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Quality assessment of different polyethylene-packaged groundwater in the llorin metropolis of Nigeria for compliance with standards

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ABSTRACT

There is a notable proliferation of sachet water brands in the Ilorin Metropolis of Nigeria. However, more information regarding their safety for human consumption is required. This study aimed to determine the quality attributes of sachet water produced within the Ilorin metropolis of Nigeria to ascertain their conformity with regulatory standards. Three sachet water companies were each selected from three Local Government Areas within the study area. Physicochemical and microbial analyses were conducted on the collected water samples within 24 hours of production and monthly during 4-month storage. The results showed that temperature (25.10-30.13 °C) and turbidity (0.37-2.84 NTU) were within permissible limits. Total hardness (36.0-136.33 mg/L) and pH (6.42-8.86) significantly ($p \le 0.05$) increased during storage and were above standards in some samples. There were significant ($p \le 0.05$) variations in chlorides (2.83-8.57 mg/L), nitrates (0.15-0.64 mg/L), sulphates (4.70-7.12 mg/L), and sodium contents (10.16-18.74 mg/L) but all were within standards. While five of the nine sachet water brands complied (2-100 CFU/mL) with the limit of 100 CFU/mL for the total viable count, all the samples failed the zero-tolerance requirement for coliform. Possible fecal contamination was concluded as the main concern with the sachet water brands produced within Ilorin Metropolis, suggesting their unfitness for human consumption. The water brands may, therefore, pose severe health hazards to consumers and threaten public health. In conclusion, the study demonstrated the likely unfitness of some sachet water brands in the Ilorin Metropolis of Nigeria for human consumption, owing to fecal coliform. This underscores the need for regulatory agencies to intensify the certification process of prospective sachet water factories. Furthermore, regular inspections of certified factories and their practices and products are recommended to ensure continued compliance with regulatory standards.

Keywords: sachet water, physicochemical, microbial, regulatory standards, conformity

INTRODUCTION

Water is essential for various aspects of life, such as drinking, domestic, industrial, and agricultural purposes [1]. Therefore, a reliable supply of clean, wholesome water is vital to promote healthy living in any community. According to a review [2], drinking up to 8 glasses of water may prevent certain chronic diseases. However, models for delivering safe drinking water in developed countries need to be improved, partly due to poverty [3]. The World Health Organisation (WHO) and the United Nations International Children's Emergency Fund (UNICEF) jointly reported that about 66 million people lack access to improved drinking water in Nigeria [4]. Thus, there is an increasing search for low-cost but safe drinking water schemes in the country, a typical example of which is polyethylene-packaged water (subsequently referred to as sachet water). While this strategy is

recommended to ensure sustainable access to potable water in rural and peri-urban settings of developing Nations **[5]**, its effectiveness would depend on consumers' prior awareness of the potential health risks associated with unsafe drinking water **[6]**.

Water pollution seriously threatens public health due to already established contamination sources and emerging ones [7]. While the typical water pollutants may be agricultural, industrial, or environmental, there are growing concerns about evolving chiral pollutants from pharmaceutical and agrochemical residues [8], [9]. Some of these pollutants could significantly impact the safety of the environment and water resources, increasing their associated risks of carcinogenicity and antifertility to humans [10]. As a result, the suitability of ground and surface waters for human use, especially for drinking purposes, becomes doubtful. Meanwhile, groundwater is a common source of sachet water production in Nigeria.

The consumption of sachet water is prevalent in Nigeria. The innovation, popularly known as 'pure water' in local parlance, represents a direct response to the yearnings of many Nigerians for potable water [11]. Akinsola et al. [12] described sachet water as Nigeria's most ubiquitous form of water marketing. Since its introduction in the 1990s, it has constituted one of the fastest-growing industries in the country [6]. During hot weather conditions, consumers typically desire chilled water. To meet this demand, vendors sell chilled sachet water on roadsides and in traffic build-ups [11]. The popularity of sachet water in Nigeria may also be associated with its affordability and perceived hygienic quality. However, the sachet water sector has become very attractive to would-be small-scale investors whose interests lie in making quick profits, leaving much to be desired regarding product quality [11]. To safeguard the safety of sachet water, the National Agency for Food and Drug Administration and Control (NAFDAC) scrutinizes intending manufacturers and their facilities before certification and registration numbers are granted. After approval, the agency conducts follow-up regulatory checks, mainly by end-product monitoring, to ensure that the companies comply with WHO's standards. This approach was, however, deemed insufficient to ensure the safety of sachet water and guarantee public health [5].

According to some past studies, the quality of sachet water brands in some major cities of Nigeria is still being determined. For instance, Yusuf et al. [13] reported that all sachet water samples analyzed in Zaria, Nigeria, were biologically unfit based on the WHO standard for permissible coliform count (0 per 100 mL). More recently, a similar study conducted in one of the Nigerian Federal Universities revealed that two brands of sachet water failed coliform tests [14]. On the other hand, available literature suggests that Nigerian sachet water may generally comply more with physicochemical standards than microbiological standards. Notwithstanding, reports indicated that a notable number of sachet water brands in the country exceeded regulatory limits in some physicochemical parameters, including temperature, pH, Iron, and total hardness [6], [11], [15].

Moreover, prolonged storage of sachet water beyond 12 weeks could result in non-compliance with chemical standards [16]. While these reports present valuable data about the quality of sachet water in some famous cities of Nigeria, their relevance may go only as far as the peculiarities of the studied regions. Due to geographical differences, several agents may influence water quality in a large country like Nigeria. Some reports on the quality of packaged water in Ilorin, Nigeria, are available. However, these were not focused on polyethylene-based packaging, nor were freshly produced samples directly from the production sources considered. For example, Olayemi [17] reported that 40% of bottled and cellophane-packaged water being hawked in Ilorin was unfit for drinking. The author found *Pseudomonas* predominant in the water samples and emphasized the need to institutionalize the drinking water supply system, which was regarded as informal then.

Similarly, 10% of bottled water obtained from different outlet centers within Ilorin did not pass total coliform tests **[18]**. Furthermore, the researchers concluded that 15% of the bottled water was unfit for drinking from the standpoints of bacteriological and physicochemical qualities. Considering the high demand and popularity of sachet water in Ilorin, the current need for more information concerning the physicochemical and microbiological safety of the product calls for more research. This identified gap underlies the novelty of the present study, which sought to provide insights into the extent of compliance of sachet water brands within the study area to regulatory standards set for safe drinking water.

Therefore, this study aimed to determine the physicochemical and microbiological attributes of different brands of sachet water produced in Ilorin metropolis of Nigeria. The water brands were studied within 24 hours of production and after every month during 4-month storage. This investigation was to ascertain their conformity with the safe drinking water standards of regulatory agencies. The research was conducted between 2020 and 2021 in Ilorin, Kwara State, Nigeria.

The three Local Government Areas under investigation are Ilorin East, Ilorin West, and Ilorin South, all located within Ilorin, the largest Metropolitan City of Kwara State, Nigeria (Figure 1). Ilorin is located within the Latitudes $8^{\circ}24'$ N and $8^{\circ}36'$ N, and Longitude $4^{\circ}10'$ E and $4^{\circ}36'$ E [19]. The city's topography was 257 m and 431 m above sea level, receiving water from the South Northward flow of the Oyun and Asa rivers [20]. In terms of climate, Ilorin is characterized by both wet and dry seasons, from April to October and November to April,

respectively [20]. Its mean annual rainfall is around 1222 mm, typically heaviest between June and September [21]. While high temperatures generally prevail between November and January (33-35 °C), as well as between February and April (35-37 °C) [20], relative humidity in Ilorin is average of 70% during the rainy season and 80% during the wet season [21]. The locations of the Local Government Areas being currently studied are as follows: $8^{\circ}61' \text{ N}$, $4^{\circ}79' \text{ E}$ (Ilorin East), $8^{\circ}49' \text{ N}$, $4^{\circ}51'$ (Ilorin West) and $8^{\circ}43' \text{ N}$, $4^{\circ}67' \text{ E}$ (Ilorin South).

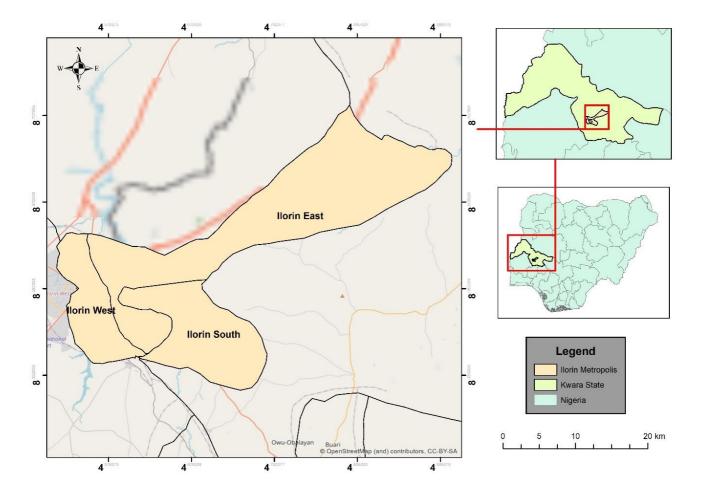


Figure 1 Map of Ilorin, Nigeria, showing the study areas.

