

Antimicrobial Susceptibility Pattern of Microorganisms from Some Selected Swimming Pools in Awka, Anambra State, Nigeria

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Abstract

The research was aimed at determining the sensitivity and/or resistance of isolated microbes from swimming pools to the various antimicrobial drugs in the case of infections arising from use of infected pools. Water samples were aseptically collected from ten different outdoor swimming pools and microbiologically analysed using standard methods. Using standard disc diffusion method, the antimicrobial susceptibility pattern of isolates was determined against antibacterial agents (Rocephin (30µg), Pefloxacin (10µg), Gentamycin (10µg), Ciprofloxacin (10µg), Streptomycin (30µg), Ampiclox (30µg), Septrin (30µg), Zinnacef (20µg), Erythromycin (19µg), Amoxicillin (30µg)) and antifungal agents (Nystatin, Fluconazole, Ketoconazole). The isolates include; *Bacillus* spp, *Proteus* spp, *Pseudomonas* spp, *Salmonella* spp, *Staphylococcus* spp, *Citrobacter* spp, *Klebsiella* spp, *Escherichia coli*, *Enterobacter* spp, *Aspergillus niger* and *Fusarium* spp. Statistical analysis showed there was no significant difference in the microbial count between the 10 different outdoor pools sampled. The effectiveness of the antimicrobials to the isolates were defined in percentage as follows: Rocephin (7.7%), Pefloxacin (10.9%), Gentamycin (9.3%), Ciprofloxacin (8.8%), Streptomycin (10.4%), Ampiclox (9.5%), Septrin (11.0%), Zinnacef (10.7%), Erythromycin (11.8%), Amoxacillin (10.0%) and Nystatin (37.0%), Fluconazole (36.0%), Ketoconazole (27.0%). With respect to the antimicrobial susceptibility patterns of the isolates, results showed that most of the test antimicrobials were effective against the microbes, with Erythromycin and Nystatin being the most effective for the bacteria and fungi isolates respectively. Thus, Erythromycin and Nystatin should be the first line of prescription in cases of swimming related infection and disease. It should be noted that bacterial and fungal resistance was seen among drugs such as Rocephin and Ketoconazole respectively.

Keywords

Antibiotics Resistance, Microorganisms, Swimming Pool, Microbial Count, ANOVA

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1. Introduction

Water, is undoubtedly, a vital need of man for food and recreation. Water is basic to life on this planet earth, and for man, no substitute has ever been found to serve man in diverse ways that are crucial for mankind's survival and societal advancement as a whole [1-2]. Water is very basic to life and also functions among others, in, transportation,

domestic activities, recreation, cooling of irrigation systems, and food production processes [3]. Swimming pool waters derive their source from natural waters. Swimming pools are increasingly being used by man for swimming and other recreational based activities such as canoe polo, underwater rugby, volleyball, and sports diving. They are also used in certain cultural practices such as baptism, where the individual's head is submerged for a while as prayers are

