Original Article

Microbiology

Pulmonary Bacterial and Fungal Infections in Cancer Patients with Neutropaenia

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ABSTRACT

Background: The term, "immunocompromised host" describes a patient who is at an increased risk of life-threatening infections as the consequence of a decrease in the immunity. This study dealt with patients who were on radiotherapy or chemotherapy for underlying malignancies. A pulmonary infection is the most common complication in these patients. This study was conducted with the objective of recording the occurrence of opportunistic pulmonary infections in immunocompromised patients.

Materials and Methods: A total number of 100 cancer patients who were undergoing chemotherapy and radiotherapy, with neutropaenia, who were clinically diagnosed to have lower respiratory tract infections, who attended a medical college hospital, were included in the study during December 2008 to May 2010. Their sputum was collected and processed for the detection of various bacteria and fungi.

Results: Out of 100 patients, the cultures of 44 patients showed significiant growth. Among the isolates, *Pseudomonas aeruginosa* was the predominant pathogen which was found to cause pneumonia. Other pathogens which were found were gram negative bacilli and Methicillin Resistant *Staphylococcus aureus.* . A rare bacterium, *Corynebacterium macginleyi,* was isolated from a patient with carcinoma of the lung.

Interpretation and Conclusion:

Patients with malignancy are prone to get infected with highly resistant strains of various bacteria which make the treatment difficult and increase the morbidity and mortality. We need to look out for rare organisms which cause pneumonia in immunocompromised patients.

Key Words: Immunocompromised, Malignancy, Neutropaenia, Pulmonary infections

INTRODUCTION

The term, "immunocompromised host" describes a patient who is at an increased risk for life-threatening infections as the consequence of a decrease in the immunity [1]. During the past few years, the population of immunocompromised hosts has expanded enormously, thus reflecting the increased use of immunosuppressive agents for the treatment of malignancies and collagen vascular diseases, the prevention of rejection in organ transplant recipients and also an increased incidence of HIV infections and malignancies. Unfortunately, the gain in years of useful life through the successful management and treatment of diseases is offset by serious effects on the immune system. As a result, infections, rather than the primary illness, become the leading cause of death in immunocompromised patients. There are many factors which predispose to infections in this patient population, including local factors due to the tumour, specific deficiencies in the host defense mechanisms due to certain malignant processes, and deficiencies in the host defense mechanisms which are secondary to cancer chemotherapy. Neutropaenia is probably the most important factor which predisposes to infections in cancer patients [1]. The lung is one of the most frequently involved organs in a variety of complications, infections being one of the most common ones and they account for about 75% of the pulmonary complications and are associated with high morbidity and mortality [2]. Pneumonia, which is caused by gram negative bacilli, is a major problem for neutropaenic patients and as they cannot mount an adequate inflammatory response, the pulmonary infection may spread rapidly, resulting in extensive necrosis and a high fatality rate.

The present study was undertaken to record the occurrence of various bacterial and fungal agents which caused lower respiratory infections in malignant patients who were on radiotherapy and chemotherapy, with neutropaenia. No viral pathogens were included due to lack of diagnostic facilities.

MATERIALS AND METHODS

A total number of 100 cancer patients who were on chemotherapy and radiotherapy, with neutropaenia, who were clinically diagnosed to have lower respiratory tract infections, who attended a medical college hospital, formed the study group. The study was conducted during December 2008 to May 2010. Out of 100 sputum samples, 44 showed significant growth.

MICROBIOLOGICAL WORKUP

Early morning, expectorated sputum samples were collected from all the patients in separate, sterile, wide mouthed, screw-capped and disposable plastic containers.

The samples were processed immediately in the Microbiology laboratory. The expectorated sputum was used to detect bacterial and fungal pathogens. The quality of the expectorated sputum was assessed both by macroscopic and microscopic examination. Specimens which were clear, thin and watery, with no purulent material, were rejected. Microscopically, Bartlett's scoring method [3] was used to assess the quality of the sputum.

