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# Frequency and Pattern of Bacterial & Fungal Infections in Neutropenic Patients Undergoing Bone Marrow Transplantation

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## Abstract

**Objective:** To determine the frequency and pattern of bacterial & fungal infections in neutropenic patients undergoing bone marrow transplantation.

**Methodology:** The descriptive study was conducted in clinical hematology & stem cell transplant department, AFBMTC/NIBMT, Rawalpindi from 1/1/2019 to 31/12/2019. A total of 84 patients were observed. All patients were monitored six hours for neutropenic fever. Blood samples were taken with aseptic technique for bacterial & fungal cultures in these patients. Serum galactomannan and beta-D glucan tests were carried out in patients with suspected fungal infection. Chest X-Ray was done for lower respiratory tract infection. These infections were categorized as primary bloodstream infections or secondary bloodstream infections as per operational definitions. Data was analyzed using SPSS version 22.

**Results:** Out of the 84 patients, 68 (80.9%) patients fulfilled the criteria for primary or secondary bloodstream neutropenic infections. The median age of the study population was 8 years (SD ± 1.25). Seventy-one percent of patients were male and 29% of patients were female. Ninety-three percent of patients had a bacterial infection, while 7% of patients had a fungal infection. Fifty-six patients had primary bloodstream infections while 12 patients had secondary bloodstream infections. Klebsiella pneumoniae, Pseudomonas aeruginosa & Stenotrophomonas maltophilia were the most common bacteria isolated from cultures. Out of 12 patients who had secondary bloodstream infections, 8(12%) patients had radiological findings (consolidations, opacities) while 4(6%) patients had positive serum galactomannan.

**Conclusion:** Our study concludes that bacterial infections are more common than fungal infections in neutropenic patients undergoing bone marrow transplantation. Klebsiella pneumonia is the most common pathogen in pre-engraftment neutropenic phase.

**Keywords:** Hematopoietic Stem cell transplantation (HSCT), Neutropenic infections, Primary bloodstream infections.

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## Introduction

Hematopoietic Stem cell transplantation (HSCT) is an established therapeutic modality for numerous malignant and non-malignant conditions,[1] considered as a standard of care in the management of two broad categories of bone marrow failures. The first category consists of functional failure of bone marrow or marrow-derived cells e.g. aplastic anaemia, myelodysplastic syndrome (MDS), immunodeficiency syndromes or hemoglobinopathies (Thalassaemia, sickle cell anaemia).

The second category includes hematologic malignancies such as acute or chronic leukaemia, multiple myeloma, lymphomas, and myeloproliferative neoplasms.[2] The former is a more common indications for HSCT in our set up.

Opportunistic infections are the leading cause of morbidity and mortality in patients undergoing HSCT. An opportunistic infection is caused by pathogens such as (bacteria, viruses, fungi, or protozoa) that take advantage of an immune-compromised host, an altered microbiota, and breached integumentary barriers. Many of these pathogens do not cause disease in an immune-

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competent host. A compromised immune system resulting from conditioning lowered resistance to infection, a penetrating injury, and a lack of competition from normal microbiota presents an opportunity for the pathogen to cause infection in the recipient of HSCT.[3] Post-HSCT sepsis is divided into three phases (1) early pre-engraftment phase (2 to 4 weeks after HSCT) (2) early post-engraftment phase (2 to 3 months) after the HSCT and (3) late phase > 3 months after engraftment.

The morbidity & mortality associated with neutropenic sepsis is variable, depend on the degree, type, and duration of immunosuppression, use of prophylactic strategies.[4, 5]

Previous studies data from Pakistan in post-transplant settings showed a frequency of 77.9% based on clinical and laboratory analysis. A combined 1mortality due to viral, fungal and bacterial infections was 13%.[6] Internationally studies showed even higher frequency up to 90% opportunistic infections.[7] Fatal opportunistic infections have been reported between 4-15% matched sibling donors/recipient and 12-28% of unrelated transplant setting. [8] In an Italian study of 553 HSCT recipient, 30% incidence of severe opportunistic infections were reported in the early pre-engraftment phase comprising 97% bacterial and 3 % fungal infections[9]. Infectious complications in HSCT recipients (allogeneic & autologous) early pre-engraftment phase are associated with major mortality and morbidity .[10]

Studies on opportunistic infections in HSCT recipient, from South Asia (Pakistan) in the recent past is nearly non-existent. This study will help to establish patterns of bacterial and fungal infections in our patients helping in the early initiation of treatment.

