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**ORIGINAL ARTICLE****Bacteriological Spectrum of Post Operative Orthopedic Implant Infections and Their Antibiogram***Satya Chandrika V<sup>\*</sup>, Surya Kirani KRL<sup>1</sup>**<sup>1</sup>Department of Microbiology, Rangaraya Medical College, Kakinada-533001 (Andhra Pradesh) India*

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**Abstract:**

*Background:* Infection is a major problem in orthopedics leading to implant failure and in severe cases, amputation and even mortality. *Aim and Objectives:* The aim of this study is to isolate and identify organisms from postoperative Orthopaedic implant infections and determine their antibiogram. *Method and Materials:* This prospective study was conducted on 50 cases of infected implants from Orthopaedics ward, Government General Hospital, Kakinda from 1<sup>st</sup> June 2014 to 1<sup>st</sup> November 2014 after obtaining clearance from Institutional Ethics Committee. Pus samples were collected using two sterile swabs. One is used for Gram's stain and the other for inoculation on MacConkey and Blood agar. Isolates were identified according to the standard protocols and antibiotic sensitivity was done by Kirby Bauer's disc diffusion technique. *Results:* Out of 50 samples, 45(90%) were culture positive and 5(10%) were sterile for aerobic bacteria. *Staphylococcus aureus* (30%) is common isolate followed by Coagulase Negative Staphylococci (CONS) (20%), *Escherichia coli* (16%), *Klebsiella* sps (10%), *Pseudomonas aeruginosa* (6%), *Acinetobacter* species (4%), *Proteus* species (4%). Most of Gram positive cocci are resistant to Methicillin (64%) followed by Ciprofloxacin (36%) and Cotrimoxazole (28%). They were mostly sensitive to Vancomycin, Linezolid, Amoxicillin Clavulanic acid and Cefotaxime. Gram negative bacilli are resistant to Cotrimoxazole (65%)

followed by Ciprofloxacin (60%) and Ceftriaxone (60%) and sensitive to Carbapenems (80%), Piperacillin Tazobactam (60%) and Amoxicillin Clavulanic acid (60%). Extended Spectrum Beta Lactamase (ESBL) production is seen in 60% of Gram negative bacilli. *Conclusions:* The incidence of multidrug resistance pathogens as a cause of implant infections is rising. Adequate preventive measures should be enforced to prevent the spread of antibiotic resistant organisms.

**Keywords:** Orthopedic implant infections, Bacteriological profile, Antibiogram, Extended Spectrum Beta Lactamase

**Introduction:**

Infection is a major problem in orthopedic implantations leading to implant failure. It is a challenging task to treat orthopedic implant infections which may lead to implant replacement and in severe cases, amputation and even mortality. Sources of infectious bacteria include environment of the operating room, surgical equipment, clothing worn by medical and paramedical staff and resident bacteria on the patient's skin. Implant-associated infections are the result of bacterial adhesion to an implant surface and subsequent biofilm formation at the implantation site. Each hospital has its own

unique bacterial flora to which patients are at risk for acquiring infection during hospitalization. In such situations; microorganism exhibit unique pattern of antimicrobial activity during a certain period of time [1]. Since initial antibiotic therapy is empirical, it is important to know prevailing antibiotic susceptibility patterns of individual institutions by routine surveillance [2].

In the recent years the organisms isolated from these infected cases are showing increased resistance to commonly used first line antibiotics and multi-drug resistance. Methicillin resistance has become most common and most organisms isolated are also Extended Spectrum Beta Lactamase (ESBL) producers. The objectives of this study are to determine the spectrum of aerobic bacterial isolates from postoperative orthopedic implant infections and to determine their antimicrobial susceptibility patterns.

#### Material and Methods:

This prospective study was conducted on 50 cases of infected implants from Orthopedic ward, from June 2014 to November 2014 after obtaining institutional ethical committee clearance.

