

Microbiological Analysis of Chlorinated Water Supplied in Jaipur

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Abstract:

Water pollution is the contamination of water bodies (e.g. lakes, rivers, oceans, aquifers and groundwater). In almost all cases the effect is damaging not only to individual species and populations, but also to the natural biological communities. All people live downstream of water supplies. Hence, the quality of water depends on the maintenance of water supply network. During the lateral travel of water it may be contaminated by various microorganisms. This polluted and contaminated water affects the consumer's health and cause various water borne diseases. Therefore, in present study, the level of faecal coliform in water distribution network of Jaipur is evaluated in 12 different areas of Jaipur city. Residual chlorine and faecal coliform bacteria was analyzed. Interestingly, it was found that residual chlorine was present in permissible limit in all areas, however, showing the presence of microbes and coliforms. Out of 12 sites selected, 8 sites were found to have coliform bacteria showing the possibility of contamination of water supply by sewage discharges which come in contact with supplied water due to leakage in pipes. In 4 areas, however microbial contamination by Gram positive bacteria was observed instead of coliforms. In these four areas water may be polluted during its flow off from polluted soil and area. It is possible that microbes may enter through damaged pipes. To provide good quality water to consumer, it should be protected from microbial contamination by maintaining the water supply network.

Keywords: APHA-American Publication Health Association, EC-Electrical conductivity, IMViC-Indole, Methyl-red, Voges-Proskauer, Citrate-utilization test, O.H.S.R-Over Head Service Reservoir, RC-Residual Chlorine

1.0 Introduction:

Jaipur (Pink city) is situated in eastern part of Rajasthan which is known as fast developing city. Its population is expected to reach about 42 lacs by 2011. Jaipur is not having its own water source. A very large section of population uses raw water, surface water and ground water bodies for human consumption. Ramgarh lake (Surface source), Amanishah tank (Ground water source) are the two major sources of water supply in wall city area of Jaipur. Besides this, Jaipur city has two water filter plants for purification of water i.e., Laxman Dungri Filter plant of 54 MLD capacities (Rapid gravity filter), Bund gate filter plant of 18 MLD capacity. Both filter plants have rapid gravity filter for purification. Filtered water is found to be disinfected by chlorine in Clear Water Reservoirs. This chlorine treated water is now ready to supply to the consumer through pipelines. Underground water sources (Tube wells) forms the major water source of supply system in Jaipur. The water is pumped from tube wells all around the city and collected in Clear Water Reservoirs (C.W.R). This water is further chlorinated before supply to the

consumer. These major sources of water are supplying treated water to Jaipur city. All consumers live downstream to these water filtration plants and reservoirs. Due to lateral travel of treated water in the pink city, it becomes contaminated. This treated water is also contaminated due to microbes present in the local environment. The contaminated water not only affects the health of the public but also the consumption of polluted water may cause various water borne diseases such as diarrhea, dysentery and complaints of skin, teeth and other abdominal diseases (Bhardwaj, 2005).

In this way, water available to public may be unfit for drinking purposes due to chemical and microbiological contamination which in terms need a proper and regular monitoring of water quality for making a proper remedial strategy. In all known forms of life continuation, water is essential substance of the natural sources. The spoiling of the water quality and water's natural balance is directly or indirectly affected by pollution (Kosygin *et al.*, 2007). In developing

countries, pollution of the surface and underground water by liquid and solid wastes is widespread, here by rendering them unsuitable for human use. With the development of Urban, the necessity of water usage and obtaining the clean water showed up as a major problem. Infections may be due to poor ground water protection, poor quality sources, poor construction, structural deterioration, poor sanitation or poor site. Chatterjee et al. (2011) have done bacteriological examination of drinking water in Burdawan, India with reference to coliforms .MPN was done to detect coliform in water samples from mobile vendors, sweet shops and tap water supplied from Burdawan municipality. Keeping in this view, the present study was planned to assess the physicochemical and microbiological qualities of water of Jaipur city from 12 different locations. The bacteriological contamination of water resources have been reported by various researchers; Gangal, 2007; Chatterjee et al., 2007; Bhardwaj, 2005).

Besides this, many reports have been published on microbiological or bacteriological quality of drinking water. According to WHO, water pollution has been reported to cause 80% of human diseases and 30% of human mortality in developing countries (Chakraborty, 1999; Pradhan et al., 2003) reported the water borne diseases like dysentery, diarrhoea and gastroenteritis were very common in people consuming microbially contaminated water. Although the water is supplied after chlorination to Jaipur city, lack of proper amenities may rendered the water unsafe for drinking as well as domestic purpose. This has resulted in contamination of water supplies with different pathogenic bacteria which cause various water borne diseases and abdominal problems. To protect the consumers from these diseases it is necessary to prevent water contamination at its source by regular monitoring presence of coliforms.