The study aimed to determine the frequency and pattern of bacterial & fungal infections in neutropenic patients undergoing bone marrow transplantation.

## Methodology

A prospective study was conducted at Clinical Hematology & Stem Cell Transplant Department of Armed forces bone marrow transplant centre/ National institute of blood and marrow transplantation AFBMTC/NIBMT, Rawalpindi from 1/1/2019 to 31/12/2019. After approval from AFBMTC ethical review committee and informed consent was taken from all patients or guardians in case of minors(<18yrs). A total of 84 patients HSCT recipients of both genders and all age groups, developing a fever during HSCT and were tested by blood cultures, swabs, radiological investigation, indwelling catheters tips cultures.

All HSCT recipients were monitored for infections during the pre and post-transplant period. Daily CBC of patients on conditioning chemotherapy was done to identify the neutropenic phase. Temperature of the patients was monitored at six hourly intervals in neutropenic patients. C Reactive Protein (CRP) was used as a surrogate marker for early identification of infections daily. On admission skin swabs of all patients were done for MRSA colonization. A blood culture sample was taken with an aseptic technique from febrile neutropenic patients and was sent for bacterial & fungal culture, Serum galactomannan and beta-D glucan were carried out in patients with suspected fungal infection. Chest X-Ray of the patient was performed to rule out lower respiratory tract infection, and identify the pattern of involvement. Stool cultures were performed in all patients pre-transplant to identify ESBL, CRE, VRE, and post-transplant in patients with diarrhoea. Urine cultures were done in patients with urinary tract symptoms. Infections were categorized into primary or secondary bloodstream infections depending on operational definitions. The neutropenic phase was divided into Pre & Post -HSCT neutropenia.

Data was entered and analyzed using SPSS Version 22. Mean and SD was calculated for continuous variables like age, blood counts including absolute neutrophilic count, CRP, and temperature. Frequency and percentages were calculated for categorical variables like sex, type of bone marrow transplantation, presence or absence of primary or secondary bloodstream infections, type of infection, blood/tissue cultures & radiological findings. Effect modifiers like age, sex, type of bone marrow transplant, type of infection, and timing of infection was controlled by stratification. Post-stratification chi-square test was applied, a p-value of ≤0.05 was considered significant.

## Results

A total number of 68 patients undergoing HSCT that fulfilled the criteria for primary or secondary bloodstream infections were included in the study.

Out of 68 patients, 48 (71%) were male and 20 (29%) were female, with a median age of 8 years (SD ±1.25). More than half of the patients, i.e., 56 (82%) were in the age bracket of (5-20) years, 12 (18%) patients were in the age bracket of (21-40 years). Sixty-three (93%) patients had Allogeneic HSCT, 4% (3) Auto HSCT and 3% (2) had Haplo HSCT. Mean absolute neutrophilic count (ANC) was 0.1 x$10^9$/L (± 0.01), mean CRP level was 60 mg/dl (±10.73) and mean highest temperature was 102°F (± 2.11).

