## The Effect of a Nigerian Brewery Effluent on Two Receiving Streams

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Abstract: This study assessed the microbiological and physico-chemical parameters of water samples from Omi-Asoro and Imo Hills streams into which effluent from a Nigerian breweries empties. Samples collected along the wastewater discharge path and receiving streams were analyzed for microbial count, coliforms, pH, Temperature, dissolved organic matter, BOD, DOC, total solid content, Total Nitrogen, Phosphorous, chlorine, iron and heavy metals. The antibiograms of bacterial isolates were determined using the disk diffusion method. Results showed that the factory effluent and contaminated streams had higher microbial loads and coliform counts than the uncontaminated streams. The bacteria recovered from the samples included antibiotic resistant *Pseudomonas* and *klebsiella* species while the prominent fungal genera were *Aspergillus, Penicillium, Microsporium, Saccharomyces,* and *Cladosporium* species. The dissolved organic matter, biochemical oxygen demand and total solid contents of the effluent and contaminated streams were beyond acceptable limits while the dissolved oxygen concentration in the contaminated stream was generally below acceptable levels. The concentration of lead and mercury were also above recommended limits. The study showed that the brewery effluent adversely affected the quality of the receiving streams rendering the water unfit for use at some locations along the course of the receiving streams.

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## 1. Introduction

Water is vital for human life and it is indispensable for agriculture. manufacturing, transportation and many other human activities. It is the most precious natural resource occurring abundantly on Earth (Karikari and Ansa, 2006). Water for human use can be pipe-borne water generated from surface waters like streams and rivers or ground waters like wells and boreholes. In many developing countries, however, treated public pipeborne water is scarce and even when available regular short or long interruptions in water supply do occur (Ukhun et al., 2005). This makes people opt for alternative sources of water from unconventional sources particularly streams and rivers (Leroy et al., 2002; Van der Bruggen and Braeken, 2006). Water though indispensible has a high potential to spread various microbial pathogens to a large number of people (Ahmed et al., 2004). The inadequate supply of potable water from public taps is therefore contributory to the high morbidity and mortality associated with water-borne diseases in developing countries (Bloomfield, 2009). About a decade ago, it was estimated that about 1,400 people die every hour due to waterborne diseases (Bouwer, 2003). This is of concern to consumers, water supplies regulators and public authorities (WHO, 2004). One reason for this is that rivers, the alternative source of water, are increasingly being exposed to pollution by human activities (Skoulikidis et al., 2004). Pollution of the

aquatic environment has been defined by UNESCO /WHO/UNEP as the introduction by man of substances or energy into the marine environment which results in deleterious effects on the living resources, hazards to human health, hindrance of marine activities including fishing and impairment of quality of water. Most of the rivers in the urban areas of the developing world are the end points of effluents discharged from many industries (Olayinka, 2004) causing irreversible degradation in the surface water systems (Rajaram and Ashutosh, 2008). Industrial effluents contain materials from the wastes that settle in river water as bottom sediments and constitute health hazards to the population that depend on the water as source of supply for domestic uses (Akaniwor et al, 2007). Brewery plants cause pollution by discharging effluents into receiving streams; ground water and soil in the locality in which breweries are located making such water bodies unsuitable for drinking and irrigation (Hari et al., 1994). Drinking water should be odorless, tasteless, colorless and devoid of particulate matter. It must be free of disease causing organism, poisonous substances and excessive amount of minerals and organic matter. Some minerals and dissolved substances are permissible only at certain levels (WHO, 2004; NESREA, 2011). The chemical investigations of water from some Nigerian rivers reveal that the quality is deteriorating in many places (Ajavi and Osibanjo, 1981; Adeniji and Mbagu,

1983; Asuquo, 1989) and there is little information on the biological effects of brewery effluent discharge in some areas in the country. This study was therefore undertaken to determine the effect of waste effluents from the International Breweries, Ilesa on the biological, biochemical, and physicochemical parameters of Omi-Asoro and Imo Hills streams which receive effluents from the brewery.

# 2. Material and Methods

## 2.1. Study Area

The study was carried out on Omi-Asoro and Imo hills streams into which the brewery effluent discharges. The sampling points are shown in Figure 1. Samples were collected once every two months from September 2008 to August 2009.



Figure 1: Sampling points

A: Effluent discharge point outside the factory.

B: Neat Omi-Asoro stream before mixing with the brewery effluent.

C: About 100m downstream from the point of mixture of the brewery's effluent and Omi-Asoro stream.

D: About 1.5km from the mixing point of brewery's effluent and Omi-Asoro stream.

E: Neat Imo Hill stream before mixing with discharged effluent and Omi-Asoro stream.

