ANTIBIOTIC RESISTANCE PATTERN OF ISOLATES FROM WOUND AND SOFT TISSUE INFECTIONS

Surg Cdr RN MISRA ^{*}, Lt Col YOGESH CHANDER ⁺,
Lt Col NK DEBATA (Retd) [#], Brig VC OHRI ^{**}

ABSTRACT

Two-hundred and eighty bacterial isolates from wound and soft tissue infections were studied for species identification and antibiotic resistance pattern. Amongst them 122 isolates were from community acquired infection and 158 were from nosocomial infections. The common community acquired pathogens were *Staphylococcus aureus* (67.8%) and *Streptococcus pyogenes* (10.7%), whereas *Staphylococcus aureus* (60.1%) and *E. coli* (8.9%) were common in nosocomial infection. Only two anaerobes (*Clostridium perfringens*) were isolated. Penicillin resistance was found to be 87% and 92% for *Staphylococcus aureus* in community acquired and nosocomial infections respectively. 85% of *Proteus* isolates were resistant to ampicillin. There was relatively lower level of resistance by all isolates to cefotaxime. Gentamicin showed higher rate of resistance than netilmicin and amikacin. Resistance of *E. coli* isolates to fluoroquinolones being 79% for norfloxacin, 81% for ciprofloxacin and 60% for ofloxacin. The study showed a higher resistance of methicillin resistant *Staphylococcus aureus* (MRSA) to other antibiotics. Amikacin and ofloxacin were the best recommended drugs for empirical therapy for all organisms, the susceptibility rate being 80.7% and 80.4%.

MJAFI 2000, 56 : 205-208

KEY WORDS: Antibiotic resistance; Soft tissue infections; Wound infections.

Introduction

Despite the fundamental role of antiseptics and antibiotics in the development of modern surgery, implementation of these practices have reduced but not eliminated post surgical infections. The wound infection depends on a complex interaction between host factors like immunity, nutritional status and age, wound related factors like magnitude of trauma, dead space, devitalization and presence of hematoma and microbial factors like toxins, invasion and resistance to antibiotics. [1]. Different species assume importance in different procedures viz. *Staphylococcus aureus* and coagulase negative staphylococci in clean surgical procedures [2,3] and polymicrobial flora e.g. *Escherichia coli* and bacteroides spp in contaminated procedures like colorectal surgery. The success of prophylaxis in clean surgery correlates directly with susceptibility of bacteria to antibiotics in vitro. This study aims to find out the prevalence of bacterial isolates and their antibiotic resistance pattern so as to facilitate the treating clinicians to select appropriate antibiotics for prophylaxis and treatment and also to develop the bacterial database to study the changes in bacterial behaviour in future.

Material and Methods:

The study was conducted in the Department of Microbiology, Armed Forces Medical College, and Command Hospital (SC), Pune a tertiary care hospital during the period Jan to Jun 1997.

The pus swabs and body fluid swabs which were received from wound and soft tissue infections from patients attending the out-patient department and those admitted to Command Hospital (SC) were inoculated onto Blood agar, MacConkey agar and Mannite salt agar and incubated at 37°C for 18-24 hours. Pus specimens received in suitable containers i.e. pus swabs in thioglycolate medium or RCM medium or aspirated pus in closed syringe were also inoculated onto blood agar for anaerobic culture. A direct smear for assessment of the cellularity and presence of bacteria was carried out in all cases. The isolates were identified by standard identification methods [4]. Antibiotic sensitivity was carried out by Stokes' disc diffusion method using commercial antibiotic discs or those prepared in our laboratory using pure powder [5]. The standard strains used for this purpose were *S aureus* NCTC 6571, *E coli* NCTC 10418 and *P aeruginosa* NCTC 10662 [5].