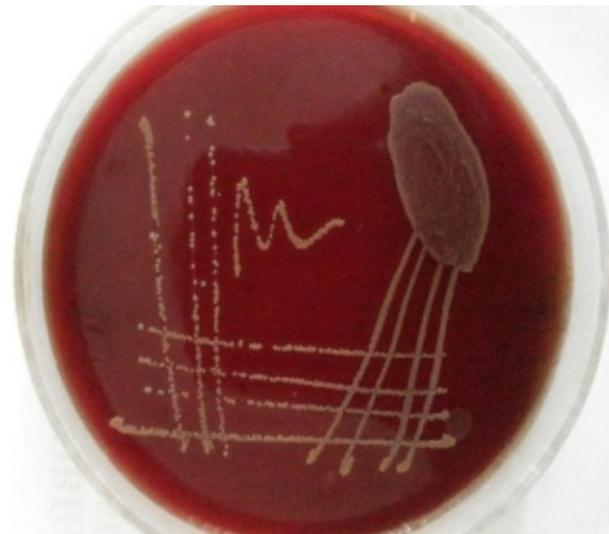
#### Inclusion Criteria:

Patients with purulent discharge from incision or drain within a week after surgery and also after few weeks after discharge from hospital of all age groups and both sexes were selected.

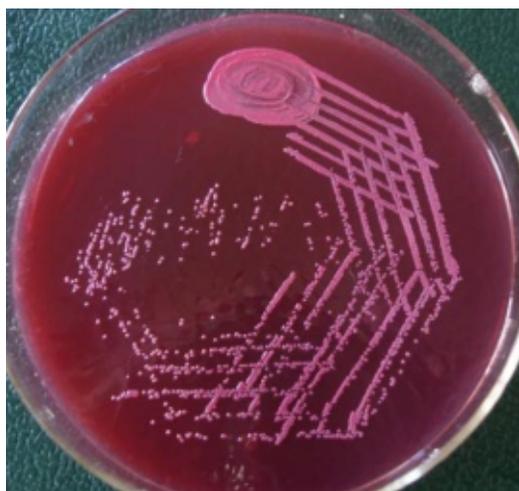
#### Exclusion Criteria:

- 1) Use of antibiotics after diagnosis of infection.
- 2) Implantation done through already infected wound.

The pus samples were collected aseptically on the first day when patients presented with clinical evidence of infection (purulent drainage from incision or drain), using two sterile cotton swabs from surgical site without contaminating with skin commensals and transported to the laboratory immediately. Specimens collected were subjected to direct microscopy by Gram's staining. For culture, the specimens were inoculated onto MacConkey and blood agar (Fig.1 and Fig. 2). Preliminary identification of bacteria was done by the colony characteristics and Gram staining. Further identification was done by a set of biochemical tests. Antimicrobial susceptibility pattern of isolated bacterial pathogens were determined by Kirby Bauer Disc Diffusion Method (KBDDM) according to CLSI guidelines [3 - 5].

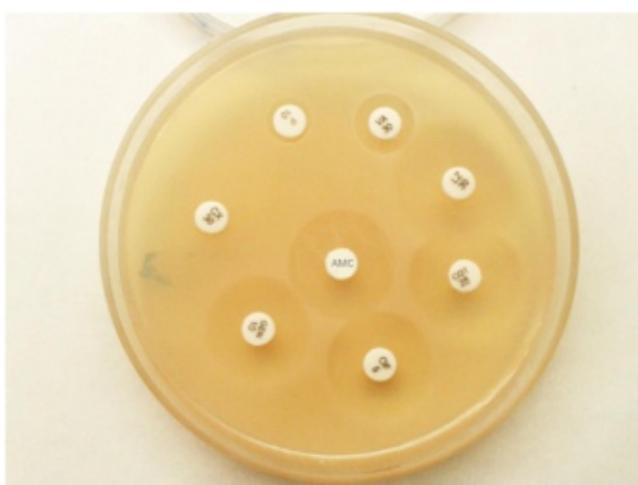


**Fig.1 Golden Yellow Colonies of *Staphylococcus aureus* on Blood Agar**



**Fig. 2: Lactose Fermenting Colonies on MacConkey Agar**

Methicillin resistance was identified by using Cefoxitin (30 µg) disc. According to the Clinical and Laboratory Standards Institute (CLSI) a zone of growth inhibition around the Cefoxitin disk –  $\geq 22$  mm was identified as Methicillin Sensitive *Staphylococcus aureus* and  $< 22$  mm was identified as Methicillin Resistant *Staphylococcus aureus* [6] (Fig.3).