F: About 500m downstream from the point of mixture of Brewery's effluent and Omi-Asoro with the Imo Hill stream.

## **2.2.** Collection of Water Samples

Samples were collected at about 10cm below the water surface into 2 L kegs pre-disinfected with 75% ethanol and rinsed with sterile distilled water. Water from each sampling point was used to rinse the appropriately labeled keg twice before sample collection. Samples were brought into the laboratory directly from the sampling field and analyzed within three hours of collection.

# 2.3. Microbial Analysis of Water Samples 2.3.1. Total Microbial Counts

The enumeration of total microbial count was carried out using the serial dilution and pour plate technique. Serial 10 fold dilutions in sterile water were carried out and 1ml of each dilution was aseptically placed in sterile petri-dishes in duplicates. 20 ml of molten nutrient agar (Oxoid) cooled to  $45^{\circ}$ C for bacterial population or Potato Dextrose Agar (Oxoid) for fungi was later added to each of the plates. The mixture was allowed to solidify and incubated appropriately for 24 hours for bacterial and 7 days for fungal populations.

# 2.3.2. Presumptive Coliform Count

The water samples were processed using the multiple fermentation tube method to determine the Presumptive coliform count or most probable number (MPN) of coliforms based on standard methods (Senior, 2006). Suspensions from positive tubes were subcultured on MacConkey agar and incubated at 37<sup>o</sup>C for 24-48 hours. The resulting colonies were identified following standard procedures (Forbes et al., 2007).

# **2.3.3. Detection and Enumeration of Faecal** Coliforms

The Eijkman's test involving the use of elevated temperature for the differentiation of faecal and non-faecal coliforms was employed. Measured volume of water samples were inoculated into lactose broth in similar combinations as in the presumptive test for total coliforms and incubated at 44°C for 48 hours. The number of positive tubes was recorded at the end of the incubation period and the MPN of faecal coliform was read from the MPN index table.

# 2.3.4. Isolation of Pure Cultures of Microorganism

Distinct colonies from the mixed culture on each of the nutrient agar and the potato dextrose agar plates were picked and streaked onto fresh plates. Pure colonies of each bacteria and fungi were then picked for further characterization. The isolates were then taken through a set of standard biochemical tests and identified to the genus level based on the interpretation of results of the biochemical tests (Farmer, 1999).The antimicrobial testing of the isolates against commonly used antibiotics was performed using Kirby-Bauer's disc diffusion method and was interpreted according to Clinical Laboratory Standards Institute (CLSI, 2007) guidelines.

## 2.3.5. Physicochemical Analysis

Standard methods for the examination of water and waste waters (APHA, 2001) were used in the determination of the physicochemical parameters of the water samples. The color, temperature, pH, and Dissolved Oxygen were measured on site.

The phosphate content was determined using phosphate Test Kit (model PO-23/PO-23A). For clear

water samples, PO-23 was used while PO-23A was used for turbid water samples. The nitrate content in the water samples was determined using Nitrate Test Kit of model N1-11. Iron Test Kit Model IR-18 was used for the determination of iron content while the chlorine content of the water samples was determined using Free and Total Chlorine Test Kit Model CN-70.

The concentrations of heavy metals (Mercury, Arsenic, Lead, Chromium and Cadmium) in the water samples were determined using Atomic Absorption Spectrophotometer (AAS) at the Central Science Laboratory of Obafemi Awolowo University, Ile-Ife and also at the Centre for Energy Research and Development, (CERD), O.A.U., Ile-Ife.

Data obtained from the study was analyzed using Microsoft Excel package.

#### 3. Results

Presented in Table 1 are the microbial parameters at the sampling points within the period of the study.

