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Microbiological and Physicochemical Evaluation of Roof-Harvested Rainwater obtained from Hezekiah University, Umudi, Imo state, Nigeria

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ABSTRACT

Roof-harvested rainwater (RHRW) has remained one of the oldest sources of alternative potable water in most rural communities in the world. In this study, two major samples were obtained from the cafeteria/canteen and female hostel at the Hezekiah University, Umudi. Total heterotrophic bacterial count, total coliform count and *Staphylococcal* count. The coliform composition was determined using the Most Probable Number (MPN) 3-tube technique. Biochemical tests and colonial morphology were employed in the identification of the bacterial isolates associated with the RHRW samples. Total heterotrophic bacterial and coliform counts for the samples obtained from the cafeteria/canteen was 5.0 Log₁₀CFU/mL and 3.0 Log₁₀CFU/mL while the coliform content was 10 and 36 CFU/100mL was observed for the samples obtained from the female hostel and cafeteria respectively. The physicochemical composition of the RHRW from the cafeteria was observed to have a pH of 6.9, electrical conductivity of 110.56 (µS/cm), total dissolved solids was 11.97, total hardness was 13.49 mg/L while total acidity was 15 mg/L. The bacterial flora identified from the RHRW were *E. coli*, *Enterococcus* sp., *Klebsiella* sp. and *Pseudomonas* sp., while *E. coli* was identified to be the most frequent. There is a need for the University management to sensitize both staff and students on the potential dangers of using roof-harvested rainwater for domestic purposes. Non-governmental organizations must sensitize rural communities to the need for the treatment of rainwater before usage.

Keywords: Roof harvested rainwater; Potable water; Most probable number; Bacterial flora; Coliform; Roof; Hostel; Physicochemical; Rural communities; Colonial morphology; Cafeteria; Most frequent isolates.

1.0. Introduction

1.1. Background to the Study

Sanitation and hygiene are pivotal for man's survival as it has taken a central stage in the global discussion. Although water is an essential resource for existence, potable water has become a major challenge in developing countries like Nigeria for quite some time now. The wholesomeness of water suitable for domestic use is at variance with a global demand for safe water. The availability of clean and potable water reduces the fatalities associated with water-borne diseases to about 65% and reduces mortality by 26% in the world today. Inadequate supply of 'safe water' has been correlated with several health challenges which have plagued Africa in recent times. In a related study, some researchers have reported that the availability of clean water can be linked to the level of civilization which has resulted in the widely accepted cliché access to clean water is a fundamental human right [1].

The practice of harvesting rainwater is an old one with persons of both socio-economic classes indulging in it. Although this activity is pronounced in areas with poor topology and water level; this may involve placing containers at specific dropouts while some people construct gutters and channel this water to storage thanks for domestic uses while others may place their earthen pots or containers at a distance away from the roof and use them as drinking water source. According to Saidu *et al.* [2] the physicochemical indices of water in any location is reflective of activities that may hamper on the quality. Hence, the standard of rainwater collected is primarily controlled by age and quality of roofing materials, topology, aerodynamics of the environment, pollution, microbial activities and anthropogenic a from industries, automobiles and anthropogenic activities, the presence of dirt, debris and birds or rodents dropping on roofs and rainwater catchments. Adeniyi and Olabanji [3] identified that the



bacteriological quality of roof surfaces was an indicator of the roof harvested rainwater making it a prime source of contamination of harvested water.

Hart and White [4] reported that the leaching of the chemical components of roofing sheets and containers was a prime reason for the presence of carcinogenic compounds in most harvested rainwater. Their study identified that storage tanks stationed at the point of collection correlated the presence of metals like Aluminum, Copper and Zinc in roof-harvested rainwater, contrary to their study, EPA [5] identified that the presence of organochlorides and polymers were primarily caused by leaching of organic compounds into the water.

