer of such contaminated fish. This study aimed at identifying the bacterial flora present in the gut and gills of *Oreochromis niloticus* and *Clarias gariepinus*. **Materials and Methods:** This research was conducted to assess the bacterial load on fish species found in Ureje Reservoir in Ekiti State. The most popular freshwater fish species in Ureje Reservoir are *Oreochromis niloticus* and *Clarias gariepinus*. Samples of each fish species were kept in sterile bags and transported to the laboratory for bacterial analysis. **Results:** Eight bacteria species were identified (*Staphylococcus aureus*, *Salmonella typhimurium*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Klebsiella pneumonia*, *Proteus* spp., *Enterococcus* spp. and *Streptococcus* spp.). The highest colony count was found in the gut of *Clarias gariepinus*. The presence of these bacteria species could pose a potential public health threat to consumers. **Conclusion:** It is recommended that better handling and processing methods should be adopted to eliminate health risks to fresh fish consumers.

Key words: Bacterial load, *Clarias gariepinus*, *Oreochromis niloticus*, reservoir, consumers

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

## INTRODUCTION

Fish is considered one of the most nutritive and highly desirable foodstuff, as fish meat has excellent nutritional value being rich in proteins, vitamins and unsaturated fatty acids. It is also extremely perishable and the safe consumption requires adequate sanitary conditions from the moment of catch, through preparation, sale and consumption[1]. The most popular freshwater fish species in Ureje Reservoir are *Oreochromis niloticus* and *Clarias gariepinus*. On a global scale, fish and fish products are the most important source of protein and it is estimated that more than 30% of fish for human consumption comes from aquaculture[2,3]. Fishery products are important not only from a nutritional point of view but also as an item of international trade and foreign exchange earner for some countries in the world[3]. Fish and shellfish are highly perishable and prone to vast variations in quality due to differences in species, environment and feeding habits[3]. Fish is a vital source of food for people and contributes about 60% of the world's supply of protein[4]. The 60% of the inhabitants of developing countries derive 30% of their animal protein requirement from fish[4,5]. Today, fish provides more than one billion poor people with most of their daily animal protein. Fish provides nutrients and micronutrients that are essential for cognitive and physical development, especially in children and are an important part of a healthy diet. As an affordable animal source of protein in some of the poorest countries, fish is the primary source of nutrition, creating a growing demand for this staple. However, fish supplies are failing to meet demand and there are major shortages in some critically poor countries where they are needed most. A greater part of the pollutants exhibits biomagnification and bioaccumulation capabilities with a broad spectrum of impacts and stresses on aquatic organisms[6]. This leads to a steady decline in aquatic flora and fauna, particularly fishes. Aquatic organisms, like fish, accumulate pollutants directly from contaminated water and indirectly through the food chain[7,8]. Entry of contaminants into the body of fish may lead to the disruption of physiological functions.

Ureje Reservoir is located on the Ureje River, Ado Ekiti. This reservoir is important in controlling floods and releasing water in the dry season to the surrounding areas. Ureje Reservoir is an artificial lake in Ado Ekiti, it was established by the government to provide water for domestic use to residents of the town. In recent times, there has been the rapid expansion of the town which is also accompanied by poor disposal of sewage and garbage. Runoff may wash some of these poorly disposed of wastes into the water body and contaminate the water and fish species that inhabit it with pathogenic bacteria. This can be a potential threat to the health of people who consume such fish. This work aimed at identifying the bacterial flora present in the gut and gills of *Oreochromis niloticus* and *Clarias gariepinus* which are two of the most abundant fish species found in it.

## MATERIALS AND METHODS

**Study area:** This study was carried out in January to March, 2020. The study area is the Ureje Reservoir. Ureje Reservoir is located on the Ureje River, Ado Ekiti, with a latitude and longitude of 7°35'59"N and 5°12'46"E, respectively.

**Collection of fish samples:** Freshly caught samples of *Oreochromis niloticus* and *Clarias gariepinus* were purchased at the fish landing site from artisanal fishermen operating at Ureje Reservoir. Samples of each species were kept in sterile nylon bags and preserved in an ice chest before being transported to the laboratory for bacterial analysis. Sterile dissecting instruments were used to take samples *Clarias gariepinus* and *Oreochromis niloticus* from the mouth, gill and gut of the fishes' bodies for analysis.

