

MICROBIOLOGICAL EVALUATION OF STREAM WATER FOR DOMESTIC USE IN RURAL AREAS: A CASE STUDY OF IJEBU NORTH LOCAL GOVERNMENT, OGUN STATE. NIGERIA

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ABSTRACT

This study aimed at evaluating the microbial quality of stream water sources for domestic purposes by rural communities in Ijebu North Local Government to determine their fitness for human consumption. The evaluated streams include Erilobinla, Imosun, Okenugbo, Odoralamo, Odoye, loji, Mamu and Tekunle oga. Physicochemical parameters were determined; pour plate method using selective media were employed to determine the enteric bacteria present in water samples. Bacterial isolates were characterized adopting the standard methods, and isolates were further subjected to antimicrobial sensitivity testing using the disc diffusion technique. The result of physicochemical parameters showed that temperature value varied from 25 - 29°C, pH varied from 7.30 - 8.50, and total dissolved solid (TDS) of samples were not in agreement with WHO standards. Two of the eight streams analysed had odour, three had taste and two had colour. The total bacteria count revealed that Erilobinla stream water had the highest total bacteria count of 9.0×10^4 cfu/ml while Okenugbo and Odoye had the lowest total bacteria count of 1.0×10^1 cfu/ml. The microorganisms isolated were *Staphylococcus aureus*, *Enterococcus faecalis*, *Escherichia coli*, *Campylobacter* species, *Salmonella* species, *Klebsiella* species, *Proteus* species and *Pseudomonas aeruginosa*. The antimicrobial sensitivity testing showed that these organisms were resistant to some antimicrobials. In conclusion, most of the stream waters are unsafe for drinking as they are of low quality thresholds. Thus, the stream waters require further purification to ensure suitability for human consumption and there is urgent need for provision of potable water to prevent outbreak of waterborne diseases.

Key words: Gastroenteritis, stream water, evaluation, microbes.

INTRODUCTION

Water is the most vital element among the natural resources; it is the most indispensable need for existence of all living things. Its decreasing availability in terms of quality and quantity has been a major public health concern in Africa, particularly in Nigeria (WHO, 2004; Saravanan and Peter, 2009). Water fit for consumption is called drinking

water or potable water (Egberongbe *et al.*, 2010). According to a recent UNICEF report, about 80 million people in Asia and Africa are living without access to safe water. Consequently, this has caused many people to suffer from various diseases (Tanwir *et al.*, 2003). In developing countries such as Nigeria, most of the rural communities lack access to potable water supply and rely mainly

on river and stream sources for their household use and other purposes (Banwo, 2006). Many water sources in developing countries are unhealthy because they contain harmful physical, chemical and biological agents. Unfortunately, many of the available water sources are not potable without some form of treatment which is seldom or not available in most rural settings which expose the rural populace to water borne diseases (Oketola *et al.*, 2006). The major proportion of all water quality degradation worldwide is due to anthropogenic causes (Scott *et al.*, 2003). In some rural areas in Nigeria, domestic wastes, sewage and faeces are being discharged into streams which also serve as their water sources for daily needs. When the load of organic matter or wastes is too heavy, the self purification power of the stream are unable to remove these materials added and there will be pollution of these water sources which can be dangerous to human and the environment as a whole (Adetokunbo and Gilles, 2003). These multiple sources of contamination are compounded by limited environmental awareness in rural areas (Lehloesa and Muyima, 2004). The microbiological quality of drinking water is of a great primary importance, and the monitoring of bacterial indicators such as total coliform and thermotolerance coliforms should be given the highest priority. Microbial indicators have been used worldwide to indicate if human wastes have contaminated water body. Microbes typically utilized are those that are found in elevated concentrated in human faecal coliform, *Escherichia coli* and *enterococci* (Brooks *et al.*, 2006). An additional indicator, *Clostridium perfringes* can be used for monitoring stream water quality (Egberongbe *et al.*, 2010). The

outbreaks of diarrhoea or gastroenteritis in rural communities have all been attributed to the consumption of water of poor microbial quality (Ashbolt, 2004). It is therefore not an option but an imperative to critically monitor the quality of water supply in rural areas in order to further highlight their despicable water supply situation and to provide the impetus for sustainable government intervention (Gucker *et al.*, 2006).

