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Bacteriological and Physicochemical Quality Of Bottled Water Brands In Ota, Ogun State, Nigeria: Public Health Implications

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ABSTRACT

Water is essential for human survival and must be managed carefully to prevent health risks associated with industrial or bacterial contamination. Hence, this study provided a baseline assessment of the quality and microbiological safety of the most widely consumed bottled drinking water brands in Ota, Ogun State, Nigeria.Nine individual bottled water samples representing nine entirely distinct brands were analysed. Each distinct brand was processed in duplicate for viable bacterial population, pH and chloride content. Gram staining, biochemical tests, and the Analytical Profile Index (API) were used to identify the bacteria. The pH and the chloride content of the samples ranged from 5.19 to 6.96 and 6.92 to 28.58 mg/L, respectively. Six of the nine water samples were below the recommended pH range (6.5 to 8.5) for drinking water. Although a notable negative trend was observed between pH and TVC ($r_s = -.567$, $p = .112$) and a minor positive trend between chloride and TVC ($r_s = .133$, $p = .732$), neither relationship was statistically significant at the 0.05 level. However, causality cannot be inferred from this dataset due to the small sample size.In addition, growth of 7 isolates was observed: *Aeromonas hydrophila*, *Klebsiella pneumoniae*, *Providencia rettgeri*, *Chromobacterium violaceum*, *Enterobacter intermedius*, *Enterobacter cloacae*, and *Escherichia coli*. Even though bottled water was supposed to be devoid of microorganisms, various bacteria were discovered in it, according to these findings. Water distribution systems must be adequately maintained, and awareness of the potential risks of consuming unsanitary water should be improved.

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INTRODUCTION

Water is indispensable for human survival and ecological sustainability, and it serves as a medium for chemical synthesis, including the production and consumption of food and pharmaceutical products. As a result, water must be treated with care at all times, as it is inextricably linked to life, without which there is no existence (Hofstra *et al*., 2026). Maintaining human life and health requires access to high-quality water. Water is essential for several physiological and metabolic processes in the human body (El Baroudi *et al*., 2024). Being able to access clean drinking water is crucial for survival. People at all stages of life, especially young ones, the elderly, and vulnerable populations, including those with impaired immune systems, should not be at risk from unsafe water. Toxins, turbidity, taste, odour, colour, and pathogenic bacteria must all be absent from drinking water to maintain health. These criteria are based on the guidelines of the World Health Organisation (WHO), which are constantly revised to reflect the latest scientific discoveries and data, thereby protecting public health (WHO, 2022). Potable water in Nigeria must adhere to the Standards Organisation of Nigeria (SON) and the National Agency for Food and Drug Administration and Control (NAFDAC) permissible limits of 6.5 to 8.5 (pH) and below 250 mg/L for chloride (Oiganji *et al*., 2020).

The availability of various contaminants in water and the lifespan of bacteria may be influenced by pH and can have a substantial effect on numerous aspects of water quality (Dewangan *et al*., 2007). Furthermore, certain types of water, particularly those in bottles that seem to be safer than tap water, are actually acidic and may cause dental damage (Schmidt & Huang, 2022). Drinking acidic water could be harmful to a person's general and dental health and could lead to enamel erosion (de Paiva *et al*., 2026).

In drinking water, residual chlorine typically exists as a mixture of $HOCl$ and OCl^-, with the exact ionic ratio dependent on solution pH. An increase in the normal chloride content of water may indicate possible pollution from human sewage, animal manure, or industrial wastes. High concentrations of chloride ions can cause water to have an objectionable salty taste and corrode hot-water plumbing systems. High-chloride waters have a laxative effect on some people (Tesfalem *et al*., 2019).

Serious gastrointestinal disorders may result from chemical or microbial contamination. Contamination of bottled water may arise from tainted water sources, including springs, or from factors encountered by workers, equipment, and the environment during bottling (Hamad *et al*., 2022). Water devoid of pathogenic organisms is critical for disrupting one of the primary pathways of disease transmission. Microorganisms play a crucial role in water quality, with *Escherichia coli*, *Salmonella* species, *Vibrio cholerae*, and *Shigella* species among the most commonly implicated in waterborne infections. The presence of *Escherichia coli* and other bacteria is a sign of polluted water (Ibrahim *et al*., 2025). While bottled water is thought to be a safe substitute for tap water, numerous studies reported that the standard limits for bacteria, fungi, and viruses in bottled water have been surpassed (Odeyemi, 2015; Udoh *et al*., 2021; Hamad *et al*., 2022).