**Morphometric of the fish:** Morphometric parameters of the fish samples such as weight, standard length, head length, gill length and buccal depth were measured with the use of top-loading balance for weights and graduated measuring ruler for lengths. This was done for all the samples of fish taken from the reservoir and their values were recorded to two decimal places.

**Preparation of the serial dilutions:** Distilled water 90-100 mL was dispensed into the conical flask as diluents for each sample and 9 mL of these diluents was dispensed into McCartney bottles for serial dilutions. The diluents were autoclaved at 121°C for 15 min. Nutrient agar was also autoclaved along with the diluents and both were kept to cool.

**Fish samples:** Using sterile dissecting tools, mortar and pestle, samples were taken from the fish (mouth, gill and gut). Each sample was pounded into pieces and properly mixed. Each of these samples was added into 9ml sterile distilled water in the test tubes and thoroughly mixed to make a dilution of $10^{-1}$. Sevenfold serial dilution was made using a sterile pipette ($10^{-1}$ to $10^{-7}$). However further dilution till $10^{-9}$ was made and used where required to allow easy colony counting.

**Inoculation into the solid medium:** The 1 mL of inoculum was pipette into sterile Petri dishes. This was done in duplicates and also labelled sequentially. Using the pour plate method, about 15 mL of sterilized molten Nutrient agar medium, cooled to about 45°C was poured into the inoculated Petri dishes within 15 min of original dilution. Both the sample dilution and agar medium were mixed thoroughly and uniformly and allowed to gel.

Some plates were also prepared as a control to check on the sterility of the diluents, glassware and agar medium. The possibility of air contamination was also assessed with the use of control plates. All poured Petri dishes were incubated in an inverted position at 37°C for 24 hrs.

Using the same procedure described above for the total bacteria count with nutrient agar as a general-purpose medium, the following list of indicator bacteria of faecal and industrial pollution were isolated from the fish organs (mouth, gill and gut) using their respective selective medium: Coliform bacteria and *E. coli* were isolated with MacConkey agar, *Staphylococcus* spp., were isolated with Mannitol salt agar, *Salmonella* spp. and *Shigella* spp., were isolated with *salmonella shigella* agar (SSA), *Vibrio* spp., were isolated with thiosulphate citrate bile salt agar (TCBS), *Streptococcus faecalis* were isolated with blood agar whose 5% is horse blood and lastly, *Clostridium* spp., were isolated with reinforced clostridium agar (RCA).

**Media preparation:** Media used in this study were prepared according to the manufacturer's instruction and sterilized using the autoclave at 121°C for 15 min.

**Nutrient agar:** Dehydrated powder (28 g) of nutrient agar was dissolved in 1000 mL of distilled water in a conical flask, mixed, properly corked and sterilized using the autoclave at 121°C for 15 min.

**Colonial and microscopic examination:** From the isolated colonies, the colonial characteristics were first determined with the colony counter magnifying lens, which was also used to count the numbers of the colony in each plate. Further clarification was then conducted with the use of a light microscope, especially morphological characteristics. The shape and arrangement and some other characteristics of the colonies were examined and recorded[9]. Also, Gram's staining was carried out.

**Biochemical test:** The following biochemical tests were carried out at the microbiology laboratory in Ekiti State University and used to further identify the bacteria isolated and also to identify any other bacteria that could be present.

**Catalase test:** A drop of 3% hydrogen peroxide was placed at the centre of a slide and a sterile wire loop was used to take a small portion of the micro-organism to be identified from the nutrient agar plate into the hydrogen peroxide for immediate gas bubble formation. Quick production of gas bubbles or foaming indicates positive result[9].

**Coagulase test:** A drop of physiological saline was placed on two separate slides. A colony of the test organism was emulsified in each drop to make a suspension. A drop of plasma was then added and mixed gently with the suspension. Clumping (due to coagulation) of the organisms within 10 sec, when viewed under the microscope, indicates a positive result. This was done for the plate suspected to be *Staphylococcus aureus*[9].

**Motility test:** A loopful of growth was inoculated into peptone water broth and incubated overnight. A wet preparation from the peptone water culture was prepared and examined under a microscope at ×40 objective lens. Darting movement of the organism indicate a positive result[9].

