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Antimicrobial susceptibility testing of *Staphylococcus aureus* isolated from apparently healthy humans and animals in Maiduguri, Nigeria

J. Y. Adamu*, A. I. Raufu, F. C. Chimaroke and J. A. Ameh

Bacteriology Research Unit, Department of Veterinary Microbiology and Parasitology, Faculty of Veterinary Medicine, University of Maiduguri, Nigeria.

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ABSTRACT: *Staphylococcus aureus* was isolated from apparently healthy humans, sheep, goats and cattle. A total of 250 samples were collected, 130 was from humans, 40 each from sheep, goats and cattle respectively. The prevalence of *Staphylococcus aureus* from humans was 52.3%, sheep 42.5%, goats 30.0% and cattle 37.5%. The overall isolation rate was 44.8%. Human were found to harbour more *S. aureus* than the animals. The isolates were tested against 13 antimicrobial agents. Of these, Ciproxin, Norfloxacin, Rifampicin, Streptomycin, and Erythromycin showed the highest activity against *S. aureus* while Cefotaxime, Ceftazimide and Ampiclox showed least activity. It was concluded that Ciproxin, Norfloxacin, Rifampicin, Streptomycin, and Erythromycin may be the drug of choice in the treatment of infections caused by *S. aureus*.

Keywords: *Staphylococcus aureus*; Antimicrobial susceptibility; Human; Cattle; Sheep; Goats.

Introduction

Staphylococcus aureus is an important pathogen of humans and animals and is implicated in a wide variety of infections. Apparently normal humans and animals harbour *S. aureus* but become virulent when the immune mechanisms are compromised. It is a pathogen of greater concern because of its virulence (Chambers, 2005), its ability to cause a diverse array of life threatening infections and its ability to adapt to different environmental conditions (Lowy, 2003). *S. aureus* has been found to be the most frequently isolated pathogen causing bloodstream infections, skin and soft tissue infections, and pneumonia (Doern *et al*., 1999; Sader *et al*., 1999; Jones *et al*., 2003).

The discovery of antimicrobial agents has been a critical element of the therapeutic armamentarium of modern medicine. Unfortunately this pathogen has been particularly efficient at developing resistance to antimicrobial agents and treatment of infections caused by *S. aureus* is still a challenge to clinicians. Since the first isolation of methicillin-resistant *S. aureus* (MRSA) in the United Kingdom in 1961 (Jevons, 1961), increasing rates of methicillin resistance among *S. aureus* have been a cause for concern especially in developed countries. Resistance to penicillin in *S. aureus* is due to the production of β-lactamases. Community-acquired strains of *S. aureus* have been shown to be uniformly resistant to β-lactam antibiotics and also show cross-resistance to other antimicrobial agents (Olayinka *et al.*, 2004).

*To whom correspondence should be addressed. Tel: 08023767116
Email: adamuyaz@yahoo.com

The ability to characterize *S. aureus* and monitor antimicrobial susceptibility patterns is important for clinicians, formulation of public healthcare policies, and providing useful information on the global surveillance of this pathogen. Data on the antimicrobial susceptibility patterns of this pathogen in north-eastern Nigeria are inadequate. It was against this backdrop that this study was carried out to determine the prevalence and antimicrobial susceptibility pattern of *S. aureus* isolated from apparently healthy humans and animals in Maiduguri.

Materials and Methods

Sampling

Simple random sampling technique was employed to collect samples from apparently healthy humans, cattle, sheep and goats within the Maiduguri metropolis. A total of 250 samples were aseptically collected using a sterile swab sticks from the nasal mucosa of humans, cattle, sheep and goats and immediately brought on ice pack to the veterinary microbiology laboratory of the University of Maiduguri for bacteriological assay.

Bacteriology

On arrival at the laboratory, each swab stick was immediately inoculated onto mannitol salt agar plates and incubated at 37 ^{0}C for 24 h. The characteristic isolates were aseptically isolated and characterized using established microbiological methods that include colonial morphology, Gram stain characteristics and catalase and coagulase tests (Cheesbrough, 2002). Isolates that were Gram-positive cocci, catalase positive and coagulated human plasma were considered as *S. aureus* in addition to other standard biochemical test (Cowan and Steel, 2004).