**Fig. 3: Cefoxitin Resistance on Muller – Hinton Agar Plate**

Enterobacteriaceae isolates with Cefotaxime and Ceftazidime zones of inhibition less than 27 mm, 22 mm, respectively were suspected to be ESBL producing [3, 4]. A confirmatory test using the Double Disc Synergy technique was carried out according to CLSI guidelines [7] (Fig.4).



**Fig. 4: Double Disc Synergy Test on Muller – Hinton Agar Plate**

**Results:**

Out of the 50 samples, 45(90%) culture were positive and 5(10%) culture were negative. Out of 45 culture positive cases 25(55.6%) were Gram positive cocci and 20(44.4%) were Gram negative bacilli. *Staphylococcus aureus* (33.3%) was the most common isolate followed by Coagulase negative Staphylococci (22.2%), *Escherichia coli* (17.8%), *Klebsiella* species (11.2%), *Pseudomonas* species (6.7%), *Acinetobacter* species (4.4%) and *Proteus* species (4.4%). (Fig. 5)

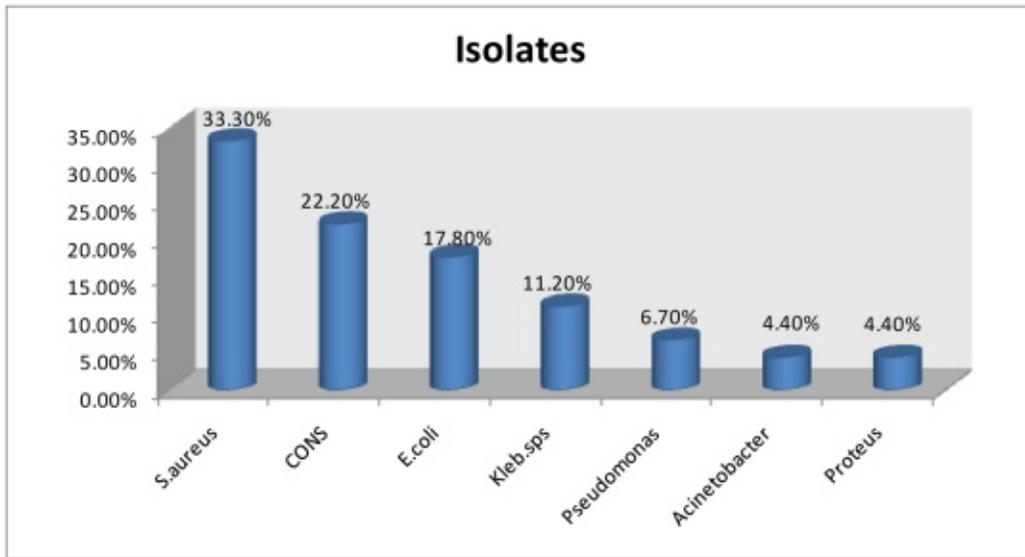


Fig. 5: Organisms Isolated from Postoperative Orthopaedic Implant Infections

Antibiogram of Gram positive cocci showed highest resistance to Cefoxitin (64%) followed by Penicillin (60%), Ciprofloxacin (36%), Cotrimoxazole (28%), Levofloxacin (16%),

Amikacin (8%) and Gentamicin (4%). Amoxyclav, Cefotaxime, Linezolid and Vancomycin did not show any resistance. (Fig. 6)