Nigerians consume more than 48 billion litres of water annually (Agbasi *et al.*, 2026), and the water sector has grown significantly due to easy access to packaged drinking water. Bottled drinking water is a ubiquitous and essential commodity, presumed to be a safe source of hydration. However, the potential presence of bacterial isolates in the bottled waters raises concerns about water quality and continues to be a challenge in developing countries, particularly in Africa (Anghileri *et al*., 2024).

While previous studies have documented contamination in bottled water, few have quantitatively examined the relationship between physicochemical parameters and bacterial loads in the same samples. Hence, investigating the effects of pH and chloride on total viable bacterial counts in the water is crucial to addressing concerns about the quality of bottled drinking water. This study was designed to provide a critical baseline assessment of the current status and quality of bottled water in circulation in Ota, Ogun State.

MATERIALS AND METHODS

A total of nine distinct commercial brands of bottled water were analysed. All physicochemical and bacteriological analyses were performed in duplicates for each brand. The brands were purchased from different shops, including local outlets in Ota, Ogun State, Nigeria. The bottled water samples were thoroughly checked and confirmed to be in good condition, with the caps and protective seals still in place before purchase. The brands were coded as shown in Table 1. They were taken to the laboratory and analysed for potentiometric pH, chlorinity, total viable counts, and bacterial identification.

Table 1: Codes, Identifiers, and Descriptions of the Tested Bottled Water for Different Brands of Tested Bottled Water

S/N	Sample Code	Anonymised Brand Identifier	Bottle Volume
1	BA	Brand A	75 cL
2	BB	Brand B	75 cL
3	BC	Brand C	75 cL
4	BD	Brand D	75 cL
5	BE	Brand E	75 cL
6	BF	Brand F	75 cL
7	BG	Brand G	75 cL
8	BH	Brand H	75 cL
9	BI	Brand I	75 cL

Brands A-I represent distinct manufacturers. All experiments for codes 1 through 9 were carried out in duplicate (n=2).

Chemical Analysis

Chloride Content

The chloride content was determined as described by Asfaw and Belete (2025). Briefly, potassium chromate (K_2CrO_4) indicator was added to 200 mL of each water sample (at pH 7.0-10). The solution was then titrated with standard silver nitrate ($AgNO_3$). A red-brown colour was observed, and the chloride concentration was calculated.

Hydrogen Ion Concentration

The pH reading of the water samples was carried out using a pH meter, and it was calibrated using standard buffer solutions. The electrode was dipped into the water sample, fully submerged, and the pH displayed on the screen was recorded.

Isolation of Water-Borne Bacteria From Drinking Water / Enumeration of Total Variable Count

Aseptically, 1ml of each of the water samples was suspended in 9 ml of sterile distilled water. Seven serial dilutions: 10^{-1}, 10^{-2}, 10^{-3}, 10^{-4}, 10^{-5}, 10^{-6}, 10^{-7} were prepared. 1 ml of each dilution was poured onto Petri dishes containing sterilised nutrient agar, Eosin Methylene Blue agar, and MacConkey agar. The plates were swirled gently to thoroughly mix the samples with the agar and incubated at 37 °C for 24 hours.

Using the pour plate method with nutrient agar, the bacterial load of the water sample was determined. Microbial growth was closely observed, and the total number of colonies was recorded as the colony-forming unit per millilitre (cfu/ml).

Biochemical Tests

According to the procedures outlined in the Microbiology Laboratory Manual, biochemical tests were carried out on isolates (Cheesbrough, 2006). The biochemical tests carried out on the samples include catalase, urease, citrate, SIM (Sulphur Indole, Motility), and Methyl Red.

Analytical Profile Index (Api)

The API 20E strips contain 20 microtubes with dehydrated biochemical substrates. The microtubes were inoculated with a bacterial suspension and then incubated at 36 °C for 24 hours. After incubation, each microtube showed a colour change or other visible reaction that indicated a positive or negative result for the specific biochemical test. The results were then interpreted using an identification table or a numerical coding system, which can be cross-referenced with an identification database. The key biochemical tests in API 20E are ONPG (detects β-galactosidase activity), ADH (detects arginine dihydrolase activity), LDC (lysine decarboxylase activity), ODC (ornithine decarboxylase activity), CIT (citrate utilization), H_2S (hydrogen sulphide production), URE (urease activity), TDA (tryptophan deaminase activity), IND (Indole production), VP (acetoin production), GEL (gelatinase activity), and sugar fermentation (fermentation of glucose, mannitol, inositol, sorbitol, rhamnose, sucrose, melibiose, amygdalin, and arabinose).

