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Seasonal Influences on the Distribution of Bacterial Pathogens and Waterborne Diseases Transmission Potentials of Imo River, Nigeria.

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ABSTRACT:

The occurrence and population of pathogenic bacteria at certain points of Imo River, were determined. Water samples were collected for two seasons at major points of human activities along the river and subjected to standard microbiological and statistical analyses. Total heterotrophic bacterial count ($2.9 \times 10^9 - 3.7 \times 10^3$ CFU/ml) and total coliform bacterial counts ($9.0 \times 10^6 - 2.5 \times 10^2$ CFU/ml) showed significant variation at $P<0.05$ for the two seasons. Percentage occurrence of individual isolates across the sampling points showed the presence of *Escherichia coli* (100%), *Klebsiella* spp. (71.0%), *Shigella* spp. (71.0%), *Salmonella* spp.(71.0%), *Proteus* spp.(42.9%), *Vibro* spp. (42.9%), *Pseudomonas* spp. (42.9%), *Staphylococcus* spp. (85.7%), *Bacillus* spp. (100%), *Enterobacter* spp. (57.1%), *Citrobacter* spp. (14.3%), *Serratia* spp.(14.3%) and *Streptococcus* spp.(14.3%). These organisms are of public health importance and imply that Imo River should be protected from pollution to avoid possible diseases outbreak and transmission.

Keywords:
Imo River, pathogenic bacteria, percentage occurrence, diseases outbreak and transmission.

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INTRODUCTION

Imo river in Nigeria cuts across three eastern states and beyond, it serves as a source of water for domestic activities, agricultural irrigation, industrial and recreational activities for more than 4 million inhabitants. Many tributaries, domestic sewage, and industrial effluents empty into Imo River causing the river to serve as a reservoir of pathogenic organisms and toxic chemicals.

Microbial pathogens are ubiquitous in nature and are the second – leading cause of water body impairment in the United States (USEPA, 2004), Nigeria and other developing countries were not left out. Once microbial pathogens are in a stream, lake, or estuary, they are capable of causing gastrointestinal, respiratory, skin, eye, ear, nose, and throat diseases in humans (USEPA, 1986). Pathogenic diseases outbreaks following recreational contact with pathogen – contaminated surface waters have been well documented. For instance, in 1982, an outbreak of gastrointestinal illness occurred among New York City police and firefighter scuba divers who swam in the Hudson and East rivers (USEPA,1998). In 1998, a review of 22 studies of recreational waters showed that the indicator organisms correlate most closely with enteric illness are fecal *Enterococcus* and *Streptococcus* spp for fresh water and *Escherichia coli* for fresh water only *Streptococcus* spp alone for marine water (Pruss, 1998).

Microorganisms in surface waters may originate from several sources (Schets *et al*., 2008). It has been demonstrated that pathogenic microorganisms enter surface waters through discharges of raw and treated sewage and manure runoff from agricultural land (van den Berge *et al*., 2005; Lodder). The presence of waterborne pathogens in bird feces has frequently been reported. In Europe, *Campylobacter jejuni* has been detected in gull (*Larus* spp.) feces in Northern Ireland (Moore *et al*., 2002) and Sweden (Broman *et al*., 2002). This information suggests that heavy rainfall events may contribute to increased concentrations of pathogens in Imo River.

Despite the awareness of sources possibly contributing to surface water contamination, no data were available on the seasonal water quality and distribution of pathogenic bacteria in Imo river, Nigeria. This study was therefore aimed at determining the presence and quantity of waterborne bacterial pathogens at different points of the Imo River.

Description of Study Area

The study area of Imo River and is as shown in figure 1. Imo River is one of the major rivers in the south-eastern Nigeria. It probably originates from Isiochi in Abia State and cuts across three states including Abia, Imo, and Rivers states. Imo River flows from the eastern-north to the eastern-south, emptying in the Atlantic Ocean. The river serves as a source of water for domestic uses, fishery, recreational activities, and agricultural irrigation programs for more than 5 million people settling close to the water body. Apart from the above listed uses, the river serves as a source of sand for sand excavators, recipient of industrial effluent discharges, dumping site for domestic wastes including sewage and industrial solid waste, and other rivers like Aba River emptying in Imo River. Some major human impacted points of the river include Ekenobizi, Udo, Owerrinta, Alulu, Owaza, Akwette, and Obigbo are highly notable.