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being conducted. Due to their increasing uses, swimming pools are now available at many recreation centers, hotels, motels, schools, beaches, universities, wildlife parks, homes, and many other areas. With such an increase in the numbers of swimming pools, high maintenance is necessary to protect the users from any form of infection. It may either be supervised or unsupervised [4]. Varieties of pathogenic organisms including bacteria, fungi, viruses and parasites have been found in swimming pools and similar recreational waters [5]. These organisms, often introduced from environmental sources and swimmers, have been reported as causes of infectious diseases. Outbreaks of waterborne gastrointestinal disease have been linked to faecal contamination of the water, due to faeces released by swimmers [4]. There are several reports on the microbial quality of swimming pools. Center for Disease Control and Prevention (1999) reported that 40% of the swimming pools sampled in Port-Harcourt metropolis, Nigeria, were contaminated with bacteria of the genera *Bacillus*, *Micrococcus* and *Staphylococcus* [5]. Similarly, [6] reported that the bacterial load of the swimming pools sampled in their study in Osu-Labadi Area, Accra exceeded the acceptable limits and were contaminated by *E. coli*, *Enterobacter faecalis*, *Enterobacter cloacae*, *Klebsiella pneumonia*, *Staphylococcus aureus* and *Streptococcus agalactiae*. The poor management of water supply systems to the pools and the use of unprotected water pose a high risk of pathogen transfer to humans. The microbial safety of swimming pool water is highly questionable in countries with poor regulatory framework due to the many existing possible microbial contaminant pathways. Microbial contamination of swimming pools can occur through (a) fecal contaminated source water or direct defecation from swimmers, birds, and animals, (b) nonfaecal human shedding from vomiting, sneezing, mucous, spitting, or skin, (c) poor wastewater disposal [7], (d) microbial biofilm formation along piped water networks, (e) contaminated air, dust, soil, or rainwater, and (f) individuals with contagious diseases or infectious pathogens. Although modern day swimming pools have a recirculation system for the water to be filtered, purified, and disinfected adequately, recent studies reveal that neither advanced technological systems nor can disinfectants obviate the colonization of the pool water with some dangerous pathogens. It has also been reported that the surviving disinfectant tolerant pathogens might also be antibiotic-resistant; a fact already documented for bacterial isolates from treated drinking water and purified sewage effluents. It is important to continuously test swimming pool water for the availability of pathogens that pose public health risks to humans in order to optimize water treatment methods to get the best performing chemicals that can kill or reduce these pathogens to minimal or nonworking levels.

The aim of this study is to investigate the microbial quality of some public swimming pools in Awka as well as assay for the antimicrobial susceptibility of the bacterial isolates. The objectives include: Enumeration of the bacteria and fungi found in the ten selected swimming pool water samples, Isolation and identification of the bacteria and fungi, and antimicrobial susceptibility testing of the bacteria isolates.

2. Materials and Methods

2.1. Sample Collection

Water samples were randomly collected aseptically from Ten (10) different outdoor swimming pools in Awka town. The sampling was done using sterilized wide-mouthed bottle bailers. The samples were properly labelled (A-J), preserved in ice-packs and transported to the microbiology laboratory at Nnamdi Azikiwe University, Awka.

2.2. Isolation, Identification and Characterization of Bacterial Isolates

All laboratory tests were done using standard procedures under aseptic conditions. The culture media used include Nutrient agar, Eosin methylene blue, MacConkey agar, Salmonella and Shigella agar and Sabouraud dextrose agar. The media was prepared according to the manufacturer's specification. These media were sterilized in autoclave at 121°C for 15 minutes and poured into plates to solidify. 1ml of each water samples was serially diluted and 0.1ml of an appropriate dilution was inoculated in triplicates on sterile Eosin-methylene blue (EMB) agar, Salmonella and Shigella agar, MacConkey agar and Nutrient agar plates and the plates were incubated for 24 hours at 37°C for Total coliform count, SSA count, Fecal coliform count and bacterial count. After 24 hours, sterile wire loop was used to pick the isolates from the plates and was streaked onto a freshly prepared nutrient agar and then incubated for 24 hours at 37°C in order to get pure cultures. After the incubation for 24 hours, distinct pure colonies were picked aseptically and cultured onto a fresh nutrient agar slant and incubated for 24 hours at 37°C. After 24 hours, the slants containing the pure cultures were then stored in a refrigerator at -4°C. The routine biochemical test methods were used to characterize the different isolates. The isolates were identified using their macroscopic, microscopic, cultural, and physiological characteristics [8].

2.3. Biochemical and Morphological Characteristics of the Isolates

The variation in bacterial colony morphology is seen with the naked eye. The general form of the colony and the shape of the edge or margin are determined by looking down at the top of the colony. The forms of the bacterial colony include;

punctiform, circular, filamentous, irregular, rhizoid and spindle. The elevation includes; flat, raised, convex, pulvinate, umbonate. The margin includes; entire. Undulate, lobate, rose, filamentous and curled. The biochemical tests such as Gram's straining, catalase test, coagulase test, fermentation test, citrate test, methyl-red test, voges-proskauer test, indole test and motility test were performed to identify the isolated bacteria.

