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Original Research Paper

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# ASSESSMENT OF BACTERIOLOGICAL QUALITY OF DRINKING WATER FROM VARIOUS SOURCES IN FOUR DISTRICTS OF TRIPURA: A NORTH-EASTERN STATE OF INDIA

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## ABSTRACT

**Background:** WHO estimated that, over 2 billion people live in water-stressed countries, which is expected to exacerbate in some regions as result of climate change and population growth. Globally, at least 2 billion people have been estimated to be consuming water from drinking water sources contaminated with faeces. Microbial contamination of drinking-water poses the greatest risk to drinking-water safety leading to outbreaks of various waterborne diseases such as diarrhoea, cholera, dysentery, typhoid and polio. Therefore, drinking water assessment for microbial contamination and bacteriological load must be conducted routinely to monitor and prevent waterborne illnesses. **Materials and Methods:** Study was conducted at Microbiology Department of AGMC & GBP Hospital, Agartala from four nearby districts of Tripura. A total of 157 water samples were collected from various sources following WHO guidelines from 27th July 2021 to 29th September 2021. Total coliform test was performed on all the samples by Most Probable Number (MPN) method. Subsequently the positive MPN samples were subjected to Eijkman's test for detection of thermotolerant E. coli. **Result:** 50.3% (79/157) & 42.7% (67/157) of the population from these four districts of Tripura were sourcing drinking water from tube wells & supply water systems respectively. Out of total 157 samples, 146 (93%) were positive on Total coliform count test (TCT) with 62% (Acceptable), 20% (unacceptable), 12.10% (Grossly polluted). 7% were excellent with negative TCT. 67 out of 146 TCT positive samples (46%) were positive on Eijkman's test, which signifies that 46% of the coliform were confirmed faecal thermotolerant Escherichia coli indicating that 67/157 (i.e., 43%) were having faecal contamination. **Conclusion:** The bacteriological quality analysis of the drinking water samples revealed that 69% were satisfactory. However, 93% were contaminated with coliforms which could be environmental or animal or human faecal coliforms contaminations. Therefore, all the water sources of drinking water should be properly sanitized, continuously monitored and bacteriological analysis should be performed routinely to prevent outbreaks of water borne illnesses.

**KEYWORDS :** Coliforms, Thermotolerant E. coli, Multiple tube method, Eijkman's test

## 1. INTRODUCTION

The majority of population in developing countries has no access to clean water. As a consequence, millions of people are suffering from diseases related to water, sanitation, and hygiene[1]. Unsafe water, inadequate sanitation, and poor hygiene are linked to 88% of diarrhoea cases worldwide [2]. Ingestion of water contaminated with human or animal faeces or urine containing pathogenic bacteria or viruses causes waterborne diseases such as cholera, typhoid, bacillary dysentery, adenoviruses, retroviruses, and other diseases. Studies showed that despite the worldwide efforts to deliver safe drinking water, the transmission of waterborne diseases is still a matter of major concern.

According to World Water Development, India ranks 120th out of 122 countries regarding water quality and ability and commitment to improving it.[3] The importance of water, sanitation and hygiene (WASH) has been proven to be a crucial factor in the control of water-borne diseases (Bartram & Cairncross et al.; Freeman et al; Patil et al.; JMP et al.).[4,5,6,7] An estimate by Joint Monitoring Programme (JMP) suggests that from 2000 to 2017, 10% of the total world population gained safely managed water services, yet 3 out of 10 people lack it. Also, in the same period a quarter of the total population in the world got access to basic sanitation services.

It was estimated that in 2017, 60% of the world's population had access to basic handwashing facilities available at home (JMP et al.).[7] India's Swachh Bharat Abhiyaan, launched in 2014, helped 500 million people, which represent 25% of the total world population, to gain access to basic sanitation. Even though more than 690 million people got access to toilets where India represents more than 50% of this population, 670 million people still practice open defecation in the world (JMP et al.).[7] Total WASH-related disability-adjusted life years (DALYs) for India were estimated to be 28,213.3, which is one of the highest in the world (Prüss-Üstün et al.).[8]

In western part of India, cases of water-borne diseases were reported to range between 0.25 and 4.18% of the total population with an average of 1.7%.[9] Study by Bhattacharya H. et al. detected that 11% of the ethnic and 8% of the non-ethnic households in rural area of West Tripura District were collecting water from sources which were heavily contaminated with coliform organisms.[10] Mukhopadhyay C et al. in Karnataka have found that 22 (27.5%) water samples and the majority (92.5%) of the water sources were contaminated with coliforms.[16] Tambe PV et al in Western Maharashtra have found that overall 49.8% of the water samples were polluted and 45.9% of the samples from piped water supply were also polluted.[18] Malhotra S et al have found that 42.9% (565/1,317) samples from various sources were unfit for human consumption.[17]