In a related study, Sule *et al.* [1] suggested that the age, handling, and quality of storage tanks played a significant role in the bacteriological and chemical quality of harvested rainwater. Lack of potable water has also been identified as a key index of child labour as it causes these teaming children to trek long distances in search of water for most domestic activities [6] these increase the over-reliance on roof-harvested rainwater. Yufen *et al.* [7] observed that the level of ignorance of people to waterborne diseases was quite astronomical and the primary concern of most people having access to the water and less concern on the safety qualities; making water budget and needs a core challenge. Bacteriological and chemical contamination of potable water sources and its usage have been linked to an increased trend in the spate of disease burden that has plagued most developing countries like Nigeria today these buttresses the report of Gbadegesin and Olorunfemi [8] whose investigation identified that the lack safe drinking water sources was a major pointer for water-borne diseases.

Roof-harvested rainwater (RHRW) has remained a source of both potable and non-potable water supplies [9,10,11]. The harvest of rainwater conserves energy and harnesses availability of natural water as alternatives [13] which in turn may serve as climate change adaptation measures [12]. Although the benefits of the collection of rainwater abound in most communities and settlements, several persons have objectionable opinions of rainwater as a source of potable water either due to the sanitary quality or safety of the collection [14]. According to Evans [15], harvested rainwater can harbour chemical pollutants of concern and pathogens these also align with the report of Yufen *et al.* [7]. This study was aimed at evaluating the microbiological and physicochemical qualities of roof-trapped rainwater obtained from Hezekiah University, Umudi, Imo State, Nigeria.

1.2. Objectives of the study

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This study sought to meet the following objectives:

- a) Aseptically obtain the roof-harvested rainwater obtained from Hezekiah University, Umudi, Nigeria.
- b) Ascertain the physicochemical parameters of the roof-harvested rainwater obtained from Hezekiah University, Umudi, Nigeria.
- c) Determine the microbiological qualities of the roof-harvested rainwater obtained from Hezekiah University, Umudi, Nigeria.
- d) Identification of the bacterial isolates associated roof harvested rainwater obtained from Hezekiah University, Umudi, Nigeria.

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e) Determination of frequency of occurrence of the bacterial isolates associated roof harvested rainwater obtained from Hezekiah University, Umudi, Nigeria.

2.0. Materials and Methods

2.1. Collection of Roof trapped Rainwater samples at Hezekiah University, Umudi

Four roof-trapped rainwater samples were collected from aluminium roofs at the University canteen, hostel, administrative block and works departments of Hezekiah University, Umudi. Imo State. A clean-catch of the samples was aseptically obtained during a heavy downpour in August 2024. The samples were labelled and transported to the Laboratory Department of Microbiology.

2.2. Physicochemical analysis of the roof-trapped rainwater

The samples were transported to the Austino Laboratory in sampling and amber bottles with an ice block placed in the flask containing ice blocks to help keep the samples stable. The following parameters were determined using standard and analytical reagents

2.3. Microbiological evaluation of the roof-trapped rainwater

2.3.1. Determination of Total Heterotrophic Bacteria Count

Total heterotrophic bacterial counts for the different water samples were determined using the spread plate method on nutrient agar. The sample was serially diluted, vortexed and plated out using 0.1ml of the sample solution. The plates obtained after incubation were plated in duplicates and the average counts were obtained and converted mathematically. After 24 hours of incubation, colonies count between 30-300 then these values were expressed as Log₁₀CFU/ml [16].

2.3.2. Determination of Total Coliform Count

The Most Probable Number (MPN) was utilized to ascertain the faecal and total coliform composition of rainwater. The method comprises presumptive, completed and confirmatory tests as described by [17;18]. The preparation of lactose broth was done to amount to double and single strengths of the broth, 9.0 mL of the broth was dispensed into three tubes, while the 6 sets were prepared using 50% dilution of the broth for single strengths. The tubes were made to receive 10 ml, 1.0 ml and 0.1 ml of rainwater. Incubation was done at 37°C and 44.5 °C, for total and faecal coliform contents respectively. The production of gas and utilization of the lactose broth due to fermentation [16].

