The Health-Related Microbiological Quality of Bottled Drinking Water Sold in Gondia District of Maharashtra, India

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ABSTRACT

Bottled water is drinking water (e.g., well water, distilled water, mineral water, or spring water) packaged in plastic or glass water bottles. Now a day there is an increased demand and consumption of packaged drinking water hence there has been a growing concern about the quality of this product. The safety of water depends on various aspects from its production to final consumption in such a way so that either any microbial contamination can be prevented or it will be reduced to levels not harmful to health. The present study deals with the bacteriological quality of packaged drinking water bottles sold in Gondia, Maharashtra to know microbiological quality of locally produced packaged drinking water brands. Out of 50 bottled waters were collected from Gondia City, 28 (56.0%) bottled water were contaminated with the pathogenic microorganisms, whereas 22 bottled water (44.0%) did not show any pathogenic microorganisms. Surveillance of bottled water manufacturing industries in the settings on a regular basis needs to be done by regulatory agencies which will ensure safe and wholesome bottled water for public usage and health authorities should do intensify efforts in the routine monitoring of activities in the purified bottled water industry in order to supply safe and healthy water to the public. Hence there is a need for strict and routine monitoring of the packaged drinking water with the view of raising their standards which may pose higher risk of microbial exposure.

INTRODUCTION

Water is the utmost important requirement of life. It is essential for the health and survival of human beings. Water is a key to social equity, environmental stability, and cultural diversity. If we see the culture of ancient times, with all the great religions of the world, it will be seen that water is much more than an economic issue. Water is directly linked with spiritual values. Access to safe drinking water is important as a health and development issue at a national, regional, and local level [1]. Packaged drinking water can be described as any product, including natural spring or well water, taken from municipal or private utility systems or other water, distilled water, or any of the foregoing to which chemicals may be added and which are put into sealed bottles (food-grade), packages or other containers, to be sold for domestic/human consumption (potable water) or cooking used [2,3]. Consumption of bottled water is rapidly increasing in developing countries especially, middle- and high-income earners as it is generally perceiving to be pure, clean, and of good quality [4]. Today, bottled water is primarily used for drinking but has also been considered for the preparation of baby formula, cleaning contact lens or used in humidifiers. Bottled water is generally perceived by consumers as being of better quality and taste than tap water. However, the good quality of bottled water is sometimes a misconception. Furthermore, the collection, packaging, transport, and disposal of plastic bottles hurt the environment. Water is a necessity of human life but it has to be pure and safe. It is, therefore, necessary that everyone has access to a safe, affordable, and sufficient quantity of water for drinking, other personal use, and sanitation. The main objective of the introduction of bottled water in society was its better safety, taste, and convenience over tap water [5]. Traditionally, pipe water distributed by the municipalities has been the trusted water supply for drinking purposes. In the earlier days, water available from the wells and springs used to be considered safe and was stored in earthen pots or brass containers. The tradition and style of serving drinking water in India, has however changed quite dramatically during the last decade. Almost a decade ago, the introduction of bottled water or “packaged mineral water” has changed the tradition of serving and consuming drinking water. This has ushered in very strongly, the use of polymers or plastics as materials for water storage and distribution. Almost all the major international and national brand's water bottles are available in the Indian market right from the malls to railway stations, bus stations, grocery stores, and even at panwala's shop. Before a few years ago, bottled water was considered as the rich people's choice, but now it is penetrated even in rural areas. The non-availability of safe drinking water around the world and particularly in third world countries has opened new avenues for the bottled water Industry. The growing number of cases of water-borne diseases, increasing water pollution, increasing urbanization, increasing scarcity of pure and safe water, etc. has made the bottled water business just like any other business in consumer items. The scarcity of potable and wholesome water at railway stations, tourist spots, and the role of the tourism industry in promoting bottled water, etc. have also added to the growth in the consumption of bottled water [6]. Increasing health concerns and unavailability of clean drinking water have led to the growth of the bottled water market in India. The major bottled water brands operating in India are Bislery, Kinley, and Aquafina. In India, bottled water is sold in four main types of stock-keeping units (SKUs) - one-liter bottles, two-liter bottles, 500-milliliter bottles, 250-milliliter bottles, pouches, and barrels of 15-20 liters. Among the different SKUs, one-liter bottles have acquired the largest market share of ~42% in 2018, followed by 500-milliliter bottles and 250-milliliter bottles. The packaged bottled water industry is segmented into:

Packaged drinking water

Packaged drinking water means water derived from any source of potable water and subjected to various treatments to meet the prescribed standard before being packed in a plastic or glass container.

