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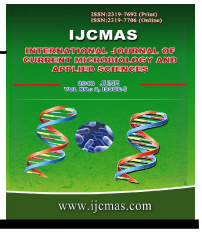


Original Research Article

Antifungal susceptibility pattern of *Candida dublinienses* recovered from HIV infected patients

S. R. More, P. C. Sharma*, S. S. Raut and V. S. Rathod

Department of Microbiology, Dr. Shankarrao Chavan Government Medical College, Nanded, Maharashtra, India- 431601
**Corresponding author e-mail: pcsharma.501@gmail.com*

Keywords

Candida dublinienses; HIV; Fluconazole.

A B S T R A C T

Candida dubliniensis is a recently identified yeast, mostly isolated in HIV-positive individuals. it shares diagnostic characteristics with *Candida albicans*. Oropharyngeal and oesophageal samples from clinically suspected cases of candidiasis were processed in the study. Identification of *Candida dublinienses* was done by conventional standard techniques using Gram's stain, cultural character on Saboraud dextrose agar, germ tube test, morphology on corn meal agar, colour difference on Hichrome Candida differential agar (CHROM agar) and sugar assimilation test by using HIMEDIA candida identification kit KB006. 25.33% of the Candida isolates were *Candida dublinienses*. 15.79% were resistant to Fluconazole, 13.16% were resistant to Clotrimazole and 10.53% were resistant to both Ketoconazole and Itraconazole. Emergence of *C. dubliniensis* infection in HIV seropositive patients is a matter of concern due to the emergence of resistance to commonly used azole antifungals.

Introduction

There is a high interest in *Candida* species other than *Candida albicans* because of the rise and the epidemiological shifts in candidiasis. These emerging Candida species are favored by the increase of immunocompromised patients and new medical practices (Gutiérrez *et al*., 2002). *Candida dubliniensis* is recently identified yeast, mostly isolated in HIV-positive individuals. It is germ tube- and chlamydospore-forming yeast. Thus, it shares diagnostic characteristics with *Candida albicans*. These similarities pose problems in the identification of isolates and have previously led to misidentification of these species. Moreover, an increased resistance to antifungal drugs has been described. This shows the importance of identification of *Candida dubliniensis*. (Schorling *et al.*, 2000) As a result, several identification techniques based on phenotypic and

genotypic characteristics have been developed to differentiate between these Candida species. (Ells *et al*., 2011). Aim and Objectives is to study the prevalence of oropharyngeal and oesophageal *C. dublinienses* species in HIV infected patients. To elucidate the phenotypic characteristics of *C. dublinienses*. To do the antifungal drug susceptibility testing of *C. dublinienses* species isolated.

Materials and Methods

200 hundred HIV positive cases with clinically suspected oropharyngeal and oesophageal candidiasis of all age group and both sexes attending ART Centre of our tertiary care hospital were selected for this study. Patients were explained about the study with informed consent. Clinical details were noted in the proforma and samples were obtained (Figures 1a-d).

Identification of Candida species was done by conventional standard techniques using Gram's stain, cultural character on Saboraud dextrose agar, germ tube test, morphology on corn meal agar, colour difference on Hichrome Candida differential agar (CHROM agar) and sugar assimilation test by using HIMEDIA candida identification kit KB006.

C. dublinienses were identified by culture on SDA which showed White cream, soft, smooth to wrinkled colonies and on microscopy showed globose /oval yeast cells of varying size. Germ tube formation was seen on incubation in serum. Colonies from corn meal agar revealed abundant branched pseudohyphae with blastoconidia and abundant chlamydospores arranged in singles, chains or clusters, terminal or intercalary.

It produced dark green coloured colonies on CHROM agar. On HIMEDIA candida identification kit KB006 the fungus was urease negative and assimilated Maltose, Galactose and Trehalose, while xylose assimilation was strain variable.

