Determination of Antimicrobial Sensitivities of Different Microorganisms in Post Surgical Wound Infection.

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ABSTRACT

Background : In this era of availability of multiple antibiotics, still surgical site infection poses a great challenge to surgeons in managing complicated wounds. Antibiotic resistance is also a big challenge resulting in increased morbidity and mortality. This study was done to get an insight of the sensitivities of microbes in our population and to get how much antibiotic resistance exist in our population.

Objective: Antimicrobial sensitivity of different organisms in surgical site infection.

Material and Methods: This cross sectional descriptive study was carried out at Department of Plastic Surgery, Saidu Teaching Hospital Swat and Northwest General Hospital and Research Center Peshawar, between January 2016 and February 2018.Laboratory services of Hyatabad medical complex were used. Seventy-five patients with SSI were included in the study and antimicrobial sensitivity of different organisms was determined by culture and sensitivity through disc diffusion method. Organisms were also stained for identification

Results: Out of 75 patients, 61 (81.3%) were males while 14 (18.7%) were females. The mean age of the patients was 31.7 ± 16.9 years. The most common age group was between 14-40 years (n=42, 56%). Minimum interval between surgery and development of infection was four days while maximum was 22 days, with the mean of 7.5 ±3.2 days.

Most common site of wound infection was lower limb (n=25, 34.7%) followed by upper limb (n=19, 25.3%).

Staphylococcus aureus was the most common isolate (n=25, 33.3%) followed by Escherichia coli (n=15, 20%) and Pseudomonas species (n=10, 13.3% %).Polymicrobial growth was present in 8 patients (10.7%).Drug sensitivity pattern of Staphylococcus aureus showed that Vancomycin and Linezolid were active against 100% of isolates followed by Fucidic acid 84 %.Staphylococcus aureus was highly resistant to Penicillins. E.coli isolates were sensitive to Meropenem in 100% cases while sensitivity to Amikacin and Ciprofloxacin was 60%.

Conclusion: Commonest organisms isolated from surgical wound infections were Staphylococcus aureus, Escherichia coli and Pseudomonas species. Isolates of staphylococcus aureus were sensitive to Vancomycin and Linezolid while E coli was sensitive to Meropenem in 100 % cases .

Key words: *Surgical site infection, Antimicrobial sensitivity, Drug resistance, Staphylococcus aureus, Linezolid.*

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INTRODUCTION

Surgical site infection is characterized clinically by increasing pain, redness, warmth or discharge from wound and confirmed by growth of 10^5 or more organisms[1]. The Centers for Disease Control and prevention (CDC) National Nosocomial Infection Survey (NNIS) includes a surgical site infection (SSI) if one out of four criteria is fulfilled: (1) purulent drainage (2) a positive culture result from wound swab (3) local symptoms and opened by a surgeon, unless culture result is negative (4) when the diagnosis is made by a surgeon or physician[2]. SSI are common, comprising 5 % of all hospital acquired infections and can lead to increased morbidity and mortality[3].

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SSI increase the hospital stay, mortality and chance of admission to ICU by 60%. In addition, SSI is responsible for five times more chance of readmission . Reduction of SSI can substantially decrease morbidity and reduce the financial burden for patient and the institution[4]. There is evidence that some interventions are useful in reducing SSI risk for people undergoing surgery, such as antibiotic prophylaxis for caesarean section and hernia repair, and also the timing of prophylactic intravenous antibiotics administered before caesarean incision. Also, there is evidence that adhesive drapes increase SSI risk [5]

The rate of SSI varies from hospital to hospital and depends on the type of surgery. It is becoming more serious issue in developing countries because of non adherence to general hygienic measures and mass production of low quality antiseptic solutions[1].

Post-operative wound infection may occur shortly after surgery or several days postoperatively. The occurrence of infection depends upon the virulence of the organisms, the physiological state of the wound.

