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BACTERIOLOGICAL ASSESSMENT OF WATER FROM TANKS IN VARIOUS LABORATORIES OF FEDERAL UNIVERSITY DUTSIN-MA, KATSINA STATE, NIGERIA

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ABSTRACT
Water is essential in laboratories for conducting various activities. The lack of a steady supply of water has mandated the need to store water in tanks prior to usage; this poses a great risk that may interfere with authentic research outcomes. This study determined both the physicochemical and the bacteriological quality of water from tanks in various laboratories of Federal University Dutsin-Ma, Katsina State, Nigeria. A total of f 15 water samples were collected from 5 laboratories using sterile screw-capped 100ml capacity bottles. Some of the physicochemical parameters measured include: temperature, pH, electrical conductivity, and total dissolved solids. The samples were serially diluted before inoculating unto the appropriate media. After 24 h of incubation, the colonies were gram stained and biochemically identified. Antibiotic susceptibility profiles of the isolates were also conducted using the disc diffusion method. The pH of the water samples in all the laboratories was found to be (5.2-6.8), whereas the mean temperature was recorded at (29.7-30.1 ºC). However, the electrical conductivity of the water samples fell below the limit (0.07-0.11). Based on colonial characteristics, gram reaction, and biochemical tests, the organisms isolated were confirmed to be *Staphylococcus aureus* and *Escherichia coli*. *S. aureus*. Staphylococcus aureus was found to be more sensitive to gentamicin, while *E. coli* proved to be more sensitive to ciprofloxacin. The results obtained in this research highlight the need for regular and proper monitoring of the water stored in tanks of the various laboratories.

Keywords: Water, Research laboratories, Antibiotics, Physicochemical parameters, Tanks, Pollution, Public health

INTRODUCTION
An adequate water supply is essential in major facilities, including laboratories. However, insufficient supply often necessitates water storage in various types of tanks. While this practice provides a temporary solution, it can introduce significant risks and operational challenges, including potential contamination and interruptions that may hinder or compromise laboratory research activities. From a sanitary point of view for domestic water usage, water can be classified as polluted and unpolluted water (Twort *et al.*, 2010). Potable water is colourless, odourless, tasteless and free from poisonous, corrosive and staining substances as well as disease-causing organisms (Diersing, 2009). The pollution of tap water could originate from contamination from water pipes and storage tanks, biological agents from human faeces, especially bacterial pathogens (Yasin *et al.*, 2015). The coliform bacteria have always been the principal concern in setting health-based targets for microbiological quality of drinking water (Nwinyi *et al.*, 2020). Common indicator bacteria are coliforms, which include *Escherichia coli*, *Enterobacter and Citrobacter* species. These bacteria are excreted in larger numbers by man and animals. Their presence in water indicates the contamination of water by faecal matter (Ochei & Kolhatkar, 2007).

Based on the recommendation of World Health Organization (WHO) and the National Agency for Food, Drug and Control (NAFDAC), the standard for potable water should be as such; for every 100 ml of water, coliform bacteria should be absent, and the total plate count for bacteria should be 100cfu/ml of water (United Nations [UN], 2018). Most Nigerian higher institutions strongly rely on plastic tanks as the main storage source for their water needs, including the laboratories; as such, detecting the physicochemical parameters such as pH, temperature, electrical conductivity, and biological oxygen demand is very crucial so as to get an idea about the quality of water being used to perform the various tasks in the laboratories. Hence, this study presents a bacteriological assessment of water stored in various tanks within the laboratory facilities of Federal University Dutsin-Ma, Katsina State, Nigeria.

MATERIALS AND METHODS
Study Area
The study was conducted at Federal University Dustin-Ma Katsina State. Dutsin-Ma Local Government Area is

one of the administrative divisions of Katsina State, Northwestern Nigeria, located within the Sudan savanna ecological zone. It lies approximately between latitudes 12°27′N and 12°30′N and longitudes 7°29′E and 7°37′E, characterized by a tropical continental climate with distinct wet and dry seasons. The area experiences a mean annual rainfall of about 600–800 mm, concentrated between May and September, while the dry season is marked by the Harmattan influence from November to February. Temperatures are generally high throughout the year, often ranging from 25 to 40 °C (Abaje *et al.*, 2014).

