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Original article

PHYSICO-CHEMICAL AND BACTERIOLOGICAL QUALITY OF WATER FROM WATER SOURCES AND HOUSEHOLD DRINKING WATER AT NAKIVALE REFUGEE BASE CAMP, SOUTH-WESTERN UGANDA, A DESCRIPTIVE CROSS-SECTIONAL STUDY.

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ABSTRACT.

Introduction:
Safe drinking water and sanitation are indispensable to sustain life and health, adequate, safe, and accessible water supply are fundamental to the dignity of all. This study determined the physico-chemical and bacteriological quality of water supplied to the fragile community in Nakivale Refugee Base Camp, Southwestern Uganda.

Methodology:
Physico-chemical properties were measured in situ using Hanna Instrument HI 98129. Multiple tube technique was employed to determine the presumptive coliform counts. Positive samples for coliforms were incubated at 44oC for 48 hours in Single Strength MacConkey broth. Only positive samples were followed by conventional biochemical tests of indole, motility, and citrate to identify *E. coli*.

Results:
The mean temperature, Total dissolved solids, pH, and electrical conductivity for both the water sources and household water samples were 23.79±1.04oC and 25.25±0.38, 289.07±153.45 and 433.31±519.13, 7.81±0.31 and 6.65±0.35 and 547.8±315.73 and 768.52±397.71 respectively. Among the water sources sampled, 43% (13) were contaminated with total coliform bacteria, and 10% (3) were positive for *E. coli*. 90% of household samples were contaminated with coliform bacteria with 11% positivity for *E. coli*. 61% (103) of the participants did not use water treatment. 19% (32) used boiling and 19% (32) used chlorination methods while 1% used filtration.

Conclusion:
Physico-chemically, all water samples had temperatures above the WHO guidelines for palatable water i.e. <15oC. Bacteriologically, Water samples were contaminated and not fit for human consumption hence the need for effective water treatment and proper post-treatment practices.

Recommendation:
Nakivale refugee base camp occupants need continuous training on water treatment methods, safe water handling practices, and proper handling of fecal waste to minimize fecal water contamination. Further studies should be conducted to assess the effect of storage of clean vessels as an intervention on the bacteriological quality of water.

Keywords:

Physicochemical, bacteriological, quality, water sources, household drinking water, descriptive, cross-sectional.

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INTRODUCTION.

Water is the essence of life, safe drinking water and sanitation are indispensable to sustain life and health, and this is fundamental to the dignity of all humans (WHO, 2019a). A satisfactory (adequate, safe, and accessible) water supply must be available to all since improving access to safe drinking water results in tangible health benefits therefore every effort should be made to provide drinking water that is as safe as practicable (WHO, 2017). In their 2002 summit, the United Nations Committee on Economic, Social and Cultural Rights resolved on the water right, as a fundamental right to every individual "to safe, adequate, clean, physically accessible and available water for use" (Palmer et al., 2018).

The water required for personal or domestic use must be safe, thus free from microorganisms, chemical substances, and radiological hazards that constitute a threat to a person's health (UN, 2015). WHO/UNICEF Joint Monitoring Program on Water Supply, Sanitation and Hygiene (JMP) reported that, as of 2017, 2.2 billion people lacked safely managed water and 4.2 billion people lacked safely managed sanitation (UNICEF, 2019). Globally, approximately 884 million people lack access to basic drinking water, and even their few available water sources are deemed contaminated (Hernandez, 2020). About 88% of diarrhoea-associated deaths are attributable to unsafe water, inadequate sanitation, and insufficient hygiene and diarrhoea accounts for 1 in 9 child deaths worldwide, making it the second leading cause of death among children under the age of 5 years (CDC, 2015). Together, unclean water and poor sanitation are the world's second biggest killers of children as postulated by the (UN, 2015). 42% of people in rural sub-Saharan Africa lack basic water supply while 72% share their domestic water sources with animals or rely on unprotected water sources that suffer from the accumulation of surface run-offs from uplands and are thus breeding grounds for pathogens (Kumpel et al., 2016).