The patient risk factors also play a major role in the development of infection. The various patient risk factors include immunological integrity of the host, diabetes, tobacco smoking, obesity and concomitant remote site infections or colonization.[6, 7] The bacteriological agents involved in wound infection vary with the geographical location, bacteria resident on the skin and clothing at the site of wound. The native flora of the skin, mucous membranes, or hollow viscera is usually responsible for SSI[8].

The common organisms include Staphylococcus aureus (21.7%), Klebsiella species (16.7%) Pseudomonas auruginosa (13%), followed by, E.coli (11.7%), atypical coliform, Proteus species (6.7%), Streptococcus pyogenes (1.7%) and Enterococcus faecalis (1.7%). Multiple pathogens may be isolated in 25% of wound culture results[9-11].The sensitivity and resistance patterns also vary in different studies but the incidence of Methicillin resistant Staphylococcus aureus (MRSA) is 32-38%[12, 13]. According to the National Nosocomial Infection Surveillance survey report, 61% of all the S. aureus in the health care setting is multi-drug-resistant, with mortality rates ranging from 20 to 25 percent[14]. Similarly, Pseudomonas auruginosa has showed up to 100% and 91% resistance to Ampicillin and Ceftazidime respectively. As there is growing incidence of drug resistance and misuse of antibiotics in a country like ours, microbiological studies of wound infections are of crucial importance. This study will identify the common organisms in our population. This will help in selecting correct antimicrobial agents, thereby improving the post surgical outcome.

MATERIALAND METHODS

The study was carried out at Department of Plastic surgery Saidu Teaching Hospital Swat and Northwest General hospital peshawar , between January 2016 and February 2018. Laboratory services of hayat abad medical complex was used. This was Cross sectional study and the duration of the study was 2 years in a total of 75 cases of post operative wound infection were included. Post-operative wound infection was defined by the clinical signs on examination. Informed consent was l taken from the patients. A pre designed proforma was filled in for each patient to collect information about age, sex, type of surgery and interval between surgery and wound infection (in days). Examination of wound was carried out for clinical signs of infection. In case of many infectious sites, bacteriological culture was taken from all infected sites. Wound samples (swabs) were collected aseptically with sterile cotton wool swab. These samples were immediately cultured for aerobes and their antimicrobial sensitivity pattern was studied. For aerobes, sample was inoculated into blood agar, Mac Conkey agar and chocolate agar. These plates were incubated at 37°C for 24-48 hours. Isolates were identified and characterized by their culture, morphological and biochemical staining characteristics. Disc diffusion test was used for drug sensitivity testing. Culture reports were collected from the laboratory. Strict clinical definition of wound infection was followed in order to avoid selection bias. Similarly care was taken in the extraction of information from the data so that information bias is reduced. Data was entered in computer by using SPSS version 17.Mean ±Standard deviation were calculated for age and interval between surgery and wound infection. Frequencies and percentages were presented for gender, type of surgery, organism and antibiotics sensitivity. Results were presented on tables and graphs.

RESULTS

A total of 75 patients of post operative wound infection were included in the study. Out of these 75 patients 61 (81.3%) were males while 14 (18.7%) were females.

The mean age of the patients was 31.7 ± 16.9 years. Minimum age was six years, while maximum age was 75 years. In majority of patients 42 (56%), age was ranging from 14- 40 years, followed by 21 patients (28%) were more than 40 years of age. Children (less than 14 years) make 16 % of the total population.

Minimum interval was four days while maximum was 22 days, with the mean interval of 7.5 ±3.2 days.

Most common site of wound infection was lower limb (n=25, 34.7%) followed by upper limb (n=19, 25.3%) as shown in table 1.

Staphylococcus aureus was the most common organism isolated from infected wounds accounting for 33.3% of the total organisms yielded. The next common organisms were E. coli and Pseudomonas which make 20% and 13.3% respectively. More than one organism was also isolated from eight patients. Moreover, fungus was also found in one specimen sent for culture and sensitivity (see table 2).

