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Bacteriological Profile of Post Operative Orthopedic Implant Infections and Their Antibiotic Susceptibility Pattern in a Tertiary Care Hospital

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Abstract:
Background: Orthopedic implant site infection is one of the major constituent of surgical site infection associate with high morbidity and mortality. Incidence of orthopedic implant infection varies from 0.5-2% in fixation of closed fractures and 30% after fixation of open fractures. Infection can occur by infected implant, aseptic failure, predisposing patient conditions, biofilm production on implant by bacteria.
Methods: A prospective study was carried out on the patients who had undergone orthopedic implant surgeries and present with signs and symptoms of implant site infections admitted a tertiary care hospital in Punjab from January 2020- June 2021. The aim of this study was to determine the bacteriological profile of orthopedic implant site infections, to study the antimicrobial susceptibility pattern and biofilm production by bacterial isolates.
Results: The present study reported more than 80% culture positivity of orthopedic implant-site infections. The most common bacteria which were isolated included Escherichia coli, followed by Pseudomonas aeruginosa and Klebsiella. A majority of them are resistant to the commonly used antibiotics, leading to treatment failures which necessitated an implant removal.
Conclusions: The practice of starting empirical therapy to be avoided and the routine identification of bacteria through culture methods and conducting drug susceptibility testing should be practiced in addition to direct microscopy as a routine diagnostic procedure especially in the centres where orthopedicians have access to microbiology lab facilities.

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I. Introduction:

Surgical site infection (SSI), as defined by US Center for Diseases Control (CDC) is an infection occurring within 30 or 90 days after a surgical operation (or within 1 year if an implant is left in place after procedure) and affecting either incision or deep tissues at the operation site.[1]Orthopedic infections are one of the commonest which can occur in approximately one percent of all orthopedic operations. Incidence of SSI is reported to vary from 3.6% to 22.5% and the implant infection varies from 0.5-2% in fixation of closed fractures to 30% after fixation of open fractures.[2] Implant surgery has become one of the commonest orthopedic operations, because of the success of this procedure in restoring function to the affected joint.According to Trampuz and Zimmerli, less than 10% of prosthesis recipients develop implant-associated complications during their lifetime, predominantly as aseptic failure and bacterial infection has been a significant part of this complication implicated in 22% of revision operations.[3]

Implants being foreign bodies compromise the local host defence mechanisms and become highly susceptible to microbial infections. Microorganisms rapidly adhere to the implant and resist elimination, which leads to the most disastrous consequences. Numerous challenges associated with these infections include the need for multiple surgeries, long periods of disability for the patient, and occasionally sub-optimal outcomes. Due to ineffective means of definitive diagnosis, inappropriate use of antibiotics and hence drug resistance as well as the ability of microorganisms to evade the host response through biofilm formation, treatment of orthopaedic implant infections is often inadequate, leading to chronic infections and other complications.[4] The isolates and the pattern of their susceptibility and resistance to the antibiotics are continuously changing. So,

periodic survey of isolation and the susceptibility pattern of isolates, in patients with implant infections become essential.

This study was conducted on patients with orthopaedic implant infections admitted in orthopaedic wards, of a tertiary care hospital in Punjab from January 2020 to June 2021. The purpose of this study was to isolate the bacterial pathogens causing implant infection, know their resistance to various antimicrobials and detection of biofilm production

II. Material And Methods:

The present prospective study was carried out in a tertiary care hospital in Punjab for a period of 18 months from January 2020 to June 2021. The patients who had undergone an implant surgery and presented with signs and symptoms of implant-site infections were included in the study. The study was conducted after obtaining the ethical clearance from the institution ethical committee and informed consent from the patients. The demographic data like age, sex, type of surgery, type of implant, time of infection, co-morbidities and other risk factors were recorded.

Inclusion criteria:

1. Patients with implant infections in orthopaedic wards
2. Patients of all age groups and both sexes with implant infections
3. Patients with open/ closed fracture
4. Surgeries including emergency and elective procedures

Exclusion criteria:

1. Implantation done through already infected wound
2. Patients on antibiotics, long term steroids and immunosuppressive therapy
3. Patients with chronic illness like tuberculosis, malignancy, etc.