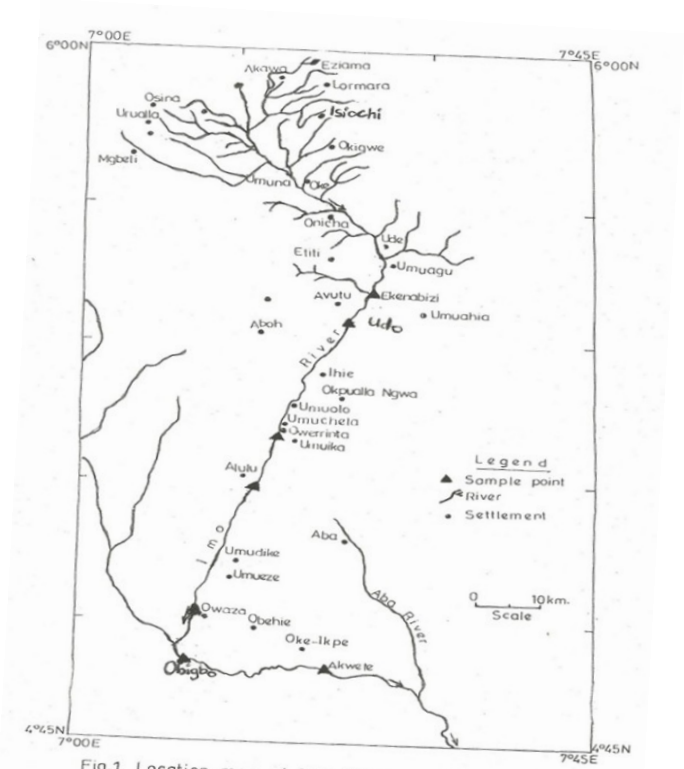

Fig.1 Location map of Imo River showing the sampling points.

MATERIALS AND METHODS:

Sample collection:

Surface water samples were collected from 7 major human impacted points of Imo River. Samples were collected in triplicates with the aid of sterile 1 liter water sampling cans. Collected samples were transported to the laboratory within 4

hours. The samples were collected in two seasons – dry and rainy seasons for two years. The dry season was between November and March while the rainy season was between May and September.

Microbiological analysis

Sterilization of media was carried out by moist heat sterilization method using autoclave at 121^0C, 15psi for 15 minutes. Heat stable materials were sterilized using hot air oven at 160^0C for 1 hour as described by Cruickshank *et al*. (1982). Heat labile materials were aseptically rinsed with alcohol and distilled water. The water samples were aseptically subjected to 10 fold serial dilutions to dilute the population of microorganism sufficiently in sterile blanks of 9ml peptone water and then plated to produce discrete colonies for easy enumeration. The media used include Nutrient agar, MacConkey agar, Eosin Methylene Blue agar, TCBS, and *Salmonella – Shigella* agar. All media were prepared as directed by the manufacturer. The method was adopted for the inoculation of media. Spread plates of appropriately diluted samples were incubated at 37^0C for 24 hours for heterotrophic bacterial count (THBC) while total coliform bacterial count (TCBC) were determined after incubation at 45^0C for 24 hours in MacConkey agar. Identification of isolates was based on the scheme described by Cheesborough (1984).

The results were subjected to Analysis of Variance (ANOVA) by using Statistical Program for Social Sciences (SPSS) and percentage occurrence.

RESULTS AND DISCUSSION:

Results:

The result of seasonal variation of Total Heterotrophic Bacterial Count (THBC) and Total Coliform Bacterial Count (TCBC) at all sampling locations of the Imo River is as shown in **Table 1.**The THBC during the dry season ranged from 3.6 x 10^3 -1.23 x 10^6cfu/ml while the TCBC during the dry season ranged from 1.65 x 10^3 – 8.7 x 10^6 cfu/ml. These results were above the limits of EPA Maximum Contaminant Levels (MCLS) of <100cfu/ml in drinking water (USEPA, 2003).

Table 2 shows the % occurrence[a] of bacterial isolates from the different sampling point or location of Imo River. A total of 14 organisms were isolated from the different locations. These organisms include *Escherichia coli* (100%), *Klebsiella* spp (71.0%), *Shigella spp* (71.0%), *Salmonella* spp (71.0%), *Proteus* spp (42.9%), *Vibro* spp(42.9%), *Pseudomonas* spp(85.7%), *Staphylococcus* spp (85.7%), *Bacillus* spp (100.0%), *Enterobacter* spp (57.1%), *Citrobacter* spp (14.3%), *Serratia* spp (14.3%) and *Streptococcus* spp (14.3%). This implied that *Escherichia coli* and *Bacillus* spp were isolated from all the 14 sampling points, *Pseudomonas* spp and *Staphylococcus* spp isolated from 6 out of the 7 sampling points and so on.

The % occurrence[b] of total bacterial isolates at each sampling point showed Owerrinta (69.2%), Udo (61.5%), Alulu (61.5%), Akwette (38.4%), Ekenobizi (76.9%), Owaza (53.8%), and Obigbo (53.8%).