The problems of contamination of water distribution system are diverse, the major sources of water contaminants are mostly wastes from improper sanitation and agricultural and other activities that make their way to the water distribution networks.[11] Furthermore, break in the distribution system, age and improper maintenance of the distribution system, and low level of chlorine usually compromise the integrity of the distribution system and quality of potable water. Therefore, this requires proper protection of water supply from contamination and regular surveillance of water sources to reduce water-related diseases.

## 2. MATERIALS AND METHODS

### 2.1. Study Site:

Study was conducted in Tripura, a North-eastern State of India, from four nearby districts (Gomati, Khowai, Sipahijala and South Tripura) in the Department of Microbiology, Agartala Government Medical College & GB Pant Hospital, Agartala, Tripura.

### 2.2. Study Design and Sampling Procedures:

A cross-sectional study was conducted at AGMC & GBP Hospital to assess the bacteriological and physical parameters. Duration of study was roughly two months from 27th July 2021 to 29th September 2021. A total of 157 water samples were collected from sampling points following the WHO guidelines.[13] Of these 157 samples, 35 were from Gomati District, 57 were from South District, 25 were from Sipahijala District and 40 were from Khowai District. Sites were selected by a simple random sampling method. Structured proforma was used to collect the information of the water samples. Samples (250 ml) from pipelines were collected into sterilized glass bottles rinsed thoroughly with nitric acid and sterilized distilled water. For the chlorinated water samples, about 2.5 ml sodium thiosulphate was added into each sampling bottle to stop the chlorination process during transportation. Samples were transported to Bacteriology Laboratory of the Department of Microbiology, AGMC & GBP Hospital, Agartala in a cold box containing ice freezer packs for physical parameters analysis and bacteriological assessments respectively.

### 2.3. Sample Analysis

### 2.3.1. Total Coliform Count Test:

Drinking water samples were received in the Department of Microbiology after collection by a Health Staff of AGMC & GBP Hospital for the assessment of bacteriological quality of water from various public places of rural areas of four districts of Tripura. A total of 157 drinking water samples were collected from various water sources from rural areas of four nearby districts of Tripura between July 2021 to September 2021.

The samples were collected aseptically in sterilized containers and tested following the guidelines of the WHO.[13] 250ml of water samples from each source were collected in sterilized glass stopper bottles for microbiological examination. The samples were transported and stored strictly in accordance with the procedures and guidelines described in the WHO's guidelines for drinking water quality. Water samples from pipelines were collected into sterilized bottle added with 0.1 mL sodium thiosulphate (1.8% w/v) per 100 mL of water sample. The samples were processed within two hours of collection or stored at 2°C –8°C in a dark area when collected from far hard to reach areas. The total coliform count test which is a multiple tube fermentation method was performed to estimate the most probable number (MPN) of coliform organism in 100 mL of water for diagnosis of bacteriological contamination.

The test was carried out by inoculating measured quantities of sample water (1.0, 10, 50 mL) into tubes of double- and single-strength McConkey broth accordingly with bromocresol purple as an indicator and the tubes were incubated at 35°C for 48 hours. The tubes showing gas formation in Durham's tubes along with colour change were considered to be presumptive coliform positive. The results of MPN were interpreted based on McCrady's probability tables from the number of tubes showing acid and gas to define the sample as excellent, acceptable, unacceptable & grossly polluted [15].

From the positive presumptive tests, Eijkman's test was performed by incubating subcultures at 37°C in lactose bile broth and the other subculture at 44°C in tryptophan broth. The presence of coliform bacilli was confirmed by the production of gas from lactose at 37°C, and that of E. coli was confirmed by the production of gas from lactose and indole from tryptophan at 44°C, followed by subculture on MacConkey agar. All the media and reagents were procured from Himedia Pv. Ltd., Mumbai, India.

### 2.3.2. Determination of Physical Parameters.

The following basic physical parameters were determined in all samples following the procedures described in APHA [14]. Temperature (mercury thermometer, pH (SYSTRONICS, Digital pH meter 335) & turbidity (DesiCHEK plus) were measured at sites of sample collection. Turbidity of the samples was determined using a turbidity meter in the laboratory.