Table 1: Microbial parameters at sampling points

$\begin{array}{c c c c c c c c c c c c c c c c c c c $	Table 1: Microbial parameters at sampling points								
Sept.A $4.20x10^5$ $2.10x10^4$ 0 $2.10x10^4$ 08B $4.60x10^2$ $2.50x10^2$ 0 $5.10x10^2$ C $2.60x10^4$ $7.70x10^3$ 0 $3.70x10^3$ D $3.10x10^3$ $3.30x10^3$ 0 $4.00x10^2$ E $3.00x10^3$ $5.00x10^2$ 0 $3.20x10^2$ F $4.30x10^2$ $2.70x10^3$ 0 $4.70x10^2$ Nov.A $7.20x10^6$ $2.40x10^4$ 0 $4.60x10^4$ 08B $5.50x10^2$ $2.30x10^2$ 14 $2.00x10^2$ C $5.80x10^4$ $3.60x10^4$ 0 $4.10x10^2$ D $3.60x10^4$ $6.40x10^3$ 0 $2.20x10^2$ E $6.20x10^2$ $2.60x10^2$ 0 $2.80x10^2$ F $6.80x10^3$ $3.20x10^3$ 0 $3.60x10^4$ D $3.60x10^4$ $3.80x10^3$ 0 $3.60x10^2$ F $6.80x10^3$ $3.20x10^3$ 0 $2.80x10^2$ F $6.80x10^3$ $3.20x10^3$ 0 $2.80x10^2$ C $6.70x10^4$ $3.80x10^4$ 0 $2.90x10^4$ D $4.70x10^4$ $1.40x10^3$ 0 $1.60x10^4$ D $4.70x10^4$ $1.40x10^3$ 0 $1.60x10^4$ D $4.70x10^5$ $1.10x10^3$ 0 $3.60x10^3$ F $1.90x10^5$ $1.10x10^3$ 0 $3.60x10^3$ D $1.00x10^6$ $1.60x10^4$ 0 $2.90x10^3$ D $1.00x10^6$ $1.10x10^3$ 0 $3.60x10^3$ D $1.00x10^5$	Period	Site	TBC	TCC	FC	FGC			
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	Sept.	А	$4.20 \times 10^5$	$2.10 \times 10^4$	0	$2.10 \times 10^4$			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	08	В	$4.60 \times 10^2$	$2.50 \times 10^2$	0	$5.10 \times 10^2$			
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $		С	$2.60 \times 10^4$	7.70x10 <sup>3</sup>	0	3.70x10 <sup>3</sup>			
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $		D	3.10x10 <sup>3</sup>	3.30x10 <sup>3</sup>	0	$4.00 \times 10^2$			
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $		Е	$3.00 \times 10^3$	$5.00 \times 10^2$	0	$3.20 \times 10^2$			
Nov. 08         A $7.20x10^6$ $2.40x10^4$ 0 $4.60x10^4$ 08         B $5.50x10^2$ $2.30x10^2$ $14$ $2.00x10^2$ C $5.80x10^4$ $3.60x10^4$ 0 $4.10x10^4$ D $3.60x10^4$ $6.40x10^3$ 0 $2.20x10^2$ E $6.20x10^2$ $2.60x10^2$ 0 $2.80x10^2$ F $6.80x10^3$ $3.20x10^3$ 0 $3.60x10^4$ B $1.80x10^3$ $8.80x10^2$ 11 $9.20x10^2$ C $6.70x10^4$ $3.80x10^4$ 0 $2.90x10^4$ D $4.70x10^4$ $1.40x10^3$ 0 $1.60x10^4$ E $2.70x10^3$ $1.20x10^3$ 0 $1.50x10^3$ F $1.90x10^3$ $4.20x10^2$ 0 $2.30x10^4$ 09         B $1.00x10^6$ $1.60x10^4$ 0 $5.60x10^4$ D $9.10x10^5$ $1.11x10^3$ 0 $3.60x10^4$ 09         A $1.00x10^8$ $1.10x10^3$ </td <td></td> <td>F</td> <td><math>4.30 \times 10^2</math></td> <td><math>2.70 \times 10^3</math></td> <td>0</td> <td><math>4.70 \times 10^2</math></td>		F	$4.30 \times 10^2$	$2.70 \times 10^3$	0	$4.70 \times 10^2$			
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	Nov.	А	$7.20 \times 10^{6}$	$2.40 \times 10^4$	0	$4.60 \times 10^4$			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	08	В	$5.50 \times 10^2$	$2.30 \times 10^2$	14	$2.00 \times 10^2$			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		С	$5.80 \times 10^4$	$3.60 \times 10^4$	0	$4.10 \times 10^4$			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		D	$3.60 \times 10^4$	$6.40 \times 10^3$	0	$2.20 \times 10^2$			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		Е	$6.20 \times 10^2$	$2.60 \times 10^2$	0	$2.80 \times 10^2$			
$ \begin{array}{c c c c c c c c c c c c c c c c c c c $		F	6.80x10 <sup>3</sup>	3.20x10 <sup>3</sup>	0	3.60x10 <sup>2</sup>			
$\begin{array}{c c c c c c c c c c c c c c c c c c c $	Feb. 09	А	8.60x10 <sup>6</sup>	$3.70 \times 10^4$	0	$2.80 \times 10^4$			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	`	В	$1.80 \times 10^{3}$	8.80x10 <sup>2</sup>	11	9.20x10 <sup>2</sup>			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		С	$6.70 \times 10^4$	$3.80 \times 10^4$	0	$2.90 \times 10^4$			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		D	$4.70 \times 10^4$	$1.40 \times 10^3$	0	$1.60 \times 10^4$			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		Е	$2.70 \times 10^3$	$1.20 \times 10^3$	0	$1.50 \times 10^3$			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		F	$1.90 \times 10^{3}$	$4.20 \times 10^2$	0	$2.30 \times 10^3$			
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	April	А	1.90x10 <sup>8</sup>	$1.80 \times 10^4$	0	5.60x10 <sup>4</sup>			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	09	В	$1.00 \times 10^{6}$	$1.60 \times 10^2$	0	$2.10 \times 10^3$			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		С	$1.70 \times 10^{6}$	$1.10 \times 10^{3}$	0	6.80x10 <sup>4</sup>			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		D	9.10x10 <sup>5</sup>	$1.11 \times 10^{3}$	0	$3.60 \times 10^3$			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		Е	8.80x10 <sup>5</sup>	9.30x10 <sup>2</sup>	0	7.20x10 <sup>3</sup>			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		F	$2.40 \times 10^5$	$2.20 \times 10^2$	0	$3.00 \times 10^4$			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	June 09	А	$1.00 \times 10^8$	$1.10 \times 10^3$	0	3.00x10 <sup>5</sup>			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		В	$9.40 \times 10^5$	$2.30 \times 10^2$	0	$4.30 \times 10^3$			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		С	$1.60 \times 10^{6}$	$9.30 \times 10^2$	0	$1.60 \times 10^4$			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		D	1.30x10 <sup>6</sup>	$2.50 \times 10^2$	0	5.70x10 <sup>3</sup>			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		Е	6.70x10 <sup>5</sup>	$6.30 \times 10^2$	0	$4.70 \times 10^3$			
Aug.         A $7.60x10^6$ $3.20x10^3$ 0 $2.80x10^5$ 09         B $4.20x10^5$ $1.20x10^3$ 11 $5.60x10^3$ C $9.20x10^6$ $4.20x10^3$ 0 $1.20x10^4$		F	5.90x10 <sup>5</sup>	$2.30 \times 10^2$	0	$2.00 \times 10^3$			
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	Aug.	А	$7.60 \times 10^{6}$	$3.20 \times 10^3$	0	$2.80 \times 10^5$			
C $9.20x10^6$ $4.20x10^3$ 0 $1.20x10^4$	09	В	$4.20 \times 10^5$	$1.20 \times 10^3$	11	5.60x10 <sup>3</sup>			
		С	$9.20 \times 10^{6}$	$4.20 \times 10^3$	0	$1.20 \times 10^4$			
D $8.80 \times 10^4$ $2.40 \times 10^3$ 0 $5.90 \times 10^3$		D	$8.80 \times 10^4$	$2.40 \times 10^3$	0	5.90x10 <sup>3</sup>			
E $5.20 \times 10^4$ $2.30 \times 10^2$ 0 $4.20 \times 10^3$		Е	$5.20 \times 10^4$	$2.30 \times 10^2$	0	$4.20 \times 10^3$			
$F = 2.10 \times 10^4 = 1.20 \times 10^3 = 0 = 1.00 \times 10^3$		F	$2.10 \times 10^4$	$1.20 \times 10^{3}$	0	$1.00 \times 10^3$			

