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ACTIVITY EVALUATION OF COMMONLY USED PENICILLINS ON SOME CLINICAL BACTERIAL ISOLATE

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ABSTRACT
Antibiotic resistance is one of the greatest treats to modern health and we face a feature without cures for infection if antibiotics are not used responsibly. Penicillin is one of these antibiotics which microorganism is coming resistant to. To resolve the problems of antibiotics resistance the present thesis was an effort to study the activity of commonly used penicillin on some clinical bacterial isolate. The commonly used penicillins include ampicillin, amoxicillin, oxacillin, cloxacillin and augumentin. While the clinical bacterial isolate include Escherichia coli, staphylococcus aureus, klebsiella, proteus, pseudomonas aeruginosa, streptococcus pneumonia etc. The susceptibility of the test organism to test antibiotics was determined, antimicrobial strength and spectrum of the penicillin antibiotics on the bacterial isolate was determine and also the pattern of in-vitro antimicrobial sensitivity of the test antibiotic on susceptible isolate was determine. The test isolate collected were confirmed using biochemical test and then inoculums of these organism was prepared and standardized using marcfarland, disc diffusion technique was used for the bioassay. Even though more than 50% of the organism show resistance to penicillin, the current result indicate that augumentin has the broadest spectrum of activity on both Gram positive and Gram negative organism (45.45%), pseudomonas aeruginosa emerges to be the most resistant organism (only 13.84% are sensitive to the test antibiotic), While staphylococcus aureus was the most sensitive organism. Antibiotic need to be used when it becomes necessary and there is need for proper usage according to the recommended dose.

KEYWORD: penicillin, microorganism, inoculums, resistance and sensitivity.

1.0 INTRODUCTION

The in-vitro evaluation of antimicrobial activity of an agent means to assess its antimicrobial activity outside a living system.[1] In-vitro evaluation is a biological phenomena made to occur outside a living body, traditionally in a test-tube in the laboratory.[2] Thus, in vitro evaluation is very important because it can be used to predict the *in vivo* (inside the body) activity of the given antimicrobial agent.[3] Technically, antibiotics are only those substances that are produced by one micro-organism that kills or prevents the growth of another micro-organism.[4] In today's common usage, the term antibiotic is used to refer to almost any substance that attempt to rid the body of bacterial infections.[5] Antimicrobial includes not just antibiotics, but synthetically formed compounds as well.[6] Penicillin are among the antibiotics that have been used in the treatment towards bacteria invasion.[7] Penicillin was discovered by Alexander Fleming in September 1928 while he was working at St. Mary's Hospital London. He left for holidays and left the culture of the microbe at the window of his lab. After he came back from the holidays, he found an unusual phenomenon of the culture of microbe that he had left. It was found that the absence of fully developed colonies of a common microbe, Staphylococcus aureus and a round a large colony of a common mould, Penicillium notatum. The discovery of his findings led to the use of penicillin as an antibiotic in the present days.[8] Penicillin is one of the broad spectrum antibiotic consisting of an agent that contain the β-lactam ring in their molecular structure and it was the first antibiotics to be use clinically since 1941.[9] β-lactam work by inhibiting cell wall biosynthesis in the bacteria and are the most widely use group of antibiotics.[10] Bacterial cell wall contain peptidoglycan layer and this cell wall is a rigid outer layer unique to bacterial species. And it completely surrounded the cytoplasmic membrane maintaining cell shape, rigidity and prevents cell lyses from high osmotic pressure.[11] The cell wall composes of complex cross linked polymer of polysaccharide and peptide like peptidoglycan. The polysaccharide containing altenating amino sugars N-

