

# Analysis of Bacteriological Quality of Domestic Water Sources in Kabale Municipality, Western Uganda

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## Abstract

In the present study, we assessed the bacteriological quality of water of drinking water sources in Kabale Municipality. A total of 28 water samples were collected from 14 water springs during the dry and wet season and analyzed for determination of Escherichia coli (E. coli), and Total Heterotrophic Bacteria (THB) using the membrane filtration method. Most water springs located in less than 20 meters away from residential areas were associated with bacterial contamination. The mean concentrations of E. coli, total coliforms, and THB were 24.07 CFU/100 ml, 85.71 CFU/100 ml, and 197.07 CFU/100 ml, respectively in the wet season. While in the dry season, the mean concentrations were 2 CFU/100 ml, 10 CFU/100 ml, and 91 CFU/100 ml for E. coli, total coliforms, and THB, respectively. There were significant differences between CFU of total coliforms, HTB, between wet and dry seasons (p = 0.026). Samples collected and analyzed during the wet season for total coliforms did not conform to WHO drinking water quality guideline value of no detection per 100 ml. The study concludes that the majority of spring water sources are located in less than 20 meters away from residential areas with significant paved areas, the presence of septic tanks and pit latrines. Wet season significantly affects the quality of domestic water sources than the dry season. The study recommends that spring water should be treated before drinking due to high bacteria concentrations which makes it unsafe for consumption.

## **Keywords**

Water Quality, Drinking Water Sources, Total Coliform, E. coli

## **1. Introduction**

Worldwide, water is an important component of living beings as it performs

unique and indispensable activities [1]. However, due to many anthropogenic activities, freshwater resources are getting deteriorated at a faster rate. Increasing urban population growth, industrialization, and intensive farming have led to severe disturbance of drinking water across the world, resulting in limited access to clean and safe drinking water. Access to clean and safe drinking water is vital for a healthy life and human sustainability.

Globally, estimates indicate that more than 700 million people lack access to a safe drinking water source [2]. In Africa, Sub-Saharan Africa is most remarkably and disproportionately affected by lack of access to safe drinking water. Estimates show that 327 million people without access to safe drinking water live in Sub-Saharan Africa [2] [3]. Absurdly, people still drink unsafe water in spite of improved access to safe drinking water. Drinking water can be contaminated at the source, in the distribution system, during collection, transportation, and storage.

In Uganda, access to safe drinking water sources was approximately 76% in 2015. This is so because since the 1990s efforts were made to provide safe drinking water to the people through National Water and Sewerage Corporation (NWSC), and gravity flow scheme projects (Saturday *et al.* 2016). However, the burden of waterborne diseases remains a major challenge [4]. A study by [5] reported that 18.8% of children under the age of five in southwestern Uganda had diarrhea disease before 2012 demographic health survey. The report further indicated the child mortality rate at 90 deaths per 1000 live births. Most of these deaths result from diarrhea disease due to the consumption of unsafe drinking water.

In Kable Municipality, while more than 82% of inhabitants have access to treated pipe-borne water for drinking and domestic use, a reasonable proportion of people especially in slum areas rely on water springs as their main source of water for drinking and domestic use [5]. Unless water springs are protected for human consumption, its water may become hazardous human health and transmit some waterborne diseases. The main sources of contaminants in these water sources in urban areas are pit latrines, wastewater from residential areas, animal waste and effluent from sewage system [6]. Thus, people use water from doubtful sources, which put them at risk of waterborne illnesses.

Analysis of the bacteriological quality of drinking waters is important in determining the sanitary quality of domestic water sources. Indicator bacteria are used to evaluate the quality of drinking water because it would be next to impossible to accurately enumerate all pathogenic organisms that transmitted through contaminated water [7]. The use of indicator bacteria as the means of assessing the quality of drinking water has been of paramount importance in protecting the lives of water against waterborne diseases. The principle here is that the selected bacteria indicate either contamination or deteriorating water quality upon which protection of public health from waterborne diseases has been developed. The study therefore aimed at ascertaining the bacteriological quality of drinking water sources with particular emphasis on spring water sources of Kabale Municipality.

