BACTERIOLOGICAL PROFILE OF OSTEOMYELITIS CASES WITH SPECIAL REFERENCE TO ANTIBIOTIC SUSCEPTIBILITY PATTERN OF ISOLATES IN A TERTIARY CARE HOSPITAL OF EASTERN INDIA

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ABSTRACT

BACKGROUND

Despite advances in medical and surgical therapies, the management of osteomyelitis is an increasing challenge to clinicians due to growing resistances to antibiotics. The present study was conducted to determine the bacteriological profile of cases of osteomyelitis and also to ascertain the antibiotic susceptibility pattern of these isolates to prevent unnecessary morbidity and mortality.

OBJECTIVE

The proposed study was carried out to isolate, identify, and characterize the bacterial strains isolated from cases of acute and chronic osteomyelitis and to ascertain the antimicrobial susceptibility pattern of these isolates.

MATERIAL AND METHODS

A total of 97 samples like pus or exudates or pieces of necrotic tissues were collected during surgery if possible, otherwise aspirated. All samples were subjected to Gram staining and culture. Various organisms were identified by standard methods. The Kirby-Bauer method was employed to perform the antimicrobial susceptibility on Mueller-Hinton agar [MHA]. For detection of methicillin-resistant Staphylococcus aureus [MRSA], MHA supplemented with 4% NaCl was used.

RESULTS

A total of 94 organisms were isolated from indoor patient department. Of the unimicrobial isolates, Escherichia coli (19.0%) were the main organism. Amongst the polymicrobial growth, the common combination was between Staphylococcus aureus and E. coli. The most common factor leading to osteomyelitis was trauma/accidents, 49.5% (48/97). S. aureus, was the common organism isolated from trauma/accident (83.3%) cases. Of the total, 30.2% (16/53) of strains were MRSA. 50% of MRSA isolates were found in patients with orthopaedic implants. Among the gram-negative bacilli, E. coli showed maximum resistance to amoxicillin (100%) followed by P. aeruginosa that showed maximum resistance to ciprofloxacin (77.7%).

CONCLUSION

Our study will thereby guide the clinicians in choosing appropriate antibiotics, which not only contribute to better treatment, but the judicious use of such antibiotics will also help in preventing emergence of resistance to the drug, which are still sensitive.

KEYWORDS

Methicillin Resistant Staphylococcus Aureus; Osteomyelitis.

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INTRODUCTION

Osteomyelitis is a serious deep bone infection with significant morbidity and high rate of recurrence. The infection caused by a variety of microbial agents can arise from a variety of aetiologies such as trauma, nosocomial infections, or after implant replacement surgery. The future era with modern high speed travel, use of implants, and prosthetics will add to the load of osteomyelitis Treatment of osteomyelitis is challenging particularly when complex multi-drug resistant bacterial biofilm has already been established.

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Bacteria in biofilm persist in a low metabolic phase causing persistent infection due to increased resistance to antibiotics.[1]

Thus, earlier diagnosis like x-ray and bacterial cultures are needed to prevent unnecessary morbidity and mortality. The other main issue that needs to be addressed during treatment is proper care of wound, debridement of dead tissue, earlier treatment with combination with parenteral and oral antibiotics that will help in reducing the development of resistance among organisms. Since anaerobic bacteria play an important role in chronic osteomyelitis, it should be included in antimicrobial coverage.[2]

There have been no reports of studies on the bacteriological profile of osteomyelitis in and around Katihar/Kosi region. Due to increase in road traffic accidents resulting in compound fractures and also increase in the number of orthopaedic surgeons using implants, iatrogenic and chronic osteomyelitis is being encountered more frequently.[1]

Also, due to poverty and illiteracy prevailing in this region, there are more chances of acute osteomyelitis turning into chronic cases and presenting in the hospital. The study was therefore undertaken to determine the bacteriological profile of these cases of osteomyelitis and also to ascertain the antibiotic susceptibility pattern of these isolates that will go a long way in helping the clinician in deciding upon the treatment regime for these patients. The data generated by these studies will also help in formulating hospital antibiotic policies.