2.4. Identification and Characterization of Fungal Isolates

Fungi were isolated using plating methods [9]. The plating method, 500µl of samples were plated on agar plates and incubated at 25°C for 72 hours. Colonies from primary plates were sub-cultured onto fresh sabouraud dextrose agar supplemented with 100mg Kanamycin to inhibit bacterial growth. The sub-culture was carried out to purify the fungi isolates. During the sub-culture a inoculating straight wire flamed in a Bunsen burner was used to pick the colony and smeared on the agar plate. This was further incubated at room temperature for 3days. Fungal colonies were isolated upon formation, stained with lacto phenol and observed under the microscope. Fungi so observed were identified using the appropriate taxonomic guides [9].

2.5. Antimicrobial Susceptibility Testing

Antimicrobial susceptibility testing on all the isolates was done using disc diffusion method. For bacterial isolates, sensitivity discs containing conventional antimicrobials which are Pefloxacin (10µg), Gentamycin (10µg), Ampiclox (30µg), Zinnacef (20µg), Amoxicillin (30µg), Rocephin (30µg), Ciprofloxacin (10µg), Streptomycin (30µg), Septrin (30µg) and Erythromycin (19µg). Nystatin, Fluconazole and Ketoconazole were used for the sensitivity test against the fungal isolates. The pure bacterial and fungal isolates are evenly streaked individually on the appropriate agar plates that are properly labeled. After which, the inoculums were dried at room temperature for 30 min, then antibiotic impregnated disks were applied to the surface of the inoculated plates using sterile forceps. Note; the sensitivity discs containing antibacterial drugs were placed on the surface of each Nutrient agar plate evenly seeded with test bacterial organisms and was incubated for 24 h at 37°C while the sensitivity discs containing antifungal drugs were placed on the surface of each Sabouraud dextrose agar plate evenly seeded with test fungal organisms and was incubated for 24-72 h at 25°C -27°C [10].

2.6. Measurement of Zone of Inhibition

After 24 hours of incubation in the case of bacteria and 48-72 hours incubation in the case of fungi, the antimicrobial

sensitivity results for each isolates against each antimicrobial drug was read by measuring the zones of inhibition using ruler. The diameter of the zone were measured and recorded; that is for organisms susceptible to the antimicrobials. Isolates that was not susceptible/ sensitive to the drugs showed no zone of inhibition and are thus termed resistant to the antimicrobial agents used for the sensitivity test against the fungal isolates. The pure bacterial and fungal isolates are evenly streaked individually on the appropriate agar plates that are properly labeled. After which, the inoculums were dried at room temperature for 30 min, then antibiotic impregnated disks were applied to the surface of the inoculated plates using sterile forceps. Note; the sensitivity discs containing antibacterial drugs were placed on the surface of each Nutrient agar plate evenly seeded with test bacterial organisms and was incubated for 24 h at 37°C while the sensitivity discs containing antifungal drugs were placed on the surface of each Sabouraud dextrose agar plate evenly seeded with test fungal organisms and was incubated for 24-72 h at 25°C -27°C [10].

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2.7. One-way Analysis of Variance

In statistics, one-way analysis of variance (ANOVA) is a technique that can be used to compare means of two or more samples. This technique can be used only for numerical response data as shown in [11-13], the "Y", usually one variable, and numerical or (usually) categorical input data, the "X", always one variable, hence "one-way". The Viable count of the microorganisms from the swimming pools water samples was analyzed using ANOVA. The ANOVA tests the null hypothesis, which states that samples in all groups are drawn from populations with the same mean values.

$$y_{i,j} = \mu + \tau_j + \varepsilon_{i,j} \quad (1)$$

Where,

$i = 1, \dots, I$ is an index over experimental units

$j = 1, \dots, J$ is an index over experimental units

I_j is the number of experimental units in the j_{th} treatment group

$I = \sum_j I_j$ is the total number of experimental units

$y_{i,j}$ are observations

μ_j is the mean of the observations for the j_{th} treatment group

3. Results and Discussion

Table 1. Viable count of the swimming pools' water samples.