2.3.3. Determination of Total Salmonella-Shigella counts

The *Salmonella* sp. and *Shigella* sp. composition of rainwater obtained during the study, the water samples were enriched using Selenite -F broth for qualitative analysis. The rainwater samples were diluted and plated after 10-fold serial dilution was performed on the samples. The rainwater samples were plated using 0.1 mL of dilution and enrichment. The characteristic colony of isolates were counted for the isolates after the 37°C and 48hr duration.

3.0. Results

3.1. Physicochemical qualities of roof harvested rainwater

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The physicochemical qualities of the roof-harvested rainwater obtained from Hezekiah University facilities are presented in Table 3.1. The samples obtained from the female hostel as presented with the initials HRHRW was observed to have a temperature of 18° C while the samples obtained from the cafeteria were observed to have a temperature of 20° C. The electrical conductivity of the samples was $69.1~\mu$ S/cm for the samples obtained from the female hostel while the cafeteria had $110.56~\mu$ S/cm and the NAFDAC limits for potable water were observed to be $800-2500~\mu$ S/cm. The total dissolved solids for the samples were observed to be 8.53~and~11.97~mg/L. The total hardness of the water samples was observed to be 24.77~and~13.49~mg/L for the samples obtained from the female hostel and the cafeteria respectively while the NAFDAC/WHO limits were given to be 100-300~mg/L, Calcium hardness of the water samples was observed to be 8.0~mg/L for the rainwater samples obtained from the female hostel while the samples obtained from the cafeteria had 15.02~mg/L while the limits were identified to be 20-200~mg/L.

3.2. Coliform indices of the roof harvested rainwater

The coliform content of the roof-harvested rainwater was presented in Table 3.2 below; the 3-tube Most Probable Number technique utilized showed that the presumptive studies for the roof-harvested rainwater sourced from the hostel had a positive result for the single-strength tubes inoculated with 1.0 ml of the rainwater while the sample obtained from the cafeteria showed positive results for the three concentrations of the rainwater indication two (2) tubes were positive for double strength inoculated with 10 ml of the samples. Both samples proved to be positive for indicator organism *E. coli* by showing the presence of the green metallic sheen and a CFU/100 ml of 10 and 36 was reported for the samples obtained from the female hostel and cafeteria respectively.

3.3. Bacteriological qualities of the roof harvested rainwater

The bacteriological qualities such as the total heterotrophic bacterial count, *Staphylococcal* count and coliform counts of the roof harvested rainwater are presented in Figure 3.1 below. The total heterotrophic bacterial count of the rainwater sample obtained from the female hostel had a concentration of 5.0 Log₁₀ CFU/mL while the samples obtained from the cafeteria had a concentration of 4.7 Log₁₀ CFU/mL. The total *Staphylococcal* count for the samples obtained from the female hostel and cafeteria was 3.9 and 3.0 Log₁₀ CFU/mL respectively. The coliform count for the female hostel was 3.0 Log₁₀ CFU/mL while the cafeteria had a concentration of 3.3 Log₁₀ CFU/mL.

3.4. Colonial morphology of the bacterial isolates associated roof harvested rainwater

The colonial morphology of the bacterial isolates obtained from the study was presented in Table 3.3 below. The isolate KNRWA1 was observed to have a regular edge and raised elevation, the surface of the isolate was observed to have a smooth texture, the appearance was creamy while the size of the colony was moderate. KNRWA2 had a flat form of elevation, regular edge with a punctiform size as described below.