acetyl glucosamine and N-acetyl muramic acid in which 5 amino acid peptide linked N-acetyl muramic acid sugar, which the peptide terminate in D-alanyl D-analine.[12] Penicillin binding protein is an enzymes that remove the terminal alanine in the process of forming cross linked with a nearby peptide.[13] The β-lactams inhibit the final transpeptidase forming covalent bond with penicillin binding protein that has transpeptidase and carboxypeptidase activities thus preventing formation of the cross linked.[14] The final bactericidal action is the inactivation of inhibitors of autolytic enzymes in the cell wall which leads to the lysis of the bacterial cell.[15] But the most widespread cause of resistance to β-lactam antibiotics like penicillins is production of enzymes called β-lactam. β-lactamase are family of enzymes produce many Gram positive and Gram negative bacteria that inactivate β-lactam antibiotics by opening the β-lactam ring.[16] This enzyme is responsible for many failure of antimicrobial therapy by hydrolysis of β-lactam ring of these antibiotics.[17]

The aim of this research is to evaluate activity of commonly used penicillin on some clinical bacterial isolate and this can be achieve via the following objectives.

- To determine the susceptibility of the test organism to test antibiotics.
- To determine antimicrobial strength and spectrum of the given antibiotic.
- Also to determine the in-vitro pattern of antimicrobial sensitivity of the antibiotic on susceptible isolate.

2.0 MATERIALS AND METHOD

2.1 Sample Collection

2.1.1 Collection of drugs Sample

Fixed brands of commonly used penicillins; ampicillin and amoxicllin capsules were purchased from registered pharmacies within Kano metropolis after surveying 10 shops. These were transported to the laboratory for labeling and storage. The pharmaceutical details of these drugs (Brand name, production date, expiring date, NAFDAC Reg. no e.t.c) were all noted and tabulated. Also commercially prepared sensitivity discs for oxacillin, cloxacillin and augumentin were purchased. Pharmaceutical details of these were noted and tabulated; they were stored at 4°C.

2.1.2 Collection of test Organisms

Clinical bacterial isolates were collected from microbiology department of Aminu Kano teaching hospital, located in Kano state northwestern Nigeria. These include: *Escherichia coli, staphylococcus aureus, salmonella Spp, pseudomonas aeruginosa, shigella spp, enterococcus faecalis, neisserria gonorrhoeae, streptococcus pneumoniae, proteus spp.* They were isolated from various samples which include urine, stool, urogenital, wound etc.

2.2 Sample Size

A total of 200 clinical bacterial isolated were collected for this study.

2.3 Identification of the Test Organisms

2.3.1 Gram Staining

Using test organism a smear was made on a clean glass slide. The smear was allowed to air dry and passed three times over the flame with the smear side upper most to heat fix the preparation to the slide. The smear was stained using Gram staining technique and examined microscopically under x100 objectives, which Gram positive and Gram negative isolate were identified.[18]

2.3.2 Biochemical identification

The bacterial isolates collected were confirmed using the following biochemical tests, coagulase, catalase, motility, urease, indole and triple sugar iron test according to the procedures described by.[18]

Catalase Test

Catalase production by the test organism was detected by picking the colony with the edge of clean microscopic slide and immersing it on drop of hydrogen peroxide contained on a center of a clean microscopic slide.

Staphylococcal specie shows active bubbling while non staphylococcal specie does not show bubbling.

Coagulase Test

A drop of distilled water was placed on each end of slide; a colony of the test organism was emulsified on each of the drops to make two thick suspensions. A loop full of plasma was added to one of the suspensions and mixed gently. No plasma was added to the second suspension as a control.

Staphylococcal aureus shows clumping within seconds while other staphylococci do not clump within seconds.

Motility Test

The test organism was placed on a drop of normal saline contained in a center of clean microscopic slide. The preparation was covered with a cover slip and examined microscopically for motile organisms using X10 and X40 objectives.

Urease Test

The test organism was grown on a medium containing urea and indicator phenol red for 24hours urease positive organism show pink coloration among which are *proteus* spp and *klebseilla* spp.

