

## Microbiological and Physicochemical Analysis of “Pure Water” Samples From 5 Villages in BORGU L.G.A Of Niger State, NIGERIA.

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**Abstract:** Microbiological and physicochemical analysis of 30 samples of Sachet-packaged drinking water popularly called ‘Pure Water’ procured from 5 villages in Borgu Local Government Area of Niger State Nigeria, namely: New Bussa, Wawa, Malale, Dogongari and Sabo were carried out to evaluate their suitability for drinking. Microbiological examination for total counts of bacteria such as faecal Coliform, *Escherichia coli*, *Vibrio sp*; *Salmonella sp*; *Streptococci* and *Staphylococci* spp was performed. Coliform counts of 10-200cfu/ml; *E. coli* counts of 0.2-8.0cfu/ml; *Staphylococcus aureus* counts 10-2000cfu/ml and total plate count of 120-1400cfu/ml were obtained in 4, 2, 3, 3, and 5 samples respectively out of the 6 tested from each village. Most of the physicochemical properties of the water samples tested had values that were above the acceptable values. Using the standards supplied by National Agency for Food and Drug Administration and Control [NAFDAC] in Nigeria and as specified by World Health Organization [WHO], only 43.3% of the water samples were suitable for drinking. This research confirms the speculations about the impure state of the sachet packaged drinking water sold in Nigeria as ‘Pure Water’ of which only 40.0% had NAFDAC registration numbers.

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### I. Introduction

Water is one of the most abundant commodities of man and occupies about 70% of the earth’s surface yet a great percentage of the world’s population live without access to safe drinking water especially in the developing countries. In the light of this, many sachet packaged water popularly known as ‘pure water’ businesses has sprung up.

Unsafe water is a global public health threat placing persons at risk for a host of diseases such as diarrhea, cholera, typhoid fever etc. It has been reported that two million persons mostly children below the age of five have died of diarrheal diseases and according to the world health organization (WHO) 2004, about 90% of diarrheal –related deaths have been attributed to unsafe or inadequate water supplies and poor sanitation Halpern et al (2008).

In Nigeria, there has been a shortage in public piped water supply that made the populace depend on open dug wells, streams and boreholes as sources of portable water. However, an increased outbreak of cholera, typhoid and other water –borne diseases has led to the emergence of sachet-packed water called “pure water” so called to give an assurance of safety Adeleke et al (2000). As a regulatory body in Nigeria, National Agency for Food and Drug Administration and Control (NAFDAC) has set up limits of microbiological standards as guidelines on the quality of water suitable for drinking purposes (NAFDAC 2000). The metallic composition of water is important however the emphasis here is on the microbiological standards because of the serious health implications of ingesting microbial pathogens in water. World Health Organization (WHO) and NAFDAC permit  $\leq 100$ cfu/ml as total count and no faecal contaminants (WHO, 1996; NAFDAC, 2000). As *E. coli* is the most numerous facultative organism in the gut which can be detected at very low dilution, this makes it a sensitive indicator of faecal contamination. The enterobacteriaceae: *Escherichia*, *shigella* and *salmonella*, along with other organisms like *Vibrio cholera*, *faecal streptococci*, *staphylococcus aureus* and some fungi species represent the major pathogens causing water-borne disease world wide (Lewis, 1991; Richards and Batram, 1993; Hunter, 1997 and Kirkwood, 1999). For the purposes of this study 30 sachets of “pure water” will be procured randomly from different locations within New Bussa, Wawa, Malale, Dogongari and Sabo areas of Borgu Local Government Area of Niger state. This sachet water will be taken to the laboratory and subjected to analysis to verify their extent of purity and suitability for drinking.

### II. Materials And Methods

#### Water Samples

Thirty samples of sachet-packaged drinking water were procured randomly from the five different villages as mentioned above. Six samples from each village were labeled according to the commercial names on

the sachets which were coded as follows: IK, AG, MT; DA, KO, AT; DL, RB, RT; AI EW, EL; CA, KN, and SO.

### **Physicochemical Analysis**

Total hardness of the pure water samples were determined by complexometric method of disodium EDTA. The acidity, alkalinity and carbon dioxide were determined by titrimetric methods using 0.1 NaoH, 0.01m Hcl and 0.02m Na<sub>2</sub>Co<sub>3</sub> respectively.

