Microbiological Etiology of Prosthetic Joint Infections

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Abstract

Introduction: *Infection of the prosthesis remains one of the most devastating complications of arthroplasty surgery. Identification of the likely cause of early-onset PJI is particularly important given that these infections are more frequently treated with a debridement procedure where the implant is not removed.*

Aim and objectives: *To identify the microbiological aetiology of prosthetic joint infections and to identify the antimicrobial susceptibility pattern in order to propose the treatment guidelines of PJI based on the duration of onset of symptoms.*

Material and methods: *Synovial fluid collected preoperatively, periprosthetic tissue and the removed prosthesis were processed following standard laboratory protocols set up by the Mayo Clinic.*

Results: *The primary gram stain from culture positive samples was positive in 81/111 (72.97%) samples. Gram negative bacteria constituted majority of the isolates (57/75) (76%) identified from early onset of infection of which Acinetobacter baumannii was the commonest and was significantly associated with early onset infection, while MRSA was associated significantly with delayed onset infection and CONS was significantly associated with late onset infection.*

Discussion: *This study highlights the importance of gram negative pathogens (76%) in early onset of infection, which showed a high incidence of resistance to β lactam and β lactam inhibitor combinations which might be due to the high incidence of ESBL in both community and hospital acquired infections. Multi-drug resistance among the isolates advocates the case for reconsideration of existing protocols for surgeries involving prosthesis which might potentially decrease the incidence of PJIs and reduce patient morbidity and mortality. Treatment modalities can be determined based upon the database generated in individual institutions*

Keywords :*Prosthetic joint infection, biofilms, microbial flora, treatment guidelines for PJI*

I. Introduction

Infection of the prosthesis remains one of the most devastating complications of arthroplasty surgery. Prosthetic joint infections are uncommon (1% to 3%); however, they are associated with significant morbidity for patients and with health care costs.[1,2]A PJI is defined as isolation of the same microorganism from at least two cultures of joint aspirates or intraoperative tissue specimens, or as isolation in at least one intraoperative culture of microorganisms, plus evidence of infection at the site of hip or knee prosthesis (presence of a discharging sinus communicating with the joint, operative findings of purulence, or positive laboratory and histopathological tests [3]. The risk factors for prosthetic joint infection include obesity (body mass index >35), diabetes mellitus, rheumatoid arthritis, malignancy, arthroplasty revision surgery and a prolonged procedure duration. The composite risk scores attempt to aggregate a number of factors into one, more easily applied variable. The National Nosocomial Infections Surveillance (NNIS) System surgical score includes the length of the surgical procedure, the American Society of Anesthesiologists (ASA) preoperative assessment score, and surgical wound classification for each procedure An elevated ASA score alone, estimating the burden of systemic disease, has also been associated with an increased risk of infection [4- 7]. The Mayo PJI score, while not fully validated, is a numerical score to predict PJI based on assessment at the time of joint arthroplasty implantation or 1 month later [8].

The majority of PJIs occurring within 1 year of surgery are initiated through the introduction of microorganisms at the time of surgery. This can occur through either through direct contact or aerosolized contamination of the prosthesis or periprosthetic tissue. Once in contact with the surface of the implant, microorganisms colonize the surface of the implant. A significant factor in this process is the low inoculum of microorganisms needed to establish infection in the presence of the prosthetic material which has been

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explained by the formation of biofilms by the pathogen. In the biofilm state, bacteria are protected from antimicrobials and the host immune system [9], making treatment of infection difficult without a biofilm-directed treatment strategy, which today mandates surgical intervention, in many cases including prosthesis removal, to achieve a cure. The reduced antimicrobial susceptibility of bacteria in biofilms is related to their low growth rate, the presence of resistant bacterial subpopulations (so-called "persisters"), and a microenvironment within the biofilm that impairs antimicrobial activity [10, 11].

