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BACTERIOLOGICAL ASSESSMENTS OF SELECTED FISH PONDS AT OWERRI METROPOLIS

Pondei J.O ¹, Ukpong SE ^{2,*}, Stanley JC ³, Amuneke E ² and Stanley H.O ^{1,4}

¹ Department of Microbiology, Faculty of Science, University of Abuja, Federal Capital Territory, Nigeria.

² Department of Microbiology, Faculty of Science, Madonna University, Elele, Rivers State.

³ Department of Obstetrics and Gynecology, Niger Delta University Teaching Hospital, Bayelsa State, Nigeria.

⁴ Department of Microbiology, Faculty of Pure and Applied Science, University of Port Harcourt, Rivers State, Nigeria.

* Corresponding Author

ORCID Details

Ukpong S.E: <https://orcid.org/0009-0001-5769-3819>

Stanley J.C: <https://orcid.org/0009-0007-5972-2832>

Stanley H.O: <https://orcid.org/0000-0001-5280-4869>

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ABSTRACT

This study assessed the bacteriological quality of fish pond water from selected fish ponds in Amakohia Area, Owerri Metropolis, Imo State, Nigeria, to determine the level of microbial contamination and its public health implications. A total of nine (9) pond water samples were collected from three sampling points (A, B, and C) and analyzed using standard microbiological methods. Total Heterotrophic Bacterial Count (THBC), Total Coliform Count (TCC), Fecal Coliform Count (FCC), and bacterial isolation and identification were carried out using conventional culture and biochemical techniques. The THBC ranged from 4.15×10^4 to 5.64×10^4 CFU/mL, while TCC ranged from 11.5 to 18.7 CFU/100 mL and FCC ranged from 5.8 to 9.5 CFU/100 ML. Point B consistently recorded the highest bacterial counts, whereas Point C had the lowest. A total of 42 bacterial isolates were recovered, comprising *Escherichia coli* (23.8%), *Staphylococcus aureus* (19.0%), *Pseudomonas aeruginosa* (16.7%), *Bacillus* spp. (14.3%), *Klebsiella* spp. (11.9%), *Proteus* spp. (9.5%), and *Salmonella* spp. (4.8%). The predominance of *E. coli* and the detection of fecal coliforms indicate fecal contamination of the pond water, while the presence of potential pathogens suggests poor sanitary conditions and possible health risks to fish consumers. The study concludes that the investigated fish ponds were bacteriologically contaminated to varying degrees, with Point B showing the highest level of contamination. Regular microbiological monitoring, improved pond sanitation, proper waste management, routine water replacement, and strict biosecurity measures are recommended to improve water quality, ensure healthy fish production, and protect public health.

Keywords: Bacteriological assessment, Fish-pond water, Aquaculture, Bacterial isolates, Water quality, Public-health.

1 INTRODUCTION

Fish farming, also known as aquaculture, is an essential sector in global food production, providing a substantial source of protein and livelihood for millions of people. In Nigeria, fish farming has gained prominence as a means of meeting the

increasing demand for fish, particularly in urban areas where wild fish stocks are depleting. The town of Owerri, located in the south-eastern part of Nigeria, has witnessed a notable rise in fish farming due to favorable climate conditions and the growing demand for fish in the region (Okoye et al., 2017). However, the growth of this industry is often threatened by various environmental factors, including water quality and microbial contamination, which can negatively affect fish health, productivity, and the safety of the end product for human consumption.

Fish ponds are the primary aquatic habitats in fish farming systems, and their quality is critical in determining the success of the farming process. Water quality parameters such as pH, dissolved oxygen (DO), temperature, salinity, turbidity, and ammonia levels, alongside microbial contaminants, can significantly impact the biological processes of fish and other aquatic organisms. Poor water quality due to improper pond management practices, contamination from surrounding environments, or improper feeding regimes can lead to fish diseases, mass mortality, and compromised nutritional content in farmed fish (Ufodike et al., 2019). Additionally, the presence of harmful bacteria such as *Escherichia coli*, *Salmonella* spp., *Vibrio* spp., and *Aeromonas* spp. in aquaculture environments poses a risk to both aquatic life and human health, particularly when fish from contaminated ponds are consumed (Ogunyemi et al., 2020).