Sample Collection

Fifteen (15) water samples in total were gathered; three samples from each tank were purposively taken from Biochemistry, Biology, Chemistry, Microbiology, and Physics laboratories, respectively. The water samples were collected in sterile screw-capped 100 ml capacity bottles. Temperature and pH of the water were measured at the point of collection, whereas the other physicochemical parameters were determined later. The samples were immediately transported to the Microbiology Laboratory, Federal University Dutsin-Ma for further analysis.

Determination of Physicochemical Parameters

The physicochemical analysis was carried out to determine the quality of the water samples in accordance with the methods of the American Public Health Association (Afroz *et al.*, 2017). The parameters analyzed include: temperature, pH, electrical conductivity, and dissolved oxygen (DO).

Temperature and pH

The temperature and pH of the water samples were determined at the point of collection using the HANNA Combo (H198130) instrument. Following the manufacturer's instructions, the electrodes connected to each meter were submerged in a clean beaker containing the sample. Selecting a specific test via the mode buttons, the values read for each parameter tested were observed and recorded.

Electrical Conductivity

Electrical conductivity (EC) of water samples collected from storage tanks was determined using a calibrated digital conductivity meter. Prior to analysis, the instrument was calibrated according to the manufacturer's instructions using standard conductivity solutions. Water samples were thoroughly mixed and allowed to reach room temperature before measurement. The conductivity probe was immersed directly into each sample, ensuring complete submersion of the sensing element. Measurements were recorded in microsiemens per centimeter (µS/cm), and each sample was analyzed in triplicate to ensure accuracy and reproducibility. The mean conductivity value for each sample was calculated and compared with established drinking water quality standards

Dissolved Oxygen (DO)

Dissolved oxygen (DO) was determined in situ using a Hanna 3200 multiparameter meter by inserting the probe directly into the water samples at the point of collection.

Isolation and Characterization of the Bacteria Present in the Water Samples

The bacteria present in the water samples were isolated and identified following these steps: serial dilution; inoculation onto Nutrient agar, MacConkey, and EMB agar; incubation at 37 ℃. After 24 h of incubation, colonial morphology was observed macroscopically, Gram's stain was performed, and biochemical tests were done on appropriate media.

Serial Dilution

The water samples were serially diluted using the procedure adopted from Arnold and Colford (2007). Six tubes were arranged in a test tube rack and used for the dilution. From the water samples, 1ml was withdrawn using a sterile syringe and introduced into a test tube containing 9ml of sterile distilled water, to make the 10^{-1} dilution or stock. The test tube was gently homogenized to ensure even mixing of the aliquot and the diluent. Thereafter, successive dilutions were made into 5 subsequent tubes by withdrawing 1 ml from each tube and introducing it into the next, until 10^{-2}, 10^{-3}, 10^{-4} and 10^{-5} dilutions were obtained (Arnold & Colford, 2007).

Media Preparation and Inoculation

All media were prepared and dispensed according to the manufacturer's instructions.

Culture and Incubation

Sterile wire loops were used to withdraw 0.1 ml of each dilution from the samples, which were subsequently inoculated onto the prepared solid culture plates. The prepared culture plates were inoculated onto the prepared media. The plates were incubated aerobically at 37 °C for 24 or 48 h, depending on the medium used. Colonial growth was observed and recorded macroscopically.

Morphological Characteristics of the Isolates

The morphological features of the isolates were observed by assessing the colonies appearance, form, texture, consistency, shape, pigmentation, margin and elevation of the obtained colonies on agar plates.

Gram Stain

The procedure of Thursby and Juge (2017) was used, as follows: smears of the isolates were prepared from the overnight culture plates on a clean glass slide, and allowed to air dry. These were then fixed by passing the slide three (3) times through the flame of a Bunsen burner. The slide was then allowed to cool before staining. The crystal violet stain was applied to the smear for 30-60 seconds. The plates were then washed using tap water. Lugol's iodine was added for 1 minute, and then washed using tap water. The slides were decolorized rapidly for 15 seconds with acetone alcohol and then immediately washed using tap water. The back sides of the slides were wiped clean and placed in a staining rack for the smear to air dry. Drops of oil immersion (100x) were added to the dried smears and examined under the electron microscope (Thursby & Juge, 2017).

Biochemical Identification

Conventional methods were carried out to further ascertain the isolates; these include:

Catalase, Citrate utilisation test, Indole**,** Methyl Red-Voges Proskaeur (MR-VP), Motility, and Oxidase test.

