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Assessment of Bacteriological Quality of Public Swimming Pool Water in Ile-Ife, Nigeria

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ABSTRACT: Swimming pools are important recreational resources in urban areas, contributing to physical fitness, social interaction, and mental well-being. Consequently, the objective of this study is to assess the bacteriological quality of public swimming pool water in Ile-Ife, Nigeria using standard methods. The pH and temperature of the water samples ranged from 7.1 to 7.4, and 23.6°C to 26.5°C respectively. Eight of the ten pools tested positive for bacterial contamination. Six different Gram-negative bacteria (GNB) were isolated, with *Escherichia coli* (32%), Klebsiella *pneumoniae* (16%), Salmonella subsp. (12%) and Pseudomonas *aeruginosa*. (8%), being the most prevalent. The coliform bacteria count ranged from 1.0×10^2 CFU/100ml to 9.8×10^2 CFU/100ml and the *Salmonella* count ranged from 1.0×10^2 CFU/100ml to 3.0×10^2 CFU/100ml. Most of the GNB isolates were resistant to commonly administered antibiotics like Augmentin (62.5%), and ampicillin (57.6%) but were totally susceptible to quinolones and carbapenem. This study highlights the significant public health threat posed by fecal contamination and the presence of antibiotic-resistant bacteria in outdoor swimming pools in Ile-Ife, underscoring the need for improved pool maintenance and monitoring.

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Swimming pools serve as valuable recreational resources in urban areas, promoting physical activity, social interaction, and mental well-being (Overbury *et al.*, 2023). However, inadequate pool maintenance can lead to microbial contamination, posing significant health risks to users (Cabral, 2010; Fantuzzi *et al.*, 2023). Contaminated recreational water, including swimming pools, can serve as a transmission route for waterborne pathogens like *E. coli*, a fecal indicator organism, raising concerns about the potential spread of diarrhoeal diseases, ear infections, and skin irritations (Fantuzzi *et al.*, 2023; Hassanein *et al.*, 2023). The emergence and spread of antibiotic-resistant bacteria further complicate the management of waterborne infections and pose additional challenges for public health (Bobate *et al.*, 2023). Previous research has underscored the public health risks associated with microbial contamination in swimming pools. Lohff *et al.* (2001) reported significant bacterial contamination levels in recreational waters, emphasizing the need for regular monitoring and proper maintenance practices; Omotayo *et al.* (2016) identified various sources of microbial contamination in swimming pools, including bathers, environmental factors, and inadequate disinfection, Saberianpour *et al.* (2015)

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and Yedeme *et al.* (2017) further highlighted the health risks posed by contaminated recreational waters, underscoring the importance of stringent disinfection protocols and routine water quality assessments. Emerging antibiotic resistance among bacterial pathogens in recreational waters is an escalating challenge highlighting the potential for treatment complications and the need for more effective strategies to mitigate this threat (Zhang *et al.,* 2015). Consequently, the objective of this paper is to assess the bacteriological quality of public swimming pool water in Ile-Ife, Nigeria

MATERIALS AND METHODS

Study Area: The study was a cross-sectional study conducted on outdoor swimming pools in Ile-Ife, Osun State, Nigeria, at the Microbiology Unit of the Biological Sciences Department, Oduduwa University. Ile-Ife, a city with a population exceeding 500,000 (NPC, 2009) hosts numerous public swimming pools, making it an ideal location to assess the bacteriological quality of recreational waters.

Sample Collection: Ten (10) outdoor swimming pools (labeled A-J) were collected using random sampling technique between July and September 2023 for the study, accompanied by a research proforma. Sterile 250ml screw-cap bottles were used to collect water samples from different points within each pool to ensure a representative sample. To avoid interference from bathers, water samples were collected when the pools were unoccupied. The collection point was approximately 50 cm away from the pool edge, at a depth of 30 cm (Onuorah *et al.,* 2017). Subsequently, the samples were transported within 1 hour of collection time, while being kept at 4°C using appropriately insulated coolers.

Inclusion Criteria: The study recruited recreational centers within Ile-Ife, Nigeria, who willingly provided consent to participate

Exclusion Criteria: Recreational centers that expressed willingness to participate by filling research pro forma but refused to allow water sample collection from their swimming pool were not considered eligible for inclusion in the study. They were consequently disqualified from participation.

Determination of pH and temperature: The pH of the swimming pool water samples was measured using a pH meter calibrated with a 7.0 phosphate buffer solution, following the manufacturer's instructions. The pH probe was directly inserted into the sample, and the reading was recorded from the digital display. Similarly, temperature was measured by immersing a thermometer into the water sample, and the temperature reading was taken once it stabilized on the device's display.