Antimicrobial susceptibility testing

The antimicrobial susceptibility pattern of all isolated *S. aureus* to Ciproxin (10 µg), Lincocin (30 µg), Gentamycin (10 µg), Ampicloxacillin (30 µg), Rifampicin (10 µg), Peflacin (10 µg), Erythromycin (30 µg), Streptomycin (20 µg), Norbactin (30 µg), Chloramphenicol (30 µg), Ceftazimide (30 µg), Norfloxacin (10 µg) and Cefotaxime (30 µg) were determined by the modified Kirby-Bauer diffusion technique (Cheesbrough, 2002). Standardized overnight culture of each isolate (containing approximately 10^6 cfu/ml) was used to flood the surface of Mueller Hinton agar plates and excess drained off and dried while the Petri dish lid was in place. The standard antimicrobial discs were aseptically placed at reasonable equidistance on the inoculated plates and allowed to stand for I hr. The plates (prepared in duplicates) were then incubated at 37 ^{0}C for 18-24 h. The diameter of the zone of inhibition produced by each antimicrobial disc was measured with a ruler in millimeters. Breakpoints and interpretative criteria for susceptibility/resistance was based on the performance standards for antimicrobial disc susceptibility tests, approved standard of the National Committee for Clinical Laboratory Standards, 1999.

Statistical Analysis

Differences in the prevalences and the susceptibilities to the antimicrobial agents was performed using ANOVA (GraphPad Instat Statistical package, 2003).

Results

A total of 250 samples were collected from the anterior nares of apparently healthy humans, cattle, sheep and goats. The prevalence of *S. aureus* from humans was 52.3%, cattle 37.5%, sheep 42.5% and goats 30.0%. Overall isolation rate was 44.8%. Humans were found to harbour more *S. aureus* than the animals though it was not significant statistically ($P>0.05$) (Table 1).

Table 1. Prevalence of *S. aureus* isolated from apparently healthy humans, cattle, sheep and goats in Maiduguri.

Species	No. of samples collected	No. of samples positive for *S. aureus*	Prevalence (%)
Humans	130	68	52.3%
Cattle	40	15	37.5%
Sheep	40	17	42.5%
Goats	40	12	30.0%
Total	**250**	**112**	**44.8%**

The result of the antimicrobial susceptibility test showed that the isolates were highly susceptible to Ciproxin (91.1%), Norfloxacin (90.2%), Rifampicin (73.2), Streptomycin (72.3%), Erythromycin (71.4%), Norbactin (64.3%) and moderately susceptible to Peflacin (57.4%), Gentamycin (51.8%), Lincocin (50.9%), Chloramphenicol (42.0%) but showed resistance to Ceftazimide (7.1%), Cefotaxime (14.3%) and Ampiclox (31.3%). Ciproxin had the greatest activity against *S. aureus* while Ceftazimide had the least activity but these differences were not significant statistically (P>0.05) (Table 2; Fig.1).

Table 2. Antimicrobial susceptibility patterns of 112 *S. aureus* isolates from apparently healthy humans, cattle, sheep and goats in Maiduguri.

Antimicrobial	Drug concentration (µg)	Susceptibility (%)	Resistance (%)
Ciproxin	10	91.07	8.93
Erythromycin	30	71.43	28.57
Lincocin	30	50.89	49.11
Ampiclox	30	31.25	38.75
Rifampicin	10	73.21	26.79
Peflacin	10	57.14	42.86
Streptomycin	20	72.32	27.68
Norbactin	30	64.29	35.71
Chloramphenicol	30	41.96	58.04
Ceftazimide	30	7.14	92.86
Norfloxacin	10	90.18	9.82
Cefotaxime	30	14.29	85.71
Gentamycin	10	51.79	48.21