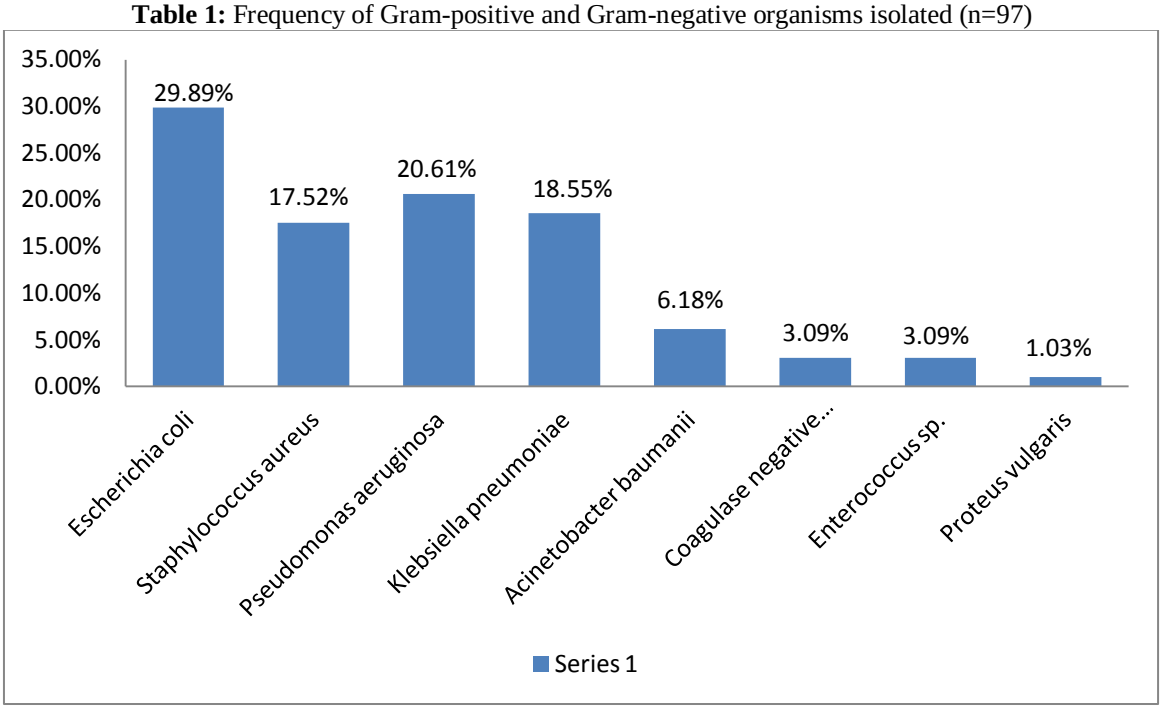
Data collection: Data collection including name, age, sex, address, date of admission, risk factors like old age, diabetes mellitus, tissue 18 damage, anemia, chronic osteomyelitis, renal failure, malnutrition, concurrent UTI, smoking and hypertension was recorded.

Sample collection and transportation: The pus samples was collected aseptically from patients presented with clinical evidence of infection (purulent discharge from incision site or drain or implant) using two sterile cotton swabs from surgical site after cleaning the surrounding area with sterile normal saline without touching the wound to prevent contamination by skin commensals. The samples were immediately transported to the Microbiology laboratory in appropriate sterile containers. Processing of specimen:55 All the specimens were processed for Gram-staining and aerobic bacterial culture by inoculating the specimen on blood agar, MacConkey agar and brainheart infusion broth and incubated at 37^{o}C for 24-48 hours. All positive cultures were identified by their characteristic appearance on the media, Gram- staining and were further identified by standard microbiological tests.[5]

III. Results:

A total of 120 samples were obtained from patients of implant site infections who attended the OPD/IPD of orthopedics department of a tertiary care hospital in Punjab and subjected to microbiological processing during the study period. Following observations were noted: Out of 120 specimens when processed, 97(80.83%) showed growth on culture. The most common organism isolated was found to be Escherichia coli 29(29.89%), followed by Pseudomonas aeruginosa 20(20.61%), Klebsiellapneumoniae 18 (18.55%), Staphylococcus aureus 17 (17.52%). (Table 1).Gram-positive isolates showed 100% resistance to azithromycin and cephalexin, 59.47% resistance to Ciprofloxacin, 47.05% resistance to Amoxicillin, 29.41% resistance to penicillin. All isolates were sensitive to Amoxacillin + Clavulanic acid, Linazolid and Vancomycin. (Table 2).Among Gram-negative organisms, 70% isolates showed resistance to Cefotaxime-Clavulanic acid, 67.01% to Ampicillin, 65.78% to Cefotaxime, 63.87% to Ciprofloxacin, 63.10% to Ceftazidime-Clavulanic acid, 53.50 % 58 to Ceftazidime, 50.02% to Piperacillin + Tazobactam, 29.45% to Amoxicillin, 14.53% to Gentamycin and 4.13% to Imipenem. (Table 3) 67 (69.07%) isolates were found to be biofilm producers by tube method while 68 (70.10%) were by congo red agar method.

