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DISTRIBUTION AND TYPES OF WATER-BORNE BACTERIAL PATHOGENS IN RIVER SOKOTO, NIGERIA AND THEIR HEALTH IMPLICATION

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ABSTRACT

The quality of water from River Sokoto was assessed to determine its bacterial load and types. Standard bacteriological techniques were used to perform the total heterotrophic bacteria, faecal coliform and enterococci counts of water samples collected from six sampling points on the river and distribution of bacteria in the water samples was also determined using standard procedures. The study indicated high heterotrophic bacteria, faecal coliform and enterococci counts above permissible limits for drinking and recreational waters according to World Health Organization (WHO) and United States Environmental Protection Agency (USEPA). A total of 434 bacteria organisms were isolated comprising nineteen different species. Among the *Enterobacteriaceae, Escherichia coli,* which are human pathogenic organisms, had the highest percentage (11.98%) followed by *Enterobacter aerogenes* and *Klebsiella pneumoniae subspecies pneumonia. Pseudomonas aeruginosa* constituted the majority of non-*Enterobacteriaceae* Gram negative organisms. *Staphylococcus aureus* was the highest among the Gram positive organisms followed by *Staphylococcus saprophyticus* (5.99%). Other isolates in significant numbers are *Streptococcus faecalis*, *Bacillus subtilis*, *Elizabethkingia meningoseptica* and *Aeromonas sobria*. Bacteria of aquatic habitat like *Providencia rettgeri, Raoultella ornithinolytica, Staphylococcus cohnii subspecies urealyticus* and *Staphylococcus chromogenes* that have not been isolated before in the study area were also isolated. River Sokoto predominantly contained *E. coli* which is an indication of faecal contamination and that makes it unsuitable for drinking and agricultural uses. People in the area should be encouraged to practice adequate sanitation.

Key words: River Sokoto, water quality, bacterial pathogens, *E. coli*, health implication.

RÉPARTITION ET TYPES DE BACTÉRIES PATHOGÈNES TRANSMISES PAR L'EAU DE LA RIVIÈRE À SOKOTO, NIGERIA ET LEUR IMPLICATION SUR LA SANTÉ

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Résumé

La qualité de l'eau du fleuve Sokoto a été évalué pour déterminer sa charge bactérienne et types. Les techniques bactériologiques standard ont été utilisés pour effectuer l'ensemble des bactéries hétérotrophes, coliformes et entérocoques chefs d'eau prélevés dans six points de prélèvement sur la rivière et la distribution des bactéries dans les échantillons d'eau a été déterminé en utilisant les procédures standard. L'étude indiquait des bactéries hétérotrophes, coliformes et entérocoques compte au-dessus des limites acceptables pour l'eau potable et des eaux récréatives selon l'Organisation mondiale de la Santé (OMS) et l'United States Environmental Protection Agency (EPA). Un total de 434 organismes ont été isolées de bactéries comprenant dix-neuf espèces différentes. Parmi les entérobactéries, *Escherichia coli*, qui sont des organismes pathogènes, présente le pourcentage le plus élevé (11,98 %) suivie de *l'Enterobacter aerogenes Klebsiella pneumoniae* et sous-espèces la pneumonie. *Pseudomonas aeruginosa* constituaient la majorité des non-*Entérobactéries* organismes Gram négatif. Staphylococcus aureus a été le plus élevé parmi les organismes Gram positif suivi de *Staphylococcus saprophyticus* (5,99 %). D'autres isolats en nombres importants Streptococcus faecalis, Bacillus subtilis, *Elizabethkingia meningoseptica* et *Aeromonas sobria* Les bactéries de l'habitat aquatique comme *Providencia rettgeri*, *Raoultella ornithinolytica, Staphylococcus cohnii Staphylococcus chromogenes urealyticus* sous-espèce et qui n'ont pas été isolés avant dans la zone d'étude ont aussi été isolés. River Sokoto principalement contenues *E. coli*, qui est une indication de contamination fécale et qui le rend impropre à la consommation et les utilisations agricoles. Les gens dans la région devraient être encouragés à pratiquer un assainissement adéquat.

Mots clés : Fleuve Sokoto, la qualité de l'eau, de bactéries pathogènes, E. coli, l'implication de la santé.