Table 3 shows the result of seasonal variation of THBC and TCBC of Ekenobizi Point of Imo River. There were significant seasonal variations ($P<0.05$) in THBC at P = 0.00 and T-31.92. The mean THBC values were 1.2 x 10^6 cfu/ml (Dry) and 5.0 x 10^5 cfu/ml (Rainy). There were no significant seasonal variations in TCBC at P = 0.197 and T= 1.54. The mean TCBC values were 8.7 x 10^5 cfu/ml (Dry) and 3.7 x 10^4 cfu/ml (Rainy).

TABLE 1: SEASONAL VARIATION OF THE THBC AND TCBC AT SOME POINTS OF IMO RIVER WATER SAMPLES

	THBC		TCBC	
	DRY cfu/ml	RAINY cfu/ml	DRY	RAINY
Owerrinta	1.8 x 10^4	5.5 x 10^4	4.8 x10^3	2.0 x 10^4
Udo	2.0 x 10^5	2.9 x 10^9	1.9 x10^4	8.9 x 10^6
Alulu	8.6 x 10^3	1.3 x 10^7	5.0 x10^3	1.0 x 10^6
Akwette	4.1 x 10^4	5.6 x 10^6	1.0 x10^4	5.0 x 10^4
Ekenobizi	1.2 x10^6	5.0 x 10^5	8.7 x10^5	3.7 x 10^4
Owaza	3.7 x10^3	4.1 x 10^3	1.7 x10^3	2.5 x 10^2
Obigbo	4.5 x 10^4	2.9 x 10^6	1.9 x 10^4	3.0 x 10^3

Table 2: Percentage occurrence of microbial isolates at different sampling points of Imo River

Sampling point	*E. coli*	*Klebsiella* spp	*Shigella* spp	*Salmonrlla* spp	*Proteus* spp	*Vibro* spp	*Pseudomonas* spp	*Staphylococcu* spp	*Bacillus* ssp	*Entrobacter* spp	*Citrobacter* spp	*Serratia* spp	*Streptococcus* spp	% Occurence [b]
Owerrinta	+	+	+	+	+	-	+	+	+	-	+	-	-	69.2%
Udo	+	+	+	+	-	-	-	+	+	+	-	-	+	61.5%
Alulu	+	+	+	+	-	-	+	+	+	+	-	-	-	61.5%
Akwette	+	-	-	-	-	+	+	-	+	-	-	+	-	38.4%
Ekenobizi	+	+	+	+	+	+	+	+	+	+	-	-	-	76.9%
Owaza	+	+	-	-	+	+	+	+	+	-	-	-	-	53.8%
Obigbo	+	-	+	+	-	-	+	+	+	+	-	-	-	53.8%
%Occurence [a]	100.0	71.0	71.0	71.0	42.9	42.9	85.7	85.7	100.0	57.1	14.3	14.3	14.3	

a = % occurrence of individual isolate across the sampling points
b = % occurrence of total bacterial isolates at each sampling point
+ = Present and
- = absent.

Table 4 shows the result of seasonal variation of THBC and TCBC of Udo point of Imo River. There were significant seasonal variations in THBC at P = 0.00 and T-99.98. The mean THBC values were 2.0 x 10^5 cfu/ml (Dry) and 2.0 x 10^9 cfu/ml (Rainy). There were significant seasonal variations in TCBC at P = 0.00 and T= 11.85. The mean TCBC values were 1.9 x 10^4 cfu/ml (Dry) and 8.9 x 10^6 cfu/ml (Rainy).

Table 5 shows the result of seasonal variation of THBC and TCBC of Owerrinta Point of Imo River. There were significant seasonal variations in THBC at P = 0.00 and T = 32.04. The mean THBC values were 1.8 x 10^4cfu/ml (Dry) and 5.5 x 10^4 cfu/ml (Rainy). There were significant seasonal variations in TCBC at P = 0.00 and T= 60.21. The mean TCBC values were 4.8 x 10^3cfu/ml (Dry) and 2.0 x 10^4cfu/ml (Rainy).

Table 6 shows the result of seasonal variation of THBC and TCBC of Alulu Point of Imo River. There were significant seasonal variations in THBC at P = 0.00 and T = 85.99. The mean THBC values were 8.6 x10^3cfu/ml (Dry) and 1.3 x 10^7 cfu/ml (Rainy). There were significant seasonal variations in TCBC at P = 0.00 and T= 32.70. The mean TCBC values were 5.0 x 10^3cfu/ml (Dry) and 1.0 x 10^6cfu/ml (Rainy).

Table 7 shows the result of seasonal variation of THBC and TCBC of Owaza Point of Imo River. There were significant seasonal variations in THBC at P = 0.01 and T = 4.47. The mean THBC values were 3.7 x10^3cfu/ml (Dry) and 4.1 x 10^3cfu/ml (Rainy). There were significant seasonal variations in TCBC at P = 0.00 and T= 23.35. The mean TCBC values were 1.7 x 10^3cfu/ml (Dry) and 2.5 x 10^2cfu/ml (Rainy).