3.5. Biochemical characterisation of the bacterial isolates obtained from the roof-harvested rainwater

The biochemical identification of the bacterial isolates obtained during the study was presented in Table 3.4 below. The isolate KNRWA4 was observed to be a Gram-negative rod with a negative test for citrate, VP while showing a positive result for Indole, Methyl red, having an acid slant and butt with the production of both gas and hydrogen



sulphide and did not show a positive result for motility. The isolate KNRWA5 was observed to be motile, was positive for citrate was identified to be a Gram-negative organism, was able to ferment glucose and sucrose with the evolution of gas was identified to be *Pseudomonas* sp. While KNRWA1 had a characteristic or unique utilization of Simon citrate as a sole source of energy; it was identified for its reactivity for VP using the Barrit A and B as it turned positive brick red unique for the *Klebsiella* sp.

3.6. The frequency of occurrence of the bacterial isolates associated with the roof-harvested rainwater obtained from Hezekiah University

The result presented in Figure 3.2 describes the frequency of occurrence and distribution of bacterial isolates reported in the roof-harvested rainwater. The isolate *E. coli* was observed to occur 23.52% while *Enterococcus* sp. was observed to be 29.41% frequent during the study. Isolate such as *Bacillus* sp. was reported to occur 17.65% while the least frequent isolate was *Klebsiella* sp. with 11.65%.

Table 3.1. Physicochemical properties of roof harvested rainwater obtained from the Hezekiah University, Umudi

Parameters	H-RHRW	C-RHRW	NAFDAC/WHO limits for potable water
Temperature (°C)	18	20	20 – 32
pH	6.5	6.9	6.5-9.5
Electrical conductivity (µS/cm)	69.1	110.56	800-2500
TDS	8.53	11.97	50-150
Colour	8	6	5-35
Turbidity (NTU)	0.12	0.18	0.25
Total hardness (mg/L)	24.77	13.49	100-300
Calcium hardness (mg/L)	8	15.02	20-200
Chlorides (mg/L)	8.1	5.3	0.1-0.5
Total acidity (mg/L)	10.11	15	47-147
Total Alkalinity (mg/L)	20.4	31.9	30-50

H-RHRW= Hostel sourced- Roof trapped Rainwater; C= RHRW- Cafeteria sourced- Roof trapped Rainwater TDS- Total dissolved solids.

Table 3.2. Coliform composition of Roof Harvested Rainwater obtained from the Female Hostel and Cafeteria at Hezekiah University Umudi

Sample	3-DS 10 mL	3-SS 1.0 MI	3-SS 0.1mL	Confirmatory	Completed	CFU/100mL
H-RHRW	0	3	0	+	A/+	10
C-RHRW	2	3	1	+	A/+	36

Confirmatory test (+) = Green Metallic Sheent.

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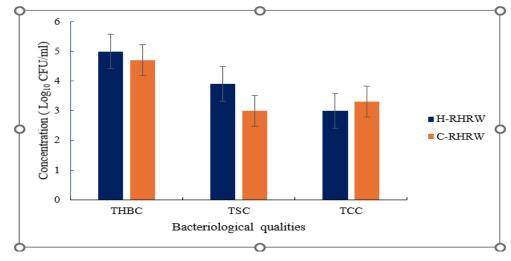


Figure 3.1. Bacteriological quality of roof harvested rainwater obtained from the Female Hostel and Cafeteria at Hezekiah University Umudi

Table 3.3. Colonial morphology of bacterial isolates obtained from the roof-harvested rainwater at Hezekiah University, Umudi

	Elevation	Edge	Texture	Pigment	Size
KNRWA1	Raised	Regular	Smooth	Creamy	Moderate
KNRWA2	Flat	Regular	Smooth	Creamy	Punctiform
KNRWA3	Raised	Regular	Smooth	Milky	Moderate
KNRWA4	Raised	Regular	Mucoid	Creamy	Moderate
KNRWA5	Raised	Irregular	Dry	Creamy	Punctiform
KNRWA6	Flat	Regular	Dry	Creamy	Moderate

Table 3.4. Biochemical characterization of bacterial isolates associated with roof-harvested rainwater obtained from Hezekiah University, Umudi

