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PREVALENCE OF POTENTIAL NOSOCOMIAL BACTERIAL PATHOGENS IN LIQUID WASTE AND WASTE DUMP SOIL OF THREE MAJOR HOSPITALS IN CALABAR METROPOLIS, SOUTH-SOUTH, NIGERIA

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ABSTRACT

Hospital environment serves as a complex ecosystem that need proper intervention in order to have adequate infection control. The prevalence of potential nosocomial bacterial pathogens in liquid waste and waste dump soil of three major hospitals in Calabar Metropolis was investigated. The liquid waste and waste dump soil samples were collected and analyzed microbiologically. A total of 179 bacteria were isolated from the samples. 114 isolates were gram negative bacteria while 65 isolates were gram positive. The potential nosocomial bacterial pathogens isolated in this study were *Staphylococcus aureus*. 41(22.9%), *Escherichia coli*. 37(20.7%), *Streptococcus* spp. 21(11.7%), *Pseudomonas aeruginosa* 23(12.8%), *Salmonella* spp 14(7.8%), *Providenia* spp 9(5.0%), *Enteronacter aerogenes* 9(5.0%), *Klebsiella pneumoniae* 12(6.7%), *Bacillus cereus* 3(1.7%), *Chryseobacterium* spp 3(1.7%) and *Serratia mercescens* 2(1.1%). Statistical analysis carried out on the data obtained shows no significant ($p>0.05$) difference in the frequency of occurrence of nosocomial bacterial pathogens in liquids waste and waste dump soil of the three hospitals studied. The finding of this study shows that the hospital liquid waste and waste dump soil may be a potential reservoir of potential nosocomial bacterial pathogen that can cause nosocomial infections.

INTRODUCTION

Nosocomial infections popularly known as “Hospital Acquired/associated infection”, are those infection that develop in a patient during his/her stay in a hospital or other types of clinical facilities which were not present at the time of admission. (Likere *et al*., 2008). The

pathogen that causes nosocomial infections are therefore termed "Nosocomial Pathogens" Nosocomial pathogens may be bacteria, viruses and fungal parasites. Bacteria are the most common pathogen responsible for nosocomial infection. Some belong to the natural flora of the patient and cause infection only when the immune system of the patient is compromised. (Bergogne – Berezin and Towner, 1996). These micro-organisms vary depending upon different patient population medical facilities and even differences in environment in which the care is given (Hassan *et al*., 2017). The manifestation of nosocomial infections can occur during healthcare delivery for other diseases. Most nosocomial infections become clinically apparent while patients are still hospitalized; however, disease onset can occur after patient has been discharged. Beside harming patients, nosocomial infections can affect nurses, physicians, aides, visitors, salespeople, delivery personal, custodians and anyone who has contact with the hospital. Infections that are incubating when patients are admitted to a hospital are not nosocomial, they are community acquired (Prescott *et al.*, 2005).

Medical devices employed in modern health are often associated to nosocomial infections (CDC, 2016). Animate and inanimate sources of nosocomial infections include hospitals staffs, other patients, visitors, food, water, formites, urinary catheler, intravenous devices, respiratory equipment and other prostheses (Prescott *et al.*, 2005).

Nosocomial infections affect large number of patients globally, elevating mortality rate and financial loses significantly. According to estimate reported by WHO, approximately 15%of all hospitalized patients suffer from these infections (Wood *et al*., 2009; Apanga *et al*., 2014). These infections are responsible for the dead neonates, with incidence rate of 75% in south-east Asia and sub-saharan Africa (WHO, 2011). The incidence rate is higher in high income countries (3,5% and 12%),whereas in middle and low income countries, it varies between 2.7% and 19.1% (Mbim *et al*., 2016). The frequency of overall infections in low income countries is three times higher than in high income countries whereas the incidence is 3-20 times higher in neonates (WHO, 2011).

Risk factors associated with nosocomial infection depends upon the environment in which care is delivered, the susceptibility and condition of the patient, and the lack of awareness of such prevailing infections among staffs and health care providers.