Table 8 shows the result of seasonal variation of THBC and TCBC of Obigbo Point of Imo River. There were significant seasonal variations in THBC at P = 0.00 and T = 44.71. The

Table 3: Seasonal variation of THBC and TCBC of Ekenobizi Point of Imo River.

Season	X	SD	X-Diff.	T-value	P-value
THBC Dry	1.2 x10^6	1.0 x 10^8	4.8 x 10^5	31.9	0.00
Rainy	5.0 x 10^5	2.6 x 10^7			
TCBC Dry	8.7 x 10^5	1.7 x 10^6	1.7 x 10^4	1.5	0.197
Rainy	3.7 x 10^4	1.9 x 10^4			

Table 4: Seasonal variation of THBC and TCBC of Udo Point of Imo River.

	Season	X	SD	X-Diff.	T-value	P-value
THBC	Dry	2.0×10^5	3.4×10^7	1.9×10^5	99.98	0.00
	Rainy	2.9×10^9	1.7×10^{10}			
TCBC	Dry	1.9×10^4	2.6×10^5	1.8×10^4	11.85	0.00
	Rainy	9.0×10^6	3.0×10^7			

Table 5: Seasonal variation of THBC and TCBC of Owerrinta Point of Imo River.

	Season	X	SD	X-Diff.	T-value	P-value
THBC	Dry	1.8×10^4	1.0×10^5	3.7×10^4	32.04	0.00
	Rainy	5.5×10^4	1.7×10^5			
TCBC	Dry	4.8×10^3	1.3×10^4	4.6×10^3	60.21	0.00
	Rainy	2.0×10^4	2.6×10^6			

Table 6: Seasonal variation of THBC and TCBC of Alulu Point of Imo River.

	Season	X	SD	X-Diff.	T-value	P-value
THBC	Dry	8.6×10^3	1.7×10^4	8.5×10^3	85.99	0.00
	Rainy	1.3×10^7	1.0×10^7			
TCBC	Dry	5.0×10^3	2.6×10^4	4.9×10^3	32.70	0.00
	Rainy	1.0×10^6	4.5×10^6			

mean THBC values were 4.5 $x10^4$cfu/ml (Dry) and 2.9 x 10^6cfu/ml (Rainy). There were significant seasonal variations in TCBC at P = 0.00 and T= 18.30. The mean TCBC values were 1.9 x 10^4cfu/ml (Dry) and 3.0 x 10^3cfu/ml (Rainy)

Table 9 shows the result of seasonal variation of THBC and TCBC of Akwette Point of Imo River. There were significant seasonal variations in THBC at P = 0.00 and T 26.47. The mean THBC values were 4.1 $x10^4$cfu/ml (Dry) and 5.6 x 10^6cfu/ml (Rainy). There were significant seasonal variations in TCBC at P = 0.00 and T= 24.49. The mean TCBC values were 1.0 x 10^4cfu/ml (Dry) and 5.0 x 10^4cfu/ml (Rainy).

DISCUSSION:

The result of Total Heterotrophic Bacterial Count (THBC) and Total Coliform Bacterial Count (TCBC) as shown in table 1 were above the USEPA Maximum Contaminant Levels (MCLS) in drinking water (USEPA, 2003). The variation in the dry and rainy seasons might be due to heavy rainfalls resulting to sewage overflow and surface runoff. This was demonstrated by previous studies (Goyal *et al.,* 1977). These results suggest that heavy rainfall events may contribute to Imo River contamination and may give rise to increased concentration of pathogens in the water.

The coliform test is a reliable indicator of the possible presence of fecal contamination and is, consequently, correlated with pathogens. The USEPA MCL is less than one coliform per 100ml (USEPA, 2003).

The Total Heterotrophic Bacteria Count (THBC) test also called “total count” or “plate count”, provides an estimate of the total number of bacteria in a sample that will develop into colonies during a period of incubation in a nutrient. This test detects a broad group of bacteria including pathogens, and opportunistic pathogens, but it does

Table 7: Seasonal variation of THBC and TCBC of Owaza Point of Imo River.

