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Candida species distribution and fluconazole susceptibility of blood isolates at a regional hospital in Passo Fundo, RS, Brazil

Distribuição de espécies de Candida *e suscetibilidade ao fluconazol de isolados de hemocultivos em hospital regional de Passo Fundo, RS, Brasil*

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ABSTRACT

Introduction: Candidemia is a bloodstream infection produced by *Candida* genus yeasts. **Objectives**: The purpose of this study was to characterize the epidemiology and the fluconazole susceptibility in *Candida* species isolated from patients at a regional hospital in Passo Fundo, RS. **Methods:** Records from the laboratory were used to identify patients with positive blood cultures for *Candida* between 2010 and 2011. The *in vitro* activity of fluconazole was determined using the disk diffusion method. **Results:** Were analyzed 24 positive blood cultures for *Candida* and found a 54.16% mortality rate. *C. albicans* was the most prevalent species, followed by *C. parapsilosis* and *C. krusei*. For susceptibility to fluconazole, *C. albicans*, *C. parapsilosis* and *C. tropicalis* showed 100% sensitivity. However, *C. krusei* was 100% resistant; and *C. glabrata*, 50% resistant. **Conclusion:** The high mortality and fluconazole resistance rates emphasize the importance of the diagnosis of candidemia in a hospital environment.

Key words: candidemia; fluconazole; *Candida albicans*.

INTRODUCTION

Bloodstream infections by *Candida* genus yeasts are the fourth cause of septicemia in US hospitals[1]. The most frequently isolated species in hospitals is the *Candida albicans*[2-4]; however, non-*albicans Candida* species may be responsible for more than half of candidemia cases[5]. And as the number of reports about the antifungal-resistant *Candida* species has been increasing[6-9], the early establishment of an adequate treatment of candidemia is associated with a favorable prognosis[10].

In the latest decades, the number of publications that document improved attention to health regarding candidemia in different populations has increased[11]. Due to variations in the geographical distribution of *Candida* species able to produce septicemia in different regions of the planet, local epidemiological data continue to be very important[12]. Knowing the resistance profiles of *Candida* species to fluconazole plays an important role in the management of candidemia, considering that this antifungal drug is used for prophylaxis in hospitalized patients[13].

OBJECTIVES

The current work was aimed at identifying species of the *Candida* genus isolated from blood cultures of hospitalized patients at Hospital São Vicente de Paulo (HSVP), in the city of Passo Fundo (RS), Brazil, and determining their antifungal susceptibility profile to fluconazole.

METHODS

Samples and study period

Samples encompassed yeasts of the *Candida* genus obtained from blood cultures of inpatients at HSVP, between December

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2010 and December 2011. Participants' data were transcribed from clinical information of candidemia patients provided by the microbiology sector of the HSVP clinical laboratory.

Inclusion criteria

All blood samples sent to the HSVP clinical laboratory for culture and Gram staining were used. They were collected from inpatients whose results were positive for *Candida* genus yeasts.

Exclusion criteria

Candida samples from the same patient that presented growth in different days were excluded, that is, just one sample from each patient was included.

Sample processing

Positive blood cultures were detected at an automated system, by a conventional method.

Yeast identification

Yeast identification was performed by Gram staining, macromorphology and micromorphology of the colonies, germ tube formation, growth characteristics of *Candida* on a chromogenic medium, auxanogram and zimogram methods[14, 15]. Samples were processed at the HSVP clinical laboratory and at the mycology laboratory of Universidade Regional Integrada (URI) Erechim.

Fluconazole susceptibility test

The susceptibility tests were carried out by the M44-A2[16] method. Fluconazole 25 mcg disks, produced by Centro de Controle e Produtos para Diagnósticos Ltda. (CECON), were used in the research. The standard strain of *Candida albicans* (ATCC 90028) was used as the control.

Ethical aspects

The project under number 085/PGH/2011 was approved by the research ethics committee of URI Erechim and HSVP.

RESULTS

From December 2010 to December 2011, 24 episodes of candidemia occurred at HSVP. Concerning patient distribution according to age group, 12 were adults (62 ± 14.6 years), and 12 were children (7 ± 2.7 years) – among these, seven were newborn. Still among the children, the female:male ratio was 4:8; among the adults, this value was 3:9. The death percentage was 54.17% (13/24), of which seven were children, and six were adults. All participants were on antibiotic drugs, and two used fluconazole. Just a third of the patients were inhabitants of Passo Fundo; the remaining patients lived in other towns of Rio Grande do Sul.

Table 1 shows the distribution of *Candida* species in the studied population. *Candida albicans* was the prevalent species, present in 41.67% of the cases, followed by *C. parapsilosis* (16.67%), *C. krusei* (12.5%), *C. tropicalis* (8.33%) and *C. glabrata* (8.33%). Non-*albicans Candida* species were responsible for 58.33% of candidemia cases. The occurrence of *C. parapsilosis* was observed only in the child population; *C. krusei*, only in adults. The identification of three non-*albicans Candida* species was not made, due to the absence of growth after replica plating.

