Original Article

Chemical and Microbial Analysis of Drinking Water in the Villages Under Rural Health Training Center, Apollo Institute of Medical Sciences and Research, Hyderabad

Sri Vishnupriya Ravipati[1], Manick Das[2], Neeta Mathur[2], B. N. Das[2], N. Balakrishna[2], Snigdha Pattnaik[2]

[1]Department of Community Medicine, Apollo Institute of Medical Sciences and Research, Hyderabad, Telangana, India [2]Department of Microbiology, Apollo Institute of Medical Sciences and Research, Hyderabad, Telangana, India

Abstract

Aim: The primary concern regarding the quality of drinking water in developing countries is microbiological contamination. The rural people of Moinabad Mandal use 20 L bubble top water cans for drinking. The purpose of the study is to check the chemical and microbial quality of the water obtained from these packaged water cans. The study analyses the basic parameters such as chlorine content and coliform count in the drinking water. **Methods:** Out of 37 villages for the study, we randomly took 10 villages and from those randomly 10 houses were selected who were using 20 L bubble top water cans for drinking purpose. From each house, 150 ml water sample was obtained in a sterile container and two tests were applied: Chlorine estimation by orthotolidine test and coliform count by single strength and double strength MacConkey broth. Basic statistical parameters such as minimum, maximum, mean, median, and deviation standard are used to analyze the data. **Results:** The study shows that none of the samples have free residual chlorine which indicates that the chances of contamination of the water are high. Forty-one percentage of the total 100 samples are contaminated with *Escherichia coli* and the other 59% samples with Gram-negative bacteria other than *E. coli* which indicates environmental contamination making it unfit for drinking. **Conclusions:** As most of the people in Moinabad Mandal depend on packaged water, and hence authorities need to monitor drinking water quality to safeguard consumers' health.

Keywords: Chlorine, coliform count, drinking water quality, *Escherichia coli*, microbial contamination, packaged drinking water

Introduction

Inadequate water supply is one of the major challenges in developing countries. The joint monitoring program for Water Supply and Sanitation, implemented by the World Health Organization and UNICEF, states that 783 million people in the world (11% of the total world population) have no access to safe drinking water, 84% of them are who live in rural areas.[1] In Chad, 56% of people in rural areas and 30% of people in urban areas have no access to safe water.[1]

Whether in 1 Lor 20 L bubble topped water cans, packaged drinking water has been in great demand, and India is rated among the top 10 countries in the world. Bottled water production companies are one of the fastest-growing industrial sectors in the world. Presently, there are almost more than 3400 bottling plants in India. Half of these are concentrated in the Southern regions of India.[2]

Access this article online

Quick Response Code:

Website: https://journals.lww.com/IJEH

DOI: 10.4103/ijehe.ijehe_58_20

Demand for bottled water has caused a rise in several small-scale entrepreneurs involved in their production and distribution. However, as the demand has been increasing, serious concerns about its quality and safety have arisen subsequently. The chemical and microbiological quality of packed drinking water of some manufacturers has been violating some of the national standards.[3] Studies from India and other parts of the world have shown that bottled water has been contaminated with harmful

Address for correspondence: Dr. Neeta Mathur, Department of Community Medicine, Apollo Institute of Medical Sciences and Research, Hyderabad, Telangana, India.
E-mail: mathurneeta90@gmail.com

How to cite this article: Ravipati SV, Das M, Mathur N, Das BN, Balakrishna N, Pattnaik S. Chemical and microbial analysis of drinking water in the villages under rural health training center, apollo institute of medical sciences and research, Hyderabad. Int J Env Health Eng 2025;14:8.

Received: 15-12-2020, **Accepted:** 13-10-2021, **Published:** 31-03-2025

disease-causing microorganisms at various stages of its production.[4-6]

Most companies generally use bore wells as their source of water. There, water is pumped out from depths of 80–500 ft below the ground.[7,8] The least likely sources of packaged water are from public drinking water systems such as municipality supply water.[9] The primary concern regarding the quality of drinking water in developing countries is microbiological contamination. Moreover, inorganic contaminants, concerning both health and other aspects, can be present in the waters.