Hospital environment plays an important role in nosocomial infection because the environment contains diverse population of microorganisms. These micro-organisms are

always present in great numbers in organic and moist environments, though some can also survive under harsh environmental condition (Ummu *et al.,* 2013). The hospital environment also serves as a potential reservoir of infectious agent since it houses both patients with diverse pathogenic micro-organisms and large number of susceptible/immuno-compromised individuals (Rhomberg *et al.,* 2006; Zhanel *et al.,* 2008). Nosocomial pathogens can come from either endogenic source or exogenous source. The endogenic source are those that are from the patient's own normal microbial flora while the exogenous source are those that comes from the surrounding environment. The bacteria that commonly cause nosocomial infections include; *Staphyloccus aureus, Streptococcus* spp, *Bacillus cereus, Acinetobacter* spp, coagulase negative *Staphylococci*, *Enterococci*, *Pseudomonas aeruginosa*, *Legionella* and members of the enterobacteriaceae (Esposito and Leone, 2007; Zhanel *et al*., 2008). The most frequently reported nosocomial pathogen have been *Escherichia coli*. *Staphylococcus aureus*, *Enterococci* and *P. aeruginosa*. (McCraig *et al*., 2006; Pitout *et al.,* 2005 and 2007). The occurrence of multidrug resistance in hospital-associated pathogens has resulted in emergence and re-emergence-of-difficult to treat nosocomial infection in patients.

Hospital waste are one of the most dangerous causes of pollution. Hospital waste refers to all biological or non-biological waste from hospitals which are discarded directly to the soil. Hospital wastes are generated during diagnosis and treatment or immunization of human being or animals. Hospital wastes are so infectious/hazardous that every mean of improper disposal pose a threat to the environment (Chikere *et al*., 2008). In developing countries like Nigeria, the incidence of Nosocomial infections can be devastating resulting in major disease outbreak in hospitals and other healthcare facilities. This may be attributed to poor infrastructure, overcrowding inadequate personal management in most hospitals and improper disposal of untreated hospital waste. This study was undertaken to investigate the prevalence and distribution of potential nosocomial bacterial pathogens in liquid waste and waste dump soil of three major hospitals in Calabar Metropolis-south-south, Nigeria.

MATERIALS AND METHODS

Sample Collection

Hospital Liquid waste: The hospitals liquid wastes were collected from three hospitals, from the outer chambers before discharging into the drainage system, following standard procedures. Samples were collected into a sterile container.

Hospital waste Dump Site soil: The soil samples were collected from the waste dump site of the hospitals. Surface soils were randomly collected using a sterile trowel. The samples were collected in triplicates.

Isolation of Bacterial Strain: A 10fold serial dilution of the samples was done and 10^{-4} and 10^{-5} dilutions was plated in duplicates by pour plate method using freshly prepared nutrient agar, it was incubated aerobically at 23^{0}C for 24hours.

Enumeration of Bacteria: After the period of incubation, the bacterial colonics the showed visible growth were counted by using colony forming unit (CFU) formula and average counts for duplicate cultures as total viable bacteria in the samples.

$$\text{Formula: CFU} = \text{Average count} \times \frac{1}{Dilution\ factor} \times \text{Amount plated (volume of sample)}$$

Purification of Bacterial Isolates: Bacterial colonics differing in size, shape and colour in different plates were selected and purified through repeated sub-culturing method. Streak plate methods were used. Nutrient agar was used as the media. When the culture plate yielded only one type of bacterial colony, the isolates were considered to be pure.

Characterization and Identification of Bacterial Isolates: The isolates were characterized based on colonial and cell morphology, growth on differential selective media and biochemical test which include gram's reaction, indole test, MR-VP test, citrate utilization, motility, utilization of carbohydrates such as glucose, sucrose, manitol, lactose and fructose, oxidase, catalase, coagulase, and starch hydrolysis test. The bacterial isolates were then identified by comparing their characteristics with those of known taxonomy using Bergey's manual of systematic Bacteriology.

STATISTICAL ANALYSIS

Replicate readings were managed using Microsoft Excel 2010. All the replicates readings for the various analyses were subjected to one way factor analysis of variance (ANOVA). Student t-test were used to compare paired mean readings. The results were presented as mean plus or minus standard deviation (Mean±SD). Mean values with probability values less than 0.05 ($p < 0.05$) were considered significant at 95% level of significance while those greater than 0.05 were not significant ($p > 0.05$). Confidence interval was set as described

previously (Uusippaikka, 1985) while analysis of variance of variance was done as reported by Nelson (1983).