Note: TBC: Total bacterial count; TCC: Total coliform count; FC: Faecal coliform; FGC: Fungal count.

As shown in Table 1, the factory effluent and contaminated streams had higher microbial loads and coliform counts than the uncontaminated streams. The factory effluent (point A) had total bacterial, total coliform, faecal coliform and fungal densities of 4.2x10<sup>5</sup> - 1.9x10<sup>8</sup> cfu/mL, 1.1x10<sup>3</sup> -3.7x10<sup>4</sup> cfu/100mL, 0cfu/100mL, and 3.0x10<sup>3</sup> -2.8x10<sup>5</sup> cfu/mL respectively. The uncontaminated Omi-Asoro stream (point B) had total bacterial, total coliform, fecal coliform and fungal densities of  $4.6 \times 10^2$  -  $1.0 \times 10^6$  cfu/mL,  $1.2 \times 10^2$  -  $8.8 \times 10^2$ cfu/100mL, 0-14 cfu/100mL, and 3.7x10<sup>3</sup> - 6.8x10<sup>4</sup> cfu/mL respectively while those of uncontaminated Imo Hills stream (point E) were  $6.2 \times 10^2 - 8.8 \times 10^5$ cfu/mL, 2.3x10<sup>3</sup> - 1.2x10<sup>3</sup> cfu/100mL, 0cfu/100mL, and  $2.8 \times 10^2$  -  $7.2 \times 10^3$  cfu/mL respectively.