The important tests used in water quality surveillance or quality control in small communities are those for microbiological quality (by measuring the indicator bacteria) and turbidity, and for free chlorine content and pH where chlorination is used. These tests should be carried out whenever a sample is obtained, regardless of how many other physical or chemical variables are to be measured.[1] Coliform bacteria should not be detectable in treated water supplies and if found, suggest inadequate treatment, posttreatment contamination, or excessive nutrients. The coliform test can therefore be used as a basis for both the treatment efficiency and the integrity of the distribution system.[1]

The Ranga reddy district in Telangana is mainly dependent on groundwater for its irrigation and domestic needs. Groundwater utilization for daily purposes in these areas is through deep bore wells in the noncommand areas and dug wells and shallow bores in other areas. The wells yield low during the summer months.[10] The rural people mainly use packaged 20 L water cans for drinking purposes. Hence, we planned to take the study to assess the chlorine content and coliform count of drinking water of the villages under Rural Health Training Center, AIMSR, Hyderabad.

Materials and Methods

The study is carried in the Moinabad Mandal, Rangareddy district. This is the rural field practice area of the Community Medicine Department, Apollo Institute of Medical Sciences and Research, Hyderabad. The study was conducted from July 2019 to September 2019. Approval was obtained from the institutional ethics committee before the commencement of the study.

As of 2018, the Mandal has 37 villages covering 60,181 populations. Out of 37 villages for study purposes, we randomly took 10 villages: Chilkur, Chinna Shapoor, Himayat Nagar, Kanakamamidi, Kanchananiguda, Kethireddypally, Moinabad, Peddamanglaram, Surangal, and Venkatapur. Among these 10 villages, 10 houses are selected from each village that was using 20 L water cans for drinking purposes for the past 6 months. Hence, the total sample will be 10 × 10 = 100 water samples (10 samples from each of the 10 villages).

From each house, 150 ml water sample was obtained in the sterile container and transported to the medical college at room temperature. On each water sample, two tests were applied in the Community medicine and microbiology department, respectively

1. Chlorine estimation: By orthotolidine test
2. Coliform count: By single strength and double strength MacConkey broth.[11]

Type of study

The study was a cross-sectional study.

Collection of water samples

Sterile glass bottles of 200 ml capacity were used to collect water samples. Samples were obtained from the 20 L bubble top water cans used for drinking purposes. The sampling bottle was not filled up to the brim and 50 mm space was left for effective shaking of the bottle. Water samples were properly labeled and transported to the medical college within 4 h at room temperature which ensured that there was no change in the contents of the sample. In the Microbiology Department, bacteriological test on water samples was started as early as possible to avoid unpredictable growth of microbial population.

Chlorine estimation

Orthotolidine test determines both free and combined chlorine in the water. The test is carried out by adding 0.1 ml of the reagent to 1 ml of water. The reagent consists of orthotolidine, dissolved in a 10% solution of hydrochloric acid. This reagent when added to water containing chlorine, turns yellow and the intensity of the color varies with the concentration of the gas and this color is produced by both free and combined chlorine residuals. The yellow color produced is matched against suitable standards or color discs. The reading is obtained within 10 s after the addition of the reagent to estimate free chlorine in the water.[12] The orthotolidine test reacts with free chlorine instantaneously but reacts more slowly with combined chlorine.[13] The color that is produced after a lapse of 15–20 min is due to the action of both free and combined chlorine.

Coliform count estimation

It was done by multiple tube fermentation test, to find the total or presumptive coliform count. This test is based on estimating the most probable number (MPN) of coliform organisms in 100 ml of water.

Multiple tube method

The test is carried out by inoculating measured quantities of the sample water (0.1, 1.0, 10, 50 ml) into tubes of MacConkey's lactose bile salt broth with bromocresol purple as an indicator. The tubes are incubated for 48 h. From the number of tubes showing acid and gas, an estimate of the MPN of coliform organisms in 100 ml of the sample water can be obtained from statistical tables.[14] The results were interpreted as excellent, acceptable, unacceptable, and grossly polluted as given in Table 1. The principle of the coliform test is the detection of *Escherichia coli* in water samples as its presence will indicate that there is contamination with animal or human feces. It does not exist as a saprophyte in the environment. Other coliforms

such as Klebsiella, Enterobacter, and Citrobacter are found as saprophytes in the environment apart from human and animal feces [Table 1].

Confirmatory tests

The confirmation is done by subculturing each presumptive positive tube in two tubes of brilliant green bile broth, one of which is incubated at 37°C for up to 48 h for the confirmation of the presence of coliform organisms, and the other incubated at 44°C and inspected after 6 and 24 h to decide whether or not *E. coli* is present. *E. coli* is capable of producing gas from lactose at 44°C. Further confirmation of the presence of *E. coli*, can be obtained by testing for indole production at 44°C.

Data analyses

Basic statistical parameters such as minimum, maximum, mean, and standard deviation (SD) were used to analyze the data.