The majority of rural communities in Uganda lack access to improved water sources and the few available water facilities are often non-functional (Mulogo et al., 2018). Refugees in Uganda have only 38% access to safe drinking water, 27% (5,232 m3/day) of which is supplied through costly and inconsistent trucking (UNHCR, 2017). Nakivale Refugee Camp, one of the oldest refugee settlements in Uganda, was opened in 1958 and officially established as a settlement in 1960 (O'Laughlin et al., 2014). The settlement hosts more than 100,000 refugees from Burundi, The Democratic Republic of Congo, Eritrea, Rwanda, Somalia, Sudan, and South Sudan (Meyer et al., 2017) and the population of the settlement greatly increased during the Burundian crisis of 2015 (Bapolisi et al., 2020). One of the greatest challenges faced by occupants of this camp is limited access to water and the questionable quality of a few existing water sources and water supplied by tucking and this is associated with the high incidence of reported diarrhoeal cases (UNHCR, 2018).

The demand for water in Nakivale Refugee Base Camp is fulfilled by ground and surface water sources (Ivanova et al., 2019). Greater than 80% of this demand is met by water channeled from Lake Nakivale and is supplied at 57 unprotected water points that are susceptible to likely contamination as a result of poor human waste management, irregular distributions of soap to households, lack of hand-washing facilities at key locations, pollution after heavy rains and sharing of the sources with domestic animals. Only 4% (64.3m3/day) of the water that refugees in Nakivale Refugee Base Camp have access to is supplied via water trucking (UNHCR, 2017).

The physicochemical quality of water affects its acceptability to consumers (Sheibani and Mohammadi, 2018). Turbidity, color, taste, and odor, whether of natural or other origin affect consumers' perceptions and behaviors. Turbidity over 5 nephelometric turbidity units is objectionable, color is indicative of organic matter, and odor is due to mainly the presence of organic substances and an indication of increased biological activity in the water sources (Soros et al., 2019).

Bacteriologically, drinking water should not contain microorganisms known to be pathogenic or any bacteria indicative of fecal pollution (Ferrer et al., 2020). The detection of *E. coli* provides definite evidence of fecal contamination; in practice, the detection of thermotolerant (fecal) coliform bacteria is an acceptable alternative (Foster et al., 2019). All water intended for drinking, including treated water for distribution, should have no *E. Coli* or thermotolerant coliform bacteria detectable in any 100ml samples (WHO, 2019b). WHO 2019, a guide to equitable water safety planning provides for safe and accessible drinking water as an effective pathway to health promotion and poverty reduction as safe drinking water and hygienic toilets protect people from disease and enable societies to be more productive economically (WHO, 2019b). National government should therefore work towards achieving the Sustainable Development Goals (SDG 6) which aims at ensuring available and sustainable management of water and sanitation for all by 2030 (Park & Ph, 2018) to achieve this target, active monitoring of microbial water quality of drinking water through the enumeration of *Escherichia coli* in water samples is inevitable (Verbyla et al., 2019). This study determined the physical-chemical and bacteriological quality of water sources and household drinking water at Nakivale refugee base camp, Isingiro district, South-western Uganda

MATERIAL AND METHODS.

Study area.

This study was conducted in Nakivale Refugee Camp located in the Isingiro district in South-western Uganda and covering a total area of 185km2, approximately 42 kilometers from Mbarara with a population of about 61,000 people by 2019 (Ivanova et al., 2019). The settlement camp is divided into three zones; Juru zone, Rubondo zone, and Base camp zone which has the highest concentration of the refugee populous (Nara et al., 2020).

Sample size determination.