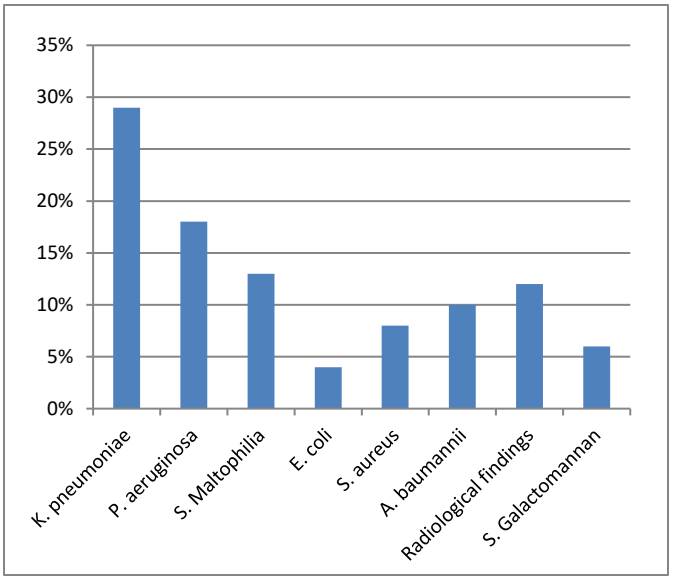


**Figure 1. Blood/Tissue Cultures & Radiological Findings of Patients having Infections**

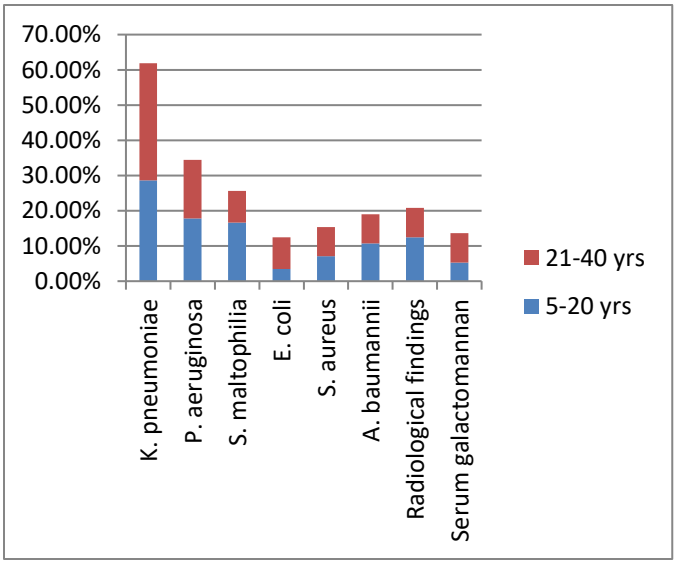


**Figure 2. Stratification of Blood/Tissue Cultures & Radiological Findings based on Age**

Bacterial infections constituted (93%) as compared to fungal infections (7%). Patients were more prone to infections in the pre-engraftment phase (98%) as compared to post-engraftment phase. Detailed blood, tissue culture, and radiological findings of these patients are detailed in Figure 1.

Stratification of blood/tissue cultures, positive serum galactomannan & radiological findings for age is given in Figure 2 while sex, type of bone marrow transplant, type of infection, and timing of infection is given in table number I-V.

**Table no I: Stratification of Blood/Tissue Cultures & Radiological Findings based on Gender.**

| Blood/Tissue Cultures & Radiological Findings | Male | Female | Total | P value |
|---|---|---|---|---|
| ***Klebsiella pneumoniae*** | 14(29.1%) | 6(30%) | 20(29.4%) | 0.945 |
| ***Pseudomonas aeruginosa*** | 8 (16.6%) | 4(20%) | 12(17.6%) | 0.742 |
| ***Stenotrophomonas maltophilia*** | 7(14.5%) | 2(10%) | 9(13.2%) | 0.611 |
| ***Escherichia coli*** | 2(4.1%) | 1(5%) | 3(4.4%) | 0.878 |
| ***Staphylococcus aureus*** | 3(6.2%) | 2(10%) | 5(7.3%) | 0.589 |
| ***Acinetobacter baumannii*** | 5(10.4%) | 2(10%) | 7(10.2%) | 0.958 |
| **Radiological findings (consolidations / opacities)** | 6 (12.5%) | 2(10%) | 8(11.7%) | 0.770 |
| **Serum galactomannan** | 3 (6.25%) | 1(20%) | 4(5.8%) | 0.841 |
| **Total** | 48 | 20 | 68 | |

**Table no II: Stratification of Blood/Tissue Cultures & Radiological Findings for Type of Bone Marrow Transplantation**

| Blood/Tissue Cultures & Radiological Findings | Auto BMT | Allo BMT | Haplo BMT | Total | P-value |
|---|---|---|---|---|---|
| Klebsiella pneumoniae | 1 (33.3%) | 18 (28.5%) | 1 (50%) | 20 (29.4%) | 0.797 |
| Pseudomonas aeruginosa | 1 (33.3%) | 10 (15.8%) | 1 (50%) | 12 (17.6%) | 0.352 |
| Stenotrophomonas maltophilia | 1 (33.3%) | 8 (12.6%) | 0 | 9 (13.2%) | 0.502 |
| Escherichia coli | 0 | 3 (4.76%) | 0 | 3 (4.4%) | 0.882 |
| Staphylococcus aureus | 0 | 5 (7.9%) | 0 | 5 (7.4%) | 0.807 |
| Acinetobacter baumannii | 0 | 7 (11.1%) | 0 | 7 (10.3%) | 0.733 |
| Radiological findings(consolidations / opacities) | 0 | 8 (12.7%) | 0 | 8 (12.7%) | 0.697 |
| Serum galactomannan | 0 | 4(6.3%) | 0 | 4(5.9%) | 0.844 |
| Total | 3 | 63 | 2 | 68 | |

