ORIGINAL ARTICLE

Bacteriological Analysis of Drinking Water from Services Hospital Lahore and Services Institute of Medical Sciences Lahore

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ABSTRACT

Objective: To evaluate fifty-six water samples for bacteriological contamination in Services Hospital and Services Institute of Medical Sciences (SIMS), Lahore from 15 December 2011 to March 2012.

Materials and method: A total of 56 water samples were collected from water tanks, taps of toilets, kitchens and water coolers of different wards and laboratory of Services Hospital, Lahore. Samples were also collected from taps of all the laboratories, kitchen and toilets of SIMS Pathology Department and student hostels.

Samples were collected in 100ml sterile bottles and brought to the laboratory within two hours of collection. All the samples were tested for bacterial contamination by the Multiple Tube Method to determine the Most Probable Number of "total coliforms" and "faecal coliforms" using the standard procedure.

Results: Coliforms were grown from 16 (28.6%) water samples both for total and faecal coliforms

Key words: Drinking water, bacterial contamination, Multiple tube method, coliforms.

INTRODUCTION

Sanitation and drinking water are universally accepted as essential for human life, dignity and human development. Urban and rural sanitation and drinking water policies have been adopted and published by 63% developing countries in 2011 from 40% in 2009 including Pakistan[1].

An improved drinking water source is one that by nature of its construction adequately protects the source from outside contamination, in particular from faecal matter. In 2010, 89% of the world's population (6.1 billion people) used improved drinking water sources, whereas 92% are expected to have access to the same in 2015. Presently 783 million people (11%) are without access and 605 million will still be without access in 2015[2].

Globally, improving water, sanitation and hygiene has the potential to prevent at least 9.1% disease burden. Diarrohea is caused by ingestion of pathogens as cholera, typhoid, dysentery by faeco-oral route in unsafe drinking-water (globally 80%), contaminated food and from unclean hands. One and half million deaths occur annually mainly in children. In Pakistan the death rate due to water, sanitation and hygiene is 13.6% of the total deaths, whereas diarrohea accounts for 103.3* 10^3 cases[3]. From a literature review on 50 studies and analysis of 38 of these studies showed that 56% reduction in diarrheal disease can be brought about by improving water quality and its supply[4].

Pakistan is the world's sixth most populous country (estimated population of 18.7 million) with 15-18% population having access to safe drinking water[5].

WHO Guidelines for Drinking-water Quality recommend water safety plans, encompassing whole water-supply chain from catchment to consumer. Pakistan is one of the countries where there is no data available of a national policy to develop or implement a water safety plan or preventive risk management[6]. Studies on water are few and that too on the urban closed pipe system. The rural setting is a totally neglected area.

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MATERIAL & METHOD

This study was conducted at the Microbiology Section, Department of Pathology, Services Institute of Medical Sciences, Lahore from 15 December 2011 to March 2012. Fifty- six drinking water samples were collected from Services Hospital and SIMS, Lahore.

Sampling technique for water collection: A total fifty-six water samples were collected; 51 from taps, 4 from water coolers, 01 from distillation plant. They were transported immediately to the Microbiology Laboratory or as soon as possible within 2 hours of collection. Heat-sterilized, screw capped glass sample bottles of 100ml capacity were used. The external surface of the taps were cleaned with alcohol and water allowed to run to waste for 2-3

minutes before running into the bottle. Care was taken to avoid touching the opening with hand. The bottle was stoppard, labelled with full details of water source, time and date of collection and delivered to the laboratory as early as possible, at least within 2 hours of collection, kept protected in a container away from heat and light.

Multiple tube test: 50ml and 10ml volumes of indicator broth, MacConkey Broth Purple at double strength concentration (8%) were placed into 100ml capacity bottle and 20ml capacity test tubes containing an inverted Durham tube. The tubes were sterilized at 121^0 C for 15 minutes in an autoclave. After sterilization, the Durham tubes were checked that it was free from air bubbles. The water was examined for any visible change in colour, transparency or solid /suspended particles or impurities.