Natural mineral water

Water containing less than 250 ppm of total dissolved solids is defined as natural mineral water. Packaged drinking water, which holds about 85% of the market, is witnessing strong growth owing to various factors such as changes in lifestyle, increase in Foreign Tourist Arrivals (FTAs), health awareness, etc. Natural mineral water on the other hand, which falls under the premium water segment and is primarily consumed by the urban populace owing to its high cost, will witness the growth that will be restricted to urban consumers. The Bureau of Indian Standards (BIS) had formulated Indian Standard (IS) for packaged drinking water (PDW) and packaged natural mineral water (PNMW) under mandatory BIS certification as per Food Safety and Standards Regulation (FSS). This implementation led major players to acquire ISI and BIS certifications to avoid any cancellation of licenses. The increase in the number of domestic and foreign tourists in the past years, and in the years to come is expected to be a major driver for bottled water sales in India. India is rated among the top ten countries in the world concerning bottled water consumption. The fastest-growing industrial sectors are the bottled water production companies, presently in India; more than 3400 bottling plants are in operation [7]. Contaminated water poses a massive health risk to India, as millions suffer from diseases such as diarrhea, dysentery, typhoid, jaundice, and gastroenteritis every year. Several studies have documented the detection of coliforms and heterotrophic bacteria in bottled/ sachet drinking water counts which far exceeded the national and international standards set for potable water for human consumption [8]. Heterotrophic bacteria are non-coliform species of bacteria that utilize organic substances for their development. The presence of heterotrophic bacteria in drinking water is not an indication that the water presents a health risk but poses a significant health risk in immune compromised individuals. Similarly, total coliform bacteria may cause a health risk for infants, young children, elderly people, and especially to the immunocompromised individual [9]. The most reliable source of drinking water is bottled water which is of good (bacteriological) quality [10], but it is expensive and thus only within the means of prosperity in the society. While some apply sophisticated techniques such as ozonation and reverse osmosis, most use ordinary boiling of well water sources, and exclusion of particles by waterborne diseases constitutes one of the major public health hazards in developing countries. Worldwide, in 1995, contaminated water and food caused more than 3 million deaths were recorded, of which more than 80% were among children under age 5 [11-14].

Aim and objectives

Nowadays there is an increased demand and consumption of packaged drinking water hence there has been a growing concern about the quality of this product. From a microbiological point of view, the safety of water depends on various aspects from its production to final consumption in such a way that either any microbial contamination can be prevented or it will be reduced to levels not harmful to health [1]. Therefore, the present study deals with the facts and awareness about bacteriological contamination present in the packaged drinking water, Comparative analysis of the known and locally produced packaged drinking water brands, and assess the bacteriological quality of packaged drinking water samples sold in Gondia, Maharashtra.

MATERIALS AND METHODS

The Gondia city is a district place of Maharashtra State, belongs to the Vidarbha region. The city is surrounded by Goregaon Taluka towards the south, Amgaon Taluka towards the east, Kirnapur Taluka (Balaghat District, MP) towards the north and Tirora Taluka towards the west. In the present study, both analytical and descriptive methodology was adopted and The generation of data was made by using qualitative and quantitative approaches which were carried out under three steps i.e., survey, sample collection, and analysis of samples, which was followed by statistical analysis and interpretation of results.

Collection of sample

A total of 50 samples, Intact and sealed water bottles of the 1-liter capacity of different brands were collected from different shops present in Gondia City.