This project deals with water quality assessment of "Inside walled city area of Jaipur "having different morphological functional areas, by considering microbiological parameters. To study microbial contamination of water bodies, **the present work was planned to protect human being from water borne diseases.** To detect water pollution indicator microorganisms i.e. *E.coli*, Imvic test was performed to confirm its presence. The main objective of this study is to identify coliform organisms as *Escherichia coli*.

2.0 Materials and Methods:

2.1 Study Area: Sampling Sites:

Twelve sampling points was selected from inside walled city area of Jaipur for the collection of water samples. Water samples were collected from i)Laxmandungari water filters plant (surface water source) and ii)Amanishah Tank (ground water source) before chlorination and after chlorination. Other sites for collection of water samples involved iii)Transport Nagar area iv) Suraj Pole area v)Topkhana Hazoori area vi) Badi Chaupar area vii) Purani-Basti area viii) Toup-Khana-Desh ix) Modi-Khana Chaukri and x)Chaura-Rasta. These are the main covering areas of Jaipur and people are suffering from microbiological diseases in these areas so these sites were selected for survey.

2.2 Collection of Samples:

Pre- and post-chlorinated Water samples were collected from filter plants, tanks and C.W.R in duplicates. Samples were also collected from consumer end point areas in duplicates fortnightly during 2 months.

2.3 Physico-chemical Analysis:

Collected water samples were analyzed for Colour, taste, Odour and Residual Chlorine as given in APHA (2005). Residual chlorine was estimated by chloroscope and volumetric titration analysis method.

2.4 Bacteriological analysis:

The microbiological analysis was done by inoculating the samples on Nutrient Agar plates. Total number of colonies were counted and recorded as colony forming units. Evaluation of coliforms were done by IMViC test which is a combination of four tests i.e., Indole production, Methyl-red test, Voges-Proskauer test and citrate utilization test (Cappuccino JamesG., Sherman Natalie (2005).

2.4.1 Indole Test- Coliforms hydrolyze tryptone, when they were grown in tryptone broth, with the production of Indole. Presence of indole was detected by adding Kovac's reagent, which produced a cherry red reagent layer.

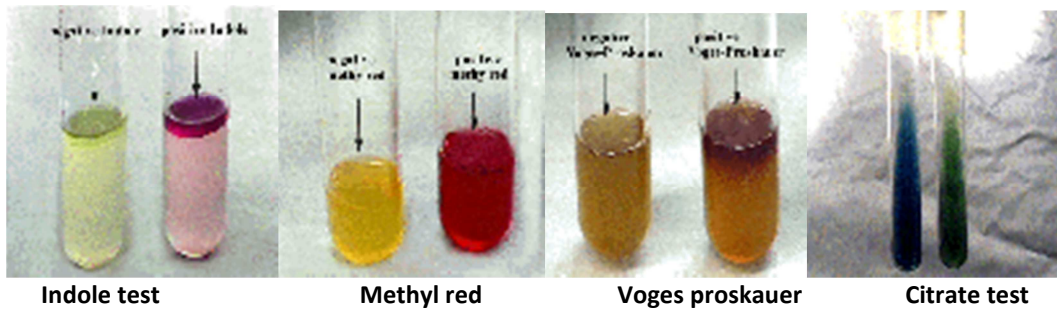
2.4.2 Methyl Red Test- Glucose, present in MR-VP broth, was oxidized by all enteric organisms for energy production. End products, however, will vary depending on enzymatic pathways present in bacteria. Therefore, pH indicator Methyl red was used to detect the presence of acid end products of glucose fermentation.

2.4.3 Voges- Proskauer Test- Coliforms cultured in MR-VP Broth, produce neutral end products such as acetylmethylcarbinol from organic acids that result from glucose metabolism. These neutral products were detected by Barritt’s reagent which consists of alpha-naphthol and potassium hydroxide solution. Acetylmethylcarbinol is oxidized to diacetyl compound in presence of alpha-naphthol catalyst and a guanidine group of peptone of MR-VP Medium and produced red colour in the medium.

2.4.4 Citrate Utilization Test- Coliforms are capable of using citrate of Simmon Citrate Agar as carbon source for their energy which led to the production of oxalaacetic acid and acetate. These products converted to pyruvic acid and carbon dioxide. Carbon dioxide reacts with sodium and water to form sodium carbonate, an alkaline product which changes the colour of medium from green to deep Prussian blue due to the presence of bromothymol blue indicator in medium.



