# BACTERIAL AETIOLOGY OF OSTEOMYELITIS CASES AT FOUR HOSPITALS OF LAHORE

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**Background**: The conclusive diagnosis of osteomyelitis requires isolation of pathogen in aspirate from bone lesion, bone debridement and blood culture. The present research was undertaken to study the microbiological pattern of cases of osteomyelitis reporting to four hospitals in Lahore. Method: One hundred and fifty patients of osteomyelitis were selected from outpatient departments and Orthopaedic wards of Lahore General Hospital, Sir Ganga Ram Hospital, Services Hospital and Mayo Hospital, Lahore. Specimens of pus from bone, blood and bone debridement were collected. All samples were inoculated onto two Blood Agar and one MacConkey agar plates. One Blood Agar plate was incubated anaerobically for 48 hours and the other two plates aerobically for 24 hours. Smears were made from samples and stained by the Gram's stain. The colonies obtained were processed according to the technique of Mackie and MacCartney. Results: The commonest isolates belonged to the Enterobacteriaceae (32.8%), followed by Staphylococcus aureus (29.5%), Pseudomonas aeruginosa (15.5%), anaerobes (2.6%) and miscellaneous (19.3%). Five (2.7%) anerobic bacteria were isolated. Anaerobic bacteria were peptostreptococci, peptococci and bacteroides either alone or as a mixed infection. Conclusion: The present study highlights the importance of microbiological examination of bone in cases of osteomyelitis. Different types of bacteria either alone or as a mixed infection could be the causative agent(s).

**Key Words:** Bacteria, Aerobes, Staphylococci, Enterobacteriaceae, Pseudomonas, Anaerobes, Osteomyelitis.

# **INTRODUCTION**

Infections of the bone have been known for a long time. Attempts at understanding the disease began a century ago, when in 1884 Rodet reported to the Academy of Sciences in Paris, his experimental production of haematogenous osteomyelitis in animals by means of intravenous injection of Staphylococci<sup>1</sup>. The micrococci were injected into a rabbit which developed typical lesion of osteomyelitis in long bones<sup>2</sup>.

The most common route by which bacteria reach the bone is blood stream<sup>3,4</sup>. However, traumatic modes as penetrating injury<sup>5</sup>, fractures and intramedullary nailing<sup>6</sup> and post-surgical complications<sup>7</sup> have been identified. Intravenous drug users<sup>8,9</sup> and foreign body presence<sup>10</sup> also predispose to bone infection. The initial diagnosis of osteomyelitis is usually made on physical signs<sup>11</sup> and by sonography for early soft tissue changes<sup>12,13</sup>. Magnetic resonance imaging<sup>14</sup> and bone scans<sup>15</sup> are most sensitive and specific. Conclusive diagnosis requires isolation of pathogen in aspirate from bone lesion, bone debridement and blood culture<sup>15</sup>.

In Pakistan, only a few workers have studied the bacteriological pattern of osteomyelitis. The reports from different cities have shown different bacteriological pattern. In

a study at Karachi<sup>16</sup>, out of 125 cases, 68.6% were reported to be infected with Staphylococci. Karamat *et al*<sup>17</sup> from Rawalpindi have also reported a high frequency (79%) of Staphylococci, whereas Farooq and Ahmad<sup>18</sup> have reported a very low (37.5%) frequency of Staphylococci. Studies from abroad by Karwowska *et al*<sup>19</sup> Alonge *et al*<sup>20</sup> and Lobati *et al*<sup>10</sup> also support the predominant role of Staphylococci in bone infection.

The present research study was undertaken to study the microbiological pattern of cases of osteomyelitis reporting to four hospitals in Lahore.

# MATERIAL AND METHODS

The study was conducted at the Department of Microbiology, Postgraduate Medical Institute, Lahore. One hundred and fifty patients were selected from outpatient departments and Orthopaedic wards of Lahore General Hospital, Sir Ganga Ram Hospital, Services Hospital and Mayo Hospital, Lahore.

The patients included were from both sexes and all age groups. The only exclusion criterion was those patients who were on antibiotic therapy.

Specimens of pus from bone, blood and bone debridement were collected. All samples were inoculated onto two Blood Agar and one MacConkey agar plates. One Blood Agar plate was incubated anaerobically for 48 hours and the other two plates aerobically for 24 hours.

Smears were made from samples and stained by the Gram's stain. The colonies obtained were processed according to the technique of Mackie and MacCartney<sup>21</sup>.

## RESULTS

The age of the patients ranged from 2–90 years with a mean age ( $\pm$ SD) of 28.73 $\pm$ 16.64 years. There were 105 males and 45 females.

There were four specimens of bone curetting, two specimens of blood and 144 specimens of pus. Out of these 150 specimens 186 isolates were identified.

The positivity rate according to various hospitals is shown in Table-1. Table-1: Distribution of positive cultures according to the hospitals.