RESULT AND DISCUSSION

The mean counts of bacteria isolated from the liquid waste and waste dump site soil of the three hospitals studied is presented in Table 1 & 2. The number of bacteria isolated from waste dump soil was higher (52.5%) compared to that of liquid waste (47.5%). The statistical analysis carried out on the values showed no significant different ($p>0.05$) in the number of bacterial isolated from the sampling points in the hospitals.

Table 1: Mean counts of bacterial isolates in liquid waste and waste dump soil from different sampling points in the three hospitals.

		Sampling points		
Hospitals	**Samples**	**A**	**B**	**C**
H1	WDS(CFU/g)	$1.46^{a} \pm 4.00 \times 10^{7}$	$1.46^{b} \pm 7.64 \times 10^{7}$	$1.48^{c} \pm 5.51 \times 10^{7}$
	LW (CFU/ml)	$1.40^{a} \pm 2.00 \times 10^{6}$	$1.39^{b} \pm 3.06 \times 10^{6}$	$1.68^{c} \pm 1.00 \times 10^{6}$
H2	WDS(CFU/g)	$1.89^{a} \pm 4.16 \times 10^{7}$	$1.88^{b} \pm 5.29 \times 10^{7}$	$1.72^{c} \pm 6.93 \times 10^{7}$
	LW (CFU/ml)	$1.67^{a} \pm 1.15 \times 10^{6}$	$1.62^{b} \pm 2.52 \times 10^{6}$	$1.59^{c} \pm 4.16 \times 10^{6}$
H3	WDS(CFU/g)	$1.52^{a} \pm 3.61 \times 10^{7}$	$1.36^{b} \pm 5.29 \times 10^{7}$	$1.33^{c} \pm 2.65 \times 10^{7}$
	LW (CFU/ml)	$8.7^{a} \pm 4.16 \times 10^{6}$	$8.2^{b} \pm 2.00 \times 10^{6}$	$8.2^{c} \pm 2.08 \times 10^{6}$

Superscripts a, b and c represents non-significant one way ANOVA p values ($p > 0.05$)

Table 2: Total mean count of bacteria isolated from 3 hospitals studied.

Hospitals	WDS(%)	LW(%)	Total(%)
H1	$147(49.7)^{a}$	$149(50.3)^{b}$	296(34.1)
H2	$183(53.9)^{a}$	$163(47.1)^{b}$	346(40.0)
H3	$140(62.5)^{a}$	$84(37.5)^{b}$	224(25.9)
TOTAL	$470(54.3)^{a}$	$396(45.7)^{b}$	866(100)

Superscripts a and b represents non-significant paired Student t-test p values ($p > 0.05$) across the various rows.

Table 3: Biochemical characterization of bacterial isolates.

Numbers of Isolates showing similar reaction	Gram's reaction	Catalase	Motility	Oxidase	Citrate	Indole	MR	VP	Coagulase	Gas	H2S	Manitol	Glucose	Sucrose	Lactose	Probable organism
37	-	+	+	-	-	+	+	-	NA	+	-	+	+	+/-	+	*Escherichia coli*
12	-	+	-	-	+	-	-	+	NA	+	-	+	+	+	+	*Klebsiella pneumoniae*
5	-	+	+	-	-	+	+	-	NA	+	+	-	+	-	-	*Proteus spp*
23		+	+	+	+	-	-	+	NA	-	-	+	-	-	-	*Pseudomonas aeruginosa*
41	+	+	+	-	+	+	+	+	+	+	-	+	+	+	+	*Staphylococcus aureus*
21	+	-	-	-	-	-	-	+	NA	+	-	-	+	+	+	*Streptococcus spp*
3	-	+	-	-	+	+	+	-	**NA**	-	-	+	+	-	+	***Chryseobacterium spp***
3	+	+	+	+	+	-	-	+	NA	-	-	+	+	+	+/-	*Bacillus cereus*
9	-	+	+	+	+	+	-	-	NA	+	+	+	+	+	-	*Providencia spp*
14	-	+	+	-	-	-	+	-	NA	+	+	+	+	-	-	*Salmonella spp*
9	-	+	+	-	+	-	-	+	NA	+	-	+	+	-	-	*Enterobacter aerogenes*
2	-	+	+	-	+	-	-	+	NA	-	-	+	+	+	-	*Serratia marcescens*

