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Antibiotic Sensitivity Pattern of Bacteria from Selected Hospitals in Akungba Akoko, Ondo State, Southwest Nigeria

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ABSTRACT

In recent times, there has been an astronomical increase in the cases of bacterial resistance to commonly used antibiotics by medical personnel in Akungba Akoko, Ondo State, Nigeria. It is just right for a study of this nature to be carried out in order to provide the important microbiological pieces of information to help medical practitioners to tackle this menace. Bacteria were isolated from inanimate object in hospitals in Ondo State which include: General Hospital, Iwaro-Oka; Reva Clinic, Akungba Akoko; University Health Centre, Akungba Akoko and Comprehensive Health Centre, Akungba Akoko. In this study seven organism were isolated, *Staphylococcus aureus*, *Streptococcus* sp., *Pseudomonas* sp., *Aeruginosa* sp., *Klebsiella* sp., *Bacillus* sp. and *Escherichia coli*. All these organisms were identified based on their morphology and biochemical characteristics. The incidence of *Staphylococcus aureus* (34.8%) was found to be dominant out of the gram-positive bacteria isolated. The incidence of *E. coli* (44.4%) was found dominant out of the gram-negative bacteria isolated. Of the 70 isolates recovered, 43 (61.4%) were gram-positive while 27 (38.6%) were gram-negative. Multiple resistances were recorded against antibiotics used in this study. This study reveals that *staphylococci*, *streptococci*, *bacillus*, *pseudomonas* and *enterbacteriaceae* are predominant organism associated with inanimate object in hospitals. Effective hospital control and aseptic precaution will help reduce the incidence.

Key words: Bacteria, hospital, antibiotics, inanimate objects, antimicrobial

INTRODUCTION

Antibiotics are antimicrobial agent produced by microorganism; they could be better described as any chemical of natural origin (i.e., from any type of cell) which can prevent the growth of other types of cell (Todar, 2009). It has been found that soil dwelling organisms are the most predominant amongst organisms that produce antibiotics as secondary metabolites. Most of these organisms form spores or other forms of dominant cells in the cell there are as many as seven enzymes (depending on the bacteria sp.) which bind peptidoglycan unit via their D-alanyl-D-alanine residues. Enzyme binding β -lactam antibiotic are known as penicillin-binding protein (Adebayo-Tayo *et al.*, 2012; Colodner *et al.*, 2007; Depardieu *et al.*, 2007).

Antibiotics are either bacteriostatic or bacteriocidal based on the mechanism of selective toxicity as the human cells are left alone. They may act on cell-wall, ribosome, cell membrane. Those that act on the 30 sec ribosomes are nitrofurans, gentamycin, streptomycin and neomycin while

antibiotics that act on the 50S protein of the ribosome include, *chloramphenicol*, *lincomycin*, *clindamycin*, *erythromycin* and the inhibitor of metabolic pathways via competitive antagonism (Doublet *et al.*, 2003; Cheesbrough, 2006).

Antibiotic sensitivity is a term used to describe susceptibility of bacteria to antimicrobial. Susceptibility are usually carried out in vitro to determine the efficacy of an antibiotic to determine its suitability for treatment of a particular ailment. Testing for the antimicrobial sensitivity is often done by the Kirby-Bauer disc diffusion technique (Bauer *et al.*, 1966). A circular clear ring, devoid of growth around the antibiotic disc on the petri dish indicates sensitivity by the organism under test (Halawani and Shohayeb, 2008; Doublet *et al.*, 2003). Antibiotic resistance may occur through natural selection or mutation (Hsu *et al.*, 2007).

The study is aimed at determining the incidence of gram-positive and gram-negative bacteria isolate and the antibiotic sensitivity pattern of the bacterial isolated.

MATERIALS AND METHODS

Sample collection: The collection sample was from inanimate hospital source, sterile swab sticks were used to swab the surface of inanimate object such as bed hold, medical trolleys, door knobs, drip stands, wash hand basin and water closet inlets. The swab sticks were moistened with sterile peptone water and were transported to the laboratory within an hour of sampling with proper labelling with information such as area sample serial number and hospital name etc.

Preparation of culture media: The culture media used was solids which were used in isolation and subsequent identification of the organism. The media were in dehydrated form obtained from nutrient agar, Marconky, biochemical 70148; lab (E05 m methylene blue mannitol salt agar).