MATERIAL AND METHODS

Study Population: In this study, a total number of 97 cases were studied to ascertain the bacteriological profile of Osteomyelitis in patients admitted in the Department of Orthopaedics of Katihar Medical College for a period of 18 months extending from December 2014 to May 2015 of which 89 showed growths of various organisms and remaining 8 samples were sterile. Informed consent from all cases was taken before collection of samples. The study sought for and obtained ethical clearance from the Institutional Ethics Committee before it was started.

Isolation and Identification: Samples like pus or exudates or pieces of necrotic tissues were collected during surgery if possible, otherwise aspirated. Swabs were collected only when collection during surgery and aspiration was not possible.

Two samples of the same specimen were collected aseptically using sterile cotton swabs. The swabs were transferred to sterile test tubes and transported to the laboratory as soon as possible.[3]

Pus, exudates/swab was inoculated on 5% Blood agar [BA], Nutrient agar [NA], MacConkey's agar [MA] without crystal violet, and Mannitol salt agar, aerobically at 37^0C for 24 hours. After incubation, the plates were examined for growth and colony morphology. If the culture showed different types of colonies, subculture was done from single colony to obtain pure growth.

Bacterial growth on BA, NA, MA, and MSA were processed for identification and characterization up to species level. Identification and characterization of gram-negative organisms up to species level were done by a battery of standard tests.[3]

Antimicrobial Susceptibility Testing: Antibiotic sensitivity testing was done by modified Kirby-Bauer's disc diffusion test on Mueller-Hinton agar as per the Clinical and Laboratory Standards Institute [CLSI, 2013]. For detection of MRSA strains, MHA [HiMedia, Mumbai, India] supplemented with 4% NaCl was used. Inoculum was prepared and adjusted to 0.5 McFarland's standard. Antibiotic discs were obtained from the HiMedia Laboratories [Mumbai].[4] the concentration of each antimicrobial agent [In µg] tested per disc are mentioned in the Table 1.

Antibiotic Discs for Gram Positive Bacteria	Strength in µg	Antibiotic Discs for Gram Negative Bacteria	Strength in µg	Antibiotic Discs for non-Fermenters	Strength in µg
Amoxicillin	10	Amoxicillin	10	Ceftazidime	30
Cephalexin	30	Cefuroxime	30	Piperacillin	100
Cefoxitin	30	Cefotaxime	30	Ciprofloxacin	5
Netilmicin	10	Cefoperazone	75	Cefoperazone	75
Ciprofloxacin	5	Cefepime	30	Cefepime	30
Erythromycin	15	Amikacin	30	Amikacin	30
Amikacin	30	Gentamycin	30	Gentamycin	30
Vancomycin	30	Ciprofloxacin	5	Tobramycin	10
Linezolid	30	Piperacillin/Tazobactam	100/10	Piperacillin/Tazobactam	100/10
Clindamycin	10	Cefoperazone/Sulbactam	75/10	Cefoperazone/Sulbactam	75/10

Table 1: Antibiotic Discs and its Potency

RESULTS

A total of 97 cases were studied to ascertain the bacteriological profile of Osteomyelitis in patients admitted in the Department of Orthopaedics of Katihar Medical College, of which 89 [91.8%] showed growth of various organisms and remaining 8 [8.2%] samples were sterile. Out of the 97 samples, 84 [86.6%] showed unimicrobial growth and 5 [5.2%] showed polymicrobial growth. A total of 94 organisms were isolated from indoor patient department.

Out of the total of 94 isolates, 61.7% were gram-positive organisms including [53 isolates of S. aureus and 5 isolates of CONS] whereas 38.3% were gram-negative organisms that included [18 isolates of *E. coli,* 7 isolates of *Klebsiella pneumonia*, 9 of *Pseudomonas aeruginosa*, and 2 of Proteus mirabilis]. Of the unimicrobial isolates, *Escherichia coli* [19.0%] was the main organism followed by *Pseudomonas aeruginosa* [8.3%], *Klebsiella pneumoniae* and CONS [5.9% each] and *Proteus mirabilis* [2.3%]. Amongst the polymicrobial growth, the common combination was between *S. aureus* and *E. coli.* Of the unimicrobial cases, 27.4% (23/84) were found in the age group 16-20 years followed by 16.7% (14/84) in 21-25 years and 13.1% (11/84) in 26-30 years. Amongst the polymicrobial cases, 20.0% (1/5) each were found in 21-25 years followed by 26-30 years and 31-35 years. Most of the sterile growths were in the age group 0-5 years, 50.0% (4/8).