Samples	Total bacterial count (cfu/ml) (10^3)	Total coliform count (cfu/ml) (10^3)	Fecal coliform count (cfu/ml) (10^2)	Salmoella and Shigella count (cfu/ml) (10^2)
A	1.41	0.50	0.00	0.00
B	0.05	0.00	0.00	0.00
C	0.53	0.71	0.40	0.92
D	1.58	0.90	0.45	0.51
E	1.20	0.67	0.51	0.49
F	1.32	1.60	0.04	0.00
G	1.00	0.14	0.04	0.05
H	1.00	0.05	0.00	0.10
I	1.23	0.45	0.09	0.04
J	0.35	0.05	0.00	0.00

Table 1 showed the viable counts of bacteria isolated from the ten swimming pool water samples in Awka metropolis. They were counted and grouped according to their different growth rate on nutrient agar and Eosin-methylene blue agar, MacConkey agar and *Salmonella* and *Shigella* agar. The

viable count on nutrient agar ranges from 0.05×10^2 to 1.41×10^2 cfu/ml. Sample A and B showed the highest and lowest number of bacteria count on nutrient agar respectively. There was no growth on four SSA and EMB plates.

Table 2. Morphological characteristics of the bacterial isolates.

Isolate	Color	Form	Margin	Elevation
1	Cream	Circular	Irregular	Flat
2	Cream	Irregular	Entire	Raised
3	Golden yellow pigment	Circular	Entire	Convex
4	Greyish white	Circular	Entire	Convex
5	Cream	Irregular	Entire	Convex
6	Colorless with black center	Irregular	Entire	Convex
7	Cream	Filamentous	Filamentous	Flat
8	Brown	Irregular	Irregular	Raised
9	Green metallic sheen	Circular	Entire	Convex

Table 2 shows the morphological characteristics of the bacteria isolates. The morphological features were grouped based on their color, form, margin and elevation on the plates. Green metallic sheen on EMB agar showed the presence of *E. coli* in the water samples.

Table 3. Microscopic and biochemical characteristics of the bacterial isolates.

Isolates	Gram reaction	shape	form	O	I	C	Co	Cu	M	Mr	Vp	G	S	M	F	L	Probable organism
1	+	Rod	Single	-	+	+	-	+	-	+	+	AG	A	A	A	AG	<i>Bacillus</i> spp
2	-	Rod	single	-	-	+	+	+	-	-	+	AG	AG	A	A	A	<i>Enterobacter</i> spp
3	+	coccus	chain	+	-	+	+	+	-	+	-	-	-	-	-	-	<i>Staphylococcus</i> spp
4	+	coccus	single	-	-	+	-	+	-	-	+	AG	A	A	A	A	<i>Klebsiella</i> spp
5	-	Rod	single	-	-	+	-	+	-	+	-	AG	AG	A	AG	A	<i>Citrobacter</i> spp
6	-	Rod	single	-	-	+	-	-	+	+	-	AG	-	A	A	-	<i>Salmonella</i> spp
7	-	Rod	single	-	+	+	+	-	+	+	-	AG	AG	AG	A	-	<i>Proteus</i> spp
8	-	Rod	single	+	-	+	+	+	+	-	-	AG	A	A	A	AG	<i>Pseudomoas</i> spp
9	-	Rod	single	-	+	+	+	-	+	+	-	AG	AG	A	AG	AG	<i>Escherichia coli</i>

Key: O-Oxidase test, I-Indole test, C- Catalase test, Co- Coagulase test, Cu-Simmon citrate utilization test, M- Motility test, Mr-Methyl red test, Vp-Voges Proskauer test, G-Glucose, S- Sucrose, M- Maltose, F- Fructose, L- Lactose, AG- Acid gas production, A- Acid production only,+ Positive and -Negative

Gram stain reaction and biochemical test results carried on the bacterial isolates were tabulated in Table 3. IMViC test and other biochemical test were carried out to aid in identification of the bacteria. Bergey's manual was used to trace the probable manual.