INTRODUCTION

Water is an indispensable natural resource essential for the existence of all living creatures. It is required for various human daily activities such as drinking, cooking, tooth-brushing, bathing, washing utensils and also for agricultural and industrial purposes (1 ,2). However, poor water quality continues to be a leading cause of health problems especially in developing countries where it is estimated that 80% of all illnesses are linked to water and sanitation and 15% of all child deaths under the age of 5 years result from diarrhoeal diseases (3, 4). Currently, an estimated 884 million people worldwide do not use improved sources of drinking water and 2.6 billion are not provided with adequate sanitation. The majority of these are in Southern Asia (25%) and sub-saharan Africa (37%) (5). In Nigeria, increasing population and infrastructural breakdown have made municipal pipe borne water to be inadequate in quantity and quality (6). Today, less than 30% Nigerians have access to safe drinking water due to these inadequacies and most of the populations have to resort to drinking water from wells and streams especially in the rural and suburban communities. These water sources are largely untreated and might harbour waterborne and vector-borne diseases such as cholera, typhoid fever, diarrhoea, hepatitis and guineaworm (7-9). These diseases are caused by pathogenic bacteria, viruses, protozoa and other microbes which are shed in human faeces and pollute water supplies which people utilize for drinking and washing purposes. Many rivers, streams and wells worldwide are affected by faecal contamination leading to increased health risks to persons exposed to the water, degradation of recreational and drinking water quality (10-17).

Pollution of river waters with pathogenic microorganisms has been on steady increase in the recent past. The major source of microbes in water is faeces from human and other mammals (18). Entry of pathogens into rivers can occur either from a point source, non- point sources or both. Non-point source microbial pollution of rivers occurs from rainwater surface run-offs, storm sewer spillages or overflow, while point-source pollution comes from discharge of untreated or partially treated effluents from wastewater treatment plants (19). One of the most frequent types of contamination in rural communities is faecal pollution from different sources, most frequently livestock and inadequate on-site human waste disposal systems (20). Microbiological contamination, is therefore dispersed, sporadically and influenced by a range of interacting environmental factors such as the watersheds physical characteristics, climatic conditions and the activities of man like waste disposal and agricultural management practices (21). The quality of water from River Sokoto was therefore assessed to determine its bacterial load.

MATERIAL AND METHODS

Study area

The segment of River Sokoto around Kalanbaina industrial area of the metropolis where there are industries, human settlements and irrigation farming activities was used in the study. Six sampling points were chosen namely; a point 5 kilometres away from farmland (P1), a point close to farmland about 2 kilometres from P1 (P2), a point close to residents along the riverside about 4 kilometres from P2 (P3), a point on stream drainage immediately from Sokoto Cement Factory (P4), a point on the stream just about to enter the river about 3 kilometres from P4 (P5), and a point 2 kilometres away from P5 on the river (P6) (Figure 1).

Bacteriological analysis

Water samples collected from six sampling points on River Sokoto were analysed for their bacterial load. Heterotrophic bacteria, coliforms and enterococci counts; and types of bacteria were determined by serial dilution and plating of water samples on differential culture media following the method of (22). Discrete isolates from heterotrophic plate agar, kept on nutrient agar slant were subjected to Gram Staining. Gram negative bacteria and Gram positive *Staphylococcus* species were isolated and identified using identification kits – ID 32E (BioMerieux, France) and MICROBACT STAPH 12S (Oxoid Ltd, England). Other Gram positive bacteria (*Streptococcus faecalis* and *Bacillus subtilis*) were identified and characterized biochemically following the methods described in Bergey's Manual of Systematic Bacteriology (23).

RESULTS

The heterotrophic plate counts (HPC), faecal coliforms (FC) and faecal streptococci (FS) counts of water samples collected from River Sokoto for the sampling period between January and December, 2014 are presented in Table 1. The result showed HPC in tens of thousands CFU/ml, FC counts in thousands CFU/ml and FS counts in hundreds CFU/ml. As shown in the table, sampling point P1 recorded the highest heterotrophic and total coliforms counts during the sampling periods with highest count in July, 2014. On the other hand, lowest heterotrophic and coliform counts were recorded at site P4 which is the point through which effluents from Sokoto Cement Factory entered River Sokoto.

TABLE 1: HETEROTROPHIC BACTERIA AND COLIFORM COUNTS OF RIVER SOKOTO WATER AT DIFFERENT SAMPLING POINTS IN JANUARY TO DECEMBER, 2014

