Parasitological and Bacteriological Evaluation of Sachet Drinking Water Sold in Selected Markets in Yenagoa, Bayelsa State, Nigeria

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Abstract
Water is very important in sustaining life. For water to be drinkable, its quality must be high. This study is focused on isolation and identification of parasites contaminating sachet water sold in selected markets within Yenagoa metropolis, Bayelsa State, Nigeria. One hundred sachets of water were collected randomly from five brands sold in four different markets within the study area. Physical features of the sachet water sampled such as labeling, volume, NAFDAC registration number, batch number, production date, expiry date and company address were visualized and recorded. The parasites were identified using sedimentation technique. Of the 100 samples examined, all the five brands showed the presence of the physical features listed above. An overall prevalence of 34% from five parasite species of the genera *Ascaris lumbricoides* (5%), *Entamoeba histolytica* (11%), *Gardia lamblia* (11%), *Necator americanus* (4%) and *Trichuris trichuria* (3%) respectively was recorded. There was no significant difference in the distribution of parasites among the different brands of water sampled ($P>0.05$). The physiochemical properties fell within the World Health Organization (WHO) standards for drinking water except for the pH of two brands that were below standard. The overall percentage difference among the bacteria species isolated from the samples analyzed was also not significant. The presence of parasites and bacteria in the samples could have unhealthy implications when consumed. Hence, there is a need for regular and periodic monitoring of the water quality before and after production.

Keywords: Parasites, Sachet water, Water contamination, Water quality, Bacteria

1 Introduction

Water is very important in sustaining life. It is an integral part of the human body making up 75% of the brain, 82% of blood, 25% of bone and over 70% of body tissues [1]. For water to be drinkable, its quality must be high. Such water is called potable drinking water which is free from contaminants and dissolved substances [2]. If water is not potable, it constitutes health risks to consumers [3]. About 1.1 billion people do not have access to potable drinking water, according to a WHO report [4]. In Nigeria, Yenagoa, the Bayelsa State capital is not exempted from this problem. Similar to what is obtainable in other areas, surface and underground water are often contaminated and these are the sources often used by sachet water producing factories. Water needs to be properly treated to make it suitable for human consumption [5]. National Agency for Food, Drug Administration and Control (NAFDAC) is the agency saddled with the responsibility to ensure that sachet water producers meet the required standard.

Sachet water popularly called "Pure Water" is consumed because of its low cost and it also provides an easy access to drinkable water. This is quite appreciated but the means of production needs to be hygienic to prevent transmission of water related enteric parasites and their cysts [6]. Sachet water is exposed to contamination either at the source, during production, transportation or sales by vendors. It is usually available in 60 cl polythene plastic and sold in shops, parks, street corners and by vendors [7]. Some brands of sachet water available in Yenagoa include Fido, Ingo, NDU, Ayalla, Aqualina, Bosy, Frefa, Ranz, Regal, Prextam, Bruzz, Kumo, ET, Esbel, Fountain, Ezewode, and Yemeb. As a result of increasing demand and low cost of sachet water, the quality/safety of some sachet water is now questionable because many of the producers fail to properly treat water before selling to the public [8]. As such, these beautifully labeled and well packaged sachet water often contains contaminants which may be injurious to health [9].

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It has been reported that parasites and bacteria are usually associated with contamination of drinking water [10]. Some of these parasites are resistant to water treatment processes mainly because they form cysts, hence remain in water for a long time [11]. *Cryptosporidium parvum, Giardia lambia* and *Entamoeba histolytica* have been reported to cause deaths in children [12]. Furthermore, bacteria such as *Escherichia coli*, *Staphylococcus aureus*, *Bacillus sp*. and *Streptococcus sp*. also portend serious deleterious effects on humans [9]. Generally, there is yet insufficient information on parasites and bacteria that contaminate sachet water. Hence, this study was designed to investigate the presence of parasites and bacteria in five selected sachet water brands consumed in Yenagoa metropolis, Bayelsa State, Nigeria.

2. Materials and Methods

2.1 Study Area

The research was carried out in four markets namely Akenfa Market (GPS Coordinates: 4'9212ºN, 6'2748ºE), Bayelsa Palm Market (GPS Coordinates: 4'8567ºN, 6'3436ºE), Swali Market (GPS Coordinates: 4'9186ºN, 6'2628ºE) and Tombia Market (GPS Coordinates: 4'9975ºN, 6'2628ºE) located in Yenagoa metropolis, Bayelsa State, Nigeria. The study area lies between longitude 6º15'51E and latitude 4º55'29N (Figure 1). Yenagoa metropolis has a geographical area of 706 km^2 with a human population of 352,285 [13]. The climate of the area is the equatorial type with an annual rainfall of over 2500 mm per annum and double maxima of rainfall between June/July and September/October. The area experiences conventional rainfall and it is known for hot, humid, and wet seasons with an average temperature of approximately 27ºC [14].

