Antifungal Susceptibility Testing and Candidaemia at a Tertiary Referral Hospital

K O'Connell, M Lyons, B Hanahoe, M Cormican
University College Hospital Galway, Newcastle Road, Galway

Abstract

Candidaemia is associated with a high mortality. We have reviewed cases of candidaemia over a 2-year period at a tertiary referral hospital in association with the introduction of routine antifungal susceptibility testing. The aim of the study was two fold; firstly to establish the typical profile of a patient who might experience a Candida bloodstream infection and secondly, to evaluate methods of antifungal susceptibility testing. In 2008-2009, 31 patients with candidaemia were retrospectively identified using the Laboratory Information Systems (Apex). Clinical data were obtained by chart review. Antifungal susceptibility testing to fluconazole and voriconazole was carried out on 20 of the clinical isolates using three different methods. These isolates were also sent to the mycology reference laboratory at Bristol and results were compared. The male-to-female ratio was 2.1:1 with an age range from 6 weeks to 89 years. Candida albicans was the predominant species (n = 17). Patients were predominantly general surgical (39%), oncology (16%) and urology (13%). Identified risk factors included treatment with broad-spectrum antimicrobial agents (89%), central venous catheters (CVCs) (89%), and surgery during the current admission (54%). The crude mortality rate (death prior to discharge) was 42%. Only 1 of the 20 isolates tested, a Candida glabrata, tested resistant to fluconazole. Of 3 antifungal susceptibility test systems evaluated (VITEK® 2, TREK Sensititre® YeastOne® and CLSI disk diffusion), the VITEK® 2 system was considered most appropriate for routine use in our laboratory. Retrospective review of therapy identified 7 patients treated with echinocandins in whom susceptibility testing indicated that fluconazole could have been used with significant reduction in cost of therapy.

Introduction

Candidaemia is defined by the detection of Candida species in blood. Candida species are the fourth most common cause of nosocomial bloodstream infections. Candida albicans is the most common cause of candidaemia, however, there has been an increase in the number of non-albicans species in recent years. It has been suggested that this may be related to selective pressure associated with use of fluconazole. It has been an increase in the number of non-albicans species in recent years. It has been suggested that this may be related to selective pressure associated with use of fluconazole. Of 3 antifungal susceptibility test systems evaluated (VITEK® 2, TREK Sensititre® YeastOne® and CLSI disk diffusion), the VITEK® 2 system was considered most appropriate for routine use in our laboratory. Retrospective review of therapy identified 7 patients treated with echinocandins in whom susceptibility testing indicated that fluconazole could have been used with significant reduction in cost of therapy.

There are an increasing number of agents available for treatment of candidaemia including the newer azoles and the echinocandins. Many of these agents have a spectrum of activity that includes consistent activity against fluconazole-resistant Candida species. Accordingly, these agents are often used in preference to fluconazole for therapy of infection with non-albicans species and for therapy of C. albicans where there is a history of exposure to fluconazole. In recent years, significant advances have been made in the development of standardised methods for antifungal susceptibility testing. Against this background we report a review of two years experience (1st January 2008-31st December 2009) of candidaemia at a tertiary referral hospital in the context of the introduction of routine antifungal susceptibility testing for invasive candidiasis. We sought to establish the typical profile of a patient who might experience a Candida bloodstream infection and also to evaluate methods of antifungal susceptibility testing.

Methods

University Hospital Galway (UHG) is a 546-bed tertiary referral hospital with a 15-bed general intensive care unit/ high dependency unit (ICU/HDU), a 4-bed cardiothoracic ICU, and provides a broad range of services including haematology and oncology. Blood cultures are performed on clinical indication using the Bactec 9240 system. Aerobic, anaerobic and mycotic broth bottles are available, however, in practice clinicians rarely use mycotic bottles. Isolates of yeast are provisionally identified using germ tube and "Quick Brilliance" chromogenic agar, and species is then confirmed biochemically using the VITEK® 2 system. Cases of confirmed candidaemia during the time period 1st January 2008-31st December 2009 were retrospectively identified from the Laboratory Information Systems (Apex).

Clinical data such as risk factors for infection, likely source of infection, management and outcome were obtained by chart review. Twenty recent Candida bloodstream isolates were available from frozen stocks for evaluation of susceptibility test methods. Antifungal susceptibility testing to fluconazole and voriconazole was performed by the mycology reference laboratory at Bristol using the Clinical and Laboratory Standards Institute (CLSI) broth dilution method. Results were compared to susceptibility data generated locally using 3 different antifungal susceptibility test systems, the systems evaluated in our laboratory were: CLSI disk diffusion, TREK Sensititre® YeastOne®, and VITEK® 2 systems. CLSI disk diffusion performed in accordance with CLSI document M2-A9. Clinical systems were used in accordance with the manufacturer's instructions. The American Type Culture Collection (ATCC) quality control strains used were as follows: C. albicans ATCC® 90028™, C. parapsilosis ATCC® 22019™, C. krusei ATCC® 6258™, C. tropicalis ATCC® 750™ and C. glabrata ATCC® MYA 2950™.