Isolation, maintenance and preservation of cultures: The sample were cultured using the streak plate method media such as nutrient agar, incubated at 37°C overnight and the isolates were collected as growth on these media. The cultures were maintained in slant of nutrient agar. The slants were kept in the refrigerator at 4°C until when needed in further experiments. The characterization and identification of various organisms were done by carrying out biochemical characterization, catalase test, motility test, copulas test, methyl red voges-prostcaur test (the methyl red test is used to detect acid production while the voges-proslcaur test is used to show acetyl production). Sugar fermentation and antibiotic sensitivity tests were also carried out.

RESULTS

Isolates identification was based on their cultural morphology and biochemical characteristics as shown in Table 1. The gram-positive organisms were smooth, opaque, irregular, entire, raised whitish, yellowish circular representing the morphology characteristics of *staphylococcus*, *streptococcus* and *Bacillus*. These gram positive do not show growth characteristics on Eosin Methylene Blue agar (EMB) but show growth characteristics on nutrient agar. However, gram negative stained scored negative to gram reaction showing different morphological characteristics as smooth, opaque, whitish round translucent, greenish, convex short rods and curved rod representing the genera of *Klebsiella*, *Pseudomonas* and *Escherichia coli* they show growth characteristics on EMB agar.

Out of the total gram positive organism 32 (74.42%) were smooth, convex, opaque, yellow with entire edges, they appeared as cocci in irregular clusters and chain 11 (25.58%) isolates were rough opaque with irregular edges and long rods with rounded ends. Of the 27 (38.6%) gram negative

Table 1: Characterization of bacteria isolate cultured from hospital inanimate object

Isolates	Cultural morphology			Gram			Growth										Identification
	on nutrient agar	Shape	Gram stain	Motility	Catalase	Coagulase	Indole	on EMB	MA	MR	VP	Glucose	Sucrose	lactase	Maltase	Galactase	
A	Green, smooth flat dull	Curved rod	-	+	+	-	+	Translucent colourless	Mucoid purple	-	-	A	-	-	-	-	<i>Pseudomonas</i> sp.
B	Whitish, smooth entire, opaque	Cocci in chain	+	-	+	-	-	NG	NG	-	-	AG	A	A	A	A	<i>Streptococcus</i> sp.
C	Creamy smooth, opaque, entire	Short rod	-	+	+	-	+	Metallic sheen	Pinkish	+	+	AG	A	AG	AG	A	<i>E. coli</i>
D	Whitish, entire, smooth, flat	Long rod	-	+	+	-	-	Mucoid pinkish	NG	+	+	AG	A	A	A	A	<i>Bacillus</i> sp.
E	Opaque, entire, convex, grayfish white	Rod	-	-	+	NG	-	Mucoid brown	Red	-	-	AG	AG	AG	AG	A	<i>Klebsiella</i> sp.
F	Golden yellow, glossy raised	Cocci in cluster	+	-	+	+	-	NG	NG	+	+	A	A	-	A	A	<i>Staphylococcus aureus</i>
G	Whitish, smooth, entire, opaque	Cocci in cluster	+	-	+	-	-	NG	NG	+	+	A	A	-	A	A	<i>Staphylococcus</i> sp.

NG: No growth, +: Positive, -: Negative, A: Acid, Ag: Acid and gas

Table 2: Bacteria isolates from hospital inanimate object

Isolate	Incidence	%
<i>Pseudomonas</i> sp.	9	12.9
<i>Streptococcus</i> sp.	11	15.7
<i>Escherichia coli</i>	12	17.1
<i>Bacillus</i> sp.	10	14.3
<i>Klebsiella</i> sp.	7	10.0
<i>Staphylococcus aureus</i>	15	21.4
<i>Staphylococcus</i> sp.	6	08.6
Total	70	100.0

Table 3: Incidence of gram positive bacterial isolated from hospital inanimate object

Isolate	Incidence	%
<i>Staphylococcus aureus</i>	15	34.8
<i>Staphylococcus</i> sp.	6	14.0
<i>Staphylococcus</i> sp.	6	25.6
<i>Bacillus</i> sp.	10	25.6

Table 4: Incidence of gram negative bacterial isolated from hospital inanimate object