Citrate Utilization Test

A sterile wire loop is used to inoculate the citrate medium with a saline suspension of the test organism and then the butt was stabbed. This was incubated at 35°c for 24 hrs. Enteric bacteria show blue coloration on the media after incubation.

Indole Test
The test organism was inoculated in a bijou bottle containing 3ml of sterile peptone water and incubated at 37^0c for 24hours. 0.5ml of kovac's reagent was added and shacked gently after incubation. A red surface layer is produced by indole positive organisms such as *E. coli*.

Triple Sugar Iron test
Triple sugar iron agar slant was inoculated with the test organism. Covered with a cotton plug to allow aeration. After incubation for 24 hours at 37°c. Sugar fermentation by the test organism was observed as follows;
(No sugar fermentation); Red slant (alkaline) & red butt (alkaline) ……. *pseudomonas aeruginosa*.

(Glucose fermentation); Red slant (alkaline) and yellow butt with small amount of acid released.....*salmonella spp and shigella spp.*

(Lactose and sucrose fermentation); Yellow slant (acid) and a yellow butt (acid) gas may appear at the bottom…….. *E. coli* and *klebsiella.*

Oxidase Test
A clean glass rode was used to emulsify a colony of the test organism on a piece of filter paper in a clean petridish containing 3 drops of freshly prepared oxidase reagent. Oxidase positive organism show blue or purple coloration e.g. *Pseudomonas aeruginosa* & Neisseria gonorrhea.

2.4 Preservation of the test organisms
Nutrient agar slants were prepared as instructed by the manufacturer, autoclaved at 121^0C for 15 minutes. These were allowed to dry in slanted position and stored at 4^0C. The test organism was inoculated on the surface of the slant using a sterile wire loop. This was stored at 4^0C.

2.5 Preparation of Sensitivity Disc
Discs of about 6.25mm diameter from whatman No. 1 filter paper were punched with the aid of paper puncher. Batches of 50 discs were dispensed in a clean screw capped bottles (bijou bottles) and sterilized by dry heating in hot air oven at 140°C for 60 minutes.[18]

2.5.1 Preparation of Sensitivity Discs for Ampicillin Drugs
Using the formula

$$W = \frac{10^3 \times C \times V}{P}$$

Where P = potency of the drug expressed in mg/g
V= volume of the solvent in milliliter (ml) i.e. DMSO
C = concentration of the stock solution in mg/L
W = Wight of the drug powder in mg to be dissolved in volume V
Conversion factor: 1mg = 1000µg
Discs of 25µg/disc potency were prepared for Ampicillin drugs as follows.
P = 250mgg $^{-1}$
W = 0.5g
V = 0.5ml

$$C = \frac{WP}{10^3 x} = \frac{0.5g \times 250mmg^{-1}}{1000 \times 0.5ml} \quad \frac{125mg}{500ml}$$

$$= 0.25mgml^{-1} \qquad = 25 \times 10^1 \mu gml^{-1}$$

Therefore, a sock solution of 25 × 10^1 µgml^{-1} was prepared by dispensing 0.5g of the drug powder into 0.5ml of DMSO. A final solution of 25 µgml^{-1} was achieved by transferring 1ml from the stock solution into 9ml of DMSO in a test tube, 1ml from the resulting solution was transferred into a bijou bottles containing 100 sterilized paper discs. So that as the whole volume is absorbed each disc is assumed to absorb 0.01ml this is represented diagrammatically below.

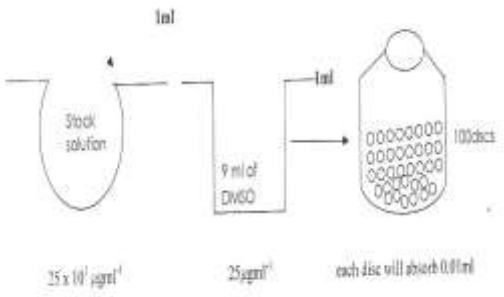

Fig 8: preparation of sensitivity discs for Ampicillin.