Jaipur map showing areas covered for survey



Indole test

Methyl red

Voges proskauer

Citrate test

3.0 Results and Discussion:

Organoleptic properties and residual chlorine amount of entire 12 sites are given in Table 1. Among all the 12 sites, water of 10 sites was found to be colourless, sweetish and odourless. Water of Laxmandungri filter plant and Amanishah tank were found to be yellowish, sweet and having pungent odour before chlorination. This water, however, became colourless, sweetish and odourless after chlorination (Table 1). Residual chlorine concentration was found to be very high before and after chlorination in the Laxmandungri filter plant and Amanishah tank. However, during the lateral travel of water from its source i.e. Laxmandungri filter plant and Amanishah tank to the various area of city, chlorine concentration reduced to accepted level i.e. 0.2-0.3 unit such as mg/l etc.) according to World Health Organization(WHO)1984, CPHEEO(MOH) Ministry of Works & Housing-Govt.of India(PH Manual)1975.

All the city area of Jaipur studied in the present work was found to have appropriate amount of chlorine in the water. Chlorine is well known disinfecting agent which is used to kill all the microbes present in drinking water Pandit et al. (2006). However, all the 12 areas showed very high microbial load in the supplied water as revealed by their colony forming units per 100ml (cfu/ml) of water sample (Table 2). The colony forming units were found to be in the range of 200 to 750 cfu/100ml which showed that chlorination is inefficient in killing the microorganisms or different microbes was acquired by water from the surrounding atmosphere. All the 12 sites showed the presence of different types of bacteria in the

supplied water which was differentiated on the basis of cultural and microscopical characteristics as shown in Table 2. Out of 12 sampling sites, eight sampling sites were found to have gram-negative bacteria which were further analyzed by IMViC test for the confirmation of coliforms. Modikhana-Chaukri, Chaura-Raasta and Laxmandungri filter plant (before chlorination) were found to have *Salmonella spp.* in the supplied water. Besides this, *Klebsiella sp.* was found to be present in Tokhana Hazoori area, Laxmandungri filter plant and Amanishah (CWR) even after chlorination. Similarly, *Pseudomonas sp.* was found to be present in the water of Transport nagar area and Amanishah (C.W.R.) after chlorination. Presence of coliforms in the water revealed that chlorination is not sufficient to remove the entire microbial load from the water. *Shigella sp.* was found to be present in Amanishah tank before chlorination which is reduced by chlorine, however, *Pseudomonas* appear in the water after chlorination showing its resistance to chlorine. All these 8 sites were showing the presence of coliforms, Gram-negative bacteria in spite of having appropriate amount of chlorine. This indicates that there is some leakage in the pipeline or may be exposed area through which faecal coliforms gets their way to water supply. Remaining other 4 sites was found to have Gram-positive bacteria. Surajpole and Topkhana desh was found to have *Staphylococcus sp.* However, Badi-Chaupar and Purani-basti was found to have *Streptococcus sp.* and *Diplobacillus sp.* respectively. This also indicates that due to poor maintenance microbe may enter the pipeline and contaminate water during its lateral travel towards consumer point area.

Table 1: Physicochemical-Characteristics of water samples collected from 12 sample sites.

Sample Name	Colour	Taste	Odour	Residual Chlorine (mg/liters)
Laxmandungari Filter plant (Before Chlorination)	Yellow	Sweet	pungent	0.7
Laxmandungari Filter plant (After Chlorination)	Colourless	Slightly sweet	odourless	0.64
Amanishah Tank	Yellowish	Sweet	pungent	0.71
Amanishah (C.W.R)	Colourless	Slightly sweet	odourless	0.62
Transport Nagar	Colourless	Slightly sweet	odourless	0.20
Surajpole	Colourless	Slightly sweet	odourless	0.30
Topkhana Hazoori	Colourless	Sweet	odourless	0.25
Badi-Chaupar	Colourless	Sweet	odourless	0.2
Purani-Basti	Colourless	Slightly sweet	odourless	0.20
Toupkhana-Desh	Colourless	Sweet	odourless	0.25
Modikhana-Chaukri	Colourless	Slightly sweet	odourless	0.20
Chaura-Raasta	Colourless	Sweet	odourless	0.2