Scientific Hypothesis

The compliance of some Nigerian sachet water brands to regulatory standards is still being determined. We expect changes in collected sachet water brands' physicochemical and bacterial properties during ambient storage.

MATERIAL AND METHODOLOGY Samples

Nine brands of NAFDAC-certified sachet water, sourced from groundwater, were collected within 24 hours of production from nine randomly selected pure water companies in the three local government areas. Samples collected were labelled ILM1, ILM2, ILM3, ILM4, ILM5, ILM6, ILM7, ILM8, and ILM9.

Typically, the production of the various sachet water brands involves pumping groundwater from a confined source (bore-hole), followed by its transfer into a storage tank subjected to biological and chemical treatments. In line with the report of Ajala et al. **[2]**, these treatments include chlorination, sedimentation, flocculation, and filtration. On the other hand, the packaging process is automated, i.e., treated water is filled into a sterile polyethylene packaging material by an electronic machine and then sealed by the same machine. Next, the water is manually packed, 20 sachets per bag for distribution. The water treatment and filling occur at the same sites (the factories) where the water is sourced.

Chemicals

All reagents and chemicals used were of analytical-grade quality. Culture media (TVC and MacConkey) were from Hi-media (Mubai, India).

Instruments

The were mercury-in-glass thermometer, Hach portable colorimeter (Model DR/890, Hach Corporation, USA), multipurpose water quality tester (model EZ-9908, Jinan Runjie Electronic Technology, China), spectrophotometer (Hach DR/2000, Hach Corporation, USA), flame photometer (Systronics Flame Photometer 128, Systronics Ltd, India), and atomic absorption spectrophotometer (model 210 VGP, Buck Scientific incorporation, USA).

Laboratory Methods

A mercury-in-glass thermometer was used to determine the temperature of the water samples. The nephelometric method was used to determine the turbidity of the water samples. Before turbidity measurement, the Hach portable colorimeter was first zeroed with 10 mL of deionized water in a cuvette. Each of the water samples (10 mL) was placed in the cell compartment of the Hach portable colorimeter, and the, program 95' was pressed to measure turbidity. Turbidity was expressed in the Nephelometric Turbidity Unit (NTU) [22]. For pH, total dissolved solids, and electrical conductivity, the multipurpose water quality tester was calibrated with Buffer solutions of pH 4, 7, and 10, after which the electrode was rinsed with deionized water. The electrode was then immersed in 50 mL of each of the water samples and allowed to stabilize for 1-2 min. pH, total dissolved solids (mg/L), and electrical conductivity (μ S/cm) were then recorded appropriately.

Total Alkalinity was determined by titration [23]. Two drops of phenolphthalein indicator were added to the 50 mL of water sample in a 250 mL conical flask. This was titrated against 0.2 N H_2SO_4 until the pink colour disappeared. Next, 2 drops of methyl orange were added and the mixture was further titrated with 0.2 N H_2SO_4 until a reddish-orange colour appeared indicating the endpoint.

 $Total alkalinity (mg/L) = \frac{Titre value (mL) x Normality of acid x 50.00x1000}{Volume of sample (mL)}$

Total hardness was assessed using the ethylenediamine tetraacetic acid (EDTA) titrimetric method **[23]** as follows: The water sample (50 mL) was mixed with 1 mL of NH_4Cl-NH_4OH buffer solution and 2 mL of Eriochrome Black T. This was then titrated against 1 M ethylenediamine tetraacetic acid (EDTA) until a blue end point was reached.

Total hardness
$$\left(\frac{\text{mg}}{\text{L}}\right) = \frac{\text{Titre value (mL) x Normality of EDTA x 1000}}{\text{Volume of sample (mL)}}$$

Test for the concentration of nitrates was done using the spectrophotometric sodium salicylate method described by Onweluzo and Akuagbazie [24]. Standard solutions of potassium nitrates (0-5 mg/L) were each mixed with 1 mL of 1% sodium salicylate solution and 2 mL of 96% sulphuric acid and allowed to rest for 15 min. A calibration curve of the working standards was obtained by plotting absorbance values taken at 420 nm in a spectrophotometer against concentrations. To each of the water samples, 15 mL of deionized water and 15 mL of sodium tartrate and absorbance read at 420 nm. Nitrate concentrations in the water samples were then extrapolated from the calibration curve of the standards.

Previously recommended methods by Baird et al. **[22]** were adopted for chloride (Argentometric), sulphate (turbidimetric), sodium (flame emission photometric), magnesium, lead, iron, and copper (atomic absorption spectrophotometric). To test for chlorides, 100 ml of each sample was measured in a conical flask and three drops of 10% potassium chromate indicator were added. The mixture was titrated against 0.014 N AgNO₃ until a pinkish-yellow colour endpoint. The reagent blank was established using distilled water. The concentration of chloride was calculated using the equation:

$$Cl^{-} (mg/L) = \frac{(A-B) \times N \times 35.45}{Volume \text{ of sample}}$$

Where:

A = Titre value of sample; $B = Titre value of distilled water; N = Normality of AgNO_3.$

For sulphates, 100 ml of sample was mixed with 20 ml of buffer solution (30 g/L MgCl₂·6H₂O, 5 g/L CH₃COONa·3H₂O, 1 g/L KNO₃, and 20 mL/L acetic acid) in a conical flask. While stirring, a spoonful of BaCl₂ crystals was added and allowed to mix for 60 s. The turbidity of the mixture developed after 5 min was then read using the spectrophotometer at 420 nm. A sample blank was also analysed using deionized water but without adding BaCl₂. Sulphate concentration was estimated using the calibration curve plotted from different concentrations (0-40 mg/L) of sulphate standards. Concerning sodium, serial dilutions (0 to 10 mg/L) from stock

sodium standard were prepared. Emmison intensities of the standards were then measured at 589 nm in a flame photometer and used to plot a calibration curve. The established calibration curve was used to determine the sodium concentrations in the water samples. As for magnesium, lead, iron, and copper, serial dilutions of 0-4 ppm were prepared from commercial annular grade their respective standard stock solutions. The absorbance values of the working standard solutions were then measured at 285.2, 283.3, 248.3, and 324 nm for magnesium, lead, iron, and copper, respectively, using an atomic absorption spectrophotometer. Calibration curves plotted from the working standard concentrations against absorbance values were used to determine the mineral concentrations in the water samples.

The pour plate method previously described [25], [26] was adapted for the isolation and enumeration of heterotrophic bacteria. From each water sample, 0.1 mL was aseptically transferred using a sterile micropipette onto Nutrient agar in sterile Petri dishes. The nutrient agar had been previously sterilised (121 °C, 0.15 mPa, 15 min) and cooled. The petri dishes were then swirled gently to mix the sample and the media before incubation at 30 °C for 24 hr. After incubation, developed colonies were counted, and the total viable count (TVC) was expressed in colony-forming units per mL (CFU/mL) of the water sample tested. For total coliform analysis, the most probable number method involving presumptive, confirmatory, and complete tests, was carried out according to the method described by Sutton [27]. Briefly, three sets of test tubes in three places (making a total of nine) were used to prepare. The presumptive test was carried out by adding 10 mL of double-strength, 5 mL of single-strength, and 5 mL of double-strength lactose broth into the first, second, and third sets of the test tubes, respectively. This was followed by the placement of inverted Durham tubes in all the test tubes, then sterilization at 121 °C and 0.15 mPa for 15 min. After being cooled to 40 °C, the sterilized test tubes were inoculated with the varied volumes of water sample: 10 mL in double-strength lactose broth, 1 mL in single-strength lactose broth, and 0.1 mL in single-strength lactose broth. These were incubated at 37 °C for 48 hr to examine acid and gas formation as indicated by the colour change of the broth. For a confirmatory test, a loopful from each of the positive fermentation tubes was inoculated in McConkey broth (containing an inverted Durham's tube) and incubated at 37 °C for 48 hr. The tubes with gas production were considered positive. Finally, the complete test was conducted on the positive tubes from the confirmatory stage. This was done by streaking the inocula from the positive tubes in Eiosine Methyle Blue followed by incubation at 37 °C for 24 hr for the growth of Escherichia coli.

