RESEARCH ARTICLE

BACTERIOLOGICAL STUDY OF PACKAGED DRINKING WATER MARKETED AS BULK PACK IN MANGALORE, KARNATAKA

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ABSTRACT

Background: *Consumption of bottled water is increasing rapidly in developing countries especially among the middle and high income earners as it is generally perceived to be pure, clean and of good quality. This has led to the sale of different brands of bottled water in the market. Although disease outbreaks due to contaminated bottled water are rare, any contamination may pose a unique hazard because of the widespread distribution.*

Objective: *The study was designed to evaluate the bacteriological quality of the bulk packaged drinking water marketed in Mangalore, Karnataka.*

Material and Methods: *One hundred ten samples of bulk packaged drinking water of 7 different brands were selected for the present study. Two hundred ml of water sample was collected from each container with sterile precautions. The water samples were subjected to bacteriological study using total viable count, presence - absence test, presumptive coliform count and differential coliform count.*

Result: *Thirty out of 110 samples had viable count much higher than specified by Bureau of Indian Standard. Twenty eight samples were found to be positive in presence absence test. Further all the 28 samples showed the presence of coliforms and 26 samples confirmed the presence of Escherichia coli.*

Conclusion: *Packaged drinking water marketed as bulk quantity need not to be safe always. It is advisable to go for the packaged drinking water sold in smaller quantity and with ISI certified brands.*

Key Words: *Bulk packaged, E.coli, Viable count, Presence - Absence test, Presumptive coliform count, Differential coliform count*

INTRODUCTION

Water is an essential requirement of all life forms. Pure and safe drinking water has always been a necessity. Traditionally pipe water distributed by municipalities has been the trusted water supply for drinking purpose. Packaged drinking water is generally considered safe and is taken granted by people without question. Water related diseases continue to be one of the major health problems globally. In developing countries 80% of all diseases are related to drinking water. In India as about 70% of surface water resources are being contaminated by biological, toxic, organic and inorganic pollutants. Rapid urbanization, industrialization and increase in human and animal population densities had a remarkable impact on global ecology and environment quality. The use of bottled water in the world includes mainly North America (30%), Europe (29%), Asia (27%) and other part of world (14%). The bureau of Indian Standards (BIS) has come out with standards for packaged drinking water. [1, 2, 3] Though, we are not aware of any reports of transmission of waterborne diseases through bottle water, such possibility cannot be denied. [4,5] People are concerned about poor quality of tap water and that is why they have switched over to bottled water perceiving it to be clean and safe. For various reason water supply authorities are unable to provide potable water in our country. Majority of population depends on packaged drinking water and further going for bulk quantity of packaged water. Bulk quantity of packaged water is the present trend because of the cost factor and convenience. In India, there is an astronomical increase in the consumption of packaged waters especially bulk pack. The increase demand for drinking water products is attributed largely to factors such as inadequate or non-availability of reliable, safe water in urban areas. The association between water and disease has been known for long. The first authenticated report of water borne disease from broad street water pump London was given by John snow. Since then water borne diseases out breaks from different parts of world including India.[6] Several studies has been documented the detection of coliforms and heterotrophic bacteria in bottled drinking water counts which far exceeded the national and international standards set portable water for human consumption. As far as our knowledge no microbiological study was dealt with till today by subjecting bulk pack of 20 litres or more purified water available in market. The main objective of the current study was designed to evaluate the bacteriological quality of packaged drinking water marketed in Mangalore as bulk pack and to check their compliance with the standards.