Bacteriological and physiochemical assessments of fish ponds are crucial for understanding the environmental conditions that influence fish health and product safety. The bacteriological quality of the pond water can provide insights into potential sources of contamination, which can arise from organic waste, agricultural runoff, or even improper pond management practices (Akinmoladun et al., 2021). Similarly, the physiochemical properties of pond water such as pH, temperature, oxygen levels, and turbidity play a vital role in maintaining a healthy aquatic ecosystem. Monitoring and managing these factors are essential to optimizing fish production and ensuring that fish from these ponds are safe for human consumption.

Furthermore, the effects of water quality on fish health are profound. Poor water conditions can cause physiological stress, which in turn weakens the immune system and increases the susceptibility of fish to infections and diseases (Boyd & Tucker, 2014). Chronic exposure to suboptimal water quality can lead to delayed growth, decreased reproductive success, and even death. In contrast, maintaining optimal water quality promotes better growth rates, healthier fish, and higher survival rates, which are critical for the economic success of fish farms (Biller et al., 2022).

Water quality also plays a significant role in controlling the spread of pathogens in aquaculture systems. Many pathogens, including bacteria, viruses, and parasites, thrive in polluted or improperly managed water environments. Therefore, regular monitoring and maintenance of water quality parameters can help reduce disease outbreaks and improve biosecurity (Pérez-Sánchez et al., 2020). Pathogenic bacteria can be introduced into aquaculture systems through contaminated feed, poor sanitation, fish faeces, and water exchange with nearby bodies of water. Regular monitoring and control of bacterial levels are crucial to prevent disease outbreaks and maintain a healthy aquaculture environment, also pathogenic bacteria can disrupt normal metabolic processes in fish, leading to stress, weakened immunity, reduced growth rates, and, in severe cases, mortality. Bacteria such as *Aeromonas hydrophilia* produce toxins that damage fish tissues, particularly gills and internal organs, leading to secondary infections. Infected fish may exhibit symptoms like erratic swimming, lesions, and reduced feeding behaviour, which ultimately compromise the economic viability of aquaculture operations (Tucker & Robinson, 2014).

Regular bacteriological testing of water is essential to detect early signs of contamination, enabling timely intervention and reducing the risk of widespread disease outbreaks (Pérez-Sánchez et al., 2020).

Recent studies on fish pond water quality in various parts of Nigeria, including Owerri, have highlighted significant variations in both bacteriological and physiochemical parameters across different farming systems (Akinmoladun et al., 2021; Olayemi et al., 2022). Despite the growing interest in fish farming in the Owerri area, there remains a gap in comprehensive research focusing on the combined bacteriological and physiochemical assessment of local fish ponds. This study aims to fill that gap by investigating the microbiological and physiochemical conditions of selected fish ponds in Owerri and providing recommendations for improving pond management practices to enhance fish health and product safety.

The findings of this research shall contribute to the body of knowledge on sustainable aquaculture practices in the region, with particular emphasis on water quality management, disease prevention, and the overall improvement of the

fish farming industry. The study also informs policymakers and aquaculture stakeholders about the importance of regular water quality monitoring to ensure a safe and healthy environment for both farmed fish and consumers.

2 MATERIALS AND METHODS

2.1 Study Area

The study was carried out in selected fish ponds within Owerri Metropolis, Imo State, Nigeria. Owerri is located at approximately 5°28'N latitude and 7°03'E longitude. The metropolis consists of both small-scale and commercial fish farming operations, primarily utilizing earthen ponds. These ponds are situated in urban and peri-urban areas, where they are influenced by environmental factors such as agricultural runoff and waste disposal practices. The region experiences a tropical climate, with distinct wet and dry seasons that affect water quality and pond management practices. The diverse fish farming practices in the area provide a representative sample for evaluating the bacteriological conditions of the ponds.

2.2 Sample Collection

Water samples were collected from nine (9) selected fish ponds in Owerri Metropolis, representing both small-scale and commercial aquaculture systems. Samples were collected during the dry and wet seasons to assess seasonal variations. For bacteriological analysis, water samples were collected using sterile bottles from three different locations within each pond: surface, middle, and bottom layers to ensure consistency. All samples were transported to the laboratory in ice-packed containers and analyzed within 24 hours to maintain sample integrity. Standard procedures were carried out for bacteriological assessments.

