Identification and Antifungal Sensitivity Pattern of Candida Species Isolated from Various Oro-dental Conditions.

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ABSTRACT

Background: Oral candidiasis is the most common oral opportunistic infection seen in immunocompromised patients. Apart from *C. albicans* the non *albicans Candida* species, which are less susceptible to the commonly used antifungal drugs are major etiological agent for candidiasis. Thus, in recent years there has been an increased interest in spectrum of infections caused by *Candida* species. However with the recognition, that *Candida* spp. differ in the production of virulence factor and sensitivity to antifungal agents, greater emphasis has been placed on identification of isolates up to species level. In the past identification of various species of Candida other than *C. albicans* has not been attempted in oral lesions. **Methods:** A total of 158 swabs were collected from oral cavity of patients having lesions suggestive of oral candidiasis. One swab was subjected for direct microscopy using Gram staining. The second swab was inoculated on two tubes of Sabouraud Dextrose agar (SDA) with antibiotics (Hi-Media). **Results:** *Candida albicans* though was the commonest species isolated. , NAC is also emerging as important opportunistic pathogens in oro-dental infections. **Conclusion:** In view of the changing pattern, it is strongly recommended that species identification and sensitivity test can help in much better treatment strategies, and thus, gain a good control over the disease.

Keywords: Candida albicans, Non albicans Candida, oro-dental infections.

INTRODUCTION

Candidiasis is the commonest fungal disease found in humans affecting mucosa, skin, nails and internal organs. Oral candidiasis is the most common form of disease, produced by colonization of *Candida* species also known as oral thrush. First known description of candida infection as oral thrush has been found in Hippocrates "epidemics" from 4th century B.C.^[1] Candida infection may be acute or chronic, superficial or deep and its clinical presentation is wide.^[2,3] *Candida* species are opportunistic pathogens that can cause disease in host who are compromised by underlying local or systemic pathological process.^[4] It is reported that it affects very young, very old and very sick.^[5]

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Fungal infections are often challenging to manage; caution has to be exercised in the use of antifungal drugs. [6] Many current antifungal therapies have

unfortunate clinical side effects and more and more clinical isolates are becoming resistant to traditional antifungal drugs such as fluconazole and amphotericin-B.^[7] The increase in the incidence of fungal infections caused by *Candida* species over the past two decades is significant and non albicans *Candida* (NAC) species continue to replace *Candida albicans* at most of the clinical sites like blood stream infection.^[1,8] *Candida glabrata* and *Candida krusei* are species that have received attention due to their enhanced resistance to certain antifungal agents. Therefore an attempt was made to identify *Candida* isolated from oro- dental lesions upto species level and further determine their antifungal sensitivity pattern.

MATERIALS AND METHODS

The present study was conducted in department of Microbiology Subharti Medical college in collaboration with department of Oral Medicine and Radiology Subharti Dental college over a period of 12 months. A total of 158 swabs were collected from oral cavity of patients attending the outpatient clinics and having lesions suggestive of

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Mohan et al; Antifungal Sensitivity Pattern of Candida Species

oral candidiasis. The patient presented with different oro-dental conditions such as denture wearers, smokers, immunocompromised patient, patients of gingivitis and periodontitis. Two swabs were collected from the oral lesions of each patient, transported and processed for isolation and identification of Candida species. One swab was subjected for direct microscopy using Gram staining. The second swab was inoculated on two tubes of Sabouraud Dextrose agar (SDA) with antibiotics (Hi-Media). The inoculated tubes were incubated at 25°C and 37°C. Any growth appearing on the tubes were identified using standard mycological technique such as colony morphology. Gram staining, germ tube test (GTT) to differentiate C. albicans from NAC species, morphological identification on corn meal agar, colour differentiation on CHROM agar and sugar fermentation tests. Antifungal sensitivity testing was done by disc diffusion method against B, clotrimazole, amphotericin fluconazole itraconazole, voriconazole and ketoconazole (Himedia). Culture was considered as negative and tubes were discarded when there was no growth for maximum of 3 weeks.

RESULTS

A total of 92/158 (58.2%) swabs yielded *Candida* species, out of which 66/92 (71.73%) isolates were identified as *Candida albicans* and 26/92 (28.26%) were identified as NAC spp. [Table 1] Among the 26 NAC species, *Candida tropicalis* (10) was the predominant *Candida* spp. isolated followed by Candida *parapsilosis* (8), *Candida glabrata* (6) and Candida *dublinensis* (2) [Table 2].

Table 1: Distribution of *Candida albicans* and Non albicans *Candida (NAC)* species in clinical isolates. (n=46)

Name	Number	Percentage
Candida albicans	66	71.73%
Non albicans Candida	26	28.26%
(NAC)		

Table 2: Distribution of various Non albicans *Candida* species isolated (n=26)

Candida species	Number	Percentage among NAC's
C. tropicalis	10	38.46%
C. parapsilosis	08	30.76%
C. glabrata	06	23.07%
C.dublinensis	02	7.69%

A total of 80% cases of *Candida* species was isolated from immunocompromised individuals in which the predominant species was *Candida glabrata* ^[4] followed by Candida *albicans* and *Candida dublinensis* (2 each). *Candida albicans*, *Candida tropicalis* and *Candida parapsilosis* were the predominant species isolated from 69% cases of

gingivitis and periodontitis. *Candida albicans* and *Candida tropicalis* were isolated from 60% cases of denture wearers. However, *C. albicans* was the common species seen in patients with habit of smoking (26%) [Table 3].