Smears were prepared and subjected to Gram's staining, Ziehl Neelsen staining [20% H_2SO_4 and1% H_2SO_4] and Toluidine O staining. A KOH mount was done for the fungi.

The specimens were cultured on 5% Sheep Blood agar, Mac-Conkey's agar, heated Blood agar, Lowenstein Jensen Medium and Sabouraud's dextrose agar. The plates were incubated at 37^0 C for 18-24 hours in humid air plus 5-10% CO_2. Sabouraud's Dextrose agar slopes were incubated in duplicates, one in room temperature and the other at 37^{0}C for 4 weeks and they were observed for growth at intervals. The Lowenstein Jensen Medium was incubated at 37^{0}C for 4 weeks and it was observed for growth. The identification of the organisms was conducted according to standard laboratory methods [3, 4].

Antibiotic sensitivity testing was done by the Kirby Bauer disc diffusion method according to the CLSI guidelines [5]. The antibiotic discs were selected as per the CLSI guidelines.

Candida, which was present in the direct smear of the sputum, on Gram's staining, appeared as gram positive yeast like budding cells with pseudohyphae. It was regarded as a pathogen after obtaining the same strain in three repeated samples as pure growth, after observing numerous polymorphonuclear leucocytes on Gram's staining of the sputum samples with pseudohyphae (thus showing its invasive nature), and after the patients who were treated with antifungals recovered. All the Candida which were obtained in the Sabouraud's Dextrose agar, were then processed for the identification of the species. The germ tube test was done. All Candida were inoculated on corn meal agar and incubated at 25^{0}C to demonstrate chlamydospore formation and to look for the typical morphology. The sugar assimilation test was done on Yeast Nitrogen Base agar for further speciation, by using the following sugars- glucose, maltose, sucrose, lactose, trehalose, raffinose and galactose.

A rare *Corynebacterium* isolate was obtained from a patient with carcinoma of the larynx. This was isolated in the pure form from the same patient in 3 different samples and the gram staining morphology gave a clue to its pathogenicity. It was identified as lipophilic *Corynebacterium* in our laboratory and was sent to the L.V. Prasad Eye Institute, Hyderabad, for identification by the commercial API-Coryne System for the confirmation of the strain. It was thus identified as *Corynebacterium macginleyi* with a very good profile acceptance of 99.4%.

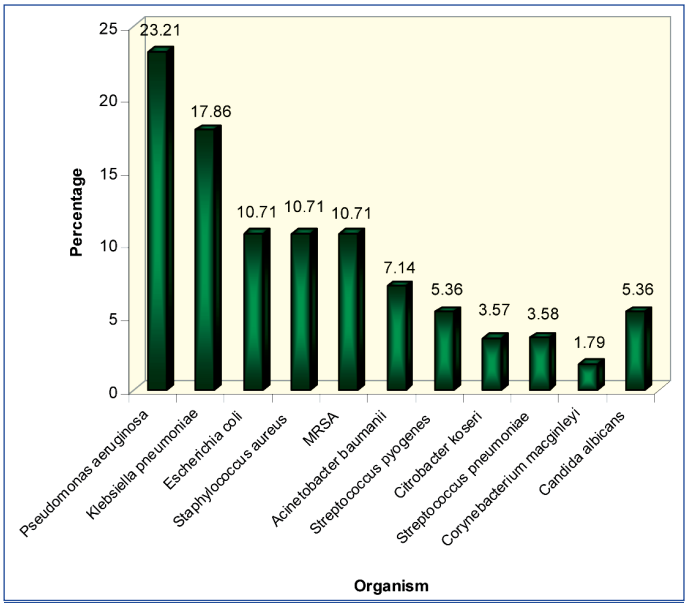
Nocardia was suspected when gram positive, thin, branching filaments were observed on Gram's staining of the sputum smears. Ziehl Neelsen staining with 1% H_2SO_4 was done, which showed acid fast bacilli with branching filaments. The specimen was then inoculated on Lowenstein Jensen medium and Sabouraud's Dextrose agar without antibiotics. The colonies which were obtained were white, glabrous, chalky and wrinkled.