2.5.2 Preparation of Sensitivity Discs for Amoxicillin
Discs of 25 µg/disc potency were prepared for amoxicillin drugs as follows:
W = 0.5g
P = 500mgg $^{-1}$
V = 1ml

$$c = \frac{WP}{10^3 \times V} = \frac{0.5g \times 500mgg^{-1}}{10^3} \quad \frac{250}{10^3 ml}$$

= 0.25mgml^{-1} = 25x10^1 µgml^{-1}.

Therefore a stock solution of 25x10^1 µgml^{-1} was prepared by dispensing 0.5g of the powder into 1ml of DMSO, I ml from the stock solution was transferred into 9ml of DMSO, 1ml from the resulting solution is transferred into 9ml of DMSO, also 1ml from the resulting solution was transferred into 9ml of DMSO and a solution of 25µgml^{-1} was achieved. 1ml from this solution was transferred into a bijou bottle containing hundred sterilized filter paper discs. This was finally stored at 4°C.

2.6 Preparation of Inoculums
Subcultures
Subcultures were made by carefully picking one colony using sterilize inoculation loop, this was inoculated on the surface of nutrient agar a confluent bacterial growth

appears on the medium. Such pure cultures were used for identification and antibiotic sensitivity tests. 3 to 5 colonies of the organism to be tested was picked up by using a sterile wire loop and emulsified in 3ml of sterile saline in which density of the bacterial suspension adjusted by matching it with turbidity standard of barium chloride (McFarland's).[18]

2.7 Bioassay Procedure

Sensitivity agar plates were dried in the hot air oven until no visible excess moisture is observed on the surface. A sterile swab stick was dipped in the suspension of the prepared inoculums and then excess fluid was removed by pressing and rotating it against the side of the tube above the level of the suspension. The swab was streaked evenly over the surface of the medium in three directions, whilst rotating the plate to ensure even distribution.

With the petridish lid in place, the surface of the agar was allowed to dry for 3-5 minutes. A sterile forceps was used mounted in a holder, to place the appropriate antimicrobial discs on the surface of the inoculated plates.

Plates were incubated at 37°C for 24 hours and then examined for the presence of zones inhibition of bacterial growth around antibiotic disc. Zone of inhibition was measured by a ruler on the underside of the plate.[18]

3.0 RESULT

Table 1: Overall Antibiotic Sensitivity Profile of Bacteria Isolates to Commonly Used Penicillin at Aminu Kano Teaching Hospital (AKTH) Kano Nigeria.