Table 1: Frequency of Gram-positive and Gram-negative organisms isolated (n=97)



The antibiotic resistance pattern of Gram positive isolates was found to be:

Table 2: Antibiotic resistance pattern of Gram-positive isolates (n=26)

Gram positive isolate	Cotrimoxazole	Penicillin	Gentamycin	Amoxicillin	Amoxicillin &Clavulanic acid	Azithromycin	Cephalexin	Ciprofloxacin	Linizolid	Vancomycin
Staphylococcus aureus (n=17)	41.17%(7)	88.23% (15)	58.82%(10)	41.17% (7)	0% (0)	100% (17)	100% (17)	11.76% (2)	0% (0)	0% (0)
Coagulase negative *staphylococci (n=6)*	0% (0)	0% (0)	0% (0)	66.66% (4)	0% (0)	100% (6)	100% (6)	66.66% (4)	0% (0)	0% (0)
Enterococcus (n=3)	0% (0)	0% (0)	0% (0)	33.33% (1)	0% (0)	100% (3)	100% (3)	100% (3)	0% (0)	0% (0)

The antibiotic resistance pattern of Gram-negative isolates was found to be:

Table 3: Antibiotic resistance pattern of Gram-negative isolates (n=71)

Gram negative isolate	Ampicillin	Amoxicillin &Clavulanic acid	Gentamycin	Ciprofloxacin	Ceftazidime	Cefotaxime	Ceftazidime& Clavulanic acid	Cefotaxime& Clavulanic acid	Pipericillin&Tazobactam	Imipenem
Escherichia coli (n=29)	51.72% (15)	34.48% (10)	31.03% (9)	48.27% (14)	44.82% (13)	51.72% (15)	65.51% (19)	100% (29)	37.93% (11)	20.68% (6)
Pseudomonas aeruginosa(n=20)	50% (10)	35% (7)	25% (5)	60% (12)	45% (9)	55% (11)	100% (20)	50% (10)	40% (8)	0% (0)
Klebsiella pneumonia(n=18)	100% (18)	44.44% (8)	16.66% (3)	44.44% (8)	44.44% (8)	55.55% (10)	50% (9)	100% (18)	38.88% (7)	0% (0)
Acinetobacterbaumanii (n=3)	33.33% (1)	33.33% (1)	0% (0)	66.66% (2)	33.33% (1)	66.66% (2)	100% (3)	100% (3)	33.33% (1)	0% (0)
Proteus vulgaris (n=1)	100% (1)	0% (0)	0% (0)	100% (1)	100% (1)	100% (1)	0% (0)	0% (0)	100% (1)	0% (0)

IV. Discussion

Orthopedic implant site infections continue to be a diagnostic and therapeutic challenge. It is much more complicated by the formation of biofilm leading to burden in antibiotic selection and prolonged antimicrobial therapy due to emergence of multidrug resistant pathogens.

In the present study, out of 120 pus samples received from patients with orthopedic implant infections, the culture positivity was found to be 80.83% (97 cases) which is similar to other studies on implant site infection. Anisha Fernandez et al reported 84%[6] and Khosravi et al and Vishwajith et al reported a culture positivity of 93.9%[7] and 94.89%[8] respectively. The incidence of culture positivity of 80.83% in the present study is relatively high. Our findings are in contrast to a study done by Lakshminarayan et al which reported a culture positivity of only 45.13%.[2]

Out of 97 culture positive cases, the most common pathogen isolated was found to be *Escherichia coli* 29(29.89%), followed by *Pseudomonas aeruginosa* 20(20.61%), *Klebsiella pneumonia* 18 (18.55%), *Staphylococcus aureus* 17 (17.52%), CONS 6(6.18%), *Enterococcus sp* 3(3.09%), *Acinetobacterbaumanii*3 (3.09%)and *Proteus vulgaris* 1(1.03%) (Figure 10). Our findings correlate with a study by Okoro K et al that identified Escherichia coli (40%) as the most common cause of ORDIs in orthopaedic implant surgery, followed by *Klebsiellapneumoniae* (33%) and Staphylococcus aureus (33%).[9] Benazir et al found *Staphylococcus aureus* (37.1 %) to be the most common Gram- positive isolate followed by CoNS (coagulase-negative Staphylococcus (10.3%) and Enterococcus (7.2%). [10] It is noteworthy that ten years ago, *Staphylococcus aureus* was the most common cause of implant-associated infection. The emergence of gram-negative bacteria may be due to the preferential use of Amoxicillin-Clavulanic acid as the first-choice prophylactic antibiotic within the last few years.[9]

In 1950's and 60's *Staphylococcus aureus* used to be the most common strain. In late 70's there was a shift from gram positive infections to gram negative infections among orthopedic patients. *Staphylococcus aureus* is an important pathogen isolated in such infections. Patients, as well as healthy people, can carry this organism in their nasal passages. Healthcare workers colonized by *Staphylococcus aureus* act as a medium in transmitting this organism to patients. Hospital supplies like bed linen, instruments and dressings have also been found to act as reservoirs of these organisms.[10]CONS is a normal skin commensal which could have reached the surgical site due to improper disinfection of the skin during surgery or it might be due to improper collection of sample for the diagnosis.[11]