**Citrate utilization test:** A slant of a citrate agar was aseptically inoculated with the organisms to be identified using a sterile wire loop. The inoculated citrate agar slant was incubated at 37°C for 24 hrs and was thereafter monitored daily for up to 4 days for possible colour change. Blue colouration indicates positive test[9].

**Indole reaction test:** The micro-organisms to be identified were inoculated into tryptone broth for 48 hrs at 37°C. Five drops of Kovac's reagent was then added. The formation of deep red colour indicates a positive result[9].

**Sugar fermentation test:** Peptone water (7.5 g) was diluted to 500 mL with distilled water after which a few pinches of phenol red was added. 9 mL of broth was distributed into test tubes with Durham tubes inverted into each tube. The tubes were sterilized at 121°C (at 15 pounds pressure) for 15 min. 1% (w/v) aqueous solution of Glucose, Sucrose, Lactose and Mannitol were prepared separately and sterilized. The 1 mL of 1% of the sugar solution was added aseptically using a sterile pipette into each of the test tubes that contained broth. The test organisms were inoculated into each set of test tubes. The test tubes that served as control were, however, not inoculated. Incubation was done at 35°C for 5 days. A change in the colour of the solution from red to yellow indicates acid production and the presence of gas in the inverted Durham tubes indicates gas production[9].

**Oxidase test:** A drop of a freshly prepared oxidase reagent was added onto a strip of filter paper. A little of the test organism was rubbed into it. Change in colour to deep-blue in 5 sec indicates a positive test while non-colouration indicate a negative result[9].

**Statistical analysis:** Data obtained for an abundance of detected bacteria in the mouth, gills as well as gut were compared in the two selected fish species by subjecting them to student t-test, using Standard Statistical Package for Social Sciences (SPSS) version 20.0.

## RESULTS

**Types and abundance of detected bacteria in each sampled organ of *Oreochromis niloticus* and *Clarias gariepinus*:** The bacteria isolated from the mouth, gill and gut of *Oreochromis niloticus* and *Clarias gariepinus* such as *Staphylococcus aureus* (in the mouth of *O. niloticus* 50.0% and *C. gariepinus* 33.33%, also in the gills of *O. niloticus* 66.67% and *C. gariepinus* 33.33% in the study area while the bacterium was only found in the gut of *C. gariepinus*-28.57%), *Salmonella typhimurium* was observed only in *C. gariepinus*: Mouth-50.0%, gill-50.0% and gut-28.57%, *Pseudomonas aeruginosa* was present also only in *C. gariepinus*: Gill-16.67% and gut-14.29%. *Escherichia coli* was found only in the gut (28.57%) of *C. gariepinus* while in *O. niloticus*, it was found in the mouth (33.33%), in the gill (16.67%) and in the gut (25.0%). *Klebsiella pneumonia* was present in the mouth (16.00%) of *C. gariepinus* and the gut (25.0%) of *O. niloticus*. *Proteus* spp., was only observed in *O. niloticus*, in the mouth (16.67%) and the gut (25.0%). *Enterococcus* spp., was only present in *O. niloticus*-in the gill (16.67%) and the gut (12.50%) also, *Streptococcus* spp., was only present in the gut (12.5%) of *O. niloticus* as depicted below in Table 1 and 2.

**Biochemical properties of the bacteria isolated:** Biochemical properties of the isolated bacteria are shown in Table 3. The isolated bacteria tested positive or negative for gram stain tests carried out. Another biochemical tests to which the bacteria either tested positive or negative include, motility, sucrose, glucose, lactose, maltose, mannitol, citrate utilization, indole, catalase, oxidase, methyl red, urease, nitrate reduction.