Using the formula (Kish, 1965).

$$n = \frac{z2\ p(1-p)}{d2}$$

Where; n=Sample size

P=Estimated prevalence of water sources that meet the National Standards and WHO Guidelines for Drinking Water (29%)

d= Precision error (5%)

Z= Z statistic for a level of confidence (95%) which is 1.96

With an estimated prevalence of 29% according to the Cross-sectional descriptive study that was carried out in South-western Uganda (Apecu *et al*., 2019).

n=316.394176. This is approximately 316 samples.

Sampling Strategy.

Water sources were sampled from 30 water points within Nakivale Refugee Settlement- Base Camp. The study employed the systematic sampling technique whereby a sample was taken from every second home and a total of 168 household water samples were collected. A structured questionnaire was used to collect information on socio-demographic characteristics and certain water sanitation and hygiene factors that influenced water quality. The questionnaires were developed, translated into the local language, and pretested before administration by trained research assistants to the head of the individual household who participated in the study. 300mls of water samples were collected from each of the 168 selected households and 30 from the drinking water sources making a total of 198 water samples. Water samples were aseptically collected into sterile sample bottles and immediately placed in a cool box containing ice packs and transported to Mbarara University of Science and Technology Department of Microbiology for microbiological analysis within 6 hours of sample collection.

Measurements.

Physico-chemical examination.

The physical and chemical parameters of temperature, pH, electrical conductivity, and total dissolved salts were measured *in situ*, at the time of sample collection to avoid changes likely to occur during transportation and storage. Using a pH/electrical conductivity (EC)/total dissolved solids (TDS)/temperature meter (Hanna Instrument HI 98129) that was calibrated with pH 4.01 and 7.01 standard buffer solutions according to the manufacturer's instructions.

Bacteriological examination.

The bacteriological aspects of the water samples were measured using the Most Probable Number (MPN) Test. This method was used to estimate the concentration of viable microorganisms using replicating liquid broth growth in tenfold dilutions and the results were reported as the Presumptive Coliform Count concerning McCrady's probability table. All the positive presumptive test bottles were inoculated into sterile MacConkey broth containing Durham tubes and incubated at 440C for 48 hours. Gas and acid production confirmed the presence of thermotolerant bacteria (*E. coli*). The positive samples for Eijkman's test were sub-cultured on MacConkey agar for 24 hours at 370C and biochemical tests of Indole, Citrate, and Motility were done to confirm *E. coli* which was positive for Indole and Motile but negative for Citrate utilization test. For the MPN technique, single and double-strength MacConkey broth media, sterilized by autoclaving at 15 psi pressure (1210C) for 15 minutes were used and since the water was assumed to be treated, 50ml of double-strength MacConkey broth was dispensed in one bottle, 10ml of double strength medium in each of the 5 universal bottles and 1ml of single strength medium in each of the 5 universal bottles and properly added sterile Durham tube in an inverted position to ensure it was filled with the broth with no air bubbles inside the tubes. One bottle of double strength (50ml), double strength (10ml), and single strength (1ml) was used for each water sample tested, and an equal volume of water to the MacConkey broth was added to the respective water culture bottle aseptically using sterile pipettes, positive controls, and negative controls were treated equally as water sample to validate the tests and this was followed by 48hr incubation at 37oC. Where there was growth, the number of positive bottles indicated by airspaces inside Durham's tube were recorded and compared with the standard probability table according to McCrady to get the presumptive coliform count of a water sample. Sulphide Indole Motility (SIM) test was done by a central stab of the test organism isolated on MacConkey agar after subculture of positive water culture bottles and incubation for 24 hours at 37◦C to determine the ability of the organism to utilize an amino acid tryptophan, *E. coli* possesses tryptophanase enzyme and thus hydrolyze and deaminate tryptophan to Indole, pyruvic acid, and ammonia (Romasi and Lee, 2013). The test interpretation was by checking for bacterial motility depicted by hazy or feather-like growth away from the central stab line, while indole production was checked by the addition of ten drops of Kovac's reagent down the side of the tubes after the incubation period where cherry red/bright red color development upon addition of the reagent within seconds was a positive test, the case with *E.coli*. Citrate utilization was done by streaking Simmons citrate agar medium with the bacterial isolate followed by 24hr incubation at 37oC, a negative test shown by retention of the green color of the medium was the case for *E coli* (Mitra et al., 2020).