Out of the 97 cases, 73.2% (71/97) of patients were males and 26.8% (26/97) were female. The overall male to female ratio was 2.7:1.

The commonest bone affected in the study was tibia 62.9% (61/97) followed by femur 28.9% (28/97) and the other small bones (foot and sacrum 3.1% each and spine 2.1%).

Table 4 shows the incidence of predisposing factors in osteomyelitis cases.

The most common factor leading to osteomyelitis was trauma/accidents 49.5% (48/97) followed by orthopaedic implants 28.9% (28/97) and postsurgical wound 21.6% (21/97).

Of the trauma/accidents cases, 64.6% (31/48) were males and 35.4% (7/48) were females whereas osteomyelitis due to orthopaedic implants was seen in 78.6% (22/28) males and 21.4% (6/28) of females. Osteomyelitis due to postsurgical wound infection was seen in 85.7% (18/21) males and 14.3 (3/21) females [Table 2].

Predisposing Factors	No. of Cases	Percentage
Trauma/accidents	48	49.5
Orthopaedic implants	28	28.9
Postsurgical wounds	21	21.6
Total	**97**	**100**

Table 2: Distribution of Predisposing Factors in Osteomyelitis Cases

S. aureus was the common organism isolated from trauma/accident (83.3%) cases followed by postsurgical wound infections (96.7%) and orthopaedic implants (45.4%). Among the gram-negative organisms, *E. coli, K. pneumoniae*, and *P. aeruginosa* (4.7% each) were isolated from trauma/accident cases whereas E. coli was the main isolate from orthopaedic implants and postsurgical wound infections being 27.3% and 33.3%. On the other hand, 13.6% (3/5) isolates of CONS were associated with orthopaedic implant infections [Table 3].

Microorganisms	Predisposing Factors		
	Trauma/Accidents	Orthopaedic Implants	Postsurgical Wounds
S. aureus	35 (83.3%)	10 (45.4%)	8 (96.7%)
CONS	0	3 (13.6%)	2 (6.7%)
E. coli	2 (4.7%)	6 (27.3%)	10 (33.3%)
Klebsiella pneumoniae	2 (4.7%)	1 (4.5%)	4 (13.3%)
Pseudomonas aeruginosa	2 (4.7%)	2 (9.1%)	5 (16.7%)
Proteus mirabilis	1 (2.3%)	0	1 (3.3%)
Total = 94	**42**	**22**	**30**

Table 3: Distribution of Organism According to Predisposing Factors

The study results showed that 69.8% (37/53) of the strains of S. aureus were MSSA and 30.2% (16/53) of strains were MRSA. Majority of the MRSA (31.3%) isolates were recovered during increased length of stay in the hospital i.e. ≥30 days whereas 32.5% MSSA isolates were recovered during the initial period of stay i.e. 11-20 days [Table 4]. In case of MRSA, resistance was seen with amoxicillin (100%) followed by cephalexin, netilmicin, amikacin, and ciprofloxacin being 62.5% each [Table 5]. On the other hand, 12.5% and 6.3% strains were resistant to vancomycin and linezolid by disc diffusion method. Among the gram-negative bacilli, *E. coli* showed maximum resistance to amoxicillin (100%) followed by 50.0% resistance to cefuroxime, cefotaxime, and cefoperazone each. *Klebsiella pneumonia* showed 100% resistance each to amoxicillin, cefuroxime, and cefotaxime. However, only 16.7% strains of *E. coli* and 42.8% strains of *K. pneumonia* were resistant to gentamicin [Table 6]. Amongst the non-fermenters, *P. aeruginosa* showed maximum resistance to ciprofloxacin (77.7%), piperacillin and cefepime (55.5% each), and ceftazidime (33.3%). Maximum sensitivity was seen with tobramycin, gentamicin (77.7% each) followed by amikacin, piperacillin/tazobactam (55.5%). *P. mirabilis* were resistant to most of the antibiotics except cefepime, tobramycin, cefoperazone/sulbactam, and cefotaxime [Table 7].