The bottle containing the sample water was inverted many times to mix and distribute any deposit. A little water was aseptically discarded, the cap replaced and the bottle shook up and down 25 times. One 50ml and five10 ml water volumes were aseptically pipetted out and added to the corresponding 50ml and 10ml volumes of double strength medium. The media were incubated aerobically at 37^0C. After 24 hours and 48 hours the media were examined for any colour change and gas bubble filling the concavity of the inverted Durham tube. These features verify presence of "presumptive coliforms"

Cultures not showing production of both acid and gas were labelled as negative. By reference to tables of Most Probable Numbers (MPN) in respect of the positive and negative results, the MPN of the presumptive coliforms present in 100ml of the sampled water was read off. Confirmation of faecal coliforms was done by subculturing the positive culture of presumptive coliforms into tubes containing 5 ml broth with MacConkey Broth Purple and an inverted Durham tube. These tubes were kept at incubation at 44^0C, and then a drop of "positive coliform" was added to it and incubation continued for 48 hours. If again there was acid and gas production, it "confirmed" presence of "faecal coliforms" from the Table of MPN (51.6) read off the most probable number of coliform per 100ml. This gave the confirmed coliform count[7].

RESULTS

A total of 56 samples were tested from piped water supply of Services Hospital Lahore. These samples were collected from different areas of hospital. Water sources included 51 taps (90%), 4 from water coolers (8%), 1 distillation plant (2%) and main tanks of hospital. These water samples were collected and tested during winter season from December to March 2012. Multiple Test Tube method was used to determine the bacteriological quality of water. Thirteen samples (26%) of the 56 had a colour change from purple to yellow at 37^0 C and 44^0C

Table 1: Showing water quality at SIMS and Services Hospital

Water location	(n=56)	+ve13(23.2 %)	Water source
Surgical	8(14.3%)	5(8.9%)	Tap water
Medical	2(21.4%)	3(5.3%)	Tap water
Ortho	3(5.3%)	1(1.7%)	Tap water
Gynea	8(14.3%)	1(1.7%)	Water cooler
Peads	8(14.3%)	1(1.7%)	Water cooler
SIMS	8(14.3%)	1(1.7%)	Tap water
Hostel	2(3.5%)	0	Tap water
Emergency	4(7.1%)	0	Tap water
ENT	2(3.5%)	0	Tap water
Main tank	1(1.7%)	0	-

Table 2: Water samples showing coliform contamination

Ward	**Facility**	**Water source**	**MPN/ 100ml**
Emergency Lab	Laboratory	tap	01
Govt. officers block	Kitchen	tap	02
Surgical **I**V	Kitchen	tap	16
Surgical **II**	Washroom; male	tap	16
Surgical II	Washroom; male	tap	>18
Surgical II	Washroom; male	tap	>18
Orthopaedics	Kitchen	tap	<01
Surgical IV	Kitchen	tap	>18
Paediatrics	Corridor	Water cooler	03
Medical I	Washroom female	Tap	06
Medical III	Kitchen	Tap	01
Gynea		Water cooler	01
Pathology; SIMS	Media preparation	tap	>18

Fig. 1: Broth cultures used in bacteriological examination of water.

DISCUSSION

The bacteriological quality of drinking water can be determined by the presence of indicator bacteria like coliforms and *E coli* by the Membrane Filteration and Multiple Tube Method[7]. Although we tested for coliforms by the conventional methods, newer, more rapid methods are being evaluated for the early detection of and prevention from ill effects of contaminated waters. These include Polymerase Chain Reaction (PCR), Transcription Mediated Amplification (TMA) of RNA with fluorogenic probe and Dual Wavelength Audiometry[8].

In the present study 13(24%) water samples from SHL/SIMS yielded positive results (Table 2). Five samples were from the tap water of surgical floor and two from the laboratories. One sample from Department of Pathology yielded positive result for coliform, *E coli*. Two sources with positive results were water coolers as well.

SHL is a 1200 bedded hospital. In 2011, 678267 patients were managed on emergency basis, 10,85637 patients visited the outpatient and 62638 were admitted as inpatients and 71000 were operated upon. Such influx of an immuno-compromised population at the public sector hospital would utilize water supplies as well. SHL and SIMS are supplied by and stored in 4 storage tanks at the floor. They are regularly chlorinated on daily basis. The present building of Services Hospital Lahore started in 1960 and new blocks were annexed in 1986. SIMS College is a new building functioning since 2010. It has a closed water pipe system. The pipes are subject to hot and moist climate[9].

