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# Water-Supply Potability Status of Bangladesh University of Health Sciences (BUHS)

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*This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.*

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## ABSTRACT

Water that is free of disease-producing microorganisms and chemical substances harmful to health is called potable water. Drinking water quality has a microbiological and a physicochemical dimension. In public water supply systems, water should be disinfected**.** This observational study was conducted through a multiple-tube method. And the water supply channel of Bangladesh University of Health Sciences (BUHS) served as the study's sample. Using an aseptic procedure, 105 ml of water from various sources is collected for each sample on a conical flask. The selected sample was from the four different water supply points at BUHS institution. From sample 1, the MPN (Most Probable Number) index we found the combination of positive sets (0-1-0). The MPN value per 100ml water is (1). From sample 2, the MPN index we found the combination of positive sets (1-1-1). The MPN value per 100ml water is (5). From sample 3, the MPN index we found the combination of positive sets (1-2-1). The MPN value per 100ml water is (7). From sample 4, the MPN index we found the combination of positive sets (0-1-0). The MPN value per 100ml water is (1). In Dhaka city, there is a possibility of contamination of the water sources. Bangladesh University of Health Sciences is a renowned institution with a big number of staff. So, it is important to examine the water condition in this institution. Our work and research reflect a little about it. Gladly and hopefully, we found that there is no pathogenic organism in the water of this institution which is used every day and everybody drinks.

*Keywords: BUHS; E. coli; coliforms; macconkey; potablility.*

## 1. INTRODUCTION

Water supply is a channel by which we have water that we do drink or use. According to the WHO [1] water has got two distinctive characteristics, whether it is safe for use and drink termed “potable” and which is not safe is termed as “non-potable”. The classification is based on the presence or absence of some selective indicator microorganisms in water. This research will be conducted on the Bangladesh University of Health Sciences (BUHS) water supply to assess the potability status of this institute’s water. After this study, we will have an assessment of our institute’s water supply, which will tell us whether we are using safe and pure water or using unsafe and impure water.

“It is impractical to attempt the routine isolation of pathogens because they are present in relatively small numbers compared with other types of microorganisms. Moreover, there are many types of pathogens and each requires a unique microbiological isolation technique stated by International Standard organization” [2]. The approach that has been adopted is to analyze for indicator organisms that inhabit the gut in large numbers and are excreted in human feces. One study [3] suggested that “the presence of these indicator organisms in water is evidence of fecal contamination and, therefore of a risk that pathogens are present. If indicator organisms are present in large numbers, the contamination is considered to be recent and/or severe. Bacteria in water are, in general, not present individually, but as clumps or in association with particulate matter”. Department of the U.S. environment [4] reported that “when enumerating bacteria in water it is not the number of individual bacteria present which are counted, but the number of clumps of bacteria or the particles and their associated bacteria. Each clump or particle may have many bacteria associated with it. Historically, most of our concern about water purity has been related to the transmission of diseases that are mostly waterborne diseases like shigellosis, salmonellosis, etc. Therefore, tests have been developed to determine the safety of the water”.

“The tests for water purity in use today are aimed instead at detecting particular indicator organisms. This can be seen that there are several criteria for an indicator organism. The most important criterion is that the microbe be consistently present in human feces in substantial numbers so that its detection is a good indication that human waste is entering the water” [5]. In the United States, the usual indicator organism in freshwater is “*coliform bacteria”.* Coliforms are defined as aerobic or facultative anaerobic, gram-negative, non-endospore forming, rod-shaped bacteria that ferment lactose to form gas within 48 hours of being placed in lactose broth at 35°c, but there is another thermotolerant coliform named fecal coliform has got the capability to grow on >44°C [6].

## 2. METHODS AND MATERIALS

This observational study was conducted through a multiple-tube method. And the sample for this study is the water supply channel of Bangladesh University of Health Sciences (BUHS). From different sources, 105 ml water for each sample is been collected on a conical flask with an aseptic technique. Observation on the procedure will be that, either the sample containing fecal coliform or not.

### 2.1 The Multiple-Tube Method

"The technique has been used for the analysis of drinking water for many years with satisfactory results. It is the only procedure that can be used if water samples are very turbid or if semi-solids such as sediments or sludges are to be analyzed. The procedure followed is fundamental to bacteriological analyses and the test is used in many countries. It is customary to report the results of the multiple fermentation tube test for coliforms as a most probable number (MPN) index" [7]. This is an index of the number of coliform bacteria that, more probably than any other number, would give the results shown by the test. It is not a count of the actual number of indicator bacteria present in the sample.

