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Prevalence of *Acinetobacter spp* isolated from clinical specimen in University of Ilorin Teaching Hospital Ilorin, Nigeria.

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Abstract

The genus Acinetobacter currently contains 34 species, the vast majority of which are not regularly implicated in causing infection. However, incidences of hospital acquired infection with Acinetobacter species are on the increase due to the rise in the number of infections caused Acinetobacter baumannii. The objective of this study was to determine the prevalence of Acinetobacter species isolated from clinical samples from University of Ilorin Teaching Hospital This study is a descriptive cross-sectional study was conducted in UITH located in the North Central Nigeria. Ten (10) strains of Acinetobacter spp. were isolated from clinical samples between February to July 2018. The isolated strains were identified using standard microbiological methods, API20NE. The Prevalence of Acinetobacter spp in this study was 6% high. Moreso, it is also similar to a work done in France, which reported 9% and about 7% in Nigeria. They are mostly multi-drug resistant. The prevalence has shown to be significantly connected with age. Majority of the patients 43(38.1%) were in the range of 31-40 years. This eventually shows that patients in the age bracket 31-60 years were more susceptible to Acinetobacter infection. The prevalence rate of Acinetobacter spp in this study is high. This study is of high significance particularly all health workers to know that there is high prevalence of Acinetobacter spp. There is need to maintain a clean and healthy environment at all times.

Keywords: Acinetobacter species, Acinetobacter baumannii, Intensive care unit, Carbapenemase enzymes, Cerebrospinal fluid, Blood agar.

Introduction

Acinetobacter spp are gram negative, coccobacillus and strict aerobes (Allen and Hartman, 2000). They play significant role in the colonization and infection of patients admitted to the hospital. In the past three decades, it emerged from organisms of questionable pathogenicity to an infectious agent of importance worldwide (Schneckenburger et al., 2003). Acinetobacter infection has long been clinically prominent in tropical countries. It has been a recurrent problem during war, natural disasters and recently it has been multi-hospital outbreak in temperate climate (Lolans et al., 2006). Acinetobacter spp is easily isolated in standard cultures but is relatively non-reactive in many biochemical tests commonly used to differentiate gram negative bacilli.

Acinetobacter can survive for more than four months in the environment, which is longer than seven days survival period of Staphylococcus aureus (Villegas and Hartstein, 2003). Because of this and afore-mentioned features, it has successfully been involved in several outbreak across the globe. In early 1980s, Acinetobacter baumanni nosocomial outbreaks were described in South Europe particularly in France, Germany, England, Netherland and Spain (Fournier et al., 2006). Data collected from 1984 to 2003 by the National Nosocomial Surveillance Infection System from several hospitals in New-York, USA shows that the prevalence of multi-drug resistant of Acinetobacter strains are more manifested in intensive care unit acquired pneumonia due to Acinetobacter baunnamii which was found to be 4% in 1986 and 7% in 2003 (Crawford et al.,

1997). This substantial rise in United States was observed to have been contributed by the injured military personnel returning from war in Iraq and Afghanistan (Iregbu *et al.*, 2002).

The activity of some third generation cephalosporins, aztreonam, imipenem, ciprofloxacin, and amikacin against isolates of Acinetobacter baumannii of various biotypes was studied in Chilean hospitals (Bello et al., 1997). Isolates exhibited a broad multi resistance against cephalosporins. Ceftazidime was the most active and cefoperazone the least active compound. Back home in Nigeria, a study conducted by Iregbu et al. (2002) clearly revealed high prevalence of Multi-drug Resistance Acinetobacter baunnamii found to be 50/58(86.2%) in Lagos University Teaching Hospital (LUTH) Lagos. Also, in a recent study conducted in the University College Hospital (UCH) Ibadan, Nwaduke et al. (2014) documented a prevalence of 11/14(79.0%) for MDR Acinetobacter baunnamii species among other species of Acinetobacter investigated.