Observations and Results

Hundred drinking water samples were collected from packaged 20 L water cans of 10 villages of Moinabad Mandal, namely, Chilkur, Chinna Shapoor, Himayat Nagar, Kanakamamidi, Kanchananiguda, Kethireddypally, Moinabad, Peddamanglaram, Surangal, and Venkatapur. The mean duration of packaged water can be used for drinking purpose is 7.69 years ± 3.5 SD. The maximum duration of water can be used for drinking purpose is in villages Himayat Nagar and Moinabad and the minimum duration is in Chikur and Kethireddypally. Table 2 shows the duration of the use of packaged drinking water cans. Free residual chlorine was absent in all 100 water samples [Table 2].

Figure 1 shows the percent distribution of coliform count. Among 100 water samples, 59% samples had no coliform count but other Gram-negative bacteria were present, whereas 7% of samples were acceptable having coliform count between 1 and 10 MPN, 13% samples were unacceptable with coliform count between 11 and 50 MPN, and 21% were grossly polluted having coliform count >51 MPN. Percent distribution of coliform count in the villages is given in Table 3 [Figure 1 and Table 3].

Table 4 represents the frequency distribution of coliform count in the water samples. The mean coliform count is 28.78 ± 53 SD. Among 21 water samples grossly polluted for drinking, five showed coliform count 180, four showed coliform count 161, ten showed coliform count 92 and two showed coliform count 54. The 75th percentile of the coliform count is 21 [Table 4].

Discussion

The study shows that the free residual chlorine was absent in all the samples whereas the coliform count of the 100 samples shows that 7% contain coliform count <10 MPN; hence, they are acceptable for drinking. About 13% of the samples contain coliform content ranging from 10 to 50 MPN making it unacceptable for drinking purposes and 21% of samples contain a coliform count of more than 50 MPN indicating that they are grossly polluted and are not fit for drinking. The recovery of 59% samples showing Gram-negative bacteria other than *E. coli* indicates that there is environmental saprophyte contamination of drinking water which is also unfit for drinking purposes.

The study of Joseph N *et al*.[15] on bacteriological analysis of bottled drinking water at major transit places in Mangalore shows that the number of colony-forming units (CFU) per ml was <100 in 15 samples, 101–200 in 3 samples, 201–400 in 4 samples, and 401–500 in 2 samples. In other studies on packaged drinking water done in India, the acceptability of bottled water ranged from 60%,[14,16,17] 83%,[18] 90%,[19] and even 100%.[20-22] In another study conducted by Haulage *et al*.[23] on bacteriological and physic-chemical analysis of locally packaged drinking water state that most (73.3%, 11/15) of sachet water and a few (30%, 3/10) of bottled water brands had a total coliform count above the acceptable limit of 0 CFU/100 ml. A study conducted by I. Salehi[24] on the evaluation of the microbial and physicochemical quality of bottled water reports that none of the samples, had coliforms or fecal coliforms (<1cfu/100 ml). This helps us understand that the packaged drinking water is mostly contaminated and is not fit for drinking purposes.

The practice of filling the bottle directly from the tap water and sealing it without any pretreatment is generally done by the bottled water manufacturers for their financial benefit is one

Table 1: Interpretation of most probable number values/100 ml result

Coliform count	Comments
0	Excellent
1-10	Acceptable
10-50	Unacceptable
>50	Grossly polluted

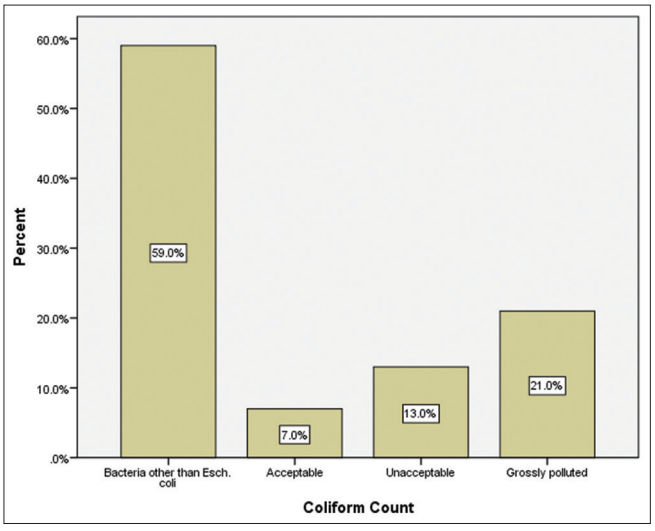

Figure 1: Distribution (%) of coliform count

Table 2: Duration of the use of packaged drinking water can (years)