2.3 Media Preparation for Bacteriological Investigation

For the bacteriological analysis of pond water samples, standard microbiological media were such as Nutrient agar (NA), MacConkey agar, and *Salmonella-Shigella* agar (SS agar) were utilised, these culture media were prepared according to established protocols, by dissolving the appropriate amounts of powder in distilled water, followed by sterilization through autoclaving at 121°C for 15 minutes. The media were allowed to cool and poured into sterile Petri dishes under aseptic conditions. These media facilitated the isolation and enumeration of bacteria such as *Escherichia coli*, *Salmonella spp.*, and *Vibrio sp.* from the water samples (Ogbu et al., 2020).

2.4 Bacteriological analysis

Serial dilution of the water sample was performed according the method described by Chukwu et al., (2022), to quantify bacterial populations in the selected fish ponds at Owerri Metropolis. Briefly a 10 mL aliquot of each water sample was mixed with 90 mL of sterile saline solution to achieve a 1:10 dilution. Subsequent dilutions (10^{-1} to 10^{-6}) was prepared by transferring 1 mL of each diluted sample into 9 mL of sterile saline solution. For bacteriological analysis, 0.1 mL of each sample from the serial dilution was aseptically spread on the surface of selective agar media. Nutrient Agar (NA) was used for the enumeration of total bacterial counts, while MacConkey Agar was employed to isolate and quantify coliform bacteria, while *Salmonella-Shigella* Agar was used for detecting enteric pathogens such as *Salmonella* and *Shigella* species. The inoculated plates were incubated at 37°C for 24-48 hours to allow bacterial growth. All plates were prepared in duplicate. After incubation, colonies were counted and recorded as colony-forming units (CFU). Colonies were counted and recorded after incubation period. The different colonies on each plate were isolated, purified and stored on nutrient agar slants for further characterization and identifications.

2.5 Characterization and Identification of isolated bacterial Species

The bacteria isolates obtained were identified based on a combination of colonial, morphological and biochemical tests; these tests were conducted to determine the cultural characteristics of the isolates. The biochemical tests were conducted as described by Chesbrough, (2006) and include; catalase test, oxidase test, glucose test, glucose fermentation tests and hydrolysis of arginine.

2.6 Total Heterotrophic and Fecal Coliform Test

Water samples from the selected fish ponds were tested for the presence of fecal coliforms and heterotrophic bacterial species using the method described by ISO, (2014). The total heterotrophic count and fecal coliform test were assessed and determined on nutrient agar using the pour plate technique. One ml of each serially diluted sample was aseptically inoculated onto various sterile nutrient agar plates in triplicate; the inoculums were evenly distributed on the superficial layer of the media. Immediately, the plates were incubated at 37°C for 24 hours. Thereafter, the distinct colony growth of the isolates that developed were adequately counted using a counting chamber and expressed as colony-forming units per milliliter (CFU/ml) (Ogbonna, 2019).

2.7 Antibiotic Susceptibility Testing

Disc diffusion method as described by Haley *et al.*, (2024) was used to determine the susceptibility of the bacterial isolates to selected antibiotics was tested on the Kirby- Bauer agar. The following antibiotic disk were used Augmentin (30µg), gentamicin (10µg), ofloxacin (5µg), ciprofloxacin (5µg), ceftazidime (30µg), cefixime (30µg), erythromycin (15µg), and tetracycline (30µg). The prepared sterile Mueller Hinton agar was adequately dispensed into sterile petri dishes and allowed to solidify. Thereafter, 0.1ml of the bacteria isolates were streaked on the surface of the well- dried agar plates using a sterilized hockey stick. The multiple antibiotic discs were gently and firmly placed on the agar plates using a sterilized forceps. The plates were incubated at 35-37° C for 18-24hours.

Zones of clearance or inhibition were observed, measured to the nearest millimeter and recorded accordingly. The zones clarity was interpreted as sensitive, intermediate or resistant for each of the assayed antimicrobial agent, based on the recommended standard (CLSI, 2017). This testing provided valuable information on the potential for antibiotic resistance in bacterial pathogens in the fish ponds, highlighting the need for prudent use of antibiotics in aquaculture practices (Shang *et al.*, 2019; Iwegbue *et al.*, 2020).