Season	X	SD	X-Diff.	T-value	P-value
THBC Dry	3.7×10^3	1.7×10^5	4.5×10^4	4.47	0.01
Rainy	4.1×10^3	1.7×10^4			
TCBC Dry	1.7×10^3	1.7×10^5	2.3×10^2	23.35	0.00
Rainy	2.5×10^2	1.7×10^3			

not pretend to report all of the bacteria in the water sample examined. High THBC may be an indicator of poor general biological quality of drinking water (USEPA, 2003). There might be presence of emerging waterborne pathogenic bacteria like *Mycobacterium avium, Legionella* spp., *Helicobacter* spp., and *Aeromonas hydrophyla* in Imo River that might not be isolated through conventional laboratory techniques. Health agencies like the USEPA and World Health Organization (WHO) have avoided setting standards for plate counts possibly for lack of pathogenicity and great variation in density, encountered (Dezuane, 1990). A recommended MCL for human drinking water has not yet been proposed, but the USEPA does recognize the water quality deterioration implied by high plate counts. The upper limit for portable water is usually 500cfu/ml. Dezuane (1990) says that water with counts under 100cfu/ml should be considered "portable" and values 100-500/ml is "questionable". Therefore Imo River samples have questionable water quality.

Among these organisms, the members of the family *Enterobacteriaceae* include *Escherichia coli, Citrobacter* spp., *Klebsiella* spp, *Proteus* spp, *Enterobacter* spp, *Salmonella* spp, *Shigella* spp, and *Serratia* spp (Prescott *et al.,* 2005). As *E. coli* was isolated from all the sampling points, it indicated recent fecal contamination of the different sampling points. This result is supported by the works of Health Canada (2006) and Cabral (2010). *E. coli* is a coliform bacterium found exclusively in the digestive tract of warm blooded animals, including humans. This might be responsible for the highest percentage occurrence (Swerdlow *et al.,* 1992). As such *E. coli* is used in the drinking water industry as the definitive indicator of recent fecal contamination of water.

Citrobacter spp, *Klebsiella* spp, *Salmonella*, *Shigella*, and *Serratia* spp are present in most individuals although in low numbers, while *Proteus* spp and *Enterobacter* spp. are only present in minority of humans (Wilson, 2005). Therefore, these organisms are not suitable as indicators of fecal pollution of the environment. This might be responsible for their <100% distribution in the waters. This is supported by the work of Cabral (2010).While most strains of *E. coli* are non-pathogenic, some can cause serious diarrheal infections in human (Health Canada, 2006), urinary tract infections (Scheatz and Strockbin, 2005), and distribution of erythrocytes (Moe, 1997).*Citrobacter* spp (14.3%) is included in a number of pathogenic bacteria capable of causing serious disease and being discharged into rivers (Donovan *et al.,*2008); has ability to produce an enterotoxin and this become an intestinal pathogen in environments such as water, sewage, soil and food (Frederisksen and Sogaard, 2003).

Table 8: Seasonal variation of THBC and TCBC of Obigbo Point of Imo River.

Season	X	SD	X-Diff.	T-value	P-value
THBC Dry	4.5×10^4	1.7×10^5	4.4×10^4	44.71	0.00
Rainy	2.9×10^6	1.0×10^8			
TCBC Dry	1.9×10^4	2.6×10^5	2.8×10^3	18.30	0.00
Rainy	3.0×10^3	2.6×10^4			

Table 9: Seasonal variation of THBC and TCBC of Akwette Point of Imo River.

Season	X	SD	X-Diff.	T-value	P-value
THBC Dry	4.1×10^4	2.6×10^5	4.0×10^4	26.47	0.00
Rainy	5.6×10^6	1.7×10^7			
TCBC Dry	1.0×10^4	1.0×10^5	4.0×10^4	24.49	0.00
Rainy	5.0×10^4	2.6×10^5			

The presence of *Citrobacter* spp. is significant since the species - *C. freundii* can cause meningitis with high morbidity and mortality (Donovan *et al.,* 2008).

Klebsiella spp (71.0%) are ubiquitous in the environment. They have been found in a variety of environmental situations, such as soil, vegetation, or water, and they influences, many biochemical, and geochemical processes (Cabral, 2010). They have been recovered from aquatic environments receiving industrial wastewaters, plant products, fresh vegetables, food with a high content sugars and acids, frozen orange juice concentrate, sugarcane waste and living trees (Grimont *et al.,* 2005). It is because *Klebsiella* spp. has high % occurrence (71.0%) in Imo River and especially its isolation from Owerrinta Point of Imo River that receives effluents from paper mill industries. *Klebsiella* spp can cause human diseases, ranging from asymptomatic colonization of the intestinal, urinary, or respiratory tract to fatal septicemia. *Klebsiella* are mostly considered nosocomial pathogens (Grimont *et al.,* 2005).

The presence of *Salmonella* spp in Imo River samples might be due to contamination from municipal sewage agricultural pollution, and storm water runoffs. This argument is supported by the reports of WHO (2008) and Arvanitidov *et al.* (2005). Though *Salmonella* spp can survive in water bodies, its presence and multiplication can be influenced by seasonal conditions, temperature, humidity, and pH, which might be contributory to the <100% (71.1%) occurrence of *Salmonella* spp in Imo River water samples. This is supported by the work of Le Minor (2003). *Salmonella* spp are responsible for two types of salmonellosis: (1) Typhoid and paratyphoid fever; (2) gastroenteritis (Le Minor, 2003). This implies that controlled water sewage systems, pasteurization of foods and personal hygiene will reduce the incidence of typhoid fever (Popoff *et al.,* 2005) that might result from the use of Imo River.

