

# A study on bacterial and fungal isolates and their antimicrobial susceptibility pattern in patients with chronic osteomyelitis in a tertiary care hospital

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## ABSTRACT

Chronic osteomyelitis is a major challenging problem in our country, and it is a persistent disease difficult to treat and eradicate completely. The aim of the study is to analyze the predisposing factors associated with chronic osteomyelitis and to study the causative organisms and their antimicrobial susceptibility pattern and check the resistance pattern in common isolates. It is a cross-sectional study done during a time between October 2011 and September 2012 and was included in the analysis of the data. Total of 120 patients were included prospectively. In 120 patients detailed history were recorded. Collection of samples was done under strict aseptic precautions. Pus, swabs from sinus tract and sequestrum were the samples collected. Processing of samples was done by culture both bacterial and fungal, catalase test, oxidase test, biochemical reactions, and antimicrobial susceptibility was done by Kirby-Bauer disc diffusion method according to CLSI guidelines. All the tests were done as per protocol. ATCC strains *Staphylococcus aureus* - ATCC 25923, *Escherichia coli* - ATCC 25922, and *Pseudomonas aeruginosa* - ATCC 27853 were used as controls. Detection of  $\beta$ -lactamase enzymes in Gram-negative bacilli (GNB), detection of methicillin resistance in *S. aureus* were also done phenotypically. Minimum inhibitory concentration (MIC) of vancomycin was done to detect vancomycin resistance against *S. aureus*. Fungal cultures were identified by macroscopic appearance, microscopy analysis (Gram staining, LCB), germ tube test, CHROM agar media, and sugar fermentation. MIC determination by microbroth dilution method was also done. ATCC *Candida albicans* 90028 was used as quality control. In this study, 97 (80.83%) were males and 23 (19%) were females. 40% of the patients had a duration of the illness from 7 to 12 months. 35.8% of patients had illness ranged from 13 to 24 months. 50.8% of patients had compound fracture leading to infection. Among the samples collected, 63 (52.5%) were sequestrum/per-operative collections of pus and tissue fluids, and 57% (47.5%) were swabs. Culture positivity was 83.3%, an increased number of polymicrobial (12.2%) infections were noted in swabs, though monomicrobial infection was the most common type even in swabs (57.8%). The common organism isolated was *S. aureus* (36.7%), *S. aureus* which was the most common bacteria isolated in this study showed 100% sensitivity to Rifampin, 97.4% sensitivity to vancomycin, 64% sensitivity to amikacin, chloramphenicol and Erythromycin, 51.2% to Penicillin, and 51.2% were sensitive to cefoxitin. Among Gram-negative isolates, *P. aeruginosa* was the most common isolate, which showed 100% sensitivity to imipenem, 76.4% to cefoperazone sulbactam, 52.9% to amikacin, and 35.2% to cefotaxime and ceftazidime. One isolate of *Mycobacterium tuberculosis* was sensitive to all first and second line drugs. The one fungal isolate *Candida tropicalis* was sensitive to fluconazole, amphotericin B, itraconazole, and voriconazole. 40.5% of aerobic bacteria were multidrug-resistant. 56.6% were aerobic Gram-positive cocci (GPC) and 43.3% were aerobic GNB, one acid-fast bacillus *M. tuberculosis*, and one yeast *C. tropicalis* were isolated in the study. Among GPC, *S. aureus* (36.7%)

was the most common pathogen isolated followed closely by *Staphylococcus epidermidis* (10.5%). All GPC except one were sensitive to vancomycin and rifampin. Among GNB, all were sensitive to imipenem and 90% to cefoperazone sulbactam. *Pseudomonas* had lower sensitivity (76.4%) to cefoperazone. Various factors in open fracture leading to chronic osteomyelitis

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each patient has to be routinely monitored after trauma and treatment for developing osteomyelitis. Treatment given in the early stage will prevent dreadful complications and sequelae.

**Keywords:** Antimicrobial susceptibility, osteomyelitis, cross-sectional study, gram-positive cocci

## Introduction

The word osteomyelitis is a combination of Greek word “osteon” meaning bone and “myelos” meaning marrow plus the suffix. “It is” meaning inflammation. Osteomyelitis is acquired in three ways. They are direct seeding of microorganisms into bone due to trauma or surgery, hematogenous spread of microorganisms from the focus of infection elsewhere in the body and spread from surrounding infected soft tissue and joints.