A total of 28 strains of bacteria belonging to five genera and 11 fungi were isolated from the 6 sampling points in the study as shown in Table 2. The bacterial strains isolated were members of the genera *Streptomyces, Bacillus, Pseudomonas, Klebsiella, Actinomyces and Enterobacter.* Strains *Bacillus, Pseudomonas* and *Klebsiella* were recovered from each of the sampling points.

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	V I gamsins	UNNAHIEU	110711	Sammung	DOTINS
10010	01841101110	000000000		Samp mg	pomo

Organism	Sampling points						
	Α	В	С	D	Е	F	
Bacteria							
Actinomyces spp.	+	+	-	-	1	-	
Bacillus spp.	+	+	+	+	+	+	
Enterobacter spp.	+	-	-	-	-	-	
Klebsiella spp.	+	+	+	+	+	+	
Pseudomonas spp.	+	+	+	+	+	+	
Streptomyces spp.	-	+	-	-	+	-	
Fungi							
Absichia spp.	+	+	+	+	+	+	
Aspergillus spp.	+	+	+	+	+	+	
cladosporium spp.	+	+	+	+	+	+	
Gliocladium spp.	-	-	I	+	+	-	
Microsoprium spp.	+	+	+	+	+	+	
Mucor spp.	+	+	+	+	+	+	
Penicillium spp.	-	-	+	+	+	+	
Phialophora spp.	-	-	-	+	+	+	
Saccharomyces spp.	+	+	+	+	+	+	
Scepulariosis spp.	+	+	+	+	+	+	
Trichophyton spp.	+	+	+	+	+	+	
Absichia spp.	+	+	+	+	+	+	

The results of the susceptibility of the medically significant gram negative bacteria isolated from the river at point F showed that the contaminants were multi-resistant to the commonly used antibiotics as shown in Table 3.

6						<u> </u>		
Organism	Ant	Antibiotics						
	Α	AC	С	Ν	G	Ср	0	Р
Klebsiella spp. 1	R	S	S	R	R	S	S	S
Klebsiella spp. 2	S	S	R	S	R	R	S	S
Pseudomonas spp. 1	R	R	S	R	R	S	S	S
Pseudomonas spp. 2	R	R	R	R	R	S	S	S

Table 3: Antibiotic susceptibility profile of Gram negative bacteria strains isolated at sampling point F

Note: A: Amoxicillin; AC: Amoxicillin+Clavulanic acid; C: Trimethoprim + Sulphamethoxazole; Cp: Ciprofloxacin; G: Gentamicin; N: Nitrofurantoin; Of: Ofloxacin; P: Pefloxacin

The results of the biochemical and physical parameters of the samples are presented in Tables 4a and 4b.

Table 4a:	Biochemical	and	physical	parameters a	ŧt
sampling	points				

Month	Site	T. °C	DO	BOD	DOM		
			Concentration in mg/L				
Sept. 08	А	32	3.2	60	5.77		
	В	26	3.2	40	3.85		
	С	29	1.8	20	4.20		
	D	27	4.8	3.2	3.60		
	Е	26	5.3	3.6	3.72		
	F	26	6.4	4.0	3.40		
Nov. 08	А	33	1.8	80	33.93		
	В	26	2.4	60	13.10		
	С	30	1.6	160	17.36		
	D	27	4.8	80	14.46		
	Е	26	5.2	40	5.20		
	F	27	6.4	12	3.62		
Feb. 09	А	32	1.2	120	14.40		
	В	27	2.2	60	6.70		
	С	30	1.6	80	8.43		
	D	27	4.8	40	4.80		
	Е	27	3.2	60	5.60		
	F	27	5.2	16	2.80`		
Apr 09	А	32	1.6	80	9.20		
*	В	26	34	40	4.62		
	С	29	1.8	60	5.96		
	D	27	5.2	40	4.88		
	Е	25	4/8	12	3.66		
	F	26	6.4	4	3.40		
Jun 09	А	33	1.8	220	26.40		
	В	25	3.2	32	8.40		
	С	29	2.4	16	14.00		
	D	27	5.6	12	8.40		
	Е	26	4.8	10	4.00		
	F	25	6.2	4	2.60		
Aug 09	А	32	1.8	180	9.95		
-	В	25	3.2	40	3.82		
	С	28	1.6	20	5.36		
	D	26	4.8	36	4.21		
	Е	25	5.2	40	3.82		
	F	25	6/4	40	3.44		

T (<sup>o</sup>C): Temperature; DO: Dissolved oxygen; BOD: Biochemical oxygen demand; DOM: Dissolved organic matter.

