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Quinolone Resistant *Staphylococcus aureus* in Okigwe, Imo State Nigeria

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Abstract: The antimicrobial spectrum of quinolones namely cephalexin, ciprofloxacin, tarivid, peflacin and nalidixic acid on *Staphylococcus aureus* isolated from hospital patients in Okigwe was studied. Out of 79 samples examined *Staphylococcus aureus* was isolated from 50(71.42%). Out of 17 urethral swab isolates 15(88.27%), 10(58.8%), 12(70.5%), 8(47.05%) and 8(47.05%) were resistant to cephalexin, ciprofloxacin, tarivid, peflacin and nalidixic acid, respectively. The percentage of the of the *S. aureus* isolates resistant to each quinolone was 62, 64, 74, 44 and 88% for cephalexin, ciprofloxacin tarivid, peflacin and nalidixic acid, respectively. Isolates from urine, semen and vagina were less resistant to the quinolones.

Key words: Quinolones, *Staphylococcus aureus*, resistant, Okigwe, inhibition zone diameter

INTRODUCTION

Attempts to control diseases by chemotherapy through the use of antimicrobial agents particularly antibiotics have results in increased prevalence of resistance to these agents (Levy, 1998). *Staphylococcus aureus* is a Gram-positive, catalase positive, coagulase positive non motile coccus bacterium that causes a variety of human infections and is a major cause of surgical wound infections and epidermal skin diseases in newborn infants. They live as commensals in anterior nares of more half the population of humans (Doig, 1981) and the cocci spread from these sites into the environment by hands, handkerchief, clothing and dust. *S. aureus* is an opportunistic pathogen in the sense that it causes infection most commonly in tissues and sites with lowered host resistance (Burnett *et al.*, 1990). Wound infections can be severe and the organism can invade the blood stream with consequent seeding of other sites such as heart valves, causing endocarditis (Prescott *et al.*, 1996).

Various studies have been conducted to study the antimicrobial resistance pattern of *S. aureus* and it has been shown to be resistant to β -lactam antibiotics, aminoglycosides and macrolides (Atkinson and Lorian, 1984; Maple *et al.*, 1989). *S. aureus* strains carry a wide variety of multidrug resistance genes on plasmids, which can be exchanged and spread among different species of Staphylococci (Neihart *et al.*, 1988).

The quinolones are a group of synthetic antimicrobial agents that have gained wide usage (Boreherding *et al.*, 1996). Shortly after the introduction of the

fluoroquinolones into clinical practice, strains expressing resistance to these compounds were detected in particular among isolates of methicillin-resistant *S. aureus* (MRSA), which frequently exhibit multiple additional resistance determinants to unrelated antimicrobial agents (Trucksis *et al.*, 1991).

In developing countries such as Nigeria where drugs are readily available to consumers across the counter with or without prescription from medical practitioners, drug resistance is a serious problem. Misuse of antimicrobial drugs has been associated with high prevalence of drug resistance among the Staphylococci (Nnochiri, 1973; Paul *et al.*, 1982). Researchers have reported that some hospital strains of *S. aureus* are resistance to all clinically useful antibiotics except vancomycin. Some workers have also reported the presence of vancomycin resistant *S. aureus* strains (Aury-Damon *et al.*, 1998).

This study reports the resistance of *Staphylococcus aureus* hospital isolates in Okigwe to quinolones.

MATERIALS AND METHODS

Study area: This research was carried out in Okigwe Imo state, Nigeria between March and October 2006.

Antibiotics and media used: Antibiotic discs used and their concentrations were as follows: Tarivid (10 mg/disc), cephalexin (30 mg/disc), peflacin (10 mg/disc), nalidixic acid (30 mg/disc) and ciprofloxacin (30 mg/disc). These discs were obtained commercially. Optun disc (Optun laboratories, Aba Nigeria) were used. The media used

were Nutrient agar (Oxoid), Mannitol Salt agar (Difco), Bacto-peptone (Oxoid) and Mueller Hinton agar (Antec Diagnostics India) and were prepared according to manufacturers instructions.

Sample collection: Ear, vaginal, urethral, wound and carbuncle swabs were collected from patients attending General hospital Okigwe and Winners Hospital Okigwe using sterile swab sticks (Evepon). Urine, semen and stool samples were also collected from patients in the same hospitals using sterile specimen bottles. All specimens were transported to the Microbiology Laboratory of Abia State University and cultured within 3 h of collection. A total of 79 samples were isolated and characterized for presence of bacteria.