Table 4. Frequency of distribution of the bacterial isolates.

Organisms	Number isolated	Frequency of occurrence (%)
<i>Bacillus</i> sp	2	6.5
<i>Enterobacter</i> sp	2	6.5
<i>Staphylococcus</i> sp	4	12.9
<i>Klebsiella</i> sp	3	9.7
<i>Citrobacter</i> sp	2	6.5
<i>Salmonella</i> sp	6	19.4
<i>Proteus</i> sp	2	6.5
<i>Pseudomonas</i> sp	2	6.5
<i>Esherichia coli</i>	8	25.8
TOTAL =	31	100

Table 4 represents the percentage distribution of the bacterial isolates in the swimming pool water samples. *E. coli* is the most occurred bacteria isolated from the different samples followed by *Salmonella* spp.

Table 5. Macroscopic identification of fungi isolates.

Sample	Cell wall	Type of spores	Fungi
A	Septate	Chlamydospore	<i>Fusarium</i> spp
B	Septate	Conidiospore	<i>Aspergillus niger</i>
C	Septate	Chlamydospore	<i>Fusarium</i> spp
D	Septate	Chlamydospore	<i>Fusarium</i> spp
E	Septate	Chlamydospore	<i>Fusarium</i> spp
F	-	-	-
G	-	-	-
H	Septate	Conidiospore	<i>Aspergillus niger</i>
I	-	-	-
J	-	-	-

Fungi isolated were grouped based on the macroscopic identification after lactophenol test in table 5. Only two genera of fungi (molds) were isolated on SDA and they are *Aspergillus niger* and *Fusarium* spp.

Table 6. The zone of inhibitions (cm) shown by the bacterial isolates.

Isolate	R	CPX	S	SXT	E	PET	CN	APX	Z	AM
1	1.35	1.5	1.4	1.5	1.7	1.5	1.3	1.25	1.5	1.2
2	1.45	1.5	1.6	1.6	1.8	1.5	1.6	1.8	1.8	1.45
3	1.4	1.0	1.5	1.8	1.8	1.75	0.8	1.25	1.6	1.4
4	R	0.9	1.6	1.5	1.5	1.6	1.5	1.65	2.0	2.0
5	1.3	1.4	1.2	1.6	1.9	1.7	1.5	1.4	1.5	1.3
6	1.5	1.65	2.1	2.0	2.0	1.8	1.75	1.25	1.3	1.75
7	1.2	2.0	1.5	1.5	1.5	1.5	2.1	1.5	R	1.5
8	1.45	1.3	1.9	1.5	1.9	1.6	1.5	1.6	1.3	1.5
9	1.4	1.75	2.0	1.75	2.0	1.2	1.9	1.2	1.5	1.75

Key- R: Rocephin (30µg) PEF: Pefloxacin (10µg) CN: Gentamycin (10µg) CPX: Ciprofloxacin (10µg)

S: Streptomycin (30µg) APX: Ampiclox (30µg) SXT: Septrin (30µg) Z: Zinnacef (20µg)

E: Erythromycin (19µg) AM: Amoxicillin (30µg)

Table 7. The zone of inhibitions (cm) shown by the fungal isolates.