2.8 Statistical Analysis

The data obtained from the bacteriological assessment of the fish ponds in Owerri Metropolis was analysed using descriptive statistics, including mean, standard deviation, and range, to summarize the variations in water quality parameters and bacterial load. The differences in microbial counts across different pond samples were analysis using one-way analysis of variance (ANOVA), with post-hoc tests to identify significant differences between the groups. Correlation analysis was done to assess the relationship between physicochemical parameters and bacterial contamination. All statistical analyses were conducted using SPSS version 20, and results was considered statistically significant at $p < 0.05$.

2.9 Ethical Considerations

This study adhered to ethical guidelines to ensure the welfare of the environment and public health. Informed consent was obtained from the fish farmers involved, ensuring their voluntary participation. The collection of water and sediment samples from the fish ponds was carried out with minimal disruption to the farming activities. All procedures were conducted with the aim of enhancing the sustainability of aquaculture practices, and confidentiality regarding the identity and location of the ponds was maintained. The study also adhered to safety standards in handling microbial samples to prevent contamination and harm to researchers and the surrounding community.

3 RESULTS AND DISCUSSION

3.1 Mean Total of Bacterial Counts of Fish Pond water samples collected from three sampling points in Amakohia Area, Owerri Metropolis.

Table 1 presents the results of the mean total bacterial counts of fish pond water samples collected from three sampling points in the Amakohia area of Owerri Metropolis. Sampling Point B recorded the highest Total Heterotrophic Bacterial Count (THBC) ($5.64 \pm 0.20 \times 10^4$ CFU/mL), Total Coliform Count (TCC) (18.7 ± 1.5 CFU/100 mL), and Faecal Coliform Count (FCC) (9.5 ± 1.0 CFU/100 mL). In contrast, Sampling Point C had the lowest values for all bacteriological parameters, with a THBC of $4.15 \pm 0.12 \times 10^4$ CFU/mL, TCC of 11.5 ± 0.9 CFU/100 mL, and FCC of 5.8 ± 0.6 CFU/100 mL. Overall, the results indicate that bacterial contamination was highest at Point B, followed by Point A, while Point C exhibited the lowest microbial load.

3.2 Frequency and Percentage Occurrence of Bacterial Isolates Recovered from Selected Fish Ponds in Amakohia Area, Owerri Metropolis.

Table 2 presents the frequency and percentage occurrence of bacterial isolates recovered from selected fish ponds in Amakohia Area, Owerri Metropolis. A total of 42 bacterial isolates were identified, with *Escherichia coli* recording the highest occurrence (10 isolates; 23.8%), followed by *Staphylococcus aureus* (8 isolates; 19.0%) and *Pseudomonas aeruginosa* (7 isolates; 16.7%). *Bacillus* spp., *Klebsiella*, and *Proteus* spp. accounted for 14.3%, 11.9%, and 9.5%, respectively, while *Salmonella* had the lowest occurrence (2 isolates; 4.8%). Overall, the predominance of enteric and opportunistic bacterial species suggests varying levels of microbial contamination within the fish pond water samples.

3.3 Distribution of Bacterial Isolates According to Sampling Points in Amakohia Area, Owerri Metropolis.

Table 3 shows the distribution of bacterial isolates recovered from the three sampling points in the selected fish ponds located in Amakohia Area, Owerri Metropolis. A total of 42 bacterial isolates were identified, with Point B recording the highest number of isolates (20), followed by Point A (12), while Point C had the lowest (10). *Escherichia coli* was the

most frequently isolated bacterium (10 isolates), whereas *Salmonella* spp. was the least frequently recovered (2 isolates). The findings indicate that Point B had a greater level of bacterial contamination than the other sampling points.

3.4 Antibiotic susceptibility resistance profile of bacterial isolates from the fish ponds

Table 4 showed the antimicrobial susceptibility pattern of the probable bacteria in mm of positive disk with *Bacillus* specie having the zone of inhibition of 34mm on (PEF) pefloxacin followed by (OFX) tarivid 32mm, (SXT) septrin 30mm, (SP) sparfloxacin 28mm, (CPX) ciprofloxacin 28mm, (AM) amoxicillin 24mm, (CH) 16mm and resistance to (CH) chloramphenicol, (AU) augmentin and streptomycin. *Staphylococcus aureus* with the zones of bition of 32mm on (SP) sparfloxacin, (CPX) ciprofloxacin, and (AM) amoxicillin, followed by (SXT) septrin 30mm, (CN) gentamycin 30mm, (PEF) pefloxacin 30mm, AU) augmentin 28mm, CN gentamycin 24mm. *Staphylococcus* showed no resistance to the antibiotics. **Table 5** showed the antimicrobial susceptibility pattern of the probable bacteria in mm of negative disk with *Salmonella*, *Vibrio* species having the highest zone of inhibition of 30mm on (SXT) septrin. All the gram-negative isolates showed resistance to (AM).