Hospital Stay	MRSA	Percentage	MSSA	Percentage
1-10	3	18.8	7	18.9
11-20	4	25.0	12	32.5
21-30	4	25.0	10	27.0
≥ 30	5	31.3	8	21.6
Total	**16**	**100**	**37**	**100**

Table 4: Duration of Hospital Stay with Reference to MRSA/MSSA Infections

Antibiotics	Methicillin Sensitive S. Aureus (MSSA), n=37		Methicillin Resistant S. Aureus (MRSA), n=16		CONS, (n= 5)	
	Sensitive n (%)	Resistant n (%)	Sensitive n (%)	Resistant n (%)	Sensitive n (%)	Resistant n (%)
Amoxicillin	0	37 (100)	0	16 (100)	0	5 (100)
Cephalexin	7 (18.9)	30 (81.8)	6 (37.5)	10 (62.5)	2 (40.0)	3 (60.0)
Netilmicin	10 (27.0)	27 (72.9)	6 (37.5)	10 (62.5)	3 (60.0)	2 (40.0)
Ciprofloxacin	7 (18.9)	30 (81.8)	6 (37.5)	10 (62.5)	1 (20.0)	4 (80.0)
Erythromycin	15 (40.5)	22 (59.5)	7 (43.8)	9 (56.2)	1 (20.0)	4 (80.0)
Amikacin	30 (81.0)	7 (18.9)	6 (37.5)	10 (62.5)	3 (60.0)	2 (40.0)
Vancomycin	37 (100)	0	14 (87.5)	2 (12.5)	3 (60.0)	2 (40.0)
Linezolid	37 (100)	0	15 (93.8)	1 (6.3)	4 (80.0)	1 (20.0)
Clindamycin	30 (81.0)	7 (18.9)	12 (75.0)	4 (25)	3 (60.0)	2 (40.0)
Gentamicin	30 (81.0)	7 (18.9)	7 (43.8)	9 (56.2)	3 (60.0)	2 (40.0)

Table 5: Antibiotic Sensitivity/Resistant Pattern of Staphylococcus Species

Antibiotics	Escherichia Coli, n (18)			Klebsiella Pneumoniae, n (7)		
	S	I	R	S	I	R
Amoxicillin	0	0	18 (100%)	0	0	7 (100%)
Cefuroxime	0	0	18 (50.0%)	0	0	7 (100%)
Cefotaxime	3 (16.7%)	6 (33.3%)	9 (50.0%)	0	0	7 (100%)
Cefoperazone	6 (33.3%)	3 (16.7%)	9 (50.0%)	1 (14.2%)	1 (14.3%)	5 (71.4%)
Cefepime	6 (33.3%)	0	0	3 (42.8%)	3 (42.9%)	1 (14.2%)
Amikacin	9 (50.0%)	6 (33.3%)	3 (16.7%)	4 (57.1%)	0	3 (42.8%)
Gentamicin	15 (83.3%)	0	3 (16.7%)	4 (57.1%)	0	3 (42.8%)
Ciprofloxacin	3 (16.6%)	0	0	1 (14.2%)	3 (42.9%)	3 (42.8%)
Piperacillin/Tazobactam	3 (16.6%)	9 (50.0%)	6 (33.3%)	1 (14.2%)	4 (57.1%)	2 (28.5%)
Cefoperazone/Sulbactam	12 (66.6%)	0	0	1 (14.2%)	1 (14.3%)	5 (71.4%)

Table 6: Antibiotic Sensitivity/Resistant Pattern of Gram-Negative Bacilli/Fermenters

