

Original Research Article

Bacteriological assessment of drinking water sources in an institutional campus in Northeast India using the most probable number method

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ABSTRACT

Background: Safe drinking water is essential for preventing water-borne diseases and maintaining public health. Microbial contamination of drinking water remains a concern, and coliform bacteria are widely used as indicator organisms of microbiological safety.

Methods: A laboratory-based cross-sectional study was conducted in May 2023 at the Regional Institute of Paramedical and Nursing Sciences (RIPANS), Aizawl, Mizoram. Forty-two drinking water samples were collected aseptically from hostels, academic blocks, administrative offices, library and canteen areas. Samples were analyzed using the Most Probable Number (MPN) technique for coliform detection. Positive samples were further examined by culture on Eosin Methylene Blue (EMB) agar and Gram staining.

Results: Of 42 water samples, 8 (19.0%) were positive for coliform organisms and 34 (81.0%) showed no bacteriological contamination. Positive samples showed gas production in lactose broth, green metallic sheen colonies on EMB agar and Gram-negative rod-shaped bacteria consistent with *Escherichia coli*. Contamination was detected in hostels, academic blocks, canteen, library and administrative areas.

Conclusions: Detection of coliform organisms indicates possible fecal contamination in parts of the institutional water supply system. Regular microbiological surveillance, maintenance of water purification systems and periodic sanitation of storage facilities are needed to support safe drinking water in institutional settings.

Keywords: Coliform bacteria, Drinking water quality, *Escherichia coli*, Water contamination

INTRODUCTION

Access to safe drinking water is a fundamental requirement for maintaining human health. Contaminated drinking water is associated with the transmission of infectious diseases and remains an important public health concern.¹ Fecal contamination of drinking water has been documented in different settings in India, including community and rural water sources.² Microbiological contamination is an important indicator of drinking water safety, and studies have reported bacterial contamination, including *Escherichia coli*, in

drinking water sources.³ The presence of fecal contamination in drinking water is a recognized environmental health risk, particularly in low- and middle-income settings.⁴

Systematic evidence has demonstrated the widespread occurrence of fecal contamination of drinking water in low- and middle-income countries.⁵ Coliform bacteria, particularly *Escherichia coli*, are widely used as indicator organisms for assessing the microbiological safety of drinking water.^{6,7} The microbiological quality of water is also relevant to the occurrence of water-borne disease,

making routine assessment of drinking water an important preventive measure.^{8,9}

The Most Probable Number (MPN) technique is a multiple-tube fermentation method used for estimating coliform organisms in water samples, and standard methods describe its use for microbiological examination of water.¹⁰ For drinking water intended for human consumption, national drinking-water specifications and international guidance emphasize microbiological safety and appropriate monitoring.^{11,12} Assessment of drinking-water quality requires appropriate microbiological parameters and standardized testing approaches.¹³ MPN-based qualitative and quantitative assessment has also been described in studies of drinking water, supporting its use for bacteriological evaluation.¹⁴ Basic microbiological principles underlying culture and identification of bacteria provide the laboratory basis for such assessment.¹⁵

Institutional campuses such as educational institutions rely on centralized water supply systems, storage tanks and purification units to provide drinking water to a large number of individuals. Failure or inadequate maintenance of these systems may lead to microbial contamination; therefore, periodic monitoring is important for protecting students and staff. The present study was conducted to assess the bacteriological quality of drinking water sources within the campus of the Regional Institute of Paramedical and Nursing Sciences (RIPANS), Aizawl, Mizoram.

METHODS

Study design and setting

A laboratory-based cross-sectional study was conducted in May 2023 to evaluate the bacteriological quality of drinking water sources within the campus of the Regional Institute of Paramedical and Nursing Sciences (RIPANS), Aizawl, Mizoram, India. RIPANS is located at Zemabawk on the outskirts of Aizawl city and provides educational and healthcare services to a large population of students, staff members, and visitors.

Sample collection

A total of 42 drinking water samples were collected from different locations within the institutional campus, including boys hostels, girls hostels, academic blocks, administrative block, library, and staff canteen.

Water samples were collected aseptically in sterile screw-capped containers using clean gloves to prevent contamination. Prior to sample collection, water taps were allowed to run for 1-2 minutes to remove stagnant water from the pipelines. Any external attachments such as filter screens or aerators were removed before collection to avoid interference with the results.