On antifungal sensitivity testing it was observed that *Candida albicans* isolated was 100% sensitive to amphotericin-B and voriconazole but showed high level of resistance to clotrimazole (30%) and ketoconazole (42%). However, the NAC spp. has shown high level of resistance to fluconazole (61%) and ketoconazole (69%). *Candida glabrata* and *Candida dublinensis*, isolated from case of immunocompromised patient showed resistance to all azoles and was sensitive only to amphotericin-B.

Table 3: Prevalence of *Candida* species in different orodental conditions.

S. no	Condition	Number
		(%)
1.	Immunocompromised status (n=10)	8 (80%)
2.	Patients with Gingivitis and	36(69%)
	Periodontitis(n=52)	
3.	Denture wearers(n=30)	18(60%)
4.	Smokers(n=38)	10(26%)

DISCUSSION

Candida spp. lives commensally in the human oral cavity with high carriage rate (>70%); responsible for candidiasis, the most common opportunistic infection seen in the immunocompromised patients. Although, *C. albicans* is the predominant etiologic agent of candidiasis, numerous records indicated that epidemiology of candidemia has changed with increased incidence of non-Candida albicans Candida species among immunocompromised patients.

In the present study, the prevalence of oro dental candidiasis was 58.2%. This is slightly higher to that reported by Hamester *et al*,^[10] where they reported yeast of genus *Candida* in 52% of their samples. This may be because our study included symptomatic patients with some predisposing factors responsible for oro-dental lesions.

In our study *Candida albicans* (72%) was the predominant species of candida isolated from patients with oro-dental lesions compared to NAC species (28%). This is in accordance with the findings of Batool et al^[8] who reported 75% prevalence of *Candida albicans* and 25% prevalence of NAC species.

Among NAC species *Candida tropicalis* was the predominant species with prevalence of 11%. Similar findings have been reported by Hamester et al,^[10] and Shaheen et al^[11] who reported prevalence of 12% for *Candida tropicalis*. In the present study the second predominant NAC species isolated was *Candida parapsilosis* (8%) followed by *Candida*

Mohan et al; Antifungal Sensitivity Pattern of Candida Species

glabrata and Candida dublinensis with prevalence of 6% and 2% respectively. Similar finding has been reported by Shaheen et al,^[11]. Hamester et al,^[10] reported 2% occurrence of Candida glabrata and Shaheen et al reported 4% occurrence of Candida glabrata. However, Candida dublinensis was not isolated in their study. Batool et al,^[8] reported 4% occurrence of Candida dublinensis in their study.

The positivity rate of Candidiasis was 60% among the denture wearers with Candida albicans and Candida tropicalis as the predominant species isolated. Similar finding has been reported by Salerno et al.[12] Hamester et al.[10] they reported 52% prevalence of Candida species in partial or complete denture wearers with predominance among women. Incidence rate of only 26% was found among smokers showing little effect of smoking on candidiasis, that is lower than that reported by Oliver et al,[13] who found 35% prevalence of Candida spp. among smokers. Candida albicans was the predominant species. among patients with periodontitis and gingivitis incidence rate was 69% which is quite lower than the findings of Batool et al who reported 93% incidence rate of Candidiasis in patients with periodontitis and gingivitis. Among immunocompromised patient incidence οf Candidiasis was found in 80% patients in the present study which is similar to that reported by Badarkar et al, who reported 83.3% incidence rate among immunocompromised individuals.

The antifungal sensitivity pattern of Candida species isolated from samples was studied. The susceptibility pattern of these isolates revealed all isolates of *Candida albicans* were sensitive to amphotericin-B. However, *Candida albicans* has shown high level of resistance to clotrimazole (30%) and ketoconazole (42%) while NAC species has shown high level of resistance to fluconazole (61%) and ketoconazole (69%). Resistance pattern obtained was similar to that reported by Berry et al. [6] Peter G Pappas et al, [9] also reported high level of resistance to fluconazole in NAC species.

CONCLUSION

Candida albicans though was the commonest species isolated. , NAC is also emerging as important opportunistic pathogens in oro-dental infections. NAC species show increase in the resistance to various commonly used antifungal drugs posing a therapeutic problem which is a matter of concern. In view of this changing pattern, it is strongly recommended that species identification and sensitivity test can help in much better treatment strategies, and thus, gain a good control over the disease.^[15] Moreover in future similar studies on a larger group of patients and for

a longer duration needs to be carried out to exactly know the prevalence of different *Candida* species in orodental conditions.

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