Khosravi et al also found aerobic Gram-negative bacilli as the most prevalent microorganisms (64.5%)[7]Lakshminarayan et al reported *Escherichia coli* as a commensal of gut and as many patients are admitted for prolonged periods, contamination of wounds, dressings, linen, clothes and hands during perineal hygiene plays a major role in increasing chances of transmission of infection.[2].In our study *Pseudomonas aeruginosa* (20.61%) was found to be the second most common cause. Pseudomonas can multiply on common objects in hospital environment such as dressings materials, buckets used for soaking Plaster of paris bandages and foreceps.[12]

Infections are a frequent cause of implant failure and the mainstay of treatment is the administration of appropriate antibiotics with or without the removal of implants. The antimicrobial susceptibility testing of the organisms isolated in our study showed variable results. Gram-positive isolates showed 100% resistance to azithromycin and cephalexin, 59.47% resistance to Ciprofloxacin, 47.05% resistance to Amoxicillin, 29.41% resistance to penicillin, 19.60% resistance to Gentamycin, 13.72% resistance to Cotrimoxazole and 0% resistance to Amoxacillin + Clavulanic acid, Linazolid and Vancomycin.. Similar pattern was shown in a study by Chandrika S et al which showed highest resistance to Penicillin (40%), Ciprofoxacin (60%), Cotrimoxazole (15%) Gentamicin (20%).Amoxyclav, Linezolid and Vancomycin did not show any resistance.[13] In a study by Roopashree et al the Gram positive bacteria showed 100% sensitivity to Linezolid, and Vancomycin.[11] Methicillin resistance was found in 11.76% of isolates which correlates with a study by Roopashree et al who found 13.33% of S. aureus to be Methicillin resistant (MRSA)[11] and a study undertaken by Anisha et al which found 12.7% to be MRSA.[6]

Gram-negative organisms also showed variable resistance patterns in our study. 70% showed resistance to Cefotaxime-Clavulanic acid, 67.01% to Ampicillin, 65.78% to Cefotaxime, 63.87% to Ciprofloxacin, 63.10% to Ceftazidime-Clavulanic acid, 53.50 % to Ceftazidime, 50.02% to Piperacillin + Tazobactam, 29.45% to Amoxicillin, 14.53% to Gentamycin and 4.13% to Imipenem.. A study by Chandrika S et al also found among Gram negative bacilli highest resistance to Ampicillin (70%), Ciprofloxacin (60%),Cefotaxime (55%), Ceftazidime (45%), PiperacillinTazobactum (30%), Gentamicin (10%) and Imipenem (5%).[13] Another study by Jyoti et al found 100% to be sensitive to imipenem and 70 % sensitive to nitrofurantoin. However it was found resistant to ceftriaxone (90%), Ciprofloxacin (80%) and cotrimoxazole (80%). Amikacin and gentamicin showed good sensitivity (70 %).[14]

Biofilms play a pivotal role in healthcare associated infections (HAIs), especially those related to the implantation of medical devices. A worrying feature of biofilm-based infections is represented by the higher

antibiotic resistance of bacterial cells growing as biofilms as compared with planktonic cells. Biofilm-associated bacterial infections are difficult to eradicate using antibiotics. It has been observed that biofilm makes the micro-colonies impermeable to antibiotics, hydroxyl radicals, and superoxide anions which bind at the outer surface of the matrix layer of the biofilm.[15]67 (69.07%) isolates were found to be biofilm producers by tube method while 68 (70.10%) were by congo red agar method. These findings correlate with a study performed by Roopashree S et al which found that 72% isolates were biofilm producers by Congo Red Agar method, and 76% were biofilm producers by Tube method.[11]

Orthopaedic device related infection (ODRI) is a diagnostic and therapeutic challenge which can pose a serious threat to the patient leading to high morbidity. This study has given us a wide knowledge about ODRI and their incidence in our hospital and also helps us in finding out the bacteriological profile of organisms causing implant infection and their sensitivity pattern.

V. Conclusion

To prevent implant associated infections appropriate pre and postoperative wound care for the dirty wounds should be done with more caution. The practice of starting empirical therapy to be avoided and the routine identification of bacteria through culture methods and conducting drug susceptibility testing should be practiced in addition to direct microscopy as a routine diagnostic procedure especially in the centres where orthopedicians have access to microbiology lab facilities. To mitigate the burden of bacterial orthopedic implant site infections, physicians should regard on risk reduction and comply with etiologic approach of diagnosis.

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