The specimens were inoculated on mannitol salt agar plates and Nutrient agar plates with sterilized wire loop to obtain discrete colonies. The plates were incubated at 37°C for 24 h under aerobic conditions. After 24 h of incubation, the culture plates were examined recording appearance, size, colour and morphology of colonies. Grams stain reaction, catalase test and coagulase tests were carried out on isolates. Isolates that were gram-positive cocci, catalase positive and coagulated human plasma were considered as *Staphylococcus aureus* (Chigbu and Ezeronye, 2003; Uaboi-Egbenni, 2003).

SUSCEPTIBILITY OF ISOLATES TO THE QUINOLONES

Antimicrobial sensitivity test was carried out on all isolates using the paper diffusion technique. A 0.2 mL of the 12 h peptone water culture of the test organism was used to inoculate on sterile Mueller Hinton agar plate. This was spread over the entire surface of the agar plate using a sterile glass spreader and allowed to dry for about 15 to 30 min. The antibiotic discs were then placed on the agar using sterile forceps. Each disc was placed far from each other so as to avoid their zones of inhibition from coalescing into the other. The set up was then incubated at 37°C for 24 h to observe the zones of growth inhibition. The Inhibition Zone Diameter (IZD) was measured in millimeters and interpreted by the standard of National Committee for Clinical Laboratory Standards (NCCLS) (Cheesbrough, 2002).

RESULTS

Out of 79 samples collected, *Staphylococcus aureus* was isolated from 50 representing 71.42%. Thirty one (62%) of the isolates were resistant to ceporex, 32(64%) and 37(74%) were resistant to ciprofloxacin and tarivid, respectively, 22(44%) and 44(88%) were resistant to peflaccine and nalidixic acid, respectively (Table 1 and 2). Fifteen isolates from the different sites were uniformly

resistant to the quinolones (Table 3). The number of isolates from different sites and the percentage resistant to each antibiotic is summarized in Table 4.

Table 1: Antibiogram of *Staphylococcus aureus* isolates from Okigwe to quinolones

Plate No.	Quinolones				
	Ceporex	Ciprofloxacin	Tarivid	Peflaccine	Nalidixic acid
01	R	R	R	R	R
02	R	R	R	R	R
03	I	R	R	I	R
04	R	R	R	S	R
05	R	R	R	R	R
06	S	R	R	S	R
07	I	R	S	R	R
08	R	R	R	R	R
09	R	R	R	R	R
10	S	S	S	S	R
11	R	R	R	R	R
12	R	R	R	R	R
13	R	R	R	R	R
14	R	R	R	R	R
15	S	S	R	S	S
16	R	R	R	R	I
17	R	R	R	R	I
18	R	R	R	R	R
19	R	R	R	R	R
20	R	R	R	R	R
21	R	R	R	I	R
22	R	R	R	I	I
23	R	R	R	S	R
24	R	S	S	I	R
25	I	S	R	I	R
26	S	S	S	S	R
27	I	I	R	R	R
28	R	R	R	S	R
29	R	R	R	R	R
30	R	R	R	R	I
31	I	I	R	I	R
32	S	R	R	S	R
33	R	R	S	I	R
34	R	R	R	R	R
35	R	S	R	R	R
36	R	R	R	R	R
37	R	R	R	I	R
38	S	I	S	S	R
39	I	I	S	S	R
40	R	R	R	I	R
41	I	I	I	I	R
42	I	I	R	S	R
43	S	I	R	I	R
44	R	I	R	I	R
45	S	S	S	S	S
46	R	S	S	S	R
47	R	S	S	S	R
48	S	R	S	S	R
49	S	R	R	S	R
50	S	S	I	R	R

R = Resistant, I = Intermediate, S = Sensitivity, <12 mm = Resistant, 12-15 = Intermediate >16 mm sensitive

Table 2: Number and percentage of *Staphylococcus aureus* isolates that were resistant, sensitive or intermediate zone to the quinolones

Quinolones	No. of isolates resistant	No. of intermediate	No. of isolates sensitive
Ceporex	31(62)	8(16)	11(22)
Ciprofloxacin	32(64)	8(16)	10(20)
Tarivid	37(74)	3(6)	10(20)
Peflaccine	22(44)	11(22)	17(34)
Nalidixic acid	44(88)	4(8)	2(4)

N = 50, Numbers in bracket are in percentages

Table 3: Profile of isolates (*Staphylococcus aureus*) uniformly resistant to the 5 quinolones

