



BACTERIOLOGICAL STUDY OF DRINKING WATER : A STUDY FROM RURAL NORTH INDIA.

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ABSTRACT **BACKGROUND:** The routine monitoring and assessment of the microbiological quality of water is the key priority for both water suppliers and surveillance agencies. Microbiological quality is of principal concern because of the acute risk to health posed by viruses, bacteria and helminths in drinking-water. Therefore, monitoring and assessment of drinking-water is primarily a health-based activity which emphasises the protection of public health through ensuring that the water supplied is of a good quality.

AIM: The aim of this study is to analyze the bacteriological quality of the available drinking water collected directly from various sources, namely, water source supply outlets, as well as drinking water sources from different areas of Kangra district received in the Department of Microbiology of Dr.Rajendra Prasad Government Medical Collage & Hospital Kangra at Tanda.

METHODS: A total of 274 water samples from different sources received from april 2016 - may 2019. The bacteriological analysis of water was done by the multiple tube technique. Results were interpreted after 48 hours of incubation of the water sample in MacConkey bile broth medium in accordance with Mc Crady probability table.

RESULTS: Out of a total of 274 water samples were received in the Microbiology laboratory 5 (1.85%) sample were rejected due to improper transport, 112(41.63%) were unsatisfactory, 13(4.8%) suspicious, 117(44%) were satisfactory and 27(10 %) were excellent. Nearly 5.5% samples from water tanks, 16.7% from public taps were unsatisfactory but water samples from all the aqua guards were excellent. *Escherichia coli* (71%) was the commonest isolate followed by, *Citrobacter freundii*(24%), *Klebsiella pneumoniae* spp.(3.6%) and *Pseudomonas aeruginosa* (1.4%).

CONCLUSIONS: Bacteriological assessment of drinking water is essential and should be carried out on regular basis so as to prevent outbreaks of water borne diseases.

KEYWORDS : Water Bacteriology, Coliforms, Escherichia Coli, Multiple Tube Technique

INTRODUCTION

Water is essential to life and access to safe drinking water is essential for health. Drinking water is defined as water intended for human consumption for drinking and cooking purposes from any source. The current WHO bacteriological guidelines for drinking water recommend zero faecal coliforms for 100ml of water.^{1,2}

According to WHO, globally approximately 1.1 billion drink unsafe water¹ it is estimated that nearly 88% of diarrhoeal diseases are attributed to unsafe water, sanitation and hygiene. It is well established that infectious diseases are transmitted primarily through water supplies contaminated with human and animal excreta, particularly faeces³. In India, 37.7 million people suffer due to waterborne diseases annually and nearly 1.5 million children die due to diarrhoea alone¹. Main bacteria that can be transmitted through the faeco-oral route, include *Salmonella spp.*, *Shigella*, pathogenic *Escherichia coli*, *Yersinia enterocolitica*, *Campylobacter spp* etc. Other organism include viruses such as *Hepatitis A* and *E. rotavirus, poliovirus*, and parasites such as *Entamoeba histolytica* and *Giardia spp*. Waterborne pathogens like *Vibrio cholerae*, *Enteropathogenic E.coli*, *Salmonella spp* and *Hepatitis E virus* may cause outbreaks and have a high mortality rate.^{3,4}

For public and environmental health protection, it is mandatory to provide safe drinking water. Even with well-operated drinking-water treatment systems, there is growing concern that aging drinking water distribution systems (DWDSs) are vulnerable to higher rates of mains

breaks/repairs and related pressure losses that may lead to pathogen intrusion scenarios. A key realization is the need for ongoing system-wide vigilance, coupled with a preventative rather than just responsive management approach.^{5,6}

MATERIALS AND METHODS:

A retrospective analysis was done for water sample received in the Department of Microbiology, Dr.Rajendra Prasad Medical College, Kangra at Tanda for the assessment of bacteriological quality of water from various water sources in the Kangra district. From January 2016 to December 2018, a total of 274 water samples from various water sources in and around Kangra were received and processed in the Department of Microbiology, in accordance with WHO and the Indian Council Medical Research (ICMR) guidelines.^{5,7,8} Taking all aseptic precautions 100 millilitres of water samples from each source were collected in sterile glass stopper bottles for microbiological examination as per guidelines⁸. Water samples containing residual chlorine were neutralized by adding presterilized 0.1 mL sodium thiosulphate (1.8% w/v) per 100 mL of water sample. The samples were stored at 2°C–8°C in the refrigerator to avoid changes in bacterial count until sample was processed far away sources the water sample were transported in a vaccine carrier from point of collection to the laboratory with maintain the cold chain. The total coliform count test was based on the multiple tube technique method to estimate the most probable number (MPN) of coliform organism in 100 mL of water for diagnosis of bacteriological contamination¹¹. The test was carried out

