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Microbiology

MONITORING AND ASSESSMENT OF DRINKING WATER FOR SAFE WATER SUPPLY IN RURAL HARYANA TERTIARY CARE HOSPITAL

Muskaan Garg	Research Assistant, Department of Microbiology, Adesh Medical College and Hospital, Shahabad, Haryana.
Mahenaz Khan	Tutor, Department of Microbiology, Adesh Medical College and Hospital, Shahabad, Haryana.
Dr. Swati Mittal	Assistant Professor, Department of Microbiology, Adesh Medical College and Hospital, Shahabad, Haryana.
Dr. Vanita Mittal*	Assistant Professor, Department of Microbiology, Adesh Medical College and Hospital, Shahabad, Haryana. *Corresponding Author
Dr. Ashwini Manhas	Associate Professor, Department of Microbiology, Adesh Medical College and Hospital, Shahabad, Haryana.
Dr. KC Rathish	Professor and Head, Department of Microbiology, Adesh Medical College and Hospital, Shahabad, Haryana.

ABSTRACT **Background**-The aim of our study is to determine the drinking water quality monitoring and assessment of safe water supply in tertiary care hospital. Water samples which are collected from different sites of Adesh Medical College and Hospital, Shahabad, Haryana were processed.

Material and methods- Most Probable Number (MPN) test was done to detect the coliform bacteria in drinking water, samples were collected from different sites of AMCH, bacterial isolate was identified by culture, colony morphology, Gram's staining and biochemical characterization of bacteria.

Results- Out of 40 samples of drinking water 7(17.5%) were positive in which coliform bacteria were present, while 33(82.5%) were negative. In which 1(14.2%) of the total water samples were contaminated with mix growth organism, 1(14.2%) harbored *Klebsiella sp.*, while 5(87.5%) among 7 water samples were highly contaminated with *E.coli* species. Out of 7 positive samples, 4(57.1%) water sample were satisfactory, 1(14.2%) were unsatisfactory sample and 2 (28.5%) were suspicious samples.

Conclusion- On the basis of the results obtained, the quality analysis revealed that most of water samples were satisfactory and were not contaminated with coliforms and various pathogenic bacteria. The water cooler samples showing positive results have *E.coli* while less of *Klebsiella* species. Therefore, we suggest that all the water sources of drinking water should be properly sanitized, continuously monitored and bacterial load should be estimated on daily basis.

KEYWORDS : Drinking water quality, Most Probable Number, coli

INTRODUCTION

Water is the natural resource crucial to sustain life on earth. For maintaining a healthy life water is required in revival and cells composition [1]. Water is an inorganic blend mainly used for household purposes, essentially for drinking. Since the demand is increasing, not only economic expansion and social welfare is influenced by the availability of fresh clean water, it more dominantly incites the public health [2]. Despite this, drinking water is more likely to be polluted, posing burden on human species.

In Ethiopia over 60% of the communicable diseases are due to poor environmental health conditions arising from unsafe and inadequate water supply [4]. Viruses, pathogenic microorganisms and parasites primarily spread via water to humans. Moreover, it visually looks acceptable in taste and looks but may cultivate in human intestinal tract. Hence, it may not be safe to drink and use. However, microbial survival is observed in both ground and surface water which may cause chronic sickness in humans if not treated immediately [3]. Thus, coliform count, act as index for bacteriological quality of water.

Generally microbes are present in small numbers that they can escape detection. Therefore, it is essential to test fecal pollution. Perhaps, if the water sample is detected with fecal pollution, it is inferred that water sample might harbor enteric pathogens. The presence of coliform bacteria is present in high dilutions in feces of human beings and other warm blooded animals. [12]

Widespread risk to human health is present, as 80% of drinking water contain microbes such *Shigella spp., Salmonella spp, Yersinia entercolitica, Escherichia coli, Campylobacter spp, Vibrio cholerae, Pseudomonas spp.,* when contaminated. Some of the viruses and parasites are even found in water which includes *hepatitis A, hepatitis E*, *Giardia spp and Entamoeba histolytica* [6,7].

