

Physicochemical and Bacteriological Analysis of Wawan Dam Rafi Water, Karaftai, Kazaure Local Government Area at the Early and Late Rain Fall

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Abstract: *Water quality is crucial for human well-being and existence, but its availability remains a challenge in underdeveloped and developing countries. The demand for quality water in urban cities in Nigeria has increased due to limited public water supplies. This study conducted physicochemical and bacteriological analysis of Wawan Rafi Dam water during early and late rain fall. Results showed fluctuating temperature and pH levels, high dissolved ion concentrations, and within WHO permissible levels. Turbidity, nitrate, and phosphate levels were also found. Heavy metal concentrations were higher during the early rain fall compared to the late rain fall. However, zinc levels did not exceed the maximum permissible level. Manganese, Co, Pb, and Cu concentrations varied between 0.05 to 0.15 mg/L and 0.01 to 0.05mg/L, respectively. Gram staining identified two bacteria from Wawan Rafi Dam water: E. coli and E. cloacea. E. coli had a rod shape, motile, variable capsule, non-sporing, and flagellated appearance, while E. cloacea had straight rod-like cells bound in clusters with few laterally inserted flagella with pili and mucoid material and numerous bubbles on the surface. DNA sequencing analysis revealed that MA1 merged with Escherichia coli and MA2 merged with Enterobacter cloacae. All two strains showed sensitivity to gentamycin, chloramphenicol, erythromycin, and Ciprofloxacin. E. Cloacae and E. coli showed high resistance to trimethoprim and ampicillin, while E. cloacae was sensitive to trimethoprim and ampicillin. These findings contradict the belief of Karaftayi people who believe the water has medicinal properties.*

Keywords: Water Quality, Early and Late Rain Fall, Heavy Metals, Bacteriological Analysis and Physicochemical Analysis

I. INTRODUCTION

The quality of water is a critical factor affecting people's health(Hicham *et al.*, 2022), and waterborne diseases such as typhoid fever(Fida, *et al.*, 2022), cholera, and dysentery are caused by microbiological contaminants and pollutants. The demand for quality water in urban cities in Nigeria has been increasing(Oyedejiet *al.*, 2010) due to limited public water supplies, leading to the production of packaged drinking water by private companies(Isikwue, 2014). However, contamination of these quality waters is well-known from physical, chemical, and microbiological factors(Danso-Boateng and Frimpong, 2013). Most impurities are known to have originated from Dam water and might persist due to limited operative techniques to get rid of them.

Waterborne diseases are essential to human well-being and perpetual existence(Saxena *et al.*, 2022), but more than one billion people worldwide do not have access to safe drinking water(Olawuyi, 2006). Infectious diarrhea is responsible for the greatest burden of this morbidity and mortality(Fenwick, 2010: Jeje and Oladepo, 2012), with children less than five years of age (WHO, 2000) being the most severely affected. In 2001, infectious diseases accounted for an estimated 26% of deaths worldwide (Kindhauser, 2003: Davison *et al.*, 2005). Gastro-intestinal water-borne infections

are among the most emerging and re-emerging infectious diseases throughout the world, commonly associated with Dam water.

Untreated Dam water is not recommended for consumption as potable water (WHO, 2004), as it lacks acceptable quality in terms of its physical, chemical, and bacteriological parameters (Mukate *et al.*, 2018). It should be colorless, tasteless, and odorless, free from fluorine, arsenic, nitrates, and nitrite and lead contents, and not be associated with waterborne disease during and after use. Dam water lacks all these features

The health aspects of environmental quality were among the first to receive scientific attention through the recognition of waterborne diseases (Nwabore *et al.*, 2016). Waterborne diseases are caused by pathogenic microorganisms which are directly transmitted when contaminated fresh water is consumed (Batalini de Macedo *et al.*, 2022). Contaminated Dam water, used in the preparation of food, can be the source of food-borne diseases through consumption of the same microorganisms (Onyango and Angienda, 2010).

In Karaftayi village, the majority of households obtain their drinking water from Wawan Rafi Dam in order to survive. They base their belief that the water from this Dam is safe to drink and uncontaminated on organoleptic criteria. However, the water they obtain is occasionally contaminated, and often not protected or poorly maintained. Dam pollution is typically made worse by various human activities, including defecating in public places, using pit latrines, dumping trashes, farming, and discharging heavy chemicals at industrial sites adjacent to the dam.

Health and water go hand in hand; two million people, most of them children, die each year as a result of water-borne illnesses. Unsafe water, poor sanitation, and sanitary issues are at blame for almost 88% of all diarrheal illnesses. Unfortunately, just 39% of Nigerians have access to clean water (Kumar *et al.*, 2022).

This research work was carried out in Wawan Rafi Dam of Karaftayi in Kazaure local government area of Jigawa State between six months. The research aimed to assess the quality of Wawan Rafi Dam water in Karaftayi by analyzing physicochemical parameters and bacteriological analysis of the water. The objectives of the current study include determining the physicochemical properties of the Wawan Rafi Dam water, isolating and characterizing isolated bacteria of public health importance, and determining the antibiotic susceptibility of the isolated bacteria.

