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Epidemiology of candidaemia and antifungal susceptibility patterns in an Italian tertiary-care hospital

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ABSTRACT

The epidemiological and antifungal susceptibility data for 94 episodes of candidaemia in an Italian tertiary-care hospital between January 2000 and August 2003 were evaluated by prospective laboratory-based surveillance. The incidence of fungaemia was 0.90 episodes/10 000 patient-days, and the most common species isolated were *Candida albicans* (40.4%), *Candida parapsilosis* (22.3%), *Candida tropicalis* (16.0%) and *Candida glabrata* (12.8%). Among 24 patients who received antifungal prophylaxis, non-*albicans Candida* spp. were more prevalent than *C. albicans* (p 0.012). The 30-day mortality rate was high (38.2%), particularly for haematological (71.4%) and solid-organ transplant patients (50.0%), and in individuals with *C. tropicalis* and *C. glabrata* bloodstream infections (60.0% and 50.0%, respectively). In-vitro susceptibility tests demonstrated that 95% of the isolates were susceptible to amphotericin B (MIC < 2 mg/L), 98.1% to posaconazole (MIC < 1 mg/L), 95.8% to flucytosine (MIC < 32 mg/L) and fluconazole (MIC < 64 mg/L), and 94.7% to itraconazole (MIC < 1 mg/L). Posaconazole was active (MIC 0.5 mg/L) against all three isolates of *Candida krusei*, which had reduced susceptibility to both fluconazole and itraconazole. Overall, non-*albicans Candida* spp. accounted for 60% of the episodes of candidaemia, which could be related to the use of antifungal prophylaxis. Resistance is still uncommon in *Candida* spp. recovered from blood cultures. The in-vitro activity of posaconazole is encouraging, and this agent could play an important role in the management of invasive candidiasis, including episodes caused by inherently less susceptible species such as *C. krusei*.

Keywords Amphotericin B, antifungal susceptibility, bloodstream infections, *Candida* spp., posaconazole, susceptibility

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INTRODUCTION

During the past two decades, the frequency of invasive fungal infections has increased dramatically in hospitalised patients throughout the world, and *Candida* has now emerged as one of the leading causes of bloodstream infections (BSIs) [1,2]. Data from North American and European surveillance programmes of hospital-acquired bacteraemia have revealed that isolates of *Candida* spp. are the fourth most common cause [3,4], accounting for 8–10% of nosocomial BSIs [1]. Risk-factors for invasive candidiasis include improvements in intensive care strategies (i.e., central venous catheters, mechanical ventilation, hyper-alimentation), prolonged stays in intensive care units (ICUs), the development of more aggressive surgical techniques, and the prolongation of survival of critically-ill patients [5]. Two other important factors, observed mainly in cancer patients, are colonisation of mucous membranes by yeasts, and neutropenia, resulting from increased use of antibiotics and anti-neoplastic agents, respectively [6].

The crude mortality rate of candidaemia is high (38–75%) [3,4,7], and the attributable mortality has been estimated at 25–38% [8,9]. During the past 15 years, the prevalence of infections caused by non-*albicans Candida* spp. has increased

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exponentially, so that these organisms now account for >50% of episodes of fungaemia in various surveys. The widespread use of azoles has been suggested as the main factor responsible for this changing epidemiology [10–15].

Candida albicans is generally susceptible to all the antifungal agents available currently. Among the non-*albicans Candida* spp., *Candida tropicalis* and *Candida parapsilosis* are both generally susceptible to azoles, but *C. tropicalis* is less susceptible to fluconazole than is *C. albicans*. *Candida glabrata* is intrinsically more resistant to antifungal agents, particularly to fluconazole. *Candida krusei* is intrinsically resistant to fluconazole, and infections caused by this species are associated strongly with previous fluconazole prophylaxis and neutropenia. *Candida lusitaniae*, accounting for 1–2% of candidaemias, is susceptible to azoles, but has higher intrinsic resistance to amphotericin B [16]. The increase in invasive fungal infections, the associated high mortality rate, and the emergence of antifungal resistance, have all driven the search for more potent antifungal drugs. Posaconazole is a novel antifungal triazole, with potent activity against the yeasts and filamentous fungi that cause systemic infections [17–19].