Description of the Experiment

Sample preparation: Bags (packs) of the labeled sachet water were immediately conveyed to the laboratory, where subsamples were subjected to physicochemical and microbial analyses. From each pack, a sachet was taken, and cut open from the edge using a pair of sterilized scissors. The content of the sachet was then poured into a sterilized beaker just before analyses.

Number of repeated analyses: Analyses were conducted in three replicates except where otherwise specified.

Number of samples analyzed: 18 samples (i.e., 2 from each brand) were analysed.

Number of experiment replication: The experiment was conducted twice. This involved sample preparation before analyses, as already manufactured sachet water brands were studied.

Design of the experiment: The experiment was designed to determine the compliance of sachet water produced within the study area with physicochemical and microbial standards. First, selected samples were collected within 24 hrs of production and tested for physicochemical and microbial parameters. Next, unopened samples were stored for 4 months, and the same physicochemical and microbial parameters were monitored. Results obtained were evaluated against the permissible limits stipulated by the World Health Organisation and Nigerian Industrial Standards.

Statistical Analysis

Statistical Package for Social Sciences (SPSS, version 20) was used for the statistical analysis. Physicochemical and microbial parameters were statistically subjected to the Analysis of Variance (ANOVA) and the Duncan Multiple Range Comparison Test. Results were reported as means of triplicate determinations \pm SD. Means were considered significantly different at 5%.

RESULTS AND DISCUSSION

The temperature of the water samples significantly ($p \le 0.05$) varied from 25.10 to 27.40 °C before storage and from 26.41 to 30.13 °C after four months of storage (Table 1). This is in line with the findings of Unegbu [28], who also observed a temperature rise while storing sachet water samples in their study. The predominant increase in temperature may be attributed to metabolic activities [27], such as the growth of mesophilic microbes within the optimal temperature range of 20-45 °C [29]. On the other hand, the general temperature drop observed in most

of the samples after the first month could be attributed to some external influences from the storage ambient temperatures [29]. According to NIS, the water temperature should not exceed that of the environment (ambient) in which it is stored [30]. The ambient temperature of the storage environment $(27 \pm 3 \text{ °C})$ in this study is similar to the range of temperature values recorded for the various water brands.

The pH of the sachet water samples ranged from 6.42 to 7.65 during the 24 hours of production (Table 2). The majority of the brands satisfied the WHO recommended limits (6.5-8.5), but the significantly ($p \leq 0.05$) lower pH of ILM1 and ILM9 may impact an acidic taste. pH increased as the storage period progressed, probably due to microbial activities and changing chemical components. However, the pH of all the sachet water samples, except ILM4 and ILM5, remained within the acceptable range throughout the storage period. While a relationship exists between pH and other chemical properties of water, such as copper and sulphates, the interaction is somewhat complicated and varies with different water sources [31]. Therefore, it may not be immediately clear to directly link the changing pH to a particular chemical or physicochemical property, except if such a property is studied in isolation [31]. The simultaneous influence of microbial activity is another factor that may complicate any attempt to predict the relationship between water's changing chemical components and pH. Nonetheless, the pH values obtained showed that most of the sachet water brands were slightly alkaline, which could potentially cause acidosis. The pH results from the present research are consistent with an earlier study by Duru et al. [16] on sachet water brands studied in Owerri metropolis of Nigeria. According to the authors, most of the water samples had pH in the range of 6.85-7.37, with a steady increase during storage. The results are also in line with what was reported by Unegbu [28]. The pH of drinking water is critical to its sensory properties as pH outside the WHO/NIS acceptable range of 6.5-8.5 may constitute an undesirable taste [32]. Water with an acidic pH can be corrosive to metallic pipes [14].

Sample	Storage period (month)									
	0	1	2	3	4					
ILM ₁	$27.30\pm\!0.00^{\rm c}$	$25.60\pm\!0.01^d$	$27.37\pm\!0.05^{\rm f}$	$28.33 \ {\pm} 0.05^{\rm f}$	29.35 ± 0.01^{b}					
ILM ₂	27.07 ± 0.06^{d}	$25.53\pm\!0.05^e$	27.60 ± 0.00^{e}	$28.40\pm\!0.00^{e}$	28.35 ± 0.01^{d}					
ILM ₃	$27.60\pm\!\!0.00^a$	$25.80\pm\!\!0.01^{c}$	$27.80\pm\!0.00^d$	28.90 ± 0.00^{b}	30.08 ± 0.00^{a}					
ILM ₄	$26.80\pm\!\!0.00^{\rm e}$	$25.80\pm\!\!0.01^{c}$	$28.00 \pm 0.00^{\circ}$	$28.80 \pm 0.00^{\circ}$	28.29 ± 0.09^{e}					
ILM ₅	27.00 ± 0.00^{e}	$25.40 \pm 0.01^{\rm f}$	27.60 ± 0.00^{e}	$28.40\pm\!0.00^{e}$	26.41 ± 0.01^{f}					
ILM ₆	27.40 ± 0.00^{b}	26.40 ± 0.01^{b}	$28.10\pm\!0.00^{\text{b}}$	$28.50\pm\!0.00^d$	$28.50 \pm 0.01^{\circ}$					
ILM ₇	25.10 ± 0.00^{g}	$26.80\pm\!\!0.01^a$	27.30 ± 0.00^{g}	29.20 ± 0.00^{a}	30.13 ± 0.02^{a}					
ILM ₈	$26.40 \pm 0.00^{\rm f}$	26.40 ± 0.01^{b}	$28.40\pm\!\!0.00^a$	$27.80\pm\!0.00^g$	28.45 ±0.01°					
ILM ₉	$27.38 \pm 0.06^{\mathrm{b}}$	$26.78\pm\!\!0.04^a$	28.39 ± 0.03^{a}	28.62 ± 0.03^{e}	29.33 ± 0.04^{b}					
LIMITS	Ambient °C (WHO/NIS)									

Table 1 Temperature of selected sachet water brands produced in Ilorin Metropolis over a 4-month storage.

Note: Values are Means \pm SD of triplicate determinations (n = 3); Means with different superscripts along the same column are significantly ($p \le 0.05$) different.

Table 2 pH of selected sachet water brands	produced in Ilorin Me	etropolis over a 4-month.
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Sample	Storage period (month)									
	0	1	2	3	4					
ILM ₁	$6.42 \pm 0.03^{\text{b}}$	$7.83 \pm 0.04^{\circ}$	$7.55 \pm 0.05^{\rm d}$	$7.59 \pm 0.07^{\rm f}$	8.00 ± 0.09^{e}					
ILM_2	$7.65 \pm 0.05^{\rm a}$	7.94 ± 0.03^{b}	$7.55 \pm 0.06^{\text{d}}$	$7.90 \pm 0.05^{\text{d}}$	$8.16\pm\!\!0.06^d$					
ILM ₃	$7.65 \pm 0.04^{\rm a}$	$7.72 \pm 0.06^{\circ}$	$7.88\pm\!0.03^{\rm b}$	$8.10\pm0.03^{\circ}$	$8.33 \pm 0.05^{\circ}$					
ILM_4	7.65 ± 0.01^{a}	$8.26 \pm 0.03^{\rm a}$	$8.62\pm\!\!0.10^{\rm a}$	$8.48 \pm 0.06^{\text{b}}$	$8.52 \pm 0.07^{\text{b}}$					
ILM ₅	$7.65 \pm 0.06^{\rm a}$	$7.88 \pm 0.07^{\rm b}$	$8.40\pm\!\!0.06^a$	8.77 ± 0.07^{a}	$8.86\pm\!\!0.08^a$					
ILM ₆	7.65 ± 0.03^{a}	7.94 ± 0.06^{b}	$7.84 \pm 0.05^{\mathrm{b}}$	$7.97 \pm 0.07^{\rm d}$	$8.32\pm0.05^{\circ}$					
ILM ₇	$7.65 \pm 0.05^{\rm a}$	$7.74 \ \pm 0.05^{d}$	$7.90\pm\!\!0.07^{b}$	$7.97 \pm 0.03^{\text{d}}$	8.09 ± 0.04^{e}					
ILM ₈	7.65 ± 0.08^{a}	$7.64 \ \pm 0.06^{d}$	$7.72\pm 0.08^{\circ}$	$7.82 \pm 0.04^{\text{e}}$	$8.16\pm\!\!0.03^{d}$					
ILM ₉	$6.49 \pm 0.03^{\text{b}}$	$7.00 \pm 0.08^{\rm e}$	$7.73 \pm 0.07^{\rm c}$	7.67 ± 0.05^{d}	8.03 ± 0.10^{e}					
LIMITS			6.5-8.5 (WHO))						

Note: Values are Means \pm SD of triplicate determinations (n = 3); Means with different superscripts along the same column are significantly ($p \le 0.05$) different.