MATERIALS AND METHODS

Water samples were collected aseptically from eight streams in different communities in Ago-Iwoye and Oru. The samples were collected in sterile container and analysed within 24 hours. Water samples were analysed for Physicochemical and bacteriological qualities. The Physicochemical properties examined include temperature, pH, taste, appearance, odour, total solids, total dissolved solids and total suspended solids. The media and reagents used for bacteriological analysis of water were weighed out and prepared according to manufacturers' specification. Nutrient agar (NA), *Salmonella* and *Shigella* agar (SSA), Eosin methylene blue agar (EMBA), Mannitol salt agar (MSA) and Blood agar (BA) plates were inoculated with 1ml of 10⁻³ dilution factor of each water sample using sterile pipette to determine the colony forming units per ml (cfu/ml) of samples. Discrete colonies after 48 hours of incubation at 37°C were sub-cultured to obtain pure cultures and were identified according to Cheesbrough (2000). Gram staining and biochemical tests were conducted to characterize the bacterial isolates. Antimicrobial sensitivity testing was carried out using the disc diffusion sensitivity technique.

RESULTS

Table 1: Physicochemical characteristics of the stream water samples in Ijebu North Local Government Area, Southwestern Nigeria.

Sample source	Temperature (°C)	pH	Appearance	Taste	Odour	Total solids (mg/l)	Total dissolved Solids (mg/l)	Total suspended solids (mg/l)
Erilobinla	29	8.32	Brown	Has taste	Irritating	0.52	0.21	0.32
Odoralamo	29	8.36	Colourless	Tasteless	Odourless	0.30	0.10	0.20
Imosun	27	8.50	Brown	Has taste	Irritating	0.40	0.10	0.30
Odoye	27	7.40	Creamy	Tasteless	Odourless	0.30	0.10	0.10
Okenugbo	28	7.52	Colourless	Tasteless	Odourless	0.31	0.00	0.31
Loji	29	7.53	Brown	Has taste	Irritating	0.20	0.10	0.20
Tekunleoga	25	7.60	Creamy	Tasteless	Odourless	0.26	0.10	0.26
Mamu	27	7.90	Brown	Has taste	Irritating	0.16	0.20	0.16

Table 1 shows the various Physicochemical characteristics exhibited by the stream water samples. Okenugbo and Odoralamo were colourless; Odoye and Tekunle oga had creamy colour while Imosun, Loji, Mamu and Erilobinla were highly polluted with brown appearances. Odoye, Okenugbo, Tekunle oga and Odoralamo samples were tasteless and odourless while Erilobinla, Loji, Mamu and Imosun samples had taste and irritating smell. The pH range of the streams varied from 7.30 in Odoye to 8.50 in Imosun. The temperature ranged from 25°C in Tekunle oga and Imosun to 29°C in Erilobinla and Odoralamo samples. The

Total Dissolved Solids (TDS) ranged from 0.0 mg/l in Odoralamo to 0.21mg/l in Erilobinla samples, and total suspended solids ranged from 0.10mg/l in Odoye and Okenugbo samples to 0.32mg/l in Erilobinla sample.

The colonial characteristics of each isolates on Nutrient agar, Blood agar, Eosin methylene blue agar, *Salmonella* and *Shigella* agar and Manitol salt agar are shown in Table 2. The isolates include *Staphylococcus aureus*, *Enterococcus faecalis*, *Escherichia coli*, *Salmonella* species, *Klebsiella pneumoniae*, *Pseudomonas* species and *Campylobacter* species.

