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Bactericidal Potency of Common Therapeutic Antibiotics Sold In Pharmaceutical Stores In A Southwest Community of Nigeria

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Abstract

Twenty-four (24) samples of antimicrobial agents were purchased from four major pharmaceutical stores in Abeokuta, Southwestern Nigeria. The effectiveness of the antibiotics was tested on *Staphylococcus aureus* and *Escherichia coli* by agar diffusion method. The micro-biological assay of the antibiotics revealed that Ampicillin from two of the pharmaceutical stores had the highest potency for *Staphylococcus aureus*, while the least potency for ciprofloxacin was observed in only one of the two. Complete resistance for chloramphenicol was observed in all outfits ($P < 0.05$). *Escherichia coli* exhibited significant resistance ($P > 0.05$) to all the antibiotics except for tetracycline where over 100% potency was observed in all the outfits.

Keywords: Antibiotics, Potency, Pharmaceutical, *Staphylococcus aureus, Escherichia coli*

1.0 Introduction

Chemotherapeutic agents are chemical substances used for treatment of infectious diseases caused by the proliferation of malignant cells. These substances are prepared in chemical laboratories or obtained from microorganisms and some plants and animals. In general, naturally occurring substances are distinguished from synthetic components by the name antibiotics (Pelczar *et al*. 2004). Antibiotics are special kind of chemotherapeutic agents usually obtained from living organisms. An antibiotic refers to a metabolic product of one microorganism that in very small amount is detrimental or inhibitory to other microorganism (Pelczar *et al*. 2004).

The first antibiotic compounds used in modern medicine were produced and isolated from living organisms. For example the penicillin class was produced by fungi in the genus *Penicillium* and *streptomycin* from bacteria of the genus *Streptomyces*. However Maskel and William (1987) reported that the first known use of antibiotic was by the ancient Chinese over 2500 years ago. The antibiotic described by Ernest Duchesne in 1893 however was without much notice until Alexander Fleming's discovery of penicillin in 1928 (Prescott *et al*. 2002).

Antibiotics can be categorized based on their target specificity: Narrow-spectrum antibiotics target particular types of microorganism such as Gram-negative or Gram-positive bacteria, while broad-spectrum antibiotics affect a wide range of microorganisms. Antibiotics can be classified as either bactericidals or bacteriostatics. Bactericidals kill bacteria directly where as bacteriostatics prevent them from dividing. However both are capable of ending a bacterial infection (Dubois and St. Pierre 2000).

Antibiotics disrupt microbial processes or structures that differ from those of the host. They may damage pathogens by hampering cell wall synthesis, inhibiting microbial protein and nucleic acid synthesis, disrupting microbial membrane structure and blocking metabolic pathways through inhibition of key enzymes (Prescott *et al*. 2002). Also effectiveness of individual antibiotic varies with the location of the infection, the ability of the antibiotic to reach the site of infection and the ability of the microbes to inactivate or excrete the antibiotic. Antibiotics have varying side effects. These side effects depend on the antibiotics used and the microorganisms targeted. Adverse effects can range from fever and nausea to major allergic reaction including photo-dermatitis. One of the most common side effects is diarrhea, sometimes caused by the

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anaerobic bacterium *Clostridium dificile* which results from antibiotics disrupting the normal balance of the intestinal flora (Speciale *et al.* 1995).

Antibiotic resistance is as a result the of use or misuse of antibiotics. Use or misuse of antibiotics can result in the development of antibiotic resistance by the infecting organism, failure to take the entire prescribed course of antibiotics or failure to resist for sufficiency recovery allowing clearance from infecting organism. These practices may cause the development of bacterial population with antibiotic resistance (Alhambra *et al*. 2004, Moran *et al*. 2006). Therefore main objective of this study is to determine the effectiveness and significant of antibiotic potency sold in various pharmaceutical outfits in Abeokuta against specific clinical pathogenic microorganisms.

2.0 Materials And Methods

2.1 Study Area

The study was conducted in Abeokuta -historic Yoruba city founded by the Egba, located in the tropical rain forest zone of southwest Nigeria and lies within longitude 3°21" East and latitude 7°11" North.

2.2 Antibiotic Samples

Antibiotic samples were purchased from four pharmaceutic stores located in North and South Local Government areas of Abeokuta, Nigeria: Precious (S_1), Leotapharm (S_2), Raybambs (S_3) and Anuoluwapo (S_4). The antibiotics include Ampicillin (Amp) – 250mg, Amoxyllin (Amox) – 500mg, Ofloxacin (Ofl) – 200mg, Ciprofloxacin (Cipro) – 200mg, Ampiclox (Ampl) – 500mg and Chloramphenicol (Chlora) – 250mg.

2.3 Preparation of Media

The media were prepared according to the manufacturer's directive.Nutrient broth (PSAMATEC, UK) was weighed (5.5mg), dissolved in 250mL distilled water and 20mL each was dispensed into McCartney bottles. They were autoclaved at 121°C for 15min at 15lb/sq inch, allowed to cool in a slanting position and then stored in the refrigerator at 4°C.