The turbidity values of the sachet water samples were originally in the range of 0.37-0.57 NTU (Figure 2). There was, however, a significant ($p \le 0.05$) difference throughout the storage period, though these were in varying degrees. Ayegbo et al. [33] reported 0.02-0.35 NTU values for sachet water samples sold in Sango-Otta, Ogun State of Nigeria. These are lower than the values recorded in this study. It is, however, worth noting that Ayegbo et al. [33] conducted their study on samples stored at 4 °C, and the age of the samples at the point of collection was not indicated. The rising turbidity recorded during the 4-month storage in this study is comparable to observations reported elsewhere by other scholars. For instance, Ajekunle et al. [23] noted a turbidity rise from 0.39-0.7NTU to 0.74-1.85NTU for sachet water samples after three months of storage. Regarding regulatory compliance, all the sachet water samples conformed with WHO's turbidity limit of less than 5 NTU, implying good physical quality. Turbidity is the physical property of water associated with reduced light transmission owing to absorbance and scattering by solid particles in suspension. Materials causing turbidity may include clay, silt, finely divided organic and inorganic matter, and soluble colored organic compounds. These can come from soil erosion, excess nutrients, various wastes and pollutants, and the action of bottom-feeding organisms [34]. Since the water samples were packaged, the increasing turbidity may be attributed to increased total dissolved solids from microbial activities. Initial turbidity in water may protect some microorganisms from disinfection during water treatment, thereby promoting microbial regrowth during storage [35]. This phenomenon will lead to a further rise in turbidity.

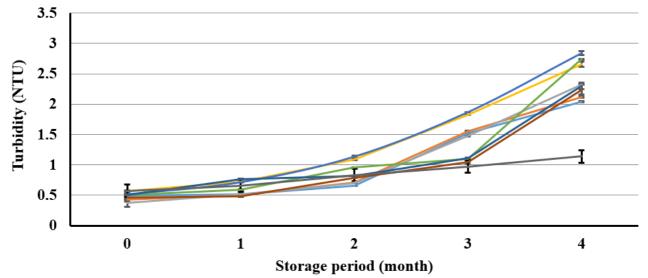


Figure 2 Turbidity of selected sachet water brands produced in the Ilorin metropolis over a 4-month storage. Note: Error bars represent standard deviations of triplicate determinations (at $p \le 0.05$).

There was a significant ($p \le 0.05$) difference in the total hardness of all the sachet water samples within 24 hr of production, varying from 31.33 mg/L to 90.67 mg/L (Figure 3). This is comparable with values (65.21 mg/L-90.08 mg/L) earlier reported for sachet water brands marketed in the Bauchi region of Nigeria [1]. The values also comply with the WHO set limit of 100 mg/L. However, total hardness increased in some samples after the second month of storage, exceeding the WHO limit. The increased total hardness may signal an increased availability of metal ions in the sachet water [36]. Differences in total hardness could be attributed to varying levels of magnesium and calcium in water as influenced by the geology of its source [37]. Water hardness can imply adverse effects such as corrosion and impaired taste [38]. Since it is a function of cations, hardness can also influence the electrical conductivity of water [39].

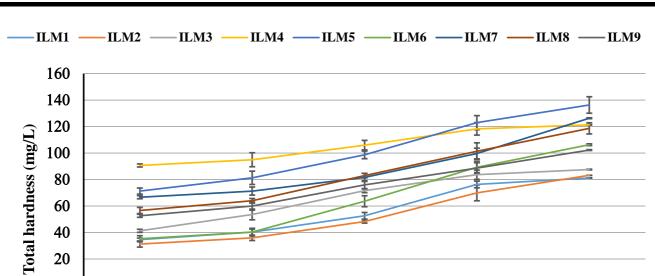


Figure 3 Total hardness of selected sachet water brands produced in the Ilorin metropolis over a 4-month storage. Note: Error bars represent standard deviations of triplicate determinations (at $p \le 0.05$).

2

Storage period (month)

3

4

1

Figure 4 shows the electrical conductivity of all sachet water samples over 4 months of storage. Before storage, electrical conductivity values significantly ($p \le 0.05$) varied between 110.67 µS/cm and 238.00 µS/cm. Though these were followed by some significant ($p \le 0.05$) increase as storage progressed, the values were still below the WHO permissible limit (1000 µS/cm). The increasing electrical conductivity could be associated with rising levels of dissolved materials in the water samples during storage [40]. An elevated temperature of stored sachet water has also been linked to higher electrical conductivity [28]. Electrical conductivity denotes the ability of water to conduct electricity. Previously, Ojekunle et al. [41] similarly reported a significant ($p \le 0.05$) increase in the electrical conductivity of sachet water in the Ibadan metropolis during storage. The authors recorded 60.98-117.35 µS/cm within the first week of storage, but these rose to 63.10-121.05 µS/cm after three months. An electrical conductivity higher than an acceptable level in drinking water may lead to unpleasant taste or gastrointestinal distress [1].

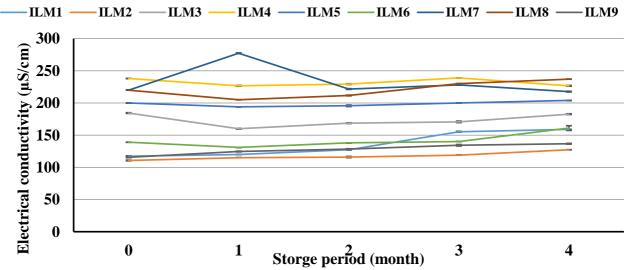


Figure 4 Electrical conductivity of selected sachet water brands produced in the Ilorin metropolis over a 4-month storage. Note: Error bars represent standard deviations of triplicate determinations (at $p \le 0.05$).

Figure 5 shows the total dissolved solids (TDS) in the studied sachet water brands. TDS varied significantly ($p \le 0.05$) from 55.33 mg/L to 120.00 mg/L and further rose during storage. At the end of the storage period, the

0

0

values were 70-125.7 mg/L. The variation in the levels of metabolites from growing microorganisms during storage may account for the changing TDS. Higher TDS may also be associated with possibly increasing metal availability during storage. TDS recorded in this study is higher than values (50-70 mg/L) reported for some sachet water brands marketed in a Nigerian Federal University [14], probably due to geographical and environmental differences. However, the TDS of the sachet water samples remained below the WHO limits of 500 mg/L throughout the storage period. Water can dissolve many inorganic and organic minerals or salts, such as potassium, calcium, sodium, bicarbonates, chlorides, magnesium, and sulphates [40]. These minerals produce undesirable taste and appearance in water. While TDS in water may not directly affect consumers health, it can impair sensory attributes like taste, smell, and color [42].

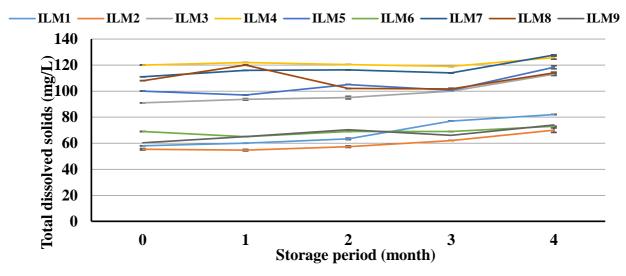


Figure 5 Total dissolved solids of selected sachet water brands produced in the Ilorin metropolis over 4-month storage. Note: Error bars represent standard deviations of triplicate determinations (at $p \le 0.05$).

The chloride concentration of the water samples varied significantly ($p \le 0.05$) from 2.83 mg/L to 5.21 mg/L before storage and from 4.10 mg/L to 8.57 mg/L after the fourth month (Table 3). ILM1 demonstrated the highest increasing rate in chloride concentrations throughout the storage period. However, despite the observed increase in chloride content, its level in all the sachet water samples was within WHO/NIS limits of 200-300 mg/L throughout the investigation period [43]. A similar case was observed by Ajekunle et al. [23] on the sachet water produced in Ogun State, where the chloride concentration of all the sachet water increased with the length of storage. The rise in chloride concentrations may be associated with possible leaching from chlorine-containing compounds into the packaged water during storage. For example, Farinu et al. [36] had earlier hypothesized the possible contribution of leached chemicals, including trichloromethane from disintegrating polyethylene packaging materials, on the increased chloride concentration of stored sachet water. Chlorides are inorganic compounds resulting from the reaction between chlorine gas and metals. Some common chlorides include sodium chloride (MaCl) and magnesium chloride (MgCl₂). Chlorides may get into surface water from several sources, such as rock-containing chlorides, agricultural run-off, industrial wastewater, oil well wastes, and effluent wastewater from wastewater treatment plants [44]. The significance of chlorides in water has been linked with heart and kidney damage, and the sensory properties of water on the other hand [43].