STUDY AREA

Wawan Rafi, located in the Karaftayi of Kazaure L.G.A, spans 1,780 km² and has an undulating landform with dunes and loamy soil. The inhabitants are mostly farmers and cattle rearers, who engage in tie and dye activities during the dry season. The Wawan Rafi Dam, surrounded by hills and high land, is a natural resource that transports essential and toxic minerals and wastewater. The water has various domestic uses, including irrigation, fishing, and recreational activities. (Sagagiet *al.*, 2022), which signifies the importance of this work. The description of the sample site is presented in Fig. 1:

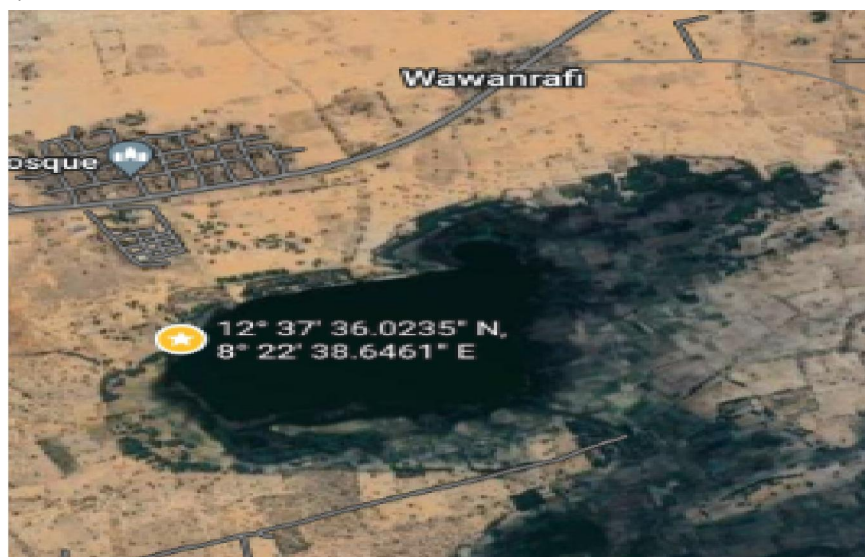


Fig. 1 Google Map of Wawan Rafi

II. METHODOLOGY

Sample Collection

Water samples were aseptically collected in clean sterile one 1 litre plastic containers, rinsed with de-ionized water and subsequently with the dam water before it was finally collected using stratified sampling techniques at 0.1, 1.0 and 1.5 meter depth, and stored in a freezer to keep them in prime condition prior to analysis as described in Onianwa and Fakayode, (2000). The coordinate location of each sampling site was read and recorded using Garmin GPS (table 1). Dam water was collected during the early and late rain fall (Early and late rain fall). The sampling points were tagged as Point A (Up dam), Point B (Mid dam) and Point C (Down dam). The water was collected and filled to capacity to prevent entrapment of air and was promptly closed and transported to the laboratory of Hussaini Adamu Federal polytechnic Kazaure in a Cool Box container at 4°C (Amoo *et al* 2018)

Table 1. GPS reading of water sample collected from Wawan Rafi Dam Water between early and late rain fall (June and September) of 2023

Sample	Water Sample ID	Depth(m)	Longitude and Latitude
A1	W ₁ , W ₂ and W ₃	0.5	Long: N12° 37'35.9"
			Lat: E008° 22'23.5"
A2	W ₄ , W ₅ and W ₆	1.0	Long: N12° 37'35.9"
			Lat: E008° 22'23.5"
A3	W ₇ , W ₈ and W ₉	1.5	Long: N12° 37'35.9"
			Lat: E008° 22'23.5"
B1	W ₁₀ , W ₁₁ and W ₁₂	0.5	Long: N12° 37'36.0"
			Lat: E008° 22'248"
B2	W ₁₃ , W ₁₄ and W ₁₅	1.0	Long: N12° 37'36.0"
			Lat: E008° 22'248"
B3	W ₁₆ , W ₁₇ and W ₁₈	1.5	Long: N12° 37'36.0"
			Lat: E008° 22'248"
C1	W ₁₉ , W ₂₀ and W ₂₁	0.5	Long: N12° 37'418.0"
			Lat: E008° 22'19.6"
C2	W ₂₂ , W ₂₃ and W ₂₄	1.0	Long: N12° 37'418.0"
			Lat: E008° 22'19.6"
C3	W ₂₅ , W ₂₆ and W ₂₇	1.5	Long: N12° 37'418.0"
			Lat: E008° 22'19.6"

Key: W_w = Wawan Rafi

Physicochemical Analysis

The water samples were collected monthly at three locations during early and late rain fall in 2023. Physicochemical parameters such as temperature, pH, oxygen demand, biological oxygen demand, conductivity, turbidity, total dissolved solid, nitrate, and phosphate were determined using standard methods described by the American Public Health Association (APHA, 1998).

Temperature was measured using a mercury-filled glass thermometer, while pH was measured using a digital PH meter instrument. Conductivity was determined using an AOAC (2004) approved standard procedure, with a Hach model CO150 meter calibrated to 14.7ms/m at 25°C as stated in González and Herrador, (2007). Biological oxygen demand was determined using Ademoroti (2006)'s standardized methods, with samples incubated for five days at 20°C. The dissolved oxygen content of the samples was measured before and after the incubation period.

Total Dissolved Solid (TDS) was determined using a vacuum pump, glass beaker, membrane filter paper, and glass fiber filter disk. The weight of each dish was then measured before being transferred into evaporating dishes. The TDS was calculated using the equation (Mohammed *et al.*, 2016).

$$\text{TDS (mg/L)} = \{(A - B) \times 1000\} \div \text{Volume of sample}$$

Where, A = Weight of evaporating dish and dried residue

B = Weight of evaporating dish

Transparency (Turbidity) was determined using a 25cm diameter Secchi disc, which was lowered into the water at each station until it became visible. The depth of appearance and disappearance were measured to the nearest cm, and the average was taken.

Chemical Parameters

Heavy metals such as lead (Pb), copper (Cu), zinc (Zn), manganese (Mn) and cobalt(Co), copper (Cu) and Nickel (Ni) was analyzed by Atomic Absorption Spectroscopy (AAS) with the Perkin Elmer PinAAcle 900 (United States). All analysis for metal elements was carried out in triplicate. Phosphate and nitrate were analysed by UV (Sachdeva *et al.*, 2023).

