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ANTIBIOTIC SUSCEPTIBILITY PATTERN OF BACTERIAL ISOLATES IN WOUND INFECTION IN A TERTIARY CARE HOSPITAL

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ABSTRACT

Objectives: To determine the causative aerobic bacteria causing wound infection and to determine their antimicrobial sensitivity pattern.

Material and methods: Pus samples were collected from patients with wound infections and subjected to gram staining and aerobic bacterial culture . The isolates were identified by standard identification procedures .Antimicrobial susceptibility testing of the isolates was done by Kirby Bauer disc diffusion method.

Results : A total of 112 samples were processed over a period of 6 months. The most common isolate was Stapylococcus aureus followed by Klebsiella. Vancomycin(100%) followed by Amikacin (88.6%) were the most effective antimicrobial agents effective against S. aureus. The gramnegative isolates showed maximum sensitivity to amikacin (80-100%) and gentamicin (40-100%) .

Conclusion : Vancomycin and Amikacin are the most effective drugs in wound infection due to grampositive and Gram- negative bacteria respectively . Penicillins, macrolides and third generation cephalosporins should not be used as empiric therapy of wound infection

KEYWORDS : Bacteria ,wound infection, Antimicrobial susceptibility .

Introduction :

A wound infection is defined by the US Centre for Disease Control and Prevention (CDC) as surgical site infection (SSI).Skin and soft tissue infections(SSTIs) are a common type of infection that may cause longer hospital stay , increase the cost of medical care and play and important role in development of antimicrobial resistance. They are a common cause of morbidity in both community and hospitals.[1] SSTIs include cellulitis, abscesses, impetigo, folliculitis, furuncle, arbuncle,necrotizing fasciitis , diabetic foot infections and surgical site infections .Superficial infections can be dealt with by oral antibiotics and topical care. Complicated SSTI may turn out to be fatal and need hospitalization , intravenous antibiotics and / or surgery . SSTIs/Wound infections may be caused by a wide range of pathogens. these include grampositive pathogens like Staphylococcus aureus, CoNS, Enterococcus spp. Gram negative organisms include Pseudomonas aeruginosa, Escherichia coli, Klebsiella spp, proteus spp, acinetobacter spp, enterobacter spp and Serratia marcescens depending upon the prevalence of organism in the specific community. Even drug resistant organisms like Methicillin-resistant S. aureus ,Vancomycin resistant S. aureus and Enterococcus and ESBL producing gram negative pathogen are prevalent in these infections which cause a significant problem for physicians in deciding empiric therapy. [2,3,4] Each hospital has its own bacterial flora to which patients are at risk for acquiring health care associated infection. A working knowledge of the most likely causative organisms and their antibiotic sensitivity is essential for the physicians to deal with these infections. It is essential to monitor the changing trends in bacterial infection and their antibiotic susceptibility pattern to guide the physicians to deliver appropriate treatment to these patients. Moreover data concerning soft tissue infection is limited .[5] The objective of the present study was to determine the causative aerobic bacteria causing wound infection and to determine their antimicrobial sensitivity from pus specimens.

Materials and Methods

The study was conducted at Mediciti institute of medical sciences , RR district, Hyderabad over a period of 6 months from July 2012 to December 2012.The study population included in -patients as well as out patients of all ages.A total of 112 samples were processed. For patients with wounds with copious discharge, the area around the wound was cleaned with 70% ethyl alcohol and the exudates was collected from the depth of the wound using a sterile syringe.If adequate amount of exudate was not present, the sample was collected using two sterile cotton swabs by gently swabbing the surface of the wound. The swabs were used for Gram stain and culture, a third swab was also collected and was put in nutrient broth and incubated at 37°C.. All the specimens were processed immediately after being transported to the laboratory. A smear was made on a clean glass slide using one of the swabs and stained by Gram s staining. Gram stained smears were analyzed under oil immersion magnification. Presence of pus cells and microorganisms was determined. For each morphologically distinct microorganism seen, the Gram reaction , size ,shape and arrangement of bacteria were determined. The other swab was inoculated on nutrient agar, 5% blood agar and MacConkey agar by rolling the swab over the agar and streaking from primary inoculums using a sterile bacteriological loop. These plates were incubated aerobically at 37°C for 24- 48 hours. Primary plates were observed for any visible growth after overnight incubation and if there was no growth after 24 hours, subcultures were done from nutrient broth. Primary plates were further incubated for another 24 hours. Plates were observed for growth. The isolates were identified following standard identification procedures like colony morphology, Gram stained smear from the colony, motility and biochemical tests.Antimicrobial susceptibility testing of the bacterial isolates was done by Kirby Bauer disc diffusion method. The strengths of antibiotic discs used was as follows: Ampicillin 10mcg, Amikacin 30 mcg, Gentamicin 10mcg, Cotrimoxazole 25mcg, Levofloxacin 5mcg, Ofloxacin 5mcg, Ciprofloxacin 5mcg, Ceftriaxone 30mcg, Cefotaxime 30mcg, Cefoxitin 30mcg, Azithromycin 15mcg, Erythromycin 15mcg.,Vancomycin30mcg and ceftazidime30mcg, .