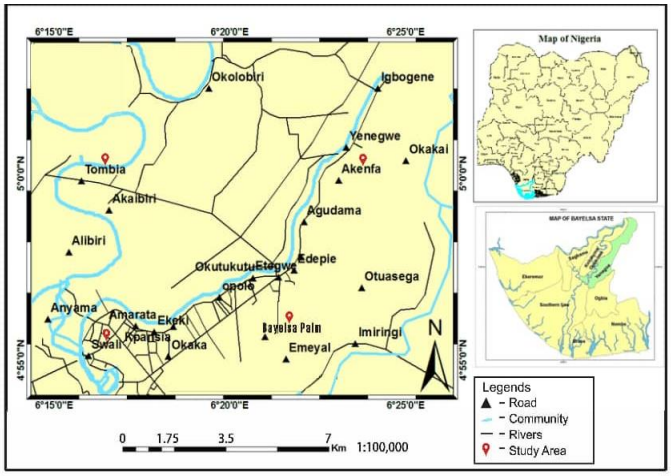

Figure 1: Map of Yenagoa Metropolis Showing the Study Area (Source: Google Map)

2.2 Collection of Water Samples

Five brands of sachet water (labelled A to E) were selected for the study from the four markets (Akenfa, Bayelsa Palm, Swali and Tombia) based on level of patronage by consumers and distributors. Four (4) sachets were selected from each bag giving a total of 100 sachets to be analyzed (20 sachets per market). All the water samples were registered with NAFDAC, sourced from ground with a volume of 500 mL.

2.3 Physicochemical Analysis of Sachet Water Samples

The collected sachet water samples were analyzed for physicochemical parameters including total dissolved solids (TDS), pH, Chlorides, hardness and nitrite based on standard methods [15].

2.4 Examination of Water Samples for Parasites

Water samples were examined for presence of parasites using sedimentation method [16]. Briefly, a drop of suspended sediments was placed at the centre of a clean, labeled, grease-free glass slide and covered using a cover slip without trapping air bubbles. It was then observed microscopically under x10 objective lens. A drop of Lugols iodine was added on the glass slide to determine the presence of the parasite cysts [17].

2.5 Bacteria Isolation and Identification from Water Samples

MacConkey purple broth was used. The water sample (50 mL each) water sample was measured and added to a bottle of 50 mL double strength medium for serial dilution. Test tubes were incubated at 37ºC for 48 hours. A positive test was indicated by colour change in medium from purple to yellow. Samples were streaked for the growth of isolated colonies on MacCkonkey's agar. Culture plates were further incubated at 37ºC for 48 hours for bacteria. Plates were examined for their morphology and identification [17].

2.6 Statistical Analysis

The results were analyzed with a two-way analysis of variance (ANOVA) test, using SPSS version 23.

3. Results

The physicochemical properties of sachet water sampled are presented in Table 1. All the sampled water met WHO standards except for the pH of brands D and E which fell below standards. The prevalence of parasites across the four selected markets based on the sampled water sachet are presented in Table 2. Out of 100 sachet water samples examined, 34% were positive for parasitic contamination.

Table 1: Physiochemical properties of sachet water samples

	Brands					
Parameters	**A**	**B**	**C**	**D**	**E**	**WHO standard**
TDS (ppm)	21.06	21.84	24.77	11.25	10.00	1000.00
pH	7.2	6.9	7.2	5.3	5.2	6.5-8.5
Cl^- (ppm)	0.03	0.03	0.04	0.04	0.03	250.00
Total Hardness (mg/L)	52.80	7.40	6.30	3.40	3.10	200
Nitrite (mg/L)	0.01	0.01	0.02	0.01	0.02	0.1

KEY: A-E=Sachet water brand code, TDS=Total dissolved solids, Cl=Chloride, ppm=part per million, mg/L=milligram per litre (P=0.005; P< 0.05)

Table 2: Prevalence of parasites in sampled Sachet Water in Yenagoa, Bayelsa State, Nigeria

Markets	**Number examined**	**Number infected (%)**
Akenfa	25	8(32)
Bayelsa Palm	25	8 (32)
Swali	25	10 (40)
Tombia	25	8 (32)
Total	**100**	**34 (34)**

The prevalence and parasite distribution in sachet water samples consumed in Yenagoa according to species is presented in Table 3. Five parasites were identified namely *Ascaris lumbricoides* (5%), *Entamoeba histolytica* (11%), *Giardia lamblia* (11%), *Necator americanus* (4%) and *Trichuris trichuria* (3%) respectively. *Entamoeba histolytica* and *Giardia lamblia* were found in all the brands. *Ascaris lumbricoides* was found in A, B, C and E. *Necator americanus* was found only in brands A, C, and D, while *Trichuris trichuria* was found in brands A, B, and C respectively.