Results

There are an increasing number of agents available for treatment of candidaemia including the newer azoles and the echinocandins. Many of these agents have a spectrum of activity that includes consistent activity against fluconazole-resistant Candida species. Accordingly, these agents are often used in preference to fluconazole for therapy of infection with non-albicans species and for therapy of C. albicans where there is a history of exposure to fluconazole. In recent years, significant advances have been made in the development of standardised methods for antifungal susceptibility testing. Against this background we report a review of two years experience (1st January 2008-31st December 2009) of candidaemia at a tertiary referral hospital in the context of the introduction of routine antifungal susceptibility testing for invasive candidiasis. We sought to establish the typical profile of a patient who might experience a Candida bloodstream infection and also to evaluate methods of antifungal susceptibility testing.

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Over the two-year period 1st January 2008-31st December 2009, there were 31 patients with Candida bloodstream infections. The total number of positive blood cultures was 54 as several patients had multiple positive blood cultures during a single episode of candidaemia. In total, 33 isolates of Candida spp. were detected. One patient experienced a mixed infection with Candida albicans and Candida glabrata, and one patient experienced two episodes of candidaemia with a Candida albicans initially, followed 6 weeks later by a Candida glabrata. The male-to-female ratio was 2:1. Patient ages ranged from 6 weeks to 89 years. Candida albicans was the predominant species (n= 17), with C. glabrata second (n=8) and other species including C. parapsilosis, C. tropicalis, C. lusitaniae, C. krusei and C. guilliermondii.

Patients were predominantly general surgical: 12 (39%), oncology: 5 (16%) and urology: 4 (13%). Medical notes were obtained for 28/31 patients. Identified risk factors are displayed in Table 1. It should be noted that many patients had more than one risk factor. The presumed source of infection was CVC in 36% (10/28), gastrointestinal tract in 25% (7/28) and urinary tract in 18% (5/28). The CVC was removed promptly in 84% (21/25) of patients. The antifungal agents that patients received included the echinocandins (caspofungin or anidulafungin): 71% (20/28) of patients, fluconazole 64% (18/28), liposomal amphotericin B: 18% (5/28) and flucytosine; 4% (1/28). Many patients received more than one antifungal agent due to changes in therapy. The crude mortality rate (death prior to discharge) was 42% (13/31). The duration of antifungal treatment was not completely documented in the medical notes in many cases. Two patients were treated with antifungals for 6 weeks. Two patients (6%) had recurrence of their candidaemia. The first of these had a 14-day course of fluconazole and candidaemia recurred 2 weeks after discontinuation of antifungal therapy. The second patient received a 14-day course of caspofungin and candidaemia recurred one month after cessation of therapy.

There is often a delay in making a diagnosis of candidaemia as Candida species are relatively slow growing in standard blood culture media. Once identified, the availability of on-site antifungal susceptibility testing has been reported as having an important role to play in patient care. The costs of antifungal susceptibility testing (3.70 per VITEKF2 card) are dwarfed by the difference in the cost of the newer antifungal agents compared with fluconazole. A 14-day treatment course of iv fluconazole costs of the order of 200 whereas a 14-day course of the least expensive echinocandin costs of the order of 5,000 to 6,000. Antifungal susceptibility testing using the VITEK2 has now been introduced to our laboratory as a routine service in patients with invasive Candida infections that have been put in place or enhanced include antimicrobial stewardship to optimise antimicrobial use, and central line surveillance to monitor CVC-related infection.

Discussion
Bloodstream infections with Candida species are associated with a high mortality. Although Candida albicans was the predominant species reported, there was almost an equal number of non-albicans Candida identified. Many patients in this study had more than one risk factor for candidaemia including surgery, ICU stay, CVCs and receipt of broad spectrum antimicrobials. Interventions to modify identified risk factors for candidaemia that have been put in place or enhanced include antimicrobial stewardship to optimise antimicrobial use, and central line surveillance to monitor CVC-related infection.

Antifungal susceptibility testing was carried out on 20 of the clinical isolates which had been stored using the CLSI disk diffusion method, TREK Sensisititre YeastOne method and VITEK2. Reference laboratory results revealed that the vast majority of Candida spp. were susceptible to fluconazole. Only 1 of the 20 isolates, a Candida glabrata, tested resistant to fluconazole. CLSI disk diffusion gave a 95% (19/20) agreement with the reference laboratory results; however, the one disagreement was a very major error (VME) in that a fluconazole-resistant isolate was falsely identified as susceptible. VITEK2 gave 85% (17/20) agreement but the discrepancies were minor and the resistant isolate was correctly categorised. This method had the shortest turnaround time with the added advantage of 100% agreement with the VME. Results from the disk diffusion method were not readily available for many patients as the MIC results were required in the clinical setting.

CLSI disk diffusion gave a 95% (19/20) agreement with the reference laboratory. The TREK Sensititre YeastOne and VITEK2 methods agreed in 100% of results. Of the three, the VITEK2 method was the most rapid, requiring 18 hours for an isolate to be tested, with the other two methods requiring 24 hours. There was 100% agreement between the reference laboratory results and VITEK2. Reference laboratory results revealed that the VITEK2 results were 100% true in that no discrepancy occurred.

There were 33 isolates of Candida spp. The species distribution was as follows: Candida albicans first (n=17), followed by C. glabrata second (n=8) and other species including C. parapsilosis, C. tropicalis, C. lusitaniae, C. krusei and C. guilliermondii.

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