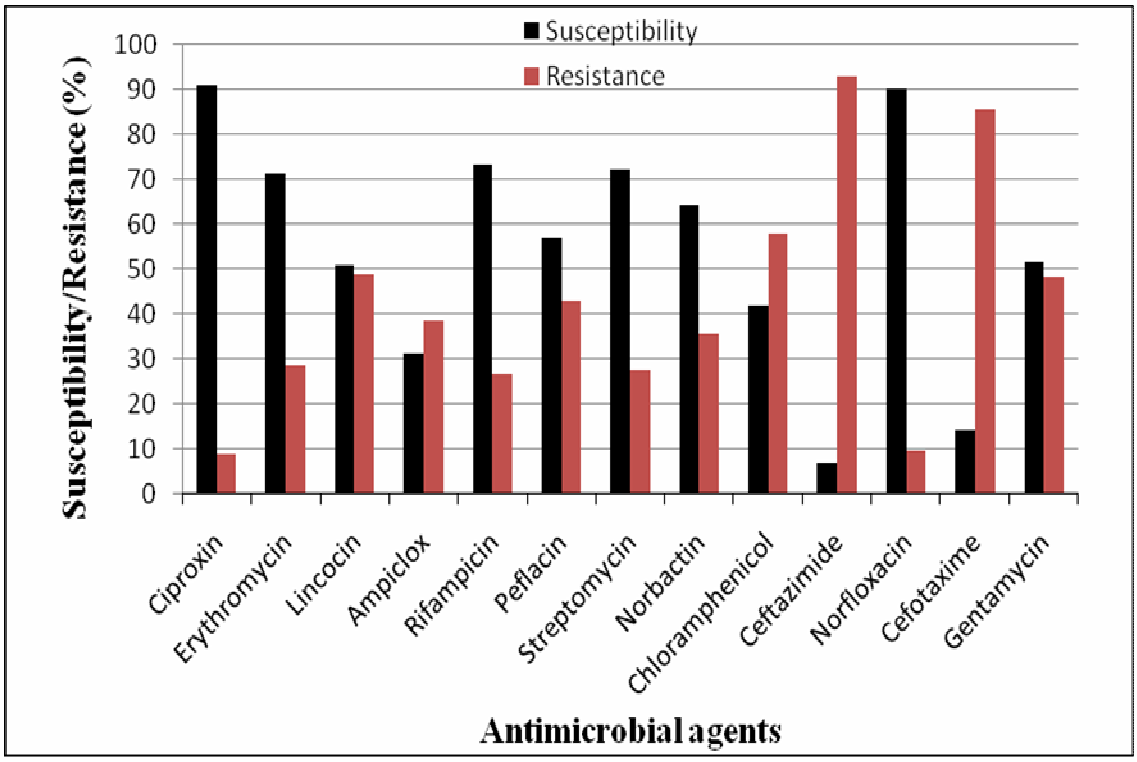

Fig. 1. Antimicrobial susceptibility pattern of *S. Aureus* isolated from humans, cattle, sheep and goats in Maiduguri.

Discussion

S. aureus is a ubiquitous microorganism and is implicated as causing infections of skin and soft tissues (Nordmann and Nass, 2005) especially when immune mechanisms of the host are compromised. There is increasing evidence of drug resistance by this microorganism (Lowy, 2003). An overall prevalence of 44.8% of *S. aureus* was obtained from nasal swab samples of apparently healthy humans, cattle, sheep and goats. This is slightly higher than earlier reports of Ehinmidu (2003) who reported prevalence of 43.3% and Onanuga *et al.* (2005) who reported a prevalence of 40.0%. Nasal carriers are indeed the most important reservoir of staphylococcal infection especially in man which can be spread within community and hospital environment. Domestic animals live in close association with human beings; this may explain the higher prevalence of *S. aureus* in humans than animals.

Most isolates were susceptible to Ciproxin, Rifampicin, Streptomycin and Erythromycin which is in agreement with previous reports (Ehinmidu, 2003; Olayinka *et al.*, 2004). The fluoroquinolones are newer drugs with mode of action central on inhibition of DNA replication which stops the multiplication of the bacteria cells and are relatively expensive and therefore are less available for abuse.

The resistance to Cefotaxime and Ceftazimide may not be unconnected with the production of β-lactamase or cephalosporinase enzymes by *S. aureus* which destroy the antimicrobial agents. Another observation is that most isolates of *S. aureus* are resistant to a large number of commonly prescribed antibiotics (Olukoya *et al.*, 1995).

The isolates were also moderately resistant to Peflacin, Gentamycin and Chloramphenicol which may not be unconnected to the complexity of aminoglycosides and the route of administration (Onanuga *et al*, 2005).

From the result of the study, it is therefore recommended that Ciproxin, Norflacin, Rifampicin, Erythromycin and Streptomycin may be the best choice of antimicrobials for treating infections caused by *S. aureus*. Antimicrobial susceptibility patterns are important for clinicians in selecting empiric antimicrobial therapy. Nationwide survey should be undertaken for rational formulation of public healthcare policies and providing useful information on the global surveillance of this pathogen.

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