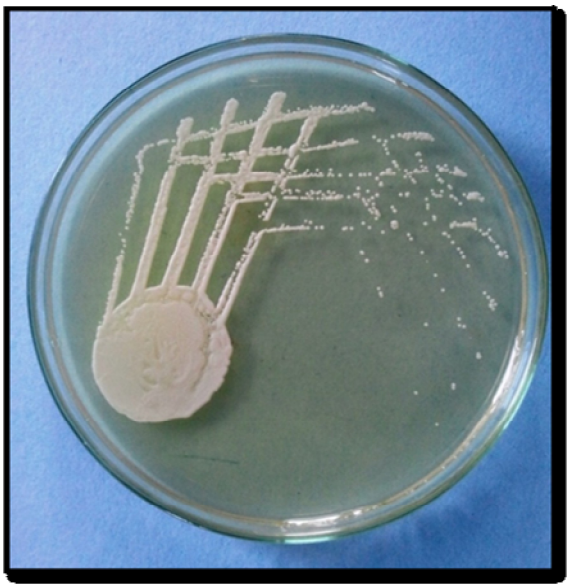
The Kirby Bauer disk diffusion method was used to test the susceptibility of Candida isolates following the Clinical and Laboratory Standards Institute (CLSI) guidelines M44-A for antifungal disk diffusion susceptibility testing of yeasts. Discs applied were Fluconazole (25 µg), Voriconazole (1 µg), Amphotericin B (20 µg), Itraconazole (10 µg), Clotrimazole (10 µg) and Ketoconazole (30 µg). Plates were incubated for 24 hours.

Result and Discussion

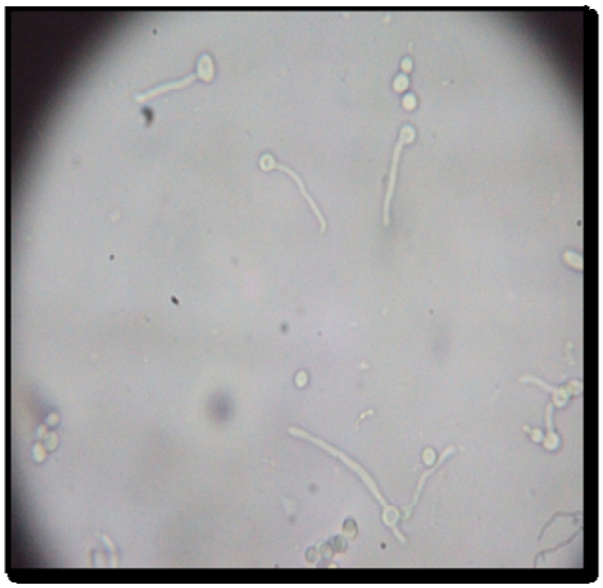
From 200 samples, 150 *Candida* species were isolated. Out of these 150 isolates 38 were *C. dublinienses*, the prevalence being 25.33%. The resistance pattern of *C. dubliniensis* was 15.79% (6/38) resistant to Fluconazole, 13.16% (5/38) resistant to Clotrimazole, 10.53% (4/38) resistant to both Ketoconazole and Itraconazole, 07.89% (3/38) resistant to Voriconazole and 02.63% (1/38) resistant to Amphotericin B (Figure 2).

The prevalence of *C. dubliniensis* in our study was 25.33% . It was similar to study by Nadagir *et al*., (2008) and Sastry *et al*., (2012) who reported the prevalence to be 16.29% and 25% respectively. While it was lower (6.3%) in study by Patel *et al*., (2006). Fluconazole is the most commonly used drug for candidiasis. In our study 15.79% of *C. dublinienses* were resistant to fluconazole which was similar to other studies by Sastry *et al*., (2012) and Deorukhkar *et al*., (2012), who reported it to be 23% and 18.1% respectively. But

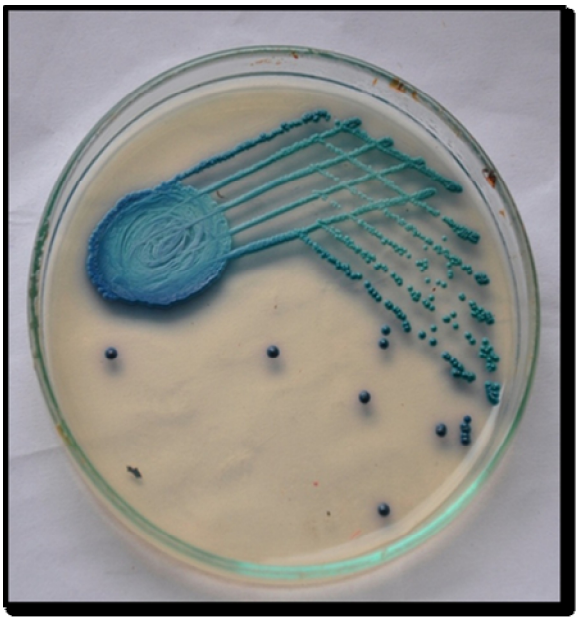
Figure. 1 Photographs of *C. dublinienses*