Table 2: Colonial characteristics of isolates from stream samples in Ijebu North Local Government Area, Southwestern Nigeria on different growth media

	Organisms		Media Plates		
	NA	EMB	MSA	SSA	B.A
<i>Staphylococcus aureus</i>	tiny whitish colonies, slightly raised	-	yellow to cream slightly raised mucoid colonies	-	cream colonies slightly raised
<i>Enterococcus faecalis</i>	whitish, round raised, glistening colonies	light pink raised large colonies	-	-	cream non- haemolytic colonies
<i>Escherichia coli</i>	cream mucoid opaque colonies	characteristic of metallic sheen, non-opaque smooth colonies	-	-	cream, mucoid colonies.
<i>Salmonella</i> sp	Pink to red colonies without black centres	-	-	Black colonies with offensive odour	Grey to white colonies
<i>Klebsiella</i> sp	Mucoid colonies actively spreading across the plates, opaque whitish raised colonies	-	-	-	large grey to white mucoid colonies
<i>Proteus</i> sp	Small colourless colonies with fishy smell	-	-	-	Small swarming colonies with fishy smell
<i>Pseudomonas aeruginosa</i>	large dark greenish blue colonies	-	-	-	Large flat, haemolytic dark greenish- blue colour colonies.
<i>Campylobacter</i> sp	whitish droplet colonies	-	-	-	Non-haemolytic spreading droplet like

Key: - = No growth; NA = Nutrient agar; BA = Blood agar; EMB = Eosin methylene blue agar; SSA = Salmonella and shigella agar; MSA = Manitol salt agar

Table 3: Total bacterial counts (cfu/ml) obtained from the stream water samples from eight villages in Ijebu North Local Government Area, Southwestern Nigeria

Sample source	Organisms in cfu/ml				
	NA	BA	EMBA	SSA	MSA
Eriobinla	4.2x10 ⁴	3.3x10 ⁴	9.0x10 ⁴	2.0x10 ⁴	4.0x10 ⁴
Imosun	4.2x10 ⁴	5.6x10 ⁴	1.2x10 ⁴	2.0x10 ⁴	2.0x10 ⁴
Odoralamo	4.5x10 ⁴	3.5x10 ⁴	4.0x10 ⁴	2.0x10 ⁴	2.0x10 ⁴
Odoye	4.2x10 ⁴	4.9x10 ⁴	4.5x10 ⁴	1.0x10 ⁴	2.0x10 ⁴
Okenugbo	3.6x10 ⁴	3.2x10 ⁴	3.0x10 ⁴	1.0x10 ⁴	1.0x10 ⁴
Loji	3.0x10 ⁴	4.8x10 ⁴	2.2x10 ⁴	2.2x10 ⁴	1.7x10 ⁴
Tekunleoga	7.0x10 ⁴	3.1x10 ⁴	5.2x10 ⁴	4.8x10 ⁴	1.5x10 ⁴
Mamu	4.2x10 ⁴	3.2x10 ⁴	1.5x10 ⁴	5.2x10 ⁴	1.1x10 ⁴

Table 3 shows the total bacterial count of the stream samples. The total bacteria count revealed that Eriobinla stream water had the highest total bacteria count of 9.0×10^4 cfu/ml while Okenugbo and Odoye had the lowest total bacteria count of 1.0×10^1 cfu/ml. This indicated that Odoye and Okenugbo stream waters had the lowest bacterial indicators compared with all other stream waters investigated in this study.

Staphylococcus aureus, *Proteus* species, *Pseudomonas* species, *Escherichia coli* and *Klebsiella pneumoniae* were present in all samples; *Enterococcus faecalis* was present in Imosun and Eriobinla stream water samples; *Salmonella* species was present in all the stream water samples except from Odoralamo, and *Campylobacter* species was present in Eriobinla, Tekunleoga, Mamu and Imosun stream water samples (Table 4).

Table 4: Morphological and biochemical characteristics of bacteria present in stream water samples from eight villages in Ijebu North Local Government Area, Southwestern Nigeria.