Approximately 100 mL of water was collected from each sampling point. The containers were immediately sealed and transported to the microbiology laboratory for analysis. Samples were processed as soon as possible, preferably within 5 hours of collection. If immediate processing was not feasible, samples were stored at 2-6°C and examined within 24-30 hours.

Bacteriological analysis

The bacteriological quality of water samples was assessed using the Most Probable Number (MPN) technique, which is recommended by the American Public Health Association (APHA) for detection of coliform bacteria in drinking water. The MPN method is a multiple-tube fermentation technique that estimates the concentration of coliform organisms in water samples through three sequential stages presumptive test, confirmatory test and completed test.

Presumptive test

The presumptive test was performed using lactose broth containing inverted Durham tubes for detection of gas formation. Nine tubes of lactose broth were arranged in three sets, each set containing three tubes. The tubes were sterilized by autoclaving at 121°C for 15 minutes. The water samples were inoculated as follows: 10 mL of water sample into the first set of tubes, 1 mL of water sample into the second set of tubes, and 0.1 mL of water sample into the third set of tubes. All inoculated tubes were incubated at 37°C for 24 hours and observed for gas production and turbidity. Tubes that did not show gas formation after 24 hours were further incubated for an additional 24 hours and re-examined. The presence of gas in Durham tubes within 48 hours was considered a positive presumptive test indicating possible coliform contamination.

Confirmatory test

Samples showing positive presumptive results were subjected to confirmatory testing. A loopful of culture from positive lactose broth tubes was streaked onto Eosin Methylene Blue (EMB) agar plates. The plates were incubated at 37°C for 24 hours and examined for characteristic colonies. Colonies showing a green metallic sheen were considered indicative of *Escherichia coli*, a common indicator of fecal contamination.

Completed test

For further confirmation, colonies suspected to be coliform organisms were subjected to Gram staining. Smears were prepared from isolated colonies, heat-fixed, and stained using the Gram staining method. The stained slides were examined under a microscope for the presence of Gram-negative rod-shaped bacteria, which are characteristic of coliform organisms.

Interpretation of results

The number of positive tubes in each set of dilutions (10 mL, 1 mL, and 0.1 mL) was recorded and compared with standard MPN statistical tables to estimate the most probable number of coliform organisms in the water samples. Water samples showing the presence of coliform organisms were considered microbiologically contaminated, whereas samples without coliform detection were considered bacteriologically safe for consumption.

Data analysis

The collected data were summarized using descriptive statistics, including frequencies and percentages. The distribution of coliform contamination across different sampling locations within the campus was analyzed.

RESULTS

A total of 42 drinking water samples were collected and analyzed. Out of these, 8 (19%) samples were found to be positive for coliform organisms while 34 (81%) samples showed no bacteriological contamination. Coliform positive samples demonstrated gas production in lactose broth and characteristic green metallic sheen colonies on EMB agar. Gram staining confirmed gram-negative rod-shaped bacteria consistent with *Escherichia coli*.

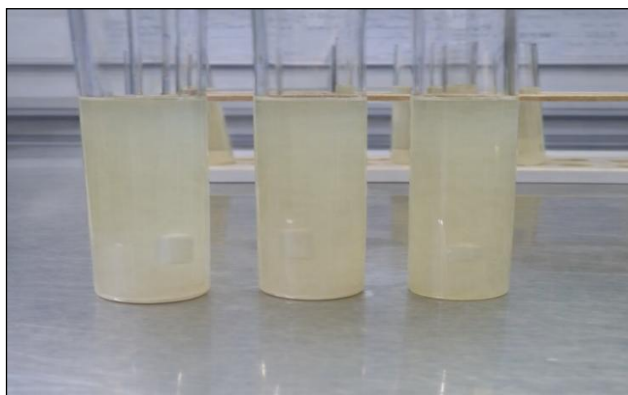


Figure 1: Positive water culture sample in lactose broth showing formation of gas in Durhams tube.