Commonly the infection is monomicrobial. Infection due to multiple organisms<sup>[1]</sup> is usually seen in patients with diabetes mellitus with an ulcer in the foot. The following six components characterize chronic osteomyelitis: Sequestrum formation or sclerosis, radiological changes seen in bone due to infection for 6 weeks or longer, relapse or persistence of infection after initial treatment, osteomyelitis due to foreign bodies, osteomyelitis in association with peripheral vascular disease, and organisms that produce chronic disease (e.g., *Mycobacterium tuberculosis*).

The most common presenting symptoms are persistent pain and chronic intermittent discharge through sinuses. Bone debris and sequestra find an exit through multiple openings in an involucrum, go through the sinus tracts and present to the surface. In children, after discharge of sequestrum, the sinus is closed, and the cavity is filled with new bone. In adults, the sinus is not closed and the persistence of viable pathogens in cavities for a longer period leads to reactivation of infection at any time.

The usual complications of chronic osteomyelitis are reduced rate of growth, pathological fracture, septic arthritis, lengthening of bone, and contracture of muscles. Other rare complications are the formation of epithelioma, secondary amyloidosis,<sup>[2]</sup> and squamous cell carcinoma in scar tissue (<1%).

Chronic osteomyelitis is a disease, which is difficult to eradicate completely. There may be subsidence of systemic symptoms, but the cavities containing purulent material, infected granulation tissue or sequestrum act as foci of infection. There may be recurrent acute flare-ups occurring at indefinite intervals over months and years. To achieve eradication of the disease, aggressive surgical debridement with curettage of cavities, filling of cavities with soft tissues and effective antimicrobial treatment is required.<sup>[3]</sup>

The pattern and behavior of organisms are constantly changing under the pressure of newer antibiotics.<sup>[4]</sup> As a result, the wonder drugs of fifties have been relegated to a position of limited usefulness today. With this background, it is felt worthwhile to study the spectrum of organisms causing osteomyelitis and their antimicrobial susceptibility pattern.

## Aim

The aim of the study was to study the predisposing factors associated with chronic osteomyelitis, study the causative organisms and their antimicrobial susceptibility pattern and check the resistance pattern in common isolates.

## Materials and Methods

It is a cross-sectional study done during a time between October 2011 and September 2012 was included in the analysis of the data. Total of 120 patients were included prospectively. The study was conducted in the Institute of Microbiology, Madras Medical College in association with Institute of Orthopaedics, Rajiv Gandhi Government General Hospital, Chennai - 600 003.

## Ethical consideration

The necessary Ethical Committee approval was obtained before the commencement of the study. Informed consent was obtained from the study population. All patients satisfying the inclusion criteria were documented. Patients were interviewed by structured questionnaire.

## Inclusion criteria

1. Patients are older than 12 years.
2. Patients admitted to orthopedic wards and those attending outpatient departments who satisfy one of the following six components of chronic osteomyelitis.
3. Osteomyelitis in association with trauma only.
4. Osteomyelitis in association with diabetes and peripheral vascular compromise.
5. Clinical evidence of chronic disease (e.g., M. tuberculosis).
6. Radiological changes suggestive of infection for 6 weeks or more.
7. Formation of sequestrum or sclerosis
8. Even after treatment, persistence or relapse of infection.

## Exclusion criteria

1. Patients with prosthetic orthopedic implant devices.
2. Pediatric age group (<12 years).

## History

Name, age, sex, date of admission, physical examination findings, history of trauma, associated predisposing factor (diabetes mellitus, intravenous drug abuse, immunosuppression, and tuberculosis) duration of illness, smoking, and alcoholism were also recorded.

## Collection, transport and processing of samples<sup>[5]</sup>

Under strict aseptic precautions, samples were collected from the patients and transported immediately to the laboratory and sample

## Conclusion

56.6% were aerobic Gram-positive cocci (GPC) and 43.3% were aerobic GNB. Among GPC, *S. aureus* (36.7%) was the most common pathogen isolated followed closely by *S. epidermidis* (10.5%). All GPC except one were sensitive to vancomycin and rifampin. Among GNB, all were sensitive to imipenem and 90% to cefoperazone sulbactam. *Pseudomonas* had lower sensitivity (76.4%) to cefoperazone. Various factors in open fracture leading to chronic osteomyelitis each patient has to be routinely monitored after trauma and treatment for developing osteomyelitis. Treatment given in the early stage will prevent dreadful complications and sequelae.

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