Key: NA: Not applicable, +/- : Variable

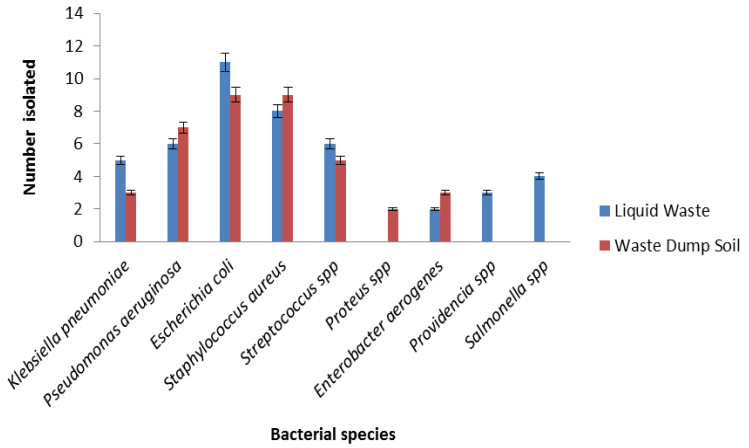

Fig. 1: Distribution of bacterial species from liquid waste and waste dump soil from Hospital H1.

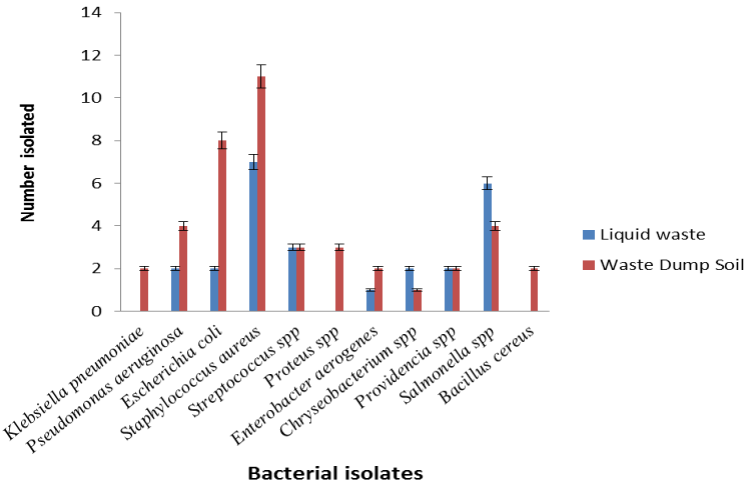

Fig. 2: Distribution of bacterial species from liquid waste and waste dump soil from Hospital H2.

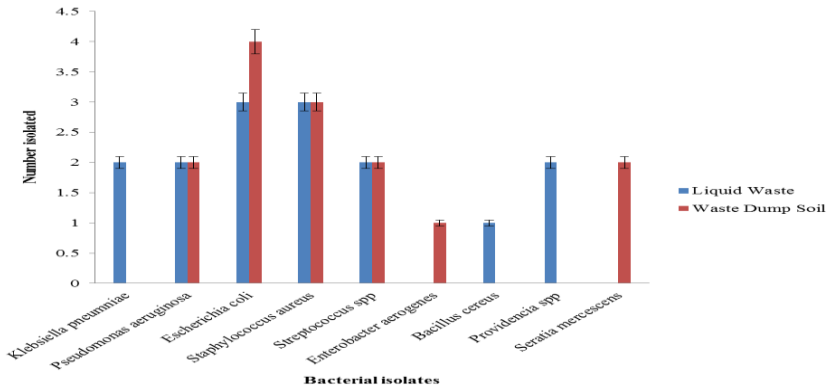

FIG. 3: Distribution of Bacterial species from liquid waste and waste dump soil from Hospital H3.

Table 4: Prevalence of bacterial isolates in Hospitals liquid Waste (%).

Hospitals	*Klebsiella pneumoniae*	*Pseudomonas aeruginosa*	*Escherichia coli*	*Staphylococcus aureus*	*Streptococcus spp*	*Enterobacter aerogenes*	*Salmonella spp*	*Providencia spp*	*Chryseobacterium spp*	Total number of isolate (%)
H1	5	6	11	8	6	2	4	3	-	45(52.9)
H2	-	2	2	7	3	1	6	2	2	25(29.4)
H3	2	2	3	3	2	1	-	2	-	15(17.6)
TOTAL (%)	7(8.2)	10(11.8)	16(18.8)	18(21.2)	11(12.9)	4(4.7)	10(11.8)	7(8.2)	2(2.4)	85(100)

KEY: = Absent

Table 5: Prevalence of bacterial isolates in hospitals waste dump soil (%).