Nitrates were not present in all the sachet water samples when analysed within 24 hr of production. However, during storage, nitrates were recorded with significantly ($p \le 0.05$) different values: 0.25-0.59 mg/L after the first month, 0.15-0.59 mg/L after the second month, 0.22-0.57 mg/L after the third month and 0.42-0.64 mg/L after the fourth month of storage (Table 3). However, there were fluctuations, with values rising during some months while dropping in others. This observation is at variance with the report of Duru et al. [16], who reported a decrease in nitrates within 24 hr of production, and this was attributed to the presence of nitrate-utilizing microorganisms. While the exact reason may account for the similarly decreasing nitrates in this study, it is likely that some contrary biochemical or microbiological processes also took place, which caused rising nitrate levels in some samples. For example, some bacteria produce nitrates from the short-lived intermediates, nitrites [1]. Nitrates in water may initiate the development of methemoglobinemia, otherwise known as a blue baby disease in infants [1]. However, concerning the sachet water samples currently under study, this condition is not of serious concern as the nitrates concentration is below the WHO permissible limits of 50 mg/L. Again, giving a sachet of water to children for drinking is not a common practice.

The values of sulphates in the various water samples differed significantly ($p \le 0.05$) between 5.83 mg/L and 6.40 mg/L within 24 hr of production (Table 4). However, these values generally increased by the fourth month, except for some samples in which lower sulphates were observed. Notwithstanding, all sulphate levels were lower than the maximum of 250 mg/L allowed by WHO/NIS **[43]**. The sulphate content of natural waters is essential in determining their suitability for public and industrial supplies. Beyond the permissible level, sulphates in water may affect human organs, leaving vermifuge consequences on the human system **[43]**. There was no steady pattern in the amount of sulphates during storage, as some fluctuating figures were recorded. Similar fluctuating trends had been previously reported for bottled and sachet water samples stored for six weeks **[45]**. The fluctuating sulphates concentrations in the water samples might be partly due to microbial activities. For instance, Fadipe et al. **[45]** explained that some microorganisms could reduce sulphates in water, which may partly corroborate the declining sulphates noticed in some samples during storage. It has been reported that the discharge of industrial waters and domestic sewage could increase the concentration of sulphates in water **[1]**. Sulphate may have a laxative effect that can lead to dehydration and is of particular concern for infants **[46]**.

The sodium contents of all the nine sachet water samples significantly ($p \le 0.05$) varied from 15.17 mg/L to 18.74 mg/L during 24 hr of production (Table 4), with samples from ILM1 and ILM4 recording the highest and lowest values, respectively. Notably, sodium contents generally decreased during storage except in the second month when about two-thirds of the sachet water samples demonstrated stable sodium. A similar decreasing pattern had been earlier documented by Ajekunle et al. [23] while comparing the sodium contents of some sachet water samples with the WHO limit of 200 mg/L. While the general drop in sodium concentrations is yet to be precisely rationalised, the likely complexation of sodium with other elements probably reduced its detectability during analysis. In general, sodium salts are not acutely toxic because of the efficiency with which mature kidneys excrete sodium. However, acute effects and death have been reported following accidental overdoses of sodium chloride [47]. An excessive salt intake seriously aggravates chronic congestive heart failure, and ill effects due to high sodium levels in drinking water have been documented [47]. Consuming food and water containing high sodium levels can contribute to high blood pressure [48].

	Chlorides (mg/L)										
Sample		Storage period (month)									
	0	1	2	3	4						
ILM ₁	3.56 ± 0.02^{cd}	$3.65 \pm 0.37^{\circ}$	$4.00\pm\!0.08^d$	$4.26\pm0.26^{\circ}$	$4.10\pm\!\!0.02^{d}$						
ILM_2	$2.83 \pm 0.01^{\text{d}}$	$3.43\pm0.33^{\circ}$	$3.80\pm\!\!0.66^e$	4.65 ± 0.36^{bc}	$4.80\pm0.52^{\circ}$						
ILM ₃	$3.56\pm\!\!0.01^{cd}$	$3.72\pm 0.04^{\circ}$	4.46 ± 0.47^{cd}	4.54 ± 0.21^{bc}	$4.54 \pm 0.20^{\circ}$						
ILM_4	4.27 ± 0.02^{bc}	$4.70\pm\!0.16^{\rm b}$	4.94 ± 0.12^{bc}	$5.20\pm\!\!0.16^{abc}$	8.57 ± 0.75^{a}						
ILM ₅	$4.26 \pm 0.71^{\text{be}}$	5.30 ± 0.23^{a}	$5.25\pm\!\!0.28^{b}$	$5.29\pm\!\!0.19^{abc}$	$5.29 \pm 0.19^{\mathrm{b}}$						
ILM ₆	$3.08\pm\!\!0.41^{d}$	$3.56 \pm 0.35^{\circ}$	3.87 ± 0.13^{e}	$4.88\pm\!\!1.18^{abc}$	$6.26\pm\!0.39^{b}$						
ILM ₇	4.97 ± 0.71^{ab}	4.97 ± 0.15^{b}	5.44 ± 0.13^{ab}	$5.86\pm0.39^{\mathrm{a}}$	6.07 ± 0.75^{b}						
ILM ₈	5.21 ± 0.41^{a}	$5.10\pm\!0.30^{\rm b}$	$5.86\pm\!0.13^a$	5.47 ± 0.95^{ab}	$6.22\pm\!0.16^{\rm b}$						
ILM ₉	3.56 ± 0.02^{cd}	$3.72\pm0.41^{\circ}$	$4.02 \pm 0.05^{\text{d}}$	4.54 ± 0.07^{bc}	$4.78 \pm 0.07^{\circ}$						
LIMITS		200-	-300 mg/L (WHO	/NIS)							
			Nitrates (mg/L)								
ILM ₁	0.00^{a}	0.34 ± 0.11^{bcd}	$0.49\pm\!\!0.08^{ab}$	0.37 ± 0.02^{b}	$0.54\pm\!0.06^{\mathrm{b}}$						
ILM_2	0.00^{a}	0.30 ± 0.04 ^{cd}	0.15 ± 0.02^d	$0.22\pm0.01^{\circ}$	$0.44 \pm 0.02^{\circ}$						
ILM ₃	0.00^{a}	$0.25 \pm 0.06^{\circ}$	$0.26\pm0.03^{\circ}$	0.42 ± 0.04^{ab}	$0.42\pm 0.09^{\circ}$						
ILM_4	0.00^{a}	0.59 ± 0.11^{a}	$0.59 \pm 0.04^{\rm a}$	0.57 ± 0.13^{a}	0.64 ± 0.02^{a}						
ILM ₅	0.00^{a}	$0.52\pm 0.16^{\mathrm{a}}$	$0.58 \pm 0.07^{\rm a}$	0.47 ± 0.09^{ab}	$0.56\pm\!\!0.07^{\rm b}$						
ILM ₆	0.00^{a}	$0.44\pm\!0.09^{abc}$	0.47 ± 0.07^{ab}	$0.42\pm\!\!0.08^{ab}$	0.47 ± 0.02^{bc}						
ILM ₇	0.00^{a}	$0.49\pm\!\!0.02^{ab}$	$0.46\pm\!\!0.10^{b}$	0.49 ± 0.09^{ab}	$0.43\pm 0.04^{\circ}$						
ILM ₈	0.00^{a}	0.28 ±0.05°	$0.50\pm\!\!0.03^{ab}$	$0.40\pm\!\!0.03^{b}$	$0.54\pm\!\!0.03^{b}$						
ILM ₉	$0.01{\pm}0.02^{\text{ a}}$	5.21 ± 0.41^{a}	$0.28 \pm 0.02^{\circ}$	$0.29\pm 0.02^{\circ}$	$0.40\pm\!\!0.41^{b}$						
LIMITS			50 mg/L (WHO)								

Table 3 Chlorides and nitrates of selected sachet water brands produced in Ilorin Metropolis over a 4-month storage.

Note: Values are Means \pm SD of triplicate determinations (n = 3); Means with different superscripts along the same column are significantly ($p \le 0.05$) different.