Isolate	NYS	FLU	KET
1	2.4	3.4	1.8
2	3.0	1.4	2.6

Key- NYS: Nystatin KET: Ketoconazole FLU: Fluconazole

3.1. Antimicrobial Sensitivity Results for the Identified Isolates from the Swimming Pool Samples

The antimicrobial sensitivity results showed that the antibacterial: Erythromycin gave the highest collective zones of inhibition, followed by Septrin while Rocephin gave the least collective zones of inhibition.

In the case of antifungals, Nystatin gave the highest collective zones of inhibition while Ketoconazole gave the least.

3.2. Analysis of the Antimicrobial Sensitivity Results

The antibacterial susceptibility patterns (%) of the bacterial isolates are as follows:

Isolate 1= R (9.5%), CPX (10.6%), S (9.9%), SXT (10.6%), E (11.9%), PEF (10.6%), CN (9.2%), APX (8.8%), Z (10.6%), AM (8.5%)

Isolate 2= R (9.0%), CPX (9.3%), S (9.9%), SXT (9.9%), E (11.2%), PEF (9.3%), CN (9.9%), APX (11.2%), Z (11.2%), AM (9.0%)

Isolate 3= R (9.8%), CPX (6.9%), S (10.5%), SXT (12.6%), E (12.6%), PEF (12.2%), CN (5.6%), APX (8.7%), Z (11.2%), AM (9.8%)

Isolate 4= R (0%), CPX (6.3%), S (11.2%), SXT (10.5%), E (10.5%), PEF (11.2%), CN (10.5%), APX (11.6%), Z (14.0%), AM (14.0%)

Isolate 5= R (8.8%), CPX (9.5%), S (8.1%), SXT (10.8%), E (12.8%), PEF (11.5%), CN (10.1%), APX (9.5%), Z (10.1%), AM (8.8%)

Isolate 6= R (8.8%), CPX (9.6%), S (12.3%), SXT (11.7%), E (11.7%), PEF (10.5%), CN (10.2%), APX (7.3%), Z (7.6%), AM (10.2%)

Isolate 7= R (9.0%), CPX (9.3%), S (9.9%), SXT (9.9%), E

(11.2%), PEF (9.3%), CN (9.9%), APX (11.2%), Z (11.2%), AM (9.0%)

Isolate 8= R (9.8%), CPX (6.9%), S (10.5%), SXT (12.6%), E (12.6%), PEF (12.2%), CN (5.6%), APX (8.7%), Z (11.2%), AM (9.8%)

Isolate 9= R (0%), CPX (6.3%), S (11.2%), SXT (10.5%), E (10.5%), PEF (11.2%), CN (10.5%), APX (11.6%), Z (14.0%), AM (14.0%)

The antifungal susceptibility patterns (%) of the fungal isolates are as follows:

Isolate 1= NYS (31.6%), FLU (39.5%), KET (28.9%)

Isolate 2= NYS (57.7%), FLU (42.3%), KET (2.3%)

Table 8. Descriptive Statistics on the Total Bacteria Count.

Min.	1st Qu.	Median	Mean	3rd Qu.	Max.	Skewness	Kurtosis	Variance
0.0500	0.6475	1.1000	0.9670	1.2975	1.5800	-0.6599747	2.185388	0.2483122

Table 9. Descriptive Statistics on the Total Coliform Count.

Min.	1st Qu.	Median	Mean	3rd Qu.	Max.	Skewness	Kurtosis	Variance
0.0000	0.0725	0.4750	0.5070	0.7000	1.6000	0.9577542	3.268666	0.2477344

Table 10. Descriptive Statistics on Fecal Coliform Count.

Min.	1st Qu.	Median	Mean	3rd Qu.	Max.	Skewness	Kurtosis	Variance
0.0000	0.0000	0.0400	0.1530	0.3225	0.5100	0.8669231	1.894296	0.04442333

Table 11. Descriptive Statistics on the Salmonella and Shigella count.

Min.	1st Qu.	Median	Mean	3rd Qu.	Max.	Skewness	Kurtosis	Variance
0.0000	0.0000	0.0400	0.1918	0.2950	0.9200	1.42798	3.696426	0.09559636

Table 12. ANOVA Table.