Results

S aureus constituted the majority of isolates both in the community and hospital infections being 67.2% and 60.1% respectively, followed by *Str pyogenes* in community infections (10.7%) and *E. coli* in nosocomial infections (8.9%). There were only two (1.3%) anaerobic infections due to *Clostridium perfringens* (Table-1). Majority of *S aureus* both from community and from hospital were resistant to penicillin being 87% and 92% respectively (Table-2). Methicillin resistance among hospital isolates was significantly more (54%) than the community isolates (43%). Resistance to Gentamicin amongst the isolate is much more than to netilmicin and amikacin (Table-3). Most of the organisms are sensitive to fluoroquinolones particularly to ofloxacin except *E coli* which were relatively more resistant to those drugs (Table-4). Many *S aureus* were sensitive to Clindamycin but *Proteus spp* were 100% resistant to tetracyclines (Table-5). Resistance pattern of MRSA strains to other antibiotics is given in table-6. Most of the MRSA were resistant to tetracycline, trimethoprim and sul-

^{*} Classified Specialist(Pathology and Microbiology), INHS Asvini, Colaba, Mumbai-400 005, ⁺ Reader, [#] Ex-Associate Professor, Department of Microbiology, Armed Forces Medical College, Pune 411 040 ^{**} Commandant, 167 Military Hospital, C/o 56 APO.

phamethoxazole and a majority to norfloxacin (Table-6). The overall susceptibility of all organisms to newly introduced antibiotics is given in table-7. Susceptibility to amikacin and ofloxacin was more being 80.7% and 80.4% respectively. The least susceptibility was shown to gentamicin (52.3%) and ciprofloxacin (53.5%). Two anaerobic isolates were sensitive to most of the antibiotics.

TABLE 1
Isolates from wound and soft tissue infections Jan.-Jun. 1997

Organism	Community Acquired	Hospital Acquired
<i>S aureus</i>	82 (67.2)	95 (60.1)
<i>CON Staphylococci</i>	06 (4.9)	08 (5.1)
<i>Strept pyogenes</i>	13 (10.7)	09 (5.7)
<i>E coli</i>	03 (2.5)	14 (8.9)
<i>Proteus spp</i>	05 (4.1)	06 (4.0)
<i>Ps aeruginosa</i>	09 (7.4)	09 (5.7)
<i>Clostridium spp</i>	-	02 (1.3)
<i>Others</i>	04 (3.3)	15 (9.5)
Total	122	158

Figures in parenthesis reflect percentage

TABLE 2
Beta-lactam resistance (%) in major isolates

	Pen	Amp	Met	Cef	Ctz	Cbn
<i>S aureus</i> (OPD) 82	87	-	43	29	-	-
<i>S aureus</i> (Hosp) 95	92	-	54	41	-	-
<i>E coli</i>	-	85	-	35	-	-
<i>Proteus spp</i>	-	63	-	11	-	-
<i>Ps aeruginosa</i>	-	-	-	33	40	31

Pen = Penicillin; Cef = Cefotaxime; Amp = Ampicillin; Ctz = Ceftazidime
Met = Methicillin; Cbn = Carbenicillin

TABLE 3
Aminoglycoside resistance (%) in major isolates

	Gen	Amk	Net
<i>S aureus</i> (OPD) 82	42	-	18
<i>S aureus</i> (Hosp) 95	44	-	22
<i>E coli</i>	65	11	46
<i>Proteus Spp</i>	54	11	10
<i>Ps aeruginosa</i>	88	40	63

Gen = Gentamicin, Amk = Amikacin, Net = Netilmicin

TABLE 4
Fluoroquinolones resistance (%) of major isolates

	Nor	Ofi	Cpx
<i>S aureus</i> (OPD) 82	35	19	33
<i>S aureus</i> (Hosp) 95	45	20	25
<i>E coli</i>	79	60	81
<i>Proteus Spp</i>	11	0	22
<i>Ps aeruginosa</i>	47	36	53

Nor = Norfloxacin, Ofi = Ofloxacin, Cpx = Ciprofloxacin

TABLE 5
Other antibacterial resistance (%) of major isolates

	Tet	Chl	Sul	Tmp	Cld	Pmx
<i>S aureus</i> (OPD) 82	56	16	61	60	20	-
<i>S aureus</i> (Hosp) 95	67	36	70	68	23	-
<i>E coli</i>	94	88	44	86	-	-
<i>Proteus Spp</i>	100	56	88	63	-	-
<i>Ps aeruginosa</i>	-	-	-	-	-	00

Tet = Tetracycline; Tmp = Trimethoprim; Chl = Chloromycetin;
Cld = Clindamycin; Sul = Sulphamethoxazole; Pmx = Polymyxin B