The different diagnostic methodologies proposed for diagnosis of PJI include peripheral blood tests for white blood cells, C reactive protein, ESR, procalcitonin and IL-6 (shows the highest sensitivity and specificity among these tests). Synovial fluid analysis for nucleated cell counts and neutrophil differentiation, leukocyte esterase markers has been showed to be of importance in the diagnosis. In addition to informing the diagnosis of PJI, preoperative synovial fluid culture is invaluable for early identification of the infecting pathogen(s) and determination of antimicrobial susceptibility. This information can inform the choice of perioperative antimicrobials and construction of antimicrobial-loaded polymethylmethacrylate (PMMA) and may impact the selection of a treatment strategy, if a particularly sensitive or resistant pathogen is present. Besides this other diagnostic methods like Preoperative periprosthetic tissue biopsy, Intraoperative periprosthetic tissue Gram staining, Intraoperative periprosthetic tissue culture can be performed. Cultures obtained by using swabs have a limited role in the microbiological detection of PJI. While the presence of a sinus tract is considered definitive evidence of PJI [12], swab culture of the drainage from the sinus tract is neither sensitive nor specific for the microbiological detection of PJI. Intraoperative cultures obtained via swabs are less accurate than tissue cultures.Sonication has emerged as a practical and effective method to dislodge biofilm and the associated bacteria from the surface of the implant. Vortexing of the prosthesis alone may be a viable alternative in laboratories in which sonication is not available. *In vitro* data suggest that vortexing alone can remove bacteria from biofilm-coated coupons [12]. Portillo and colleagues prospectively compared the results of vortexing alone to the results of vortexing plus sonication of 135 removed prostheses [12]. Using a lower cutoff of 1 to 10 CFU per milliliter, in which a centrifugation step is not used demonstrated that vortexing of the prosthesis alone may be reasonable in laboratories that do not have the equipment or personnel to perform a full sonication protocol. [13]Identification of the likely cause of early-onset PJI is particularly important given that these infections are more frequently treated with a debridement procedure where the implant is not removed as because the treatment for PJI can be divided into two main groups: (A) prosthesis removal with or without subsequent reimplantation and (B) debridement and implant retention using long-term antibiotics. Removal of all prosthesis components (resection arthroplasty) has a higher chance of eradicating infection but requires extensive surgery and often prolonged immobilization [14]. Debridement and retention of the prosthesis is an attractive alternative, which may be attempted in selected patients to salvage the joint prosthesis [14]. This less-extensive surgery is thought to be associated with a lower probability of procedure-related morbidity, less immobilization, and consequently less need for rehabilitation. The main problem of debridement and retention, however, is that a substantial number of patients will ultimately experience a relapse of infection after the less-aggressive procedure, necessitating exchange or resection arthroplasty [14]. Treatment remains challenging, with patients often requiring multiple surgical procedures and long-term antibiotic therapy [15], so the focus of PJI is prevention of the occurrence. Surgical antibiotic prophylaxis is one the strategies to prevent PJI's. At present, local and international guidelines recommend a single dose of cefazolin at the time of induction based on data from randomized control trials performed in the 1970s and 1980s [15]. The guidelines stipulate, however, that the antibiotics chosen as prophylaxis should be selected to cover the most frequently encountered pathogens [15].

II.   Aim And Objectives

To identify the microbiological aetiology of prosthetic joint infections and to identify the antimicrobial susceptibility pattern in order to propose the treatment guidelines of PJI based on the duration of onset of symptoms.

III.   Material And Methods

The samples collected for the diagnosis in suspected cases of PJI included the synovial fluid collected preoperatively, periprosthetic tissue and the removed prosthesis. A portion of the synovial fluid or prosthetic tissue was transported aseptically in sterile plastic bags for Gram stain and Zeihl Neelsen stain. The remaining portion of the aspirated synovial fluid collected preoperatively or Periprosthetic tissue collected intraoperatively were inoculated into blood culture bottles containing brain heart infusion broth at the time of collection and transported to the microbiology laboratory where subcultures were performed after overnight incubation onto blood agar and MacConkey agar. The plates were incubated 37°C aerobically for a minimum of 7 days with 5% CO_2. The colonies obtained were then identified by standard laboratory procedures [16] and antibiotic susceptibility performed as per CLSI guidelines [17] by the modified Kirby bauer disc diffusion technique.