## 2. Materials and Methods

## 2.1. Location of the Study

The study was conducted in Kabale Municipality (**Figure 1**) located in Kabale District, Southwestern Uganda, 406.78 km from Uganda's capital city, Kampala. Geographically, Kabale Municipality lies between longitudes 29°45'0"E and 30°15'0"E and latitudes 0°1'0"S and 1°29'0"S. It covers a total area of 33 km<sup>2</sup> [8]. It borders with the sub-counties of Kitumba in the south, Bubare to the north and Kyanamira to the East [9]. The fairly densely populated Kabale municipality of about 49,667 people (UBOS, 2014) is divided into Kabale North, Kabale South, and Kabale Central divisions, and 82% of households are said to have access to improved drinking water sources [10]. The main sources of drinking water are treated pipe-borne water, and water springs. However, some municipal inhabitants practice rainwater harvesting practiced [10]. In spite of relatively high accessibility to improved water sources in outskirts of the Kabale Municipality. As a result, people greatly exert pressure on springs which are free of charge. Hence 14 water springs were randomly selected from the three divisions

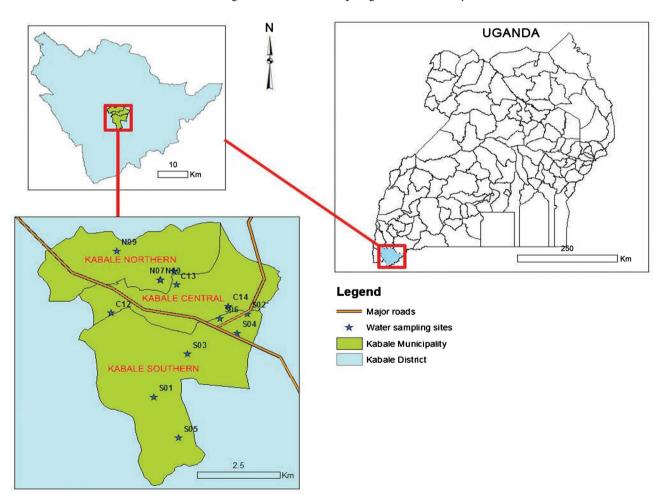


Figure 1. Map of Kabale municipality showing sampling locations.

that make up Kabale Municipality viz; 5 springs from Kabale Northern Division, 5 from Kabale Southern Division, and 4 from Kabale Central Division.

#### 2.2. Sample Collection and Preparation

A total of 28 spring water samples were collected from 14 water springs during dry and wet seasons. Samples were collected aseptically in sterilized 500 ml sterile plastic bottles and then transported to National Waster Sewage Cooperation water quality laboratory for analysis. Before the transportation of water samples, the sampling bottle corks were shielded with aluminum foil in order to avoid any form of hand contamination and adhere to aseptic techniques. All samples were assigned identification numbers and the location of the source. Samples were analyzed as soon as possible after collection to avoid unpredictable changes in the bacterial population.

#### 2.3. Bacteriological Analysis

Membrane filtration method which involves direct plating for detection and estimation of coliform was used in accordance with [11]. This method of analysis involved filtering 100 ml of water sample through a 0.45  $\mu$ m millipore filter membrane that retained total coliform (TC) and *Escherichia coli*. For, heterotrophic bacteria count; 1 ml of sample was filtered following procedures as for total coliforms. Funnel unit was placed carefully over the filter support assembly and was locked in place. Samples were poured into the funnel assembly then the filtered through the membrane-filter by applying a vacuum pump. Finally, the prepared culture dishes were incubated for 24 hours at 37°C, 24 hours at 44°C, and 48 hours at 37°C for determination of total coliforms *E. coli* and THB count, respectively. Upon completion of the incubation period, typical coliform colonies were seen on the membrane filter. All the colonies were counted with the aid of a magnifying lens [11].

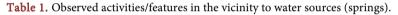
## 3. Results and Discussion

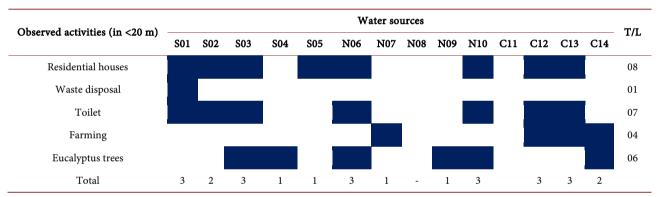
## **3.1. Characterization of Drinking Water Sources**

The potential threats and pathways of contamination of protected water springs were identified based on observed human activities in close proximity to water sources. This was accomplished during the field study visit to 14 water springs in Kabale Municipality. The study findings revealed that 8 (57.1%), 7 (50%), 4 (28.5%), and 6 (42.8%) of water springs sampled were situated in less than 20 m away from residential areas, toilet/ pit latrines, farming and eucalyptus tree plantations, respectively (**Table 1**).