Data analysis.

All statistical analysis was done using STATA version 16.0. Data was entered in Excel and then exported to Stata. Descriptive statistics of the study subjects or household characteristics and WASH factors were reported as proportions, mean, and range. We managed to analyze the data in the form of frequencies and percentages.

Quality control.

The training was organized by the team for all research assistants a week before the commencement of the study. The questionnaires were pretested in households closer to the refugee camp and necessary corrections were made accordingly by the principal investigator. Data was checked for consistency and completeness before the closer of every day of data collection. Water samples were collected in a sterile sample bottle, stored in a cool box containing ice packs, and processed within 6 hours of collection. *Escherichia coli-infused* water was used as a positive control and 100mL of sterile distilled water was used as a negative control after every ten water samples during analysis. The standard method for testing the drinking water quality was maintained as per World Health Organization guidelines (WHO, 2019a).

Ethical considerations.

The study's Principle Investigator sought permission from the office of the Prime Minister of the Republic of Uganda after approval by the Faculty Research Committee of Mbarara University of Science and Technology. The study gained informed consent from the study participants before they participated in this study.

RESULTS.

Descriptive characteristics.

A total of 198 water samples were analyzed, 30 water samples were from water sources, and 168 were from households. Descriptive characteristics for households are summarized in Table 1.

Table 1; Shows descriptive statistics of household heads and water treatment characteristics.

Variable	Freq.	%
Sex		
Female	108	64
Male	60	36
Age in Years		
20-29	104	62
30-39	53	32
40 and above	11	6
Education background		
Primary	29	17
Ordinary level	53	32
Advanced level	35	21
Higher Institution	14	8
None	37	22
Water Treatment		
No	104	62
Yes	64	28
Water Treatment method		
Boiling	32	19.
Filtration	1	1
Chlorination	32	19
None	103	61
Water Sources		
Improved	26	87
Unimproved	4	13

Physico-chemical properties.

The temperature of water sources ranged between 21.6ºC to 25.7ºC with a mean of 23.79±1.04ºC, their pH ranged from 7.1 to 8.3 with a mean of 7.81±0.31, Total Dissolved Solids ranged from 121ppm to 666 ppm with a mean of 289.07±153.45 and Electrical conductivity ranged from 227 µs/cm to 1330 µs/cm with a mean of 547.8±315.73.

Among the households water samples, the temperature ranged between 23ºC to 25.9ºC with a mean of 25.25±0.38, the pH ranged from 5.52 to 8.37 with a mean of 6.65±0.35, Total Dissolved Solids ranged from 27ppm to 4833 ppm with a mean of 433.31±519.13 and Electrical conductivity ranged from 54µs/cm to 3999µs/cm with a mean of 768.52±397.71.

Table 2 Showing Physico-chemical properties measured for Water sources and households.

Mean of physio-chemical properties for water sources and household water samples

	Ph	Temp. (^{0}C)	Total Dissolved Solids (TDS) (ppm)	Electrical Conductivity (EC) (µs/cm)
Water Sources	7.81±0.31	23.79±1.04	289.07±153.45	547.8±315.73
House Holds	6.65±0.35	25.25±0.38	433.31±519.13	768.52±397.71

Faecal Contamination in water samples.

21 of the 198 samples (11%) were positive for ***E. coli*** as shown in figure 1.