Among them, *Escherichia coli* are predominant member of facultative anaerobic portion of human colonic normal flora [8]. The outbreak of infections from *Ecoli* caused significant morbidity and mortality [9]. With some exceptions, its presence denotes faulty sanitation implementations in food and water processing facilities [10, 11]. *E.coli* 0157:H7,a fatal strain is known to be contagious to drinking water. *E. coli* is considered a more explicit indicator of fecal foulness than other coliforms. These bacteria may cause major human diseases such as diarrhea, cholera, typhoid fever, poliomyelitis in the hospitals and communities [5]. Other bacteria are also sometimes used as indicators of fecal pollution. These include fecal *streptococci* and *Clostridium perferingens.*[12]

Since, safe drinking water is the very crucial for public health; its microbial contamination level has to be checked regularly. Therefore, in this study we aimed at bacterial quality check of water samples from different sites in Adesh Medical College and Hospital, Shahabad, in order to detect the level of contamination and improve the sanitary conditions as desirable using MPN method.

MATERIAL AND METHODS

The study was conducted in Adesh Medical College and Hospital, Shahabad, Department of Microbiology and Community Medicine.

Design of study and period

For accessing the bacterial containment level in the drinking water the study was conducted from December end 2020 to February 2021 over a period of 2 months in Adesh Medical College and Hospital, Shahabad.

Sample collection

Sterilized bottles with 0.1 ml of freshly prepared 18% aqueous sodium thiosulphate in each bottle are used for water sample collection [13]. Before collection of water sample, tip of the taps were sterilized with cotton wool, soaked in 70% ethanol, and the tap was allowed to run for two minutes and then water sample was carefully collected in 250ml

sterile capped bottle. The sample bottle was neatly labeled, including time, collection site name. The sterile 250 ml container was directly transported for testing [14].

Sample size

Water samples were collected from water coolers, drinking water taps and RO systems from different locations in the college campus. The Table -1 represents the different points of sample collection.

Points of sample collection	Number of sample
Boys hostel	3
Girls hostel	3
Hospital	8
Medical College	6
Rural health centre under college	1
Urban health centre under college	1
Campus Residential Areas (CRA)	
CRA (block B)	6
CRA (block C1)	6
CRA (block C2)	6

Total coliform determination

In order to determine the coliform count in water sample the following three main protocols were processed, presumptive, confirmed and completed.

Presumptive coliform test (multiple tube method)

Presumptive coliform count –multiple tube test

To detect the indicator organisms, total and fecal coliform was carried out by bacteriological analysis with the help of most probable number (MPN),

***Procedure*:**

Measured amounts of sample of water are added by sterile graduate pipette as follows:-

1. 50 ml of water sample is added in 50 ml of Double Strength MacConkeys medium.
2. 10 ml of water sample is added in 5 tubes/bottles containing 10 ml of Double Strength MacConkeys medium.
3. 1 ml of water sample is added in 5 tubes/bottles containing 5 ml of Single Strength MacConkeys medium.

Then 24hours to 48hours of incubation was given at 37□ C for evaluation of fecal and total coliforms. The indication was observed by the entrapment of gas in durham's tube while production of acid and anaerobic bacterial growth specify change in color of broth from reddish purple to yellow. Hence, the statistical analysis i.e the MPN no was predicted by McCrady's probability table.

Confirmed test

For further analysis, a loopful of positive samples culture were transmitted to a tube containing Brilliant Green Lactose Bile Broth along with durham's tubes so to confirm the presence of susceptible organisms. The inoculated tubes were incubated at 37 □ C for 48 hours and observed for entire coliforms and gas production.

Completed test

As per the WHO 2012, an administration of complete test is done by streaking loopful of above positive broth into Eosin Methylene Blue agar and MacConkey Agar plate for isolation of pure colonies. The growth was observed after the incubation period of 24 hours at 37□C. Further, by using morphology, biochemical tests and culture characteristics, isolated colonies were differentiated and categorized as coliforms or *Ecoli* (fecal coliforms). The green metallic sheen on the EMB culture plate, while pink color colony was determined on MacConkey Agar [15,16].