Month	Sampling point	HPC	FCC	FSC	
J	P1	1.25 ± 0.48 x 10^5	1200	160	
A	P2	0.98 ± 0.27 x 10^5	1800	110	
N	P3	0.92 ± 0.15 x 10^5	1100	110	
	P4	0.18 ± 0.78 x 10^5	900	100	
	P5	1.00 ± 0.83 x 10^5	1000	100	
	P6	1.20 ± 0.33 x 10^5	1500	110	
F	P1	2.00 ± 0.28 x 10^5	1800	180	
E	P2	1.13 ± 0.18 x 10^5	1600	150	
B	P3	1.82 ± 0.21 x 10^5	1600	120	
	P4	0.19 ± 0.26 x 10^5	1100	118	
	P5	1.45 ± 0.25 x 10^5	1400	120	
	P6	1.75 ± 0.48 x 10^5	1800	160	
M	P1	3.00 ± 0.40 x 10^5	2000	220	
A	P2	2.75 ± 0.39 x 10^5	1800	180	
R	P3	1.65 ± 0.21 x 10^5	1700	150	
	P4	0.39 ± 0.47 x 10^5	1500	120	
	P5	1.20 ± 0.45 x 10^5	1700	130	
	P6	2.82 ± 0.30 x 10^5	2000	200	
A	P1	3.02 ± 0.24 x 10^5	2100	242	
P	P2	1.28 ± 0.63 x 10^5	2000	200	
R	P3	1.97 ± 0.31 x 10^5	2000	200	
	P4	0.42 ± 0.12 x 10^5	1500	130	
	P5	2.86 ± 0.38 x 10^5	1800	160	
	P6	2.82 ± 0.33 x 10^5	2100	210	
M	P1	3.45 ± 0.53 x 10^5	2700	300	
A	P2	2.91 ± 0.44 x 10^5	2500	220	
Y	P3	2.48 ± 0.34 x 10^5	2500	200	
	P4	0.95 ± 0.99 x 10^5	1800	160	
	P5	1.26 ± 0.14 x 10^5	2200	160	
	P6	3.32 ± 0.48 x 10^5	2500	300	
J	P1	5.36 ± 0.62 x 10^5	3200	400	
U	P2	3.92 ± 0.35 x 10^5	3000	250	
N	P3	3.80 ± 0.60 x 10^5	2900	220	
	P4	3.60 ± 0.56 x 10^5	2000	160	
	P5	3.68 ± 0.28 x 10^5	2700	180	
	P6	4.86 ± 0.72 x 10^5	3000	310	
J	P1	8.20 ± 0.26 x 10^5	4500	800	
U	P2	7.00 ± 0.96 x 10^5	4200	260	
L	P3	6.08 ± 0.10 x 10^5	3800	230	
	P4	3.84 ± 0.72 x 10^5	2600	200	
	P5	4.20 ± 0.68 x 10^5	3500	220	
	P6	8.05 ± 0.22 x 10^5	4200	320	
A	P1	6.02 ± 0.72 x 10^5	4000	530	
U	P2	4.89 ± 0.30 x 10^5	3200	220	
G	P3	4.20 ± 0.18 x 10^5	2700	210	
	P4	2.20 ± 0.62 x 10^5	2400	160	
	P5	3.02 ± 0.86 x 10^5	2500	200	
	P6	5.73 ± 0.78 x 10^5	3300	300	
S	P1	4.75 ± 0.60 x 10^5	2600	300	
E	P2	3.80 ± 0.57 x 10^5	2300	200	
P	P3	3.02 ± 0.48 x 10^5	2200	180	
	P4	1.96 ± 0.56 x 10^5	1800	150	
	P5	3.00 ± 0.86 x 10^5	2200	180	
	P6	4.26 ± 0.71 x 10^5	2500	290	
O	P1	3.80 ± 0.54 x 10^5	2300	280	
C	P2	2.76 ± 0.43 x 10^5	1800	180	
T	P3	2.64 ± 0.35 x 10^5	1700	160	
	P4	1.62 ± 0.62 x 10^5	1500	150	
	P5	1.94 ± 0.74 x 10^5	1600	150	
	P6	3.00 ± 0.40 x 10^5	2200	280	
N	P1	3.49 ± 0.56 x 10^5	2300	250	
O	P2	2.64 ± 0.28 x 10^5	1800	180	
V	P3	1.92 ± 0.56 x 10^5	1600	140	

	P4	$0.98 \pm 0.38 \times 10^5$	1200	120	
	P5	$1.80 \pm 0.45 \times 10^5$	1500	120	
	P6	$2.75 \pm 0.39 \times 10^5$	1800	250	
D	P1	$2.75 \pm 0.87 \times 10^5$	1800	240	
E	P2	$1.32 \pm 0.16 \times 10^5$	1700	150	
C	P3	$1.20 \pm 0.94 \times 10^5$	1200	120	
	P4	$0.85 \pm 0.62 \times 10^5$	1000	100	
	P5	$1.10 \pm 0.68 \times 10^5$	1200	100	
	P6	$1.80 \pm 0.56 \times 10^5$	1800	200	

KEY: HPC = Heterotrophic plate count; FCC = Faecal Coliforms count; FSC = Faecal Streptococci count.