Staphylococcus aureus isolates were resistant to Ampicillin and amoxicillin in 100% of cases. Fucidic acid, Vancomycin and Linezolid showed good activity against Staphylococcus aureus. Fucidic acid showed resistance in only 4% cases, while Vancomycin and Linezolid showed no resistance against any of the isolates, as shown in table No. 3.

E.coli showed highest resistance to Gentamicin (75% resistance). Tazocin ,Cefoperazone + sulbactam and a Meronem showed no resistance to any of the available isolates of E.coli. as shown in table No. 4.

Table No.01

Sites of surgical wound infection.

Site	Frequency	Percent
Head and Neck	15	20%
Upper limb	19	25.3%
Chest	8	10.7%
Abdomen	3	4.0%
Perineum	4	5.3%
Lower limb	25	34.7%
Total	75	100%

Table No. 2: Microorganisms isolated from surgical wound infection.

Microorganism	Frequency	Percent
Staphylococcus aureus	25	33.3
E.Coli	15	20.0
Pseudomonas species	10	13.3
Proteus species	8	10.7
Mixed organisms	8	10.7
Providentia	6	8.0
Enterobacter	1	1.3
Fungus	1	1.3
Citrobacter	1	1.3
Total	75	100

Table No. 3: Antibiotics sensitivity and resistance of S.Aureus

Antibiotic	Sensitivity n(%)	Resistance n(%)
Amoxicillin	0(0)	25(100)
Ampicillin	0(0)	25(100)
Co-Amoxiclav	11(44)	14(56)
Cephradine	10(40)	15(60)
Cefuroxime	8(31)	17(69)
Fucidic acid	21(84)	4(16)
Vancomycin	25(100)	0(0)
Linezolid	25(100)	0(0)

Table No. 4: Antibiotics sensitivity and resistance of E.Coli

Antibiotic	Sensitivity n(%)	Resistance n(%)
Gentacin	4(25)	11(75)
Amikacin	9(60)	6(40)
Ciprofloxacin	9(60)	6(40)
Tazocin	15(100)	0(0)
Cefoperazone/Sulbactam	15(100)	0(0)
Meronem	15(100)	0(0)

Table No. 5: Antibiotics sensitivity and resistance of Pseudomonas Sepsis

Antibiotic	Sensitivity n(%)	Resistance n(%)
Co-Amoxiclav	0(0)	100(100)
Amikacin	9(90)	100(100)
Cefoperazone/Sulbactam	7(70)	3(30)
Imipenem	8(80)	2(20)
Piperacillin/Sulbactam	8(80)	2(20)

Table No. 6: Antibiotics sensitivity and resistance of Proteus Sepsis

Antibiotic	Sensitivity n(%)	Resistance n(%)
Co-Amoxiclav	0(0)	100(100)
Amikacin	2(25)	6(75)
Cefoparazone/Sulbactam	6(75)	2(25)
Meropenem	7(87.5)	1(12.5)
Piperacillin/Sulbactam	7(87.5)	1(12.5)
Piperacillin/Tazobactam	7(87.5)	1(12.5)

Pseudomonas species was the third most common organism isolated and was highly resistant to Co-amoxiclav. Amikacin on the other hand had very good sensitivity (90%), followed by Meronem and Cefoperazone/sulbactam (see table 5).

Proteus species was isolated in 10.7% cases. No case was sensitive to Co-amoxiclav, while the organism was sensitive toAmikacin in 25% cases. Meronem, Piperacillin/Sulbactam and Piperacillin/tazobactam were the most effective drugs against Proteus species (table 6).

DISCUSSION

A wound is the disruption of the skin or mucosa_one of the major obstacles to the establishment of infections by pathogens. When bacteria breach this barrier, infection can result. Surgical site infections (SSI's) are associated not only with increased morbidity but also with mortality. Seventy seven percent of the deaths of surgical patients are related to surgical wound infections. Kirkland et al. calculated a relative risk of death of 2.2 attributable to SSI's[15].