### 2.2 Qualitative Analysis through the Mpn Method

#### 2.2.1 Presumptive test

"The presumptive test is the very first portion of the MPN test method. This is primarily done for the detection of Gram-negative coliform bacteria in the water samples. For this task, 15 series of test tubes containing 10 mL of lactose fermentation broth were needed for each sample. 10 ml, 1 ml, and 0.1 ml samples were added sequentially in 5 test tubes containing 10 mL lactose fermentation broth 2X, 5 tubes containing 10 ml lactose fermentation broth 1X, and 5 test tubes containing 10 ml 1X lactose fermentation broth. Each tube was incorporated with a Durham tube indicating gas formation after lactose fermentation by coliform bacteria" [8].

#### 2.2.2 Confirmed test

"The test tubes showing positive results by the accumulation of gas in the Durham tubes were selected for the confirmed test to determine the presence of E. coli in the respective water samples. The loopful sample from the broths which gave positive results in the presumptive test, was inoculated on EMB agar to detect as well as differentiate E. coli and other Gram-negative coliform bacteria. The plates were incubated at 37º C for 24 hours" [8].

#### 2.2.3 Completed test

This is the final part of the MPN (Most Probable Number) test procedure which was completed after the confirmation of the indicator bacteria Escherichia coli found in the EMB agar medium. "The suspected E. coli from a single colony of green metallic sheen was introduced into a lactose fermentation broth 1X again for the assurance of gas production after the fermentation of lactose. Gram staining was also performed for the confirmation of E. coli isolates" [8].

**Table 1. Reactions over different media**

| Medium | Reactions | |
|---|---|---|
| | **Total coliforms at 35 or 37 °C** | **Thermotolerant coliforms at 44 or 44.5 °C** |
| 1. Macconkey broth. | 1. Gas visible in the inverted fermentation (Durham) tube plus turbidity of the medium | 1. Same as total coliforms at 35 or 37 °C. |
| 2. Macconkey agar. | 2. Lactose fermenting pink colony. | 2. Not applicable. |
| 3. EMB agar. | 3. Greenish Metallic sheen colony will appear. | 3. Same as total coliform at 35 or 37°. |
| 4. MIU agar | 4. Indole positive with gas production and non-motile in nature. | 4. Same as 35 or 37° C |

## 3. RESULTS

The selected sample was from the four different water supply points at BUHS institution. And the sampling technique is random sampling. They are enlisted here.

Following reactions, the media plate will be assessed to generate results based on the growth of bacteria will be considered as positive results, if these characteristics will not be found then the result will be considered negative.

From the result of different testing procedures here we have one double strength set positive with coliform bacteria (*E. coli*).

**Table 2. The four samples from different water supply points at BUHS**

| Sample no. | Collection point (105 ml /sample) |
|---|---|
| 1 | Filter water |
| 2 | Tap water in the washroom |
| 3 | Cafeteria water supply |
| 4 | Vice Chancellor's office. |

Now the sample is being placed on the MacConkey broth for their further investigation and the reporting of each individual test is also sorted out along with the dates. The following chart will tell us the interpretation of each particular sample analysis. From this reporting will be able to know the status of water quality.

From each sample, we have a particular result for them.

Here's a typical classification is been presented that is based on increasing orders of magnitude of fecal contamination.

## 4. DISCUSSION

U.S Environmental Protection Agency [4] stated that "the term "total coliforms" refers to a large group of Gram-negative, rod-shaped bacteria that share several characteristics. The group includes thermotolerant coliforms and bacteria of fecal origin, as well as some bacteria that may be isolated from environmental sources. Thus, the presence of total coliforms may or may not indicate fecal contamination. In extreme cases, a high count for the total coliform group may be associated with a low, or even zero, count for thermotolerant coliforms. Such a result would not necessarily indicate the presence of fecal contamination. It might be caused by the entry of soil or organic matter into the water or by conditions suitable for the growth of other types of coliforms". "In the laboratory, total coliforms are grown in or on a medium containing lactose, at a temperature of 35 or 37 °C. They are provisionally identified by the production of acid and gas from the fermentation of lactose. One reported that thermotolerant (fecal) coliforms the term "fecal coliform" has been used in water microbiology to denote coliform organisms that grow at 44 or 44.5 C and ferment lactose to produce acid and gas. In practice, some organisms with these characteristics may not be of fecal origin and the term "thermotolerant coliform" is, therefore, more correct and is becoming more commonly used" [10]. Nevertheless, the presence of thermotolerant coliforms nearly always indicates fecal contamination. Usually, more than 95 percent of thermotolerant coliforms isolated from water are the gut organism Escherichia coli. The presence of which is definitive proof of fecal contamination.