Hospitals	*Klebsiella pneumoniae*	*Pseudomonas aeruginosa*	*Escherichia coli*	*Staphylococcus aureus*	*Streptococcus spp*	*Enterobacter aerogenes*	*Salmonella spp*	*Providencia spp*	*Chryseobacterium spp*	*Proteus spp*	*Bacillus cereus*	*Serratia mercescens*	Total number of isolate (%)
H1	3	7	9	9	5	3	-	-	-	2	-	-	38(40.4)
H2	2	4	8	11	3	2	4	2	1	3	2	-	42(44.7)
H3	-	2	4	3	2	-	-	-	-	-	1	2	14(14.9)
TOTAL (%)	5(5.3)	13(13.8)	21(22.3)	23(24.5)	10(10.6)	5(5.3)	4(4.3)	2(2.1)	1(1.1)	5(5.3)	3(3.2)	2(2.1)	94(100)

KEY: = Absent

Table 6: Frequency of occurrence of bacterial isolates in liquid waste and waste dump soil sample in 3 hospitals examined.

Bacterial isolates	Hospitals H1 (%)	H2 (%)	H3 (%)	Total (%)
Klebsiella pneumonia	8 (9.6)	2(3.0)	2(6.9)	12(6.7)
Pseudomonas aeruginosa	13(15.7)	6(9.0)	4(13.8)	23(12.8)
Escherichia coli	20(24.1)	10(15.0)	7(24.1)	37(20.7)
Staphylococcus spp	17(20.5)	18(26.9)	6(20.7)	41(22.9)
Streptococcus spp	11(13.3)	6(9.0)	4(13.8)	21(11.7)
Proteus spp	2(2.4)	3(4.5)	0(0.0)	5(2.8)
Enterobacter aerogenes	5(6.0)	3(4.5)	1(3.4)	9(5.0)
Bacillus cereus	0(0.0)	2(3.0)	1(3.4)	3(1.7)
Providencia spp	3(3.6)	4(6.0)	2(6.9)	9(5.0)
Chryseobacterium spp	0(0.0)	3(4.5)	0(0.0)	3(1.7)
Salmonella spp	4(4.8)	10(15.0)	0(0.0)	14(7.8)
Seratia mercescens	0(0.0)	0(0.0)	2(6.9)	2(1.1)
TOTAL (%)	83(46.4)	67(37.4)	29(16.2)	179(100)

Table 3 shows the biochemical characterization of the bacterial isolates form liquid waste and waste dump soil of the hospitals. It was observed that out of the 179 isolates subjected to biochemical characterization, 114 isolates were gram negative while 62 isolates were gram positive bacteria. The isolates identified were *Escherichia coli.*[37] *Klebsiella pneumoniae.*[12], *Proteus* spp.[5], *Pseudomonas aeruginosa.*[23], *Staphylococcus aureus*[41] *Streptococcus* spp.[12], *Chryseobacterium* spp.,[3] *Bacillus cereus.*,[3] *Providencia* spp.,[9] *Salmonella* spp.[14] *Enterobacter aerogenes.*[9] and *Serratia mercescens.*[2] Similar bacterial isolates were reported by Chikere *et al* (2008) as possible nosocomial organism in the hospital environment.

The distribution of bacterial isolates from liquid waste and waste dump soil from the three hospital studied is presented in Figure 1-3. The hospital with the highest number of bacterial distribution was hospital H2. *Chryseobacterium* spp. was isolated in hospital H2 only, while other bacterial isolates in this study were present in the three hospital.

Chryseobacterium spp. especially *Chryseobacterium indolegenes* is a rare pathogen in the human microflora, although it is widely distributed in nature (Chen *et al*., 2012). According to SENTRY Antimicrobial surveillance program, *Chryseobacterium* spp. represent 0.03% of the total isolates and account for 0.03% of all bacteraemia cases (Sakurada, 2008). Reported infections caused by *Chryseobacterium* spp. include ventilator –associated pneumonia and urinary trait infections, and they are often associated with a high motality rate (Abhuyar *et*

al., 2012; Souza de Souza *et al.*, 2012), nearly half of the published cases of *Chryseobacterium* spp. refers to nosocomial infections, and the vast majority of patients has underlying immunocompromising conditions (Bhuyar *et al.*, 2012). Other potential nosocomial pathogens that are often associated with nosocomial infectiosn were *Escherichia coli, staphylococcus aureus, pseudomonas aeruginosa* and *klebsiella pneumoniae. Escherichia coli* and *staphylococcus aureus* had the highest frequent of distribution in both liquid waste and waste dump soil.