Table 3: Parasite distribution in sachet water samples consumed in Yenagoa, according to species

Brand	**Number Examined**	***Ascaris lumbricoides***	***Entamoeba histolytica***	***Gardia lamblia***	***Necator americanus***	***Trichuris trichuria***
A	20	1(5)	1(5)	3(15)	2(10)	1(5)
B	20	1(5)	2(10)	2(10)	-	1(5)
C	20	1(5)	2(2)	2(10)	1(5)	1(5)
D	20	-	3(3)	1(5)	1(5)	-
E	20	2(10)	3(15)	3(15)	-	-
Total	**100**	**5(5)**	**11(11)**	**11(11)**	**4(4)**	**3(3)**

The distribution of bacterial isolates in the sampled sachet water is presented in Table 4. The information revealed five bacteria species isolated with varying percentage distribution. Of the 100 samples analyzed, *Escherichia coli* had the highest occurrence of 8 (23.5%), followed by *Bacillus sp.*, *Staphylococcus aureus* and *Streptococcus sp.* at 7 (20.6%) each, while *Micrococcus sp.* had the least in 5 samples (14.7%).

Table 4: Distribution of bacteria isolates in sachet water consumed in Yenagoa, Nigeria

Brands	*Bacillus sp.*	*Escherichia coli*	*Micrococcus sp.*	*Staphylococcus aureus*	*Streptococcus sp.*	Total
A	2 (5.9)	2 (5.9)	1(2.9)	2 (5.9)	2 (5.9)	9 (26.5)
B	1 (2.9)	2 (5.9)	-	1 (2.9)	1 (2.9)	5 (14.7)
C	1 (2.9)	1 (2.9)	2 (5.9)	2 (5.9)	1 (2.9)	7 (20.6)
D	2 (5.9)	2 (5.9)	1 (2.9)	-	2 (5.9)	9 (26.5)
E	1 (2.9)	1 (2.9)	1 (2.9)	1 (2.9)	1 (2.9)	4 (11.8)
Total	7 (20.6)	8 (23.5)	5 (14.7)	7 (20.6)	7 (20.6)	34 (100)

4. Discussion

Physiochemical parameters including TDS, pH, chlorides, total hardness and nitrite are key indices employed to determine water quality. The values recorded for most of these parameters in the present fell within WHO standards which agrees with a previous study carried in Akwa, South-Eastern Nigeria [18]. The only exception was the pH value which fell below the required standard in two brands. The slight acidity recorded for the samples may be due to microbial activities which release carbon (iv) oxide and sulphur (iv) oxide. This finding is corroborated by the study of Anake *et al*. [19] which reported physiochemical and microbial assessment of different water sources in Ota, Ogun State. According to a report, consumption of water low in pH may cause gastrointestinal irritation in sensitive individuals [20].

Parasitic and bacterial contamination of sachets water has been previously reported on a relatively higher scale in Ngwo, Enugu State, Nigeria [21]. The difference may be attributed to environmental conditions and variation in species of parasites present. However, the present findings agree with Umeanaeto *et al*. [21] with respect to high prevalence rates of both *Entamoeba histolytica* and *Gardia lamblia*. Similar findings were reported in previous studies carried out in Accra and Lagos while evaluating parasites present in sachet water sold in those cities [6, 22, 23]. This may be due to poor or inadequate treatment as the cysts of *G. lamblia* could survive outside the host for several months in cold water while *E. histolytica* survives days to weeks in an external environment [24]. However, a study carried out in Owerri metropolis of Imo State, Nigeria reported absence of enteric pathogens in sachet water in contrast to the other studies [25].

The absence of parasites in the sachet water can be attributed to effectiveness of the treatment methods adopted by manufacturers during the production process. Some of the treatment measures include UV radiation and ozone generators. These treatment measures are highly efficient by virtue of their ability to kill parasites in the water. The cysts on the other hand were present because they were highly resistant to the water treatment measures applied [26]. Similar findings were reported regarding the presence of bacteria in sachet water [27-29]. Some of the sachet water factories visited during this study made use of manual sealing machines which involves contact of operators with the water during packaging. This may be responsible for the presence of cysts in sachet water [30]. The manual sealing machine could also attract other parasites including *A. lumbricoides, N. americanus and T. trichuria* in sachet water via hand contact [30].

5. Conclusion

Parasite and bacterial species were identified in the sachet water brands examined in this study. The presence of these parasites and bacteria significantly impacted the quality of water and may have negative consequences on human health. Considering the high patronage of sachet water in the study area, it would be immensely importance that all producers adhere to NAFDAC guidelines and laws to safeguard the lives of people. Regulatory bodies including Standard Organization of Nigeria (SON), NAFDAC and State Ministry of Heath should intensify efforts at monitoring sachet water. This should be done as periodic sanitary inspection of production environment as well as the raw materials and the finished products.

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Conflicts of interest
The authors declare no conflicts of interest.

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