As a result, it is often unnecessary to undertake further testing to confirm the specific presence of E. coli. In the laboratory, thermotolerant coliforms are grown on media containing lactose, at a temperature of 44 or 44.5 °C. They are provisionally identified by the production of acid and gas from the fermentation of lactose [11]. Environments rich in nutrients may promote the development or survival of some thermotolerant coliform organisms other than E. coli. When, for instance, an exceptionally high result is produced from water that was assumed to be quite clean, this possibility should be taken into account. In this situation, a microbiology lab's guidance should be sought in order to identify the more precise indicator, *E. coli*.

WHO [12] reported that "Fecal streptococci the presence of fecal streptococci is evidence of fecal contamination. Fecal streptococci tend to persist longer in the environment than thermotolerant or total coliforms and are highly resistant to drying. It is, therefore, possible to isolate fecal streptococci from water that contains few or no thermotolerant coliforms, for example, when the source of contamination is distant in either time or space from the sampling point. Fecal streptococci grow in or on a medium containing sodium azide, at a temperature of 37-44 °C. They are usually detected by the reduction of a dye (generally a tetrazolium-containing compound) or the hydrolysis of aesculin. Routine methods may give "false positives" and additional confirmatory tests may be required".

**Table 3. Sample 1 report status**

| **Date** | **8th June 2018. Sample placed at broth.** | | **Sample no.** | **1** | **Location** | **Bangladesh University of Health Sciences campus** | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| **Date** | **10th June2018** | **Date** | **11th June 2018** | | **Date** | **11th June 2018** | **Date** | **12th June 2018** | | **Date** | **12th June2018** |
| | **MacConkey Broth purple 37 °c/48h** | | **Mac agar** | **EMB agar** | | **EMB agar 44°c/24h** | | **MIU** | | | **Result** |
| | | | | | | | | **Indole** | **Indole** | | |
| | | | **37°c/24h** | | | | | **37°c/24h** | **44°c/24h** | | |
| Tube Name | BUHS | | | | | | | | | | ----- |
| D-50m | No growth. | | ----- | ----- | | ----- | | ----- | ----- | | ----- |
| D-1 | No growth | | ----- | ----- | | ----- | | ----- | ----- | | ----- |
| D-2 | No growth | | ----- | ----- | | ----- | | ----- | ----- | | ----- |
| D-3 | No growth | | ----- | ----- | | ----- | | ----- | ----- | | ----- |
| D-4 | No growth | | ----- | ----- | | ----- | | ----- | ----- | | ----- |
| D-5 | Growth positive | | Positive | Positive | | Negative | | Positive | ---- | | Total Coliform identified. |
| S-1 | No growth | | ----- | ----- | | ----- | | ----- | ----- | | ----- |
| S-2 | No growth | | ----- | ----- | | ----- | | ----- | ----- | | ----- |
| S-3 | No growth | | ----- | ----- | | ----- | | ----- | ----- | | ----- |
| S-4 | No growth | | ----- | ----- | | ----- | | ----- | ----- | | ----- |
| S-5 | No growth | | ----- | ----- | | ----- | | ----- | ----- | | ----- |

*From the MPN index, we found the combination of positive sets (0-1-0). The MPN value per 100ml water is 1.*

**Table 4. Sample 2 report status**

| **Date** | **8th June 2018. Sample placed at broth.** | | **Sample no. 2** | | | **Location** | | **Bangladesh University of Health Sciences campus** | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| **Date** | **10th June 2018** | **Date** | **11th June 2018** | | **Date** | **11th June 2018** | **Date** | **12th June 2018** | | **Date** | **12th June2018** |
| | **MacConkey Broth purple 37 °c/48h** | | **Mac agar** | **EMB agar** | | **EMB agar 44°c/24h** | | **MIU** | | | **Result** |
| | | | | | | | | **Indole** | **Indole** | | |
| | | | **37°c/24h** | | | | | **37°c/24h** | **44°c/24h** | | |
| Tube Name | BUHS | | | | | | | | | | ----- |
| D-50m | Growth positive | | positive | positive | | positive | | negative | positive | | Thermotolerant coliform |
| D-1 | Growth positive | | positive | positive | | positive | | positive | positive | | Thermotolerant coliform |
| D-2 | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |
| D-3 | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |
| D-4 | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |
| D-5 | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |
| S-1 | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |
| S-2 | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |
| S-3 | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |
| S-4 | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |
| S-5 | Growth positive | | positive | positive | | negative | | positive | positive | | Thermotolerant coliform |