Methodology

Study Area: This study was carried out at the Medical Microbiology Laboratory Department of the University of Ilorin Teaching Hospital, UITH is located in Ilorin which provides quality health care services to the neighboring state such as Oyo, Kogi, Niger, Osun and Ekiti states UITH has bed space of about four hundred and fifty (450) and admission rate of about twelve thousand (12,000) patients per annum. Ilorin is the capital of Kwara state.

Study Population

The study population includes patients admitted into medicals, surgical wards and intensive care unit (ICU) of University of Ilorin Teaching Hospital, Ilorin, Nigeria from February to July, 2018 whose clinical samples were sent to the Medical microbiology laboratory for routine investigation.

Study Design

This is a descriptive cross-sectional study.

Sampling Method

Purposive sampling, a non-probability method was employed for this study using all *Acinetobacter spp* isolates from clinical samples collected in a repetitive manner. All patient's data of each isolate were extracted from the laboratory register and used for this study. The age, gender and unit/wards were noted in the proforma.

Ethical Clearance

Ethical approval was obtained from the Ethical Review committee of the University of Ilorin Teaching Hospital, Ilorin (ERC No: ERC PAN/2018/06/1780).

Sample Size

Sample size was determined using the sample size formula for estimating single proportion ¹⁰.

$$N = \frac{z^2 p q}{d^2}$$

Where,

N = Minimum sample size

Z = Standard normal deviation, usually set at 1.96 which corresponds to 95% confidence level P = The best estimated prevalence of target population; 8% from a study, Odewale, Adefioye and Ojo (2016)=0.08%

Q = I - p; 1 - 0.08 = 0.92

d = minimum statistically significant difference to be measured at 0.05 precision.

Hence N=
$$\frac{1.96X0.08X0.92}{0.05X0.05} = 113$$

The minimum sample size is 113. Therefore, 113 clinical samples were collected for this study. However, they were some multiple samples collected from more than one patient which made it 150 clinical samples.

Specimen collection

Various clinical samples were collected through aseptic technique by medical practitioners to prevent contamination. For optimal results, specimen was collected to clean sterile, wide bore containers. The clinical samples include urine, pus, blood, fluid (pleural, pericardial, synovial and peritoneal), wound swab, cerebrospinal fluid (CSF), sputum, central venous pressure (CVP), catheter tip, stool, throat swab, ear swab, nasal swab, from the tip of Foley catheter, tissue (wound and soft tissues), and bronchial washing samples. They were transported immediately to the Medical Microbiology Laboratory of the University of Ilorin Teaching Hospital for microbiological analysis.

Characterization of bacterial isolates

Colonial morphology of suspected colonies from both blood agar and MacConkey agar plate was characterized based on their appearances. Gram staining was done on the isolates and biochemical testing such as oxidase, catalase and triple sugar iron was further done on the gram negative, coccobacillus isolates for characterization. The identified *Acinetobacter spp*. were further confirmed by API20NE multi-test system (Falagas and Bliziotis, 2007). These tests were used according to manufacturers' protocol for Enterobacteriaceae and non-enteric bacteria.

Statistical analysis

Data obtained was fed into a computer and analyses were done using statistical package for the social sciences (SPSS) version 20.0. Data was presented in frequency tables, charts and prose. Cross tabulation of important table was done.

Results

Demography of study population: During the period of this study blood, urine, cerebrospinal fluid and wound swab of 150 samples were collected from 113 patients over a period of four months.

Age groups	Number	Male	Female	
(In years)	N (%)	N (%)	N (%)	
<10	9(8.0)	5(55.6)	4(44.4)	
11-20	6(5.3)	2(33.3)	4(66.7)	
21-30	23(20.4)	14(60.9)	9(39.1)	
31-40	43(38.1)	30(69.8)	13(30.2)	
41-50	22(19.5)	12(54.5)	10(45.5)	
51-60	6(5.3)	3(50.0)	3(50.0)	
>60	4(3.5)	1(25.0)	3(75.0)	
Total	113	67	46	

Table 1: Age range and gender distribution of Acinetobacter spp

The patient age ranges from 2 to 85 years. Majority of the patients 43 (38.1%) were in the age range of 31 to 40 years while the least 4 (3.5%) were in age range> 60 years. There were 67 males and 46 females with M: F of 1.45:1 (Table 1).