Antibiogram of the Isolated Species against some Antibiotics

McFarland Standards

Standardized microbial testing was used to determine the effectiveness of certain antibiotics against the isolates. These were prepared using a 0.5 McFarland standard, which was obtained from 24 h old cultures of each of the bacterial isolates, cultured using the lawn streaking method on nutrient agar (Thursby & Juge, 2017). Loopfuls of the growths from the culture media were made into a test tube containing 5 ml of sterile normal saline. The emulsified suspension was compared to the 0.5 McFarland Standard prepared using $BaCl_2$ and H_2SO_4, until the turbidity of the bacterial suspension matches that of the McFarland Standard (Muazu *et al.*, 2012).

Disc Diffusion Method

Susceptibility testing was carried out using the disc diffusion method, which is the standard method for assessing the efficacy of certain antibiotics against bacterial isolates (Muazu *et al.*, 2012). Mueller-Hinton agar was prepared and poured into Petri dishes; three antibiotics (streptomycin, pefloxacin, amoxicillin) were placed on the plates for each isolate, two per plate. The plates were then incubated for 24 h, after which zones of inhibition were measured and recorded in mm ± standard deviation (Clinical and Laboratory Standards Institute (CLSI), 2018).

Statistical Analysis

Descriptive statistics were used to calculate the mean zone of inhibition for the two antibiotics tested against bacterial isolates recovered from laboratory water tanks.

RESULTS AND DISCUSSION

Physicochemical Characteristics of the Water Samples

The mean physicochemical parameters of the water from five locations are shown in Table 1. The pH of all the water samples was found within the range of 5.2-6.8. The results show that water from all the locations falls below the WHO standard except for the laboratory (sample BIO). This finding is similar with the study of Araujo *et al.* (2007) which recorded a pH of water samples ranged from 5.2-6.8, also, the findings of Sule *et al.* (2011) and Yasin *et al.* (2015) recorded pH of water samples in the range of 6.53–7.45 and 7.4–8.14, respectively, all of which fall within the WHO permissible limits (6.5- 8.5). The temperature of water samples collected from the five laboratory storage tanks ranged from 29.7 to 30.1 °C. These values are within the permissible limits recommended by the World Health Organization (WHO) for potable water and reflect the prevailing tropical environmental conditions of Northern Nigeria. Water temperature plays a critical role in influencing chemical reactions, microbial metabolism, and the solubility of gases such as oxygen. Similar temperature ranges have been reported in studies of stored and drinking water sources in Nigeria, indicating that temperatures around 30 °C are common in tropical regions and do not necessarily compromise water quality when other parameters remain within acceptable limits (Allison *et al.*, 2020; Jimoh *et al.*, 2025).

The electrical conductivity (EC) values recorded in this study ranged from 0.07 to 0.11, indicating low concentrations of dissolved ionic substances in the water samples. Electrical conductivity is directly related to the concentration of dissolved salts and inorganic materials in water. The low EC values observed suggest that the water was weakly mineralized and contained relatively low levels of dissolved solids. Similar findings were reported by Okonkwo *et al.* (2024), who observed low conductivity values in borehole and stream water sources in Rivers State, Nigeria and concluded that the water samples met international drinking water quality standards. Low conductivity values are generally considered desirable for potable water because they indicate limited contamination by dissolved chemical constituents.

The dissolved oxygen (DO) concentrations varied among the sampling locations, with the highest mean value recorded in the Biochemistry Laboratory tank and the lowest value observed in the Physics Laboratory tank. Dissolved oxygen is an important indicator of water quality because it reflects the level of oxygen available for biological processes and influences the survival of aerobic microorganisms. High dissolved oxygen concentrations generally indicate good water quality and adequate aeration, whereas lower concentrations may result from microbial respiration, organic matter decomposition, or prolonged water storage. The variation in DO levels among the tanks may be attributed to differences in storage duration, frequency of water usage, tank cleanliness, and exposure to environmental conditions. Similar observations were reported by O'Hogain *et al.* (2012), who found that dissolved oxygen levels in stored water systems varied significantly depending on storage conditions and microbial activity. Likewise, Prince *et al.* (2023) reported that dissolved oxygen concentrations in stored water supplies were influenced by the level of organic matter and the extent of microbial contamination present in the water.