*From the MPN index, we found the combination of positive sets (1-1-1). The MPN value per 100ml water is 5.*

**Table 5. Sample 3 report status**

| **Date** | **13th June 2018.sample placed at broth.** | | **Sample no. 3** | | **Location** | **Bangladesh University of Health Sciences campus** | | | |
|---|---|---|---|---|---|---|---|---|---|
| **Date** | **15th June 2018** | **Date** | **16th June 2018** | | **Date** | **16th June 2018** | **Date** | **17th June 2018** | **Date** | **17th June 2018** |
| | **MacConkey Broth purple 37 °c/48h** | | **Mac agar** | **EMB agar** | | **EMB agar** | | **MIU** | | **Result** |
| | | | **37°c/24h** | | | **44°c/24h** | | **Indole** | **Indole** | |
| | | | | | | | | **37°c/24h** | **44°c/24h** | |
| Tube Name | ----- | | | | | | | | | |
| D-50m | Growth positive | | positive | positive | | positive | | negative | positive | Thermotolerant coliform |
| D-1 | No growth | | ---- | ---- | | ---- | | ---- | ---- | ---- |
| D-2 | No growth | | ---- | ---- | | ---- | | ---- | ---- | ---- |
| D-3 | Growth positive | | positive | positive | | positive | | positive | positive | Thermotolerant coliform |
| D-4 | Growth positive | | positive | positive | | positive | | negative | positive | Do |
| D-5 | No growth | | ---- | ---- | | ---- | | ---- | ---- | ---- |
| S-1 | No growth | | ---- | ---- | | ---- | | ---- | ---- | ---- |
| S-2 | No growth | | ---- | ---- | | ---- | | ---- | ---- | ---- |
| S-3 | Growth positive | | positive | positive | | negative | | positive | ---- | Total coliform |
| S-4 | No growth | | ---- | ---- | | ---- | | ---- | ---- | ---- |
| S-5 | No growth | | ---- | ---- | | ---- | | ---- | ---- | ---- |

*From MPN index we found the combination of positive sets (1-2-1). The MPN value per 100ml water is 7.*

**Table 6. Sample 4 report status**

| **Date** | **13th June 2018.sample placed at broth.** | | **Sample no. 4** | | | **Location** | | **Bangladesh University of Health Sciences campus** | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| **Date** | **15th June 2018** | **Date** | **16th June 2018** | | **Date** | **16th June 2018** | **Date** | **17th June 2018** | | **Date** | **17th June 2018** |
| | **MacConkey Broth purple 37 °c/48h** | | **Mac agar** | **EMB agar** | | **EMB agar 44°c/24h** | | **MIU** | | | **Result** |
| | | | | | | | | **Indole** | **Indole** | | |
| | | | **37°c/24h** | | | | | **37°c/24h** | **44°c/24h** | | |
| Tube Name | ----- | | | | | | | | | | |
| D-50m | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |
| D-1 | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |
| D-2 | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |
| D-3 | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |
| D-4 | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |
| D-5 | Growth positive | | positive | positive | | negative | | positive | ---- | | Total Coliform detected |
| S-1 | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |
| S-2 | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |
| S-3 | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |
| S-4 | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |
| S-5 | No growth | | ---- | ---- | | ---- | | ---- | ---- | | ---- |

*From the MPN index, we found the combination of positive sets (0-1-0). The MPN value per 100ml water is 1.*

**Table 7. Compare the combination of positive results, from the most probable number index that is values/100ml of sample for a test of one 50ml, five 10ml, and five 1 ml volumes [7].**