Hospital	Tested	Positive	Percentage	
Mayo	74 70		93.00	
Lahore General	27	26	96.29	
Services	28	26	92.85	
Sir Ganga Ram	21	18	85.71	

The commonest isolates belonged to the Enterobacteriaceae (32.8%), followed by Staphylococcus aureus (29.5%), Pseudomonas aeruginosa (15.5%), anaerobes (2.6%) and miscellaneous (19.3%).

The distribution of isolates according to various age groups is shown in Table-2. Five (2.7%) anaerobic bacteria were isolated. Anaerobic bacteria were peptostreptococci, peptococci and bacteroides either alone or as a mixed infection. The miscellaneous group comprised of Streptococci, Staphylococcus epidermidis, Diptheroids, Micrococci and Bacilli.

## DISCUSSION

In the present study, monomicrobial infections in cases of osteomyelitis were seen in two third of patients while in one third cases the aetiology was polymicrobial. Other studies, from Pakistan as well as abroad <sup>17,22,23</sup>, also report the predominance of monomicrobial aetiology (Table-2). Recent studies<sup>20,24,25</sup> all report an increasing incidence of polymicrobial infection than the series reported in the past<sup>26,27</sup> in which mainly monomicrobial infection was common.

Study	Year	Country	Monomicrobial	Polymicrrobial
Dendrinos et al	1995	Greece	57.1	42.9
Karamat <i>et</i> al	1995	Pakistan	62.5	12.5
Mousa	1997	Iraq	53.8	46.15
Present study	1999	Pakistan	70.7	29.3

Table-2: Comparison showin	g percentage of monomicrobial/	polymicrobial osteomyelitis

As shown in Table-3, Staphylococcus aureus was the predominant isolate (29.56%). Most studies as by Alonge *et al*<sup>20</sup>, Lobati *et al*<sup>10</sup>, Karwowska *et al*<sup>19</sup>, Carek *et al*<sup>28</sup>, Marsh *et al*<sup>25</sup> and Karamat *et al*<sup>17</sup> also report Staphylococcus aureus as a single organism to be the commonly isolated pathogen from bone infection.

Table-3: Distribution of isolates in various age groups of osteomyelitis patients

Patients							
Age group (yea rs)	N 0.	No. of Microb es	Staphylococ cus aureus (%)	Entero- bacteriace ae (%)	Pseudomo nas aeruginosa (%)	Anaerob es (%)	Miscellane ous (%)
<15	8	34	29.41	20.59	17.65	-	32.35
15-29	57	70	32.86	30.0	14.28	2.86	20.0
30-44	36	49	24.49	40.81	14.29	6.12	14.29
45-59	18	21	28.57	38.10	14.28	-	19.05
>60	11	12	33.33	41.67	25.0	-	-
Total	15 0	186	29.56	32.80	15.60	2.69	19.35

Although Staphylococcus aureus remains the most frequent pathogen isolated in bone, the distribution varies from two third Staphylococcus aureus to one third Enteriobacteriaceae or one third each of Staphylococcus aureus, Enteriobacteriaceae and Pseudomonas aeruginosa. This increase in Pseudomonas aeruginosa as a significant bone pathogen is related to the increasing nosocomial nature of osteomyelitis<sup>29</sup>.

Mousa<sup>23</sup> reported a slight predominance in the isolation rate of Enteriobacteriaceae. Even in our study there was a difference of only 3.24% between Staphylococcus aureus and Enterobacteriaceae group. Enterobacteriaceae are increasingly common nosocomial pathogens<sup>29</sup>. The third major group in our study was Pseudomonas aeruginosa. Pseudomonas aeruginosa remains a severe complication of hospitalization<sup>30</sup>. The total significant number of cases noted to be infected by the Entrobacteriaceae in the present study could be due to blind therapy for presumptive Staphylococcal aetiology, thus eliminating some such cases. Moreover the post-traumatic (69) and post-surgical (48) cases during their hospital stay (114 indoor cases) acquired Klebsiella and Pseudomonas as additional pathogens in the bone. Mixed infections included upto three isolates belonging to aerobes as Staphylococci, Enterobacteriaceae, Pseudomonas, miscellaneous group or with an anaerobe. Enterbacteriaeiae alone as single organism were isolated in thirty cases and as two organisms in eight cases. Klebsiella was the commonest Entertobacteriaceae to be isolated (21 cases) in single or mixed pattern.

Microorganisms as Staphylococcus epidermidis included in miscellaneous group (5 isolates) have gained importance in periprosthetic infections<sup>31</sup>.

This proves the importance of culturing pus from osteomyelitis cases for aerobes, as well as anaerobes for appropriate management and cure of chronic illness.

#### CONCLUSION

The present study highlights the importance of microbiological examination of bone in cases of osteomyelitis. Microorganisms could not be detected in only 6.6% cases and one third of cases had a polymicrobial aetiology. Any bacterium, Gram positive or negative, aerobe or anaerobe, either alone or as a mixed infection, could be responsible for osteomyelitis.

The future era with modern high speed travel, warfare, use of implants and prosthetics will add to the load of osteomyelitis.

The clinicians should first obtain a microbiological investigation and then treat their patients to halt the chronic relentless course of this crippling disease.

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