Results

Table.1 Distribution of bacterial isolates in wound infection

Bacterial isolate	No of isolates	% of isolates
Staphylococcus aureus	35	35%
Klebsiella	27	27%
Escherichia coli	21	21%
CONS	6	6%
Pseudomonas aeruginosa	5	5 %
Proteus	2	2%
Enterococci	2	2%
Citrobacter	2	2%

Table.2 Antibiogram of gram positive bacterial isolates

Antibiotic	Staphylococcus aureus		CONS		Enterococci	
	Number of Susceptible isolates (%)	Number of Resistant Isolates (%)	Number of Susceptible isolates (%)	Number of Resistant Isolates (%)	Number of Susceptible isolates (%)	Number of Resistant Isolates (%)
Ampicillin	7(20%)	28(80%)	2(33.3%)	4(66.7%)	0(0%)	100(100%)
Erythromycin	14(40%)	21(60%)	4(66.6%)	2(33.3%)	1(50%)	1(50%)
Ciprofloxacin	21(60%)	14(40%)	5(83.3%)	1(16.6%)	1(50%)	1(50%)
Levofloxacin	21(60%)	14(40%)	5(83.3%)	1(16.6%)	2(100%)	0(0%)
Ofloxacin	21(60%)	14(40%)	4(66.6%)	2(33.3%)	2(100%)	0(0%)
Azithromycin	15(42.7%)	20(57.3%)	4(66.6%)	2(33.3%)	1(50%)	1(50%)
Cefoxitin	18(51.4%)	17(48.6%)	4(66.6%)	2(33.3%)		
Amikacin	31(88.6%)	4 (11.4%)	5(83.3%)	1(16.6%)	2(100%)	0(0%)
Cotrimoxazole	15(42.7%)	21(60%)	2((33.3%)	4(66.7%)	1(50%)	1(50%)
Vancomycin	100(100%)	0(0%)	6(100%)	0(0%)	2(100%)	0(0%)

Table.3 Antibiogram of gramnegative bacterial isolates excluding Pseudomonas aeruginosa

Antibiotic	Esch.coli		klesiella		proteus		citrobacter	
	Number of Susceptible isolates (%)	Number of Resistant isolates (%)	Number of Susceptible isolates (%)	Number of Resistant isolates (%)	Number of Susceptible isolates (%)	Number of Resistant isolates (%)	Number of Susceptible isolates (%)	Number of Resistant isolates (%)
Ampicillin	2(9.5)%	19(90.5%)	3(11.5%)	24(88.5%)	0(0%)	2(100%)	0(0%)	2(100%)
Cefotaxime	5(23.8%)	16(76.2%)	7(25.9%)	20(74.1%)	1(50%)	1(50%)	1(50%)	1(50%)
Ceftazidime	6(28.57%)	15(71.4%)	9(33.3%)	18(66.7%)	1(50%)	1(50%)	1(50%)	1(50%)
Amikacin	18(85.7%)	3(14.3%)	25(92.6%)	2(7.4%)	2(100%)	0(0%)	2(100%)	0(0%)
Gentamicin	11(52.3%)	10(47.7%)	16(59.3%)	11(40.7%)	2(100%)	0(0%)	1(50%)	1(50%)
Ciprofloxacin	8(38.1%)	13(61.9%)	11(40.7%)	10(59.3%)	1(50%)	1(50%)	1(50%)	1(50%)
Cotimoxazole	10(47.6%)	11(52.4%)	18(66.6%)	3(33.3%)	1(50%)	1(50%)	1(50%)	1(50%)
Levofloxacin	8(38.1%)	13(60%)	15(55.5%)	45(44.5%)	1(50%)	1(50%)	1(50%)	1(50%)
Ofloxacin	32(32%)	68(68%)	13(48.1%)	59(51.9%)	46(46%)	54(54%)	1(50%)	1(50%)