| No of the tubes giving a positive reaction | | | |
|---|---|---|---|
| 1×50 ml | 5×10 ml | 5×1 ml | MPN/100ml |
| 0 | 0 | 0 | <1 |
| 0 | 0 | 1 | 1 |
| 0 | 0 | 2 | 2 |
| 0 | 1 | 0 | 1 |
| 0 | 1 | 1 | 2 |
| 0 | 1 | 2 | 3 |
| 0 | 2 | 0 | 2 |
| 0 | 2 | 1 | 3 |
| 0 | 2 | 2 | 4 |
| 0 | 3 | 0 | 3 |
| 0 | 3 | 1 | 5 |
| 0 | 4 | 0 | 5 |
| 1 | 0 | 0 | 1 |
| 1 | 0 | 1 | 3 |
| 1 | 0 | 2 | 4 |
| 1 | 0 | 3 | 6 |
| 1 | 1 | 0 | 3 |
| 1 | 1 | 1 | 5 |
| 1 | 1 | 2 | 7 |
| 1 | 1 | 3 | 9 |
| 1 | 2 | 0 | 5 |
| 1 | 2 | 1 | 7 |
| 1 | 2 | 2 | 10 |
| 1 | 2 | 3 | 12 |
| 1 | 3 | 0 | 8 |
| 1 | 3 | 1 | 11 |
| 1 | 3 | 2 | 14 |
| 1 | 3 | 3 | 18 |
| 1 | 3 | 4 | 21 |
| 1 | 4 | 0 | 13 |
| 1 | 4 | 1 | 17 |
| 1 | 4 | 2 | 22 |

**Table 8. The result of the samples**

| Sample no. | Isolated organism | MPN value /100 ml |
|---|---|---|
| 1 | Total coliform | 1 |
| 2 | Thermotolerant coliform | 5 |
| 3 | Thermotolerant coliform | 7 |
| 4 | Total coliform | 1 |

**MPN (Most Probable Number)*

**Table 9. Example of classification and color-code scheme for thermotolerant (fecal) coliforms or E. coli in water supplies [9].**

| Count per 100ml | Category and color code | Remarks |
|---|---|---|
| 0 | A (blue) | In conformity with WHO guidelines |
| 1–10 | B (green) | Low risk |
| 10–100 | C (yellow) | Intermediate risk |
| 100–1000 | D (orange) | High risk |
| >.1000 | E (red) | Very high risk |

**Table 10. List of bacteria reported in drinking water in Bangladesh and their possible health effects [19]**

| Bacteria | Possible health effects |
|---|---|
| *Aeromonas hydrophila* | Variety of diseases like septicemia, gastroenteritis in young children and elderly |
| *Enterobacter aerogenes* | Cause infections in urinary and respiratory tracts |
| *Enterococcus species* | Cause infection in urinary tract |
| *Escherichia coli* | Cause food poisoning, gastroenteritis, urinary tract infections |
| *Klebsiella species* | An astute pathogen that ordinarily causes nosocomial contaminations in the urinary tract, respiratory tract, lung, wound destinations |
| *Listeria species* | Variety of diseases like meningitis, endocarditis and has high mortality rate |
| *Pseudomonas aeruginosa* | Causes diseases in newborns and elderly patients |
| *Salmonella species* | Typhoid |
| *Shigella species* | Cause abdominal pain, tenesmus, watery diarrhea |
| *Staphylococcus species* | Cause superficial skin lesions, food poisoning |
| *Vibrio cholerae* | Cholera |

