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Pattern of Bacterial Profile and Drug Resistance of Pathogens Isolated from Wounds Infection at A Tertiary Care Hospital in North India

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Abstract

Background: Wounds can occur accidently or after surgery for wide variety of reasons. The exposed subcutaneous tissue is a favorable home for optimal growth of many microorganisms. Because of injudicious use of antimicrobials, getting medication without proper prescriptions there is rapid emergence of resistance to many important antimicrobials. This study was carried out to outline bacterial profile and antimicrobial drug resistance in our hospital.

Materials and Methods: All wound swabs received in microbiology laboratory were considered. These wound swabs were inoculated on blood agar in case of Gram positive bacteria and MacConkey agar for Gram negative bacteria. Series of biochemical tests were done for gram-negative and gram-positive bacterias. Antibiotic susceptibility test (AST) was performed for all isolates.

Results: A total of 496 patients with wound infection were included in this study. Out of 496 samples 387 (78.3%) were culture positive while 109 (21.97%) showed no growth. Out of 387 positive samples 121 (31.26%) were gram positive while rest 266 (68.73%) were gram negative. *S. aureus* (72) is mostly resistant to *erythromycin* 69 (95.8%), *levofloxacin* 449 (61.1%) and *cefoxitin* 44 (61.1%), but is susceptible to vancomycin, linezolid and amoxicillin. Pseudomonas aeruginosa (86) showed the highest resistance to *cefoperazone/ sulbactam* (100%) and *amikacin* 80 (93.02%), while as *ceftriaxone* and *ampicillin sulbactam* were most effective antibiotics with resistance rates of 2.3% and 4.6% respectively. *E. coli* showed highest resistance to *cotrimoxazole* 27 (79.4%) followed by *ampicillin-sulbactam* 25 (74%).

Conclusions: Antimicrobial resistance is also a serious concern in our hospital similar to other health care institutions in our country and worldwide.

Keywords: Wounds; Bacteria; Culture; Antimicrobial resistance.

Introduction

Wound is a breach in the skin and exposure of subcutaneous tissue following loss of skin integrity. The exposed subcutaneous tissue provides a favorable stratum for wide variety of organisms to contaminate and colonize and if the involved tissue is devitalized and if host immune response is compromised the conditions become optimal for microbial growth.[1,2] Wounds can be classified as accidental, pathological or postoperative.[3–5] The progression of a wound to an infected state is likely to involve a multitude of microbial or host factors.[6–8] Wounds are susceptible to contamination and colonization by a variety of both aerobic and anaerobic microorganisms. *Candida albicans* and *Candida tropicalis* have also been implicated as etiological agents.[3,5,9,10] Rapid emergence of antimicrobial resistant because of easy availability of antimicrobials without proper prescription is a public health calamity.[11,12] We have undertaken this study to discern bacteriological profile of infected wounds and antimicrobial resistance pattern which

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is very important as for as wound management is concerned.

Materials and Methods

This study was carried out in Government Medical College Jammu, a tertiary care hospital in north India. All wound swabs received in microbiology laboratory were studied and demographic information of these patients was recorded. These wound swabs were inoculated on blood agar in case of Gram positive bacteria and MacConkey agar for Gram negative bacteria. The plates were incubated in aerobic and microaerophilic atmosphere at 37°C for 24–48 hours. Positive cultures were identified by appearance on media, gram staining and biochemical reactions using standard methods.[13] Gram negative rods were identified by series of biochemical tests namely carbohydrate utilization tests, indole production, urease test, mannitol motility test, citrate utilization, oxidase test, triple sugar iron agar (TSI), H_2S production. Gram positive cocci were identified based on their gram reaction, catalase and coagulase test.