S/N	Antibiotic	Sensitivity of Isolates																					
		E. coli N=30		*Klebs. Spp N=30*		*Proteus Spp N=30*		*Staph. Aureus N=30*		*Pseu. Aeru. N=30*		*Strep. Pneomo N=10*		*N. gonorr. N=10*		*Salmonella N=5*		*Shigella SPP N=5*		*E. aerogenes N=10*		*E. Faecalis N=10*	
		No Sens	% Sens	No Sens	% Sens	No Sens	% Sens	No Sens	% Sens	No Sens	% Sens	No Sens	% Sens	No Sens	% Sens	No Sens	% Sens	No Sens	% Sens	No Sens	% Sens	No Sens	% Sens
1.	Augumentin	5	16.67	18	60.00	18	60.00	17	56.67	7	23.33	4	40.00	5	50.0	2	40.0	2	40.0	4	40.00	4	40.0
2.	Oxacillin	4	13.33	5	16.67	5	16.67	18	60.00	3	10.00	6	60.00	2	20.0	2	40.0	1	20.0	2	20.00	5	50.0
3.	Cloxacillin	10	33.33	7	23.33	13	43.33	16	53.33	3	10.00	4	40.00	2	20.0	2	40.0	1	20.0	3	30.00	4	40.0
4.	Ampicillin A	12	40.00	8	26.67	10	33.33	15	50.00	3	10.00	5	50.00	4	40.0	1	20.0	2	40.0	3	30.00	5	50.0
5.	Ampicillin B	11	36.67	7	23.33	9	30.00	14	46.67	3	10.00	5	50.00	4	40.0	1	20.0	2	40.0	2	20.00	5	50.0
6.	Ampicillin C	11	36.67	8	26.67	9	30.00	13	43.33	2	6.67	3	30.00	4	40.0	2	40.0	2	40.0	2	20.00	3	30.0
7.	Ampicillin D	12	40.00	8	26.67	10	33.33	15	50.00	3	10.00	5	50.00	4	40.0	1	20.0	2	40.0	3	30.00	5	50.0
8.	Ampicillin E	13	43.33	9	30.00	11	36.67	16	53.33	4	13.33	6	60.00	4	40.0	2	40.0	3	60.0	4	40.00	6	60.0
9.	Amoxicillin A	11	36.67	10	33.33	15	50.00	13	43.33	5	16.67	4	40.00	3	30.0	2	40.0	2	40.0	6	60.00	6	60.0
10.	Amoxicillin B	10	33.33	11	36.67	15	50.00	13	43.33	5	16.67	4	40.00	3	30.0	2	40.0	2	40.0	4	40.00	6	60.0
11.	Amoxicillin C	9	30.00	9	30.00	14	46.67	12	40.00	4	13.33	3	30.00	2	20.0	1	20.0	1	20.0	5	50.00	5	50.0
12.	Amoxicillin D	11	36.67	9	30.00	13	43.33	12	40.00	5	16.67	5	50.00	2	20.0	2	40.0	1	20.0	5	50.00	4	40.0
13.	Amoxicillin E	12	40.00	9	30.00	15	50.00	14	46.67	5	16.67	4	40.00	5	50.0	3	60.0	3	60.0	3	30.00	5	50.0
14.	Total		33.59		30.25		40.26		48.20		13.33		44.62		33.84		35.38		36.92		35.38		48.46

Table 2: Overall Sensitivity Profile of Clinical Bacterial Isolates to Commonly Used Penicillins at Aminu kano teaching hospital (AKTH) kano Nigeria.

S/N	Test Isolate	(%) Sensitive to Commonly use Penicillins
1.	*Escherichia coli*	33.59
2.	*Klebsiella Spp*	30.25
3.	*Proteus Spp*	40.26
4.	*Staphylococcus aureus*	48.20
5.	*Pseudomonas aeruginosa*	13.33
6.	*Streptococcus pneumoniae*	44.62
7.	*Neisseria gonorrhoeae*	33.84
8.	*Salmonella Spp*	35.38
9.	*Shigella Spp*	36.92
10.	*Enterobacter aerogenes*	35.38
11.	*Enterococcus faecalis*	48.46

Table 3: Overall Sensitivity Profile of Clinical Bacterial Isolates to Various Penicillins Tested.

S/N	Antibiotic	(%) of Organisms Sensitive
1.	Augumentin	43.00
2.	Oxacillin	26.50
3.	Cloxacillin	32.50
4.	Ampicillin A	34.00
5.	Ampicillin B	31.50
6.	Ampicillin C	29.50
7.	Ampicillin D	34.00
8.	Ampicillin E	39.00
9.	Amoxycillin A	38.50
10.	Amoxycillin B	37.50
11.	Amoxycillin C	32.50
12.	Amoxycillin D	34.50
13.	Amoxycillin E	39.00

4.0 DISCUSSION

It is well known that microorganisms are constantly evolving, and the existing microorganisms are constantly generating means of surviving various habitats and metabolize the substrate on that environment for growth and reproduction. It is for the reason that this study was conducted, various bacterial isolates were subjected to media in which it contains agents that does not allow them to grow. These agents are the penicillin antibiotics and it is activity on the clinical bacterial isolates was studied.