Construction of toilets/pit latrines in close proximity to drinking water puts it at risk of feacal contamination. A study by [12] reported that River Cauvery in India was loaded with coliform bacteria attributable to raw sewage. In a related study, high concentration microbes in drinking water sources located in close proximity to pit latrines (<20 m) were reported [13]. Additionally, construction of residential houses in proximity or upstream to drinking water sources poses a threat to water contamination through leaching of contaminants. As shown in **Plate 1**, a water spring was found constructed in <5 m adjacent to the residential area. It further observed that wastewater was flowing from the residential area into the water spring area. For instance, direct deposition of feces into the water body increases the survival rate of pathogens due to rapid sorption of contaminants onto bed sediments [14]. A study by [15] reported that animal manure and feces are key important sources of fecal contamination of drinking water sources in the agricultural watershed.

Farming was also another observed activity around water springs around that were studied (**Table 1**). Crops such as beans, potatoes, vegetables, and bananas were planted in close proximity to water sources. Interviews revealed the uncontrolled application of agricultural inputs such as fertilizers and pesticides. Animals were also observed in close proximity to water sources. These are potential threats to source water quality given the seemly high water table in some parts of Kabale Municipality and potential for surface runoffs and sediment transport.







**Plate 1.** Water spring constructed in <5 m away from a residential area and wastewater was seen flowing from the residential area.

Establishing crop farms in close proximity to drinking water sources puts it at risk of chemical contamination especially when fertilizers, organic manure, and pesticides applied on crop farms end up in drinking water sources by runoff water after heavy rains. In a related study conducted by [16] reported high nitrate concentration in the majority of wells with typical values of 100 - 300 ppm and exceeding 600 ppm in some other areas of the Gaza Strip.

More to farming, eucalyptus trees were observed grown in close proximity to 42.8% of water springs. Eucalyptus is a unique tree species with the ability to efficiently adjust to surplus water situations when its water requirement rises to as high as 90 liters per plant per day. The Eucalyptus trees negatively affect the water springs by drawing much water to mean its daily water requirements (90 liters per day). A study by [17] in south Karnataka, India revealed a negative correlation between distance from the eucalyptus plantation and the depth of freshly dug bore wells. The study further revealed that bore well yields had reduced by 35 to 42% in the study area during the span of 3-5 years when they were located within a distance of 1 Km from Eucalyptus plantations. A study by [18] reported that sap flow measurements showed transpiration rates of 2 - 3.7 mm d<sup>-1</sup>, lowering groundwater levels by more than 0.5 m with respect to the surrounding grassland. According to [19], Eucalyptus had deepened the water table by 0.762 m (0.833 yards) per year in the villages of Total and Kot. As a result, the average depth of the water table reached 20.116 m (22 yards) for Total village and 15.544 m (17 yards) for Kot village.

The tendency of drinking water contamination at the source depends on nature of the source, topography, and distance of septic tanks, distance from toilets, and other land use activities in close proximity to a water source. Whereas there are various agents of source water pollution, humans, animals, and precipitation (runoff) are principal agents of contaminant transport to a water source in the downstream areas. Constructed surfaces and bare soil increase the rate of runoff and provide channels between contaminants and source water [20]. Poorly positioned and improper functioning toilets and septic tanks have been linked to many outbreaks of waterborne diseases. Subsurface soil permeability or soil subject to inundation and steeply sloping sites enhance the likelihood of the presence of open pathways for contaminant transport.

#### 3.2. Bacteriological Quality of Domestic Water Samples

Bacteriological analysis of water samples from 14 protected water springs in three divisions of Kabale Municipality showed that all samples were positive for total coliforms and heterotrophic bacteria counts. Unlike the thermotolerant *E. coli* which was at times not detected in the water sample especially in samples collected during the dry spell (**Table 2**).