TEST	Erilobinla	Odoralamo	Imosun	Odoye	Okenugbo	Tekonleoga	Loji	Mamu
Gram Stain	+	-	+	-	-	-	-	-
Shape	Cocci	Rod	Cocci	Rod	Rod	Rod	Rod	Spiral
Citrate utilization test	-	-	+	+	-	-	-	-
Indole test	-	+	-	-	-	-	-	-
Catalase test	+	-	-	-	-	-	-	+
Coagulase	+	-	-	-	-	-	-	-
Motility test	+	+	-	-	+	+	+	+
Oxidase test	-	-	-	-	-	+	-	+
Urease test	-	-	-	+	+	-	-	-
Methyl red test	+	-	-	+	-	-	+	+
Dnase test	+	-	-	-	-	-	-	-
Glucose fermentation	AG	AG	AG	ANG	AG	AG	AG	NA
Fructose fermentation	NA	AG	AG	AG	AG	AG	NA	NA
Lactose fermentation	AG	AG	AG	AG	NA	AG	NA	NA
Organism present	Staphylococcus aureus	Escherichia coli	Enterococcus faecalis	Klebsiella pneumoniae	Proteus sp	Pseudomonas aeruginosa	Salmonella sp	Campylobacter sp

Table 5: Antimicrobial sensitivity testing of gram-negative bacteria isolated from stream water samples from eight villages in Ijebu North Local Government Area, Southwestern Nigeria

Types of Antibiotics	Zones of inhibition in millimeter							
	Proteus sp	Pseudomonas aeruginosa	Escherichia coli	Salmonella sp	Klebsiella pneumoniae	Campylobacter sp		
Septtrin (SXT)	-	-	-	-	-	-	-	-
Chloramphenicol (CH)	-	-	-	2.0	1.0	-	-	-
Sparfloxacin (SP)	-	-	3.0	-	5.0	3.0	-	3.0
Ciprofloxacin (CPX)	5.0	6.5	7.5	3.0	3.0	3.0	-	3.0
Amoxicillin (AM)	1.0	4.0	1.0	1.0	2.0	-	-	-
Augmentin (AU)	-	1.0	1.0	-	2.0	-	-	-
Gentamycin (CN)	-	-	-	-	4.0	2.0	-	2.0
Pefloxacin (PEF)	3.0	3.5	8.5	1.5	2.0	-	-	-
Streptomycin (S)	2.0	2.5	3.5	-	-	3.0	-	3.0

Key: - = Resistance; 1.0 – 2.0 = partial zone of inhibition; 3.0 – 5.0 = clear zone inhibition; 6.0 – 8.5 = very clear zone inhibition

Table 5 shows the antimicrobial sensitivity patterns of the gram negative isolates. All the isolates were resistant to Septrin; *Salmonella* species and *Klebsiella pneumoniae* showed partial zone of inhibition to Chloramphenicol; *Escherichia coli*, *Klebsiella pneumoniae* and *Campylobacter* species showed clear zone of inhibition to Sparfloxacin; All the isolates were inhibited by Ciprofloxacin, Amoxicillin and Pefloxacin except from *Campylobacter* species that shows resistance to Amoxicillin and Peflox-

acin; *Pseudomonas* species, *Escherichia coli* and *Klebsiella pneumoniae* shows partial zone of inhibition to Augmentin; All isolates were resistant to Gentamycin except *Klebsiella pneumoniae* and *Campylobacter* species that showed clear zones of inhibition. All isolates were resistant to Tarivid except *Klebsiella pneumoniae* which showed a partial zone of inhibition. All isolates were inhibited by Streptomycin except from *Salmonella* species and *Klebsiella pneumoniae*.

Table 6: Antimicrobial sensitivity testing of gram-positive bacteria isolated from stream water samples from eight villages in Ijebu North Local Government Area, Southwestern Nigeria.