Table 1: Mean Total of Bacterial Counts of Fish Pond water samples collected from three sampling points in Amakohia Area, Owerri Metropolis.

Sampling point	THBC (X10 ⁴ CFU/mL)	TCC (CFU/100mL)	FCC (CFU/100mL)
Point A	4.82±0.15	14.3±1.2	7.2±0.8
Point B	5.64±0.20	18.7±1.5	9.5±1.0
Point C	4.15±0.12	11.5±0.9	5.8±0.6

Key: THBC=Total Heterotrophic Bacterial Count, Total Coliform Count, FCC= Faecal Coliform Count, CFU=Colony Forming Unit.

Table 2: Frequency and Percentage Occurrence of Bacterial Isolates Recovered from Selected Fish Ponds in Amakohia Area, Owerri Metropolis.

Bacterial Isolates	Frequency (n)	Percentage (%)
<i>Escherichia coli</i>	10	23.8
<i>Staphylococcus aureus</i>	8	19.0
<i>Pseudomonas aeruginosa</i>	7	16.7
<i>Bacillus</i> spp.	6	14.3
<i>Klebsiella</i>	5	11.9
<i>Proteus</i> spp.	4	9.5
<i>Salmonella</i>	2	4.8
Total	42	100

Table 3: Distribution of Bacterial Isolates According to Sampling Points in Amakohia Area, Owerri Metropolis.

Bacterial Isolates	Point A	Point B	Point C	Total
<i>Escherichia coli</i>	3	5	2	10
<i>Staphylococcus aureus</i>	2	4	2	8
<i>Bacillus</i> spp.	2	2	2	6
<i>Klebsiella</i>	2	2	1	5
<i>Proteus</i> spp.	1	2	1	4
<i>Salmonella</i>	0	2	0	2
<i>Pseudomonas aeruginosa</i>	2	3	2	7
Total	12	20	10	42

Table 4: Diameter of Inhibition Zones (mm) Produced by Antibiotics against Gram-Positive Bacterial Isolates obtained from Selected Fish Ponds

Organisms	CIP	OFX	GEN	CRO	CAZ	AUG	TET	ERY
<i>Bacillus spp</i>	31±1.0	28±0.8	25±0.7	22±0.8	23±0.9	14±0.7	19±0.8	24±1.0
<i>Staphylococcus spp</i>	28±1.2	26±1.0	23±0.8	19±0.9	20±1.0	11±0.8	13±0.9	24±0.8

Table 5: Diameter of Inhibition Zones (mm) Produced by Antibiotics against Gram-Negative Bacterial Isolates obtained from Selected Fish Ponds

Organisms	CIP	OFX	GEN	CRO	CAZ	AUG	TET	EYY
<i>Salmonella spp.</i>	28±1.1	27±0.9	26±0.9	17±1.2	20±0.9	11±0.7	14±0.8	Nil
<i>Klebsiella spp.</i>	30±1.0	30±1.0	28±1.1	19±0.9	22±1.4	10±0.8	16±0.9	Nil
<i>E.coli</i>	31±1.2	29±1.0	25±0.8	18±8.9	20±1.1	12±0.8	14±0.8	Nil
<i>P.auroginosa</i>	29±1.3	31±1.1	26±1.0	17±0.8	18±0.9	8±0.6	11±0.9	Nil
<i>Proteus spp.</i>	29±1.0	27±1.1	24±0.9	17±0.9	19±0.1	10±0.5	12±0.7	Nil

Table 6: Morphological and Biochemical Characteristics of bacterial isolates obtained from Selected Fish Ponds in Amakohia Area, Owerri Metropolis