The study reveals that the most common gram negative rod *Escherichia coli* is 43.33% sensitive to Ampicillin, this is not surprising because Ampicillin was reported susceptible to β-lactamase enzymes produced by some gram negative bacteria like *E. coli*.[3] Also *Escherichia coli* is less sensitive to amoxicillin (36.67%). only 13.33% of the *Escherichia coli* tested are sensitive to oxacillin and 33.3% are sensitive to cloxacillin. This may be due to the fact that *Escherichia coli* is not included in their spectrum of activity as they are mainly designed for *staphylococci*, however in terms of augumentin the activity on *E. coli* has improved compared to the other drugs. This is because augumentin is a synergistic drug containing amoxicillin coupled with clavulanic acid, which protects the structure and integrity of amoxicillin and prevents it from being destroyed by beta-lactamases.[4]

The amino penicillin i.e. ampicillin and amoxicillin showed almost equal activity on *klebsiella spp* (33.3% and 36.7% respectively), while oxacillin are less active on it (16.67%) so also cloxacillin with 23.33% activity even though a little higher than oxacillim. among the five drugs, the most effective drug on *klebsiella spp* is augumentin which is 60% active on the 30 *klebsiella spp* isolates tested.

Proteus spp were 36.67% sensitive to ampicillin, which is in contrast with the work of.[2] Who studied activity comparism between penicillin and ampicillin in which majority of the *Proteus spp* isolate were sensitive to ampicillin. However 50% of the *Proteus spp* are sensitive to amoxicillin. This shows that, amoxicillin has higher activity on *Proteus spp* than ampicillin in this study. While 16.67% of the organisms are sensitive to oxacillin, 43.3% are sensitive to cloxacillin and 60% are sensitive to augumentin.

In the case of *Staphylococcos aureus* 53.33% are sensitive to ampicillins while 46.67% are sensitive to amoxicillins however 60% of these organisms are sensitive to oxacillin this is due to the fact that oxacillin is one of the pencillase resistant penicillins that are mainly designed to work on *staphylococci* that is the cloxacillin 53.33% previously show activity on, *staphylococcos aureus* that produce beta-lactamase (penicillinase). However some of the *staphylococcos aureus* are resistant to these agents and are referred to are methicillin resistant staphylococcus aureus.[12]

The activity of the penicillin drugs on *Pseudomonas aeruginosa* is very poor, only augumentin showed little activity to *Pseudomonas aeruginosa* were 23.33% of the 30 *pseudomonas aeruginosa* tested are sensitive to it, oxacillin and cloxacillin are poorly active on *pseudomonas aeruginosa* with 10% sensitivity each, while ampicillin and amoxicillin are also poorly active on it with 13.33% and 16.67% respectively. However in contrast to the work of[1] who evaluate effectiveness of penicillin group combination commonly use in Nigerian clinics on selected microorganism, which reported that *pseudomonas aeruginosa* have shown total resistance to ampicillin. *Pseudomonas aeruginosa* emerges to be the most resistant organism of all the clinical isolates tested even though augumentin has shown little activity but he percentage is very low 23.33%, this is not surprising because pseudomonas aeruginosa has been reported as a multi-drug resistant organism by various researches the problem and trouble of insensitivity or resistant worldwide.[10]

Resistant in *pseudomonas aeruginosa* may be due to the fact that it is a highly prevalent opportunistic pathogen. One of the most worrisome characteristic of *pseudomonas aeruginosa* is its low antibiotic susceptibility, which is attributable to a concerted action

of multi-drug efflux pumps with chromosomally encoded antibiotic resistant genes and the low permeability of the bacterial cellular envelopes.[17] *Pseudomonas aeruginosa* has the ability to produce HAQS and it has been found that HAQS have peroxidant effects, and over expressing modestly increased susceptibility to antibiotics.[20] The study experimented with the pseudomonas aeruginosa biofilms revealed that a disruption of relA and spoT genes produced an inactivation of the stringent response (SR) in cells who were with nutrient limitation which provide cells be more susceptibles to antibiotic.[21]