Test for Phosphate and Nitrate

Nitrogen-Nitrate, Ammonium-Nitrate, and Phosphate were digitally determined in water samples using the HANNA multi parameter logging spectrophotometer (HI83200). Standard procedures were used to determine Nitrogen-Nitrate, Ammonium-Nitrate and phosphate concentrations. The cadmium reduction metal method 8036 was used to determine nitrate as nitrogen. The cadmium metal in the added reagent converted all of the nitrate in the samples to nitrate; phosphate was measured using Sulfa Ver methods 8051.

Determination of some heavy metal

Sample digestion

Each sample received 50ml in 250ml beakers. 5cm³ of concentrated nitric acid was added and carefully heated in a fume hood chamber on a powered electrically connected sand-bath with periodic additions of 10ml and Concentrated Nitric acid until the volume was reduced to 20 ml. The sample solution was then allowed to cool before 4ml of perchloric acid (HClO₄) was added and heated until it was clear. Prior to analysis, the samples were diluted with deionized water, filtered into 100ml volumetric flasks, made up to mark, transferred into a capped labeled plastic bottle, and refrigerated. The acid ratio used for digestion, HNO₃:HClO₄, was 5:1 (Abubakar and Abdullahi; Hu & Qi, 2014).

A blank sample was also prepared by digesting the same proportion of the reagents used in the sample digestion without the sample under the same experimental conditions (Hu & Qi, 2014). The resulting solution was diluted to the appropriate concentration with distilled water, thoroughly mixed, and used to determine the heavy metals in the water sample.

AAS analysis

Standard stock solutions (1000 mg/L) of zinc, lead, cobalt, manganese, copper and Nickel of high purity were used where an appropriate volumes were diluted to obtain the working standard solutions of various concentrations for plotting calibration curves. The metals were analyzed in the digested sample and the results were presented in chapter four (Duranet *et al.*, 2007).

Analysis of bacteriological parameters

MPN Method

For bacteriological analysis of water, total bacterial count was done by standard plate count method and the most probable number (MPN) method was employed for determination of total coliforms. MPN Method was conducted in three steps:

- 1) Presumptive test
- 2) Confirmed test
- 3) Completed test

1) Presumptive test

Presumptive test functions as the primary presumption for the presence of Gram negative coliform bacteria in the samples. In this test, MacConkey broth is commonly used for lactose fermentation for the presence of the indicator bromocresol purple. The inverted Durham's tube is used for the detection of gas formation by Gram negative coliform bacteria. The color changes of media into yellow and on collection of gas in Durham's tube can be assumed that coliform bacteria are present in these samples. 0.1 ml and 1ml each of water sample were added to 5 ml of single strength MacConkey Broth. Furthermore, another 10 ml of double strength MacConkey broth were added to 10 ml of the water sample. After 48 hour incubation at 37°C, the number of positive tubes were recorded from each set and compare with standard chart to give presumptive coliform count per 100ml water sample.

2) Confirmed Test

In the confirmed test, positive samples from presumptive test were selected to determine the coliforms are of indicator bacteria of fecal origin *Escherichia coli*. Eosine Methylene Blue (EMB) media was used to differentiate *Escherichia coli* from Gram negative coliform bacteria by the production of greenish metallic sheen that confirms the presence of indicator bacteria *E. coli*. The production of color indication from colonies can be observed after 24 hours incubation at 37°C by streaking loopful sample from positive tubes.

3) Complete Test

The bacterial colonies on EMB media from confirmed test were inoculated in LB broth at 44.5°C with Durham's tube and subculture the colony on Mac Conkey agar plate. Presence of faecal indicator *E.colis* confirmed by the production of gas and color changes in media. For further complete confirmation, a satisfactory differentiation within the coliform group was done by indole, methyl red, Voges-Proskauer and sodium citrate (IMViC) tests which are commonly recommended for such differential determination according to Bergey's Manual of Systematic Bacteriology.

The study involved tenfold dilutions of Wawan Rafi water samples and plated with dilution factors of 10⁻³ and 10⁻⁵. The molten agar was poured into each plate containing the diluted inoculums, and the cultured plates were incubated for 24 hours at 37°C. After the incubation period, distinct colonies on the plates were aseptically sub-cultured onto freshly prepared solidified nutrient agar and incubated at 37°C for 24 hours. Pure gram negative bacterial isolates were observed for morphological and molecular characteristics.

Biochemical characterization of these pure gram negative bacterial isolates included gram staining, catalase test, methyl red test, Voges-proskauer test, indole production, and growth on differential media on Eosine methylene blue (EMB) agar for presumptive identification. Antibiotic sensitivity tests were conducted with Mueller Hinton agar and antibiotic discs to determine which isolates were susceptible, intermediate, or resistance.

The catalase test was performed using a slide test, indicating the presence of catalase. The indole test involved adding Kovacs' reagent to the culture in a tube containing 1 ml of indole broth, and results were observed within 30 seconds.

The gas production test involved inoculating isolated colonies on triple sugar iron agar (TSIA) and slant in the test tube, and incubating at 37°C for 18 to 24 hours.

Molecular Characterization of *E. coli* and *E. cloacae* Isolate

The genomic DNA from bacterial cultures of tentatively identified *E. coli* and *E. cloacae* strains grown overnight in nutrient broth, were extracted according to the method given by Nair et al. (2021) and Pospiech and Neumann (1995).

Primers

The 16S rRNA gene based primers namely 16E1 (GGGAGTAAAGTTAATACCTTTGCTC), 16E2 (TTCCCGAAGGCACATTCT) and 16E3 (TTC-CCGAAAGGGACCAATC) were used based on the known specific target 16S rRNA gene sequences of *E. coli* and *E. cloacae* (Tsen et al., 2006).