Gram rxn	Shape	Catalase	Coagulase	Motility	Starch hyd.	Citrate	Urease	MR	VP	Spore and hyphae	Dnase	Lip	Gala	Glucose	Malt	Xylose	Lactose	Fructose	Sucrose	Manitol	Galactose	Dextrose	Probable organisms
+ ve	Rods	+	+	-	-	-	-	-	-	A	+	-	-	A	A	A	A	AG	-	AG	A	AG	Bacillus spp
- ve	Rods	+	-	-	-	-	-	+	-	-	-	-	-	A/G	A	A/G	A/G	A	A	-	AG	A	Salmonella spp
+ ve	Cocci	+	-	+	+	-	-	-	+	+	-	-	-	A	A	A	A	A	A	-	A	A	Staphylococcus aureus
- ve	Rods	+	-	+	+	-	-	-	-	+	+	-	-	A	A	A/G	A	A	-	-	AG	-	Proteus spp
- ve	Rods	+	-	-	-	-	-	+	-	+	-	-	-	A	AG	A/G	A/G	A	AG	-	A	A	Escherichia coli
- ve	Rods	+	-	-	-	+	+	+	-	-	-	+	+	A	A	A	A	A	-	-	A	-	Klebsiella spp.
-ve	Rods	+	-	-	+		+	+	-	±	-	+	+	A	A	A	A	A	-	A	A	+	P.aeruginosa

Key: A/G = Acid/Gas positive, + = Positive, - = Negative, VP = Voges Proskauer, MR = Methyl red,

4 DISCUSSION

The bacteriological assessment of fish pond water collected from three sampling points in Amakohia Area, Owerri Metropolis revealed that all the ponds contained considerable bacterial populations, although the level of contamination differed among the sampling points. The Total Heterotrophic Bacterial Count (THBC) ranged from 4.15×10^4 CFU/mL at Point C to 5.64×10^4 CFU/mL at Point B. Likewise, the Total Coliform Count (TCC) varied between 11.5 and 18.7 CFU/100 mL, while the Faecal Coliform Count (FCC) ranged from 5.8 to 9.5 CFU/100 mL. Point B consistently recorded the highest microbial counts, whereas Point C had the lowest values. These findings indicate that microbial contamination was not evenly distributed across the ponds and may have resulted from differences in pond management practices, stocking density, feeding intensity, frequency of water replacement, and contamination from surrounding environments.

The relatively high heterotrophic bacterial counts obtained in this study indicate that the pond water contained abundant microorganisms capable of utilizing the organic materials present in the aquatic environment. Fish ponds are generally rich in organic matter due to uneaten fish feed, fish faeces, decaying vegetation, and runoff from nearby agricultural or residential areas. These materials provide nutrients that encourage bacterial growth and multiplication. The higher THBC observed at Point B indicates higher organic pollution and poorer sanitary conditions compared with the other sampling points. This observation corroborates with the findings of Sule et al. (2023), who reported significantly higher heterotrophic bacterial loads in intensively managed fish ponds due to excessive feed residues and accumulated organic wastes. The agreement between both studies suggests that poor waste management and organic enrichment remain major factors influencing bacterial proliferation in aquaculture systems.

The occurrence of *Pseudomonas aeruginosa* in the ponds was expected because species of the genus *Pseudomonas* are common inhabitants of aquatic environments. They are opportunistic pathogens capable of causing diseases in fish, especially when fish are stressed by poor environmental conditions. This finding is consistent with Aina and Olaleye (2023), who isolated *Pseudomonas aeruginosa* from fish ponds in Lagos State, Nigeria. The similarity between both studies may be explained by the ability of this organism to survive and multiply in nutrient-rich aquatic environments containing decomposing organic matter.

The isolation of *Bacillus* spp. reflects their natural occurrence in soil, sediments, and water bodies. Their ability to produce resistant spores enables them to survive under adverse environmental conditions. Consequently, their presence in the ponds is more likely to represent the normal microbial flora of the aquatic ecosystem rather than direct faecal contamination. Similar observations have been reported in previous Nigerian studies where *Bacillus* species were frequently recovered from fish pond environments. The recovery of *Klebsiella* and *Proteus* species also suggests contamination from sewage, organic wastes, or agricultural run-off entering the ponds. These bacteria are recognized as opportunistic pathogens and are commonly associated with polluted aquatic environments. Their occurrence agrees with the findings of Chitambo et al., (2024) who isolated both *Klebsiella* spp. (6.8%) and *Proteus* spp. (1.4%) from cultured Nile tilapia and their water, demonstrating that both organisms occur in aquaculture environment and linked their presence to environmental pollution and poor sanitary conditions within aquaculture facilities.