Isolation of microorganisms from water bottles

Bottles were washed externally with sterilized distilled water and thoroughly cleaned by swabbing with cotton soaked in 70% alcohol. Bottles were opened in the aseptic condition in laminar in front of the flame. 1 mL water was withdrawn from the bottle with a sterilized pipette and poured it aseptically in to test tube containing 5 mL MacConkey Broth Purple with inverted Durham's tube. The solution was kept for incubation at 37°C for 48-72 Hrs. After incubation, tubes were observed for the development of yellow color as acid and formation of gas in Durham's tube as well as turbidity. Loop full culture from positive tubes (A+G, A) was inoculated in plates containing CLED medium, whereas loop full culture from negative tubes showing turbidity was inoculated in Cetrimide Agar plate. Plates containing inoculated CLED medium and Cetrimide agar were incubated at 37°C for 24 to 48 Hr. After incubation, characteristic colonies were selected. Take out a portion of the isolated colony and inoculated it in a Nutrient broth tube for further tests. It was kept for incubation at 37° for 24 hr. After incubation, the broth culture was processed for morphological, cultural, and biochemical tests.

Identification of isolated microorganism from bottled water sample:

1. Cultural characteristics: Cultural characteristics of microorganisms were observed by colonies formed on C.L.E.D and Cetrimide Agar plates.
2. Morphological characteristics: Morphology of microorganisms was observed by performing Gram's staining and Motility test.
3. Biochemical characteristics: Biochemical characteristics of isolated microorganisms were observed by performing the IMViC test, Sugar fermentation test, Triple Sugar Iron Agar Test, Catalase Test, Urease Test, Oxidase Test, and Nitrate Reduction Test.

RESULTS AND DISCUSSION

A total of 50 bottled water samples were collected from different shops in Gondia City and a Total of 35 different microorganisms were isolated from collected bottled water samples. Table 1 illustrates information, about several positive and negative bottled water samples collected from the Gondia city market. It is evident that out of a total of 50 samples 28 (56.0%) samples indicated the presence of pathogenic microorganisms, whereas 22 samples (44.0%) (Figure 1) did not show the presence of pathogenic organisms in it. It showed that 56% of collected samples indicated the presence of bacteria in it.

Table 1. Total Positive and negative samples.

Samples	No. of sample	Percent
Positive	28	56.0
Negative	22	44.0
Total	**50**	**100**

Figure 1. Total positive and negative samples.

Table 2 shows information about brand-wise positive and negative bottled water samples collected from Gondia city. It is apparent that out of a total of 50 samples 28 samples were positive whereas 22 samples were negative concerning the presence of bacteria. Samples of a total of 13 brands (Figure 2) were collected in the study;

1. Total 11 samples were collected of brand Bisleri, out of which 7 (63.6%) samples found positive whereas 4 (36.4%) samples were found negative for the presence of bacteria.

2. Total 8 samples were collected of brand Fresh, out of which 6 (75.0%) samples found positive whereas 2 (25.0%) samples were found negative for the presence of bacteria.

3. Total 7 samples were collected of brand Bayoũ, out of which 3 (42.9%) samples were found positive whereas 4 (57.1%) samples were found negative for the presence of bacteria.

4. Total 6 samples were collected of brand Kinley, out of which 4 (66.7%) samples found positive whereas 2 (33.3%) samples were found negative for the presence of bacteria.

5. Total 6 samples were collected of brand Aquafina, out of which 2 (33.3%) samples found positive whereas 4 (66.7%) samples were found negative for the presence of bacteria.

6. Total 4 samples were collected of brand Oxyrich, out of which 3 (75.0%) samples found positive whereas 1 (25.0%) sample was found negative for the presence of bacteria.

7.Total 4 samples of brand Bailey were collected, out of which 1 (50.0%) sample found positive whereas 1 (50.0%) sample were found negative for the presence of bacteria.

8. Total 1 samples each of brands Bisleri Rockstar, Bistra, Jalpa, and Patanjali were collected and were found negative for the presence of bacteria.

9. Total 1 samples each of brand Barista and Drink Pure were collected and were found positive for the presence of bacteria.