Coliform Detection: Total coliform bacteria, including *Escherichia coli, Klebsiella pneumoniae*, *Enterobacter aerogenes*, *Citrobacter freundii*, and *Serratia marcescens*, were detected using the membrane filtration technique. Water samples (100ml) were filtered through 0.45µm, 47mm white gridded nitrocellulose filters (Merck KGaA, Darmstadt, Germany) placed on MacConkey and Eosin Methylene Blue (Oxoid Ltd., Basingstoke Hampshire, England) agar plates then incubated at 37°C for 24 hours (Mustapha *et al.,* 2020). Lactose-fermenting colonies (pink/red) were counted as coliforms. Colonies exhibiting a green metallic sheen were reported as *E. coli*.

Salmonella and Shigella Detection: For the detection of *Salmonella* spp. and Shigella spp., 100ml of water samples were filtered through 0.45µm, 47mm white gridded nitrocellulose filters (Merck KGaA, Darmstadt, Germany) and placed on Salmonella-Shigella agar (Oxoid Ltd., Basingstoke Hampshire, England) plates and incubated at 37°C for 24 hours (Mustapha *et al.,* 2020). Colonies with black centers (indicating *Salmonella*) and transparent colonies with red halos (indicating *Shigella*) were considered presumptive *Salmonella* or *Shigella*.

Pseudomonas aeruginosa Detection: For *Pseudomonas aeruginosa*, 100ml of water samples were filtered through 0.45µm, 47mm white gridded nitrocellulose filters (Merck KGaA, Darmstadt, Germany).

The membranes were placed on Cetrimide Agar (a selective medium for *P. aeruginosa)* and Nutrient agar (Oxoid Ltd., Basingstoke Hampshire, England) plates, then incubated at 37°C for 24-48 hours (Mustapha *et al.,* 2020). Colonies producing a blue-green pigment (pyocyanin) or yellow-green pigment (pyoverdin) Cetrimide Agar and greenish colonies on Nutrient agar were considered presumptive *Pseudomonas spp.*

Characterization and Identification of the Isolates: The bacterial isolates were characterized based on colonial morphology such as the colour, elevation and margin, cellular morphology such as shape of cell, arrangement of cell and Gram reaction and biochemical characteristics and confirmed using Microbact 24E identification kit (Oxoid Ltd., Basingstoke Hampshire, England) following manufacturer description.

Antibiotic Sensitivity Testing: The antibiotic resistance profile of isolated strains was evaluated using the Kirby-Bauer disk diffusion method. Mueller-Hinton agar plates were inoculated with standardized bacterial suspensions of all the recovered isolates. Commercially available antibiotic disks containing ampicillin (10µg), streptomycin (10µg), trimethoprim (5µg), tetracycline (30µg), nalidixic acid (30µg), chloramphenicol (30µg), ciprofloxacin (5µg), sulphonamide (300µg), cefotaxime (30µg), ceftazidime (30µg), cefoxitin (30µg), amoxicillin-clavulanate (20/10µg), meropenem (10µg) (Oxoid Ltd., Basingstoke Hampshire, England) were placed on the agar surface. Plates were incubated at 37°C for 24 hours. The diameter of inhibition zones surrounding each antibiotic disk was measured in millimeters. The zone diameters were interpreted using established Clinical and Laboratory Standards Institute (CLSI, 2024) guidelines to categorize isolates as susceptible, intermediate, or resistant to each antibiotic *E. coli* ATCC 25922 served as the control strain.

Analysis of Data: Proportions were tested using Fisher's exact Test in Epi Info^{TM}. Inferences were made based on the computed prevalence ratios, their 95% confidence intervals, and *p*-values. The level of significance was set at $p < 0.05$.