The PH was determined using a digital PH meter standardized with buffers 4.00 and 9.00 while the conductivity was measured with a conductometer (Cambridge). The dissolved oxygen was measured using dissolved oxygen meter. The total dissolved solids was measured using turbidity cantest. The turbidity was measured using calibrated Nephelometer.

Biological Oxygen Demand (BOD):The parameter was measured using Nephelometer, the colour was determined using turbidity can test for colour and transparency and the taste was determined manually.

All determinations were carried out following standard procedures as outlined by Olaniyi and Ogungbamila(1991) and Reynolds et al, (1996).

### **Media**

**Media used in the experiment:** This include MacConkeyagar (MCA), salt nutrient agar (SNA), Nutrient agar (NA), Alkaline peptone water (APW), Sabourand dextrose agar (SDA),Salmonellashigella agar (SSA), MacConkey broth (MB) and centrimide nutrient agar (CNA).

### **Microbiological Analysis**

**Total viable counts:** Each sample was diluted 1 in 10<sup>3</sup> in nutrient broth(oxid) for bacterial counts and sabouranddextrose broth(oxid)for fungi counts. From the dilution, duplicate pour-plates were prepared in NA and SDA for bacteria and fungi respectively. The bacterial cultures were incubated at 37°C for 24-48hours and the fungal cultures at 25°C for 5 days, both under aerobic conditions. Bacteria and fungi were estimated as colony forming units per milliliter (cfu/ml).The samples were also cultured using four differential/selective culture media namely: MCA, SSA, CNA and SNA. The plates were incubated at 37°C for 24 to 48 hrs and bacteria were estimated as colony forming units per ml (cfu/ml), this method was adopted from Harrigan, (1976) and Adeleke et al, (2000).

### **Bacterial and Fungal Isolates**

Bacterial colonies on the plate for total viable count were gram stainedafter proper macroscopic examination of the colonies. They were later sub-cultured into sterile nutrient broth of 5ml per test tube. Isolated colonies resulting from plate cultures on NA were subjected to conventional biochemical tests specifically to identify staphylococcus aureus, Escherichia coli and Salmonella sp. The fungi isolates observed on the total viable count plates were identified by their cultural characteristics such as colour, consistency and growth pattern of mycelia (Cowan, 1974,Harrigan, 1976 and Samson et al 1992).

### **Presumptive Coliform Test (PCT)**

This test gave an estimate of most probable number (MPN) of coliform bacilli per 100ml of each water sample. Each sample was diluted in 100ml of sterile distilled water and subjected to multiple tube technique of PCT. The bile-based medium used was macConkey broth (DIFCO) in double and single strength concentrations appropriately. Tests that showed acid and gas production after 48hrs of incubation at 37°C were considered positive for coliform bacilli (Lewis, 1991).

### **FaecalColiformsand Detection of E. coli**

All positive tests in the PCT and an overnightbroth culture of E. coli, were sub-cultured in loopfull, each into MacConkey broth and incubated at 45°C in an electrothermal water bath for 48hrs. The sub-cultures that produced acid and gas as evidence of faecalcoliforms were streaked each onMacConkey agar plate and incubated at 37°C for 24hrs.Reddish pink colonies that stained gram-negative rods as the E.coli control culture were subjected to indole, methyl red, voges-proskauer and citrate (IMVIC) tests for differentiating the enteric coliforms. E. coli was detected as indoleand methyl-red positive but voges-proskauer and citrates test negative (Harrigan, 1976, Richards and Bartram, 1993).

### III. Results

**Table 1: Physicochemical analysis of sachet packaged water samples "pure water" from 5 villages in BORGUL.G.A of Niger State**