In the case of implants that were removed, components were aseptically placed into a rigid sterile container, followed by the addition of 400 ml of sterile Ringer's solution. The containers were then vortexed for 30 seconds before and after, sonication for 5 minutes, followed by centrifugation of the fluid for 5 minutes. Then 1 ml of the centrifuged fluid was spread onto the plates which were processed similarly as mentionedabove. A low cutoff (any growth on agar plate, i.e., ≥1 CFU/ml) was taken as significant. [13] The bacteriological criterion was considered positive when at least one culture yielded a strict pathogen (Staphylococcus aureus, Pseudomonas aeruginosa, Enterobacteriaceae, and anaerobes) or when two cultures yielded a strain that was a skin commensal (such as coagulase-negative staphylococci [CoNS]. [18]

IV.   Results

A total of 132 samples were received during this period. Out of the samples examined, 97 (73%) were collected from male patients. The mean age of patients was 39.4 years (mean ± SD). However, no association of PJI was found with age of patient in this study. The most common clinical feature was discharge from wound site in 57 cases (43.2%) followed by pain & fever. Association of cases with diabetes mellitus as a risk factor was found to be statistically significant ($\chi2= 4.2471$, p=0.039). Most of the cases (67.6%) had an early onset of infection (<3 months), while 27% presented between 3-24 months while a few cases (5.4 %) presented after >24 months. [Table 1The primary gram stain from culture positive samples was positive in 81/111 (72.97%) samples. 111 cases (84%) gave positive cultures, while in 21 (16%), the cultures were negative. 43.2 % were Gram positive cocci and 56.8% were Gram negative bacilli. Gram negative bacteria constituted majority of the isolates (57/75) (76%) identified from early onset of infection of which Acinetobacter baumanni was the commonest and was significantly associated with early onset infection, while MRSA was associated significantly with delayed onset infection and CONS was significantly associated with late onset infection. The distribution of the other clinical isolates are shown in Table 1. MRSA was found to be significantly associated with bimalleolar and both bone upper limb prosthesis, A. baumannii was significantly associated with tibial prosthesis and *Klebsiella pneumoniae* was significantly associated with femur prosthesis PJI. [Table 2]

Table 1. Duration of onset of symptoms after Prosthesis implantation

Organism	Early Onset	Delayed onset	Late onset
	< 3 months	3-24 months	>24 months
MSSA	3 (4%)	6 (20%)	0 (0%)
MRSA	15 (20%)	12 (40%)	0 (0%)
CONS	0 (0%)	6 (20%)	**6 (100%)**
A.baumannii	24 (32%)	0 (0%)	0 (0%)
Klebsiella spp	18 (4%)	0 (0%)	0 (0%)
Proteus mirabilis	0 (0%)	3 (10%)	0 (0%)
E.coli	0 (0%)	3 (10%)	0 (0%)
P.aeruginosa	15 (20%)	0 (0%)	0 (0%)
Total	75	30	6
%	67.6%	27.0%	5.4%

(P=0.000, association significant, Fisher's exact test, SPSS ver 23)

Table 2. Table showing distribution of site of infection and organism isolated

Organism	Femur	Tibia	Bimalleolar	Humerus	Foot	BB UL	Ulna
MSSA	0 (0%)	0	0	0	3(33.3%)	0	6(100%)
MRSA	0 (0%)	6(25.0%)	12(66.7%)	0	0	12(100%)	0
CONS	9(27.3%)	0	0	3(25.0%)	0	0	0
A.baumannii	9(27.3%)	12(50.0%)	0	3(25.0%)	0	0	0
Klebsiella spp	12(36.4%)	3(12.55)	0	3(25.0%)	0	0	0
Proteus mirabilis	0 (0%)	0	0	0	3(33.3%)	0	0
E.coli	0 (0%)	0	3(16.7%)	0	0	0	0
P.aeruginosa	3 (9.1%)	3(12.5%)	3(16.7%)	3(25.0%)	3(33.3%)	0	0
Total	33	24	18	12	9	9	6
%	29.7%	21.6%	16.2%	10.8%	8.1%	8.1%	5.4%

(P= 0.001, association significant, Fisher's exact test, SPSS ver 23)

Table 3. Table showing comparison of the current available guidelines with the findings in the study and the proposed antibiotic policy in our given setup