### 2.4. Data Analysis.

The data were recorded, organized, and summarized in a Microsoft excel sheet. Data analysis was carried out by SPSS version 26.

## 3. RESULTS

### 3.1. District wise distribution of 157 water samples:

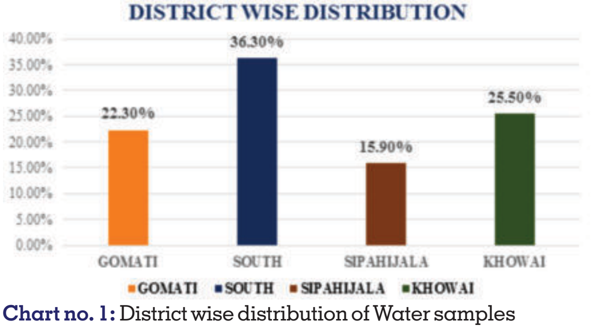


**Chart no. 1:** District wise distribution of Water samples

Highest numbers of samples were received from South District (57/157) followed by Khowai (40/157), Gomati (35/157) and Sipahijala (25/157).

### 3.2. Source Wise Distribution Of 157 Water Samples:

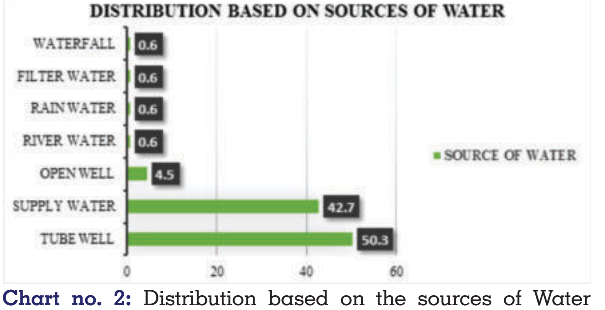


**Chart no. 2:** Distribution based on the sources of Water samples

Distribution based on the sources of water samples showed that water samples from tube well consisted 50.3% (79/157), supply water 42.7% (67/157), open well 4.5% (7/157) and 0.6% (1/157) each waterfall, filter water, rain water and river water.

### 3.3. Physical parameters Assessment of 157 water samples:

**Table no. 1: Gross Appearance of 157 water samples**

| | Frequency | Percent |
|---|---|---|
| CLEAR | 132 | 84.1% |
| TURBID | 25 | 15.9% |
| Total | 157 | 100.0% |

**Table no. 2: Temperature & pH Assessment of 157 water samples**

| Temperature (celsius) | pH |
|---|---|
| • Mean: 26.5 | • Mean: 7.5 |
| • Median: 26.300 | • Median: 7.4 |
| • Mode: 26.2 | • Mode: 7.1 |
| • Std. Deviation: 1.2905 | • Std. Deviation: .4714 |
| • Minimum: 17.2 | • Minimum: 6.0 |
| • Maximum: 30.1 | • Maximum: 9.0 |

### 3.4. Bacteriological Assessment of Samples:

In this study, 93% (146/157) and 46% (67/146) of the water samples were tested positive for total coliform (TC) and faecal coliform (FC)/ thermotolerant E. coli respectively. Therefore, the proportion of Faecal Coliform positive water samples from all four districts was 67/157 (43%).

**Table no. 3: Positivity of Total coliform Test**

| | Frequency | Percent (%) |
|---|---|---|
| POSITIVE | 146 | 93.0 |
| NEGATIVE | 11 | 7.0 |
| Total | 157 | 100.0 |

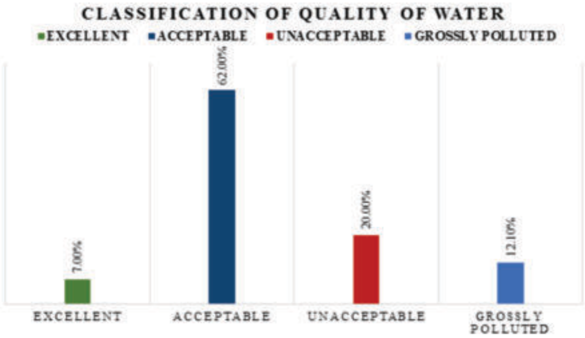


**Chart no.3:** Percentage distribution of Quality of 157 water samples.

Out of 157 water samples tested by total coliform count/MPN method, 97/157 (62%) were found to be acceptable, 31/157 (20%) unacceptable, 18/157 (11%) grossly polluted and 11/157 (7%) were excellent.