Wound infection rates (according to clinical wound classification) are 5.9% in clean wound, 10.7% in clean contaminated wound, 24.3% in contaminated wounds and 52.9% in dirty wounds. The control of wound infections has become more challenging due to widespread bacterial resistance to antibiotics and to a greater incidence of infections caused by MRSA and polymicrobial flora[16].

Our study revealed that Staphylococcus aureus is the leading etiological agent of wound infection present in 33.3% cases a bit higher than the findings of Twum-Danso K, Giacometti et al. Anguzu JR et al and Akinjogunla et al. Twum-Danso K found Staphylococcus aureus as the most common organism present in 23.7 % cases followed by E. coli in 16.9 % cases. . In a study conducted by Anguzu JR et al. staphylococcus aureus was isolated in 45.1% cases. Similar study by Akinjogunla et al reported staphylococcus aureus in 37.8% as the most common organism responsible for wound infection[17, 18].

The study by Oguntibeju OO et al observed pseudomonas as the most common organism (33.3%) followed by staphylococcus aureus (21.7 %)[9].

Nayak documented 16.8% prevalence rate for Pseudomonas aeruginosa and 5.6% for staphylococcus aureus. These differences could be due to geographical location and hygienic measures[117].We further evaluated suitable antimicrobial therapy for the common bacteria. In this study we found that Staphylococcus aureus was resistant to Ampicillin, Amoxicillin in all cases. Staphylococcus aureus has emerged as the most common organism resistant to antibiotics. Previously Vancomycin was used occasionally due to high cost but since 1980's its use increased due to the emergence of MRSA[19].

In this study Vancomycin, Fucidic acid and Linezolid were found highly effective against Staphylococcus aureus. Our results are consistent with those of Latif et al., Lacy et al. and Kouda et al. in case of Vancomycin but slightly different regarding Fucidic acid. They detected no resistance against Fucidic acid while in our case the resistance was 16% to Fucidic acid. Other studies conducted by Bukhari et al., Akiyama Trakulsomboon et al. and Tenover et al. also reported similar resistance against Fucidic acid. Our results regarding Vancomycin are also in agreement with Pirnay J et al. who also found the sensitivity of Staphylococcus aureus to Vancomycin as 100%. On the other hand, Co-amoxiclav was more effective against Staphylococcus aureus (81.7% sensitivity) as compared to our results in which the sensitivity is only 44%[20,21].

A special feature of this study is that we found Linezolid as one of the most effective agent against Staphylococcus aureus.

In this study E.coli showed no resistance to tazocin, cefoperazone+sulbactam and Meronem. Gentamicin was the least effective drug against E.coli with a resistance of 75%.Giacometti A et al. found that E.coli was sensitive to Ceftriaxone, Ciprofloxacin and Imipenem in 100% cases while Ampicillin was effective in 43.9% cases. Saini S et al. reported Cefotaxime, Amikacin and ciprofloxacin as the most effective antibiotics for E.coli. Cefotaxime revealed 77.7% sensitivity followed by Amikacin 74% and ciprofloxacin 44.4%[22].

Imipenem and amikacin should be used as the first line antibiotics for empirical treatment of post-caesarean wound infection in our centre while awaiting the result of wound swab microscopy culture and sensitivity.[23] . Imipenem was the most effective antimicrobial agent (100% effective) followed by Amikacin in 94.7% cases .We found Amikacin and Ciprofloxacin equally effective against E.coli (60% sensitivity).

*Pseudomonas aeruginosa*is a Gram-negative rod, widely known as an opportunistic organism, frequently involved in infections of immunosuppressed patients, and also causes outbreaks of hospital-acquired infections.