For DNA sequencing, bacterial 16S rRNA agent based universal primer namely 27F (5'-AGAGTTT-GATCCTGGCTCAG-3') and 1492R (5'CGGT-TACCTTGTTACGACTT-3') were used (Aminatun et al., 2024 and Kumaet al., 2021).

PCR Amplification

The reaction mixture (20µL) contained 10 pmol of each primer, 0.2 mM of each dNTP (MgCl₂), 1XPCR buffer, 20µ L of DNA solution and 1 U/µL of Taq DNA polymerase (Bangloregeinei). Amplification was carried out in a thermal cycled as follows: initial denaturation at 95°C for 5 min, then 35 cycles consisting of denaturation at 95°C for 30 s, annealing at 55°C for 30s, and extension at 72°C for 2 min, and a final extension for 10 min at 72°C. Final hold of amplified PCR product was at 4°C. Amplified product were visualized on a 2.0% agarose gel along with 100 and 500 bp DNA ladders. The PCR products amplified with universal primers were sequenced. The sequences of the 16S rRNA of the isolates were compared with available standard sequences bacterial lineages in the NCBI Genebank using nBLAST. The obtained sequences were submitted to the NCBI Genebank.

Antibiotic Susceptibility Test

Antibiotic susceptibility test of *E. coli* and *E. cloaca* were determined according to the disc-diffusion method of Kirby-Bauer et al. (1996) on Mueller-Hinton agar plate. A total of six antibiotic namely ampicillin (AMP 10µg/disc), gentamicin (GEN30µg/disc), Chloramphenicol (CHP 25µg/disc), erythromycin (Eryt15µg/disc), trimethoprim (Trim5 µg/disc) and Ciprofloxacin (CIP5µg/disc) were used. Results were interpreted according to the guidelines of CLSI, (1999).

Data Analysis

The result of the physicochemical parameter in relation to the frequency of occurrence of the Bacteria were statistically analysed using SPSS package.

III. RESULT AND DISCUSSION

The results of the physicochemical and bacteriological analysis of Wawan Rafi dam water in Karaftayi of Kazaure local government area are presented for discussions.

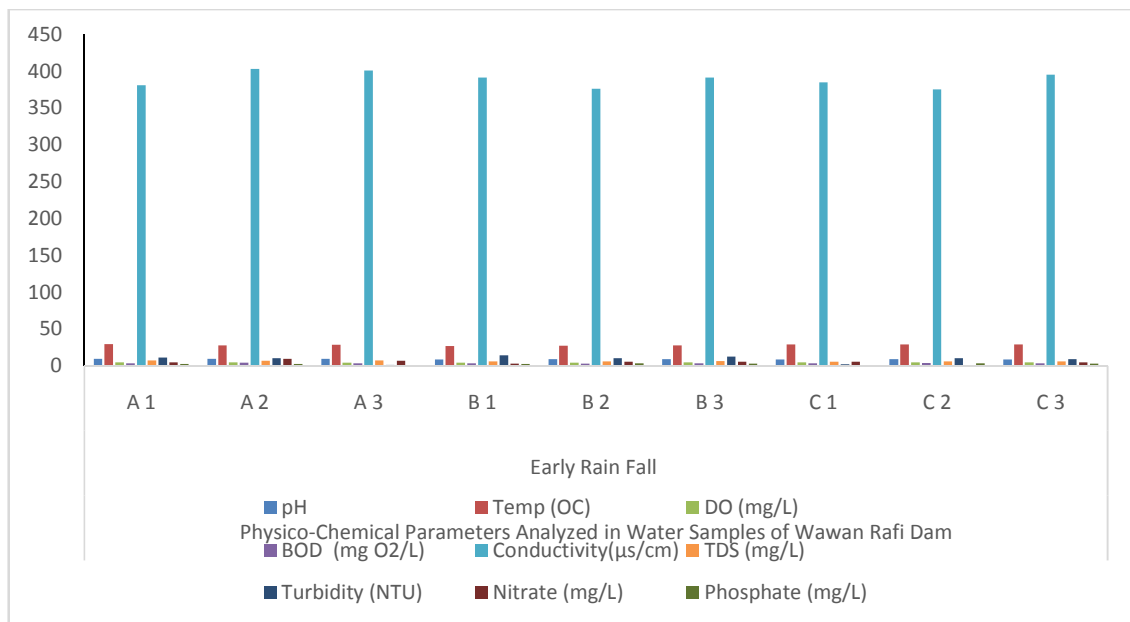


Figure 1: Result of Physicochemical Analysis of Wawan Rafi Dam during the early Rain Fall, 2023

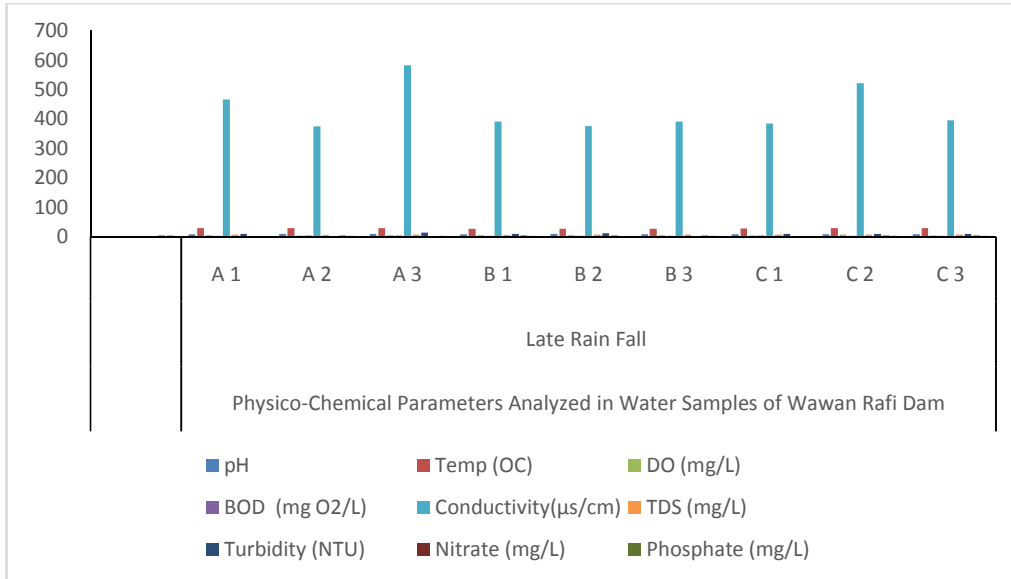


Figure 2: Result of Physicochemical Analysis of Wawan Rafi Dam during the late Rain Fall, 2023

