Prevalence of fungal infections in patients attending tertiary care teaching hospital, middle Gujarat, India

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Abstract
Introduction: With the increasing number of immuno-compromised patients suffering from different types of infections, invasive fungal infections have emerged as a most common cause of morbidity and mortality in these patients. Candida spp. and Aspergillus spp. are the most common causes of fungal infection in immuno-compromised patients. Different types of Candida non-albicans species have emerged as a major cause of candidemia in most of the countries.

Aim: The aim of this study is to detect the prevalence of fungal infection in patients and to study their sensitivity pattern to different antifungal drugs.

Materials and Methods: The study was carried out from January’17 to December’17, to know the fungal infection prevalence, Candida albicans, Aspergillus, opportunistic fungal infections in patients of our institute. During this period total number of 2867 different clinical samples received in the microbiology laboratory for fungal culture and sensitivity testing. All other specimen received in microbiology laboratory was processed using standard microbiological guidelines for yeast and mold isolation. Susceptibility of different fungal species to antifungal agents was performed by using standard guidelines for broth dilution and antifungal disk diffusion susceptibility test.

Results: During the study period total 193 fungal species were isolated from 2867 different clinical specimens received in microbiology laboratory. Candida infection was more common in patient age group between 19-60 yrs., also in male patients and also predominates in admitted patients. Invasive fungal infection was most commonly caused by Candidan-non-albicans. C. tropicalis was highest isolated followed by C.albicans.

Conclusion: All Candida isolated were susceptible to Caspofungin. Candida albicans shows 26% and non-albicans shows 4% resistance to Voriconazole.

Keywords: Fungal Infection, Candida albicans, Aspergillus, Opportunistic infection.

Introduction
With the increasing number of immuno-compromised patients suffering from different types of diseases, invasive fungal infections have emerged as a most common cause of morbidity and mortality in these patients. Also the incidence of opportunistic fungal infections has increased dramatically in the past few decades, resulting from high usage of invasive medical devices and use of broad spectrum antibiotics. The advent of major surgical procedures and its complication further increased the incidence of invasive fungal diseases and associated mortality rates.¹,²

Candida spp. and Aspergillus spp. are the most common causes of fungal infection in immuno-compromised patients.² Different types of Candida non-albicans species have emerged as a major cause of candidemia in most of the countries. Fungal infection increases mortality rates in the range of 18-50%.

To detect invasive fungal infection in febrile neutropenic patients is particularly difficult and time-consuming, but a delay treatment increases higher mortality.² Despite recognition of the clinical importance of invasive fungal infections, these infections remain difficult to diagnose and treat.¹,³,⁴

The knowledge of different candida species virulence factors and their susceptibility testing is clinically very important. Early detection of yeast species is also helpful for early selection of appropriate antifungal agents.³,⁵,⁶

Aim
The aim of this study is to detect the prevalence of fungal infection in patients and to study their sensitivity pattern to different antifungal drugs.

Materials and Methods
The study was carried out from January 17 to December 17, to know the prevalence of fungal infections in different patients of our institute. During this period total number of 2867 different clinical samples received in the microbiology laboratory for fungal culture and sensitivity testing. Blood samples collected in blood culture broths were processed manually. Then positive bottles subcultured on to Sabouraud’s dextrose agar with chloramphenicol, Sheep blood agar (Hi-Media, India) to isolate yeast cells. All other specimen received in microbiology laboratory was processed using standard microbiological guidelines. Candida colonies were confirmed by gram stain and then identified up to species level by conventional methods like colony morphology, growth on corn meal tween 80 agar, germ
tube test, sugar assimilation/fermentation tests, urea hydrolysis and hycrome media. Yeast colonies were identified by direct mount techniques. Susceptibility testing for fungus was performed by using standard guidelines for broth dilution and antifungal disk diffusion susceptibility test. Candida krusei ATCC 6258 was used for quality control.

Result
During the study period total 193 fungal species were isolated from 2867 different clinical specimens received in microbiology laboratory. Out of 193 isolates, 115 (59%) isolated from male patients and 78(41%) from females. And 23 patients were from outpatient department, 158 were indoor patients and rest of 12 patients from ICU (Graph 1).

There was a significant difference in Candida species in different clinical specimens, sex group and also in between OPD, IPD and ICU patients. In patients age group >60 years, male and admitted patients, Candida infection mainly cause by non albicans.

Out of total 193 fungal isolates, 40 (20.7%) were from urine, 37 (19.1%) from blood, 36 (18.6%) from stool, 34 (17.6%) from sputum, 32 (16.8%) from post-operative pus swab, 7 (3.62%) from bronchoalveolar lavage, 4 (2.1%) from frank pus, 2 (1.03%) from pleural fluid and 1(0.51%) from tissue sample.

Table 1 shows the sample wise distribution of fungal isolates. Total 188 different Candida species, 4 Aspergillus species and one Trichosporon were isolated. Amongst 188 Candida species, 69 (36.7%) were Candida tropicalis, 49 (26.06%) Candida albicans, 34 (18.8%) Candida glabrata, 18 (9.57%) Candida parapsilosis, 7 (3.72%) Candida krusei, 3 (1.59%) Candida haemulonii, 2 (1.06%) Candida guillermondii, 1 (0.5%) each of Candida famata and Candida calenulata. And rest of four Candida (2.05%) isolates could not be identified up to species, considered as a Candida species only.