Coliform count determination

Finally, the number of positive test tubes with yellow coloration and gas production were matched with McCrady's (MPN of coliform per 100 ml of sample) table for predicting the nature of the water sample present in the area.

RESULTS

Total of 40 samples were collected from various points of drinking water coolers, water taps and RO systems located at different sites of the college and hospital, tested by MPN method. Out of 40 samples of drinking water 7(17.5%) were positive in which coliform bacteria were present, while 33(82.5%) were negative which lacked the bacterial growth. Among all these samples including Hospital, Boys hostel, Girls hostel, Medical College, residential apartments, Rural and Urban health centre under college, the highest degree of bacterial load were observed in Hospital, Girls and Boys hostels as shown in the Table 1 and Figure.1.

Table 1- Represents distribution of positive and negative samples according to the sites respectively.

S.No.	Sample Collection sites	Number of positive samples	Number of negative samples	Total no of samples
1	Boys hostel	2	1	3
2	Girls hostel	1	2	3
3	Hospital	2	6	8
4	Medical College	0	6	6
5	Rural health centre under college	1	0	1
6	Urban health centre under college	0	1	1
7	Campus Residential Areas (B, C1,C2)			
A	BLOCK B	0	6	6
B	BLOCK C1	1	5	6
C	BLOCK C2	0	6	6
	Total samples	7(17.5%)	33 (82.5%)	40

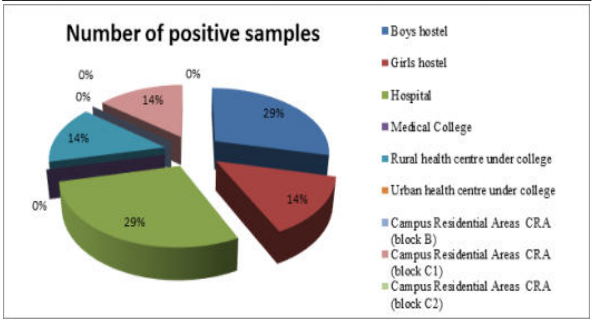

Figure.1- Site wise distribution of the positive samples is depicted respectively.

In our study, 1 (14.2%) of the total water samples were contaminated with mix organism growth, 1(14.2%) contained *Klebsiella sp.*, while 5(85.7%) among 7 water samples were highly contaminated with *E.coli* showing in Table-2. The percentage profile of isolated organisms is depicted in Figure 2.

Table 2- Distribution of organisms isolated from different positive samples

S .No.	Sample collection sites	Isolated organisms from the contaminated samples			
		Ecoli	*Klebsiella* species	*Pseudomonas aeruginosa*	*Mix organism (Ecoli+ Klebsiella)*
1	Boys hostel	2	0	0	0
2	Girls hostel	1	0	0	0
3	Hospital	0	1	0	1
4	Medical College	0	0	0	0
5	Rural health centre under college	1	0	0	0
6	Urban health centre under college	0	0	0	0
7	Campus Residential Areas (B, C1,C2)				
A	BLOCK B	0	0	0	0
B	BLOCK C1	1	0	0	0
C	BLOCK C2	0	0	0	0
	Total samples	5(85.7%)	1(14.2%)	0	1 (14.2%)

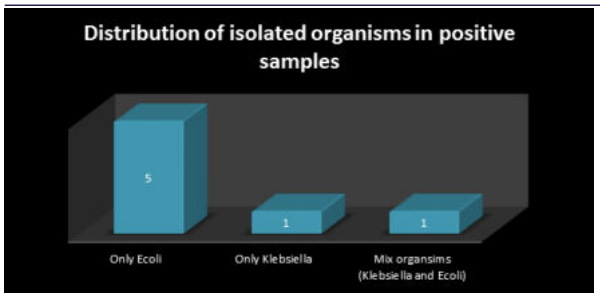

Figure.2- Shows the distribution of isolated organisms in positive water samples respectively.