It was concluded that even though the packaged water is considered a safe and potable drinking water source (in absence of alternative drinking water resources) but in actual many brands are not up to the mark and observed pathogenic bacteria when analyzed. The International and national brands such as Aquafina, Bailey, Barista, Bayoũ, Bisleri, Drink Pure, Fresh, Kinley, and Oxyrich found positive for the presence of pathogenic bacteria while the brands (including local brand) such Bisleri Rockstar, Bistra, Jalpa, and Patanjali were found negative for the presence of pathogenic bacteria hence considered as safe for consumption. The brands which are found positive for the presence of bacteria are reputed and well-known, their demand and supply are very high. The results observed in the present study are alarming and raise doubt on the quality check mechanism. It is their responsibility to sell safe packaged drinking water as it is directly associated with human health.

In the present study, 56 percent of the total (28 of 50 nos.) analyzed packaged drinking water found positive and four bacterial species are identified viz., *Pseudomonas* sp. *Streptococcus* sp., *E. coli*, and *Staphylococci* sp. The specific types of these species (except *Pseudomonas*) may cause different infections to the human body. *Pseudomonas* bacteria are generally harmless to healthy people while they may cause infection in people who have weak immunity. *Streptococcus* may cause some disorders, including strep throat, pneumonia, and wound, skin, heart valve, and bloodstream infections, *E. coli* are not harmful but some of the strains such as *E. coli* O157:H7, can cause intestinal infection including diarrhea, abdominal pain, fever, and sometimes vomiting and the species of *Staphylococcus spp*. such as S. *aureus* is the most pathogenic, it typically causes skin infections and sometimes pneumonia, endocarditis, and osteomyelitis which commonly leads to swelling. The Local brands of packaged drinking water were found unfit for human consumption and hence it was suggested that government should intensify the efforts in the monitoring of activities in this rapidly expanding industry to supply potable and wholesome water to the public.

Table 2. Brand wise positive and negative samples.

Brand	Total sample	Positive	Negative
Aquafina	6	2	4
Bailey	2	1	1
Barista	1	1	-
Bayoũ	7	3	4
Bisleri	11	7	4
Bisleri rockstar	1	-	1
Bistra	1	-	1
Drink pure	1	1	-
Fresh	8	6	2
Jalpa	1	-	1
Kinley	6	4	2
Oxyrich	4	3	1
Patanjali	1	-	1
Total	**50**	**28**	**22**

Figure 2. Brand wise positive and negative samples.