Antibiotic susceptibility test (AST): AST was performed for all isolates by using modified Kirby–Bauer disk diffusion method.[14] Standard procedures were used for AST for bacterial isolates. Mueller hinton agar was used for all gram positive and gram negative bacterias except for streptococci species. The sensitivity test for streotococci was performed on Mueller hinton agar with 5% sheep blood. Only the coventional antibiotics regularly used were included. All gram positive bacterias were tested against drugs such as *amoxicillin* (AMX), *ampicillin* (AMP), a*moxicillin/clavulanate* (AMC), *ampicillin/sulbactam* (AMS), *cefoxitin* (CX), *cotrimoxazole* (COT), *vancomycin* (VAN), *linezolid* (LZ), *levofloxacin* (LE), *clindamycin* (CD), *erythromycin* (ERY) and *tetracycline* (TE). Gram negative bacteria were tested against *ampicillin/sulbactam, ceftriaxone* (CTR), *gentamicin* (GEN), *amikacin* (AMK), t*icarcillin/clavulanate* (TCC), *ciprofloxacin* (CIP), *ceftazidime* (CTZ), *levofloxacin* (LE), *tetracycline* (TE), meropenem (MRP), *pipercillin/tazobactam* (PIT), *tigecycline* (TGC), *cefuroxime* (CFU), *cefoperazone/ sulbactam* (CFS), *aztreonam* (AT) and *cefepime* (CPM). Diameters of zone of inhibition around the disk were measured using ruler in mm and bacterial strains were classified into three groups as sensitive, intermediate and resistant as per Clinical and laboratory standard institute (CLSI).[15]

Results

A total of 496 patients with wound infection were included in this study out of which 328 (66.12%) were men and 168 (33.87%) were women resulting in overall male female ratio of 1.95:1. The infection rate was higher in men than women. Out of 496 samples 387 (78.3%) were culture positive while 109 (21.97%) of wound swab cultures showed no growth. Out of 387 positive samples 121 (31.26%) were gram positive while rest 266 (68.73%) were gram negative. *Staphylococcus aureus* 72 (59.50%) was the most frequently isolated gram positive bacteria whereas Pseudomonas aeruginosa 86 (32.33%) was most frequently isolated gram negative bacteria (Tables 1,2).

S. aureus is mostly resistant to erythromycin

Table 1: Type and Frequency of Gram Positive Isolates from Wounds

Gram positive organisms	No of isolates	Percentage
Staph. aureus	72	59.50
Enterococcus spp	25	20.66
CONS	19	15.70
Streptococcus spp	5	4.14
Total	**121**	

Table 2: Type and Frequency of Gram Negative Isolates from Wounds

Gram negative organisms	No. of isolates	Percentage
Psedomonas aeruginosa	86	32.33
Acinetobacter spp	42	15.78
Klebsiella spp	36	13.53
E.coli	34	12.78
Proteus spp	39	14.66
Enterobacter spp	15	5.63
Providencia spp	14	5.26
Total	**266**	

followed by *levofloxacin* and *cefoxitin* but is susceptible to *vancomycin, linezolid* and *amoxicillin*. *Enterococcus* species showed the highest resistance to *ampicillin, amoxicillin* and *levofloxacin*, while *CONS* showed high resistance to *cotrimoxazole* followed by *meropenem* and *ampicillin*. *Streptococcus* spp was 100% sensitive to all of antimicrobials tested.

Table 3a: Antimicrobial Drug Resistance Pattern of Gram Negative Bacteria

	Pseudomonas	*E. coli*	*Klebsiela*	*Acinetobacter*	*Proteus*	*Providencia*	*Enterobacter*
TE	13 (15.1)	8 (25.5)	20 (55.5)	32 (76)	6 (15.3)	7 (50)	NT
gen	38 (44.1)	8 (25.5)	19 (52.9)	21 (50)	16 (41)	2 (14.3)	2 (13.3)
cot	10 (11.6)	27 (79)	19 (52)	26 (61)	6 (15)	0	0
amk	80 (93)	6 (18)	8 (23)	16 (40)	6 (15)	4 (28)	5 (33)
cip	57 (66)	18 (54)	19 (53)	28 (66)	17 (43)	2 (14)	2 (13)
caz	70 (81)	10 (29)	0	2 (4)	2 (5)	0	4 (26)
ctr	2 (2.3)	13 (38)	4 (11.1)	4 (9.5)	2 (5.1)	0	NT
tcc	74 (86)	7 (20)	NT	12 (28)	4 (10.2)	0	NT
LE	35 (40.6)	13 (40)	16 (45)	27 (64)	17(43)	0	6 (44.4)
pit	74 (86)	NT	36 (100)	36 (87)	4 (10.2)	2 (14.4)	4 (26.6)
cpm	28 (32.5)	0	16 (44)	28 (66)	2 (5.1)	4 (28.5)	4 (26.6)
cfs	86 (100)	NT	16 (45)	NT	4 (10.2)	4 (28.5)	2 (13.3)
A/S	4 (4.6)	25 (74)	14 (38)	12 (28)	5 (12)	0	NT
Mrp	30 (34.8)	NT	35 (97.2)	17 (41)	14(35)	2 (14)	5 (33)
tgc	6 (6.9)	2 (5.5)	2 (5.5)	5 (11)	NT	0	NT
cfu	NT	NT	0	2 (4.7)	20 (51)	0	NT
avg	44.75%	24.4%	37.1137%	37.7%	19.1%	16.25%	16.26%