Table 4a: More biochemical	and physical parameters
at sampling points	

Month	Site	TS	TDS	TSS
		Concen	tration in n	ng/L
Sept. 08	А	360	28	80
<b>I</b>	В	130	90	40
	С	160	140	20
	D	150	120	30
	Е	60	40	20
	F	160	130	30
Nov. 08	А	300	180	120
	В	140	90	50
	С	180	120	60
	D	140	80	60
	Е	100	60	40
	F	120	100	20
Feb. 09	А	480	310	170
	В	120	80	40
	С	180	130	50
	D	140	90	50
	Е	80	60	20
	F	120	90	30
Apr 09	Α	220	180	40`
_	В	110	80	30
	С	160	140	20
	D	120	120	20
	Е	60	41	20
	F	100	70	30
Jun 09	А	280	240	40
	В	140	100	40
	С	220	160	60
	D	160	120	40
	Е	60	40	20
	F	120	90	30
Aug 09	А	230	80	150
	В	120	80	40
	С	140	120	20
	D	160	140	40
	E	80	50	30
	F	80	60	20

Note: TS: Total solids; TDS: Total dissolved solids; TSS: Total suspended solids.

The temperature of the discharged factory effluent (point A) ranged between  $32^{\circ}$ C and  $33^{\circ}$ C while those of water samples from the neat streams (points B and E) were between 25 and  $27^{\circ}$ C. The sampling point closest to the factory discharge (point C) had temperatures higher than each of the streams (28-30°C). The color of water samples nearer the factory effluent discharge point was darker than the color of the streams before mixing with the effluent and at distances further away from the effluent discharge point. The two streams at points before mixing with the factory effluent had dissolved oxygen contents ranging from 2.2 - 5.3 mg/L. On mixing with the factory effluent, the already low dissolved oxygen content of Omi-Asoro stream

(point C) was further reduced to 1.6-2.4 mg/L and as the water travelled further away from the discharge point, there was progressive increase in the dissolved oxygen contents reaching the maximum of 5.2 - 6.4mg/L at point F. The values of the biochemical oxygen demand (BOD<sub>5</sub>), dissolved organic matter and suspended solids of the samples were generally highest in the factory discharge point A with a progressive reduction as it was diluted downstream by the receiving streams.

Table 5a: Chemical parameters at sampling points

Period	Site	pH	Conce	Concentration in mg/L					
		1	Nit	PO <sub>4</sub>	CL	Fe			
Sept 08	А	7.20	4.40	14.0	0.01	2.62			
1	В	6.99	2.20	4.0	0.00	0.06			
	С	6.96	4.40	16.0	0.01	1.84			
	D	7.43	4.40	5.0	0.00	1.20			
	Е	7.04	2.20	4.0	0.00	0.04			
	F	6.83	4.40	4.0	0.00	0.20			
Nov.	А	8.06	4.40	15.0	0.01	2.80			
08	В	7.63	4.40	5.0	0.00	0.08			
	С	7.85	4.40	20.0	0.00	2.20			
	D	7.38	4.40	6.0	0.00	1.20			
	Е	7.29	2.20	5.0	0.00	0/00			
	F	7.12	4.40	4.0	0.00	0.38			
Feb. 09	Α	5.26	2.00	40.0	0,00	1.77			
	В	6.56	0.00	9.80	0.00	0.02			
	С	5.68	2.00	17.0	0.00	0.89			
	D	7.48	0.00	35.0	0.00	0.51			
	Е	6.48	0.00	14.0	0.00	0.00			
	F	7.08	0.00	27.0	0.00	0.39			
Apr. 09	Α	8.13	2.20	40.00	0.01	0.70			
	В	6.82	1.10	12.00	0.00	0.00			
	С	6.90	0.10	45.00	0.00	0.42			
	D	6.85	1.20	32.00	0.00	0.00			
	Е	6.62	0.10	12.00	0.00	0.02			
	F	6.64	1.00	24.00	0.00	0.20			
June 09	А	6.37	3.00	15.00	0.00	0.10			
	В	6.67	0.00	4.00	0.00	0.05			
	С	6.62	3.00	10.00	0.00	0.80			
	D	6.80	1.00	6.00	0.00	0.30			
	Е	6.96	0.00	5.00	0.00	0.00			
	F	6.75	0.00	4.00	0.00	0.20			
Aug.	Α	5.27	2.20	12.00	0.00	0.80			
09	В	6.57	0.00	4.00	0.00	0.00			
	С	5.69	4.40	16.00	0.00	0.66			
	D	7.49	4.40	8.00	0.00	0.32			
	E	6.49	0.00	4.00	0.00	0.04			
	F	7.09	2.20	4.00	0.00	0.20			

Nit: Nitrate; PO<sub>4</sub>: Phosphate; CL: Chloride; Fe: Iron

Tables 5a and 5b show the concentrations of nitrate, phosphate, chlorine, and the heavy metals mercury, lead, arsenic, chromium, cadmium and iron in the discharged brewery effluent and the downstream water samples.