Isolate No.	Source of sample	Zones of inhibition (mm)				
		Ceporex	Ciprofloxacin	Tarivid	Peflaccine	Nalixic acid
01	Urethral swab	4	5	0	0	5.5
02	Urethral swab	2	0	0	1	0
05	Carbuncle swab	0	0	0	0	0
08	Urethral swab	5	0	4	4	3
09	Stool	0	0	0	0	0
11	Semen	5	4	4	0	0
12	Stool	0	0	5.5	4	2.5
13	Stool	0	0	0	0	0
14	Wound	0	0	0	2	4
16	Wound	0	0	3	3	5
18	Urethral swab	4.5	2	0	0	1
19	Urethral swab	0	0	0	0	0
29	Wound swab	5	2	4	2	5
37	Urethral swab	0	0	0	0	0
39	Urine	2	0	0	0	0

Table 4: Number of isolates from different sources sensitive and resistant to quinolones

Source of sample	No. of isolate obtained	Ceporex		Ciprofloxacin		Tarivid		Peflaccine		Nalidixic acid	
		S	R	S	R	S	R	S	R	S	R
Urethral Swab	17	2	15	3	10	4	12	7	8	1	8
Carbuncle Swab	3	0	1	0	3	0	3	0	2	0	3
Discharging Ear	2	0	0	0	2	0	2	0	2	0	2
Vaginal Swab	4	1	0	1	1	1	2	1	1	0	4
Stool	4	0	4	0	4	0	4	0	4	0	3
Urine	8	4	3	2	4	1	6	2	3	0	7
Wound	6	2	4	2	4	1	4	3	3	1	5
Semen	6	2	3	2	3	2	3	2	1	0	6

S = Sensitivity (> 12 mm), R = Resistant (< 6 mm)

DISCUSSION

The high rate of isolation of *Staphylococcus aureus* in this study (71.42%) is not surprising because *S. aureus* is a normal microbial flora of humans and can therefore easily gain entry and cause infections as opportunistic pathogen. This high rate of isolation agrees with the finding of Chigbu and Ezeronye (2003). The high rate of resistance of *Staphylococcus aureus* to nalidixic acid has been reported by other researchers (Chigbu and Ezeronye, 2003). Nalidixic acid is one of the first generation quinolones, which has been on the pharmaceutical shops counter without regulation. People abuse the drug, selecting nalidixate resistant *S. aureus*. More so, nalidixic acid is a drug used against gram-negative organisms where as *S. aureus* is gram positive.

The low rate of resistance to peflaccine (44%) coupled with other quinolones used for this study could be due to the fact that peflaccine has not been long on the Nigerian market unlike the other quinolones that have been in the Nigerian market for some time now. The resistant rate of 64% reported for Ciprofloxacin in this work is similar to that reported by Buck *et al.* (2005), who reported 65% Ciprofloxacin resistant in *Staphylococcus aureus* in Minnesota, USA. The work suggested that ciprofloxacin resistant in Minnesota was a result of the use of

ciprofloxacin in the treatment of bacterial diseases of poultry. Fluoroquinolones resistance in Okigwe could likely be as a result of drug abuse. The regulation of antimicrobial agents administration is not effective in Nigeria especially with the limited health care facilities available and the high cost of health care delivery. Moreso in the rural communities drugs are easily procured across the counter.

Quinolones resistance by Gram negative and Gram positive cocci other than *Staphylococcus* has been reported in Taiwan and Alabama on *Salmonella* and *Streptococcus pneumonia* (Crystal *et al.*, 2005; Yan *et al.*, 2005) and was attributed to the use of quinolone related compounds in the manufacture of antiseptic soap but in developing countries, the major problem is drug abuse.

Staphylococcus aureus isolates used in this study were most susceptible to peflaccine (88%). This suggests that *Staphylococcus aureus* isolates have not acquired resistance at the rate at which they acquire resistance to other quinolones. This finding agrees with that of Chigbu and Ezeronye (2003) who also reported low resistance to peflaccine in *S. aureus* isolates from both hospital and non hospital subjects in Umuahia Abia State, Nigeria.

The higher prevalence of quinolone resistance *S. aureus* isolates in Okigwe could be due to widespread

indiscriminate use of antimicrobial agents. Rational drug use is important and should be emphasized in the formulation and implementation of a national drug policy by the government. Public health enlightenment on the use of drugs and drug abuse is necessary.

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