The prevalence of bacterial isolates in hospital liquid waste and waste dump soil is presented in Table 4 and 5. From the result, *Staphylococcus aureu*s had the highest prevalence of occurrence in liquid effluent sample (21.2%) while *Chryseobacterium* spp had the least occurrence of 2.4%. Among the bacterial isolates from waste dump soil, *Staphylococcus aureus* and *Escherichia coli* had the highest percentage of occurrence 24.5% and 22.3% respectively, while *Chryseobacterium* spp had the least percentage of occurrence (1.1%), followed by *Providencia* spp and *Serratia mercescens* 2.1% each. The result of this study pinpointed that *Staphylococcus aureus.* was the most prevalent pathogen isolated from the hospitals liquid waste and waste dump soil while *Escherichia coli* was the most frequent gram negative bacteria isolated. Ummu *et al.*, (2013) also reported similar organism in the hospital environment with *Staphylococcus aureus* being the most prevalent pathogen.

Table 6 shows the frequency of occurrence of bacterial isolates in liquid waste and waste dump soil samples from the three hospitals examine. The result of this study clearly spelled out *Escherichia coli* 37(20.7%) was the most prevalent gram negative bacteria isolated in the hospitals liquid waste and dump soil while *Staphylococcus aureus.* 41(22.9%) was the most prevalent gram positive bacteria isolated. Similarly, the result of this study coincided with the work of Chikere *et al* (2008) which reported *Staphylococcus aureus* as the most prevalent nosocomial pathogen in a hospital environment, followed by *Escherichia coli*. Asghar and Faidah (2009) also showed that *Escherichia coli* and *Pseudomonas* spp were the main pathogen isolated in hospital environment and according to them many international studies reported that *Pseudomonas aeruginosa* was the most prevalent gram negative bacteria isolated in the hospital environment in comparison to *Escherichia coli* and other bacterial isolates. This differences observed in the frequency of occurrence of the types of bacterial isolates between hospital is as a result of the variation that occur in maintaining standard

personal hygiene's, patient's population and lack of adequate treatment given to hospital waste before it is being disposed.

CONCLUSION

Liquid waste and waste dump soil of hospitals serves as a complex ecosystem that need many intervention in order to have adequate nosocomial infection control. The result of this study revealed a wide spread of nosocomial pathogens in the hospital waste examined. The present of this pathogen might be due to poor hygiene practice in these hospitals and lack of regular surveillance system for the control of potential nosocomial bacterial pathogens. Based on this study, it is therefore recommended that hospital management should set up national intensive surveillance program that will help to curtail the distribution of potential nosocomial bacterial pathogen in the hospital liquid waste and waste dump soil.

REFERENCES

1. Apanga, S., Adda, J., Issahaku, M., Amofa, J., Mawufemor, R. A. & Bug, r S. Post-operative surgical site infection in a surgical ward of a tertiary care hospital in Northern Ghana. Int. *J. Res. Health Sci*, 2014; 2: 207-212.
2. Asgbar, A. & Faidah, H. Frequency and antimicrobial susceptibility of gram-negative bacteria isolated from 2 hospitals in makkah, Saudi, Arabia. *Saudi Medical Journal*, 2009; 30(8): 179-185.
3. Bergogne-Berezin, E. & Towner, K. J. *Acinetobacter*spp. as nosocomial pathogens: microbiological, clinical and epidemiological features. *Clin.Microbiol.*, 1996; 19: 148-165.
4. Bhuyar, G., Jain, S., Shah, H. & Mehta, V. K. Urinary tract infection by chryseobacteria indologenes. *Indian Journal of Medical Microbiology*, 2012; 30: 370 – 372.
5. CDC. Types of healthcare- associated infection. Health care-associated infections (HAIs). 2016 (online) available from: https://www.cdc.gov/HAI/infectionType.html (accessed on 8th August, 2018).
6. Chen, F. L., Wang, G. C., Teng, S. O., Ou, T. Y., Yu, F. L. & Lee, W. S. Clinical and epidemiological features of *chryseobactrium indologenes*: Analysis of 215 cases. *Journal of Microbiology Infection,* 2012; *46:* 452-432.
7. Chikere, C. B., Omoni, V. T. & Chikere, B. O. Distribution of potential nosocomial pathogens in a hospital environment. *African Journal of Biotechnology*, 2008; 7(20): 3535-3539.