Antibiotics	Pseudomonas Aeruginosa, n (9)			Proteus Mirabilis, n (2)		
	S	I	R	S	I	R
Ceftazidime	2 (22.2%)	4 (44.4%)	3 (33.3%)	1 (50.0%)	0	1 (50.0%)
Piperacillin	4 (44.4%)	0	5 (55.5%)	1 (50.0%)	0	01 (50.0%)
Ciprofloxacin	2 (22.2%)	0	7 (77.7%)	1 (50.0%)	0	1 (50.0%)
Cefoperazone	2 (22.2)	4 (44.4%)	3 (33.3%)	1 (50.0)	0	1 (50.0%)
Cefepime	4 (44.4%)	0	5 (55.5%)	2 (100%)	0	0
Amikacin	5 (55.5%)	0	4 (44.4%)	1 (50.0%)	0	1 (50.0%)
Gentamicin	7 (77.7%)	0	2 (22.2%)	1 (50.0%)	0	1 (50.0%)
Tobramycin	7 (77.7%)	0	2 (22.2%)	2 (100%)	0	0
Piperacillin/Tazobactam	5 (55.5%)	3 (33.3%)	1 (11.1%)	1 (50.0%)	0	1 (50.0%)
Cefoperazone/Sulbactam	4 (44.4%)	4 (44.4%)	1 (11.1%)	1 (50.0%)	0	0
Cefotaxime	4 (44.4%)	4 (44.4%)	1 (11.1%)	0 (50.0%)	0	0

Table 7: Antibiotic Sensitivity/Resistant Pattern of Gram-Negative Bacilli/Non-Fermenters

DISCUSSION

Osteomyelitis is one of the vexing diseases amongst people in developing countries like India due to increase in number of drug-resistant strains that makes treatment even more complicated in addition to requirement of aggressive surgical debridement. Chronic osteomyelitis may require antimicrobial therapy for months to years sometimes with antibiotics that are invaluable for hospital environment such as glycopeptides and carbapenems. Hence, area wise studies on bacteriological profiles and monitoring of antimicrobial susceptibility pattern needs to be carried out in individual settings, which would guide to develop a policy on appropriate use of antibiotics.

Out of the 97 samples, 84 (86.6%) showed unimicrobial growth and 5 (5.2%) showed polymicrobial growth. A total of 94 organisms were isolated from indoor patient department that included (*Staphylococcus aureus,* CONS, *Escherichia coli*, *Klebsiella pneumoniae, Pseudomonas aeruginosa*, and *Proteus mirabilis* in the decreasing order of isolation). Wadekar DM et al. (2014), reported similar findings where 87.0% samples were found to be culture positive whereas 13.0% samples were culture negative; monomicrobial growth was seen in 67.0% and polymicrobial growth in 20.0% cases. Collection of specimen before the administration of antibiotics, use of proper transport media, and other factors play a role in incidence of positive culture.[5]

Of the unimicrobial growth, isolates were *Staphylococcus aureus* (58.3%) followed by *Escherichia coli* (19.0%), *Pseudomonas aeruginosa* (8.3%), *Klebsiella pneumoniae* (5.9%), and *Proteus mirabilis* (2.3%). Amongst the polymicrobial growth, most common combination of isolates was between *S. aureus* and *E. coli* being 40.0%. Kaur J et al. (2008), reported that although bone infections caused by gram-negative organisms had significantly increased, but *S. aureus* (43.0%) remained the most common cause of osteomyelitis, which was followed by *P. aeruginosa* (10.0%), Proteus spp (6.0%), Klebsiella spp and *E. coli* (5.0% each), Enterobacter spp (3.0%), *S. epidermidis* (4.0%), *Streptococcus pyogenes*, and Enterococcus spp (2.0% each).[6]

Out of the 97 cases, 73.2% (71/97) of patients were males and 26.8% (26/97) were female. The overall male to female ratio was 2.7:1. Wadekar M et al. (2014), also reported a higher incidence of osteomyelitis in male than in females with the ratio of 2.7:1. The predominance of male patients may point towards gender bias present in the society. This can also be attributed to more exposure to trauma in males.[5]