by inoculation of measured quantities of sample water (5, 10, 50 mL) into tubes of double and single-strength MacConkey lactose bile salt broth with bromocresol purple as an indicator. The water sample were incubated for 48 hrs at 37 °C. The results of MPN were interpreted based on McCrady probability tables from the number of tubes showing acid and gas (fermentation by coliform organisms) to define the sample as excellent, satisfactory or unsatisfactory. Differential coliform count (Eijkman's test) was performed by incubating subcultures from the positive presumptive tests at 44°C and 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 Hi-media Pvt Ltd, Mumbai, India. Further the coliforms and other organisms were analysed by subculture on MacConkey agar, biochemical reactions and other identification tests. Colonies from these plates were identified by conventional biochemical methods according to standard microbiological techniques.⁹

RESULTS:

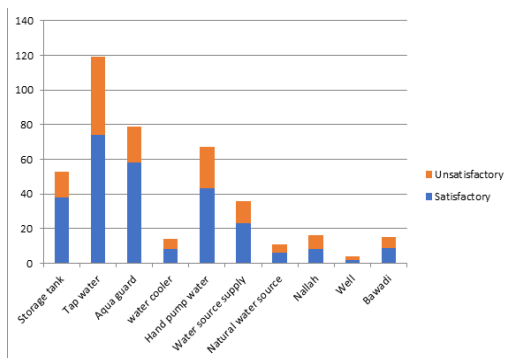
A total of 274 water samples were collected over a period of 3 years i.e. is from april 2016-may 2019 from different water sources in Kangra district .Out of 274 samples, 5 (1.85%) samples were rejected due to improper transport and the rest were processed .Out of total 269 samples, 27(10%) excellent, 117 (44%) were satisfactory, 13(4.8%) suspicious, 112(41.63%) Unsatisfactory.

(Table1) The difference sources of water was as follows comprised Hand pump water 43(15.98%), Storage tank 38 (14.12%), tap water 74 (27.50 %) , Water source supply 23(8.4%), Natural water source 6 (2.23%), Nallah 8 (2.97%), Well 2(0.74%), Bawadi 9 (3.34%) , Aquaguard/ RO 58 (21.6%), Water cooler 8 (2.97%).(Table 1), Fig.1

Table 1.The distribution of different sources of water:

Supply of water	Type /Source	Number (%)
Hospital supply	Storage tank	38 (14.12 %)
	Tap water	74 (27.50 %)
	Aquaguard/RO	58 (21.6%)
	Water cooler	8 (2.97%)
	Total = 178 (66.9%)	
Non hospital supply	Hand pump water	43(15.98 %)
	Water source supply	23(8.4 %)
	Natural water source	6 (2.23 %)
	Nallah	8 (2.9 %)
	Well	2 (0.74 %)
	Bawadi	9 (3.34 %)
	Total = 91 (33.59%)	

Figure 1: Bar chart depicting the water samples from various sources and their outcome



The purpose of water testing for bacteriological examination was divided into 2 main categories: routine water testing and testing during outbreak. Table 2 shows the purpose of water testing.

Table 2. Purpose of water testing

Routine	203
Outbreak	66

The water supply was segregated into Chlorinated and Nonchlorinated.

Table 3. Chlorinated and Nonchlorinated water

Chlorinated	%	Nonchlorinated	%
Storage tank	38 (14.12 %)	Hand pump water	43(15.98 %)
Tap water	74 (27.50 %)	Natural water source	6 (2.23 %)
Aquaguard/RO	58 (21.6%)	Nallah	8 (2.9 %)
Water cooler	8 (2.97%)	Well	2 (0.74 %)
Water source supply	23(8.4 %)	Bawadi	9 (3.34 %)
Total:	201(74.72%)	Total:	68 (25.27%)

Table 4 and fig. 2 shows the quality of water sample tested majority water sample were satisfactory(44%).