TABLE 2: DISTRIBUTION AND PERCENTAGE FREQUENCY OF BACTERIA ORGANISMS ISOLATED FROM WATER SAMPLES COLLECTED FROM RIVER SOKOTO

Organisms	Number Isolated	% Frequency
Enterobacteriaceae		
Klebsiella pneumoniae subspecies pneumonia	**38**	**8.76**
Klebsiella oxytoca	**13**	**3.00**
Enterobacter cloacae	**26**	**5.99**
Enterobacter aerogenes	**38**	**8.76**
Salmonella typhi	**28**	**6.45**
Shigella flexneri	**23**	**5.30**
Escherichia coli	**52**	**11.98**
Providencia rettgeri	**13**	**3.00**
Raoultella ornithinolytica	**26**	**5.99**
Non-*Enterobacteriaceae* Gr-ve org		
Elizabethkingia meningoseptica	**26**	**5.99**
Pseudomonas aeruginosa	**32**	**7.37**
Aeromonas sobria	**24**	**5.53**
Gram positive isolates		
Staphylococcus aureus	**30**	**6.91**
Staphylococcus saprophyticus	**26**	**5.99**
Staphylococcus epidermidis	**13**	**3.00**
Staphylococcus cohnii subspecies urealyticus	**4**	**0.92**
Staphylococcus chromogenes	**4**	**0.92**
Streptococcus faecalis	**10**	**2.30**
Bacillus subtilis	**8**	**1.84**
Total	**434**	**100**

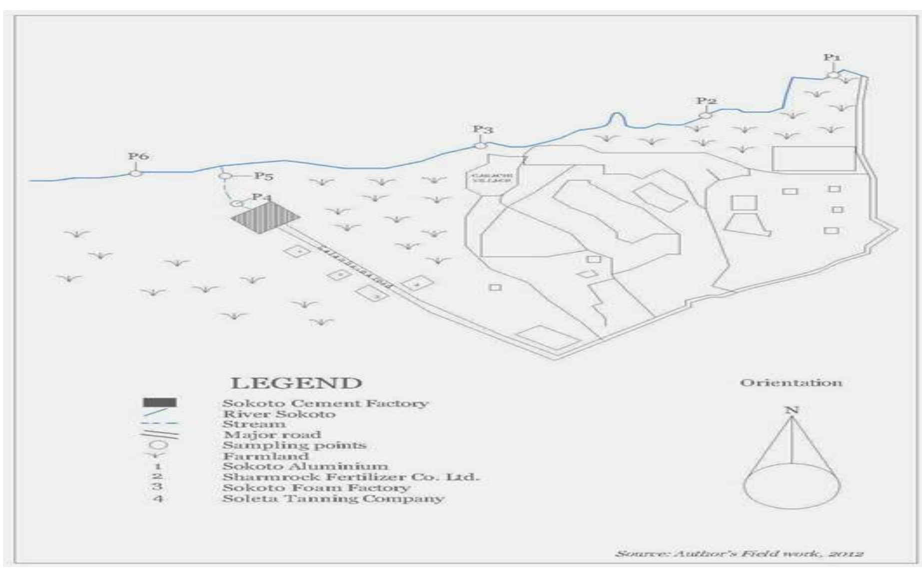

FIG. 1: MAP OF STUDY AREA OF RIVER SOKOTO SHOWING SAMPLING POINTS

Table 2 presents the distribution and types of bacteria isolated from water samples of the river. A total of 434 bacteria organisms were isolated comprising nineteen different species. Among the *Enterobacteriaceae, Escherichia coli* had the highest percentage (11.98%) followed by *Enterobacter aerogenes* and *Klebsiella pneumoniae subspecies pneumonia*. *Pseudomonas aeruginosa* constituted the majority of non-*Enterobacteriaceae* Gram negative organisms. *Staphylococcus aureus* was the highest among the Gram positive organisms followed by *Staphylococcus saprophyticus* (5.99%). Other isolates in significant numbers are *Streptococcus faecalis*, *Bacillus subtilis*, *Elizabethkingia meningoseptica* and *Aeromonas sobria*. Some rare bacteria in the study area which are of aquatic habitat like *Elizabethkingia meningoseptica, Klebsiella oxytoca, Providencia rettgeri, Raoultella ornithinolytica, Staphylococcus cohnii subspecies urealyticus* and *Staphylococcus chromogenes* were also isolated.