The commonest bone affected in the study was tibia 62.9% (61/97) followed by femur 28.9% (28/97) and the other small bones (foot and sacrum 3.1% each and foot 2.1%). Kaur J et al. (2008), reported similar findings where long bones of the lower extremity were involved in 60% cases while those of upper extremity were involved in 14.0% cases. The bones involved in rest of the cases included short bones of hand and feet (10.0%), pelvic bones (8.0%).[6]

The most common factor leading to osteomyelitis was trauma/accidents, 49.5% (48/97) followed by orthopaedic implants, 28.9% (28/97) and postsurgical wound, 21.6% (21/97). Wadekar DM. et al. (2014), reported the common predisposing factor for osteomyelitis to be trauma (44.0%), which was followed by postsurgical infections (23.0%) and orthopaedic implants (21.0%).[5]

Of the trauma/accidents cases, 64.6% (31/48) were males and 35.4% (7/48) were females whereas osteomyelitis due to orthopaedic implants was seen in 78.6% (22/28) males and 21.4% (6/28) of females. Osteomyelitis due to postsurgical wound infection was seen in 85.7% (18/21) males and 14.3 (3/21) females. Suguneswari G et al. (2013), reported that majority of accident cases was seen in males 71.6% (38/53) as compared to female 28.3% (15/53).

Similarly, 84.6% (22/26) and 100% (19/19) of postsurgical wound infections and prosthesis induced infections were seen in male patients.[7]

S. aureus, was the common organism isolated from trauma/accident (83.3%) cases followed by postsurgical wound infections (96.7%) and orthopaedic implants (45.4%). Among the gram-negative organisms, *E. coli, K. pneumoniae,* and *P. aeruginosa* (4.7% each) were isolated from trauma/accident cases whereas *E. coli* was the main isolate from orthopaedic implants and postsurgical wound infections being 27.3% & 33.3%. On the other hand, 13.6% (3/5) isolates of CONS were associated with orthopaedic implant infections.

Gilmore et al. (2009), found that the most common pathogen responsible for osteomyelitis in humans is Staphylococcus species followed by Enterobacteriaceae and Pseudomonas species.[8] Chihara S et al. (2010), reported that coagulase-negative staphylococci are often seen in association with foreign bodies such as prosthetic joints.[9] Contrasting results were shown by Agarwal AC et al. (2008), where *E. coli* was the main pathogen in 34.4% cases especially in open fractures, chronic osteomyelitis, bedsores, and patients with spinal instrumentation.[10] Authors say that *E. coli* is a commensal of the gut and as many orthopaedic patients are bedridden for prolonged periods, contamination of wounds, dressings, linen, clothes, and even hands during perineal hygiene plays a major role in increasing chances of transmission of infection.

Majority of the MRSA (31.3%) isolates were recovered during increased length of stay in the hospital i.e. ≥30 days whereas 32.5% MSSA isolates were recovered during the initial period of stay i.e. 11-20 days. Thus, the number of MRSA strains increased proportionately with increase in the length of stay in hospital due to increased exposure to hospital milieu that harbours the resistant organisms.

Majority of the MSSA showed resistance to amoxicillin followed by cephalexin and ciprofloxacin (100%, 81.8%) whereas in case of MRSA resistance was seen with amoxicillin (100%) followed by cephalexin, netilmicin, amikacin, and ciprofloxacin being 62.5% each. Of the MRSA, 12.5% and 6.3% strains were resistant to vancomycin and linezolid by disc diffusion method. In case of CONS 100%, resistance was seen with amoxicillin followed by 60.0% resistance with cephalexin and cefoxitin each. Ali M et al. (2014), reported that all the MSSA strains were susceptible to vancomycin, gentamicin, teicoplanin, ciprofloxacin, and linezolid; while the MRSA strains were sensitive to vancomycin and linezolid.[11]