An Indian study was conducted in a tertiary care Medical Centre in North Western Himalayan Region on 91 water samples in 2 years period. In 2005-2006, of the 34 tap water samples 37(91%) were unsatisfactory and in 2006-2007, of the 42 tap water samples tested 30 (71.4%) were contaminated[10].

Over the past decade there has been a marked increase in water consumption from sources alternate to tap water as coolers and dispensers. Water coolers are popular in office buildings and public sector places inclusive of hospitals to cater the need for all people visiting them. These water bodies have the potential to cause water-borne diseases especially in immunocompromised and sensitive subjects. A comparative study in Italy, on 38 commercial outlets using coolers and tap water, showed the tap water to be of good quality without any coliform isolate. Both carbonated and non-carbonated dispensers yielded higher bacterial counts but absence of *E coli* and *Enterococci* ruled out fecal contaminants[11]. However another study in Canada on coolers in residences and workplaces revealed 28% and 36% of collected samples contaminated by at least one coliform or indicator bacterium and or one pathogenic bacterium[12]. In the present study out of 3 water-coolers, 2 tested positive with isolation of *E coli* from one and *Klebsiella* from the other. This result thus does not render the water safe for consumption especially the one with fecal bacterium *E coli*. Liguori *et al* however could not detect *E coli* or *Enterococcus* spp in any of the water samples in their study. However *Pseudomonas aeruginosa* was isolated from one tap water and 28.9% and 23.7% non-carbonated and carbonated water samples from 38 stores. Thus the water samples were rendered safe for consumption.

Although currently the role of *Pseudomonas aeruginosa* as a pathogen is debatable in the community, but it is a known pathogen in the immunocompromised and weakened patients in the hospital. The ability to grow at an elevated temperature (44.5°C) separates fecal coliform bacteria from the total coliform bacteria; they are a more accurate indicator of fecal contamination by warm-blooded animals. *Escherichia coli* (E. coli) is a member of the fecal coliform group of bacteria and is distinguished by its inability to break down urease[13]. The Environment Protection Agency US sets the Maximum Contamination Limit of zero *E. coli* per 100ml of water sample. No 100ml water sample should test positive for either *E. coli* or total coliform for any water that is to be used for drinking[14.]. Water treatment can ensure that the risk of waterborne illness is negligible if an adequate source protection and/or treatment program is implemented. Drinking water treatment to public water consists of series of barriers as coagulation, flocculation, filteration and oxidation Chemical and microbiological contamination can occur in consumer's premises from plumbing material or from back flow of liquids into the distribution system due to improper connection[15].

Water borne illnesses can result from breaches in water treatment and distribution processes. They can cause gastroenteritis and other diseases. In USA total number of water borne illnesses are 19.5 million/year[16] whereas in Pakistan a survey on water revealed, bacterial causes of water contamination to be 68% giving rise to 100 million diarrheal cases seeking hospital admissions and 40% mortality associated with it[5].

Storage removes 90-95% physical impurities by suspension and oxidation of organic matter by aerobic bacteria in presence of light. The total count of bacteria can decrease by 90%. Filteration decreases the bacterial count by 98-99% and disinfection destroys all pathogens[10].

Recommendations:

1. Cleaning of sedimentation tank should be done periodically (once a week)
2. Pre-requisite purification steps of storage, filteration and disinfection should be adopted rigorously and religiously without by passing any.
3. Main supply source should be chlorinated as per guidelines. Minimum contact period between water and chlorine should be one hour. Disinfection is rapidly and directly proportional to the temperature of water and inversely with turbidity of water.
4. Cleaning and chlorination record of tanks should be maintained
5. A yearly plan should be laid down for water sampling
6. Water sampling from kitchens and hostels should be done more frequently especially in summers and with the onset of Monsoons.
7. Plumbing and water supply system should be monitored for leaks and cross-contamination[16].
8. Cooler faucets are touched by contaminated hands and give high bacterial counts, so periodic disinfection of water dispensers is needed by 3% hydrogen per oxide[17].

CONCLUSION

Water contamination especially with coliforms is a serious problem more so in a public institution and immunocompromised population. Water quality should be regularly tested particularly for microbes to prevent water borne dissemination of disease.

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