Table 5b: More chemical	parameters at sampling
points	

Period	Site	e Concentration in mg/L				
		Hg	Cd	Cr	Ar	Pb
Sept 08	А	0.46	0.068	0.04	0.40	0.14
	В	0.00	0.029	0.00	0.00	0.00
	С	0.36	0.060	0.02	0.00	0.06
	D	0.24	0.048	0.11	0.00	0.04
	Е	0.04	0.030	0.05	0.00	0.00
	F	0.18	0.018	0.07	0.00	0.02
Nov.	А	0.53	0.062	0.14	0.00	0.10
08	В	0.00	0.042	0.12	0.00	0.00
	С	0.23	0.075	0.15	0.00	0.08
	D	0.16	0.048	0.09	0.00	0.06
	Е	0.00	0.022	0.05	0.05	0.01
	F	0.08	0.020	0.00	0.00	0.02
Feb. 09	А	0.56	0.216	0.21	0.10	0.16
	В	0.01	0.042	0.18	0.00	0.00
	С	0.32	0.075	0.23	0.29	0.12
	D	0.18	0.075	0.13	0.10	0.10
	Е	0.00	0.010	0.20	0.00	0.08
	F	0.10	0.029	0.27	0.08	0.03
Apr. 09	А	0.47	0.000	0.41	0.00	0.08
_	В	0.00	0.262	0.33	0.30	0.00
	С	0.22	0.000	0.27	0.00	0.01
	D	0.14	0.357	0.19	0.50	0.00
	Е	0.00	0.000	0.00	0.20	0.00
	F	0.06	0.032	0.00	0.50	0.01
June 09	А	0.62	0.052	0.14	0.60	0.12
	В	0.00	0.000	0.12	0.00	0.00
	С	0.60	0.036	0.15	0.50	0.03
	D	0.30	0.020	0.09	0.00	0.01
	Е	0.00	0.000	0.13	0.50	0.01
	F	0.05	0.020	0.17	0.40	0.01
Aug.	А	0.36	0.16	0.18	0.40	0.12
09	В	0.02	0.001	0.07	0.00	0.00
	С	0.12	0.018	0.23	0.03	0.03
	D	0.10	0.012	0.13	0.00	0.01
	Е	0.00	0.002	0.00	0.00	0.0
	F	0.07	0.010	0.07	0.08	0.01

Hg: Mercury; Cd: Cadmium; Cr: Chromium; Ar: Arsenic; Pb: Lead

## 4. Discussions

The result of this study revealed that the brewery effluent contained the highest microbial loads throughout the sampling period. The mixture of this effluent with Omi-Asoro stream increased the microbial load of the stream indicating the microbial pollution of the stream by the factory effluent as sample of water from the stream before mixture with the factory effluent had significantly lower bacterial counts. The microbial load of the polluted stream however, decreased as the discharged effluent flowed for longer distance, mixed with Imo hills stream and became a bigger river at point F. Seasonal variations in the microbial load of water at the different sampling points was observed. A significantly higher microbial load was observed in the streams at points before and after mixing with the brewery effluent during the rainy season than the dry season which is most likely due to surface run-off which suitably located organisms into the streams at on-set of early rains. Similar findings have been reported by Bakare et al. (2003) and Adewoye (2010).

The result of the coliform count in the water sample is suggestive of the fact that the brewery effluent contributed to the presence of coliform in the water bodies. The effluent having a higher coliform count increased coliform level in the stream after mixing with the stream. None of the water bodies is safe for drinking since their coliform densities greatly exceeded the WHO standard for coliforms in drinking water. This is particularly of public health significance because a good number of people including farmers used water from point B to F for drinking, bathing, laundry and other domestic purposes throughout the period of sampling. Probable sources of coliforms in the water samples include animal, plants, soil and humans. Faecal coliforms were, however, not isolated from the brewery discharged effluent at most of the sampling time. This is an indication that faecal wastes were not being discharged through the same sewer as the effluent. In November and February, the recovery of faecal coliform in Omi-Asoro stream is an indication of recent faecal contamination of the stream (Edberg et al., 2000) while the non-recovery of faecal coliforms from the upstream suggest that there was no recent faecal pollution prior to most of the sampling periods or the result of excessive dilution of water body by rain water in the rainy season (Badge, 1982).

The bacterial strains isolated from the water samples belong to the genera *Streptomyces*. *Bacillus*. Pseudomonas. Klebsiella, Actinomyces and Enterobacter. Some of these organisms are implicated in infections of burns, wounds, ulcer and the urinary tract. The presence of these organisms is not only of public health importance because of their potentials as pathogens but also because of the fact that they are multi resistant to antibiotics which may make the choice of antibiotic for treatment of infections arising from them a difficult task. They are also important in the transfer and spread of resistance in the community when one considers the fact that water from these sources are used in the irrigation, washing, and wetting of vegetables for public consumption.