Although *Salmonella* spp. recorded the lowest occurrence (4.8%), its detection remains important because even low numbers of *Salmonella* can cause serious foodborne illnesses in humans. The organism was detected at low frequency, possibly because environmental conditions within the ponds were less favourable for its survival than for other bacterial species. Nevertheless, its presence confirms occasional faecal contamination of the ponds and highlights the need for continuous microbiological surveillance. This finding corroborates with the results of Murphy et al., (2024), who reported that only 13% of pond water samples were culture positive occasionally for *Salmonella* spp. from freshwater fish ponds.

The distribution of bacterial isolates among the sampling points further demonstrated variations in microbial contamination within the study area. Point B recorded the highest number of bacterial isolates (20 isolates), followed by Point A (12 isolates), while Point C had the lowest number (10 isolates). This distribution indicates that Point B experienced greater microbial contamination than the other ponds. The higher bacterial occurrence at Point B may be associated with accumulation of organic matter, fish waste, uneaten feed, poor water circulation, and runoff carrying contaminants into the pond. Such conditions provide favourable environments for bacterial growth. This observation agrees with Maduwuba et al. (2024), who reported differences in bacterial distribution among fish ponds in Owerri and attributed the variations to differences in pond management, waste accumulation, and environmental conditions.

Among the bacterial isolates recovered across the sampling points, *Escherichia coli* recorded the highest frequency (10 isolates), further confirming that faecal contamination was a major source of bacterial pollution in the ponds. Similar findings were reported by Jacob et al. (2024), who identified *E. coli* as one of the predominant bacterial species in fish

pond ecosystems in Nigeria and concluded that faecal contamination remains a common challenge in aquaculture environments.

The relatively high occurrence of *Staphylococcus aureus* (8 isolates) and *Pseudomonas aeruginosa* (7 isolates) also reflects poor sanitary conditions within the ponds. While *Staphylococcus aureus* is commonly introduced through human activities during pond maintenance and fish handling, *Pseudomonas aeruginosa* survives naturally in aquatic environments because of its remarkable adaptability. These findings agree with Maduwuba et al. (2024), who isolated both organisms from fish pond wastewater in Owerri and associated their presence with poor sanitation and environmental contamination.

Similarly, the recovery of *Bacillus spp.* (6 isolates), *Klebsiella spp.* (5 isolates), and *Proteus spp.* (4 isolates) demonstrates the diversity of bacterial populations present within the ponds. *Bacillus* species probably entered the ponds through soil particles, dust, and sediments, whereas *Klebsiella* and *Proteus spp.* are more closely associated with organic pollution and faecal contamination. Their occurrence suggests that poor water quality and environmental inputs contributed to bacterial contamination, a finding that is consistent with recent Nigerian studies on aquaculture water quality.

Only two isolates of *Salmonella spp.* were recovered, and both originated from Point B. Although the frequency was low, the presence of this pathogen is of considerable public health significance because *Salmonella* is a major cause of foodborne infections worldwide. Its occurrence suggests occasional faecal contamination within the pond environment and emphasizes the importance of regular microbiological monitoring. This finding agrees with Jacob et al. (2024), who also reported *Salmonella* species in selected fish pond ecosystems in Nigeria and stressed the need for proper hygiene and effective water quality management.

Overall, the findings of this study demonstrates that the selected fish ponds in Amakohia Area, Owerri Metropolis were contaminated with both environmental bacteria and organisms associated with faecal pollution. The relatively high bacterial counts, together with the predominance of *Escherichia coli*, *Staphylococcus aureus*, and *Pseudomonas aeruginosa*, indicate inadequate sanitation and poor water quality within some of the ponds, particularly at Point B. These findings are generally consistent with previous studies conducted in Nigeria, which have shown that poor pond management, contaminated water sources, excessive organic matter, and inadequate waste disposal significantly contribute to microbial contamination in aquaculture systems. Consequently, routine microbiological monitoring, proper waste disposal, controlled feeding practices, regular pond sanitation, effective water replacement, and improved biosecurity measures are recommended to enhance water quality, promote healthy fish production, safeguard consumer health, and ensure sustainable aquaculture development.