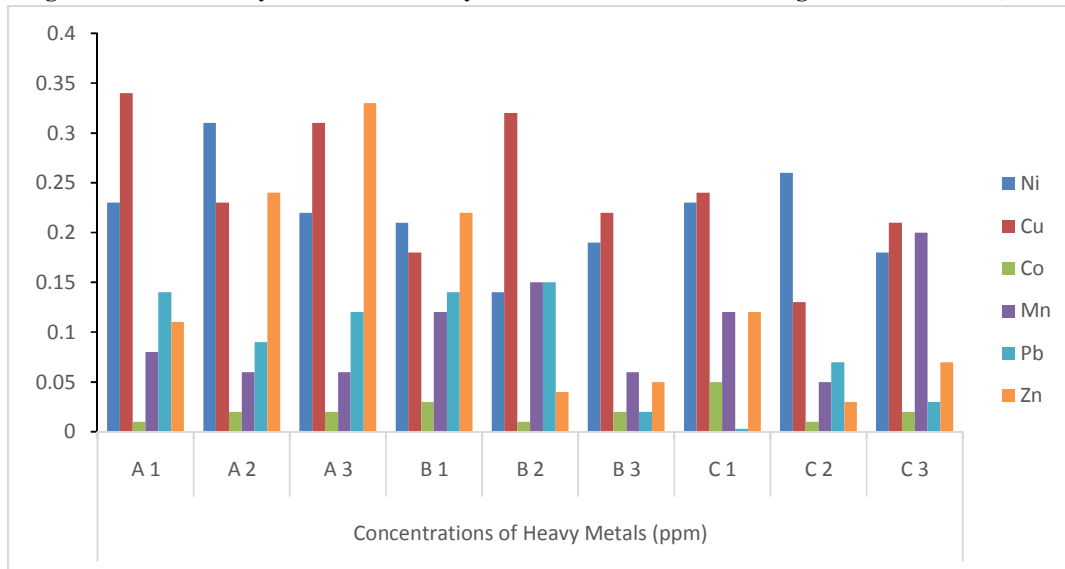


Figure 3: Result of Heavy metal Analysis of Wawan Rafi Dam during the Early Rain Fall, 2023

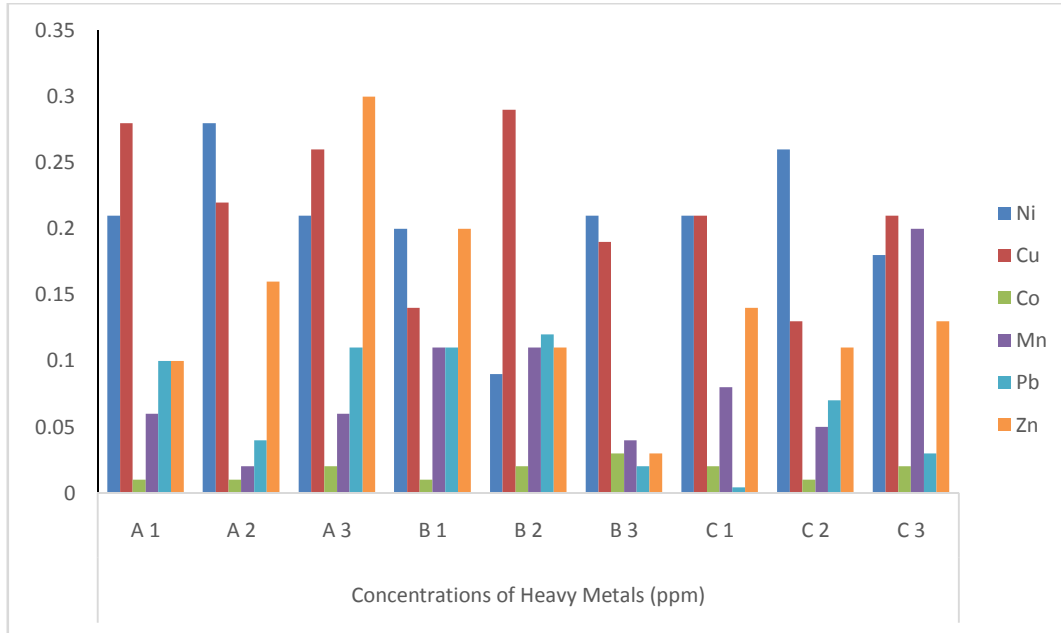


Figure 4: Result of Heavy metal Analysis of Wawan Rafi Dam during the Late Rain Fall, 2023

Table 3: Morphological Test on Acid and Gas production of bacteria from Wawan RafiDam water samples