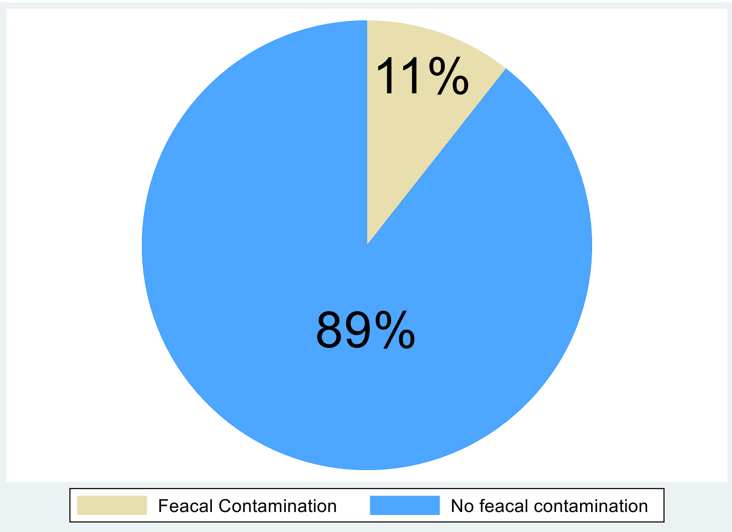

Figure 1 Pie chart showing faecal contamination in the water samples.

Faecal Contamination in water samples from water sources and households.

Bacteriologically, 3 of the 30 samples from water sources (10%) were positive for *E. coli* indicative of faecal contamination, 43% (13) of water sources had total coliform count >10cfu/ml of a water sample and 90% of the 168 household water samples were contaminated with coliform bacteria with 11% (18) contamination with *E. coli.* Generally, 82.8% of the water samples studied had total coliform count >10cfu/ml as required by the World Health Organization and 10.6% of the water samples had >0cfu/100ml of E. coli count (WHO, 2019b) and the results are summarized in the table 3.

Table 3 Showing faecal contamination in water sources and household water samples.

Water samples	Coliform Contamination	Fecal Contamination
Water sources	13 (43%)	3 (10%)
Household	151 (90%)	18 (11%)
Overall	164 (82.8%)	21 (10.6%)

Common Water treatment methods.

Water treatment methods common in Nakivale refugee Base camp included boiling, chlorination and to a lesser extent filtration. The graph below **(*Figure 2*)** indicates the percentage of use according to the questionnaire results on method of treatment from 168 households.

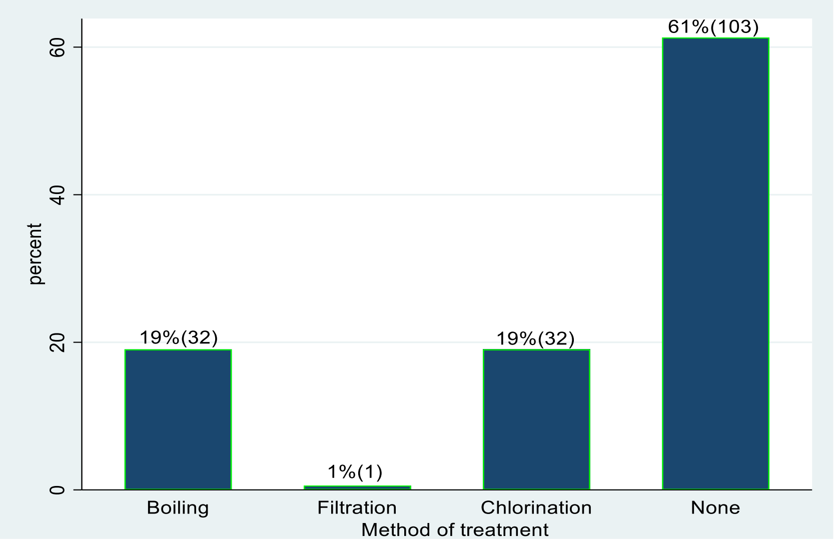

Figure *2* Bar graph showing methods of treatment used in Nakivale Refugee Base Camp.