**Table no III: Stratification of Blood/Tissue Cultures & Radiological Findings for Type of Infection**

| Blood/Tissue Cultures & Radiological Findings | Bacterial | Fungal | Total | P-value |
|---|---|---|---|---|
| ***Klebsiella Pneumonia*** | 20(31.7%) | 0 | 20(29.4%) | 0.133 |
| ***Pseudomonas Aeruginosa*** | 12(19%) | 0 | 12(17.6%) | 0.282 |
| ***Stenotrophomonas Maltophilia*** | 9(14.2%) | 0 | 9(13.2%) | 0.364 |
| ***Escherichia coli*** | 3(4.7%) | 0 | 3(4.4%) | 0.617 |
| ***Staphylococcus aureus*** | 5(7.9%) | 0 | 5(7.3%) | 0.512 |
| ***Acinetobacter baumannii*** | 7(11.1%) | 0 | 7(10.3%) | 0.431 |
| **Radiological findings(consolidations / opacities)** | 5(7.9%) | 3(60%) | 8(11.7%) | 0.000 |
| **Serum Galactomannan** | 0 | 4(80%) | 4(5.9%) | 0.000 |
| **Total** | 63 | 5 | 68 | |

**Table no IV: Stratification of Blood/Tissue Cultures & Radiological Findings for Timing of Infection**

| Blood/Tissue Cultures & Radiological Findings | Pre Engraftment Phase | Post Engraftment Phase | Total | P-value |
|---|---|---|---|---|
| ***Klebsiella pneumoniae*** | 20 (29.8%) | 0 | 20 (29.4%) | 0.515 |
| ***Pseudomonas aeruginosa*** | 12 (17.9%) | 0 | 12 (17.6%) | 0.640 |
| ***Stenotrophomonas maltophilia*** | 9 (13.4%) | 0 | 9 (13.2%) | 0.693 |
| ***Escherichia coli*** | 3 (4.5%) | 0 | 3 (4.4%) | 0.828 |
| ***Staphylococcus aureus*** | 5 (7.5%) | 0 | 5 (7.4%) | 0.776 |
| ***Acinetobacter baumannii*** | 7 (10.4%) | 0 | 7 (10.3%) | 0.732 |
| **Radiological findings (consolidations / opacities)** | 7 (10.4%) | 1 (100%) | 8 (11.8%) | 0.005 |
| **Serum galactomannan** | 4 (5.9%) | 0 | 4 (5.8%) | 0.801 |
| **Total** | 67 | 1 | 68 | |

**Table no V: Comparison with National & International Studies**

| | Study Design | Sample size | Bacterial infections (%) | Fungal infections (%) |
|---|---|---|---|---|
| **Our Study** | Prospective | 68 | 93 | 7 |
| **Khalil Ullah *et al. (2008)*** | Retrospective | 154 | 51.3 | 14.9 |
| **Anne HJ *et al. (2016)*** | Retrospective | 499 | 60 | 17.8 |
| **M. Mikulsaka *et al. (2009)*** | Retrospective | 343 | 71.6 | 5.4 |
| **M. Mikulsaka *et al. (2018)*** | Retrospective | 553 | 97 | 3 |

# Discussion

Hematopoietic stem cell transplantation (HSCT) is an established therapeutic modality for numerous malignant and non-malignant conditions.[1] It is recommended in the treatment of two broad categories of diseases. The first category consists of functional failure of bone marrow or marrow-derived cells e.g. aplastic anemia, myelodysplastic syndrome (MDS), immunodeficiency syndromes or hemoglobinopathies (thalassemia, sickle cell anemia). The second category includes hematologic malignancies such as acute or chronic leukemia, multiple myeloma, lymphomas, and myeloproliferative neoplasms.[2] The former are more common indications for HSCT in our set up.

The high number of male patients (71%) as compared to 29% females may represent the fact that ours is a male dominated society and the parents give preferential treatment to male progenies when seeking expensive treatment. Data from 18 different transplant centers in the UK for 1825 transplant patients showed a 6.2% incidence of severe infections requiring readmission to the hospital less than 100 days following HSCT.[6]

Table V shows the comparison of our study with few more from around the world in terms of the number of bacterial and fungal infections.

# Conclusions

Our study concludes that HSCT neutropenic patients have a high frequency of infections. Most of these infections occur in the pre-engraftment neutropenic phase. Bacterial infections are more common than fungal infections in these patients. Klebsiella pneumoniae, Pseudomonas aeruginosa, and Acinetobacter baumannii are the most common pathogens causing neutropenic fever.

Ninety-three percent of patients had bacterial infections while 7% of patients had fungal infections. The lower rate of fungal infections as compared to bacterial is most likely due to positive pressure ventilated rooms with HEPA filters and regular air particle counting in our setup.

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