Table 3- Quality of total positive samples accordingly, where n=7

S. No.	Sample collection sites	Count of positive samples	count of satisfactory samples	count of suspicious samples	count of unsatisfactory samples
1	Boys hostel	2	1	1	0
2	Girls hostel	1	1	0	
3	Hospital	2	1	1	0
4	Medical College	0	0	0	
5	Rural health centre under college	1	0	0	1
6	Urban health centre under college	0	0	0	0
7	Campus Residential Areas (B, C1,C2)				
A	BLOCK B	0	0	0	0
B	BLOCK C1	1	1	0	0
C	BLOCK C2	0	0	0	0
	Total samples	7	4(57.1%)	2(28.5%)	1(14.2%)

Therefore, showing the satisfactory nature of 4 (57.1%) water samples, suspicious were 2 (28.5%) and unsatisfactory quality of 1(14.2%) samples among all the positive count acquired. The following results are depicted in Table 3.

DISCUSSION

Utilization of polluted drinking water can be responsible for a large number of water borne diseases and chronic health problems. In most developing countries, the insufficiency of pure and secured drinking water distribution is the most frequent cause of gastroenteritis. The water needs to be chemical and pathogen free, must be satisfactory for human ingestion and also contain suitable taste. Therefore, potable water becomes the immediate reservoir of transmittable diseases. Hence, water decontamination is necessarily required to secure public health.

In this study, based on the obtained results of bacteriological coliform quality, from different sites of the Hospital and medical college campus, drinking water cooler samples that collected from girls hostel, boys hostels, residents, hospital and rural health centre were satisfactory. Along with this the drinking water of boys hostel and hospital shows suspicious outcomes as well, while rural health centre shows unsatisfactory results as compared to other samples.

Escherichia coli was highly and very frequently detected in the water cooler samples of boys hostel in comparison to residential sites, rural health centre and girls hostel. Moreover, mix organisms were also spotted, but were less in comparison to other specific organisms.

Since, *Ecoli* were observed more often, the washing, cleaning and regular filter replacement became necessary as per the guidelines.

On comparing the study of Shariq.et al. (2016) in which, 22 water samples were collected from different sources and among them, 11(50%) were Satisfactory, 5(22.72%) Suspicious and 6(27.28%) were unsatisfactory. While in our study out of 7 positive samples 4(57.1%) were satisfactory, 2(28.5%) were suspicious and 1(14.2%) was unsatisfactory.[17]

Ecoli and *Klebsiella* were most common isolates. Both the organisms are considered as an indicator of water pollution [17].Although, in the similar study of Ezeugwunne et al., the majority of isolated bacteria were *E.coli* (66.2%) in the water samples while others contain *Klebsiella species*[18,19]. Comparing with our study the results depicted the Ecoli and Klebsiella presence to be 87.1% and 14.2% respectively Over the decades, the fecal contamination is determined by E.coli acting as indicator organism. The fact that E.coli has short life span in water, it is however, the foremost bacterium for detection of contamination and gastric diseases in humans [20].

CONCLUSION

In case of, bacteriological water quality regulation, the coliform testing has been successful for over fifty years. As stated by WHO (2002) the level of coliform bacteria intended for drinking in a water sample should be zero in 100ml. Hence, our study provides the base line information that, from the obtained results the microbiological parameters of most of the collected water samples from different sites of AMCH met the WHO approved standards. But, few were unable to meet the recommended standards.

The quality analysis revealed that most of water samples were satisfactory and were not contaminated with coliforms and various pathogenic bacteria. The water cooler samples showing positive results have high number of *E.coli* while less of *Klebsiella* species. Among them, the occurrence of *E.coli* majorly indicates the bacteriological quality of water as well as it becomes the most important indicator organism for fecal pollution. Therefore, a pollution free water supply in the hospital and rural care centers is necessary.

So, we suggest that all the water sources of drinking water should be properly sanitized, continuously monitored and bacterial load should be estimated on daily basis. Hence, water must pass three primary steps i.e storage, disinfection and filtration, which usually eliminates 90-95% of bacterial as well as physical contamination and provides potable water for drinking.

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