The distribution of contamination varied across the sampling locations. Among the eight samples collected from the boys hostel, 2 (25.0%) were coliform positive, while 6 (75.0%) were negative. In the girls hostel, 1 of 7 samples (14.3%) was positive and 6 (85.7%) were negative. Of the 10 samples collected from academic blocks, 2 (20.0%) were positive and 8 (80.0%) were negative. The canteen yielded 1 positive sample among 5 tested (20.0%), with 4 (80.0%) negative samples. Similarly, 1 of 4 library samples (25.0%) was positive, while 3 (75.0%) were negative. In the administrative block, 1 of 8 samples (12.5%) was positive and 7 (87.5%) were negative.

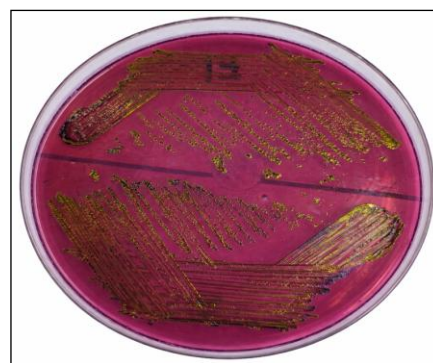


Figure 2: EMB agar showing growth of Coliform bacteria (*Escherichia coli*).

Thus, coliform contamination was detected at all six categories of sampling locations, with the highest proportion of positive samples observed in the boys hostel and library (25.0% each), followed by the academic blocks and canteen (20.0% each).

DISCUSSION

The present study evaluated the bacteriological quality of drinking water sources within an institutional campus using the Most Probable Number (MPN) method. The findings revealed that 19% of the drinking water samples were contaminated with coliform organisms, indicating possible fecal contamination in certain parts of the campus water supply system. The isolation of *Escherichia coli* on Eosin Methylene Blue agar further supports the presence of fecal contamination and highlights potential public health risks associated with consumption of contaminated drinking water.

Coliform bacteria are widely used as indicator organisms for assessing the microbiological safety of drinking water because their presence indicates possible contamination by human or animal fecal matter.^{6,7} Drinking-water standards emphasize microbiological safety, and water intended for human consumption should be free from coliform organisms in the specified test volume.¹¹

The detection of contamination in the present study therefore highlights the need for improved monitoring of institutional water supply systems.^{12,13} Similar findings have been reported in other Indian settings. Ramakrishnan et al reported contamination of drinking water sources in rural India, while Gupta et al reported an association between microbial contamination of drinking water and diarrheal disease.^{16,17} Institution-based studies have also reported comparable findings. Sidhu et al identified coliform organisms in drinking water samples from schools in northern India, while Kanth et al reported elevated MPN values in institutional water sources.^{18,19}

The contamination rate observed in the present study (19%) is comparatively lower than that reported in several community-based studies from other regions of

India. However, detection of contamination even in a limited number of samples is important in institutional environments where large populations rely on common drinking water sources.

The presence of contaminated samples in hostels, academic blocks and canteen areas suggests that microbial contamination may occur at different stages within the water supply system. Possible contributing factors include inadequate chlorination, poorly maintained filtration units, contamination of overhead storage tanks, leakage in water distribution pipelines, and secondary contamination during handling or storage.^{8,9}

Overall, the findings emphasize the importance of regular microbiological surveillance of drinking water sources, proper maintenance of water purification systems, and periodic cleaning and disinfection of water storage tanks within institutional campuses.²⁰

Limitations

This study has few limitations. The study was limited by a relatively small sample size and was conducted within a single institutional campus. Further large-scale studies including seasonal variation in water quality would provide more comprehensive data.

CONCLUSION

A proportion of drinking water sources within the RIPANS campus showed bacteriological contamination with coliform organisms. Although the majority of samples were safe, the detection of contamination indicates the need for continuous monitoring of water quality and improved maintenance of purification and distribution systems. The findings of this study emphasize the importance of regular microbiological surveillance of drinking water in institutional settings as a preventive strategy in primary care and community health.

Recommendations

Routine microbiological monitoring of drinking water sources on the campus should be conducted to ensure water safety. Regular maintenance and timely replacement of water purification systems and filters should be ensured. Water storage tanks should be cleaned and disinfected periodically to prevent microbial contamination. Furthermore, health awareness programmes should be implemented to educate students and staff about safe drinking water practices.

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Conflict of interest: None declared

Ethical approval: The study was approved by the Institutional Ethics Committee

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