DISCUSSION

River Sokoto is a major source of water for domestic, agricultural and industrial uses in Sokoto metropolis. It is the source water for the water treatment plant that supplies pipe-borne water need of the people in the metropolis. Residents in the locality use water from the river for washing and bathing. The river water is also used to irrigate adjoining farmland, where crops such as onions, sweet potatoes, carrots, millets, tomatoes and vegetables, some of which are often eaten raw, are cultivated. The factories in the locality use the river as source water for various purposes such as water for cooling and washing. People swim and fish in the river and its sand is being dredged for building construction. All these human activities with other environmental factors would negatively impact the physical, chemical and microbiological quality of River Sokoto. This could also pose serious health and environment hazards to the community, as stated by (24), who inferred that waterborne pathogens present greater health risk to people using river water for drinking, bathing, irrigation of crops eaten raw, fishing, and recreational activities. Because of many activities going on around River Sokoto, it is therefore imperative to study the river to know the impact of these factors on the river with the aim of creating awareness on the quality of the river water for the safety of the people using it. The mean total heterotrophic counts, faecal coliform and faecal streptococci counts of the water samples of River Sokoto, shown in Table 1 were higher than the permissible limit recommended by (25) and (26).

This high bacterial load might be as a result of poor hygiene and sanitation such as bathing, cloth washing and defecating in and around the river, which are common practices in the study area. Lack of proper sanitation in urban cities has been cited as the main cause of high bacterial pathogens in rivers traversing major world cities (27). Unhygienic defecation on ground causing contamination of surface water has also been reported in other studies (28-30). The finding of this study corroborates that of (31) where high level of heterotrophic plate count (HPC), coliform and enterococci counts were also recorded on all stream water samples from sixteen sampling points on Esinmirin Stream in Ile-Ife, Nigeria. High level heterotrophic and coliform counts observed in this study also agree with the findings of (32) where adjoining two drainage streams from industrial and residential areas, impact negatively the water quality of River Kaduna in Nigeria. While sampling point (P1) about 2 kilometers away from farmland had the highest heterotrophic and coliform counts, site P4 on the stream that carried effluents from Sokoto Cement Factory into the river had the least values throughout the year. Cement factories are not normally associated with large volumes of liquid waste.

Large numbers of *Escherichia coli*, among the *Enterobacteriaceae* followed by *Enterobacter aerogenes* and *Klebsiella pneumoniae subspecies pneumonia* isolated from River Sokoto has health implications. *Escherichia coli* are gram-negative bacteria that can survive in an environment with or without air (facultative anaerobes). In fact, human faeces may consist of as much as 50-52% of *E. coli* (21). The fact that large numbers of *E. coli* were isolated from River Sokoto indicated that the river's major source of contamination is human. Presence of other *Enterobacteriaceae* (*Enterobacter aerogenes* and *Klebsiella pneumonia*) also constitute serious threat to the community. The most serious water pollutions in terms of human health worldwide are pathogenic organisms such as *Pseudomonas* and *Salmonella* (21). *Pseudomonas aeruginosa* coincidentally constituted the majority of non-*Enterobacteriaceae* Gram negative organisms isolated from the river. The most important water-related diseases these could cause include typhoid fever, cholera, bacterial and amoebic dysentery, hepatitis, malaria, yellow fever, filariosis and schistosomiasis. Also, large numbers of *Staphylococcus aureus*, *Staphylococcus saprophyticus* and others like *Streptococcus faecalis*, *Bacillus subtilis*, *Elizabethkingia meningoseptica* and *Aeromonas sobria* are of serious concern. Since the largest number of faecal coliform and faecal streptococci is always present in manure (33) then the presence of either of these microbes in a surface water sample is strong evidence of faecal contamination. The presence of coliform bacteria in water does not necessarily indicate water contamination by faecal waste; however the presence of faecal coliform in water may indicate recent contamination by human sewage or animal dropping which could contain other bacteria, viruses, or diseases causing organisms (21, 34) associated water-borne disease occurrences including acute gastrointestinal disease, cholera, dysentery, hepatitis- A, and typhoid with the use of Ganges River in India. Residents around River Sokoto use water from the river for similar purposes. Therefore, isolation of these pathogenic bacteria from River Sokoto could be the cause for rampant cases of water-borne infections in the study area as reported earlier by (35).

CONCLUSION

The fact that water from River Sokoto contained high level microbial load which were predominantly *E. coli* is an indication of serious faecal contamination and that makes it unsuitable for drinking and agricultural use. The water from this river is therefore not potable, and poses a health risk to residents that rely on it for domestic and agricultural purposes. Government should conduct surveillance and regular monitoring of rivers in order to provide good quality water and people should be encouraged to practice adequate sanitation to ensure human health and protect against a wide range of water-related diseases.

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