S/N	Sample	Growth			Acid Production			Gas Production		
		single Strength		Double Strength	single Strength		Double Strength	single Strength		Double Strength
		0.1ml/10ml	1ml/10ml	5ml/5ml	0.1ml/10ml	1ml/10ml	5ml/5ml	0.1ml/10ml	1ml/10ml	5ml/5ml
1	A1	Yes	Yes	yes	+	+	+	+	+	+
2	A2	Yes	Yes	yes	+	+	+	+	+	+
3	A3	Yes	Yes	yes	+	+	+	+	+	+
4	B1	Yes	Yes	yes	+	+	+	+	+	+
5	B2	Yes	Yes	yes	+	+	+	+	+	+
6	B3	Yes	Yes	yes	+	+	+	+	+	+
7	C1	Yes	Yes	yes	+	+	+	+	+	+
8	C2	Yes	Yes	yes	+	+	+	+	+	+
9	C3	Yes	Yes	yes	+	+	+	+	+	+

Table 4. Total bacterial count (CFU/ml) of Wawan Rafi water samples

S/N	Sample	Coliform Count	
		Early Rain fall	Late Rain Fall
		10^{-3}	10^{-4}
1	A1	3.5×10^{-5}	2.1×10^{-5}

2	A2	3.8×10^{-4}	1.2×10^{-4}
3	A3	6.7×10^{-5}	2.3×10^{-5}
4	B1	2.0×10^{-5}	1.1×10^{-5}
5	B2	2.2×10^{-5}	1.1×10^{-5}
6	B3	2.2×10^{-5}	1.5×10^4
7	C1	2.3×10^{-5}	1.1×10^{-5}
8	C2	2.1×10^{-5}	1.1×10^{-5}
9	C3	2.3×10^{-5}	1.5×10^4

Table 5: Percentage distribution of isolated bacteria from Wawan RafiDam water samples

S/N	Sample	Number of <i>E. Coli</i>	Frequency (%)	Number of <i>C. Cloacae</i>	Frequency (%)
1	A1	6.00	15.00	4.00	9.52
2	A2	8.00	20.00	6.00	14.29
3	A3	6.00	15.00	3.00	7.14
4	B1	3.00	7.50	9.00	21.43
5	B2	6.00	15.00	5.00	11.90
6	B3	4.00	10.00	7.00	16.67
7	C1	3.00	7.50	2.00	4.76
8	C2	2.00	5.00	4.00	9.52
9	C3	2.00	5.00	2.00	4.76
TOTAL		40.00	100.00	42.00	100.00

Table 6: Morphological and Biochemical Characteristics of Presumptive *E. Coli* and *Enterobacta cloacae* Isolate

Isolate Identification Cod	Morphological Characteristic		Biochemical Test					Probable Organism
	Colonial morphology Eosine methylene blue (EMB) agar	Cellular/microscopic morphology	Gram reaction	catalase test	methyl red test	Voges-proskauer test	indole production	
MA1	Smooth, circular, greenish black colour colonies	Rod shape, motile, variable capsule, non-spore, flagellated	-ve	+Ve	+Ve	-Ve)	+Ve	<i>E. Coli</i>

ODMA2	Fair growth, pink-coloured colonies, and has no sheen	Straight rod-like, cells are Bound in clusters with few laterally inserted flagella, it has a pili and mucoid material and numerous bubbles on the surface	+Ve	+Ve	-Ve	+Ve	+Ve	<i>Enterobacta cloacae</i>
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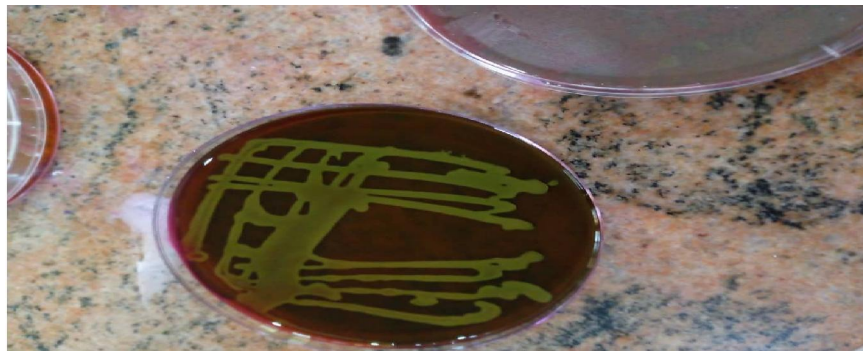


Figure 5(a) Cultured *E. coli*



Figure 5 (b) Cultured *E. cloacae*

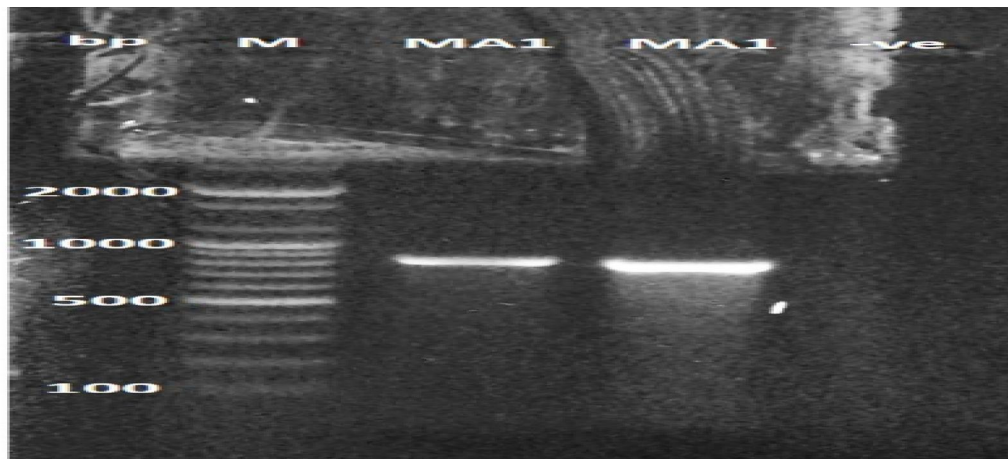


Figure 6: Sequence Characteristics of *Escherichia coli* and *Enterobacter cloacae* Strains