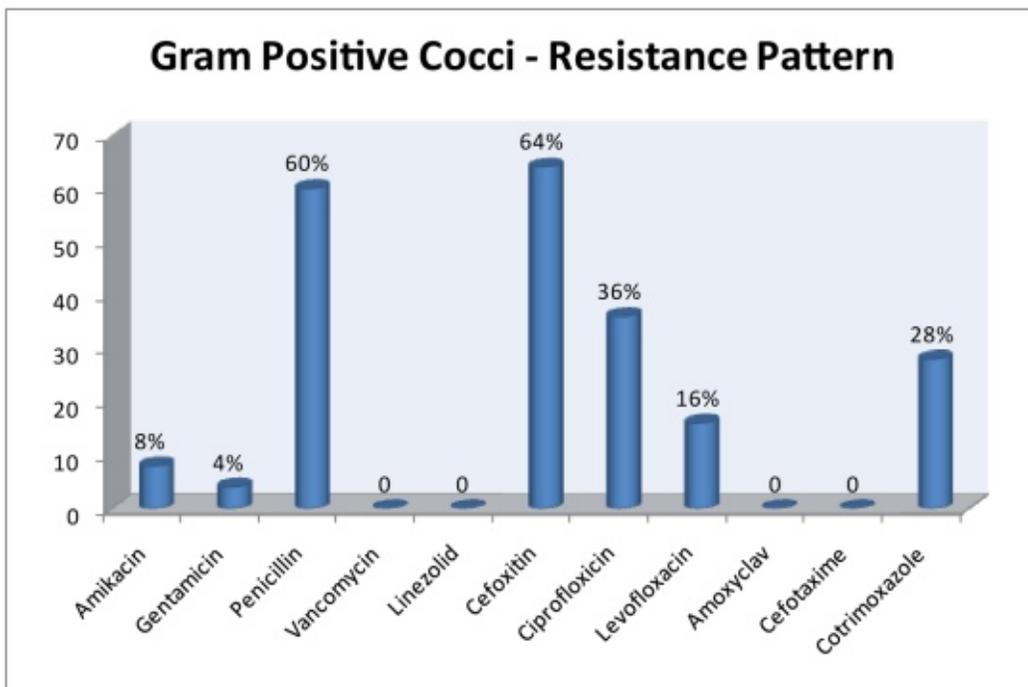


Fig. 6: Antibiogram of Gram Positive Cocci

Antibiogram of Gram negative bacilli showed highest resistance to Cotrimoxazole (65%) followed by Ceftriaxone (60%),Ciprofloxacin (60%),Cefotaxime (55%), Ampicillin (50%),

Ceftazidime (45%), Piperacillin Tazobactam (40%),Amoxyclav (35%), Imipenem (20%) and Gentamicin(10%). (Fig. 7)

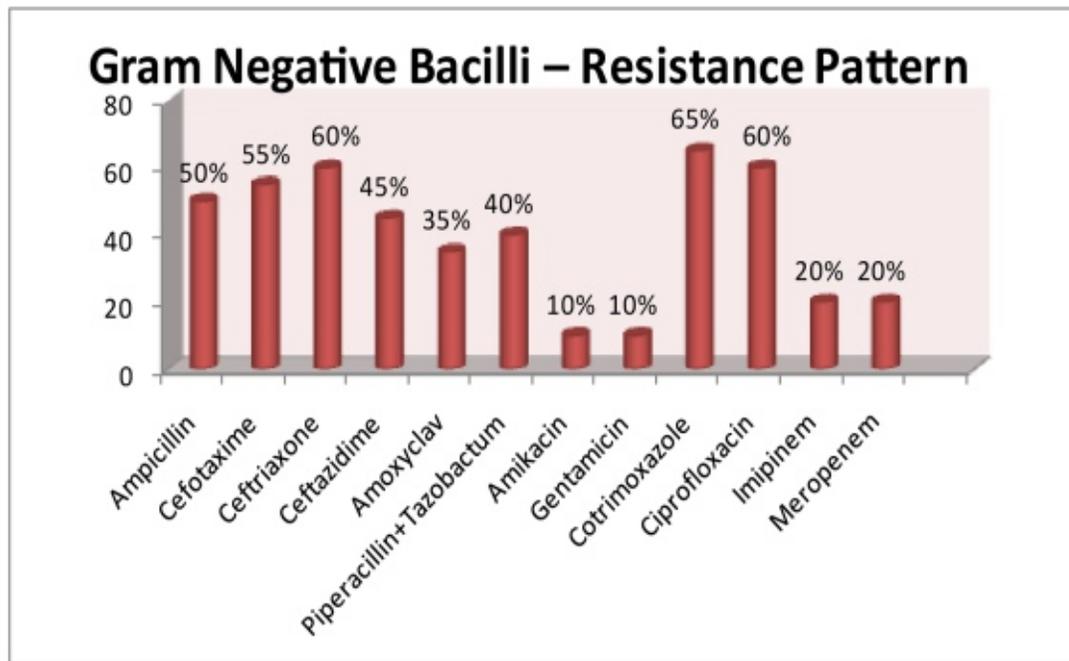


Fig. 7: Antibiogram of Gram Negative Bacilli

Methicillin resistance was seen in 66.6% isolates of *Staphylococcus aureus* and 60% isolates of *Coagulase negative Staphylococcus aureus*. ESBL production is seen in 60% of Gram negative bacilli.