Statistical Analysis

The impact of pH and chloride levels on the total viable counts (TVC) was evaluated statistically using Spearman's rank correlation coefficient (Total Viable Counts (TVC) were transformed to $\log_{10}$CFU/mL before analysis. Results were interpreted as statistically non-significant at p> .05.

RESULTS AND DISCUSSION

Chemical Parameters (pH Values and Chloride Content)

The pH and chloride content values recorded for the samples ranged from 5.19 to 6.96 and 6.92 to 28.58 mg/L, respectively (Table 2).

Total Viable Count (TVC) and Identification of Bacterial Isolates

The mean total viable count of the isolates obtained from the bottled water samples ranged from 2.12 × 10^1 to 5.6 × 10^8 CFU/mL (Table 3). Bacterial isolates identified using the Analytical Profile Index (API), were *Aeromonas hydrophila*, *Klebsiella pneumonia*, *Providencia rettgeri*, *Chromobacterium violaceum*, *Enterobacter intermedius*, *Enterobacter cloacae*, and *Escherichia coli* (Table 4).

Effect of pH and Chloride on the Bacteria's Total Viable Count (TVC)

The TVC was $\log_{10}$ transformed to normalise the distribution before analysis (Table 5). Spearman's rank correlation revealed a notable negative trend between pH and TVC (r_s = -.567, p = .112) and a minor positive trend between chloride and TVC (r_s = .133, p = .732); however, neither relationship was statistically significant at the 0.05 level(Table 6).

Table 2: pH and Chloride Content of the Bottled Water Samples

S/N	Code	pH Value	Chloride Content (mg/L)
1	BA	6.10 ± 0.01	28.58 ± 0.08
2	BB	6.45 ± 0.01	16.86 ± 0.16
3	BC	6.96 ± 0.03	16.66 ± 0.11
4	BD	5.19 ± 0.01	10.30 ± 0.07
5	BE	5.46 ± 0.01	10.83 ± 0.27
6	BF	6.06 ± 0.03	8.34 ± 0.07
7	BG	6.26 ± 0.01	6.92 ± 0.06
8	BH	6.76 ± 0.02	8.52 ± 0.06
9	BI	6.86 ± 0.01	7.63 ± 0.04

pH and chloride content were performed in duplicate and expressed as mean ±Standard Deviation

Table 3: Total Viable Count of Bacterial Isolates in Bottled Water Samples

Sample Code	TVC (CFU/mL)	CV (%)
BA	9300 ± 70.71	0.76
BB	21.20 ± 0.03	0.13
BC	70.00 ± 1.41	2.02
BD	$(5.60 \pm 0.03) \times 10^8$	0.51
BE	1230 ± 2.83	0.23
BF	44.0 ± 1.41	3.21
BG	79.0 ± 2.83	3.58
BH	36.0 ± 1.41	3.93
BI	47.0 ± 1.41	3.01

Total Viable Count (TVC) was performed in duplicate and expressed as mean ±Standard Deviation. CV- Coefficient of variation.

Table 4: Bacteria (n =9) Isolated from Bottled Water Samples

Bottled water (Code)	Bacteria (n=9)
BA	*Enterobacter cloacae*
BB	*Providencia rettgeri*
BC	*Chromobacterium violaceum*
BD	*Escherichia coli*
BE	*Klebsiella pneumonia*
BF	*Aeromonas hydrophila*
BG	*Aeromonas hydrophila*
BH	*Enterobacter intermedius*
BI	*Aeromonas hydrophila*

Table 5: Total Viable Counts (in $\log_{10}$ CFU/mL)

Isolate	Code	TVC (Log_{10} CFU/mL)
1	BA	3.97
2	BB	1.33
3	BC	1.85
4	BD	8.75
5	BE	3.09
6	BF	1.64
7	BG	1.9

Isolate	Code	TVC (Log_{10} CFU/mL)
8	BH	1.56
9	BI	1.67

Table 6: Comparative Analysis between pH, Chloride, and Total Viable Count (TVC) of the Isolates Recovered from the Bottled Drinking Water

Independent variable	Dependent variable	Spearman's rank Correlation (r_s)	P-value (p)	Significance / not significance
pH	TVC	-.5667	.112	Not significant (P>0.05)
Chloride	TVC	.1333	.732	Not significant (P>0.05)

Discussion

Clean and high-quality water plays a vital role in the existence of human life. Drinking water quality is carefully regulated and managed worldwide, with each country setting its national standards to assure the safety of drinking water for the public. Therefore, high-quality bottled drinking water should not pose any severe short-term or long-term health concerns (El Baroudi *et al*., 2024).