	Sulphates (mg/L)										
Sample	Storage period (month)										
-	0	1	2	3	4						
ILM ₁	$5.00{\pm}0.00^{e}$	4.96±0.26 ^e	5.47 ± 1.09^{b}	6.24 ± 0.25^{b}	$6.28{\pm}0.09^{ab}$						
ILM_2	$4.83{\pm}0.20^{\rm f}$	$5.05{\pm}0.28^{de}$	$5.80{\pm}0.15^{b}$	$5.35 \pm 0.32^{\circ}$	$5.72{\pm}0.05^{bcd}$						
ILM ₃	5.40±0.01°	$5.52{\pm}0.09^{\text{cde}}$	$5.86{\pm}0.02^{b}$	$6.43{\pm}0.38^{b}$	$6.34{\pm}0.06^{abc}$						
ILM_4	$6.40{\pm}0.02^{a}$	5.47 ± 0.31^{cde}	$5.53 {\pm} 0.32^{b}$	5.89 ± 0.13^{bc}	$6.41{\pm}0.03^{ab}$						
ILM ₅	$6.40{\pm}0.03^{a}$	6.94±0.81ª	$6.10{\pm}0.27^{b}$	6.30 ± 0.49^{b}	5.99 ± 0.55^{bcd}						
ILM ₆	6.03 ± 0.01^{b}	$5.77 {\pm} 0.50^{cd}$	5.46 ± 0.24^{b}	5.98 ± 0.16^{b}	5.65 ± 0.77^{cd}						
ILM_7	6.01 ± 0.02^{b}	$6.69{\pm}0.40^{ab}$	6.25 ± 0.13^{b}	$6.54{\pm}0.61^{ab}$	5.48 ± 0.25^{d}						
ILM ₈	$5.26{\pm}0.03^{d}$	5.98 ± 0.22^{bc}	$5.94{\pm}0.56^{b}$	$7.12{\pm}0.34^{a}$	6.85 ± 0.42^{a}						
ILM ₉	4.87 ± 0.01^{e}	$4.85{\pm}0.50^{\rm f}$	$4.70{\pm}0.32^{a}$	$5.42 \pm 0.50^{\circ}$	5.56 ± 0.32^{cd}						
LIMITS		250 r	ng/L (WHO/NIS	5)							
		S	odium (mg/L)								
ILM ₁	18.26±0.07 ^a	4.96±0.26 ^e	5.47 ± 1.09^{b}	6.24 ± 0.25^{b}	$6.28{\pm}0.09^{ab}$						
ILM ₂	$18.45{\pm}0.07^{\text{ a}}$	$5.05{\pm}0.28^{de}$	$5.80{\pm}0.15^{b}$	5.35±0.32°	5.72 ± 0.05^{bcd}						
ILM ₃	15.96 ± 0.25^{d}	5.52 ± 0.09^{cde}	$5.86{\pm}0.02^{b}$	$6.43{\pm}0.38^{b}$	6.34 ± 0.06^{abc}						
ILM_4	$14.51 \pm 0.15^{\rm f}$	5.47 ± 0.31^{cde}	$5.53 {\pm} 0.32^{b}$	5.89±0.13 ^{bc}	$6.41{\pm}0.03^{ab}$						
ILM ₅	$10.40{\pm}0.20^{h}$	6.94±0.81ª	$6.10{\pm}0.27^{b}$	6.30 ± 0.49^{b}	5.99 ± 0.55^{bcd}						
ILM ₆	17.22±0.15 ^b	$5.77 {\pm} 0.50^{cd}$	5.46 ± 0.24^{b}	$5.98{\pm}0.16^{b}$	$5.65 {\pm} 0.77^{cd}$						
ILM_7	12.32 ± 0.22^{g}	$6.69{\pm}0.40^{ab}$	6.25 ± 0.13^{b}	$6.54{\pm}0.61^{ab}$	$5.48{\pm}0.25^{d}$						
ILM ₈	16.42±0.15°	5.98 ± 0.22^{bc}	$5.94{\pm}0.56^{b}$	$7.12{\pm}0.34^{a}$	$6.85{\pm}0.42^{a}$						
ILM9	15.13±0.25 ^e	$4.85{\pm}0.50^{\rm f}$	$4.70{\pm}0.32^{a}$	$5.42 \pm 0.50^{\circ}$	5.56 ± 0.32^{cd}						
LIMITS		20	0 mg/L (WHO)								

Table 4 Sulphates and sodium of selected sachet water brands produced in Ilorin Metropolis over a 4-month storage.

Note: Values are Means \pm SD of triplicate determinations (n = 3); Means with different superscripts along the same column are significantly ($p \le 0.05$) different.

The sachet water samples had no lead in them before storage. However, there was little indication of lead (0.0010-0.0020 mg/L) after one month of storage (Table 5). At the end of the second month, the lead values had increased in some samples and decreased in others, but the values were not significantly different (p > 0.05). The presence of lead later during storage suggests increasing detectability owing to enhanced metal availability. An increase in lead may be associated with increasing acidity and corrosiveness of water distribution systems [32]. The lead contents of all the samples showed a steady increase throughout the rest of the storage period but were still less than the WHO limit of 0.01 mg/L [39]. In a study by Ojekunle et al. [41], lead was likewise detected in some sachet water samples. Considering the classification of lead as a toxic mineral, these findings call for more regulatory checks by concerned regulatory bodies in the operation of sachet water manufacturers.

Lead is present in water due to its dissolution from natural sources, primarily from household plumbing systems. This is especially common when lead-containing pipes, solder, fittings, or other service connections convey high-acidity water [49]. PVC pipes also contain lead compounds that can leach and produce high lead concentrations in drinking water [44]. Lead is a cumulative poison associated with several health hazards like anemia and reproductive effects [50]. According to Egbueri [32], some of the possible consequences of high lead levels in drinking water also include inhibited Vitamin D metabolism, retarded mental health, as well as cancer.

There was no presence of copper in the water samples before storage. However, after one month of storage, some level was recorded, ranging from 0.0007-0.0021 mg/L. In fact, at the end of storage, copper levels were 0.030-0.060 mg/L (Table 5). This result is similar to the observation made with lead above; presumably, there was an increased solubility of copper from its compounds during storage. There was no significant (p > 0.05) difference in copper levels in the sachet water brands throughout the storage period, and the values remained within the WHO/NIS permissible range of 0.05-0.1 mg/L [32]. Consuming water whose copper level exceeds the permissible maximum can cause gastrointestinal disturbance, including nausea and vomiting, while prolonged use may cause liver or kidney damage [51].

The magnesium values for the sachet water samples varied significantly ($p \le 0.05$) from 0.42 mg/L to 1.0 mg/L before storage (Table 6). Although some fluctuations were recorded in some samples during storage, at the end of the four-month storage period, just as noted for a good number of other chemical parameters, the magnesium contents had notably increased. Storage conditions, increasing metal solubility, and microbial activities may be responsible. The results, however, appear to corroborate an earlier report suggesting that prolonged storage of sachet water may result in increased chemical properties beyond permissible limits [16]. Results from the present research are also similar to the increasing magnesium contents noted for some brands of sachet water evaluated by Ajekunle et al. [23] during a 2-month storage. However, the magnesium values of all nine samples met the WHO standard of not more than 2.0 mg/L [30], except ILM5 and ILM7, which showed a tendency to exceed the limit after the fourth month. No evidence of adverse health effects attributed explicitly to magnesium in drinking water has been established; however, excess calcium and magnesium ions make water hard [52].