DISCUSSION

This study found that temperature ranged from 21.6 to 25.7oC and 23.0 to 25.9oC for water sources and household water respectively. The mean temperature of water samples from both the water sources (23.79±1.04) and household water (25.25±0.38) were above the WHO recommended temperature for palatable water; <15oC (WHO, 2019b). The rise in water temperature is attributable to global warming as reported by (Stocker, 2015) the global land mean surface temperature rise was 0.85°C from 1850 - 2012 (Lin and Franzke, 2015), and locally in Uganda, UNAPA reported an average temperature increase of 0.28°C per decade between 1960 and 2010 and this culminates in the rise of the surface water temperature (Hisali et al., 2011). This result was consistent with the study done in Jimma Zone Southern Ethiopia by Yasin et al., 2015 which reported that water temperature ranged between 20.67 and 25.73oC (Yasin et al., 2015) and also, a similar study in Mbarara municipality by Lukubye and Andama, 2017 reported that water temperature ranged from 21.3 and 26.4oC (Lukubye and Andama, 2017). The higher mean temperature of household water could be attributed to water storage conditions due to the low socioeconomic status of the participants making it difficult to access good water storage conditions (Abdo et al., 2019).

The total dissolved solids for water sources ranged from 121ppm to 666ppm with a mean of 289.07±153.45 and from 27ppm to 4833ppm with a mean of 433.31±519.13 for household water samples. The mean TDS for both water sources and household water were within the WHO acceptable limits; <1000ppm according to the (WHO, 2019b). Similar findings were reported in Mbarara municipality where by TDS of the studied water sources were below the WHO acceptable limit for palatable water with the mean TDs of 297.70±32.74 (Lukubye and Andama, 2017). The mean pH of water sources was 7.81±0.31 and 6.65±0.35 for household water samples and these were within the WHO guidelines for drinking water quality (WHO, 2019b). Similarly, the mean pH range for water sources and household were 7.81±0.31 and 6.65±0.35 respectively, and the mean electrical conductivity of water sources and household water were also 289.07±153.45ppm and 433.31±519.13ppm. these were in line with the WHO guidelines for palatable water with a pH range of 6.5 to 8.5 and electrical conductivity of <1500ppm (WHO, 2019b).

The study revealed that 43% of the water sources were contaminated with Coliform bacteria and 10% were faecally contaminated. According to WHO standards, the recommended Coliform count in water is <10 CFU/100mls therefore 43% of the water sources were not portable and 57% meet the WHO guidelines for drinking water quality (WHO, 2019b). The presence of thermotolerant coliform indicated fecal contamination of water sources and this was attributed to improper waste management particularly human and animal excreta which subsequently contaminate water sources when carried by surface run-off following precipitation (Nyoka et al., 2017). This study finding however was higher than the 29% coliform water contamination revealed by a study on water sources in Uganda by (Apecu *et al*., 2019). Analysis of household drinking water samples established that 83% were contaminated with coliform bacteria and 11% were faecally contaminated. This makes the water unpalatable as per WHO standards (WHO, 2019b). This high water household water contamination potential was attributed to inadequate water sanitation and hygiene inside and around water storage taps or facilities (Nyoka et al., 2017). Furthermore, a greater percentage of household water users (61%) do not treat their water and thus contamination that originates from the primary water source, transportation, and possibly at the storage site of the final water user accounts for higher bacterial contaminations (Agensi et al., 2019, Sharma and Bhattacharya, 2017). This study had similar findings with a study on the contamination potential of household water handlings and storage practices by (Agensi et al., 2019) which reported that 43.2% of samples studied from unprotected water sources had total coliforms counts >10cfu/100ml and 34.1% had *Escherichia coli* counts >0cfu/100ml of the water samples, the study further reported that household drinking water had 25% total coliforms counts >10cfu/100ml and 8.7% had *Escherichia coli* counts >0cfu/100ml (Agensi et al., 2019) as stipulated by the WHO guides for equitable safe water planning (WHO, 2019b).