RESULTS AND DISCUSSION

The temperature of the water samples ranged from 23.6°C to 26.5°C, while the pH ranged from 7.1 to 7.7 (Table 1). Eight of the ten swimming pools water tested positive for bacterial contamination (Fig. 1). A total of 113 bacterial isolates were recovered from the water samples using the standard methods; with *Escherichia coli* (32%), *Klebsiella pneumoniae* (16%), and *Salmonella subsp2*. (12%) being the most prevalent (Fig. 2). Table 2 showed the range of the coliform bacteria counts which varied significantly across the ten swimming pool water samples from 1.0×10^2 to 9.8×10^2CFU/100 ml. *E. coli* counts ranged from 1.0×10^2 to 2.7×10^2 CFU/100ml. *Salmonella* and *Shigella* counts ranged from 1.0×10 to 1.8×10^2 CFU/100ml. Most of the GNB isolates were resistant to commonly administered antibiotics like Augmentin (62.5%), and ampicillin (57.6%) but were largely susceptible to quinolones (92%). Conversely, all isolates were susceptible to both ciprofloxacin and meropenem (Fig. 4). Swimming pools serve as valuable recreational resources in urban areas, promoting physical activity, social interaction and mental well-being (Overbury *et al.,* 2023). The temperatures of the water from swimming pools were between 23.6°C and 26.5°C, and the pH levels were from 7.1 to 7.7. This result was comparable to results of a study by Rasti *et al.* (2017) which showed a mean pH value of 7.38±0.5. WHO (2003, 2006) recommends that pH values of a swimming pool fall in the range of 7.2 to 7.8. In line with this, 70% of the swimming pool water samples (pH:7.2-7.8) comply with the WHO`s standard for pH. With regard to temperature, the study indicated that swimming pool water samples had an average temperature of 24.9°C. This was a bit lower than what was reported by Rasti *et al.* (2017) which had an average value of 29.3±1.3°C. None of the swimming pool water samples in the study area had a temperature of 20°C which is below the WHO minimum recommended limit (WHO, 2006, 2015).

Table 1: pH and Temperature values of water samples from various swimming pools in Ile-Ife

Sample code	pH values	Temperature (°C)
A	7.4	24.2
B	7.7	23.6
C	7.2	24.8
D	7.3	25.2
E	7.1	26.5
F	7.3	25.5
G	7.2	24.3
H	7.1	23.6
I	7.4	26.1
J	7.1	24.8

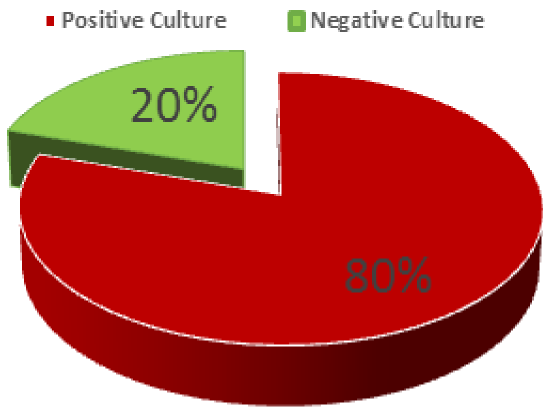

Fig. 1: Percentage of bacterial culture growth from the tested swimming pool

Microbiological analysis revealed that eight out of ten tested swimming pools contained at least one bacterial pathogen, likely due to improper maintenance, inadequate water treatment, and unsanitary conditions (Onuorah *et al.,* 2017). Identified potential pathogens included *Escherichia coli*, *Salmonella subsp. 2*, *Klebsiella spp.*, and *Enterobacter spp.*, corroborating findings by Aleru *et al.* (2021) in Port Harcourt, Rivers State, Nigeria. Similar observations were made by Onajobi *et al.* (2013) and Onifade *et al.* (2019), who reported significant levels of *E. coli* in pools in Kwara and Ekiti States, respectively. The presence of these bacteria may be attributed to insufficient personal hygiene practices following defecation, as well as the introduction of bathers' sweat, saliva, urine, and sputum, compounded by bacterial proliferation over time (George *et al.*, 2014; Sule *et al.*, 2010). The amount of this contamination is probably due to lack of maintenance, monitoring and inadequate treatment, increased use of swimming pools as the study also showed that the rate of bacterial contamination in the sampled swimming pools is significantly higher in pools that are infrequently treated (Kalantary *et al.*, 2024). These findings underscore the critical need for improved maintenance and monitoring of swimming pool environments, as inadequate hygiene practices pose significant public health concerns (Barrell *et al.*, 2000).

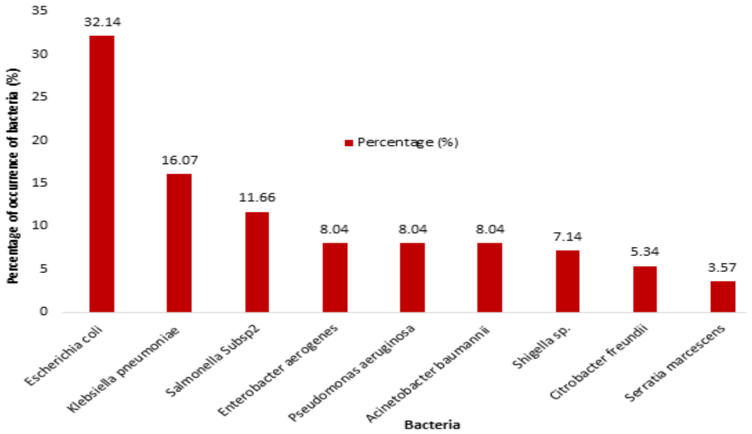

Fig. 2: Percentages of different bacteria isolates recovered from swimming pool in Ile-Ife, Osun-State (n=113)