8. Esposito S, Leone S Antimicrobial treatment for intensive care unit (ICU) infections including the role of the infectious diseases specialist. *Int. J. Antimicrob. Agents*, 2007; 29: 494-500.
9. Hassan, A. K., Fatima, K. B. & Riffat, M. Nosocomial infection: Epidemiology, prevention control and surveillance. *Asian Pacific Journal of Tropical Biomedicine*, 2017; 1: 1-5.
10. Mbin, E. N., Mboto, C. I. & Agbo, B. E. A review of nosocomial infection in sub-saharan African. *British Microbiology Research Journal*, 2016; *15*(1): 1-11.
11. McCraig LF, McDonald LC, Mandal S, Jernigan DB *Staphylococcus aureus*associated skin and soft tissue infections in ambulatory care. *Emerg. Infect. Dis., 2006; 12*: 1715-1723.
12. Nelson, P. R. A comparism of samples sizes for the analysis of means and analysis of variance. *Journal of Quality Technology*, 1983; *15:* 33-39.
13. Pitout D. D., Church D. L., Gregson D. B., Chow BL, McCracken M, Mulvey M, Laupland KB Molecular epidemiology of CTXM-producing *Escherichia coli* in the Calgary Health Region: emergence of CTX-M- 15-producing isolates. *Antimicrob. Agents Chemother*., 2007; *51*: 1281-1286.
14. Pitout JDD, Nordman P, Laupland KB, Poirel L (2005). Emergence of *Enterobacteriaceae*producing extended-spectrum -lactamases (ESBLs) in the community. *J. Antimicrob. Chemother.*, 2005; *56*: 52-59.
15. Prescott LM, Harley JP, Klein DA. Microbiology. 6th ed. McGraw- Hill, New York, 2005; 2042.
16. Rhomberg PR, Fritsche TR, Sader HS, Jones RN Antimicrobial susceptibility pattern comparisons among intensive care unit and general ward gram-negative isolates from meropenem yearly susceptibility test information collection program (USA). Diagno. Microbiol. *Infect. Dis*., 2006; 56: 57-62.
17. Sakurada, Z. A. (2008). Chryseobactrium indologenes. Revista chilena de infetologia, 25, 446 (in Spanish).
18. Souza de Souza, M., Silva, C., Paschoalin, E., Junior, J., Pascholin, R. & Oliveira, E. Bacteremia by chryseobactrium indolegenes in a diabetes patient undergoing ambulatory hemodialysis. Journal of Brazilian Pathological Medical Laboratory, 2012; *48:* 29 -31.
19. Ummu, K. M., Mustafa, A. I. & Zainab, M. A. Distribution of potential nosocomial pathogen isolated from environments of four selected hospitals in Sokoto, North Western Nigeria. *Journal of Microbiology and Biotechnology Research, 2013; 3*(1): 139-143.

20. Uusipaikka, E. Exact simultaneous confidence intervals for multiple comparison amongst three or four mean values. *Journal of the American Statistical Association*, 1985; *80*: 111-127.
21. Wood, S. M., Shah, S. S., Bafana, M., Ratner, A. J, Meaney, P. A, Malefho, C. S, Steenhoff, A. P. Epidemiology of methicillin resistance *Staphylococcus aureaus* bacteremia in Gaborone, Botswana. *Infect. Control Hosp*., 2009; *4:* 7-13.
22. World Health Organization (WHO)(2011). Report on the Burden of Endemic Health Care- Associated Infection Worldwide. (NLM classification: WX 167) Geneva: WHO; 1-34.
23. Zhanel, G. G., DeCorby, M., Laing, N., Weshnoweski, B., Vashisht, R., Tailor, F., Nichol, K. A., Wierzbowski, A., Baudry, P. J., Karlowsky, J. A., Lagace´-Wiens, P., Walkty, A., McCracken, M., Mulvey, M. R. & Johnson, D. J. (2008). The Canadian Antimicrobial Resistance Alliance (CARA), and Hoban, Antimicrobial-resistant pathogens in intensive care units in Canada: results of the Canadian National Intensive Care Unit (CAN-ICU) study, 2005-2006. *Antimicrob. Agents Chemother., 52*: 1430–1437.