**Total abundance of each bacterium:** Table 4 shows the total abundance of each species of bacteria detected in each of the fishes analyzed (*Clarias gariepinus* and *Oreochromis niloticus*). *Salmonella typhimurium* was the

Table 1: Bacteria detected in the mouth, gill and gut of *Oreochromis niloticus* and *Clarias gariepinus*

| Organs | *Oreochromis niloticus* | *Clarias gariepinus* |
|---|---|---|
| Mouth | *Staphylococcus aureus* | *Staphylococcus aureus* |
| | *Proteus aureus* | *Klebsiella pneumonia* |
| | *Escherichia coli* | *Salmonella typhimurium* |
| Gill | *Staphylococcus aureus* | *Staphylococcus aureus* |
| | *Enterococcus* spp. | *Salmonella typhimurium* |
| | *Escherichia coli* | *Pseudomonas aeruginosa* |
| Gut | *Klebsiella pneumonia* | *Staphylococcus aureus* |
| | *Proteus* spp. | *Salmonella typhimurium* |
| | *Enterococcus* spp. | *Pseudomonas aeruginosa* |
| | *Escherichia coli* | *Escherichia coli* |
| | *Streptococcus* spp. | |

Field Survey, 2020

Table 2: Abundance of detected bacteria in each examined organ of *Clarias gariepinus* and *Oreochromis niloticus*

| | *Clarias gariepinus* | | | | | | *Oreochromis niloticus* | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Mouth | | Gill | | Gut | | Mouth | | Gill | | Gut | |
| Bacteria species | No. | % | No. | % | No. | % | No. | % | No. | % | % | |
| *Staphylococcus aureus* | 2 | 33.33 | 2 | 33.33 | 2 | 28.57 | 3 | 50 | 4 | 66.67 | 0.00 | |
| *Salmonella typhimurium* | 3 | 50.00 | 3 | 50.00 | 2 | 28.57 | 0 | 0.00 | 0 | 0.00 | 0.00 | |
| *Pseudomonas aeruginosa* | 0 | 0.00 | 1 | 16.67 | 1 | 14.29 | 0 | 0.00 | 0 | 0.00 | 0.00 | |
| *Escherichia coli* | 0 | 0.00 | 0 | 0.00 | 2 | 28.57 | 2 | 33.33 | 1 | 16.67 | 25.00 | |
| *Klebsiella pneumonia* | 1 | 16.00 | 0 | 0.00 | 0 | 0.00 | 0 | 0.00 | 0 | 0.00 | 25.00 | |
| *Proteus* spp. | 0 | 0.00 | 0 | 0.00 | 0 | 0.00 | 1 | 16.67 | 0 | 0.00 | 25.00 | |
| *Enterococcus* spp. | 0 | 0.00 | 0 | 0.00 | 0 | 0.00 | 0 | 0.00 | 1 | 16.67 | 12.50 | |
| *Streptococcus* spp. | 0 | 0.00 | 0 | 0.00 | 0 | 0.00 | 0 | 0.00 | 0 | 0.00 | 12.50 | |

Table 3: Biochemical properties of isolated bacteria

| Grams reaction | Motility | Sucrose | Glucose | Lactose | Maltose | Mannitol | Citrate utilization | Indole | Catalase | Oxidase | Methyl red | Urease | Nitrate reduction | Suspected organisms |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | - | A | A | A | A | A | - | - | | - | | | | *Staphylococcus aureus* |
| - | | - | AG | - | AG | AG | | - | | - | | - | | *Salmonella typhinum* |
| - | | AG | AG | - | - | | | - | | - | - | - | | *Pseudomonas aeruginosa* |
| - | - | AG | AG | AG | AG | AG | - | | | - | | - | | *Escherichia coli* |
| - | - | | | - | | - | | - | | | - | | | *Klebsiella pneumonia* |
| - | | - | | - | - | | | - | | - | | | | *Proteus* spp. |
| | - | | | | | - | - | | - | - | - | - | - | *Streptococcus* spp. |
| | | A | AG | AG | AG | AG | | - | - | - | - | - | | *Enterococcus* spp. |

: Positive reaction of bacteria, -: Negative reaction of bacteria, A: Acid and AG: Acid and gas

most abundant in *Clarias gariepinus* with an abundance of 42.11%, while the least abundant was *Klebsiella pneumonia* (5.26%). In *Oreochromis niloticus*, *Staphylococcus aureus* was the most abundant species (35.00%), while *Streptococcus* sp., was the least abundant (5.00%).