Table 1: Mean values of physicochemical parameters of the water obtained from five locations within Dutsin-Ma Take-Off Campus, Katsina State, Nigeria

Parameter	Various Laboratories Tested					WHO Standard (2011)
	BCH	BIO	CHM	MCB	PHY	
pH	5.75±0.07	6.8±0.00	6.4±0.00	5.2±0.14	6.2±0.00	6.5 – 9.2
Temperature (°C)	30.1±0.00	30.0±0.07	30.0±0.00	29.7±0.28	30.1±0.14	20 – 30
Electrical Conductivity (µm/cm)	0.11±0.01	0.16±0.00	0.07±0.01	0.11±0.01	0.07±0.00	0.5– 3.0
DO (mg/l)	32.0±0.05	28.0±0.02	24.0±0.00	30.0±0.02	18.0±0.02	<2 mg/L for drinking water

BCH = Biochemistry Laboratory, BIO = Biology Laboratory, CHM = Chemistry Laboratory, MCB = Microbiology Laboratory, PHY = Physics Laboratory, DO = Dissolved oxygen

Table 2: Preliminary Biochemical Test of the Bacterial Isolates

Laboratory	GS	CAT	COA	CIT	IND	MR	VP	MOT	Probable Organism
Chemistry	+	+	+	+	-	+	-	-	*S. aureus*
Biology	+	+	+	+	-	+	-	-	*S. aureus*
Microbiology	-	+	-	-	+	+	-	+	*E. coli*
Biochemistry	-	+	-	-	+	+	-	+	*E. coli*
Physics	+	+	+	+	-	+	-	-	*S. aureus*

IND = Indole, GS = Gram staining, MOT = Motility test, CAT = Catalase test, MR = Methyl red test, COA = Coagulase test, VP = Voges Proskauer

Bacterial Isolation and Identification

A total of ten (10) bacteria were screened, and based on morphological similarity, five of which were selected and identified. The results of preliminary biochemical tests, as presented in Table 2, showed that 3 of the 5 bacteria were Staphylococcus aureus, while 2 were *E. coli*. This finding corroborates the findings of Poonia *et al*. (2014), who reported about 19 bacterial species associated with reservoir tanks, including *E. coli*, *Salmonella* species and *Proteus* species. These bacterial contaminants may be linked to a lack of regular cleaning of such tanks, and inefficiency in maintaining the tanks and the pipelines connected to them.

Antibiotic Sensitivity Testing of the Isolated Bacteria

Table 3 shows the antibiotic susceptibility profiles of the bacterial isolates recovered from laboratory water tanks, revealing that both Escherichia coli and Staphylococcus aureus are susceptible to ciprofloxacin and gentamicin. Among the tested antibiotics, ciprofloxacin exhibited the highest antibacterial activity against *E. coli*, producing a mean inhibition zone diameter of 21.0 ± 1.41 mm at 500 mg/mL. Gentamicin also demonstrated strong activity against the isolate, yielding an inhibition zone of 20.0 ± 1.41 mm at the same concentration. These findings suggest that the *E. coli* isolates recovered from the laboratory water tanks remain susceptible to fluoroquinolones and aminoglycosides. Similar observations have been reported in several studies where ciprofloxacin showed excellent activity against environmental and clinical isolates of *E. coli* due to its ability to inhibit bacterial DNA gyrase and topoisomerase IV, enzymes essential for DNA replication (Mwakyoma *et al*., 2023). The observed susceptibility may indicate limited exposure of the environmental isolates to selective antibiotic pressure compared with multidrug-resistant clinical strains.

Similarly, *S. aureus* exhibited considerable susceptibility to ciprofloxacin, with inhibition zones increasing from 18.0 ± 2.82 mm at 300 mg/mL to 19.0 ± 1.41 mm at 500 mg/mL. The susceptibility of *S. aureus* to ciprofloxacin observed in the present study is encouraging, particularly given the growing global concern about antimicrobial resistance in *Staphylococcal* species. The effectiveness of these antibiotics against the isolates in this study is consistent with reports demonstrating the continued efficacy of ciprofloxacin and gentamicin against susceptible strains of *S. aureus* (Isiordia-Espinoza *et al*., 2023). However, recent surveillance studies in Nigeria have reported increasing resistance among *S. aureus* isolates, emphasizing the need for continuous monitoring of susceptibility patterns (Agbo *et al*., 2024).