Also 9.5mg of Muller-Hinton agar dissolved in 250mL distilled water in a conical flask was autoclaved at 121°C for 15min at 15lb/sq inch. Thereafter it was dispensed (20mL) into Petri-dishes, allowed to solidify and then stored in the refrigerator at 4°C until ready for use.

3.0 Sample Preparation

Each sample was reconstituted by dissolving in sterile distilled water. By serial dilution, it was then diluted to required low and high dose (µg/mL) as indicated in Table 1, and then stored in the refrigerator at 4°C.

A quantity of the reference standard of antibiotic (see Table 1) was weighed and dissolved in 10mL distilled water. By serial dilution, the solution was diluted to required low and high dose (µg/mL) and then stored in the refrigerator at 4°C.

3.1 Bacteria Inoculum

Typed cultures of *Staphylococcus aureus* and *Escherichia coli* were obtained from the Drug and Vaccine Control Department of the National Agency for Food and Drug Administration and Control, Yaba, Lagos, Nigeria. A uniform suspension of an overnight pure culture of each organism was prepared by inoculating a loopful in 15 ml nutrient broth. Incubation was carried out at 37°C for 24hr.

Table 1: Reference Standard Percentage Potency Range

Antibiotics	Potency (%) B.P. Range
Ampicillin	93.0 – 108
Amoxyllin	90.5 – 110
Chloramphenicol	95.0 – 105
Tetracycline	95.0 – 105
Ciprofloxacin	95.0 – 105
Ampiclox	90.0 – 102

B.P. refers to British Pharmacopeia

Source: Drug and Vaccine Control Department, National Agency for Food and Drug Administration, Yaba, Lagos, Nigeria.

3.2 Antibiotic Potency Testing

Muller-Hinton agar plates were dried in the oven at 45°C for 30min. The agar plates were seeded with 1.5mL of bacterial inoculum. Excess of the suspension were drained off. Four (4) wells of 6mm in diameter and 2cm apart were bored in the Muller-Hinton agar using sterile cork borer and 0.2mL of each sample (antibiotic) and standard were used to fill each well (duplicate). The wells were allowed to stand for one hour at room temperature before incubation at 37°C for 24 h. Zones of inhibition around the wells were taken as a measure of antimicrobial activity. Diameters of zones inhibition were then measured in millimeter (mm) with a calibrated caliper. The percentage potency, P_A, of antibiotic was determined using:

$$P_A = \frac{\bar{IZ}_{TA}}{\bar{IZ}_{RS}} \times 100 \quad (\%)$$

where $\bar{IZ}_{TA}$ is the average zone of inhibition of test antibiotic in mm, $\bar{IZ}_{RS}$ is the average zone of inhibition of reference standard in mm.

4.0 Results And Discussion

A total of twenty-four (24) samples of antibiotics purchased from various pharmaceutical outfits in Abeokuta metropolis were analyzed for potency to *Staphylococcus aureus* and *Escherichia coli*. Results of the potency of various antibiotics for the bacterial isolates are presented in Table 2.It shows that Ampicillin in outfit S_4 (115%) and tetracycline in outfit S_1 (115%) were statistically most potent for *Staphylococcus aureus,* the least potency was with ciprofloxacin in outfit S_4 (81%) and complete resistant with chloramphenicol in all outfits ($P < 0.05$).*Escherichia coli* exhibited significant resistance ($P > 0.05$) to all the antibiotics except for tetracycline where over 100% potency was observed in all the outfits. The least potency was observed in ciprofloxacin from outfit S_1 (96%) and S_3 (89%) for *Escherichia coli* ($P > 0.05$).This study shows the effectiveness of the tested antibiotics against the bacteria (*Staphylococcus aureus* and *Escherichia coli*) used by the production of zones of inhibition. Ampicillin, amoxyllin, tetracycline, ciprofloxacin and ampiclox were found to be effective against *Staphylococcus aureus* except chloramphenicol which agrees with previous findings (Novick *et al.* 1977; Cheng *et al.* 2007). This is in contrast to the clinical reports of some fluoroquionlones (Blondeau *et al*. 2000; Grigg *et al*. 2003; Hancock 2005; Miller *et al.* 2006). Clinical studies in the study area will be needed to confirm these results.

Table 2: Potency of Antibiotics to *Staphylococcus aureus* and *Escherichia coli*

Antibiotics	Potency (%)				Potency (%)			
	Staphylococcus aureus				*Escherichia coli*			
	S_1	**S_2**	**S_3**	**S_4**	**S_1**	**S_2**	**S_3**	**S_4**
Ampicillin	96	113	103	115	R	R	R	R
Amoxyllin	100	94	95	89	R	R	R	R
Chloramphenicol	R	R	R	R	R	R	R	R
Tetracycline	115	109	96	90	104	113	111	104
Ciprofloxacin	92	91	93	81	R	R	R	R
Ampiclox	100	94	95	89	96	R	89	R

R – Resistant, S_1 – Pharmacy Store 1, S_2 – Pharmacy Store 2, S_3 – Pharmacy Store 3, S_4 – Pharmacy Store 4.

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