Table 2: Bacteria count of different swimming pool samples in Ile-Ife, Osun-state

Sample code	Coliform bacterial count (CFU/100ml)	*E. coli* count (CFU/100ml)	*Salmonella* count (CFU/100ml)
A	5.9×10^2	2.6×10^2	1.8×10^2
B	9.0×10^2	2.7×10^2	1.0×10^2
C	3.7×10^2	1.9×10^2	3.0 x 10
D	1.0×10^2	1.0×10^2	NG
E	NG	NG	NG
F	6.3×10^2	1.5×10^2	NG
G	5.8×10^2	1.1×10^2	2.0 x 10
H	NG	NG	NG
I	7.1×10^2	1.8×10^2	1.1 x 10
J	5.2×10^2	2.2×10^2	1.3 x 10

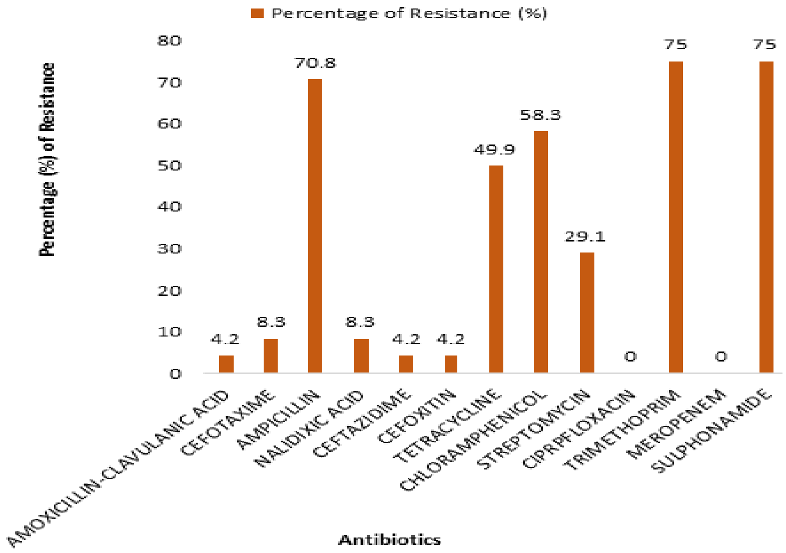

Fig. 3: Antibiotics Resistance pattern of GNB isolated from the study (n=113)

Interestingly, the low incidence of *Pseudomonas aeruginosa* and *Acinetobacter baumannii* should not minimize their potential clinical relevance; these organisms could become problematic in poorly maintained swimming pool environments. This observation is consistent with Okoruwa *et al.* (2023), who also documented low levels of *P. aeruginosa* in water samples from swimming pools in Benin City, Nigeria. The substantial variation in coliform bacteria counts among the pools indicates differing levels of contamination, as similarly noted by Awari *et al.* (2023). The consistent monitoring of coliform bacteria is crucial for assessing the sanitary quality of swimming pools, as coliforms; including *E. coli,* form a significant part of normal intestinal flora. Although many strains are harmless, their presence is a common indicator of fecal pollution and potential health risks. The American Society of Public Health emphasizes the importance of coliform standards in evaluating swimming pool sanitation (Payus *et al.*, 2018). The antimicrobial sensitivity testing showed that most of the GNB isolates were resistant to commonly administered antibiotics like Augmentin and Ampicillin but were largely susceptible to Quinolones and Carbapenem. The high rate of resistance can be due to over-the-counter availability and misused of these antibiotics by the residents of this community (AlTuraifi *et al.*, 2019; Awari *et al.*, 2023). The susceptibility of all isolated pathogens to quinolones and carbapenems suggests these classes may be effective treatment options, although carbapenems are typically reserved as a last resort. Prioritizing adequate pool treatment and promoting hygienic practices around pool environments can help reduce the possible risks of pool contamination and the spread of antimicrobial resistance (AlTuraifi *et al.*, 2019).

Conclusion: The high bacterial load and the isolation of pathogenic bacteria from the pools demonstrate the need for pool health authorities to improve surveillance, pool decontamination standards, and educate swimmers on hygiene before entering pools. This study emphasizes the need for proper hygienic maintenance of swimming pools and the need for a bacteriological standard to be drawn up for swimming pools in Nigeria.

Declaration of Conflict of Interest: The authors declare no conflict of interest.

Data Availability Statement: Data are available upon request from the corresponding author upon reasonable request

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