Streptococcus pneumoniae has shown sensitivity to the penicillins. 60% of *Streptococcus pneumoniae* are sensitive to augumentin and oxacillin. This is due to the fact that augumentin is a combination of amoxicillin drug and clavulanic acid, therefore stronger than amoxicillin alone and the fact that oxacillins are mainly designed to act on streptococci and staphylococci, they also show sensitivity to ampicillin 50% and also about 40% of *Streptococcus pneumonia* are sensitive to cloxacillin, It has been reported that Resistance in *Streptococcus pneumoniae* to penicillin and other beta-lactams is increasing worldwide. The major mechanism of resistance involves the introduction of mutations in genes encoding penicillin-binding proteins. Selective pressure is thought to play an important role.[10]

50% of *Neisseniu gonorhoeae* are sensitive to augumentin, while it is 40% sensitive to ampicillin but *Neissenia gonorhoeae* are less sensitive to oxacillin and cloxicillin with 20% sensitivity each. And 50% sensitive to amoxicillin.

Salmonella and *Shigella spp* show almost equal sensitivity to all the penicillins in overall percentage. However in the case of amoxicillin 60% of the *salmonella spp* isolate were sensitive. while oxacillin and cloxacilin have the same percentage of sensitivity on *salmonella spp* 40% each. 40% sensitivity to ampicillin on *salmonella spp* is in line with the finding of[16] who stated that salmonella spp are 40% sensilivie to ampicillin.

The activity of augumentin on *Enterococcus faecalis* and *Enterobacter aerogenes* is 40% of the organisms are sensitive to it. During the cause of study various brands of amoxiallin brands used show differences in activity on the same bacterial isolates so also ampicilliin, however these brands were assumed to contain equal active ingredient so that the extent of their activity is expected to be similar to route of manufacturing, processing, importation, half life and also manufacture and expiry date. It may also be due to chances that the drugs have hot been exposed to right environmental and storage conditions, thus may affect the active ingredient.

Also from the study carried out it is observed that the gram positive organisms are more sensitive to penicillins a (*staphylococcos aureus;* 48.20%, *Enterocuccus faecalis* 48.46% *Streptococcus pneumonia* 44.62%). Whereas gram negative bacteria are less sensitive to pencillins (*Escherichia coli* 33.59%) *klebsiella spp* 30.25%, 40.26% for *proteus spp*, *Enterobactea aerogenes* 35.38%, *Shigella spp* 36.92%, *Salmonella spp 35.38%*).

Resistance in gram negative organisms may be due to the fact that the structure of the cell wall may provides resistance to drug effects in gram negative bacteria. With their extra lipid bilayer, many antibiotics may not reach the sites of action (transpeptidase and the PBP). Therefore any antibiotic drug that is not lipid soluble enough to bilavers the outer lipid bilayer and small enough to traverse the purin channel will have no effect on the micro organism. Alternatively, a small drug with suitable solubility profile may pass through the purin channel and exert an antibacterial effect.[13]

While, gram positive bacteria do not have thick cell wall that the gram negative bacteria do. Gram positive cell walls lack the tough outer wall that the gram negative bacteria have, making them more susceptible to penicillin.[22]

It is evident from many researches carried all over the world that penicillin resistant is not just a problem in one area, it is a problem all over the world, resistance resulted from un necessary use and over use of penicillin.[2]

5.0 CONCLUSION

A study on the activity of commonly used penicillin on some clinical bacterial isolates reveals those gram positive organisms are more sensitive to penicillin than gram negative organism. Augumentin has the highest spectrum of activity on both the organisms. While *Pseudomonas aeruginosa* emerges to be the most resistant organism to penicillin *staphylococcus aureus* emerges to be the most sensitive organism, being mostly sensitive to oxacillin, cloxacillin and augumentin.

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