In other studies, the MRSA isolates showed resistance to commonly used antibiotics like cefepime (100%), erythromycin (90.9%), tetracycline (90.9%), co-trimoxazole (90.9%), piperacillin/tazobactam (81.8%), ciprofloxacin (72.7%), and levofloxacin (54.5%). However, all the MRSA strains showed 100% sensitivity to vancomycin and 91.6% sensitivity to levofloxacin.[7] It is quite clear from the studies that have been conducted so far as well as from the present study that MRSA strains are becoming alarming because of their increased resistance towards antibiotics-like amikacin, netilmicin, and to a lesser extent to vancomycin and linezolid that leaves the clinicians with less choice to use the appropriate drug for treatment of chronic osteomyelitis.

In our study, *E. coli* showed maximum resistance to amoxicillin (100%) followed by 50.0% resistance to cefuroxime, cefotaxime, and cefoperazone each. *Klebsiella pneumoniae* showed 100% resistance each to amoxicillin, cefuroxime, and cefotaxime. On the other hand, only 16.7% strains of *E. coli* and 42.8% strains of *K. pneumoniae* were resistant to gentamicin. Other authors reported that E. coli and *K. pneumoniae* showed that 100% and 80.0% resistance to ampicillin. A total of 83.8% strains of gram-negative bacilli were found to be resistant to gentamicin. The authors reported cefoperazone-sulbactam combination and amikacin to be the most effective drug for treating gram-negative organisms.[6]

In our study, *P. aeruginosa* showed maximum resistance to ciprofloxacin (77.7%), piperacillin and cefepime (55.5% each), and ceftazidime and cefoperazone being (33.3%) each. Maximum sensitivity was seen with tobramycin, gentamicin (77.7% each) followed by amikacin, piperacillin/tazobactam (55.5%). In other studies, 60.0% strains of *P. aeruginosa* were found to be resistant to ceftriaxone followed by 58.0% strains resistant to cefotaxime and 54.0% each being resistant to cefepime and imipenem. On the other hand, authors reported aztreonam and levofloxacin to be the most active drugs against non-fermenters.[5]

Fifty percent strains of *P. mirabilis* were found to be resistant to ceftriaxone, piperacillin, ciprofloxacin, cefoperazone, amikacin, gentamicin, piperacillin/tazobactam whereas 100% sensitivity was seen with cefepime, tobramycin, cefoperazone/sulbactam, and cefotaxime. Antibiotic susceptibility pattern in case of *P. mirabilis* as reported by other authors showed maximum resistance to cefotaxime (75.0%) followed by ceftazidime and ceftriaxone (63.0% each) whereas the strains retained 100% sensitivity to imipenem.[5]

CONCLUSION

The present study highlights the importance of microbiological examination of bone in cases of osteomyelitis. Microorganisms could not be detected in 8.2% of cases. The absence of growth may be due to anaerobic organisms, which flourish well in the dead tissues due to lack of oxygen supply. However, 86.6% of cases showed unimicrobial and 5.2% had polymicrobial aetiologies. Thus, any bacteria gram-positive or gram-negative either or alone or as mixed infection could be responsible for osteomyelitis.

MRSA isolation in this region was found to be slightly lower (30.2%) of all *Staphylococcus aureus* isolates as compared to the isolation rates in some other parts of India, which is probably because the medical college is situated in the rural area where the organisms are not exposed to as much antibiotic pressure as in the urban areas. Thus, the number of MRSA strains increased proportionately with increase in the length of stay in hospital due to increased exposure to the hospital milieu that harbours the resistant organisms. To prevent hospital acquired infection, prolonged hospital stay should be restricted in orthopaedic wards.

Our study revealed that overall gentamicin was the most sensitive drug among all gram-negative bacilli followed by amikacin, tobramycin, and cefepime, most of the gram-negative bacilli were found to be resistant to third generation cephalosporins like cefotaxime, cefuroxime, and ceftazidime. It is important to undertake studies to see the prevalence of bacterial isolates and their antibiotic sensitivity pattern in a particular geographical area that will enable appropriate and judicious selection of antibiotics and would limit the emerging drug-resistant strains in the future to treat the disease successfully.

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