#### 3.2.1. Total Coliforms (TC)

Total coliforms counts ranged from 1 to 419 CFU/100 ml and from 0 to 32 CFU/100 ml wet season and dry season, respectively (**Table 2**). The highest con-

centration (419 CFU/100 ml) in the wet season occurred at Rushaki water spring (S02) followed by Kamatojo water spring (S01) with 211 CFU/100 ml. Kirigime water spring recorded the highest total coliform counts (32 CFU/100 ml) in the dry season than any other source, followed by Kamatojo water spring (S01) with 20 CFU/100 ml (**Table 2**). The mean values for total coliforms were 85.71 CFU/100 ml and 10.36 CFU/100 ml in the wet season and dry season, respectively (**Table 3**). This pattern of total coliform counts indicates that water has come in contact with materials like human feces, soil, plants among others. However, their presence in drinking water indicates that disease-causing organisms could be in the water system [21].

In another related study conducted in South Africa, total bacterial count in sampled borehole water ranged from 1 to 44 CFU/100 ml and from 1.0 to 579.4 CFU/100 ml during winter and spring, respectively [22]. With the exception of only one borehole out of eight, all borehole water samples were within the permissible standards of WHO drinking water standards [22].

Location	Total C (CFU/1			<i>chia coli</i> 100 ml)	Total Heterotrophic Bacteria (CFU/100 ml)			
	Wet season	Dry season	Wet season	Dry season	Wet season	Dry season		
1.	211	20	84	6	511	221		
2.	419	0	138	0	225	7		
3.	105	32	31	12	297	208		
4.	10	15	0	1	132	88		
5.	105	6	43	1	253	30		
6.	1	24	0	2	80	225		
7.	10	5	0	1	191	11		
8.	23	0	5	0	30	10		
9.	89	9	10	1	153	75		
10.	56	17	17	5	211	168		
11.	21	2	5	0	259	10		
12.	17	0	4	0	84	10		
13.	82	13	0	2	232	201		
14.	51	2	0	0	101	4		
Max.	419	32	138	12	511	225		
Min.	1	0	0	0	30	4		
Mean	85.71	10.36	22.00	2.21	197.07	90.57		
SD	111.327	10.119	37.783	3.378	120.109	92.514		
Standards								
WHO (2006)	0	0	0	0	<500	<500		
UNBS (2008)	0	0	0	0	<500	<500		

Table 2. Results of biological analysis for spring waters from the study area.

		Mean	Ν	Std. Deviation	Std. Error Mean
Dela 1	T.C <sub>wet</sub>	85.71	14	111.33	29.75
Pair 1	T.C <sub>dry</sub>	10.36	14	10.12	2.70
	E.C <sub>wet</sub>	24.07	14	40.43	10.81
Pair 2	E.C <sub>dry</sub>	2.21	14	3.38	0.903
Pair 3	THB <sub>wet</sub>	197.07	14	120.11	32.10
	THB <sub>dry</sub>	90.57	14	92.51	24.73

Table 3. Paired samples statistics between bacteria counts in the wet and dry season.

#### 3.2.2. Thermotolerant E. coli

With regards to thermotolerant E. coli, Samples were found to contain E. coli counts ranging from 0 to 138 CFU/100 ml and from 0 to 12 CFU/100 ml in the wet season and dry season, respectively and the maximum thermotolerant E. coli was highest at Rushaki water spring (S02) during the wet season (Table 2). The highest thermotolerant E. coli (12 CFU/100 ml) were recorded at Kirigime water spring. Additionally, the study found out that 35.7% of water samples were free from thermotolerant E. coli in the dry season. The mean values for thermotolerant E. coli were 24.07 CFU/100 ml and 2.21 CFU/100 ml in the wet season and dry season, respectively (Table 2). The high level of thermotolerant E. coli recorded in this study may be attributed to the high degree of contamination of the water sources due to unhygienic practices in close proximity to water sources. The current study findings, however, disagree with [23] who reported that all water samples collected from unprotected water sources had fecal coliforms (FC), and 80% of samples had fecal coliforms ranging from 0.67 to 266.67 CFU/100 ml. A related study by [24] reported high thermotolerant *E. coli* levels up to 62,000 CFU/100 ml in drinking water sources in Central Gonja District.