**Total plate count:** Table 5 shows the bacterial load in the mouth, gills and gut of both *Clarias gariepinus* and *Oreochromis niloticus* captured from Ureje Reservoir. In *C. gariepinus*, the highest total plate count of $17.20\pm0.73\times10^8$ CFU $g^{-1}$ was recorded in the gut, while the gill has the lowest value of $7.20\pm0.16\times10^8$ CFU $g^{-1}$. Also, in *O. niloticus*, the gut had the highest total plate count of $10.23\pm2.14\times10^8$ CFU $g^{-1}$, while the gill had the lowest count of $4.17\pm0.53\times10^8$ CFU $g^{-1}$. The overall highest total plate count for the two species was recorded in the gut of *C. gariepinus* ($17.20\pm0.73\times10^8$ CFU $g^{-1}$), while the overall lowest count occurred in the gill of *O. niloticus* ($4.17\pm0.53\times10^8$ CFU $g^{-1}$).

**Interpretation of T-Test result:** The T-Test result to compare the abundance of the detected bacteria in *Clarias gariepinus* and *Oreochromis niloticus* is presented in Table 6. The result showed that the abundance of *Staphylococcus aureus*, in each of the organs examined (mouth (0.413), gills (0.242) and gut (0.951)) was not significantly different ($p>0.05$) at 5, 1 and 10% significance level in both *Clarias gariepinus* and *Oreochromis niloticus*. The occurrence of *Salmonella typhimurium* was significantly different ($p<0.05$) in the mouth (0.042) but was not significantly different in the gill (0.063) and gut (0.500) of both *Clarias gariepinus* and *Oreochromis niloticus* at 1 and 5% at significance level. However, the abundance of *Pseudomonas aeruginosa*, in both *Clarias gariepinus* and *Oreochromis niloticus*, was not significantly different in the mouth (0.500), ($p>0.05$) at 5% significance level, also in the gill (0.063) and gut (0.063) of both fishes, there was no significant difference ($p>0.05$) at 1% significance level. For *Escherichia coli*, there was no significant difference in its abundance in the mouth (0.067), gill (0.202) and gut (1.000) of both *Clarias gariepinus* and *Oreochromis niloticus* ($p>0.05$) at a 1% significance level. The abundance of *Klebsiella pneumonia*, in the mouth (0.866), gill (0.486) and gut (0.129) of both *C. gariepinus* and *O. niloticus* was not significantly different ($p>0.05$) 10, 5 and 1% significance level. There was no significant difference in the abundance of *Proteus* spp. ($p>0.05$) in the mouth (0.244), gill (0.513) and gut (0.294) of both *C. gariepinus* and *O. niloticus* at 10, 5 and 1% significance levels. The abundance of *Enterococcus* spp., in the mouth (0.513) and gill (0.500) was not significantly

Table 4: Total abundance of each bacterium detected in *Clarias gariepinus* and *Oreochromis niloticus*

| | *Clarias gariepinus* | | *Oreochromis niloticus* | |
|---|---|---|---|---|
| Bacteria | Number | Percentage | Number | Percentage |
| *Staphylococcus aureus* | 6 | 31.57 | 7 | 35.00 |
| *Salmonella typhimurium* | 8 | 42.11 | 0 | 0.00 |
| *Pseudomonas aeruginosa* | 2 | 10.53 | 0 | 0.00 |
| *Escherichia coli* | 2 | 10.53 | 5 | 25.00 |
| *Klebsiella pneumonia* | 1 | 5.26 | 2 | 10.00 |
| *Proteus* spp. | 0 | 0.00 | 3 | 15.00 |
| *Enterococcus* spp. | 0 | 0.00 | 2 | 10.00 |
| *Streptococcus* spp. | 0 | 0.00 | 1 | 5.00 |

Table 5: Total plate count for the mouth, gill and gut of *Clarias gariepinus* and *Oreochromis niloticus* (CFU $g^{-1} \pm SD) \times 10^8$

| *Clarias gariepinus* | | | *Oreochromis niloticus* | | |
|---|---|---|---|---|---|
| Mouth | Gills | Gut | Mouth | Gills | Gut |
| 11.97±0.20 | 7.20±0.16 | 17.20±0.73 | 6.90±0.43 | 4.17±0.53 | 10.23±2.14 |