Sample of pure water	New Bussa			Wawa			Malale			Dogongari			Sabo		
	IK	AG	MT	DA	KO	AT	DL	RB	RT	AI	EW	EL	CA	KN	SO
Declared Content (cl)	50	60	50	50	50	50	50	50	50	60	50	50	50	50	50
Net Content (cl)	54	62	51	52	53	55	52	55	52	64	54	52	52	54	52
NAFDAC No.	Yes	No	Yes	No	No	Yes	Yes	No	No	No	Yes	No	Yes	No	No
PH	6.9	5.0	7.0	6.7	6.9	6.6	7.0	7.2	5.6	7.1	6.8	5.5	7.0	8.4	6.2
Conductivity ohm/cm	190	185	190	350	820	260	140	245	280	120	190	520	170	120	280
Dissolved oxygen D O (mg/l)	6.20	5.10	6.20	5.00	6.20	6.00	4.20	4.50	6.10	6.40	6.20	4.20	5.60	6.00	6.20
Total dissolved solids Tds (ppm)	0.0456	0.0444	0.0456	0.163	0.197	0.0624	0.0336	0.0455	0.0378	0.194	0.0444	0.0635	0.0541	0.184	0.197
Turbidity (NTU)	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10
Colour	Cl	Cl	Cl	Cl	Cl	Cl	Cl	Cl	Cl	Cl	Cl	Cl	Cl	Cl	Cl
Taste	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Total hardness	0.8	2.2	1.1	2.1	2.4	1.1	1.0	2.4	2.0	2.9	1.4	2.9	1.6	3.0	4.0
Biological Oxygen Demand BOD (mg/l)	4.00	3.10	2.20	3.20	3.00	3.00	2.20	3.10	4.00	4.10	2.20	3.00	3.40	2.30	2.40
Total alkalinity	276	252	144	384	252	72	204	132	216	228	180	120	274	345	256

Cl = Colourless  
 - = No taste  
 + = Taste present

**Table 2: Microbiological examination of sachet packaged water samples from 5 villages in BorguL.G.A of Niger State**

Sample of pure water	New Bussa			Wawa			Malale			Dogongari			Sabo		
	IK	AG	MT	DA	KO	AT	DL	RB	RT	AI	EW	EL	CA	KN	SO
Coliformcfu/ml	Nil	50	Nil	140	60	Nil	Nil	10	80	70	Nil	20	Nil	200	120
E.coli cfu/ml	Nil	0.1	Nil	0.2	0.8	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil	0.5
Faecal streptococci cfu/ml	Nil	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Staph. Aureuscfu/ml	Nil	10	Nil	60	1400	40	20	Nil	1000	100	Nil	60	Nil	500	1200
Vibrio sp cfu/ml	Nil	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Salmonella sp cfu/ml	Nil	Nil	Nil	Nil	400	200	Nil	100	400	Nil	Nil	Nil	Nil	200	1000
Yeast/moulds	Nil	Nil	10	Nil	40	Nil	20	Nil	Nil	100	Nil	180	Nil	80	500
Total plate count (cfu/ml)	Nil	40	20	120	780	40	40	400	1400	300	Nil	200	Nil	1400	1000

cfu/ml = colony forming unit per milliliter

### IV. Discussion

Based on the result obtained from the physicochemical analysis (table 1) the electrical conductivity of most of the water samples were higher than normal while that of two samples were within the normal range for pure water samples (0.045-0.055us/cm). This is an indication of the presence of dissolved solids which may enhance redoxreaction in the water samples which may lead to discolouration of the water samples. Most of the water samples tested are within the neutral hydrogen – ion concentration except three that were highly acidic and the consumption of such water can pose health risk.

All the aesthetic requirements of good appearance colour, odour and taste were satisfactory in all but two of the samples. The WHO stipulated that all drinking water should meet the above requirements. The actual volume of content of each water sample was in excess of the 500ml and 600ml printed on the sachets. None of the samples had a batch number which means one cannot be sure of the processing period and date in case of a need for product recall. Only 12 of the 30 samples had NAFDAC numbers which is an indication that only 40% of the processing facilities were NAFDAC approved. Since NAFDAC is the regulatory body responsible for ensuring the safety of commercial drinking water in Nigeria, lack of NAFDAC number may be implicating.

The microbiological analysis (table 2) showed that E. coli was detected in 4 samples whereas total coliforms were detected in 9 samples-out of a total of 15 samples. Faecal Streptococci and Vibriosp. were absent in all the samples, but the presence of Salmonella in some samples from Wawa, Malale and Sabo may be a threat to public health (pose some serious health risk). Salmonella sp. has been implicated in gastroenteritis, typhoid and paratyphoid fevers. Similarly, the plate counts for staphylococcus aureus showed that the pathogen was present in 10 samples while total plate count revealed excess of microbes (120-1400cfu/ml) in 8 samples.

In conclusion, it is evident, based on the tested samples that most sachet-packaged drinking water (pure water) in New Bussa and environs are below standards as per purity and microbial content. Therefore compliance monitoring for approved standards of drinking water must be enforced for sachet packaged (pure water) drinking water.

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