According to [25], *E. coli* is a subgroup of the fecal coliform group. Most thermotolerant *E. coli* bacteria are harmless and exist in the intestines of warm-blooded animals and people. However, some strains can cause human illness. The presence of *E. coli* in a drinking water sample usually is indicative of recent fecal contamination [24]. *E. coli* O157: H7 strain is known to cause most outbreaks [21]. The water samples reported with *E. coli* does not necessarily mean that O157: H7 is present. It does indicate recent fecal contamination.

#### 3.2.3. Total Heterotrophic Bacteria (THB)

The concentration of THB count varied from 30 to 511 CFU/100 ml and from 4 to 225 CFU/100 ml during the wet season and dry season, respectively (**Table 2**). The mean values for THB in wet and dry seasons were 197.07 CFU/100 ml and 90.57 CFU/100 ml, respectively (**Table 2**). Various studies [26] [27] [28] have revealed major reasons for bacteriological contamination of water sources in urban centers as intermittent water supply which allows entry of any wastewater in the distribution system; layout of water pipes in close proximity to the sewer lines and overloading of sewage channels and sewers which in most cases remain

blocked.

Paired samples T-test showed a significant difference between CFU of total coliforms in water samples collected during wet season (M=85.71, SD = 111.33) and dry season (M = 10.36, SD = 10.12); t(13) = 2.5, p = 0.026. Additionally, there was a significant difference between CFU of heterotrophic bacteria count in water samples collected during wet season (M= 197.07, SD = 120.11) and dry season (M = 90.57, SD =92.51); t(13) = 3.5, p = 0.004 (**Table 3** and **Table 4**). There was no significant difference between CFU of *E. coli* in water samples collected during wet season (M = 24.08, SD = 40.43) and dry season (M = 2.21, SD = 3.38); t(13) = 2.0, p = 0.062 (**Table 3** and **Table 4**). These results indicate that during wet season, total coliforms and heterotrophic bacteria are abundant in water sources. For results showing significant no significant difference (p > 0.05) indicate that *E. coli* can be drinking water sources regardless of variations in season.

The study findings are in agreement with [29] who found out that concentrations of indicator bacteria were lower during the winter months and increased in April, which was when the first rains began. In addition, concentrations of total coliform and *E. coli* were significantly higher in the rainy seasons (ANOVA, p <0.01) [29]. However, the study findings do not concur with [30] who reported that during the rainy season, *E. coli* detection was significantly higher and most frequent compared to the dry season (all p < 0.05). Fecal contamination in improved drinking water sources showed a statistically significant seasonal trend of greater contamination during the wet season (p < 0.001) [31].

## 3.2.4. Risk Assessment of TC, Thermotolerant *E. coli*, and THB Contained in Spring Water Sources

According to [32] and [33] standards, total coliforms and *E. coli* shouldn't be detected in 100 ml of any drinking water sample and the concentration of heterotrophic bacteria should be <500 CFU/100 ml. By this declaration, samples were drawn from drinking water sources with no detectable total coliforms and *E. coli*, and Heterotrophic Bacteria Count < 500 CFU/100 ml were fit for drinking (Table 5). World Health Organization categories bacterial contamination into five different categories namely; 0 CFU/100 ml (no risk/ in compliance), 1 to 10 CFU/100 ml (low risk), 11 to 100 (medium risk), 101 to 1000 (high risk) and <1000 CFU/100 ml (Very high risk).

Table 4. Paired samples statistics between bacteria count	is in wet and dry seasons (cont'd).
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			Pa	aired Differences					
					95% Confidence Interval of the Difference		t	df	Sig. (2-tailed)
		Mean	Std. Deviation	Std. Error Mean	n Lower Upper				
Pair 1	T.C <sub>wet</sub> - T.C <sub>dry</sub>	75.36	112.49	30.06	10.41	140.31	2.51	13	0.026
Pair 2	E.C <sub>wet</sub> - E.C <sub>dry</sub>	21.86	40.06	10.72	-1.28	44.98	2.04	13	0.062
Pair 3	$\text{THB}_{\text{wet}}$ - $\text{THB}_{\text{dry}}$	106.50	115.03	30.74	40.08	172.91	3.46	13	0.004