Village name	Mean	Median	SD	*n*	Minimum	Maximum
Chilkur	5.80	6.00	2.150	10	1	10
China Shapoor	6.60	6.00	0.843	10	6	8
Himayat Nagar	10.60	10.00	4.402	10	6	20
Kanakamamidi	5.80	6.00	0.422	10	5	6
Kanchamaniguda	10.00	10.00	0.000	10	10	10
Kethireddypally	5.90	5.50	4.358	10	1	16
Moinabad	11.20	10.00	3.765	10	6	20
Peddamangalaram	9.00	10.00	3.528	10	5	17
Surangal	7.50	8.00	2.838	10	3	10
Venkatapur	4.50	4.50	1.269	10	2	7
Total	7.69	6.00	3.507	100	1	20

Table 3: Distribution (%) of coliform count in the village

Village name	Bacteria other than *Escherichia coli*, count (%)	Acceptable count (%)	Unacceptable count (%)	Grossly polluted count (%)	Total count (%)
Chilkur	6 (60)	0	1 (10)	3 (30)	10 (100)
China Shapoor	4 (40)	2 (20)	2 (20)	2 (20)	10 (100)
Himayat Nagar	5 (50)	3 (30)	1 (10)	1 (10)	10 (100)
Kanakamamidi	6 (60)	0	2 (20)	2 (20)	10 (100)
Kanchamaniguda	6 (60)	1 (10)	0 (0)	3 (30)	10 (100)
Kethireddypally	6 (60)	0	0	4 (40)	10 (100)
Moinabad	7 (70)	0	0	3 (30)	10 (100)
Peddamangalaram	5 (50)	0	5 (50)	0	10 (100)
Surangal	6 (60)	1 (10)	1 (10)	2 (20)	10 (100)
Venkatapur	8	0	1 (10)	1 (10)	10 (100)

Table 4: Frequency distribution of Coliform count

Coliform count	Frequency of water sample	Percent (%)	Cumulative Percent (%)
0	59	59	59
3	1	1	60
7	1	1	61
9	1	1	62
10	4	4	66
12	3	3	69
14	2	2	71
18	3	3	74
21	2	2	76
24	1	1	77
28	1	1	78
35	1	1	79
54	2	2	81
92	10	10	91
161	4	4	95
180	5	5	100
Total	100	100	

of the reasons behind the same bacteriological quality of tap water and bottled water found in some countries. Inappropriate disinfection, improper storage conditions, the problem of the bottled water production process in the factory, and the presence of microbial contamination in the source of bottled water, may cause undesirable changes in color and taste, and in the more severe cases, an increase in the burden of microbial contamination. Therefore, the observance of sanitary regulations during the production and packaging of bottled water, and the care and maintenance of the equipment, and the proper maintenance of packaging compounds are of great importance.

The free residual chlorine was absent in the water samples as the free chlorine concentration gradually decreases over time by storing the chlorinated water. The other studies also strongly suggest this finding.[25,26]

The advantage of this study is that the water samples of 150 ml each were appropriately collected in sterile glass bottles of 200 ml capacity which were obtained from the 20 L bubble top water cans used for drinking purposes. The sampling bottle was not filled up to the brim and 50 mm space was left for effective shaking of the bottle. Water samples were properly labeled and transported to the medical college within 4 h at room temperature so that there might not be any changes in the content of the samples. It is also very useful as it has been done in rural areas where the people are not aware of the quality of drinking water and helps them use a better quality of water for drinking purposes.

The limitation of the study is that it could not correlate the results obtained and its effects on the people using this water as their main drinking source for a long duration.

Conclusions

The study reports that none of the samples showed free residual chlorine which indicates that the chances of contamination of the water are very high. About 41% of the samples show confirmed contamination with *E. coli,* and hence they cannot be used for drinking purposes as they can cause diarrhea. The other 59% samples showed Gram-negative bacteria other than *E. coli* which indicates that there is environmental contamination which is also unfit for drinking. Hence, we will advise the people to boil and drink water.

This infers the noncompliance by the local manufactures with stipulated guidelines in the production process. The distributors, retailers, and consumers also need to be educated about the identification, reporting, and removal of problematic bottled water available at points of sale. An improved hygiene education program is needed to instill deep-rooted inherent behaviors such as hand washing before water handling as well as proper protection of container stored water as boiling before their use from environmental contamination.

Financial support and sponsorship

Financial support and sponsorship: Apollo institute of Medical Sciences and Research (AISMR), ICMR -STS.

Conflicts of interest

There are no conflicts of interest.