In our study pseudomonas auruginosa was the third common organism present in 13.3% cases. These findings are almost consistent with those of Anguzu JR et al. who reported pseudomonas auruginosa in 9.9% cases. Compared to several earlier reports on wound colonization and invasive infection, one of the most striking differences is the frequency of pseudomonas in this study, which is contrary to findings in other studies in which pseudomonas aeruginosa formed a large number of total isolates. Shankar et al. reported Pseudomonas auruginosa in 31.84% cases while Giacometti et al. found pseudomonas auruginosa in 25.2% cases. Similarly Oguntibeju OO et al. found pseudomonas aeruginosa in 33.3% cases[9]. Shankar et al. found that Pseudomonas aeruginosa showed sensitivity to Amikacin ,Meropenem and Cefoperazone/sulbactam in 62.3%,55.8%,73.9% cases respectively[24] .The majority of the *P. aeruginosa*isolated (85.7%) in a study conducted by Anguzu JR et al. were sensitive to both Gentamicin and Ceftazidime while 42.8% were sensitive to Ciprofloxacin. Oguntibeju OO et al. reported that Pseudomonas aeruginosa is sensitive to Polymyxin E in 100% cases, followed by Gentamicin in 75% cases while no isolate was sensitive to Ampicillin. Bedenic B et al. found that almost 20% of Pseudomonas aeruginosa strains were resistant to Imipenem, 13% to Meropenem, 69% to Gentamicin and 38% to ciprofloxacin. Pirnay J et al. reported Imipenem/cilastatin as the most effective antimicrobial agent against pseudomonas species (85% sensitivity). These findings are similar to our study. In our study pseudomonas was sensitive to Amikacin in 90% cases while Pirnay JP et al. reported low sensitivity as 45.5%[25].

Proteus mirabilis is a well-known cause of clinically significant infection, and several outbreaks of nosocomial infection with this species have been described. Proteus species accounted for 10.7% of isolates in our study. Nagoba BS et al. found Proteus in 18.4% cases of wound infections. Thanni LO et al. studied wound organisms and Proteus was present in 15.1% cases. Empel et al. reported the prevalence of Proteus mirabilis as 14.5%.[26,27] .Our study shows 87.5% sensitivity to Meropenem, Piperacillin/tazobactam and Piperacillin/sulbactam. According to Bedenic B et al., Proteus mirabilis is 100% sensitive to Imipenem. Mohanty et al. reported Piperacillin/tazobactam, Cefoperazone/sulbactam and

Ticarcillin/Clavulinic acid as the most effective drugs against Proteus species. The sensitivity of Proteus to these antimicrobials was 91.3%, 91.8% and 77.1% respectively.

Citrobacter is a genus of gram-negative, facultatively anaerobic, rod-shaped bacteria of the family Enterobacteriaceae. The various species of Citrobacter include C. freundii, as the most common, followed by C. koseri and C. braakii. Our study showed Citrobacter species as 1.3% of total isolates which is consistent with the findings of Empel et al. In our study, Citrobacter was sensitive to ciprofloxacin and Enoxacin.*Mohanty et al. found that Citrobacter is sensitive to Piperacillin/ Tazobactam, Cefoperazone/Sulbactam and Ticarcillin/Clavulinic acid in 80.6%, 79.5% and 49.6% respectively*. Samonis G et al. studied 38 consecutive Citrobacter isolates. Most active antimicrobial agents against Citrobacterwere Colistin (100%), Fosfomycin (100%), Imipenem (97.4%), Gentamicin (89.5%), Nitrofurantoin (89.5%), ciprofloxacin (80.6%), and cefepime (73.7%).Khorasani G et al. , reported the highest sensitivity for Ceftazidime and Cefepime (81.6% and 78.4%, respectively)[28].

CONCLUSION

Common organisms isolated from surgical site infections are Staphylococcus aureus, Escherichia coli and Pseudomonas species. Staphylococcus aureus is resistant to Penicillins. Isolates of staphylococcus aureus are sensitive to Vancomycin and Linezolid. Meropenem, Vancomycin and Linezolid can be used as good empirical antimicrobial agents in case of complicated surgical wound infections.

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