Table 2: Microbiological characteristics of 12 sampling sites

Water Sample	Total Count Cfu/100 ml	Culture characteristics	Gram Staining	IMViC Test for Gram negative (-) Bacteria				Microorganisms Identified
				Indole	MR	VP	Citrate	
Laxmandungari filter plant (Before chlorination)	750	Round, yellowish, Smooth ,small	Gram -ve	-	+	-	+	<i>Salmonella sp.</i>
Laxmandungari Filter plant (After Chlorination)	550	Round, yellowish, smooth, tiny	Gram -ve	-	+	+	+	<i>Klebsiella sp.</i>
Amanishah Tank (Before chlorination)	700	Round, yellowish, smooth, tiny	Gram -ve	-	+	-	-	<i>Shigella sp.</i>
Amanishah (C.W.R)	200	Round, whitish, smooth, tiny	Gram -ve	-	-	-	+	<i>Pseudomonas sp.</i>
		Round, whitish, smooth, tiny	Gram -ve	-	+	+	+	<i>Klebsiella sp.</i>
Transport Nagar	750	Round, whitish, rough, tiny	Gram -ve	-	-	-	+	<i>Pseudomonas sp.</i>
Surajpole	350	Round, rough, yellowish, small	Gram -ve	#	#	#	#	<i>Cocci, arranged in bunch</i>
Topkhana Hazoori	450	Oval, yellowish, smooth, tiny	Gram -ve	-	+	+	-	<i>Klebsiella sp.</i>
Badi-Chaupar	500	Round, yellowish, smooth, tiny	Gram-ve	#	#	#	#	<i>Cocci, arranged in chain</i>
Purani-Basti	400	Oval, yellow, smooth, small	Gram -ve	#	#	#	#	Bacilli, arranged in group of two
Toupkhana-Desh	200	Round, rough, yellowish, small	Gram -ve	#	#	#	#	<i>Cocci, arranged in bunch</i>
Modikhana-Chaukri	250	Round, smooth, yellowish, small	Gram -ve	-	+	-	+	<i>Salmonella sp.</i>
Chaura-Raasta	300	Oval, yellowish, rough, small	Gram -ve	-	+	-	+	<i>Salmonella sp.</i>

(+): positive test, (-): negative test, (#): no information

Residual chlorine is estimated to check the quality of water because chlorine is being added to purify the water and to make it disinfectant but when it reaches to consumer point areas it reduces because during supplying microbes in the pipelines digest it and it reduces. This also indicates the presence of microbes in the pipelines. Selected areas reduce the dose of Residual Chlorine. Microorganisms dwelling in drinking water reclaimed from water containing water in an enclosed environment were identified. The microbial species in the water reclaimed from the condensate of hydrogen peroxide decomposition products and from the air humidity condensate showed specific features and differences when compared to those in natural water. Singh et.al (2010) has analyzed groundwater samples from

hand pump of seven areas and adjacent localities of Jaipur city during monsoon session with the help of standard methods of APHA. The values obtained were compared with standards of ISR, ICMR and WHO. The concentrations of various parameters are within permissible limits in groundwater. Tatawat et.al (2010) has conducted research to evaluate the water quality of Jaipur City. Groundwater samples from hand pumps and tube wells of eleven sampling stations were analyzed during monsoon session with the help of standard methods of APHA. The analytical result shows higher concentration of total dissolved solids, total hardness which indicates signs of deterioration but values of pH within permissible limit as per WHO standards.

Sharma et al. (2011) have analyzed groundwater samples from Sanganer block of Jaipur for their structural and functional attributes, both quantitatively and qualitatively, to determine their suitability for drinking purposes. It was found that the qualities of almost all samples were within permissible limits but contents of TDS were not within permissible limits. The general characteristic of the samples can be classified under moderate category and are good for household commercial purposes. The results of suitability evaluation indicate that there is no major pollution hazard in groundwater of Jaipur. Ramteke et al.(2009) have discussed the evaluation of coliforms as indicators of water quality in India. To evaluate these counts as indicators of recent faecal contamination, the total coliforms and thermotolerant coliforms isolated have been identified like *Escherichia coli*, *Citrobacter* and *Klebsiella* species. Chatterjee et al. (2008) have done bacteriological examination of drinking water in Burdawan, India with reference to coliforms. MPN was done to detect coliform in water samples from mobile vendors, sweet shops and tap water supplied from Burdawan municipality. The study revealed that the number of coliforms was very high (> 1600) from mobile vendors. The bacteria were *Escherichia coli*. Water of mobile vendors and sweet shops was not potable while municipal tap water was found to be safe for drinking.