The aims of the present study were to investigate the epidemiology of candidaemia and to determine the antifungal susceptibility patterns of isolates of *Candida* spp. from a tertiary-care hospital in Italy.

MATERIALS AND METHODS

Surveillance

Prospective surveillance of nosocomial candidaemia was conducted at the University Hospital of Modena, Italy, between January 2000 and August 2003. The hospital has 990 beds, with two adult ICUs, one neonatal ICU, 28 medical wards and 11 surgical wards, including two centres for solid-organ transplantations (kidney and liver/multivisceral transplants). Surveillance was based on data from the microbiology laboratory. Episodes of candidaemia were defined by at least two positive blood cultures yielding *Candida* spp. during a single hospitalisation period. Demographic data, reasons for hospitalisation, antifungal prophylaxis and outcome were collected from medical records.

Organism identification

Blood specimens were processed by an automated blood culture system (BACTEC 9240; Becton Dickinson, Sparks, MD, USA), and viable yeasts were subcultured on Sabouraud dextrose agar (Labobasi, Vallauris, France). Species identification was based on germ tube production, distinctive colour, and morphology on Candida ID2 Agar (bioMérieux, Marcy l'Etoile, France), together with sugar assimilation profiles obtained using the API ID 32C System (bioMérieux). All isolates were stored at −80°C in glycerol 10% v/v.

Susceptibility testing

The antifungal agents tested were fluconazole (Pfizer Inc., New York, NY, USA), posaconazole (Schering-Plough Research Institute, Kenilworth, NJ, USA), itraconazole (Janssen Research Foundation, Beerse, Belgium), and amphotericin B and flucytosine (Sigma Chemical, Milano, Italy). MICs were determined according to NCCLS guidelines [20]. Final concentrations ranged from 0.007 to 4 mg/L for itraconazole and posaconazole, 0.125–64 mg/L for fluconazole and flucytosine, and 0.03–16 mg/L for amphotericin B. MICs were read visually; for posaconazole, itraconazole, fluconazole and flucytosine, the MIC was the lowest drug concentration that resulted in an obvious inhibition of growth (*c.* 50%) compared with that of the growth control; for amphotericin B, the MIC was the concentration in the first well that was optically clear. *C. krusei* ATCC 6258 and *C. albicans* ATCC 90029 were used as control strains in all tests. The interpretative breakpoints for fluconazole, itraconazole and flucytosine were those suggested previously [20,21]. Since in-vitro susceptibility breakpoints for amphotericin B and posaconazole have not yet been established, susceptible breakpoints of <2 mg/L for amphotericin B and <1 mg/L for posaconazole were used, based on pharmacokinetic data [22].

Statistical analyses

The chi-square test was used to evaluate the differences in prevalence between *C. albicans* and non-*albicans Candida* spp. in relation to antifungal prophylaxis and the underlying diseases of patients, to evaluate the 30-day outcome of patients in relation to different wards of admission, causes of hospitalisation and the species of *Candida* responsible for the fungaemia, and to evaluate the correlation between in-vitro susceptibility to fluconazole, itraconazole and posaconazole and the use of azole prophylaxis. To investigate the degree of cross-resistance between azoles, the correlation coefficients between the MICs of fluconazole, itraconazole and posaconazole were determined by the Spearman rho non-parametric method. $p < 0.05$ was considered significant for both analyses.

RESULTS AND DISCUSSION

In total, 94 isolates of *Candida* spp. were collected from blood cultures taken from 86 hospitalised patients. In eight (9.3%) patients, there was simultaneous or subsequent isolation of two different *Candida* spp. Forty-eight (56%) patients were male, and the median age was 61 years (range 0–89 years). The overall incidence of candidaemia was 0.90 episodes/10 000 patient-days. The most common species isolated was *C. albicans* (38 episodes, 40.4%), followed by *C. parapsilosis*