Types of antibiotics		Zones of Inhibition in Millimeter	
		Staphylococcus aureus	Enterococcus faecalis
Pefloxacin	(PEF)	6.0	3.0
Gentamycin	(CN)	1.0	-
Ampliclox	(APX)	2.0	1.0
Zinnacef	(Z)	-	-
Amoxicillin	(AM)	1.0	1.0
Recephin	(R)	2.0	-
Ciprofloxacin	(CPX)	5.0	2.0
Streptomycin	(S)	4.0	-
Septrin	(SXT)	-	-
Erythromycin	(E)	1.0	-

Key: - = Resistance; 1.0 – 2.0 = partial zone of inhibition; 3.0 – 5.0 = clear zone inhibition; 6.0 – 8.5 = very clear zone inhibition

Table 6 showed the antimicrobial sensitivity patterns of gram positive isolates. The two isolates (*Staphylococcus aureus* and *Enterococci faecalis*) were inhibited by Pefloxacin, Ampliclox, Amoxicillin and Ciprofloxacin; *Staphy-*

lococcus aureus showed partial zone of inhibition to Gentamycin The two isolates showed resistance to Zinnacef and Septrin and *Staphylococcus aureus* was inhibited by Recephin, Streptomycin and Erythromycin.

DISCUSSION AND CONCLUSION

The result from the physicochemical analysis conducted shows that the Physicochemical characteristics of the stream samples were not fully in agreement with the US EPA and WHO standards for drinking water (WHO, 2000; USEPA, 2002; USEPA, 2003). The result obtained revealed that the total bacteria count of these stream samples exceeded the recommended standards of potable water which is 1.0×10^2 cfu/ml on nutrient agar medium. This is in line with a previous report in Abeokuta settlement of Nigeria by Edema *et al.*, (2001) who reported that the qualities of waters under study were poor. The microbiological and physicochemical parameters of different fresh water system (ocean, stream, river etc.) had also been investigated by various researchers (Edema *et al.*, 2001; Bezuidenhout *et al.*, 2002; Dere *et al.*, 2006; Yayintas *et al.*, 2007a; Yayintas *et al.*, 2007b; Moskovchenko *et al.*, 2009; Egberongbe *et al.*, 2010), who all reported that their qualities were unsatisfactory.

The bacteria isolated according to morphological and biochemical characteristics include: *Escherichia coli*, *Campylobacter* sp, *Salmonella* sp, *Pseudomonas aeruginosa*, *Klebsiella pneumoniae*, *Proteus* sp and *Enterococcus faecalis*. The presence of these bacteria is in harmony with previous reports by Edema *et al.* (2001), Lehloesa and Muiyiwa (2004), WHO (2004) and Oketola *et al.* (2006). These enterobacteria are reportedly the causative agents of various diseases and complications in man (Solo-Gabriele *et al.*, 2000, WHO, 2004).

Likewise, studies conducted by Banwo (2006) and Egberongbe *et al.* (2010) have shown that in the absence of any known

sources of human or animal waste, *Enterococcus* sp and *Escherichia coli* are consistently present and recovered in high concentration in the sub – tropical environments. The isolated bacterial species in this study have been identified to be the same with those commonly encountered in water and aquatic environments as was also reported in a study on streams surface water in Wyoming, U.S.A. as reviewed by Banwo (2006). Different types of pollution characterize the different stream waters in the different communities. The antimicrobial sensitivity testing that was done for these isolates showed that they are resistant to some antimicrobials, which is a potential threat to public health. The resistant organisms may find their ways into humans system by drinking contaminated water. They may also contaminate fish and other products within the food chain, and if consumed by humans, may become part of the individual flora (Edema *et al.*, 2000; Torranzos and Marcos, 2000; Scott *et al.*, 2003). Conclusively, the microbial qualities of the evaluated stream waters were averagely poor, and are certainly not fit for human consumption as they are of low quality threshold. This may be due to direct contamination by animal and human excreta and other anthropogenic activities such as swimming, washing of clothes, farming etc., and thus, require further purification to ensure their suitability for human utility.

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