Table.4 Antibiogram of Pseudomonas Aeruginosa

Antibiotic	Number of Susceptible isolates (%)	Number of Resistant isolates (%)
cefotaxime	2(40%)	3(60%)
Ceftriaxone	1(20%)	4(80%)
ceftazidime	2(40%)	3(60%)
ciprofloxacin	3(60%)	2(40%)
levofloxacin	3(60%)	2(40%)
ofloxacin	3(60%)	2(40%)
gentamycin	2(40%)	2(40%)
amikacin	4(80%)	1(20%)
Piperacillin	2(40%)	3(60%)

Discussion

In our study ,out of the 112 pus samples processed, 100 samples were culture positive . The most common isolate was Stapylococcus aureus followed by Klebsiella and Esch.coli..But ,Ramesh et al's study[6]revealed most common organism isolated as *E. coli* (20.8%), followed by *S. aureus* (16.1%). This could be because in their study the pus samples were collected from postoperative wound infections only.

In our study pus samples were collected from infections of skin and soft tissues and also from post operative wounds. However gramnegative isolates constituted 58% of our total isolates. Other studies also revealed higher prevalence of gram negative bacilli .[7,8]

Vancomycin(100%) followed by Amikacin(88.6%), quinolones (60%)and cotrimoxazole(42.7%) were the most effective antimicrobial agents effective against S. aureus. Out of 35 S. aureus isolated, 18 isolates (51.4%) were MRSA . In Kowli et al study [9] also showed similar results which supports our study. Whereas the study of Neelesh et al [10] revealed that Gram positive organisms were most sensitive to vancomycin (100%),followed by clindamycin (83.09%), linezolid (74.64%) & gentamicin (67.60%). However, Vancomycin is the most effective drug in wound infection due to grampositive bacteria. Among the resistant drugs, ampicillin (85%), erythromycin (60%) showed maximum resistance followed by azithromycin(57.3%) and quinolones (40%) to Staphylococcus aureus . This was in accordance with the study of Shreeram et al [11] whose study also showed 90% resistance to penicillin and ciprofloxacin and 70% resistance to erythromycin . The study of Neelesh also revealed maximum resistance to penicillin group. (90.5%). [10] High resistance to penicillins and macrolides in the above studies including our study suggests overuse of these drugs which have made then less useful in the recent years. The gramnegative isolates in our study showed maximum sensitivity to amikacin (80-100%) and gentamicin (40-100%) followed by clotrimoxazole (47.6 - 66.6%). Our sensitivity pattern for gramnegative isolates is similar to the study of Madhavi et al [12]where maximum sensitivity to Amikacin & gentamicin was observed. The study of Neelesh et al 10 and Shreeram et al[11] suggested maximum sensitivity to Amikacin & Ceftazidime. Analyzing the results of all these studies it is inferred that Amikacin is the most important drug to be considered against wound infection with Gram- negative bacteria. [13] Amikacin is most often used for treating severe hospital-aquired infections with multi drug resistant Gram-negative bacteria.[14] Our study reaccertains the same . The resistant pattern of gramnegative isolates in our study reveal high resistance to penicillins and third generation cephalosporins .The study of Madhavi et al [12]showed high resistance to third generation cephalosporins and quinolones and

Sreeram et al's study [11] showed high resistance to Penicillin and quinolones group of drugs. When the resistance pattern of gramnegative isolates in different studies was analysed it is evident that the resistance pattern of drugs is different in each hospital area and there could be an the overuse of third generation cephalosporins in our area.

Considering the resistance pattern of both grampositive and gramnegative bacteria isolated in our study and also the data revealed from other studies[9,10], we recommend that penicillins, macrolides and third generation cephalosporins should not be used as empiric therapy of wound infection in our hospital area.

CONCLUSION

Vancomycin is the most effective drug in wound infection due to grampositive bacteria.

Overuse of penicillins and macrolides against grampositive bacteria in wound infection have made then less useful in the recent years. Amikacin is the most important drug to be considered against wound infection with Gram- negative bacteria. The resistance pattern of drugs against gramnegative bacteria is different in each hospital area and there could be an excessive use of third generation cephalosporins in our area. Penicillins, macrolides and third generation cephalosporins should not be used as empiric therapy of wound infection in our local area.

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