Frequency of Acinetobacter spp in hospital wards.

Forty-four of the patients were admitted via A and E with least from Intensive care unit (ICU) 5(4.4%) and they were majorly males respectively. Table 2 shows the sources of admission of the subjects.

Clinical wards	Number (%)	Male	Female
		N (%)	N (%)
Accident and	44(38.9)	30(68.2)	14(31.8)
Emergency			
Surgical ward	26(23.0)	15(57.7)	11(42.3)
Medical ward	21(18.6)	10(47.6)	11(52.4)
Intensive care unit	5(4.4)	3(60.0)	2(40.0)
(ICU)			
Paedatric ward	17(15.0)	7(41.2)	10(58.8)
Total	13(100)	67	46

Table 2: Sources of Admission of the Subjects

Frequency of Isolates in Clinical Samples

About 150 samples were collected; urine 72(48.0%) followed by blood culture 48(32.0%). A total number of 10 *Acinetobacter spp* were isolated from wound swab 6(60%) followed by blood culture 4(40%). Table 3 shows the distribution of *Acinetobacter spp* in various clinical samples.

Samples	Number of Acinetobacter	Number of <i>Acinetobacter spp</i>	
	spp		
	Samples (%)	Isolated (%)	
Urine	72(48.0)	0(0)	
Blood culture	48(32.0)	4(40)	
Wound swab	23(15.3)	6(60)	
CSF	7(4.70)	0(0)	
Total	150(100)	10(100)	

Table 3: Distribution of Acinetobacter spp in various clinical samples.

Discussion

The high rate of Acinetobacter infection being globally reported is a major concern not only to health-care provider but the population at large. The study was carried out to determine the prevalence of Acinetobacter species in clinical specimens. Acinetobacter spp is usually a healthcare-associated pathogen affecting the very ill-patients and is emerging as a cause of Nosocomial global outbreak (Fournier and Richet, 2006). Prevalence of Acinetobacter species in this study was 6.6%. The prevalence obtained in this study was similar to a work done which was carried out among the 4232 samples received at the laboratory (9.2%) were positive for A. baumannii in Casablanca (El Kettani et al., 2017) and Nabil et al. (2001) who reported 8.8%. Our finding is however at variance with findings of Iregbu et al. (2002).

Age has been discussed as a major factor associated with the acquisition of Acinetobacter. Although the sample size used in this study was smaller, patients in the age bracket 31-60 years were more susceptible to Acinetobacter infections. This is similar to the report of Mera *et al.* (2010). People of the working-class age have a greater risk of having Acinetobacter infections than the elderly people. This may be attributed to the fact that this category of people is constantly shifting their location due to their job placements and as such are more prone to diseases and accidental trauma.

In this study, seventy percent (70%) of the affected patients were male. Similar result was reported in a study conducted in a tertiary care hospital in Osogbo, Nigeria in which 63.6% of

the patients were male (Odewale *et al.*, 2016). Also, in another study conducted in tertiary care hospital in South India in which 58% of the patients were male.

Summary/Conclusion

The prevalence rate of *Acinetobacter spp* in this study is high. *Acinetobacter spp* are known to be the most persistent gram-negative bacilli on the hands of hospital staff and so can be transferred from one patient to another. This may be the reason why 6(60%) of the isolated *Acinetobacter spp* were isolated from one wound swab of patients in this study. They are the only group of Gram-negative bacteria that may be natural residents of human skin, with carriage rates of 42.5% in healthy individuals and as high as 75% in hospitalized patients. As a result of this, there should be proper and adequate health condition among people to maintain a clean and healthy environment at all times.

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