Table 7: PCR amplification of 16S rRNA Gene Bacterial Isolates

Band	Description	scientific Name	Max score	total score	query cover	E cover	per value	ACC	iden.
MA1	Escherichia coli strain HPCAQ7CR13 16S ribosomal RNA gene	partial sequence	1029	1029	96%	0.0	91.95	779	MG566068.1
MA2	Enterobacter cloacae strain ADY34 16S ribosomal RNA gene,	partial sequence	654	654	87%	0.0	0	84.42	1073,

Antibiotic susceptibility pattern of *E.coli* and *E. cloacae*

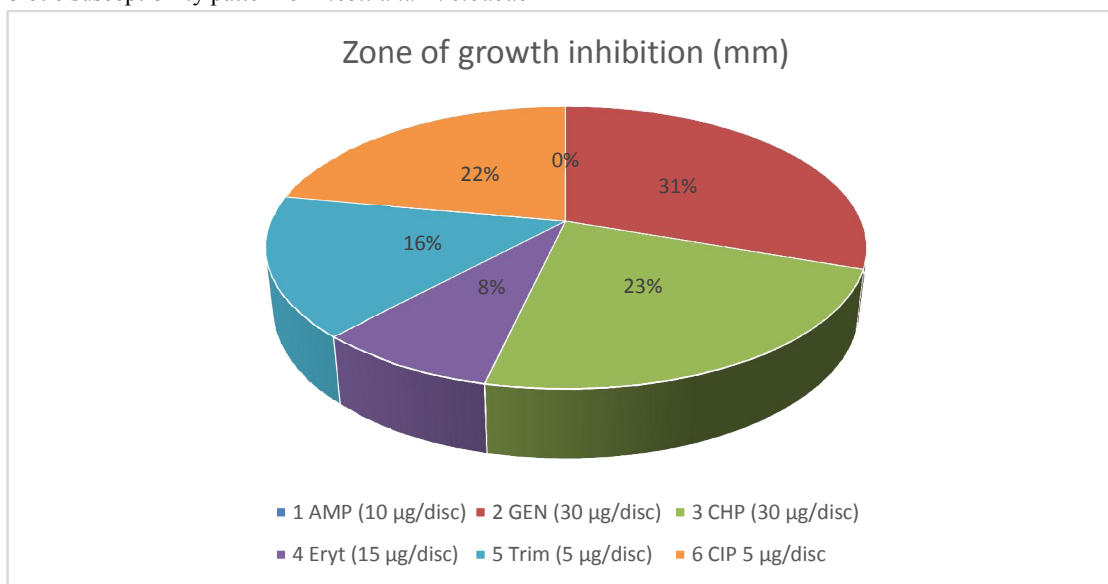


Figure 7: Antibiotic resistance pattern of *E. coli* and *E. cloacae* isolates from Wawan Rafi Dam Water

IV. DISCUSSION

Physicochemical parameters

During early and late rain fall, the temperature and pH levels fluctuated. The highest pH recorded was 9.5 in Site A during the early rain fall of 2023, while the lowest pH was 8.0 mostly across the month and all the sites in both early and late rain fall. The highest mean pH levels of 9.5 observed indicating that the Wawan Rafi Water the study areas are alkaline. Similar result was reported by Sagagiet *al.*, (2022). However, all pH values were slightly above the WHO (2011) guideline range of 6.5-8.5. The water temperature ranged from 26.40 to 30.20 in both early and late rain fall, which falls within the acceptable range.

The conductivity values showed that the water samples had the highest dissolved ion concentration, with a conductivity value ranged between 207 to 582 $\mu\text{s/cm}$. All of the conductivity values were below the WHO (1999) recommended limit of 500-1500 S/cm, but most of the highest values were recorded in the early rainfall, which if associated with dissolve ion deposited from the runoff water (Sagagiet *al.*, 2022)

The dissolved oxygen (DO) levels ranged from 4.10 to 5.80 mg/l, which were slightly higher than the WHO (2014) permissible levels of 6mg/l. the highest value of DO were recorded in the late rain fall with is attributed to sufficient oxygen released from green plants during the late period of rainy season. This is important for supporting a diverse aquatic ecosystem. The biological Oxygen demand is more during the early rain fall. The biochemical oxygen demand (BOD) values ranged from 2.87 to 4.10 mg/l, which were lower than the WHO (2014) acceptable limit of 6 mg/l. This indicates low microbial activity. The total dissolve solid levels ranged from 5.30 to 7.60 (mg/L), slightly higher than the WHO (2014) permissible level of 500mg/L.

The turbidity levels all through the period ranged from 2.04 to 14.3 NTU. The highest value obtained is slightly higher than the WHO (2014) permissible level of 5 NTU. This could be due to impurities entering the open water. This could be due to impurities entering the open water.

The levels of nitrate and phosphate in the water samples ranged from 0.09 to 9.01 mg/l and 0.15 to 2.97mg/l respectively. These values for nitrate in both early and late rain fall were lower than those found in Field *et al.* (2023)'s study and below the WHO's acceptable threshold of 50 mg/L, while phosphate exceeded the permissible limit of 1.0mg/l. High concentrations of nitrate and phosphate can lead to eutrophication (Ogundairo *et al.*, 2024) which were observed in both early and late rain fall.