Table 4.Classification of water samples

Grade of water sample	Presumptive coliform count/ 100ml	Number(percentage) of water sample
Excellent	0	27(10%)
Satisfactory	1-3	117(44%)
Suspicious	4-10	13(4.8%)
Unsatisfactory	>10	112(41.63%)

Figure 2: Bar chart for classification of water samples

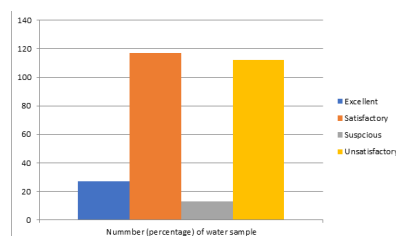
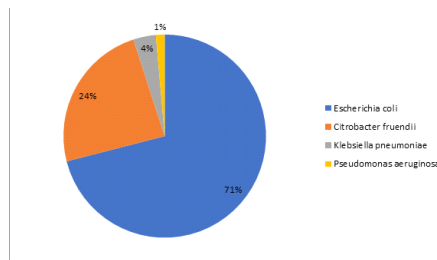


Table 5. Satisfactory and Unsatisfactory water from various sources

	Satisfactory	Unsatisfactory
Storage tank	38	15
Tap water	74	45
Aqua guard	58	21
water cooler	8	6
Hand pump water	43	24
Water source supply	23	13
Natural water source	6	5
Nallah	8	8
Well	2	2
Bawadi	9	6

Figure 3. Bacterial species in different water samples:



Organism identified were *Escherichia coli*(71%),*Citrobacter freundii* (24%),*Klebsiella pneumoniae*(3.6%)and *Pseudomonas aeruginosa*(1.4%).(Figure 3)

DISCUSSION:

Water is essential to sustain life, and every effort should be made to provide satisfactory supplies of drinking water to all. A clean and treated water supply to each house may be the norm in Europe and North America, but in developing countries, access to both clean water and sanitation are not the rule, and waterborne infections are common. Water borne diseases cause nearly one third of intestinal infections worldwide.^{3,4} Globally it has been estimated that lack of safe water, sanitation and proper hygiene measures has led to 40% of total deaths and 5.7% of total disease burden. A significant burden of disease could be prevented in developing countries through access to improved

water sources. These have been defined by WHO as piped water, public taps, tube wells, bore holes, protected springs and rain water collection and guidelines all public water supplies must be tested regularly and be free from any coliforms.¹

The present study of bacteriological assessment of drinking water sources in and around Kangra, revealed 44% of the water samples as unsatisfactory and not desirable for human consumption. Our findings are in contrast to the study of bacteriological assessment of drinking water sources in and around Shimla city which revealed 11.74% of the water samples as unsatisfactory and not desirable for human consumption.^{11,12} It is similar to other studies from North India where 48%, 47.5% and 38.6% of the water samples have been found to be unsatisfactory.^{13,14,15} In this study 5.5% of water tanks and 16.7% of tap water were unfit for human consumption. This may be explained that water tanks are more liable for contamination from leaking sewage lines, sludge, animal droppings, birds and monkey faeces, another reason for our increase rate would be because most of the sample processed in our laboratory had been during or after outbreak. According to WHO, *Escherichia coli* is the most discriminating marker for recent faecal contamination so an indicator of choice for drinking water portability in developed nations.⁵ The detection of coliforms was done by presumptive coliform count and further confirmation of *Escherichia coli* by differential coliform or the Eijkman test. The most common coliform detected was *Escherichia coli* 71% followed by *Citrobacter* 24.6%. which is similar to studies done in North India. Although other coliforms are generally not harmful themselves, they indicate the possible presence of other pathogenic bacteria, viruses and protozoans. A WHO report has stated that more people would die from consuming unsafe drinking water and unsanitary conditions by the year 2020 than would die from AIDS, if steps to improve water quality are not taken.¹⁹ It is a need of the hour to ensure provision of safe drinking water to consumers. A safe and potable water supply is ensured through three stages of storage, filtration and disinfection. Storage is the first step where 90-95% impurities are removed by sedimentation. Active intervention from public health and the health department along with raising people's awareness regarding water hygiene are required for improving the quality of drinking water.^{11,12}

CONCLUSION:

Microbiological quality of drinking water is of principal concern because of the acute risk to health posed by viruses, bacteria and helminths in drinking-water. Therefore, monitoring and assessment of drinking-water is primarily a health-based activity which emphasises the protection of public health through ensuring that the water supplied is of a good quality.

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