TABLE 6
Resistance of MRSA to other antibiotics

	MRSA (Hosp) (%)	MRSA (OPD) (%)	MRSA(Vellore) (%)
Erythromycin	69.0	69.2	84.2
Sulphamethoxazole	92.6	81.5	97.1
Trimethoprim	92.3	73.0	97.1
Tetracycline	93.3	88.5	97.9
Chloramphenicol	59.5	61.1	72.0
Cefotaxime	69.2	54.2	-
Clindamycin	41.0	21.7	-
Gentamicin	66.6	71.4	85.5
Netilmicin	30.7	64.7	12.0
Norfloxacin	80.0	66.6	90.0
Ofloxacin	33.3	40.0	53.0
Ciprofloxacin	40.0	-	92.2
Vancomycin	2.4	00	00
Amikacin	-	-	45.2

TABLE 7
Susceptibility of all organisms to newer antibiotics

Antibiotic	Susceptibility (%)
Cefotaxime	66.5
Gentamicin	52.3
Netilmicin	75.3
Amikacin	80.7
Ciprofloxacin	53.5
Norfloxacin	57.6
Ofloxacin	80.4

Discussion

S aureus was the commonest isolate from the community as well as nosocomial infections of wound and soft tissue. This could be due to the patient's endogenous flora, where the skin and the appendages are usually colonised by this organism [2]. However, rarely a particular surgeon or nurse is responsible for a cluster of staphylococcal infection. Strokes et al [6] found wound infection due to *S aureus* 17%, enterococci 13%, coagulase negative staphylococci 12% and *E coli* 10% [6]. However, selected populations such as diabetic individuals and patients on haemodialysis

may be colonised with *S aureus* at a frequency in excess of 50% [7]. Many of our patients suffered from diabetes mellitus and/or peripheral vascular disease. More than one isolate was observed in such patients.

After the discovery of β -lactamase in 1944, its production among the staphylococci is steadily increasing. Today it is produced in approximately 90% of staphylococcal isolates from community and hospital which are resistant to penicillin [8]. In this study penicillin resistance was 87% for community isolates and 92% for hospital isolates. Due to the plasmid and chromosomally mediated β -lactamase production among gram negative organisms, they were resistant to ampicillin and other β -lactams. In this study ampicillin resistance was seen among 88% *E coli* and 65% *Proteus spp*. Resistance to cefotaxime, a third generation cephalosporin was relatively low (Table-2).

Gentamicin resistance was higher among isolates as it was introduced earlier than netilmicin and amikacin (Table-3). Many strains with plasmid encoded extended spectrum β -lactamase were also aminoglycoside resistant. So large out-breaks of infection with such strains may result in an increase in aminoglycoside resistance rates [9]. Gentamicin resistance in our centre during 1996-97 was more than that found in an UK multicentric study where only 0.6% *E coli*, 6% of *K pneumoniae*, and 13% of *K oxytoca* were resistant to this aminoglycoside [10]. This could be due to more chronic nature of the cases in our study and the organisms with constant exposure to gentamicin in the hospital environment.

Resistance to fluoroquinolones occurs in organisms with high initial MIC including *Ps aeruginosa*, MRSA (with 80% resistance), coagulase negative staphylococci and *Acinetobacter spp*. Emergence of resistance during treatment with quinolones is most likely to occur when large number of organisms present at the site of infection are exposed to subinhibitory concentration of antibiotics either because of inadequate dosage or because of impaired antibiotic penetration e.g. prolonged or repeated chronic infection, impaired vascularity, osteomyelitis or presence of foreign bodies. Strategy to reduce resistance is to prescribe two to three drugs for the first 2-5 days followed by quinolones after reduction of the bacterial load [10]. In our series the resistance of *E coli* was 79% for norfloxacin, 60% for ofloxacin and 81% for ciprofloxacin but *Proteus spp* showed complete susceptibility to ofloxacin.

Among the *E coli* and *Proteus spp*, 94% and 100% were resistant to tetracycline respectively. *S*

aureus is relatively less resistant to chloramphenicol, 16% and 36% of community and hospital isolate respectively, possibly due to under-exposure to it. Clindamycin is still quite effective for *S aureus*, only 20% of isolates being resistant to it.

The MRSA strains show higher resistance to other antibiotics as well. They show 92.6% resistance to sulphamethoxazole, 92.3% to trimethoprim, 93.3% to tetracycline, and 80% to norfloxacin. The community isolates of MRSA show 81.5% resistance to sulphamethoxazole, 88% to trimethoprim. In another Indian study [12] 24% of staphylococci were MRSA which showed 97% resistance to trimethoprim, 85.5% to gentamicin, 45% to amikacin, 90% to norfloxacin and ciprofloxacin and 55% to ofloxacin. They recommend netilmicin as alternative drug for MRSA strains (Table-6), but we recommend vancomycin and ofloxacin for hospital isolates. Clindamycin and ofloxacin can be administered to community isolates of MRSA.