STATISTICAL METHODS

The results were statistically analyzed by using the SPSS version13 software for MS- Windows.

RESULTS

All the 100 patients who were included in this study were on chemotherapy or radiotherapy and had a low neutrophil count, which contributed to their immunosuppression, out of which 44 showed significiant growth. Most of the patients belonged to the age group of 51-60 years (47.7%), followed by those who were of the age group of 41-50 years (15.9%). 10(22.4%) were of the above 60 years age group. A majority of the infections were seen in the 51-60 years age group. Males were more affected (63.6%) than females (36.4%).



[Table/Fig-1]: Organisms which were isolated from malignancy

[Table/Fig-1] shows the overall rank order of the organisms which were isolated.

Among the pulmonary infections, bacterial infections accounted for the maximum number of cases [42 (94.64%)] and fungal infections accounted for 2 cases (5.36%). Gram negative bacilli were the predominant group, accounting for 36 (81%) of the isolates, Pseudomonas aeruginosa (23.21%) being the most common one. Candida albicans was only fungus which was isolated.

The antibiotic sensitivity of the bacterial isolates was as follows: Among the *Staphylococcus aureus* strains (21.42%), 10.71% were *Methicillin resistant Staphylococcus aureus* (MRSA). All the strains were susceptible to amikacin, vancomycin, teicoplanin and linezolid.

Gram negative bacilli showed a wide variety of susceptibilities. The Pseudomonas strains showed a wide variety of susceptibilities. Most of them (76.9%) were resistant to piperacillin, 46.2% were resistant to ceftazidime, 23.1% were resistant to piperacillin-tazobactum, 7.7% were resistant to cefoperazone-sulbactum, 30.8% were resistant to carbapenems and 15.4% were resistant to monobactum-aztreonam. The least resistance was seen against the drug, cefoperazone-sulbactum and thus it helped in the treatment. The resistant strains of Pseudomonas led to a bad prognosis by making the treatment difficult.

Klebsiella is known for its ESBL production. 50% of the Klebsiella strains were resistant to gentamicin, while the resistance to amikacin was only 10%. 40% were resistant to ciprofloxacin, but the resistance to gatifloxacin was only 30%. A resistance to carbapenems (40%) and cefoperazone-sulbactum (40%) was also observed.

The *Escherichia coli* strains were also resistant, with all the strains being resistant to penicillins, the 2nd and 3rd generation cephalo-sporins, the β-lactum β-lactamase inhibitor combination, trimethoprim-suphamethoxazole and ciprofloxacin. 33.3% were resistant to gatifloxacin and 66.7% were resistant to gentamicin, but all were sensitive to amikacin, 16.7% were resistant to imipenem and meropenem and 33.3% were resistant to cefoperazone-sulbactum.

Acinetobacter baumanii, which is known for its inherent capability of high degree resistance, caused severe damage to this group

of patients. All were resistant to the first line drugs. Among the carbapenems, all the strains were resistant to imipenem but only 25% were resistant to meropenem. This (Line missing)

The polymicrobial aetiology of pneumonia was noted in 18.18% of the patients and it was more commonly associated with MRSA, which increased the morbidity of the patients. Most of these were reported from the 51-60 years age group.

DISCUSSION

These patients are defined by their susceptibility to infections which are caused by organisms of low native virulence for the immunologically normal hosts. The survival has improved with the availability of newer antimicrobial agents including azole antifungals, macrolides, antivirals and antiretroviral drugs. Despite these advances, pulmonary infection remains the most common form of tissue-invasive infection in these hosts.