Organism	IDSA -IV or Highly Bioavailable Oral	IDSA-Common Antimicrobials Used	Our finding	Comments
MSSA	Ceftriaxone/vancomycin/ Linezolid	1ST Gen Cephalosporin/Clindamycin	Clindamycin Resistane high	1ST Gen Cephalosporin may be used
MRSA	Vancomycin/Linezolid	Cotrimoxazole/ Doxycycline	Vancomycin and	

			Linezolid was 100% sensitive	
Enterobacte riaceae	Sensitive B Lactam/ CIP	Cotrimoxazole/any sensitive B laclam	High Ciprofloxacin & B lactam Resistance with ESBL production noted	Meropenem can be used
P.aeruginos a	Meropenem/ ciprofloxacin	Ciprofloaxacin	ciprofloxacin resistance high	Meropenem/ Amikacin can be used

V.   Discussion

This study highlights the importance of gram negative pathogens (76%) in early onset of infection. A number of difficulties occur when treating bone and joint infections caused by gram negative bacteria: they occur primarily in immunocompromised hosts and are associated with treatment failure [6–8], only a limited number of experimental models have been described [15], and randomized controlled clinical trials are hampered by the fact that most institutions do not have sufficient patients for such studies. [14] Our data suggest GN PJIs represent a substantial proportion of all PJI occurrences. The high incidence of 76% is very high as compared to other studies. [12]*Acinetobacter baumanni* was significantly associated with early onset infection indicating nosocomial origin. *Staphylococcus spp.* accounted for 35.14% of the isolates, while MRSA accounted for 10.81% of the isolates. The gram negative isolates showed a high incidence of resistance to β lactam and β lactam inhibitor combinations which might be due to the high incidence of ESBL in both community and hospital acquired infections. [19]The distribution of isolates as per the body site shows that *Acinetobacter spp.* was significantly associated with lower joint PJIs and MRSA to upper limb PJIs which might indicate a role for detection of preoperative colonization and whether eradication of these colonizers from these areas could result in the decrease in incidence of PJIs. At present, local guidelines recommend cefazolin or flucloxacillin as prophylaxis. Vancomycin is recommended only for patients colonized with MRSA, patients undergoing revision arthroplasty, patients at high risk of MRSA colonization (patients residing in a health care facility for greater than 5 days), and patients with immediate hypersensitivity to beta-lactam antibiotics. [15] The isolation of multidrug resistant organisms also raises questions regarding the effectiveness of prophylactic antibiotics which are administered routinely like in this study, the use of Vancomycin and Meropenem could theoretically have reduced the incidence of PJIs which is in concordance with other studies by Peel *et al*. Of the different clinical samples the role of preoperative synovial culture is of paramount importance as it can give information regarding the choice of perioperative antimicrobials and PMMA. The gram stain finding though not always conclusive, when positive, is very useful for initiation of treatment with a very high specificity, as indicated in this study where 100% correlation was observed between the gram stain finding and culture obtained.

Hence, to conclude, joint replacement is a life-enhancing procedure for millions of people worldwide each year. While the majority of joint arthroplasties provide pain-free function, a minority of patients will experience device failure and will require additional surgery at some point during the life of the device. Pre and peri operative preventive measures, eg: proper glycemic control in diabetics, stoppage of DMARD's one week before the surgery and prophylactic antibiotics based on the data obtained from previous PJI reports which should guide the formulation of institutional antibiotic policy . Majority of the PJI belong to early onset category. Proper aseptic techniques to prevent iatrogenic infections. Early onset-Prompt & precise antimicrobial therapy-Prosthesis may be preserved. Given the frequency with which MRSA and *Acinetobacter spp.* causes PJI, selective identification and decolonization of patients might be important. Knowledge of commonly isolated bacteria and their sensitivity pattern in a particular area aids in choice of empirical antibiotics. Multidisciplinary collaboration with orthopaedic surgeons & clinical microbiologist is needed to reduce the incidence for orthopaedic infections. There is also need for formulation of antibiotic policy based on the institutional microbiological data, as shown in the study that many of the guidelines as proposed by IDSA might not be effective in a developing country like ours (Table 3).

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