Seven bacterial species were isolated from the pond water samples. These included *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Bacillus spp.*, *Klebsiella spp.*, *Proteus spp.*, and *Salmonella spp.* Among these organisms, *Escherichia coli* was the most frequently isolated bacterium, accounting for 23.8% of the total isolates, while *Salmonella spp.* had the lowest occurrence (4.8%). The predominance of *E. coli* indicates faecal contamination of the ponds, whereas the presence of other pathogenic bacteria suggests poor environmental sanitation and possible contamination from organic wastes, runoff, and fish handling activities.

Furthermore, in this study, all antibiotic disc diffusion were tested using the disc diffusion method using a separated disk (positive and negative disk), the antibiotic susceptibility testing among the bacterial strains obtained from selected ponds in Amakohia Area, Owerri Metropolis showed a considerable difference in their response to the tested antibiotics. The gram -negative bacterial isolates generally exhibited higher zones of clearance with ciprofloxacin, gentamycin and ofloxacin, whereas, minimal zones of clearance were noticed with augmentin, tetracycline and ceftazidime. Among the gram-positive strains, ciprofloxacin, gentamycin also produced relatively high zones of inhibition. The observed pattern shows that fluroquinolones and aminoglycosides were more active against many of the isolates, while resistance or lower susceptibility was more pronounced with some β -lactam and tetracycline antibiotics.

The significant high susceptibility of isolates to ciprofloxacin, and its resistance to tetracycline observed in this present research is similar to the findings of Apenteng et al. (2017), who discovered that bacterial isolates from fish ponds in the greater Accra region of Ghana, where ciprofloxacin, demonstrated 90% susceptibility against *Salmonella typhi* species and 80% susceptibility against *Escherichia coli*. Gentamycin also showed considerable activity with 70% susceptibility among *Salmonella typhi* and 66.66% among *Escherichia coli*. Whereas, they reported that 90% of *Salmonella typhi* and 66.66% of *E. coli* isolates developed resistance to tetracyclines, which they suggested could be as a result of prolonged exposure to antibiotics that are commonly used in aquaculture which may have contributed to the development and maintenance of resistant bacterial populations.

The relatively high susceptibility of isolates to ofloxacin also agrees with the research of Njoku et al. (2015), who worked with concrete and earthen fish ponds in Niger Delta region of Nigeria, reported 85% and 75% susceptibility to ofloxacin in isolates from earthen and concrete ponds, respectively. Gentamicin also showed potential activity, with 91%

susceptibility in concrete ponds and 50% in earthen ponds. Showing that susceptibility patterns may differ according to environment and bacterial population.

A more recent research work by Omotoso et al. (2024), further supports the occurrence of antibiotic resistance in fish-pond environments. Where they investigated fish pond water and sediment from selected ponds in Osun State and recovered organisms including *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Klebsiella* spp., and *Salmonella* spp. Their study reported considerable resistance to several antibiotics, including; ceftazidime, cefotaxime, cefuroxime, gentamycin, and ciprofloxacin, showing that resistance bacteria are present in Nigerian aquacultural environments.

This present work in this context adds new knowledge by providing baseline information on the bacterial content and antimicrobial susceptibility patterns of microorganisms associated with selected fish ponds in Amakohia Area, Owerri Metropolis. It showed the occurrence of highly disease-causing and antibiotic-resistant bacterial in aquaculture environment thereby providing useful information to fish farmers on the necessity for effective strategic management of fish ponds, good sanitation, proper and regular monitoring of water quality which will at the same time improve fish health, produce safe fish, and promote sustainable fish farming and as well as ensuring a safe environment and protecting public health.

5 CONCLUSION

The findings of this study showed that the fish ponds in Amakohia Area, Owerri Metropolis were contaminated with bacteria at different levels. The presence of heterotrophic bacteria, total coliforms, and faecal coliforms indicates poor water quality and faecal contamination. Point B had the highest bacterial count, suggesting poorer sanitary conditions than the other sampling points. Potentially harmful bacteria such as *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Klebsiella* spp., *Proteus* spp., and *Salmonella* spp. were also isolated, showing that the pond water may pose health risks to both fish and humans if not properly managed. Thus, good sanitation, proper pond management, and regular monitoring of water quality are necessary to improve fish health, produce safe fish, and promote sustainable fish farming.

STATEMENTS ON COMPLIANCE WITH ETHICAL STANDARDS

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Disclosure of conflict of interest

Authors have declared that no competing interest exist.

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