Respiratory infections are very common in cancer patients and are known for their bad prognosis. Granulocytopaenia is the major factor which predisposes to the infections [6], chiefly the bacterial infections. The next most important factor which predisposes to infections is the alteration of the anatomical barriers. Mucosal and skin protection is compromised by tumours, radiotherapy, chemotherapy, surgical diagnostic procedures and therapeutic procedures and intravenous lines and other devices.

In our study, among the pulmonary infections, bacterial infections accounted for the maximum number of infections, followed by fungal infections. *Pseudomonas aeruginosa* was the predominant pathogen which caused pneumonia, followed by *Staphylococcus aureus* and then, *Klebsiella pneumoniae*. This correlated well with the findings of previous studies [7, 8, 9]. The other bacteria which caused infections were *Escherichia coli, Acinetobacter baumanii, Streptococcus pyogenes, Citrobacter koseri, Streptococcus pneumonia* and *Corynebacterium macginleyi. Candida albicans* was the fungus which was isolated. In a study which was done by Sandro et al [8] on lung infections after cancer chemotherapy, it was found that 49% of the patients had bacterial infections. Among the bacterial infections, the pathogens which were identified most commonly were *Pseudomonas aeruginosa* and *Staphylococcus aureus,* followed by *Escherichia coli.*

A study by Sureyya et al [9] on 181 patients of lung cancer showed infections in 84 patients. Most of them suffered from sputum production (65%), cough (59%), auscultation findings (31%) and fever (31%). Gram negative bacteria were the most frequent pathogens which were isolated by culturing the samples. All our patients had presented with fever as an indicator of infection. 84.1% had cough, which indicated respiratory tract infections. 36.4% presented with breathlessness, thus indicating the severity of the infections which involved the lower respiratory tract.

Most of the bacterial strains were resistant to the first line drugs, thus making the treatment difficult. In patients from whom *Acinetobacter baumanii* was isolated, the treatment was difficult because of its inherent resistance to several drugs.

The isolation of a rare pathogen, *Corynebacterium macginleyi,* showed that immunocompromised patients are prone for a wide variety of opportunistic pathogens. Corynebacterium species, other than *C.diphtheriae,* have been referred to as diphtheroids and are considered as colonizers or contaminants. Emerging infections due to diphtheroids, mainly in immunocompromised patients, necessitate an awareness among the microbiologists to speciate them rather than discard them as contaminants.

Less virulent organisms including normal skin flora, commensal bacteria of the oral pharynx or the GIT , environmental fungi and the common community viruses of low level pathogenicity can cause severe, life threatening illnesses in immunocompromised patients. For this reason, a close communication with the diagnostic laboratory is critical, so that the laboratory does not disregard the normal flora and the organisms which are considered to be contaminants, as being unimportant.

Pneumonia is known to get complicated by the infectious agent being able to disseminate easily into the bloodstream and to thus spread to the other organ systems of the body. This makes the treatment difficult and can lead to a bad prognosis.

In the present study, 20.5% of the cases had bacteraemia. The same bacteria with the same susceptibility pattern were isolated from the blood culture of these patients, thus indicating the origin of bacteraemia. Similarly, lung pathogens were isolated from pus samples (11.4%) which were collected from certain wounds in the patient's body. 4.6% of the patients had urinary tract infections which were caused by their lung pathogens, thus suggesting the dissemination of the bacteria.

Four patients with bacteraemia expired, thus suggesting the severity of the primary pathogen.

CONCLUSION

A study on pulmonary infections in immunocompromised individuals showed that they are susceptible to a wide variety of organisms. Individuals with malignancies and those on immunosuppressive therapy are prone to develop pulmonary infections due to highly resistant strains of various bacteria, *Pseudomonas aeruginosa* being the most common one.

CRITERIA FOR INCLUSION

Dr. Meena Dias and Dr. Shreevidya were involved in conducting and analyzing the study, the manuscript preparation, the editing and the reviewing.

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