Six (66.7%) of the nine water samples recorded in these findings were below the recommended pH range for drinking water, which is 6.5 to 8.5 (Oiganji *et al*., 2020; Kiltu, 2025). As a result, these samples pose a significant public health risk of dental erosion (de Paiva *et al*., 2026). However, all the chloride content (7.63 to 28.58 mg/L) of the water samples complied with the chloride standards and are consistent with a previous study reported by Asoo *et al*. (2012). Chloride levels in public drinking water must not be more than 250 mg/L. Three of the water samples investigated had high bacterial counts exceeding the regulatory limit of $1.0X10^2$ CFU/mL for viable counts in drinking water (Tenebe *et al*., 2023).

According to these findings, pH and chloride did not statistically demonstrate a significant effect on the total viable count, indicating that microbial loads did not appear to be strongly influenced by pH and chloride. Although causality cannot be inferred from this dataset due to the small sample size.A previous study elsewhere discovered that pH values outside the neutral threshold for surface water might not favour the survival of specific groups like coliform bacteria (Aram *et al*., 2021). However, the acid-resistance mechanisms have been previously reported in *Escherichia coli*, particularly the pathogenic strain, which can survive for an extended period of time in a low pH environment (Sheikh *et al*., 2021).

The quality and microbiological safety of bottled drinking water sold in Ota, Ogun State, were the focus of the study. This study found that the bottled drinking water samples examined in the analysis had a higher prevalence of *Aeromonas hydrophila*. *Aeromonas* spp. are possible bacterial pathogens in bottled drinking water (Carusi *et al*., 2024). *Aeromonas hydrophila* detected in this study is consistent with the recovery of *A. hydrophila* from drinking water in Makurdi, Nigeria (Mnguchivir, 2021). Studies have reported *Aeromonas hydrophila* as a zoonotic waterborne pathogen linked to gastroenteritis that possesses virulence and antibiotic resistance characteristics (Abd El-Tawab *et al*., 2021; Jeamsripong *et al*., 2025). Furthermore, *Klebsiella pneumoniae* was detected in one of the bottled water samples, which was also isolated in a previous study by Maduka *et al.* (2014). The presence of *K. pneumoniae*, a possible pathogen, is worrying as they are known to have a high level of antibiotic resistance (Ike *et al*., 2025). *Klebsiella* spp., which are classified as opportunistic pathogens, are capable of environmental adaptation. Due to its adaptability and antibiotic drug resistance, there is a risk that resistance genes could spread (Araujo *et al*., 2025). One of the bacteria isolated was *Providencia rettgeri*, a developing pathogen responsible for traveller's diarrhoea, enteritis, and urinary tract infections (Kuczynski, 2016). *Enterobacter cloacae* was also detected, agreeing with an earlier published study on the microbiological and chemical quality of noncarbonated bottled drinking water, where it was reported that *Enterobacter cloacae* was identified from the bottled drinking water (Herath *et al*., 2012). *Enterobacter cloacae* is not commonly highlighted as a primary waterborne pathogen. Although its presence in water sources can indicate potential contamination and pose health risks, especially to vulnerable populations. A pathogen known to cause chromobacteriosis in humans, *Chromobacterium violaceum*, was isolated in this study. However, it was reported that, for *Chromobacterium violaceum* to be included on the WHO and US Environmental Protection Agency's list of waterborne pathogens, further research is required to determine the prevalence of the disease it causes and the occurrence of the bacteria in drinking water sources (Uba and Eze, 2004). Another bacterium that was detected in this study was *Escherichia coli*. *E. coli* in water samples is a sign of faecal contamination, which requires close observation (Duze *et al*., 2025). In addition, the presence of *Enterobacter* in water can indicate faecal contamination or poor water quality (Li *et al*., 2021).

This study provides valuable insights into the quality of bottled water samples in Ota, Ogun State, Nigeria. However, due to low sample size, future research should aim to enhance the understanding of water safety and quality by studying larger samples of bottled water. Ultimately, addressing these issues will contribute to ensuring the safety of bottled water for all consumers.

CONCLUSION

The microbiological state of the source water and the level of hygiene throughout the extraction and bottling processes affect the safety and microbiological quality of bottled water. The bottled water samples with pH below the recommended range and those with high bacterial counts pose a significant public health risk, including dental erosion and potential gastrointestinal effects for consumers. Therefore, maintaining the infrastructure of water distribution networks is vital to preventing pathogen ingress and thereby protecting community health. Personnel working on drinking water systems must be trained, and awareness of the potential risks associated with unsanitary water consumption should be improved.

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