Biochemical	KNRWA1	KNRWA2	KNRWA3	KNRWA4	KNRWA5
Gram Reaction	-/Rod	-/cocci	+/Cocci	+/Rod	+/Rod
Oxidase	+	+	+	-	+
Catalase	+	+	-	-	+
TSI	K	A	K	A	A
Slant					
Butt	A	A	A	A	A
Gas	+	+	+	+	-
H_2S	+	+	+	+	+
MR	-	+	+	+	-



VP	+	-	-	-	+
Indole	-	-	+	+	+
Citrate	+	+	-	-	-
Motility	+	+	+	-	+
Glucose	A/G	A/G	A/G	A/-	A/G
Lactose	A/-	A/-	A/G	A/G	A/-
Mannitol	A/-	A/G	A/G	A/G	A/-
Sucrose	A/G	A/-	A/-	A/G	A/G
Tentative Identity	Klebsiella sp.	E. faecalis	Bacillus sp.	E. coli	Pseudomonas sp.

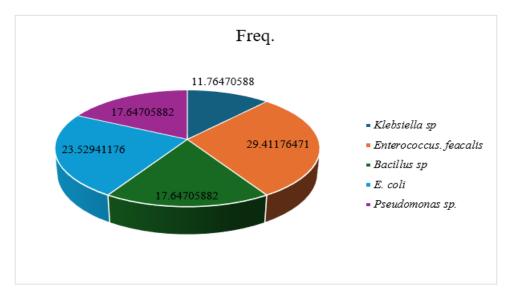


Figure 3.2. Frequency of occurrence of bacterial isolates associated with the roof-harvested rainwater from Hezekiah University, Umudi

4.0. Discussion of findings

The physicochemical composition of the roof-harvested rainwater (RHRW) correlates with the environmental condition of the area, and it translates to the well-being of the populace living within the geographical location. The pH of the RHRW obtained from the cafeteria/canteen was pH 6.5 and pH 6.8 while the NAFDAC/WHO limits for potable water is pH 6.5-9.5, the RHRW samples were within the regulatory limits. The electrical conductivity of the samples was 69.1 and 110.56 (µS/cm). The findings of the present study corroborates the earlier report of Ezemonye *et al.* [19] whose study reported a concentration value range between 5.0 to 6.5. In a similar vein, the pH concentration was also lower than the standards. The acidic pH of the samples was also identified to be contributed by the leaching of the roofing sheet and the precipitation of inorganic residues and particulates. In some extreme conditions, the acidophilic bacterial communities may resist the effect of the pH of rainwater. Electrical conductivity is a parameter that correlates with the pH of rainwater in any; this is because the aerosols and droplets may alter the pH concentration values. There are correlational studies to show that the electrical conductivity of water reflects the presence of cations and anions present in the water, these can be influenced by the leaching of



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materials into the water [16;1]. According to Dinrifo *et al.* [20] during the beginning of rainwater there is also a high proportion of pollutants. Prolonged deposition of aerosols and aerodynamics of the location during the dry season and the continuous contamination of surfaces were the primary challenges of the unwholesomeness status of rainwater. The work of Muhammad *et al.* [21] asserted that prolonged dry seasons, can be correlation with increased concentration of pollutants in the initial batch of harvested rainwater.

Total dissolved solids were observed to be 8.53 and 11.97 mg/L with a turbidity of 0.12 and 0.18 NTU both parameters were observed to be within the international and domestic standards for potable water which was 50-150 mg/L. The concentration of the dissolved solids was largely from clumps of particulate, faecal droppings from animals and debris. The presence of organic residues can impact the total dissolved solids of the water sample. There is a positive correlation between the total hardness of rainwater as this agrees with the findings of Amosah *et al.* [22] they were able to identify the chemical components that affect the concentration value of the total dissolved solids.