The association of *Shigella* spp (71.0%) with some points of Imo River is an implication of the fecal contamination of the River. This is in agreement with the reports and works of WHO (2008) and Kapperud *et al.,* (1995). That *Shigella* spp with <100% occurrence at sampling points of Imo River might imply its presence at some points but are viable and non-culturable. This argument is supported by the report of Faruque *et al.* (2002).The implication of the presence of *Shigella* spp in Imo River samples is the risk of possible outbreak of shigellosis. This was in agreement with the report of Emch *et al.* (2008).

Enterobacter spp (57.1%) might be an implication of fecal contamination at Imo River. This was supported by the works of Grimont and Grimont (2005). Apart from fecal contamination *Enterobacter* spp might have been introduced from other sources like soil, polluted water, and plants. The implication of this ubiquity is that *Enterobacter* spp cannot be used as an indicator of fecal contamination of water bodies. The presence of *Enterobacter* spp in some samples of Imo River implied possible risk of outbreak of nosocomial infections such as urinary tract infections and other healthcare- associated infections. This argument is supported by the reports of Hirdron *et al.* (2008).

Proteus spp (42.9%) is an enteric pathogen associated with the feces of animals including humans. Its low % occurrence (42.9%) might be because it exists in minority of human feces. This is supported by the reports of Wilson (2005).

Serratia spp (14.3%) is another enteric pathogen associated with feces of some humans. Its lower % occurrence might be due to low presence in feces of humans and subsequently its lower availability as a water borne pathogen. This is supported by the report of Cabral (2010).

Vibro spp (42.9%) might be present in some samples due to contamination from birds, frogs, fishes and shell fish present in aquatic environments. This argument is supported by the

reports of Ali *et al.* (2001). They detected low distribution of *Vibrio* spp (42.9%) might be due to environmental stress caused by adverse environmental conditions making some of the cells viable but non-culturable, though retaining the potential for pathogenicity for significant periods of time. This is in agreement with the report of Alam *et al.* (2006) and Chaiyana *et al.* (2001). *Vibrio* spp especially *V. cholerae* is responsible for the disease cholera in humans (Cabral, 2010).

Streptococcus spp (14.3%) were isolated in one out of the seven locations and might be due to fecal contamination of the point of Imo River. This was supported by the report of Pruss (1998). Fecal *Streptococcus* spp is responsible for gastrointestinal illness (Donovan *et al.,* 2008).

Pseudomonas spp (85.7%) were isolated from 6 out of the 7 sampling locations of Imo River. They have been isolated from many environments (Prescott *et al*., 2005). Furthermore, their presence in many sampling points of Imo River might be due to their exceptional ability to degrade wide variety of organic molecules. Thus they might be very important in the mineralization process (microbial breakdown of organic materials to inorganic substances) in nature and in sewage treatment. *Pseudomonas* has been implicated as a major crude oil degrader (Nwaugo *et al*., 2006; Amund *et al.,* 1987 and Jain, 1992). These authors supported the possibility that *Pseudomonas* spp might be isolated from Akwette oil spill contaminated Point of Imo River. *Pseudomonas aeruginosa*, *Staphylococcus aureus,* and *Bacillus cereus* have been isolated as waterborne pathogens (Ichijo *et al.,* 2010).

Bacillus spp (100%) occurrence showed that *Bacillus* spp has been implicated in the degradation of hydrocarbons (Nwaugo, *et al.,*2006), *Bacillus subtilis and Bacillus cereus* in the degradation of diesel oil (Nwaogu, *et al.,*2008). *Bacillus* spp isolated from Imo River might be involved in the degradation of the oil and grease at some points of Imo River especially at Akwette Point.

Staphylococcus spp (85.7%) isolated from 6 out of the 7 sampling points excluding Akwette point of Imo River, might result from possible contamination from bodies of human beings using the locations of Imo River as recreational and other domestic activities. This was in agreement with the report of Kayser *et al.*(2005). This was also supported by the fact that *Staphylococcus* was not isolated from the oil contaminated at Akwette Point of Imo River because the river point was not used for domestic and recreational activities.

CONCLUSION:

Seasonal influences in addition to anthropogenic activities contributed to the occurrence of water borne pathogens with high bacteria counts above established standards. The River therefore is a source organism of public health importance and should be protected from human contamination and properly treated to avoid consequences.