The heavy metal concentrations were observed to be higher during the early stage of rain fall compared with the late rain fall. Tables 4.4 reveal the concentrations of the heavy metals (Ni, Cu, Co, Mn, Pb, and Zn) found in water sample of Wawanrafi in 2023. The results indicated that Zn values in water samples varied from 0.03 mg/L to 0.33mg/L. However, the recorded Zn levels during the sampling period did not exceed the maximum permissible level in the water (5 mg/L) (Sagagiet *al.*, 2022). Even though Zn is considered to be an essential micronutrient for human metabolic processes and enzymatic activities, at higher concentrations, however, zinc can be toxic to the living organism

From the results, the manganese concentrations in water samples were found to be in the ranged of 0.05 to 0.15 mg/L between early and Late rain fall. These values were found to be higher than the allowed values according to WHO's 0.05mg/L (WHO, 1996). Manganese has been shown to have a greater tendency to accumulate in unwashed leaves (Sagagiet *al.*, 2022).

Cobalt concentration in this work varied between 0.01 to 0.05mg/L while lead varied between 0.02 to 0.15 mg/L. Lead is a poisonous metal that is used in significant quantities in many electronic devices. Lead is well known to cause a range of health risks ranging from behavior changes and pedagogical disabilities to seizures and subsequent demise (Sagagiet *al.*, 2022). Lead toxicity can affect renal and neurologic systems as most physicians report that acute lead exposure may lead to anemia which can subsequently lead to heme inhibition and destruction of erythrocytes (Sanborn *et al.*, 2002). The fact Pb concentrations were within the WHO recommended limit of 0.2 mg/L for vegetable water suggests that it is safe for

In this study, Cu was found in the range of 0.21 to 0.34 mg/L while, Ni concentration detected in the water in this study ranged between 0.14 to 0.31 mg/L, which. Nickel acts as an activator of the enzyme urease and is considered an essential trace element for human, animal, as well as plant health.

Result of Analysis of bacteriological parameters

Total bacterial count (CFU/ml) and Percentage distribution of isolated bacteria from Wawan Rafi water samples

The highest total plate counts observed in the water samples indicated the presence of high organic matter and related nutrient sources. The bacterial load were observed to be high, during the early rain fall compared with that of late rain fall. The likely primary source of bacterial loads observed might include the surface run-off, sewage treatment facilities, natural soil/plants bacteria and improper management activities of the inhabitants like washing, refuse disposal, faecal droppings, dipping of different fetching materials inside those water source which were observed. Water sample collected during the early rain fall had the highest number of coliform compared with that collected during the late rain fall. These is attributed to the environmental contamination as a result of proximity of the Dam to bush area where both people and animals defecates. *E. coli* and *E. cloacae* are the two predominate microorganisms (Table 3) isolated and identified during this study and they may have one pathogenic effect or the other. Although, water from these Dam may look clean and have no undesirable odour or taste, pathogens found in such water could be harmful by causing serious illnesses.

The total coliform counts examined during this study were exceedingly high as against the EPA maximum contamination level (MCL) for coliform bacteria in drinking water of zero total coliform per 100ml of water (Environmental Protection Agency (EPA), 2002), which may be link to faecal contamination (Environmental Protection Agency (EPA), 2002, Osunide, and Enuezie, 1999), which makes it not portable and so unfit for human consumption. The implication is that they constitute a serious risk to the public health.

Morphological and Biochemical Characteristics of Presumptive *E. Coli* and *Enterobacta cloacae* Isolate

Results gotten from Gram staining indicate the two isolates bacterial from Wawan Rafi Dam water, one having a Rod shape, motile, variable capsule, non-sporing and flagellated believe to be *E. coli*, and the other having Straight rod-like, cells Bound in clusters with few laterally inserted flagella with pili and mucoid material and numerous bubbles on the surface believe to be *E. cloacea* that were positive to Gram reaction ,catalase, methyl red, Voges-proskauer and indole production test, but *E. coli* and *E. Cloacea* were negative to Voges-proskauerand methyl red test respectively (Table 5).

Molecular Characterization of *E. coli* and *Enterobacter cloacae*

The antibiotic resistant bacteria, *Escherichia coli* and *Enterobacter cloacae* were screened for DNA associated with the resistance pattern of the antibiotics to which they were resistant to. They were found to possess different resistance genes for different antibiotic type which indicated that the resistant genes were encoded in the isolated DNA. This study has clearly shown that water from Wawan Rafi Dam harbour multiple antibiotic resistant bacteria strains and as such, individuals drinking this water or using it for other domestic purposes may ingest resistant strains which could become part of the human microflora. As a result of selection pressure, such organisms may establish themselves within the individuals and become predominant microflora (Richman, 1999). Therefore, infections caused by such organisms that can only be treated with strong antibiotics

Antibiotic susceptibility pattern of *E.coli* and *E. cloacae*

The results revealed that, all the two strain showed sensitivity to gentamicin, Chloramphenicol, erythromycin and Ciprofloxacin. The *E. Cloacae* and *E. coli* of this study showed high resistance to trimethoprim andampicillin with zero zone of inhibition respectively. On the other hand, the *E. Cloacae* showed to be sensitive totrimethoprim and *E. coli* was sensitive to ampicillin with 44 mm and 29 mm inhibition zone respectively as presented in Table 10. The result of these findings is in contrast to believe of the inhabitance of Karaftayivillage who are of the believed that the water has medicinal properties.

V. CONCLUSION AND RECOMMENDATION

he physicochemical parameters of Wawan Rafi Dam reveal fluctuating temperature and pH levels during early and late rainfall, with conductivity values below the WHO's recommended limit. The water contains high concentrations of nitrate and phosphate, which can lead to eutrophication. Heavy metal concentrations are within permissible limits, but lead is a poisonous metal used in electronic devices. The water may also contain antibiotic-resistant bacteria, which can

become part of the human microflora. These organisms can only be treated with strong antibiotics, such as trimethoprim and ampicillin. The study suggests consistent inspection of heavy metals and microbes in water bodies to avoid infections and encourages proper treatment of water before use. Environmentalists, administrators, and public health workers should create public awareness to avoid direct drinking of water in such areas to reduce health risks.

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