**Discussion:**

The purpose of the study was to isolate the aerobic organisms causing orthopedic implant infections and to know their resistance pattern with reference to Methicillin sensitivity and ESBL production.

Out of 50 samples, 45(90%) have been culture positive and 5 (10%) sterile for aerobic bacteria. In culture positive samples Gram positive cocci (55.6%) are more than Gram negative bacilli (44.4%).

In our study *Staphylococcus aureus*(33.3%) was the most common isolate and correlates to earlier studies by Goel *et al* 2013(32.8%)[8], Sonawane *et al* 2010(29.26%) [2] and Jain *et al* 2014(26.6%) [1](Table 1).

Table 1: Comparison with Other Studies

	Sonawane <i>et al</i> 2010	Goel <i>et al</i> 2013	Jain <i>et al</i> 2014	Present Study 2014
<b>Common Isolate</b>	<i>Staphylococcus aureus</i> (29.26%)	<i>Staphylococcus aureus</i> (32.8%)	<i>Staphylococcus aureus</i> (26.6%)	<i>Staphylococcus aureus</i> (33.3%)
<b>GPC Resistance</b>	Penicillin (75.32%) Cotrimoxazole (69.9%)	Ciprofloxacin (47.5%) Cotrimoxazole (32.5%)	Penicillin (89.1%) Oxacillin (93.75%)	Cefoxitin (64%) Penicillin (60%) Ciprofloxacin (36%)
<b>GNB Resistance</b>	Ampicillin (88%) Ceftriaxone (69.8%) Ceftazidime (69.8%)	Cefuroxime (81.5%) Cefotaxime (78%)	Amoxyclav (80.8%) Ceftriaxone (76%)	Cotrimoxazole (65%) Ciprofloxacin (60%) Ceftriaxone (60%)
<b>Methicillin Resistance</b>	27.85%	30%	40%	64%
<b>ESBL Production</b>	71.72%	-	-	60%

GPC-Gram positive cocci, GNB-Gram negative bacilli, ESBL-Extended spectrum beta lactamase

In our study methicillin resistance is 64% which does not correlate with other studies and ESBL production is 60% which correlates with Sonawane *et al* 2010(71.72%) [2]. The incidence of Methicillin Resistant *Staphylococcus Aureus* (MRSA) in India ranges from 30-70% [9]. The incidence of nosocomial infections which are caused by MRSA continues to increase; therefore, the importance of their detection, especially for treatment and epidemiological purposes [9].

In our study, Gram positive cocci have shown highest resistance to Methicillin, but it showed resistance to Penicillin in studies by Sonawane *et*

*al* 2010 [2] and Jain *et al* 2014 [1] and it showed resistance to Ciprofloxacin in the study by Goel *et al* 2013 [8]. In our study Gram negative bacilli showed a highest resistance to Cotrimoxazole followed by Ciprofloxacin and Ceftriaxone, whereas it is Ampicillin followed by Ceftriaxone in the study by Sonawane *et al* 2010, Cefuroxime and Cefotaxime in the study by Goel *et al* 2013 [8] and Amoxyclav and Ceftriaxone in the study by Jain *et al* 2014 [1].

Our study has shown that *Staphylococcus aureus*, *CONS*, *Escherichia coli*, *Klebsiella* sps and *Pseudomonas aeruginosa* were the major

bacterial pathogens causing implant infections in our area. Contamination from the external environment may be a possible reason for the higher rate of Surgical Site Infections (SSI) in orthopaedic surgeries [9]. The pathogenesis of infection in fractures fixation devices is related to micro-organisms, which grow in biofilm, and therefore its eradication is difficult [10].

### Conclusion:

A high incidence of methicillin resistance and ESBL production was observed in orthopedic implant infections. The incidence of multi-drug resistant pathogens as a cause of implant infections is also rising. So, adequate preventive measures should be enforced to prevent the spread of antibiotic resistant organisms.

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