The fluconazole susceptibility profile of *Candida* species is described in **Table 2**. *C. albicans*, *C. parapsilosis*, and *C. tropicalis* presented 100% sensitivity to fluconazole. In relation to *C. glabrata* isolates, 50% presented fluconazole resistance; and 50%, intermediate profile. All the *C. krusei* isolates presented resistance to fluconazole.

TABLE 1 – Distribution of Candida species in the study population

Species	Adults		Children		Total	
	n	%	*n*	%	*n*	%
C. albicans	4	16.67	6	25	10	41.67
C. parapsilosis	-	-	4	16.67	4	16.67
C. krusei	3	12.5	-	-	3	12.5
C. tropicalis	1	4.17	1	4.17	2	8.33
C. glabrata	1	4.17	1	4.17	2	8.33
Other non-*albicans C.* species	3	12.5	-	-	3	12.5

TABLE 2 – Fluconazole susceptibility test of Candida spp. isolated from inpatients at Hospital São Vicente de Paulo, in Passo Fundo (RS)

Species	*n*	Sensitive	Intermediate	Resistant
C. albicans	10	100%	-	-
C. parapsilosis	4	100%	-	-
C. tropicalis	2	100%	-	-
C. glabrata	2	-	50%	50%
C. krusei	3	-	-	100%
Other non-*albicans C.* species	3	100%	-	-

Interpretation of inhibition zone sizes – sensitive: > 19 mm; intermediate: 19-14 mm; resistant: < 14 mm.

DISCUSSION

Epidemiological surveys conducted in different countries pointed *C. albicans* as the species responsible for most candidemia episodes[17, 18]. In a recent multicentric study carried out in the USA, the most prevalent species were *C. albicans* (50.7%), followed by *C. parapsilosis* (17.4%), *C. glabrata* (16.7%), and *C. tropicalis* (10.2%). Concerning the susceptibility profile to fluconazole, 0.8% of the *C. albicans* isolates, 100% of the *C. glabrata* isolates, 2.9% of the *C. parapsilosis* isolates and 4.9% of the *C. tropicalis* isolates presented resistance[17]. *C. glabrata* is also described as the second most common *Candida* species to produce septicemia[19].

The increased incidence of non-*albicans Candida* species as causing agents of candidemia has been reported. Among these species, *C. parapsilosis* was the most frequent, a result similar to that described in other investigations conducted in Brazil[2, 4, 20, 21]; however, in another survey, *C. tropicalis* was the most frequent non-*albicans* species[3].

In this study, regarding the non-*albicans Candida* species, *C. parapsilosis* was found just in children. The prevalence of this species in the pediatric population is reported in other analyses[21, 22].

Mortality rates higher than 50% were found in other studies[2, 3, 20, 21], besides having been described in surveys in other countries. A retrospective study conducted in Africa between 1990 and 2007 identified 73% mortality in 2.066 candidemia patients[23].

The isolation percentage of *C. krusei* in different anatomical sites ranged from 0.3% to 7.6%[24]. This species is intrinsically resistant to fluconazole[25], as well as *C. glabrata*[26]. Echinocandin has shown to be the most active antifungal drug[24]. *C. tropicalis* resistance has also become a matter of concern, so that in a survey conducted in the state of Ceará, 5.9% of the *C. tropicalis* isolates were resistant to fluconazole and itraconazole[9].

The results of this analysis emphasize the importance of surveillance programs to assess the distribution of antifungal resistance trends in *Candida* species. The differences in the resistance profiles associated to different species stress the need of local epidemiological data capable of guiding treatment at a hospital level.

CONCLUSION

C. albicans was the most common species found in candidemia in patients hospitalized at HSVP, followed by *C. parapsilosis*, *C. krusei*, *C. tropicalis* and *C. glabrata*. This group of pathogens is associated with mortality indices higher than 50% in septicemia. The occurrence of *C. parapsilosis* was observed only in children; while *C. krusei*, only in adults. *C. glabrata* and *C. krusei* may present problems in relation to their profile of resistance to fluconazole.

RESUMO

Introdução: *Candidemia é a infecção da corrente sanguínea produzida por leveduras do gênero* Candida*.* ***Objetivo:*** *Este estudo teve por objetivo caracterizar a epidemiologia e o perfil de suscetibilidade ao fluconazol em espécies de* Candida*, isoladas em pacientes internados em um hospital regional de Passo Fundo, RS.* ***Métodos:*** *Registros laboratoriais foram utilizados para identificar pacientes com hemocultura positiva para espécies do gênero* Candida*, entre 2010 e 2011. A atividade* in vitro *do fluconazol foi determinada por meio do método de difusão em disco.* ***Resultados:*** *Foram analisadas 24 hemoculturas positivas para* Candida*; a taxa de mortalidade encontrada foi 54,16%.* C. albicans *foi a espécie mais prevalente, seguida por* C. parapsilosis *e* C. krusei*.* C. albicans*,* C. parapsilosis *e* C. tropicalis *apresentaram 100% de sensibilidade ao fluconazol, entretanto* C. krusei *mostrou-se 100% resistente; e* C. glabrata*, 50% resistente.* ***Conclusão:*** *A elevada taxa de mortalidade e resistência ao fluconazol enfatiza a importância do diagnóstico de candidemia em ambiente hospitalar.*

Unitermos: *candidemia; fluconazol;* Candida albicans*.*

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