REFERENCES:

Alam M, Hasan NA, Sadique A, Bhuiyan NA, Ahmed KU, Nusrin S, Nair GB, Siddique AK, Sack DA, Sack DA, Huq A and Colwell RR. 2006. Seasonal cholera caused by *Vibrio cholerae* Serogroups 01 and 0139 in the Coastal Aquatic Environment of Bangladesh. *Appl. Environ. Microbiol.,* 72:4096-4104

Ali M, Emch M, Yunus M and Sack RB. 2001. Are the environmental Niches off *Vibrio cholerae* 0139 different from *Vibrio cholerae* 01EI Tor? *Int. J. Infect.,* 5:214-219.

Amund OO, Adebowale AA and Ugorji EO. 1987. Occurrence and characteristics of hydrocarbons utilizing bacteria in Nigeria soils contaminated with spent motor oil. *Indian J. Microbiol.,* 27:63-87.

Arvanitidou M, Kanellou K and Vagiona DC. 2005. Diversity of *Salmonella.* Spp. And Fungi in Northern Greek Rivers and their Correlation to fecal indicators.*Environ. Res.,* 99:278-284.

Broman T, Palmgren H, Bergstram S, Sellin M, Waldenstram J, Danielson-Tham ML and Olsen B. 2002. *Campylobacter jejuni* in black headed gulls *Larus ridibundus* prevalence, genotypes, and influence on *C. jejuni* epidemiology. *J. Clin. Microbiol.,*40:4594-4602.

Cabral JP. 2010. Water Microbiology. Bacteria Pathogens and Water. *Int. J. Environ. Res. Public Health* 7:3657-3703.

Chaiyanan S, Chaiyanan S, Huq A, Maugel T and Colwell RR. 2001. Viability of nonculturable *Vibrio cholerae* 01 and 0139 system. *Appl. Microbiol.,* 24:331-341.

Cheesbrough M. 1984. *Medical Laboratory Manual for Tropical Countries*. Butterworth Co. Ltd.

Cruickshank R, Dugauid JP, Marmoin BP and Swain RHA. 1982. *"Medical Microbiology"*. The practice of Medical Microbiology 13th ed. Churchill Livingstone, Edinburgh. 2:273-284.

DeZuane, J. (1990). Handbook of Drinking Water Quality Standard and Controls, Van Nostrand Reinhold, New York.

Donovan E, Unice K, Roberts JD, Harris M and Finley B. 2008. Risk of gastrointestinal disease associated with exposure too pathogens in the water of the lower Passaic River. *Appl Environ Microbiol,* 74(4):994-1003.

Emch M, Ali M and Yunus M. 2008. Risk areas and Neighborhood- Level Risk Factors for *Shigella dysenteriae* 1and *Shigella flexneri. Health Place.* 14:96-104.

Faruque SM, Khan R, Kamruzzman M, Yamasaki S, Ahmad QS, Azim T, Nair GB, Takeda Y and Sack DA. 2002. Isolation of *Shigella dysenteriae* type 1 and *S. flexiner* strains from water in Bangladesh: Comparative molecular analysis of environmental *Shigella* isolates versus clinical strains. *Appl. Environ. Microbiol.,* 68:3908-3913.

Frederisksen W & Sogaard P. 2003. The genus *Citrobacter*. In *the Prokaryotes*: *An Evolving Electronic Resource for the Microbiological Community*, electronic release 3.14, 3rd ed.; Dworkin, M., Falkow, S., Rosenberg, E., Eds.; Springer- Verlag: New York, NY, USA.

Goyal SM, Gerba CP and Melnick JL. 1977. Occurrence and distribution of bacterial indicators and pathogens in canal communities along the Texas coast. *Appl. Environ. Microbiol.,* 34:139-149.

Grimont F and Grimont PAD. 2005. Genus *Klebsiella.* In *Bergey's Manual of Systematic Bacteriology,* 2nd ed. Brenner, D.J., Krieg, N.R., Staley, J.T., Eds Springer: New York, NY,USA. 2 (Part B):685-693.

Health Canada. 2006. Bacterial Waterborne Pathogens-Current and Emerging organisms of Concern. *Guidelines for Canadian Drinking Water Quality: Guideline Technical Document*, Ottawa, Ontario.

Hirdon AI, Edwards JR, Patel J, Horan TC. Sievert DM and Pollock DA. 2008. Antimicrobial resistance pathogens associated with Healthcare-associated infections: Annual summary of data reported to the National Safety Network at the Centers for disease control and prevention, 20062 007. *Infect. Control Hosp. Epidemiol.,* 29:996-1011.

Ichijo T, Yamaguchi N, Tani K and Nasu M. 2010. Oligonucleotide. Probes for phylogenetic detection of waterborne bacteria. *Journal of Health Science* 56(3):321-325.