Samples	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	1.306	9	.145	.546	.807
Within Groups	2.126	8	.266		
Total	3.432	17			

Table 12 shows the output of the ANOVA analysis and whether there is a statistically significant difference between the samples. We can see that the significance value is 0.807 which is above 0.05 and therefore indicates there is no statistical significant difference in the microbial count between the 10 different outdoor swimming pool.

Generally, the microbiological assessment of swimming pool water collected from ten different hotels in Awka metropolis carried out shows that the total viable count, fecal coliform count, total coliform count and *Salmonella* and *Shigella* agar count ranges from 0.05×10^2 to 1.5×10^2 cfu/ml, 0.00×10^2 to 0.51×10^2 cfu/ml, 0.00×10^2 to 1.60×10^2 cfu/ml, and 0.00×10^2 to 0.92×10^2 cfu/ml respectively. The sample from A showed the highest bacterial count as shown in Table 1. A total of 31 bacteria from 9 genera were identified and they are *Bacillus* spp, *Enterobacter* spp, *Staphylococcus* spp, *Klebsiella* spp, *Citrobacter* spp, *Salmonella* spp, *Proteus* spp, *Pseudomonas*

spp and *Escherichia coli*. A total of 9 fungi from 2 genera were identified and they are *Aspergillus niger* and *Fusarium* spp. The *E. coli* is the most occurring bacterium and the presence of coliform in these swimming pools showed that there is deficiency in the treatment of the swimming pools or inadequate protection of the swimming pools [14]. Ayandele *et. al* (2015) carried out an assessment of microbiological quality of outdoor swimming pools in Ilorin metropolis, Kwara state [15]. The total viable count of all the pools water analyzed were relatively high and the following microorganisms were isolated: *Pseudomonas* spp, *Proteus* spp, *Staphylococcus* spp, *Shigella* spp, *E. coli*, *Bacillus* spp, *Citrobacter* spp, *Aspergillus niger*, *Rhizopus* spp. *Pseudomonas* spp and *E. coli* showed highest multi-drug resistance in his study. Buot *et. al* (2010) carried out microbial assessment of swimming pools from selected hotels in Osogbo metropolis, Osun state, Nigeria [16]. The

bacteria isolated include *Aeromonas* spp, *Citrobacter* spp, *Klebsiella* spp, *Proteus* spp, *Pseudomonas* spp, *Shigella* spp, *Staphylococcus* spp. And the fungi isolated include *Aspergillus niger*, *Aspergillus repens*, *Absidia cylindrospora* and *Fusarium* spp. In his study, multi-drug resistance was common among the bacteria isolated from the swimming pool water. [17] evaluated of the bacteriological quality of outdoor public swimming pools in Awka metropolis, Anambra state. The bacteria isolated include *Klebsiella* spp, *Proteus* spp, *Enterobacter* spp, *Citrobacter* spp, *Salmonella* spp, and *Bacillus* spp. [18] investigated the microbiological and physiochemical characteristics of swimming pools in South eastern states of Nigeria. The bacteria isolates include *Bacillus* spp, *E. coli*, *Staphylococcus* spp, and *Proteus* spp. The results of this study agreed with the reports of these earlier investigators. The isolation of different species of bacteria and fungi which are known human pathogens from these pools might be due to faecal contamination from both humans and animals [6, 19]. *Pseudomonas* species are associated with surface run-off water, while *E. coli*, *Staphylococcus epidermidis* and *S. aureus* are usually contributed by bathers in the swimming pools [14]. Most of these bacterial isolates are known enterotoxin producers when ingested into the body, therefore the presence of these bacteria in pools is a threat to public health [20]. The presence of *E. coli* and *Citrobacter freundii* is a strong indication of faecal contamination. It has also been reported by some workers that *E. coli* should be totally absent in 100ml of pool water [21]. The presence of these organisms may constitute a public health hazard because swimmers can accidentally swallow contaminated pool water during swimming which can result in outbreaks of diseases like cholera, shigellosis, typhoid fever, gastroenteritis and diarrhea [22, 18]. The presence of *Aspergillus*, *Penicillium* and *Rhizopus* were also detected from swimming pools water in Greece [21]. *Aspergillus niger* has been reported to cause aspergillosis which is an infection of the external ear [23]. *Aspergillus* has also been reported as a starting point for dissemination of infections in immune compromised patients [16]. The antibiotic resistance of bacterial strains isolated from recreational waters, like swimming pools, lake etc. have been reported by many investigators [24]. Antimicrobial susceptibility/sensitivity patterns of these organisms were determined using specific antibiotics (Rocephin, Pefloxacin, Gentamycin, Ciprofloxacin, Streptomycin, Ampiclox, Septrin, Zinnacef, Erythromycin, Amoxicillin) and antifungal (Nystatin, Fluconazole, Ketoconazole), some of which were used by [25, 26].