Table 3: Mean zones of growth inhibition (mm) exhibited by gentamicin and ciprofloxacin against bacterial isolates recovered from laboratory water tanks

Bacterial Isolates	Gentamicin (Zone of Inhibition, mm)			Ciprofloxacin (Zone of Inhibition, mm)		
	100 mg/mL	300 mg/mL	500 mg/mL	100 mg/mL	300 mg/mL	500 mg/mL
E. coli	9.0 ± 1.41	17.5 ± 3.53	20.0 ± 1.41	6.5 ± 2.12	13.0 ± 2.80	21.0 ± 1.41
S. aureus	11.0 ± 1.41	12.5 ± 0.70	16.0 ± 1.41	9.0 ± 1.41	18.0 ± 2.82	19.0 ± 1.41

Mean ± Standard deviation for zone of inhibition (mm)

The absence of resistance to ciprofloxacin and gentamicin among the isolates recovered from the water tanks suggests that these antibiotics remain effective treatment options against potential infections caused by these organisms. Nevertheless, the presence of *E. coli* and *S. aureus* in stored water remains a public health concern because these bacteria may serve as reservoirs of antimicrobial resistance genes that could be transferred to other microorganisms. Previous studies have shown that environmental water systems can contribute to the dissemination of antimicrobial resistance determinants even when isolates remain phenotypically susceptible to commonly used antibiotics (Mwakyoma *et al.*, 2023). Therefore, regular microbiological surveillance and proper maintenance of water storage facilities are necessary to ensure water safety and prevent the emergence and spread of resistant bacterial populations. Plates 1 and 2 show the antibiotic susceptibility patterns of both *E. coli* and *S. aureus*; each plate shows the zones of growth inhibition at various concentrations as well as the positive and negative controls adopted.

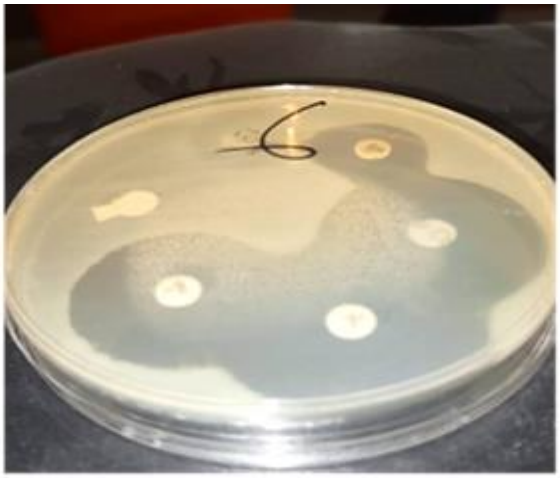

Plate 1: Kirby-Bauer disc diffusion assay showing antibiotic susceptibility pattern of *E. coli* at different ciprofloxacin concentrations

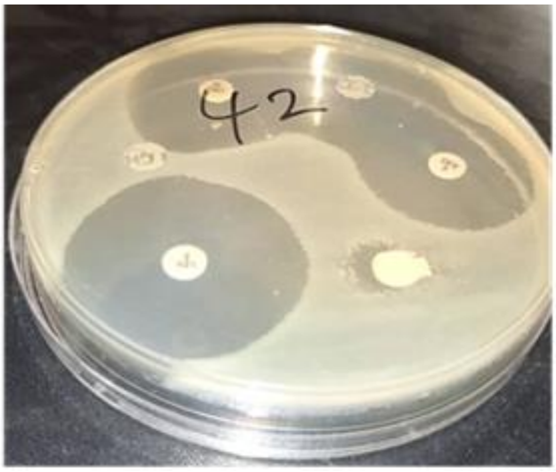

Plate 2: Kirby-Bauer disc diffusion assay showing antibiotic susceptibility pattern of *S. aureus* at different ciprofloxacin concentrations

CONCLUSION

This study highlights the critical concerns regarding water safety and fitness for use in the laboratories to conduct research activities. While other physicochemical parameters such as temperature, pH and electrical conductivity fell within the WHO permissible limits, the high OD observed is, however, worrisome and a good indicator of the significant presence of organic pollutants. In addition, the greatest concern regarding the efficiency and reliability of laboratory outcomes from research conducted using such stored water was the isolation of pathogenic microorganisms and the coliform indicator (*E. coli*). These findings therefore bring to focus the urgent need for regular monitoring, disinfecting and proper handling of these water tanks so as to reduce the possibility of interference with the interpretation of research due to the contamination of these microorganisms in the water tanks. The bacterial pathogens isolated in this study were found to be susceptible to ciprofloxacin and gentamicin antibiotics. Hence, no antibiotic-resistant strain was isolated.

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

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