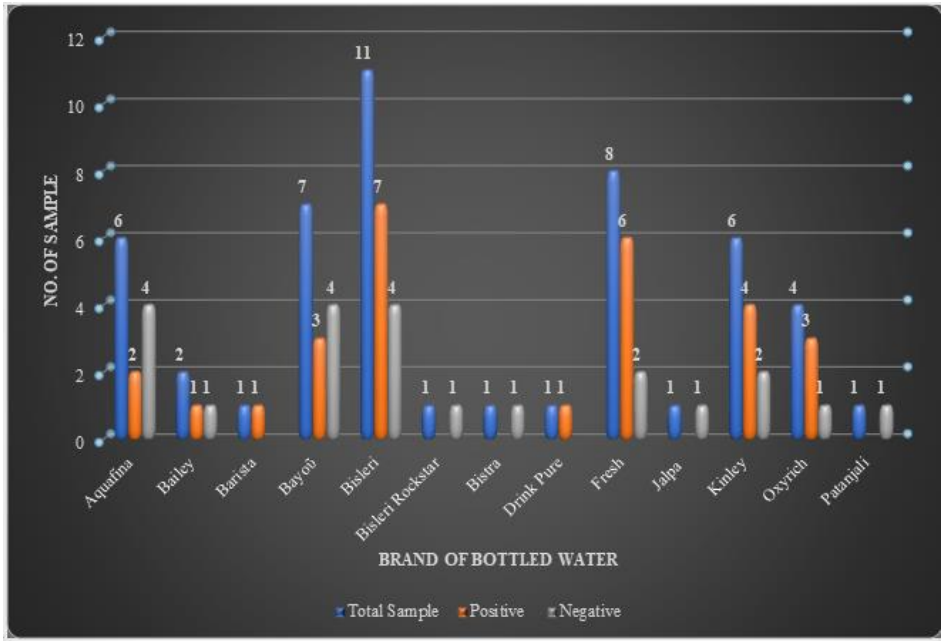

Table 3 shows the results of cultural, morphological, and biochemical analysis of microorganisms isolated from bottled water samples of different brands. In the present study, out of a total of 50 bottled water samples of different brands, *Pseudomonas* spp. was identified in 27 samples, *Streptococcus* spp was observed in 6 samples, *E. Coli* was observed in 2 samples whereas *Staphylococci* spp was observed in 1 sample. So, excess bacterial count in packaged drinking water could pose serious threats to consumers. Therefore, Surveillance of bottled water manufacturing industries is needed to be done by regulatory agencies.

Table 3. Identification of bacteria isolated from bottled water sample.

Characteristics		Type1	Type2	Type3	Type 4
Morphological characteristics	Gram staining	Negative	Positive	Positive	Negative
	Shape	Rod	Cocci	Cocci	Small rod
	Arrangement	Pair, chain	Pair, chain	Cluster	Single, pair
	Motility	Motile	Non-Motile	Non-Motile	motile
Colony Characterstics	Color	Greenish white	Yellowish white	Pale Yellow	Yellow
	Shape	Circular	Circular	Circular	Circular
	Texture	Matted	Smooth	Smooth	Smooth
	Elevation	Raised	Raised	Raised	Flat
	Margin	Rough	Smooth	Smooth	Smooth

Biochemical characteristics	Indole	Negative	Negative	Negative	Positive
	MR	Negative	Positive	Positive	Positive
	VP	Negative	Negative	Positive	Negative
	Citrate	Positive	Negative	Positive	Negative
Sugar fermentation	Glucose	Negative	Positive	Positive	Positive
	Sucrose	Negative	Positive	Positive	Positive
	Lactose	Negative	Positive	Positive	Positive
	Oxidase	Positive	Negative	Negative	Negative
	Catalase	Positive	Negative	Positive	Positive
	Urease	Negative	Negative	Positive	Negative
	Nitrate Reductase	Positive	Negative	Positive	Positive
	TSI	Negative	Negative	Negative	Positive
Inference		***Pseudomonas* spp.**	***Streptococcus* spp.**	***Staphylococcus* spp.**	***E. coli***

CONCLUSION

The supply of safe drinking water to the community is always a major concern. It is a well-known fact that the microorganisms multiply in the pipe of the water distribution system but it is still the subject of research that why they grow in packaged drinking water. It was concluded that these bottled waters were not fit for human consumption and are hazardous to health and strict and routine monitoring of the packaged drinking water is required. Thus, a more stringent standardization of the bottled water market with special attention to quality, identity, and licensing by concerned authorities is recommended to safeguard the health of consumers. It was recommended that the products should be properly stored and bacteriological tests of the air quality and environment of the factory should also be carried out regularly.

It is concluded that the high rate of contamination observed has been of public health importance. There has been a need for use of molecular-based methods to understand microbial ecology, epidemiology, virulence factors, and survival of isolated water-borne pathogens in packaged drinking water sold in Nigeria. Access to safe drinking water is considered to be a human right, not a privilege, for every man, woman, and child. Many microbial pathogens usually spread through the drinking water. It's a common practice nowadays for many people to have packaged drinking water when they are out, considering it the safe and reliable source of drinking water in absence of alternative drinking water sources. The markets also flourished with international, national, and local brands as the demand for the packaged drinking water is increasing day by day.

RECOMMENDATIONS

Carry your bottle of water; do not entirely rely on the packaged drinking water available in the market. The local government or the respective authorities must check the quality of packaged drinking water available in the local market, the government should increase the efforts in the monitoring of manufacturers activities and must inspect regularly, and Industries must assess the quality of bottled water at different stages (production and post-production) to ensure quality and safety. Industries must follow the standard method of consumable water packaging and inspect regularly. Need improved system in the processing and bottling operations to eliminate microbial contamination.

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