The bacteriological quality of the RHRW obtained from the study showed that the total heterotrophic bacterial count of the rainwater sample obtained from the female hostel had a concentration of 5.0 Log₁₀ CFU/mL while the samples obtained from the cafeteria had a concentration of 4.7 Log₁₀ CFU/mL. The total *Staphylococcal* count for the samples obtained from the female hostel and cafeteria was 3.9 and 3.0 Log CFU/mL respectively. The coliform count for the female hostel was 3.0 Log₁₀ CFU/mL while the cafeteria had a concentration of 3.3 Log₁₀ CFU/mL. These findings were in tandem with the report of Hamilton *et al.* [23] reported a microbial population of 2.0 x 10^7 CFU/mL for the total heterotrophic bacterial count.

The bacterial diversity associated with the RHRW obtained from Hezekiah University was dominated by coliforms such as *E. coli* and *Klebsiella* sp. These organisms have been implicated in some forms of gastroenteritis. The RHRW obtained from the canteen was laden with more coliforms; the major concerns lie in the potential utilization of such water in essential domestic activities such as cooking and washing vegetables or dishes which can eventually lead to cross-contamination. The report of Shergill and Pitt [24] agreed very strongly when they observed that out of 246 samples, average total coliforms (57.7%), *E. coli* (30.5%), and total aerobic bacteria (100%) were detected after rainfall. The prevalence of coliforms, *Enterococcus* and *Pseudomonas* from all sites was not significantly different before and after rainfall. Their findings also corroborate the observation of the current study as total coliforms were more frequent in the study.

5.0. Conclusion

Roof-harvested rainwater has remained a major water source for staff and students who reside in Umudi, Nkwerre, and Imo state. The geolocation of this first private University in Imo State is plagued with an insufficient water supply to the teeming populace that makes up the University community. The RHRW has remained a major source of potable water resources for students and staff. The microbial quality and the physicochemical indices have shown that the resource is tainted with both biological and chemical pollutants, which may have harmful effects on the patron of this resource. The diversity of bacterial pathogens that were identified namely *Klebsiella* sp., *E. coli*, *Enterococcus* sp. and *Bacillus* sp. was indicative of possible pollution.



6.0. Recommendations

The findings of the study quite succinctly arrived at the following recommendations:

Hezekiah University community must be educated on the dangers associated with the continued use of roof-harvested rainwater for domestic purposes due to the possibility of cross-contamination of food and food products and the cafeteria must be sensitized on the dangers of using RHRW as an alternative resource for potable water as it fell below recommended international and domestic standards and limits for drinking water.

A community-wide sensitisation will be necessary on the need to boil or treat RHRW before use in the event of extreme water scarcity. This will help reduce incidences of food-borne diseases, which may be prevalent in rural communities like Umudi.

The university can be encouraged to sponsor source-tracking studies to identify the possible sources of coliforms in RHRW, especially in the cafeteria section of the school, and identify the possible epidemiological concern that may have gone unreported among the students who patronise the University canteen/cafeteria.

7.0. Contributions to Knowledge

This study has beamed and added to the wealth of knowledge on the presence of indicator organisms of faecal pollution, namely *Escherichia coli* and *Enterococcus* sp., in roof-harvested rainwater collected from the female hostel and the university cafeteria/canteen of Hezekiah University. The bacteriological quality of RHRW obtained from Hezekiah University was severely tainted with bacteria of possible health concerns. The heterotrophic bacterial count of over 1.0×10^5 CFU/ml and a coliform quality of 10 and 36 CFU/100 ml suggests poor bacteriological quality and objectionable standards for potable water.

Declarations

Source of Funding

This study did not receive any grant from funding agencies in the public, commercial, or not-for-profit sectors.

Competing Interests Statement

The authors declare no competing financial, professional, or personal interests.

Consent for publication

The authors declare that they consented to the publication of this study.

Authors' contributions

Both the authors made an equal contribution to the conception and design of the work, data collection, experimental analysis, writing of the article and critical revision of the article. Both the authors have read and approved the final copy of the manuscript.

Availability of data and material

Authors are willing to share the data and materials according to relevant needs.



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