Table 6: Bacterial Composition of *Clarias gariepinus* and *Oreochromis niloticus* in mouth, gill and gut

| Bacteria | Organs | T-value | Std deviation | Mean value of catfish and tilapia | Significant level (2-tailed) |
|---|---|---|---|---|---|
| *Staphylococcus aureus* | Mouth | -1.317 | 1.33 | -1.24** | 0.413 |
| | Gill | -2.500 | 0.56 | -1.800*** | 0.242 |
| | Gut | -0.077 | 1.838 | -0.100* | 0.951 |
| *Salmonella typhimurium* | Mouth | 15.000 | 0.141 | 1.500*** | 0.042 |
| | Gill | 15.000 | 0.141 | 1.500*** | 0.063 |
| | Gut | 10.000 | 0.141 | 1.000** | 0.500 |
| *Pseudomonas aeruginosa* | Mouth | 1.0000 | 0.007 | 0.000** | 0.500 |
| | Gill | 10.000 | 0.07 | 0.500*** | 0.063 |
| | Gut | 10.000 | 0.07 | 0.500*** | 0.063 |
| *Escherichia coli* | Mouth | -9.47 | 0.14 | -0.995*** | 0.067 |
| | Gill | -3.04 | 0.31 | 0.670*** | 0.202 |
| | Gut | 0.00 | 0.84 | 0.00*** | 1.000 |
| *Klebsiella* spp. | Mouth | -0.214 | 0.989 | -0.15* | 0.866 |
| | Gill | 1.047 | 0.304 | 0.225** | 0.486 |
| | Gut | -4.854 | 0.289 | -0.995*** | 0.129 |
| *Proteus* spp. | Mouth | -2.478 | 0.487 | -0.855*** | 0.244 |
| | Gill | -0.959 | 0.346 | -0.235** | 0.513 |
| | Gut | -2.010 | 0.700 | -0995*** | 0.294 |
| *Enterococcus* spp. | Mouth | -0.961 | 0.360 | -0.245** | 0.513 |
| | Gill | -1.000 | 0.240 | -0.170** | 0.500 |
| | Gut | -131.00 | 0.007 | -0.655*** | 0.005 |
| *Streptococcus* spp. | Mouth | -1.000 | 0.311 | -0.0220** | 0.500 |
| | Gill | -1.000 | 0.007 | -0.005** | 0.500 |
| | Gut | -1.047 | 0.304 | -0.225** | 0.500 |

*, **, ***Mean±standard deviation ($p<0.05$) are significantly different

different ($p>0.05$) in both *C. gariepinus* and *O. niloticus* at a 5% significance level while that of the gut (0.005) in both fishes was significantly different at 1% significance level. The abundance of *Streptococcus* spp., was not significantly different ($p>0.05$) in the mouth (0.500), gut (0.500) and gill (0.500) of both *Clarias gariepinus* and *Oreochromis niloticus* at a 5% significant level.

## DISCUSSION

The consumption of fresh African catfish (*Clarias gariepinus*) is on the increase in both rural and urban centres of Nigeria[4]. Fish is an important food commodity that is traded internationally but rapidly deteriorates, in terms of quality, when not properly preserved.

Fish, in their natural habitat, may not be free from bacterial infection but the rate however is highly dependent on some key environmental and climatic factors as well as the species of fish and bacteria in question. Certain environmental factors tend to encourage the rate of infection while others affect bacterial survival and development negatively. The timings of bacterial infection on fish is also, to some extent, controlled by seasonal changes with the consequent changes in climatic conditions. The result from this research shows that

the bacterial load varied in the three parts of the fishes analyzed- the mouth, gills and gut.