Antimicrobial resistance pattern of gram negative isolates: *Pseudomonas aeruginosa* showed the highest resistance to *cefoperazone/sulbactam* and *amikacin*, while *ceftriaxone* and *ampicillin sulbactam* were most effective antibiotics with resistance rates of 2.3% and 4.6% only. *Acinetobacter* species showed highest resistance to *pipercillin-tazobactam* followed by *tetracycline* whereas *ceftazidime* and *cefuroxime* were the most effective antibiotics with resistance of only 4.7%. *E.coli* showed highest resistance to *cotrimoxazole* followed by *ampicillin-sulbactam* but were most sensitive to *tigecycline* and *aztreonam* with resistance rate of 5.5%. L*evofloxacin* is the most resistant antibiotic in *Enterobacter* species folowed by amikacin and meropenem. *Providencia* showed highest resistance to *tetracycline* (Tables 3a, 3b).

Table 3b: Antimicrobial Drug Resistance Pattern of Gram Positive Bacteria

Antibiotics	***S. aureus* (72) In %**	***Streptococcus* (5) In %**	***CONS* (19) In %**	***Enterococcus* (25) In%**
ERY	69 (95.8)	0	6 (31.5)	2 (8)
AMP	17 (23.6)	NT	9 (47.3)	12 (48)
AMX	5 (6.9)	NT	NT	12 (48)
TE	8 (11.1)	NT	NT	12 (48)
COT	8 (11.1)	0	12 (63.1)	0
CD	39 (54.16)	0	NT	2 (8)
VAN	0	0	0	4 (16)
LZ	0	0	0	0
LE	44 (61.1)	0	7 (36.8)	12 (48)
AMC	25 (43.7)	0	5 (26.3)	12 (48)
A/S	23 (31.9)	0	NT	2 (8)
CX	44 (66.1)	NT	6 (31.5)	NT
Average %	33.37%		19.70%	20%

Discussion

In this study, a total of 496 clinical samples were collected from patients with wound infections. 387 (78.03%) samples yielded pure bacterial isolates. Though the prevalence rate of wound infections in the present study was mostly in concordance with other studies conducted in other countries like India (80%) and Nigeria (82%).[16,17] In study carried out by Ohalete higher rate of wound infections was observed in men than in women.[18]

In present study 68.73% gram negative bacteria and 31.26% gram positive were isolated. *Pseudomonas aeruginosa* was most common gram negative bacteria (32.33%), while *Staphylococcus* was most common (59.50%) among gram positive bacteria. Similar results were observed in study by Khosravi Ahmadi *et al.* and Akinjogunla OJ *et al*.[19,20] Higher frequency of *Staphylococcus aureus* and pseudomonas aeruginosa infection possibly could be because these bacterias might increase wound infection rate and cross-contamination among admitted patients. *S. aureus* showed average resistance rate of 33.3% to most of antimicrobial drugs tested which is analogous to other studies in literature.[21,22] *S. aureus* is mostly resistant to *erythromycin, levofloxacin* and *cefoxitin* but is susceptible to *vancomycin, linezolid* and *amoxicillin* which is in conformity with other studies.[22,23]

Pseudomonas showed complete reisitance to *cefoperazone/sulbactam* and very high level of resistance to *amikacin* while as *ceftriaxone* and *ampicillin-sulbactam* were most effective antibiotics.[24] *Acinetobacter* showed high resistance to *pipercillin/ tazobactam* followed by *tetracycline* while *ceftazidime* and *cefuroxime* were most effective antibiotics.[25]

Our study have shown complete resistance of *Klebsiella* spp to *pipercillin/tazobactam* and high level of resistance to meropenem which is similar to study by Yishak Abraham *et al*.[23] A very common gram-negative isolate in our study, *E.coli* was having highest resistance rate against *cotrimoxazole* followed by *ampicillin/sulbactam* which is in concurrence with other studies.[23–25]

Conclusion

For optimal wound management, regional knowledge and regular review of bacterial profile and drug susceptibility pattern is very important because of rapid increase in drug resistance worldwide.

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