Jain D. 1992. Effect of addition of *Pseudomonas* on oil spilled soil. *Indian J. Microbiol.* 3:5-8.

Kapperud G, Rorvik LM, Hasseltvedt V, Hoiby EA, Iversen BG, Staveland K, Johnson G, Leitao J, Herikstad H, Andersson Y, Langeland G, Gondrosen B and Lassen J. 1995. Outbreak of *Shigella sonnei* infection traced to imported iceberge lettuce. *J. Clin. Microbiol.,* 33:609-614.

Kayser FH. 2005. Bacteria and human pathogens. *In Medical Microbiology.* Kayser, F.H., Bienz, K.A., Eckert, J. and Zinkernagel, R.M (Eds.). Thieme, New York. 229-245.

Le Minor LE. 2003. In *the prokaryotes: An Evolving Electronic Resource for the Microbiological Community*, electronic release 3.14, 3rd ed.; Dworkin, M., Falkow, S., Rosenberg, E., Eds.; Springer- Verlag: New York, NY, USA.

Moe CL. 1997. Waterborne transmission of infectious agents, In: *Manual of Environmental Microbiology*. C. J. Hurst, G. R. Knudsen, M. J. Mclnerney, L. D. Stetzenbach, and M. V. Walter (eds.) ASM Press, Washington, DC.

Moore JE, Gilpin D, Grothers E, Canney A, Kaneko A and Matsuda M. 2002. Occurrence of *Campylobacter* spp. and *Crptospooridium* spp. in seagulls (*Larus* spp.) *Vector Borne Zoonotic Dis.* 2:111-114.

Nwaogu LA, Onyeze GOC and Nwabueze RN. 2008. Degradation of diesel oil in a polluted soil using *Bacillus subtilis. Afr. J. Biotechnol.,* 7 (12):1939-1943.

Nwaugo VO, Onyeagba RA and Nwachukwu NC. 2006. Effects of gas flaring on soil microbial spectrum in parts of Niger Delta area of Southern Nigeria. *Afr. J. Biotechnol.,* 5(19):1824-1826.

Popoff MY, Le Minor LE. 2005. Genus *Salmonella*. In: *Bergey's Manual of Systematic Bacteriology*, 2nd ed; Brenner, D.J., Krieg, N.R., Staley, J.T., Eds. Springer: New York, NY, USA. 2(Part B):764-799.

Prescott LM, Harley JP and Klein DA. 2005. *Microbiology*. 6th edition. McGraw Hill.

Pruss A. 1998. Review of epidemiology study of health effects from exposure of recreational water. *Int. J.Epidemiol.,* 27:1-9.

Schets FM, van Wijnen JH, Schijven JE, Schhoon H and de Roda Husman AC. 2008. Monitoring of waterborne pathogens in water in Amsterdam, The Netherlands, and the potential health risk associated with exposure to. *Appl Environ Microbiol.,* 74(7):2069-2078.

Scheutz F and Strockine NA. 2005. Genus *Escherichia.* In *Bergey's Manual of Systematic Bacteriology,* 2nd ed. Brenner, D.J., Krieg, N.R., Staley, J.T., Eds Springer: New York, NY, USA. 2 (Part B):6076:23.

Swerdlow DL, Woodruff BA, Brady RC, Griffin PM, Tippen S, Donnel HD, Jr., Geldreich E, Payne BJ, Meyer A, Jr., Wells JG, Greene KD, Bright M, Bean NH and Blake PA. 1992. A waterborne outbreak in Missouri of *Escherichia coli* O157: H7 associated with bloody and death. *Ann. Intern. Med.*, 17(10):812-819.

USEPA (United States Environmental Protection Agency). 2004. Report to Congress. Impacts and control of CSOs and SSOs. U.S Environmental Protection Agency, Washington, DC.

USEPA (United States Environmental Protection Agency). 2003. *Drinking Water Quality Standards*. Edstrom Industries, Waterford, Wisconsin.

USEPA (United States Environmental Protection Agency). 1998. *Giardia: human health criteria.* U.S Environmental Protection Agency, Washington, DC.

USEPA (U.S Environmental Protection Agency). 1986. *Ambient water quality criteria for bacteria.* U.S Environmental Protection Agency, Washington, DC.

Van den Berg HS, Lodders W, van der Poel W, Vennema H and Roda Husman AM. 2005. Genetic diversity of noroviruses in raw and treated sewage water. *Res. Microbiol.,* 156:532-540.

WHO (World Health Organization). 2008. *Guideline for drinking – water quality*. Incorporating 1st and 2nd Addenda, Recommendations, 3rd ed; WHO: Geneva, Switzerland. Volume 1.

Wilson M. 2005. *Microbial Inhabitants of Humans.* Their Ecology and Role in Health and Disease: Cambridge University Press: Cambridge, UK.