MATERIAL AND METHODS

The present study was designed to collect a total of 110 samples from 7 brands of available water as 20 litres bulk pack marketed in Mangalore. Sample were categorized in 3 types, category I included sample from the place where more than 10 bulk packs of different brands of water were used per day/occasion. These were obtained mainly from large scale domestic/academic/official functions like weeding, conferences and workshops etc. Category II represented sample from the places where more than 2 bulk pack of water were used per day/occasion mainly from birthday parties and other small scale functions. Category III consisted of sample from places where less than 1 bulk pack of water was used per day like offices, shops and houses. All the sample of above 3 categories was further classified as national/ local brands and ISI / non ISI certified. Out of seven brands, one was national brand and six were local brand. Further out of six local brands one was non-labelled and non ISI certified, rest all were ISI certified. Two hundred ml of water from each source were collected in sterile container as an end user. The samples were transported to the department of Microbiology Kasturba Medical College Mangalore and processed immediately. All the water samples were subjected to their bacterial analysis as per the standard procedure. [6, 7]

Viable count –spread plate method: One hundred µl of water sample each were spread out by glass rod on two well dried Nutrient agar plates. One plate was incubated at 37°C and the other one at room temperature. After overnight incubation the numbers of colonies were counted and viable count per ml of water was estimated. Samples showing the viable count greater than 200 CFU/ml of water were considered as unsatisfactory as per BIS norms.

Presence - Absence test: *E.coli* was taken as indicator organism for faecal contamination of water. Fifty ml of test water sample was added to 50 ml of double strength MacConkey broth with bromocresol purple in a glass bottle with inverted Durham's tube. After that the inoculated bottle was incubated at 37^0C for 24 to 48 hours. The bottle showing both acid and gas production was considered as positive and the bottle without acid/gas production was considered as negative for presence- absence test.

Multiple tube method: The samples showed positive result in the presence absence test were further subjected to multiple tube method for quantitative estimation of coliforms.

Presumptive coliform count: Five bottles of 10 ml double strength MacConkey broth and 5 bottles containing 5 ml of single strength MacConkey broth with Durham's tube were taken and to which 10 ml and 1 ml of test water samples were added respectively. The bottles were incubated at 37^0C for 24 -48 hours and examined for the production of acid and gas as positive result. Based on the numbers of bottles shown positive results the presumptive coliform count was estimated and expressed as MPN/100 ml of water.

Differential coliform count: The tubes showed positive results in presumptive coliform count were further subjected to differential coliform count by Eijkman's test. One loop full of the inoculum from each positive tube was inoculated into test tube containing single strength MacConkey broth with Durham's tube and the tubes were incubated at 44^0C in water bath for overnight. The tubes shown both acid and gas production, were considered as positive and the differential coliform count was expressed as MPN/100 ml of water. At the end the results were tabulated and subjected to statistical analysis.

Table 1: Bacteriological Analysis of Bulk Packaged Drinking Water

Serial No. Brands - Local /National ISI certified or not	No. of samples tested	No. of samples showed colony count with in BIS specification	No. of samples showed colony count > than BIS specification	No. of samples showed positive result in Presence - Absence test.	No. of samples positive for presumptive coliform test	No. of samples positive for Confirmed *E.coli* test
A. Local Brand, ISI certified	50	-	-	-	-	-
Brand 1	12	6	6	6	6	6
Brand 2	18	14	4	4	4	3
Brand 3	8	8	0	0	0	0
Brand 4	4	4	0	0	0	0
Brand 5	6	4	2	1	1	1
Brand 6	2	0	2	2	2	2
B. National Brand, ISI certified	56	-	-	-	-	-
Brand 7	56	42	14	13	13	12
C. Unlabeled Non ISI certified	4	2	2	2	2	2

Table 2: Category Wise Bacteriological Analysis of Bulk Packaged Drinking Water.