Graph 2 represents that all Candida albicans were susceptible to Caspofungin and Fluycytosin. Candida albicans shows low resistance to Amphotericin B and Fluconazole. 26% albicans strains shows resistance to Voriconazole.

All Candida non albicans shows sensitivity to Caspofungin. And all strains show less than 8% resistance to rest of the anti-fungal drugs. (Graph 3)

| Organism         | urine | Periperal blood | Catheter blood | Stool | Sputum | Swab | BAL | Pus | Pleural fluid | Tissue | Total |
|------------------|-------|-----------------|----------------|-------|--------|------|-----|-----|---------------|--------|-------|
| C. albicans      | 8     | 2               | 0              | 7     | 16     | 11   | 4   | 0   | 1             | 0      | 49    |
| C. tropicalis    | 20    | 7               | 12             | 6     | 8      | 9    | 5   | 1   | 0             | 1      | 69    |
| C. parapsilosis  | 2     | 3               | 7              | 0     | 3      | 2    | 0   | 0   | 1             | 0      | 18    |
| C. glabrata      | 8     | 2               | 1              | 12    | 4      | 2    | 1   | 3   | 1             | 0      | 34    |
| C. krusei        | 2     | 0               | 0              | 1     | 4      | 0    | 0   | 0   | 0             | 0      | 7     |
| C. guillermondii | 1     | 0               | 0              | 0     | 1      | 0    | 1   | 0   | 0             | 0      | 2     |
| C. haemulonii    | 1     | 0               | 0              | 0     | 1      | 0    | 1   | 0   | 0             | 0      | 3     |
| C. famata        | 0     | 0               | 0              | 0     | 1      | 0    | 0   | 0   | 0             | 0      | 1     |
| C. calenulata    | 0     | 0               | 0              | 0     | 1      | 0    | 0   | 0   | 0             | 0      | 1     |
| Candida spp.     | 2     | 1               | 1              | 0     | 0      | 0    | 0   | 0   | 0             | 0      | 4     |
| Aspergillus flavus | 0   | 0               | 0              | 3     | 0      | 1    | 0   | 0   | 0             | 0      | 4     |
| Trichosporon     | 1     | 0               | 0              | 0     | 0      | 0   | 0   | 0   | 0             | 0      | 1     |
| Total            | 40    | 37              | 36             | 34    | 32     | 158  | 6   | 7   | 4             | 2      | 193   |

Graph 1: Demographic distribution of fungal species (n=193)
Graph 2: Susceptibility to antifungal agents pattern of *Candida albicans* (n=49)

Graph 3: Susceptibility to antifungal agents pattern of *Candida non albicans* (n=139)

**Discussion**

Although significant geographic variation is observed among different parts of the world, there appear to follow a specific pattern. In our study, *Candida* non-albicans were more common than *C.albicans*. This finding is correlating with other studies where non-albicans spp. predominates like in Europe, and also in the subcontinents of India.7-11

*Candida* infection was more common in patient age group between 19-60yrs., also in male patients and also predominates in admitted patients. Invasive fungal infection was most commonly caused by Candida non-albicans. Furthermore, invasive Candida infection was mostly caused by non-albicans Candida, whereas the *C.albicans* was found non-invasive infection. The data in our study is highly comparable with other studies from India.3,12,13

In our study, amongst the non albicans *Candida, C.tropicalis* was highest isolated followed by *C.parapsilosis*. This is in the line with previous data from India, which that *C.tropicalis* as the most commonly isolated fungus.

*Candida* species differs in their susceptibility to antifungal agents. All *Candida* isolated were susceptible to Caspofungin. *Candida albicans* shows 26% and non-albicans 4% resistance to Voriconazole. A study from India shown very high resistance to Voriconazole (56%).14 Also *Candida* non-albicans isolates showed less susceptibility to Fluconazole, Amphoterin B and Voriconazole also. The decrease susceptibility of Candida isolates to Voriconazole and some extent to Fluconazole is matter of concern although Amphoterin B, Flucytosin continue to have shown good efficacy. With various types of anti fungals available in the market, it has become necessary to perform anti-fungal susceptibility testing and reporting for effective therapeutic. Evaluation of newer anti-fungal agents is needed.

Resistance to the drugs like Voriconazole and Fluconazole as observed in this study is critical issue in treatment of immune-compromised patients with serious infection.

The Changing epidemiology of candidaemia requires monitoring of different *Candida* species and their susceptibility to use proper therapy and good result. We should also develop guideline for prophylactic empiric therapy based on the epidemiology of India.
Conclusion

Candida infection was more common in patient age group between 19-60 yrs., also in male patients and also predominates in admitted patients. Invasive fungal infection was most commonly caused by Candida non-albicans. C.tropicalis was highest isolated followed by C.parapsilosis. All Candida isolated were susceptible to Caspofungin. Candida albicans shows 26% and non-albicans 4% resistance to Voriconazole.

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