4.0 Conclusions:

In the present study, microbial load in water at its source and consumer point areas were assessed because contaminated or polluted water may lead to the various health related problems. The present study emphasized on the two major findings. First, chlorination is not sufficient to reduce the microbial load of water. Second, microbial load at consumers point area indicates that water become contaminated when it is supplied to these area. The possible reason of this may be the poor maintenance of water supply network due to which water may exposed to various microbes. There is a possibility that these microbes may tolerate the level of residual chlorine. When this polluted water is consumed it lead to various water borne diseases, abdominal problems etc. Therefore, it is necessary that supplied water should be regularly assess for the presence of coliforms at its source and consumer point areas.

References:

- 1) APHA (2005): Standard methods for the examination of water and waste water, American public health association, Washington D.C.
- 2) Agarwal G.D. (1999): Diffuse agricultural water pollution in India. Water science and Technology IWA publishing 39(3):33-47.
- 3) Aneja K.R (2003) :Experiments in Microbiology plant Pathology and Biotechnology.New Age International(P)Limited Publishers Ed 4:102-106.
- 4) Bharadwaj R.M. (2005): Water quality monitoring in India Achievements and constraints. International work session on water statistics Vienna 2(1):20-22.
- 5) Dhindsa S.S (2007): A manual on water and waste water analysis.CPHEEO Ministry of urban development and poverty Alleviation govt. of India New Delhi:84-196.
- 6) Forget G, Sanchez-Bain WA (1999): Managing the ecosystem to improve human health integrated approaches to safe drinking water . International journal occupation Environmental health 5(1):38-50.
- 7) Gangal K (2007): Geochemical study of ground water of Sanganer area of Rajasthan. Journal of water, Environment and Pollution 4(2):93-97.
- 8) Jain P., Sharma J.D, Sohu D., Sharma P. (2006): Chemical analysis of drinking water of villages of Sanganer Tehsil, Jaipur(District).International Journal of environmental science and Technology 6(2):67-69.
- 9) Khan T.I, Kaur Navneet, Vyas P.C. (1995): Effects of industrial effluents on physico-chemical characteristics of Amanishah Nalla – A case study. Journal of environment and Pollution 2(3):147-150.
- 10) Lechevallier MW, Cawthon CD, Lee RG (1988) :Factors promoting survival of bacteria in chlorinated water supplies. Applied environment microbial 54(3):649-654.
- 11) Mathur Priti, Kalia Sarina(2007): Water quality assessment and control- Jaipur city. Indian Journal of Env. Sc 11(1):37-39 Green Earth foundation.
- 12) Marco E., Aieta Roberts, Paul V, ASCE M. (1983): Disinfection with chlorine & chlorine dioxide. Journal of Environmental Engineering 109(4):783-799.

- 13) Naik S.R, Aggarwal R, Salunke P.N, Mehrotra N.N, (1992): A large waterborne viral hepatitis E.epidemic in Kanpur. India Bulletin of world health organization 70(5):597- 604.
- 14) Ramteke P.W, Bhattacharjee J.W, and Pathak S.P, Kalra N (1992): Evaluation of coli forms as indicators of water quality in India. Journal of applied microbiology 4(72):352-356.
- 15) Shah Mayur C. ,Shilpkar PrateekG. ,Acharya PradipB. (2008):Groundwater quality of Gandhinagar Taluka,Gujrat,And India.E-journal of chemistry 5(3):435-446.
- 16) Sharma S.K, Chandel. C.P.S (2006): Ground water pollution of sanganer block of Jaipur district in Rajasthan.MKK Publication 5(6):10-11.
- 17) Sharma M.K., Choubey V.K (2007) :Groundwater quality status of Jaipur, District, Rajasthan .The Icfai Journal of Environmental Sciences1(2):48-57.
- 18) Singh Vijendra, Singh Chandel CP (2005) : Water quality of groundwater and wastewater of Jaipur city for irrigation purpose.Aquacult 6(1):25-31.
- 19) Srivastava I.C, Sharma Bhagirath, Kulshristha S.K (2005) :Sector policy for rural drinking water and sanitation(Draft).Water science and Technology 2(5):1-2.
- 20) Tatawat R.K, Chandel Singh C.P (2007) : Quality of groundwater of Jaipur city, Raj(India) and it's suitability for domestic and irrigation purpose. Applied ecology and environmental research 6(2):79-88.
- 21) Tatawat R.K, Chandel Singh C.P (2007): Quality of ground water of Jaipur city, Raj(India) and it's suitability for domestic and irrigation purpose. Applied ecology and environmental research 6(2):79-88.
- 22) Venkatasubramani R, Murali K, Meenambal T (2005) :Groundwater quality index for Coimbatore east zone. Nature environmental pollution technology 4(2):199-202.
- 23) Wagh SP, Shrivastava VS (2005) :Impact of heavy metals on soils and groundwater. Nature environmental pollution Technology 4(1):93-96.