Category	Size of the samples tested	No. of samples showed viable count higher than BIS specification and percentage	No. of samples positive for presence absence test	No. of samples positive for presumptive coliform	No. of samples positive for confirmed *E.coli*
I	10	5(50%)	4	4	3
II	42	14 (33.33%)	13	13	12
III	58	11(18.96%)	11	11	11

DISCUSSION

This study was designed to assess the potable quality of bulk packaged bottled drinking water with regard to their qualitative and quantitative bacterial contamination. Bottled water is considered to be treated water. The steps of treatment include chlorination, exposure to UV, membrane filtration and ozonation. Hence as per BIS standard the permissible viable count of bottled water should not exceed 100 CFU/ml at room temperature and 20 CFU/ml at 37^0 C. Total of 80 out of 110 samples had viable count with in BIS norms, thus suitable for consumption. Rests of the 30 samples had a viable count much higher than BIS specification and were considered as unsatisfactory. Further 28 were positive in presence -absence test and also showed presumptive coliform. Twenty six of them confirmed the presence of *E.coli* suggesting fecal contamination. The large majority (50%) of the samples belonged to category I, 33.33% under Category II and 19% under Category III. It is highly unexpected to have coliforms in packaged drinking water. It clearly indicates the laxity in exercising stringent quality control measures by the manufacturers. The presence of *E.coli*, that too in the large majority of the bulk pack bottled water of category I, can't deny the racket of bulk pack bottled drinking water market. Further the presence of *E.coli* in water samples bottled more than one month back strongly supports the possibility of just filling untreated water in the containers of different brands and pushing on to the consumers. In occasions like large functions there may not be proper monitoring of the quality of the water supplied by the vendor. The microbial contamination of packaged drinking water could be influenced by factors such as their raw water source, treatment process employed and hygienic practices observed in production water manufacturers are observed to utilize well water or at best shallow, contaminated boreholes and municipal tap water as raw water source. Well water is usually contaminated by surface waters especially during the rainy season and inadequate attention paid to the environmental sanitary qualities these wells. Wild animals and birds may also constitute natural sources of zoonotic pathogens contaminating surface and well water. In this study, the coliform positive samples were also tested for faecal coliform faecal coliform is considered more as an indicator of faecal contamination because coliform can exist in the environment, faecal coliform are non-disease causing organism which are found in the intestinal tract of animals. Hence its presence is indicative of contamination with animal or human water. The presence of *E.coli* in water is nearly always associated with recent faecal pollution and it is the preferred indicator organism for this purpose. This study showed the presence of some microorganism in the water samples. Previous studies in other parts of the country reported similar bacterial contamination indicative of poor water quality. Scarcity for potable water has pressed people to go for bottled drinking water. The general acceptance that the bottled water is safe for drinking rarely makes a consumer to suspect its potable quality. Water intended for human consumption should not only be wholesome but also safe. The safety of bottled drinking water should be ensured through comprehensive regulatory programs at both central and state levels before its release for public use.

RESULTS

Table 1 shows the result of bacteriological analysis of 110 samples of water belonging to 7 different brands. Thirty out of the 110 samples had viable count higher than specified by Bureau of Indian Standard. Twenty eight samples were found to be positive in presence-absence test. Further all the 28 samples showed the presence of coliforms and 26 samples confirmed the presence of *E.coli*. Table 2 shows the result of bacteriological analysis of bottled water category wise. Five out of 10 samples of category I found to be unsatisfactory with the count higher than BIS norms. Four were positive in presence- absence test, 4 showed the presence of coliforms and 3 confirmed the presence of *Esch.coli*. Coming to category II, 14 out of 42 samples tested were unsatisfactory with 13 and 12 positive in presence - absence test, presumptive and differential coli form count respectively. The water quality of category III found to be slightly better with 11 out of 58 numbers of samples were found to be unsatisfactory.

CONCLUSION

Our study concluded that bulk pack bottled drinking water cannot be taken for granted as safe for consumption all the time. The manufacturer should exercise stringent quality control measures at all stages of treatment of water. ISI has a system of random sample checking of all their licensee products, which should be strictly implemented. It may be advisable to go for bulk pack bottled water manufactured by reputed brands and having ISI certification and further not to use bulk pack bottled drinking water in large functions where the quality of the same is not assured. These measures need to be strictly followed to prevent water borne diseases.

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