**Original Research Article**

**In-vitro susceptibility of fluconazole resistant candidemial isolates to mycafungin and anidulafungin**

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**ABSTRACT**

Echinocandins like Mycafungin and Anidulafungin exhibit broad-spectrum activity against Candida spp. The intrinsic or acquired resistance of Candida species to fluconazole is of increasing nowadays. We determined the MICs of Micafungin and Anidulafungin against 21 bloodstream isolates of Fluconazole-resistant Candida species obtained from medical centers between 2011 and 2013. MICs were determined using E test method according to the CLSI reference method M27-A2. RPMI 1640 was used as the test medium, and we used the MIC endpoint of prominent growth reduction at 24 h. Among the 21 Fluconazole-resistant Candida isolates, 5 (23%) were C. krusei, 6 (29%) were C. glabrata, 4 (19%) were C. albicans, and C. tropicalis, 2 (10%) were C. parapsilosis. Micafungin and Anidulafungin had good in vitro activity against all Fluconazole-resistant Candida species tested, the MICs at which 50% (MIC50) and 90% (MIC90) of isolates were inhibited were 0.03 µg/ml and 0.06 µg/ml, respectively. All the Fluconazole-resistant Candida spp. were inhibited at a Micafungin MIC that was <1 µg/ml. So Echinocandins like Micafungin and Anidulafungin has excellent in vitro activity against 21 candidemia isolates of fluconazole-resistant Candida species.

**Keywords**  
Echinocandins; Candida; Antifungal susceptibility; Mycafungin; Anidulafungin; Fluconazole.

**Introduction**

Over the two decades, Non-albicans Candida represented 10-40% of all candidemia and the ratio of Non-albicans Candida species among Candida species is increasing Gudlaugsson et al., (2003). “Little is known about the incidence of Echinocandin- resistance in Candida species. The Echinocandins (Anidulafungin [ANF], Caspofungin [CSF], and Micafungin [MCF]) are lipopeptide antifungal agents that inhibit the synthesis of β1,3-d-glucan in the fungal cellwall and exhibit concentration-dependent fungicidal activity against most species of Candida Cappelletty (2007), Chandrasekar and Sobel (2006).
Echinocandins inhibit β 1,3-d-glucan synthase (GS), which catalyzes the biosynthesis of β 1, 3-d-glucan, the major glucan component of Candida cell walls Douglas (2001). We examined the MICs of Micafungin and Anidulafungin against 21 bloodstream isolates of fluconazole-resistant Candida species. MICs were determined using E test method according to the CLSI reference method M27-A2.

Materials and Methods

Over the two decades, Non-albicans Candida represented 10-40% of all candidemia and the ratio of Non-albicans Candida species among Candida species is increasing Gudlaugsson et al (2003). Little is known about the incidence of Echinocandin-resistance in Candida species. The Echinocandins (Anidulafungin [ANF], Caspofungin [CSF], and Micafungin [MCF]) are lipopeptide antifungal agents that inhibit the synthesis of β 1,3-d-glucan in the fungal cell wall and exhibit concentration-dependent fungicidal activity against most species of Candida Cappelletty (2007), Chandrasekar and Sobel (2006). Echinocandins inhibit β 1,3-d-glucan synthase (GS), which catalyzes the biosynthesis of β 1, 3-d-glucan, the major glucan component of Candida cell walls Douglas (2001). We examined the MICs of Micafungin and Anidulafungin against 21 bloodstream isolates of fluconazole-resistant Candida species. MICs were determined using E test method according to the CLSI reference method M27-A2.

Results and Discussion

The susceptibilities of Anidulafungin, and Micafungin to 21 Fluconazole resistant Candida species from blood stream infections were determined. From the E test readings at 24 h, MIC 90 values for Anidulafungin and Micafungin were 0.004, and 0.008 µg/ml, for C. albicans; 0.004, and 0.008 µg/ml for C. tropicalis, 0.008 and 0.008 µg/ml, for C. glabrata. No resistance was detected for these species at 24 hours.

C. parapsilosis MIC values were relatively high, especially for Anidulafungin. Among the Fluconazole-resistant Candida spp. tested, C. glabrata exhibited Micafungin MICs (MIC90, <0.016 µg/ml), followed by C. albicans (MIC90, 0.07 µg/ml) and C. krusei (MIC90, 0.06 µg/ml).

The two Echinocandins like Micafungin and Anidulafungin has excellent in vitro activity against 21 invasive clinical isolates of Fluconazole-resistant Candida species. Micafungin and Anidulafungin exhibited potent activity againstazole-resistant Candida species. Pfaffer et al. (2010a) Ostrosky-Zeichner et al., (2003) who reported similar activity of Micafungin against Candida spp despite using a more prolonged incubation time of 48 h. It is now well established that cross-resistance between Echinocandins and Fluconazole does not exist Pfaffer and Diekema, (2007); Richards et al., (2008); Silver et al., 2008; Reboli et al., (2007); Pfaffer et al., (2011). The fluconazole resistance and echinocandins sensitive that was observed in our study was considered a consequence of increased drug use, so a close monitoring in the future has been strongly recommended.

The two Echinocandins like Micafungin and Anidulafungin has excellent in vitro activity against 21 Fluconazole-resistant Candidemia isolates. The sporadic
Table 1: Micafungin and Anidulafungin MIC profiles of Fluconazole resistant Candida isolates by E TEST read at 24 hrs

<table>
<thead>
<tr>
<th>Species</th>
<th>No. of isolates tested</th>
<th>Antifungal agent</th>
<th>MIC (µg/ml)</th>
<th>0.002</th>
<th>0.007</th>
<th>0.016</th>
<th>0.032</th>
<th>0.064</th>
<th>0.125</th>
<th>0.25</th>
<th>0.50</th>
<th>1</th>
<th>2</th>
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<th>8</th>
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<tr>
<td>C. albicans</td>
<td>4</td>
<td>Micafungin</td>
<td>Range</td>
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<td></td>
<td></td>
<td></td>
<td>0.007–0.25</td>
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<tr>
<td></td>
<td></td>
<td>Anidulafungin</td>
<td>0.002–0.016</td>
<td>1</td>
<td>3</td>
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<tr>
<td>C. tropicalis</td>
<td>4</td>
<td>Micafungin</td>
<td>0.008–0.5</td>
<td>3</td>
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<td></td>
<td></td>
<td>Anidulafungin</td>
<td>0.008–2</td>
<td>3</td>
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<tr>
<td>C. glabrata</td>
<td>6</td>
<td>Micafungin</td>
<td>0.007–0.06</td>
<td>1</td>
<td>5</td>
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<td>C. krusei</td>
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<td>Micafungin</td>
<td>0.064–0.125</td>
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<tr>
<td>C. parapsilosis</td>
<td>2</td>
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<td>0.125–32</td>
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occurrence of breakthrough infections associated with acquired resistance mechanisms is becoming increasingly recognized. Antifungal susceptibility testing of the Echinocandins against Candida species is becoming recognized as a useful aid in optimizing treatment of Candidemia and testing should be carried out routinely for all invasive infections caused by Candida species and other opportunistic fungal pathogens to monitor epidemiological trends and to detect the emergence of antifungal resistance.

The E-test procedure seems to be a feasible and trustworthy alternative. Standardized MIC of these agents can be performed accurately in the routine clinical laboratory providing real-time results for difficult clinical infections.

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References