The study also revealed that more than half of the participants (61%) do not practice water treatment measures and utilize the raw water as supplied and of those that practice water treatments, the majority (49.23%) employed chlorination and boiling as water treatment methods and lesser percentage (1.54%) employed filtration method.

CONCLUSION.

Physico-chemically, all water samples had temperatures above the WHO guidelines for palatable water: <15oC (WHO, 2019b). However, the mean total dissolved solids, electrical conductivity, and pH were within their respective ranges for the WHO guidelines for safe water for utilization as postulated in the guide to equitable safe water planning (WHO, 2019b). Bacteriologically, the Water supply in the Nakivale refugee base camp was contaminated and not fit for human consumption without effective water treatment and proper post-treatment practices. Also, the detection of E. coli in the water samples was an indicator of fecal water pollution (Kirschner et al., 2017).

RECOMMENDATIONS.

Water supply and treatment agencies in refugee base camps should improve water treatment as the majority of the refugee population does not treat water before use due to various reasons such as the demands for water and socioeconomic status. Where possible, the refugees need to be continuously trained and supported on water treatment methods and practices to improve the effectiveness of the already existing water treatment methods. There is a need for continuous assessment of the quality of both water sources and household drinking water.

Further research can be done on the storage and appropriate water handling practices as these may be responsible for the problem of water contamination.

GENERALIZABILITY OF THE STUDY FINDINGS.

This study was conducted in a refugee base settlement and the results may apply elsewhere where the water supply sources are similar even in non-refugee base camps with similar socioeconomic and sociodemographic factors like economic and social class of the society.

LIMITATIONS

There was delayed access to the study site due to the lockdown.

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DATA SETS.

The data set for this study is available on request from the corresponding author

LIST OF ABBREVIATIONS AND ACRONYMS.

CFU: Colony Forming Units
DRC: Democratic Republic of Congo
E. coli: Escherichia Coli
EC: Electrical conductivity
FRC: Faculty Research Committee
HI: Hanna Instrument
JMP: Joint Monitoring Program
MLS: Medical Laboratory Science
MPN: Most Probable Number
MRRH: Mbarara Regional Referral Hospital
MUST: Mbarara University of Science and Technology
MWE: Ministry of Water and Environment
NTU: Nephelometric Turbidity Units
NWSC: National Water and Sewerage Corporation
ppm: Parts per million
PI: Principal investigator
SDG: Sustainable Development Goals
SIM: Sulphide Indole Motility
TDS: Total Dissolved Solids
UDHR: Universal Declaration of Human Rights
UDHS: Uganda Demographic and Health Survey
UN: United Nations
UNAPA: Uganda National Adaptation Program of Action
UNHCR: United Nations High Commissioner for Refugees
UNICEF: United Nations Children's Fund
WASH: Water Sanitation and Hygiene
WHO: World Health Organisation

OPERATIONAL DEFINITIONS

Coliform bacteria: Defined as rod-shaped gram-negative non-spore forming and motile or non-motile bacteria that can ferment lactose with the production of acid and gas when incubated at 35-37OC.
Improved water sources: Includes water sources that by nature of their construction or through active intervention are unprotected from outside contamination, particularly fecal contamination.
Unimproved water sources: Include water sources that by nature are unprotected from outside contamination.
Thermotolerant coliforms: The group of coliform bacteria that produce gas from lactose when incubated at 44.5OC for 48 hours.
Eijkman's test: This is a test used for the identification of coliform bacteria from warm-blooded animals based on the bacteria's ability to produce gas when grown in glucose media at 46OC.
Portable water: Also known as drinking water is water that is safe to drink or use for food preparation.
Contamination: Any impairment of the quality of water in the state by sewerage or industrial waste to a degree that creates an actual hazard to public health through poisoning or spread of disease.

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