			Lead (mg/L)								
Sample	Storage period (month)										
	0	1	2	3	4						
ILM_1	0.000 ^a	0.0017 ± 0.00^{a}	0.00067 ± 0.00^{a}	0.001 ± 0.00^{a}	0.0026 ± 0.00^{a}						
ILM_2	0.000^{a}	0.0020 ± 0.00^{a}	$0.00167 \ \pm 0.00^a$	0.0026 ± 0.00^{a}	0.0050 ± 0.00^{a}						
ILM ₃	0.000^{a}	0.0013 ± 0.00^{a}	0.00200 ± 0.00^a	0.0026 ± 0.00^{a}	0.0033 ± 0.00^{a}						
ILM ₄	0.000^{a}	0.0013 ± 0.00^{a}	$0.00100 \pm \! 0.00^a$	0.003 ± 0.00^{a}	0.0040 ± 0.00^{a}						
ILM ₅	0.000^{a}	0.0010 ± 0.00^{a}	$0.0023 \ \pm 0.00^{a}$	0.003 ± 0.00^{a}	0.0040 ± 0.00^{a}						
ILM ₆	0.000 ^a	0.0013 ± 0.00 a	0.0013 ± 0.00^{a} 0.0010 ± 0.00^{a} 0.0026 ± 0.00^{a}								
ILM ₇	0.000^{a}	0.0013 ± 0.00^{a}	$0.0020 \pm 0.00^{\rm a}$	0.0026 ± 0.00^{a}	0.0046 ± 0.00^{a}						
ILM ₈	0.000^{a}	0.0013 ± 0.00^{a}	0.00200 ± 0.00^a	0.0026 ± 0.00^{a}	0.0043 ± 0.00^{a}						
ILM ₉	0.000^{a}	$0.00^{a} \qquad 0.00200 \pm 0.00^{a} \qquad 0.0020 \pm 0.00^{a} \qquad 0.002$									
LIMITS	0.01 mg/L (WHO)										
			Copper (mg/L	L)							
ILM ₁	0.000 ^a	0.0007 ± 0.00^{a}	0.0007 ± 0.00^{a}	0.0017 ± 0.00^{a}	$0.030\pm\!\!0.00^a$						
ILM ₂	0.000^{a}	$0.0010 \pm \! 0.00^a$	$0.0010 \ {\pm} 0.00^{a}$	0.0023 ± 0.00^{a}	0.023 ± 0.01^{a}						
ILM ₃	0.000^{a}	$0.0010 \pm \! 0.00^a$	$0.0010 \ {\pm} 0.00^{a}$	0.002 ± 0.00^{a}	0.013 ± 0.01^{a}						
ILM ₄	0.000 ^a	0.0020 ± 0.00^{a}	$0.0020 \ {\pm} 0.00^{a}$	0.002 ± 0.00^{a}	0.033 ± 0.01^{a}						
ILM ₅	0.000 ^a	$0.0210{\pm}0.03^{\rm a}$	$0.0210 \pm 0.03^{\rm a}$	0.003 ± 0.00^{a}	$0.035\pm\!\!0.02^a$						
ILM ₆	0.000^{a}	$0.0013 \pm \! 0.00^a$	$0.0013 \ {\pm} 0.00^{a}$	0.003 ± 0.00^{a}	0.040 ± 0.01^{a}						
ILM ₇	0.000^{a}	$0.0010 \pm \! 0.00^a$	$0.0010 \ {\pm} 0.00^{a}$	0.003 ± 0.00^{a}	$0.056\pm\!0.01^a$						
ILM ₈	0.000^{a}	0.0007 ± 0.00^{a}	0.0007 ± 0.00^{a}	0.003 ± 0.00^{a}	0.060 ± 0.01^{a}						
ILM ₉	0.000^{a}	0.000^{a}	$0.0010 \ {\pm} 0.00^{a}$	0.001 ± 0.00^{a}	0.0010 ± 0.00^{a}						
LIMITS			0.05-0.1 mg/L (WH	O/NIS)							

Table 5 Lead and copper of selected sachet water brands produced in Ilorin Metropolis over a 4-month storage. Note: Values are Means \pm SD of triplicate determinations (n = 3); Means with different superscripts along the

same column are significantly ($p \le 0.05$) different.

No iron was detected in the water samples before storage, but values of 0.001-0.0023 mg/L were obtained at the end of the first month (Table 6). There was no significant (p > 0.05) difference in the iron contents. Different trends were observed in the iron contents during the rest of the storage period. This observation contrasts the findings of Ajekunle et al. [23], who reported decreasing iron contents while storing sachet water. This may be attributed to iron-utilizing species of microbes in their study, whereas more iron-releasing metabolic or biochemical processes probably occurred in the current study. All the sachet water samples contain iron levels less than the maximum (0.3 mg/L) required by WHO and NIS.

Iron in water may indicate the presence of coliforms, which can, in turn, indicate sewage contamination and could cause dysentery and typhoid [34]. The affected water may also demonstrate discoloration, bitter and astringent flavour, and encourage microbial growth [32]. Excessive consumption of iron can also result in red hot disease [39].

Table 6	Magnesium	and	iron	of	selected	sachet	water	brands	produced	in	Ilorin	Metropolis	s over a	4-month
storage.														

	Magnesium (mg/L)									
Sample			Storage period (mo	onth)						
	0	1	2	3	4					
ILM_1	$0.42 \pm 0.00^{\rm g}$	$0.5033 \pm 0.04^{\circ}$	0.6367 ± 0.08^{b}	$0.69 \pm 0.04^{\text{b}}$	0.82 ± 0.03^{ef}					
ILM_2	$0.43\pm\!\!0.01^{g}$	0.5533 ± 0.07^{c}	$0.5600 \pm 0.03^{\text{b}}$	0.59 ± 0.04^{cd}	$0.74\ {\pm}0.03^{\rm fg}$					
ILM ₃	$0.46\pm\!\!0.01^{\rm f}$	$0.5433 \pm 0.18^{\circ}$	0.5700 ± 0.08^{b}	0.63 ± 0.06^{bcd}	$0.65 \pm 0.07^{\text{g}}$					
ILM_4	0.77 ± 0.01^{b}	$0.7833 \ {\pm} 0.12^{b}$	$0.6500 \pm 0.09^{\text{b}}$	0.67 ± 0.05^{bc}	$1.37 \pm 0.05^{\circ}$					
ILM ₅	1.0 ± 0.00^{a}	$0.7667 \pm \! 0.09^{b}$	$1.0233 \ {\pm} 0.03^a$	1.02 ± 0.03^{a}	$2.43\pm\!\!0.12^a$					
ILM ₆	$0.64\pm 0.01^{\circ}$	1.220 ± 0.19^{a}	0.5300 ± 0.10^{b}	$0.69 \pm 0.06^{\text{b}}$	$1.18 \pm 0.09^{\rm d}$					
ILM_7	$0.49 \pm 0.01^{\text{e}}$	$0.5133 \pm 0.03^{\circ}$	0.6067 ± 0.04^{b}	$0.55 \pm 0.09^{\circ}$	$2.13\pm\!0.03^{\text{b}}$					
ILM ₈	$0.52 \pm 0.01^{\rm d}$	$0.5600 \pm 0.04^{\circ}$	$0.5300 \pm 0.08^{\rm b}$	0.73 ± 0.03^{b}	$0.88 \pm 0.04^{\text{e}}$					
ILM ₉	$0.53 \pm 0.01^{\text{d}}$	$0.5900 \pm 0.01^{\circ}$	$0.6500 \pm 0.03^{\rm b}$	0.74 ± 0.04^{b}	$0.74 \pm 0.00^{\rm fg}$					
LIMITS		2.0 mg/L (WHO)								
			Iron (mg/L)							
ILM_1	0.00^{a}	$0.0023 \ {\pm} 0.00^a$	0.00200 ± 0.00^{a}	0.0020 ± 0.00^{b}	0.005 ± 0.00^{a}					
ILM_2	0.00^{a}	$0.0010 \ {\pm} 0.00^a$	0.00133 ± 0.00^{a}	0.0030 ± 0.00^{a}	0.003 ± 0.00^{a}					
ILM ₃	0.00^{a}	$0.0013 \ {\pm} 0.00^a$	0.00233 ± 0.00^{a}	0.0020 ± 0.00^{a}	0.004 ± 0.00^{a}					
ILM_4	0.00^{a}	0.0020 ± 0.00^{a}	0.00233 ± 0.00^{a}	$0.0026{\pm}0.00^{a}$	0.006 ± 0.00^{a}					
ILM ₅	0.00^{a}	$0.0107 \ {\pm} 0.01^a$	0.00167 ± 0.00^{a}	0.0023 ± 0.00^{a}	$0.002\pm\!0.00^a$					
ILM ₆	0.00^{a}	0.0017 ± 0.00^{a}	0.00200 ± 0.00^{a}	$0.0023 \pm \! 0.00^a$	0.004 ± 0.00^{a}					
ILM_7	0.00^{a}	0.0017 ± 0.00^{a}	$0.00267 \pm \! 0.00^a$	$0.0020 \pm \! 0.00^a$	$0.019\pm\!\!0.03^a$					
ILM ₈	0.00^{a}	0.0020 ± 0.00^{a}	0.00267 ± 0.00^{a}	$0.0026{\pm}0.00^{\rm a}$	0.003 ± 0.00^{a}					
ILM ₉	0.00^{a}	0.0020 ± 0.00^{a}	0.00233 ± 0.00^{a}	0.0023 ± 0.00^{a}	0.002 ± 0.00^{a}					
LIMITS			0.3 mg/L (WHO))						

Note: Values are Means \pm SD of triplicate determinations (n = 3); Means with different superscripts along the same column are significantly ($p \le 0.05$) different.