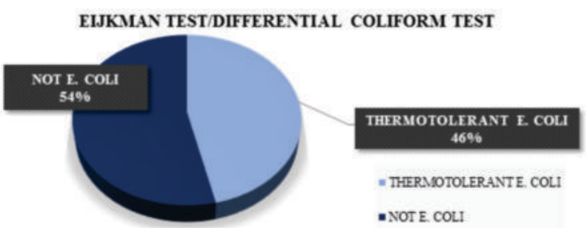


**Chart no. 4:** Positivity of Eijkman Test/ Differential coliform test of 146 coliform positive samples for Thermotolerant E. coli.

67/146 (46%) were positive for Thermotolerant E. coli/ Eijkman Test (faecal coliform) and 79/146 (54%) were negative in Eijkman Test.

### 3.5. District Wise Positivity Rate:

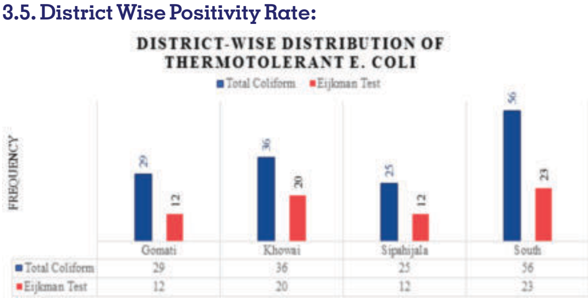


**Chart no. 5:** District wise frequency distribution of Total coliform positivity and Eijkman test positivity

Positivity for Thermotolerant E. coli by Eijkman test was highest from South district 23/146 (16%) followed by Khowai 20/146 (14%), and 12/146 (8%) each from Gomati and Sipahijala districts.

## 4. DISCUSSION

Waterborne diseases have been persistent public health challenges in Tripura. There is a high risk of drinking water pollution in all districts of Tripura, especially the rural areas. This study assessed the quality of water samples from the sources of collection from 4 nearby districts of Tripura. The detection of faecal coliform of 43% (67/157) is a strong evidence for faecal contamination, while 20% unacceptable and 12.1% grossly polluted high level of total coliform implies inadequate chlorination. Almost all water samples (93%) for total coliforms and 43% of the samples for faecal coliforms, respectively, failed to comply with the WHO and national guidelines for drinking water quality.

In the present study, 12.10% of the samples were found to be grossly polluted similar to the finding of a recent study Bhattacharya H. et al conducted in AGMC & GBP Hospital.[10] However, this finding is lower than the finding of Mukhopadhyay C et al. in Karnataka, where he found 22.7% grossly polluted. 93% of our study water samples were positive in Total coliform test which is in accordance with the finding of Mukhopadhyay C et al. study (92.5%).[16] The present study found that 32.1% of the water samples were unfit for human consumption which is lesser than the finding of Malhotra S et al. (42.9%). [17] All these variations may be attributable to the quality of construction of the sources and local practices of water sanitation and hygiene.

## 5. CONCLUSIONS

Bacterial drinking water quality in 4 chosen districts of Tripura fails to comply with WHO guidelines.

i. The most important risk factors for water quality deterioration in rural areas of Tripura include:
 a) Frequent breakage in the distribution line,
 b) Poor waste disposal system,
 c) Agricultural activities that use pesticide and herbicide, and
 d) Unhygienic water handling practices at the household level.

ii. In order to reduce waterborne health burden, the block development administration in cooperation with responsible departments and the community should take immediate actions such as:
 a) Proper chlorination,
 b) Regular inspection and maintenance of the waterline,
 c) Environmental sanitation,
 d) Limiting excessive use of agricultural inputs, and
 e) Education and awareness of the community on how to properly handle drinking water at the household level.

iii. The presence of water-quality indicator bacteria (thermotolerant E. coli) might be associated with poor waste disposal systems and management of water sources. Therefore, there is a need to design an efficient waste disposal system and a catchment area management system around the water sources of rural areas of Tripura.

iv. In addition, regular drinking water-quality assessment of the source, main distribution tanks, distribution systems, and pipes should also be employed to ensure that the water is safe for human use.

v. The present study was limited to assessing bacteriological and physical parameters (only temperature, pH and turbidity). Chemical analysis of water samples should also be considered in future studies in collaboration with other necessary departments. Also, samples from all the 8 districts should be assessed in future studies. In addition, further study should consider additional water-quality parameters such as heavy metals and their source.

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