Isolate	Incidence	%
<i>Pseudomonas</i> sp.	9	33.3
<i>Escherichia coli</i>	11	40.7
<i>Klebsiella</i> sp.	7	25.9

Table 5: Antibiotic resistance of bacteria isolate from inanimate object

Isolates	Identification	Antibiotic to which isolate was resistance
A	<i>Pseudomonas</i> sp.	AUG, CEX, COT, AMX, PFX
B	<i>Streptococcus</i> sp.	NIT, COT, CPX, PFX
C	<i>Escherichia coli</i>	AUG, TET, PEX
D	<i>Bacillus</i> sp.	AUG, NIT, TET, PEX
E	<i>Klebsiella</i> sp.	CEX, CPX, NIT, TET, PEX
F	<i>Staphylococcus aureus</i>	AUG, TET
G	<i>Staphylococcus</i> sp.	AUG, OLF, PFX

AUG: Augmentin, GEN: Gentamicin, CHL: Chloramphenicol, AMX: Amoxicillin, TET: Tetracycline, COT: Co-Trimoxazole, PFX: Pefloxacin, OLF: Ofloxacin, NIT: Nitromicin, CPX: Ciprofloxacin, CEX: Cephalexin, PEX: Trimethoprim (Trimpex)

isolates 14 (51.85%) appeared smooth convex and opaque with entire edges. They were short rods demonstrating a greenish metallic sheen, pinkish, large mucoid growth on EMB agar. The biochemical reactions of the bacterial isolates encountered in this study were also studied. Result of carbohydrate fermentation tests show that out of 70 isolates tested all reduced glucose to acid. Antibiotics resistance pattern of bacterial isolate, a total of 70 isolated with the various antibiotics enumerated in materials and all isolates were resistant to one or more antibiotics.

Table 2 showed bacteria isolates from hospital inanimate object in which *Staphylococcus aureus* was found to be 21.4%, *Escherichia coli* was 17.5% and *Streptococcus* sp. was 15.7%.

Table 3 showed the incidence of gram positive bacterial isolated from hospital inanimate object with *Staphylococcus aureus* having the highest incidence of 34.8%. Whereas, Table 4 showed the incidence of gram negative bacterial isolated from hospital inanimate object with *Escherichia coli* having the highest incidence of 40.7%.

Table 5 showed the antibiotic resistance of bacteria isolate from inanimate object in which five the bacterial isolates are resistant to Augmentin (AUG) and four are resistant to Tetracycline (TET).

DISCUSSION

According to Madigan *et al.* (2000) a hospital environment may not be a place where people get well but may also be a place where sick people get sicker. Infections in hospital environment are as result of the following factors: Microorganism on hospital inanimate object, the compromised immune status of patients and the transmission chain of infections in hospitals.

The present study elucidate that certain pathogens were isolated from hospital inanimate objects. The result shows that the frequency of gram positive is higher than the gram negative as shown in Table 3 and 4. The observation of Cowan *et al.* (1960) shows that *Staphylococcus* was in consonance with the isolation of *Staphylococci* and *Streptococci* in the study however, the isolation of *E. coli* and *Klebsiella* sp. in this study is not apparently clear but could be associated with exposure to colonized patients. This present study reported that bacteria isolated from hospital object are mainly gram positive (Table 3).

The alarming frequency with which microorganism on the hospital inanimate objects acquired resistance to antibiotics particularly by the mechanism of transmissible drug resistance and the fact that the antibiotics to which they remain sensitive are often highly toxic, it then means that concerted effort must be made to correct this in other to avoid nosocomial disaster in this area.

CONCLUSION

This study shows that gram positive bacteria particularly *Staphylococcus aureus*, *Staphylococcus* sp., *Bacillus* sp. and *Streptococci* sp. were present on hospital objects and gram negative bacteria isolate, the pathogens isolated particularly *pseudomonas* sp., *E. coli* and *Klebsiella* sp. can lead to serious health problem if not curb, education and awareness to paramedical staff, on simple hospital hygiene such as hand-washing using soap, periodic disinfection of hospital ward can reduce the incidence of pathogen in hospitals.

Antibiotics susceptibility tests indicate the presence of highly resistant bacteria in the hospitals sampled (Albrich *et al.*, 2004) and it may serve as a risk factor for the treatment of patients colonized with multiple-resistant strains of the bacteria with commonly used antibiotics.

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