References

1. WHO; UNICEF. Progress on Sanitation and Drinking Water. United Nations Plaza, New York, USA: World Health Organization (WHO);2012.
2. Bottled Water India. The Indian Bottled Water Market: Unveiling its Thirst. IKON Marketing Consultants India. Available from: http://bottledwaterindia.org/indian-bottled-water-market/. [Last accessed on 2019 Sep 15].
3. Kean E, Walker LT, Ogutu S, Shackleford L, Verghese M. Chemical and Microbial Contaminants in Bottled Water from Northern Alabama. New Orleans, USA: Culled from Presentation at the 2005 Institute of Food Technologists Meeting; 2005. p. 15-20.
4. Gangil R, Tripathi R, Patyal A, Dutta P, Mathur KN. Bacteriological evaluation of packaged bottled water sold at Jaipur city and its public health significance. Vet World 2013;6:27-30.
5. Semerjian LA. Quality assessment of various bottled waters marketed in Lebanon. Environ Monit Assess 2011;172:275-85.
6. Kassenga GR. The health-related microbiological quality of bottled drinking water sold in Dar es Salaam, Tanzania. J Water Health 2007;5:179-85.
7. Agrawal A, Pandey RS, Sharma B. Water pollution with special reference to pesticide contamination in India. J Water Resour Prot 2010;2:432-48.
8. Mathur HB, Johnson S, Mishra R, Kumar A, Singh B. Analysis of Pesticide Residues in Bottled Water. New Delhi, India: Centre for Science and Environment; 2003.
9. Bureau of Indian Standards. Manual for Packaged Drinking Water. New Delhi, India: Bureau of Indian Standards; 2005.
10. Ministry of Water Resources, GOI. Ground Water Brochure. Ranga Reddy District, Andhra Pradesh: Central Ground Water Board; 2013.
11. Greenwood D, Barer M, Slack R, Irving W. Medical Microbiology: A Guide to Microbial Infections: Pathogenesis, Immunity, Laboratory Investigations. UK: Elsevier; 2012.
12. Pal R. Genetics of insect vectors of disease. WHO chronicle. 1967; 21:348.
13. WHO. Vector Control in International Health. Geneva: World Health Organization (WHO); 1972.
14. Collee JG, Fraser AG, Marmion BP, Simmons A. Mackie and McCartney Practical Medical Microbiology. 14th Edition. New Delhi: Elsevier;1996.
15. Joseph N, Bhat S, Mahapatra S, Singh A, Unissa A. Bacteriological Assessment of Bottled Drinking Water Available at Major Transit Places In Mangalore City of South India. Journal of Environmental and Public Health. 2018.
16. Gangil R, Tripathi R, Patyal A, Dutta P, Mathur KN. Bacteriological evaluation of packaged bottled water sold at Jaipur city and its public health significance. Vet World 2013;5:27-30.
17. Jain U, Bist B, Lalwani DD. Assessment of microbiological quality by coliform estimation in drinking water sources of Mathura region. IOSR J Pharm 2012;2:500-03.
18. Sharma B, Kaur S. Microbial evaluation of bottled water marketed in North India. Indian J Public Health 2015;59:299-301.
19. Reddy PS. Microbiological analysis of bottled water. Indian J Med Microbiol 2000;18:72-6.
20. Rao KY, Anjum MS, Reddy PP, Monica M, Hameed IA, Sagar GV. Physico-chemical and bacterial evaluation of public and packaged drinking water in Vikarabad, Telangana, India – Potential public health implications. J Clin Diagn Res 2016;10:C01-7.
21. Singla A, Kundu H, Basavaraj P, Singh S, Singh K, Jain S. Physico-chemical and bacterial evaluation of packaged drinking water marketed in Delhi – Potential public health implications. J Clin Diagn Res 2014;8:246-50.
22. Venkatesan KD, Balaji M, Victor K. Microbiological analysis of packaged drinking water sold in Chennai. Int J Med Sci Public Health 2014;3:472-76.
23. Halage AA, Ssemugabo C, Ssemwanga DK, Musoke D, Mugambe RK, Guwatudde D, *et al.* Bacteriological and physical quality of locally packaged drinking water in Kampala, Uganda. J Environ Public Health 2015;2015:942928.
24. Salehi I, Ghiasi M, Rahmani AR, Sepehr MN, Kiamanesh M, Rafati L. Evaluation of microbial and physico-chemical quality of bottled water produced in Hamadan province of Iran. J Food Qual Hazards Control 2014;1:21-4.
25. Al-Jasser AO. Chlorine decay in drinking-water transmission and distribution systems: Pipe service age effect. Water Res 2007;41:387-96.
26. Chowdhury S, Rodriguez MJ, Serodes J. Model development for predicting changes in DBP exposure concentrations during indoor handling of tap water. Sci Total Environ 2010;408:4733-43.