Bacteria contamination	Remarks	Total Coliform (CFU/100 ml)		Total Coliform (CFU/100 ml)		Thermotolerant <i>E. coli</i> (CFU/100 ml)		Thermotolerant <i>E. coli</i> (CFU/100 ml)		Total Heterotrophic Bacteria (CFU/100 ml)		Total Heterotrophic Bacteria (CFU/100 ml)	
		Wet	Dry	Wet	Dry	Wet	Dry	Wet	Dry	Wet	Dry	Wet	Dry
		No.	No.	%	%	No.	No.	%	%	No.	No.	%	%
<1	No risk/Compliance	0	4	0	28.6	5	5	35.7	35.7	0	0	0	0
1 - 10	Low	3	3	21.4	21.4	3	8	21.4	57.1	0	0	0	0
11 - 100	Medium	7	7	50.0	50.0	5	1	35.7	7.1	0	9	0	64.3
101 - 1000	High risk	4	4	28.6	28.6	1	0	7.1	0	14	5	100	35.7
>1000	Very high risk	0	0	0		0	0	0	0	0		0	0

Table 5. Risk Assessment of TC, thermotolerant E. coli, and THB.

Majority of the water samples (50%) tested for total coliforms fell within the medium risk category (11 to 100 CFU/100 ml) in the both dry and wet season. While 28.6% of water samples fell within the high-risk category in the wet season, none of the water samples fell within a high-risk category (101 to 1000 CFU/100 ml) in the dry season (**Table 5**). The study findings further indicate 57.1% of water samples tested for thermotolerant *E. coli* concentration fell within the low-risk category (1 to 10 CFU/100 ml). Additionally, 35.7% and 28.6% of samples had no thermotolerant *E. coli* detected during the dry season and wet season, respectively. The study findings closely with [30] who found out that 25% of source water samples had *E. coli*. The current study findings further revealed that only 7.1% of water samples fell within the high-risk category (101 - 1000 CFU/100 ml) during the wet season (**Table 5**). For heterotrophic bacteria counts, (64.3%) and (35.7%) had colon forming units <100 CFU/100 ml and 100 - 1000 per 100 ml, respectively during the wet season. All water samples collected during dry season had THB counts ranging from 100 - 1000 CFU/100 ml (**Table 5**).

A study by [34] found out that 80% of filtered water samples had no *E. coli* per 100 ml and 17% were within a low-risk category (1 to 10 CFU/100 ml) in addition, [35] found out that all test water samples had bacteria counts < 10 CFU/100 ml (low-risk category). In addition, [36] revealed that a total of 66% of all outfall samples had fecal indicator levels greater than 10,000 CFU/100 ml (high-risk level category).

## 4. Conclusions

Major of water sources are constructed with less in less than 20 meters away from residential areas. The mean concentrations of *E. coli*, total coliforms, and THB were 24.07 CFU/100 ml, 85.71 CFU/100 ml, and 197.07 CFU/100 ml, respectively in the wet season. There were significant differences between CFU of total coliforms, HTB, between wet and dry seasons (p = 0.026). The study concludes that:

1) Drinking water collected and analyzed during the wet season for total coli-

forms did not conform to WHO drinking water quality guideline value of no detection per 100 ml.

2) The quality of water from selected drinking water sources was compromised by anthropogenic activities such as farming latrines, landfills, and animal and municipal wastes, constructed in <20 meters away from these water sources.

3) Majority spring water sources (57.1%) are located in less than 20 meters away from residential areas with significant paved areas, the presence of septic tanks and pit latrines.

4) Seasonal variations influence the concentration of bacteria in drinking water sources. For instance significant differences (p = 0.026) between CFU of total coliforms and heterotrophic bacteria count in water samples collected during wet seasons were noted.

5) Majority drinking water sources analyzed during wet season were in excess of WHO recommended guidelines for drinking water (WHO, 2006). This implies that water from these sources is not suitable for drinking without treatment.

#### **5. Recommendations**

Based on the study findings, the following are the recommendations to the policymakers, NGOs, and households in Kabale Municipality.

1) Spring water should be treated before drinking due to high bacteria concentrations which makes it unsafe for consumption. The responsibility to improve household drinking water must generally fall on individual households.

2) The government and NGOs should take an active role to ensure that residential areas are constructed at a distance of 20 meters away from spring water sources to prevent possible water contamination.

3) Nowadays, there are available cheap water testing methods to evaluate the bacteriological quality of water. The local people should always be involved in testing for bacteriological quality of water before use.

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#### **Conflicts of Interest**

The authors declare no conflict of interest.

#### **Data Availability Statement**

Data used to support the findings of this study are can be availed to you upon request.

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