The coliform counts of the various water brands at the start of storage ranged from 2 to 26 CFU/mL (Table 7). After the first month of storage, water sample ILM1 demonstrated the highest coliform count of 280 CFU/mL, even though it had the lowest count before storage. This could be due to higher residual chlorine in other samples, which prevented coliform from multiplying. The increasing total coliform count is at variance with the loss of viability expected of coliform during storage [23]. However, other scholars have reported similar reports. For example, Duwiejuah et al. [35] reported increased coliform counts in some Ghanaian sachet water brands during refrigeration and ambient storage. Coliform counts also showed a progressive rise in different brands of sachet water during an 8-week indoor storage in Enugu, Nigeria [53]. While the reason for this observation is yet to be ascertained, an increase in coliform count after storage might suggest insufficient water treatment [54]. It is also plausible that the packaging and bagging materials were contaminated with coliform before being used, thereby serving as sources of coliform in the packaged water during storage. Accordingly, it might be necessary for the manufacturers of sachet water brands to ensure more thorough water treatment during production. In addition, more frequent cleaning and sanitation of the storage area and facilities might be required to create sanitary conditions that discourage the proliferation of coliform. A detailed risk assessment that includes inspection of packaging materials and their handlers may help improve the microbial safety of the water brands. The permeability of polyethylene materials has been featured in discussions about the possible reasons for an increasing coliform count in sachet water during storage [23]. The fecal contamination of drinking water has severe health implications [24]. Total coliforms are widely used to indicate the general sanitary quality of treated drinking water, while fecal coliforms give a much closer indication of fecal pollution [55]. Consequently, the WHO requires a zero count for fecal coliform in drinking water [23]. In a similar study by Ndamitso et al. [51], a high presence of coliform was reported for some stored sachet water brands from Minna; hence, the water brands were deemed unfit for consumption. Sule et al. [18] also investigated bottled water brands in the Ilorin metropolis. The authors found one of the brands to contain coliform as high as 121 CFU/mL. Coupled with the findings in

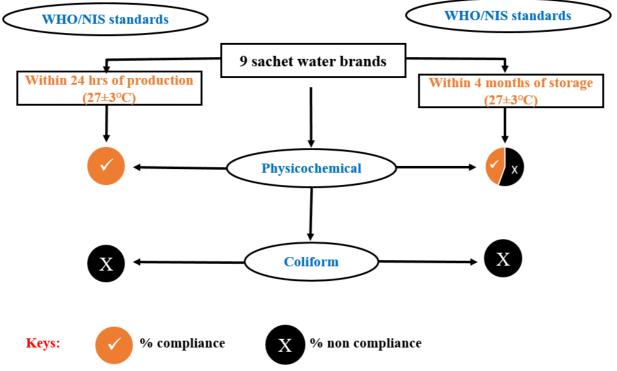
the current research, this suggests that packaged water manufacturers within the Ilorin Metropolis may require more conscious efforts to protect their water sources and operational systems from fecal contamination.

At the end of storage, the total coliform count in all sachet water samples had increased markedly, especially in samples ILM1 and ILM5 (\geq 2400 CFU/mL each), indicating coliform growth. The recorded coliform counts in the various water samples constitute a severe health hazard, and fecal contaminations of the different production sites are strongly suspected.

Bacterial loads in the various sachet water brands at the start of storage ranged between 2 and 50 cfu/ mL (Table 7), and the values were below the permissible limit of 100 CFU/mL. However, these counts showed varied rates of increase until after the second month, when bacterial loads in a few samples declined. Samples ILM7 and ILM8 notably had bacterial counts that exceeded the permissible limit from the second month of storage.

		Total via	ble coun	ts (CFU/n	nL)	Total coliform counts (CFU/mL)						
Sample		Stora	ige perioo	d (month)		Storage period (month)						
	0	1	2	3	4	0	1	2	3	4		
ILM ₁	8	28	58	75	70	4	240	280	350	920		
ILM ₂	2	8	12	32	2	≤2	280	350	920	≥2400		
ILM ₃	10	30	57	70	67	2	33	43	180	220		
ILM ₄	10	48	100	70	80	4	34	33	49	110		
ILM ₅	20	68	106	100	105	2	240	220	350	1600		
ILM ₆	21	52	80	90	108	8	240	280	350	540		
ILM ₇	50	100	120	130	120	23	45	130	350	≥2400		
ILM ₈	38	70	111	110	105	26	34	46	110	180		
ILM ₉	8	26	30	38	30	≤ 8	110	220	220	280		
LIMITS		100	CFU/mL	(WHO)			0 CF	U/mL (V	VHO)			

Table 7 Total coliform and viable counts of selected sachet water brands produced in Ilorin Metropolis over 4month storage.



WHO: World Health Organisation NIS: Nigerian Industrial Standards

Figure 6 Graphical representation of regulatory compliance.

Based on results obtained from the current investigation, especially as it concerns microbiological safety on the one hand and physicochemical instability during storage on the other hand, concerted efforts may be required on three dimensions:

- i. Addressing consumers' perceived purity of sachet water: Consumers' interpretation of sachet water as being 'pure' may represent some potential vulnerability. Nigerians typically refer to sachet water as 'pure water'. There are tendencies for many, including residents of the local government areas under study, who are not yet well-informed, to misconceive this terminology as water 'free of any probable hazard risks.' It is necessary to educate consumers that some sachet water brands may not be as safe for consumption as generally deemed. Increased awareness in this regard is helpful for consumers to be more logical by paying more attention to factory details, such as siting and operations when choosing the brands to patronize. The proposed sensitization efforts should also discourage any prolonged ambient storage of sachet water beyond two months; otherwise, physicochemical safety may not be guaranteed. Although this is in line with the two months 'Best Before Date' commonly claimed by some producers, information relating to the shelf life has been reported missing on some sachet water brands [56].
- ii. Manufacturers' consciousness on quality and safety: Contaminated water sources, inadequate water treatments, and unsanitary storage and packaging conditions are suspected to be responsible for the declining quality of the sachet water brands during storage. Producers are therefore encouraged to intensify surveillance on the microbiological status of raw water from the source, as well as during and after processing, extending this to the packaging materials and their handlers. In addition, storage areas and facilities should be regularly cleaned and sanitized to discourage microbial proliferation. If matched with appropriate measures to avoid fecal contamination along the line, this may check the presence of coliform in sachet water. Current and prospective sachet water producers are required to comply with standards relating to factory location and proximity to fecal sources.
- iii. Importance of regulatory surveillance and penalties: Right from initial certification through to subsequent operations, relevant regulatory agencies, including NAFDAC and SON, in the country may be required to review and intensify their monitoring approaches, and focus, such that certification and presence of any sachet brand in the market can be more reliably trustable by consumers. While keeping illegal producers out of the market may be cumbersome, enacting and implementing severe sanctions on any illegal operations may be strategic.
- iv. More documentation of waterborne illnesses: Some data on outbreaks of waterborne diseases are available in some different states in Nigeria, including Kwara State, where the current city under study is located. Kwara State was previously reported as the third State with the highest cholera outbreak [57]. This suggests that the microbial results from the current study may be strongly linked to the occurrence of certain diseases in the study area. In the same vein, leakages of storage tanks from nearby petrol filling stations have been identified as possible causes of increasing contaminants such as heavy metals found in groundwater sources in the llorin Metropolis [58]. This pollution was suggested as a possible threat to public health. However, more documentation of waterborne illnesses in the study area is recommended. This will provide for more scientific assessments of the likely roles of sachet water.

CONCLUSION

The study showed that sachet water brands from selected local government areas of the Ilorin Metropolis of Nigeria largely comply with the physicochemical requirements of WHO and NIS for drinking water. These included temperature (25.10-30.13 °C), turbidity (0.37-2.84 NTU), electrical conductivity (115.6-277.33 µS), total dissolved solids (55.33-125.70 mg/L), chlorides (2.83-8.57 mg/L), nitrates (0.15-0.64 mg/L), sulphates (4.70-7.12 mg/L), sodium (10.16-18.74 mg/L), copper (<0.01 mg/L), lead (<0.006 mg/L), and iron (<0.03 mg/L). However, two-thirds of the products exceeded the permissible level (100 mg/L) for total hardness at the end of four months of storage (102.30-136.33 µS/L), indicating their increased risk to human health when stored beyond three months. Similarly, two of the nine sachet water brands exhibited magnesium contents of 2.13-2.43 mg/L after the 4th month of storage, which was higher than the permissible level of 2.0 mg/L and may imply increased hardness during further storage. The bacterial loads of some brands demonstrated the tendency to rise (>105 CFU/mL) beyond the permissible limit of 100 CFU/mL after two months of storage. From a bacterial point of view, this may suggest that such brands are safe for consumption within the first two months of storage. However, this likelihood cannot be validated, considering that coliforms were detected (2-26 CFU/mL) in all the water brands even before the commencement of storage. As this strongly implies faecal contamination and renders the products unsafe for human consumption, it is pertinent that the sachet water producers in the study area, as well as concerned regulatory bodies, intensify efforts to check faecal contamination. It is therefore advised that the various producers put in place measures to prevent faecal contamination so as to meet the coliform zero tolerance for drinking water. Periodic inspection of sachet water factories by concerned regulatory bodies is vital to ensure conformity with set standards.

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