The bacterial population isolated from the two fish species analyzed were, *Staphylococcus aureus*, *Salmonella typhimurium*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Klebsiella pneumonia*, *Proteus* spp., *Enterococcus* spp. and *Streptococcus* spp. and this was in agreement with the work of several researchers[10,11] isolated *Shigella* species, *Staphylococcus aureus*, *Salmonella* spp. and *Pseudomonas aeruginosa* from Catfish samples, while another study[12] isolated *Staphylococcus* spp. and *Salmonella* spp., from fresh tilapia fish. Researcher[13] isolated *Staphylococcus aureus*, *Klebsiella* species, *Salmonella* species, *Shigella* species, *Acinetobacter baumannii*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Proteus vulgaris* and *Moraxella catarrhalis.* The distribution of bacteria in the mouth, gut and gill of *Clarias gariepinus* and *Oreochromis niloticus* revealed the occurrence of *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Salmonella typhimurium*, *Klebsiella pneumonia*, *Proteus* spp., *Enterococcus* spp. and *Streptococcus* spp. (Table 1). Although the bacterial flora found in this study did not cause mortality to the fishes probably because the fishes have a strong host defence response, the species have opportunistic and pathogenic tendencies which could make them incite fish disease[14]. In addition, these organisms could also be involved in the transmission of diseases to human beings. Fish and fish products have been reported as vehicles of foodborne bacterial infections in humans[2,15].

Total plate count is an index for measuring total bacterial load. In this study, the highest total plate count was recorded in *C. gariepinus*. This shows that it is more susceptible to bacterial invasion than *O. niloticus* in Ureje Reservoir. Both species of fish were also found to have the highest total plate count in the gut region comparable to the result[16]. The high level of bacterial infection of the gut could be due to low acid concentration in the region, prolific multiplication of naturally occurring bacteria in the region or movement of bacteria from other parts to the gut region where the environment is more conducive for their survival.

In the two fish species examined in this study, the gills had the lowest total plate count compared to the gut and mouth. The quantum of bacteria found on the gills is maintained at a low level, thus implying that fish may have a mechanism that enables it to keep the bacteria number low and therefore be conferred with some degree of protection against bacteria invasion by the gill microflora[12].

*Salmonella* spp., was the most abundant in *C. gariepinus* (with a percentage occurrence of 42.1%) but was not detected in *O. niloticus*. The presence of *Salmonella* spp., indicates faecal contamination of water from which the fishes were harvested[17]. *Salmonella* is a very pathogenic bacterium[18]. It had been reported to cause enteritis and systemic disease[12].

*Escherichia coli* strains have percentage occurrence of 10.53 and 25% in *C. gariepinus* and *O. niloticus*, respectively. This bacteria species have been implicated as the causative agent of urinary tract infection[19]. *Staphylococcus aureus* had the highest percentage occurrence in *O. niloticus* and the second-highest *C. gariepinus*. It has been associated with different clinical conditions. For instance, it is still one of the most frequently encountered single bacteria species in hospitals and continues to be one of the common causative agents of burn wound infections[20].

*Pseudomonas* is a soil bacterium that requires a high water activity for growth, it is known to cause food spoilage (meat, fish) and this is done by secreting lipases and proteases that cause off-odours and the formation of slime[12].

The presence of the isolated organisms was not surprising since the occurrence of human pathogenic bacteria in fish, according to another study[21], can be traced to its direct contact with a contaminated aquatic environment or ingestion of bacterial flora from water sediments. Bacteria flora associated with Nigerian water includes the genera: *Bacillus*, *Lactobacillus*, *Staphylococcus*, *Escherichia*, *Micrococcus* and *Proteus*[12]. Some of these bacteria species such as *Pseudomonas* sp. and *Klebsiella* spp., are pathogenic[22] to exhibit extensive drug resistance.

## CONCLUSION

This research has brought to light the bacterial species associated with *Clarias gariepinus* and *Oreochromis niloticus* and has shown that they are potentially pathogenic to humans. Therefore, all necessary measures must be taken inadequately processing these fishes before consumption. Based on the findings of this study, it is recommended that the government, through the Ministry of Environment, should ensure that industrial and municipal effluents are properly treated before being discharged into water bodies. The banks of Ureje Reservoir and its tributaries should be free of domestic animals, litter and other waste materials. Besides, fish should be properly processed before consumption in other to prevent human beings from being infected with pathogenic bacteria that might have contaminated the fish.

## SIGNIFICANCE STATEMENT

This study discovered the presence of pathogenic bacteria on *O. niloticus* and *C. gariepinus* captured from Ureje Reservoir. This discovery can be beneficial in assisting government to regulate the indiscriminate discharge of industrial effluents and sewage into the Reservoir as well as its tributaries. This study will help researchers to carry out further investigations that will help in devising strategies that are required to prevent further contamination of the Reservoir.

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