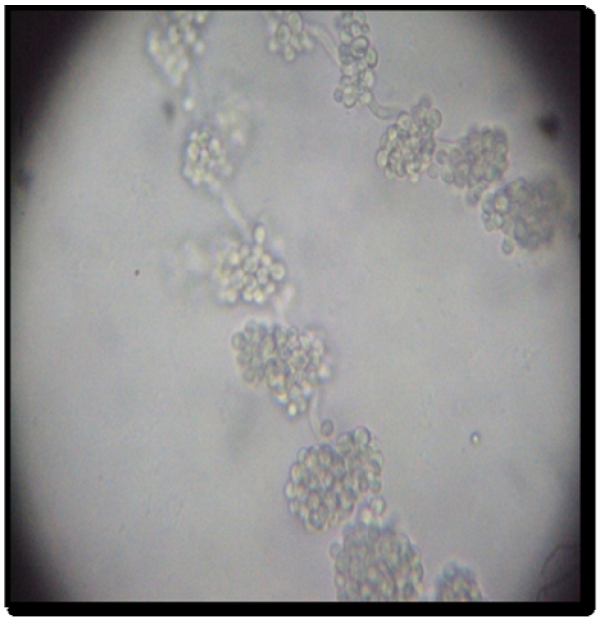
a. *Candida* on SDA Agar plate



b.Germ Tube Formation

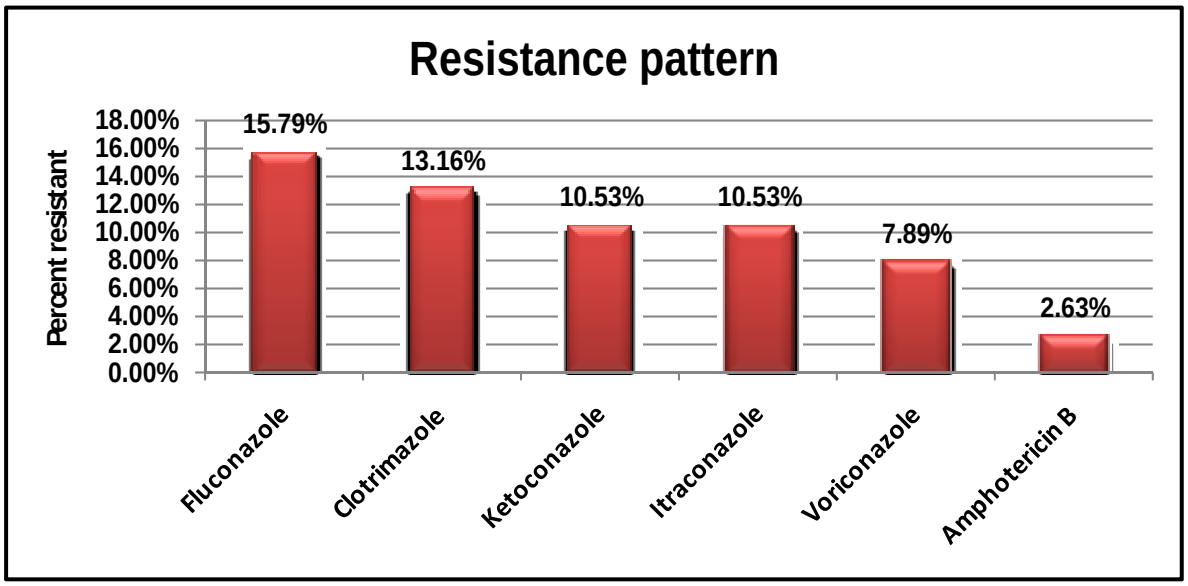


c. *C.dublinienses* Colonies on CHROM agar



d. *C.dublinienses* chlamydoapores

Figure.2 Shows the resistance pattern of the *C. dublinienses*



was higher (33.3%) in study done by Nweze *et al*., (2011). However, isolates of *C. dubliniensis* resistant to Fluconazole may still be sensitive to other azole compounds such as Itraconazole and Voriconazole (Sastry *et al*., 2012), which matches our study results.

The increasing emergence of non-Candida albicans seems to be associated with HIV pandemic. Since *C. dubliniensis* closely resembles *C.albicans* phenotypically it is possible that it is being missed in most of laboratories where only germ tube is solely used of the identification of *C.albicans*. Emergence of *C. dubliniensis* infection in HIV seropositive patients is a matter of concern due to the emergence of resistance to commonly used azole antifungals. Replacement of *C. albicans* by *C. dubliniensis* is known to occur in patients treated with Fluconazole. The antifungal pressure exerted by this drug influences the microbial ecology in these patients, as species that are better able to adapt to antifungal pressure persist over those that are suppressed by the treatment.

Acknowledgement

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