The characteristic of MRSA is that they produce altered penicillin binding proteins (PBP) 2a or 2 which are chromosomally transmitted. The other characteristics of MRSA include penicillinase production and a full armamentarium of pathogenic factors that include protein A, coagulase and DNase. Numerous in vitro studies have shown MRSA to be fully virulent and showed intraphagocytic killing potential as well resistant to a multitude of antibiotics [13-16].

The percentage susceptibility of all organisms to various newer antibiotics shows that highest number of organisms are susceptible to amikacin (80.7%), followed by ofloxacin (80.4%) and netilmicin (75.3%) (Table-7). This will help the treating physicians to decide on empirical antibiotic therapy until the sensitivity report is available.

REFERENCES

1. Kemodle DS, Kaiser AB. Post operative infections and antimicrobial prophylaxis. In: Mandell GL, Bennett JE, Dolin R, eds. Principles and Practice of Infectious Diseases. 4th ed. New York, Churchill Livingstone 1995:2742-56.
2. Ketcham AS, Lieberman JE, West JJ. Antibiotic prophylaxis and cancer surgery and its value in staphylococcal carrier patients. Surg Gynaecol Obstet 1963;117:1-6.
3. Weinstein HJ. The relation between the nasal staphylococcal carrier state and the incidence of post operative complications. N Engl J Med 1959;260:1303-7.
4. Collee JG, Fraser AG, Marmion BP, Simmons A. Mackie and McCartney Practical Medical Microbiology. 14th ed. New York, Churchill Livingstone, 1996:95-150.
5. A guide to sensitivity testing: Report of working party on antibiotic sensitivity testing of the British Society for Antimicrobial Chemotherapy. J Antimicrob Chemotherap 1991;27: Supplement D: 1-50.

6. Stokes EJ, Ridgway GL, Wren MWD. Clinical Microbiology. 7th ed. Boston, Edward Arnold 1993:251-2.
7. Schaberg DR, Culver DH, Gaynes RP. Major trends in the microbial aetiology of nosocomial infections. Am J Med 1991;91(Suppl 3B): 72-5.
8. Kirmani N, Tuazon CU, Murray HW. *Staphylococcus aureus* Arch Intern Med 1978;138:1657-9.
9. R Suderland. β -lactams: penicillins. In: O'Grady F, Finch RG, Lambert HP, Greenwood D, eds. Antibiotic and Chemotherapy. 7th ed. New York, Churchill Livingstone 1997:256-305.
10. Fisher GM, Kelsey MC, Corky EM. An investigation of the spread of gentamicin resistance in a district general hospital. J Med Microbiol 1986;22:69-77.
11. Brown EM, Reeves DS. Quinolones. In: O'Grady F, Lambert HP, Finch RG, Greenwood D, eds. Antibiotic and Chemotherapy. 7th edn. New York, Churchill Livingstone 1997:419-52.
12. Pulimood TB, Lalitha MK, Jesudasan MV, Pandian R, Selwyn J, John TJ. The spectrum of antimicrobial resistance among Methicillin Resistant *Staphylococcus aureus* in a tertiary care centre in India. Indian J Med Res 1996;103:212-5.
13. Hartman BM, Tomasz A. Low affinity penicillin binding protein associated with β -lactam resistance in *Staphylococcus aureus*. J Bacteriol 1984; 158: 513-7.
14. Peacock JE, Moorman DR, Wenzel RP. Methicillin resistant *Staphylococcus aureus*: Microbiologic characteristics, antimicrobial susceptibilities and assessment of virulence of an epidemic strain. J Infect Dis 1981;144:575-7.
15. Kinsman O, Naidoo J, Noble WC. Some effects of plasmid coding for antibiotic resistance on the virulence of *Staphylococcus aureus*. Pathology 1985;66:325-30.
16. Shalit I, Berger SA, Gorea A. Widespread quinolone resistance among methicillin resistance *Staphylococcus aureus* isolates in a general hospital. Antimicrob Agents Chemother 1989;33:595-7.