For the antibiotics: Isolate 1 was more susceptible to Erythromycin but least susceptible to Amoxicillin. Isolate 2

was more susceptible to Erythromycin, Ampiclox and Zinnacef but least susceptible to Rocephin and Amoxicillin. Isolate 3 was more susceptible to Septrin and Erythromycin but least susceptible to Gentamycin. Isolate 4 was more susceptible to Zinnacef and Amoxicillin, least susceptible to Ciprofloxacin and resistant to Rocephin. Isolate 5 was more susceptible to Erythromycin but least susceptible to Streptomycin. Finally, Isolate 6 was more susceptible to Septrin and Erythromycin but least susceptible to Zinnacef. These susceptibility patterns are in agreement with [27]. Isolate 7 was more susceptible to Erythromycin, Ampiclox and Zinnacef but least susceptible to Rocephin and Amoxicillin. Isolate 8 was more susceptible to Septrin and Erythromycin but least susceptible to Gentamycin. Isolate 9 was more susceptible to Zinnacef and Amoxicillin, least susceptible to Ciprofloxacin and resistant to Rocephin. Furthermore, considering the comparison (in %) of each of the test antibiotics, Erythromycin is the most effective antibiotic drug to be used in the case of swimming pool related infection/ disease cases caused by these aforementioned bacterial isolates.

For the antifungals: Isolate 1 was more susceptible to Fluconazole but least susceptible to Ketoconazole. Isolate 2 was more susceptible to Nystatin, least susceptible to Ketoconazole but resistant to Fluconazole. Nystatin is the most effective antifungal drug to be used in the case of swimming pool related infection/ disease cases caused by these aforementioned fungal isolates.

With respect to the antimicrobial susceptibility patterns of the isolates, results showed that most of the test antimicrobials were effective against the microbes, with Erythromycin and Nystatin being the most effective for the bacteria and fungi isolates respectively. Thus, Erythromycin and Nystatin should be the first line of prescription in cases of swimming related infection and disease. It should be noted that bacterial and fungal resistance was seen among drugs such as Rocephin and Ketoconazole respectively.

4. Conclusion

The results obtained in this study showed that most of the swimming pools did not comply with the WHO standard for recreational activities due to the presence of microorganisms and therefore constitute serious problems to the bathers due to high microbial counts, resistance of some isolated microorganisms to antibiotics. With respect to the antimicrobial susceptibility patterns of the isolates, results showed that most of the test antimicrobials were effective against the microbes, with Erythromycin and Nystatin being the most effective for the bacteria and fungi isolates respectively. Thus, Erythromycin and Nystatin should be the

first line of prescription in cases of swimming related infection and disease. It should be noted that bacterial and fungal resistance was seen among drugs such as Rocephin and Ketoconazole respectively. Statistical analysis shows that there was no statistical significant difference in the microbial count between the 10 different outdoor swimming pools sampled.

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