The organic matter contents of all water samples were relatively high. The high dissolved organic matter content of the brewery wastewater is expected, resulting from the presence of grains, sugars, adjuncts, yeasts and other additives used in production of beer and malt drink. While it was observed that the organic matter level of the stream was increased by mixture with the factory effluent, the levels in the water samples decreased downstream probably due to microbial degradation and the settling of the organic matter along the course of the streams. Higher levels were observed during the period of malt drink production and in November and February. The seasonal variation could be attributed to concentration of the water bodies during the dry season.

The dissolved oxygen contents in the water samples at most of the sampling points were less than the WHO recommended standard of 6mg/L in drinking water except at last sampling point F. Based on this, water samples from these points are unfit for drinking. Vereijken et al. (1999) reported that excess BOD loading to a stream may cause a depression in dissolved oxygen concentration to the point of causing stress to some stream lives. This impacts high organic pollutants on receiving waters consequently creating high competition for oxygen within the ecosystem (Osibanjo and Adie, 2007). The temperatures of the discharged brewery effluent could also be responsible for the low level of dissolved oxygen in the water samples. These high temperature values were likely have been generated by the brewery activities which included boiling of wort, pasteurization of water as well as the washing and rinsing of returned bottles in hot and warm water respectively. These high temperatures were observed and recorded at some distance in the water samples downstream and may be lethal to the aquatic biota. The low level of dissolved oxygen is a hindrance to the rapid decomposition of organic matter (Atlas and Bartha, 1981) which could be responsible for the development of unpleasant odor in some parts of the water downstream. The high BOD<sub>5</sub> of the brewery effluent led to the depletion of the dissolved oxygen content of the water body to some distance even after mixing with Omi-Asoro stream. One of the characteristics of potable water is that it must be colorless. The observed coloration of the water samples at locations after the mixture of the factory effluent with the streams is an indication of the high level of organic matter and inorganic wastes present in the discharged effluent and subsequently at such locations. The foul odor and unsightly color observed in the affected destroyed the aesthetic and recreational values at some points along the course of the Omi-Asoro stream

The mean value of dissolved solids of the discharged brewery effluent was lower than the stipulated limit of 2000 mg/L (FEPA, 1991) while results showed that some water samples at some periods had levels of suspended solids which greatly

exceeded the stipulated limit for suspended solids (30 mg/L). The high level of total solids in the discharged brewery effluent is most likely to be responsible for this.

The mean concentrations of nitrate and phosphate in the sample were relatively high while very low total chlorine content was observed. According to Kunze (1996), nitrate is generated as waste product in brewery as nitric acid is used in the cleaning in process plant to dissolve beer stone. Phosphate is generated also from the use of cleaning agents. These values could also be a reflection of high organic matter contents of the water samples which probably liberate nitrate and phosphate upon decomposition and degradation.

The concentrations of lead, cadmium, chromium, and arsenic were low in the water samples analyzed during this study except in the discharged brewery effluent where levels were above the limits allowable in water for these metals. The presence of mercury in the effluent is likely to be from the chemicals and equipments that were being used in brewing and bottling activities, while high level of iron might be from pipes and sewers that carried both raw and treated water. The high concentrations of these heavy metals are unhealthy rendering the water bodies unsafe for drinking and for other agricultural purposes.

# 5.0. Conclusion

This study has shown that the discharged brewery effluent greatly exceeded the National Environmental Standards and Regulations Enforcement Agency's (NESREA) stipulated limits for industrial effluent discharge into surface waters with respect to total coliform density, biochemical oxygen demand, (BOD<sub>5</sub>), suspended solids and concentrations of heavy metals such as mercury and lead. The discharged brewery effluent has thus overburdened the receiving streams of Omi-Asoro and Imo-Hill.

This study also showed that farming activities along the banks of the stream of Omi-Asoro could increase the risk of communicable diseases since antibiotic resistant pathogenic organisms were isolated from the stream. As the factory effluent travelled further away along the streams, the magnitude of the pollution appeared to reduce giving rise to colorless and odorless water with increased dissolved oxygen, reduced BOD<sub>5</sub> that met the prescribed standards and subsequently greatly reduced organic matter. Water samples from the last sampling station which was about 500m from the point of mixing of Omi-Asoro, discharged effluent and another stream of Imo-Hill could have been recommended for drinking but for the total bacterial load and total coliform density that greatly exceeded the specified and acceptable international standards.

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