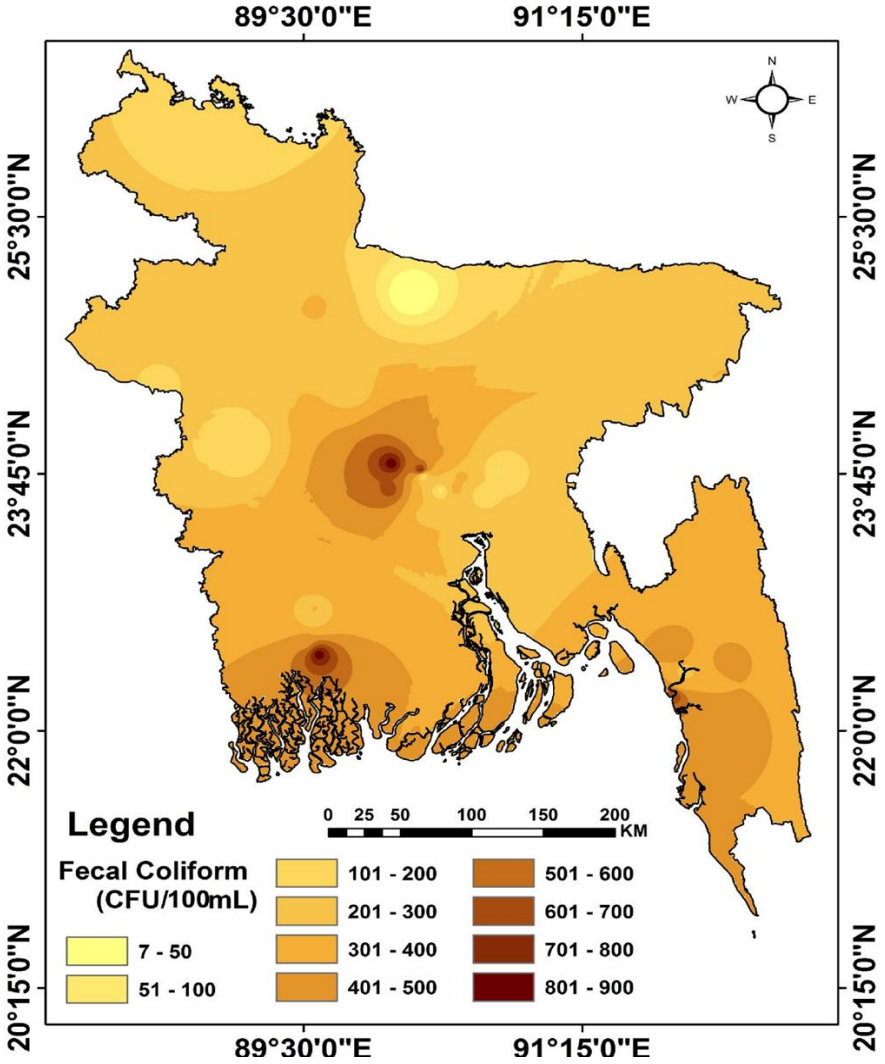


**Fig. 1. Spatial distribution of fecal coliform (CFU/100 mL) in the surface water of Bangladesh [17,18]**

In Bangladesh both the surface water and groundwater contain different contaminants like biological and chemical pollutants Waterborne pathogens, in the form of disease-causing bacteria and viruses from human and animal waste, are a major cause of illness from contaminated drinking water. “Total coliform includes bacteria that are usually found in human or animal waste and in water that has been influenced by surface water. The main microbial risks are usually associated with drinking water that is contaminated with human or animal feces. In Bangladesh, the presence of fecal coliform and other pathogenic bacteria has been frequently found in ponds and river water” [11].

A high number of fecal coliform has also been reported in the household-based rainwater harvesting systems (465 CFU/100 mL) and community-based rainwater harvesting systems (856 CFU/100 mL) of southwest coastal areas of Bangladesh.[16] Further, “very high number of fecal coliform in the water samples collected from Hemayetpur CETP (Horindhora) (780 CFU/100 mL), Uttar Mitora (860 CFU/100 mL) located at the bank of Dhaleshwari River, Barha Ghat of Padma River (640 CFU/100 mL) and CUFL, Karnaphuli River (680 CFU/100 mL) (DoE, 2016). In Sherpur, fecal coliform was found in pond water, and supply water in a range of 5–10 CFU/100 ml” [16]. The presence of biological contaminants possesses a threat to human health this is why regular monitoring and evaluation of water quality is required in order to ensure safe drinking water.

Diseases spread by unsafe water in Bangladesh include cholera, diarrhea, viral fever and typhoid.[13] In Bangladesh typhoid incidence rate is 252 per 100,000 people affected yearly [14]. With more than 100,000 cases estimated each year, Bangladesh is one of the countries with the highest number of people at risk for cholera, *Vibrio cholerae* along with rotavirus, campylobacter, enterotoxigenic *E.coli* are the main organisms responsible behind cholera outbreak [15]. There is a significant correlation between the microbiological quality of water and gastrointestinal diseases.

Since both the groundwater and surface water has been contaminated by different organism from the very earlier period which had been mentioned by different studies conducted throughout the country. This is why water quality analysis is required on a regular basis.

## 5. CONCLUSION

Water quality, however, may vary at any time in different areas of the system when piped small-community water supplies are being evaluated and samples are being obtained at various points in the system. Again, if these variations are the consequence of cross-contamination or contamination due to piping leaks, the causes may become clear during the sanitary inspection or after resampling.

A microbiological analysis of water quality from the institute, Bangladesh University of Health Sciences (BUHS) shows that its water supply is potable. In addition, contamination through fecal coliform is the least. The drinking water here is safe to drink and the most probable number of fecal coliform bacteria in per 100 ml of water is less than the risk limit. Nevertheless, regular inspection is necessary here to provide safe water to drink for all the personnel here.

## COMPETING INTERESTS

Authors have declared that they have no known competing financial interests or non-financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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