Antifungal activity of posaconazole compared with fluconazole and amphotericin B against yeasts from oropharyngeal candidiasis and other infections

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Objectives: The in vitro antifungal activity of posaconazole was compared with that of fluconazole and amphotericin B.

Materials and methods: A microdilution method (M27-A2) was used with 331 clinical yeast isolates.

Results: The geometric mean MICs of posaconazole, fluconazole and amphotericin B were 0.16, 0.91 and 0.15 mg/L, respectively. Posaconazole was markedly more active than fluconazole and was active against 9/11 fluconazole-resistant Candida albicans, and five Candida glabrata had an MIC of posaconazole of 16 mg/L.

Conclusions: These data indicate that posaconazole is a potentially effective antifungal agent for the treatment of mycoses caused by yeasts.

Keywords: susceptibility, opportunistic, Candida

Introduction

Posaconazole is a novel triazole with broad-spectrum in vitro activity against pathogenic fungi, including Aspergillus spp., Candida spp., Cryptococcus spp. and Histoplasma spp.1–20 Posaconazole has also proven to be efficacious in a wide variety of animal models of candidiasis, disseminated aspergillosis, pulmonary histoplasmosis, coccidiodomycosis and disseminated fusariosis.21–24 In the clinic, posaconazole has been effective in the treatment of human oropharyngeal candidiasis, invasive aspergillosis, candidiasis and fusariosis, and posaconazole is unique among the azoles in having activity in the treatment of zygomycosis.21–26

The purpose of this study was to compare the in vitro antifungal activity of posaconazole with that of fluconazole and amphotericin B against a collection of 331 clinically significant yeasts using the standardized microdilution method as described in the NCCLS document M27-A2.27

Materials and methods

The isolates were cultured from patients, none of whom had received posaconazole therapy. The Candida albicans strains were isolated from oropharyngeal samples from HIV-positive patients and the others were from urine, skin and nail sources. The strains included C. albicans (n = 191) (124 C. albicans serotype A and 28 C. albicans serotype B, the remaining 39 isolates were not sero-typed), Candida tropicalis (n = 42), Candida glabrata (n = 32), Candida lusitaniae (n = 30), Candida parapsilosis (n = 20) and Candida famata (n = 16). In addition, strains of C. albicans (ATCC 1001, ATCC 90028), C. glabrata (ATCC 90030) and C. lusitaniae (ATCC 200950, ATCC 200951, ATCC 200952, ATCC 200953,
Results

The in vitro susceptibility results obtained for posaconazole, fluconazole and amphotericin B against the quality control and reference strains are within the ranges that are considered normal for these strains in our internal and the external antifungal susceptibility tests. Table 1 shows the in vitro susceptibility values observed for the clinical strains; shown are the MIC$_{50}$ and MIC$_{90}$, as well as the geometric mean MIC values and the range of MIC values.

The posaconazole geometric mean MIC was 0.16 mg/L, whereas for fluconazole and amphotericin B, the geometric mean MICs were 0.91 and 0.15 mg/L, respectively. Also, the MIC$_{50}$ and MIC$_{90}$ values of posaconazole and amphotericin B (0.125 and 0.5 mg/L, respectively) were lower than the corresponding values of fluconazole (0.5 and 16 mg/L, respectively). The MIC$_{50}$ and MIC$_{90}$ values of posaconazole and amphotericin B inhibited 82.8% and 90% of the tested strains against posaconazole and 56.2% and 93.4% against amphotericin B. Posaconazole was similar or slightly superior to amphotericin B against all isolates. Compared with fluconazole, posaconazole was either similar or superior against all isolates. The incidence of fluconazole-susceptible-dose-dependent and fluconazole-resistant isolates was 12.1% and 5.1%, respectively; the mean fluconazole MIC for these isolates was >16 mg/L. Only five of these isolates had a posaconazole MIC of 16 mg/L. None of the isolates in our collection had an MIC > 2 mg/L of amphotericin B.

### Discussion

In this study, we demonstrate that posaconazole has a superior in vitro activity profile when compared with fluconazole. Previous studies reported MIC$_{50}$ values of posaconazole that differed from ours by 1–3 dilutions. This could be due to geographical and source variations in patterns of in vitro susceptibility, but also to the spectrophotometer readings which may at least partly explain the differences in the reported MIC$_{90}$s. Other authors have found similar MICs of posaconazole for pathogenic yeasts although some interspecies differences have been observed. The in vitro antifungal activity of posaconazole reported here is greater than that reported by

### Table 1. Geometric mean MIC (GM), MIC range, MIC$_{50}$ and MIC$_{90}$ (mg/L) of amphotericin B, fluconazole and posaconazole for 331 Candida isolates

<table>
<thead>
<tr>
<th>Species</th>
<th>Drug</th>
<th>GM</th>
<th>MIC range</th>
<th>MIC$_{50}$</th>
<th>MIC$_{90}$</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Candida albicans</em> (n = 191)</td>
<td>amphotericin B</td>
<td>0.123</td>
<td>0.01–1</td>
<td>0.125</td>
<td>0.5</td>
</tr>
<tr>
<td></td>
<td>fluconazole</td>
<td>0.92</td>
<td>0.06–&gt;256</td>
<td>0.25</td>
<td>32</td>
</tr>
<tr>
<td></td>
<td>posaconazole</td>
<td>0.15</td>
<td>0.015–&gt;16</td>
<td>0.125</td>
<td>1</td>
</tr>
<tr>
<td><em>C. tropicalis</em> (n = 42)</td>
<td>amphotericin B</td>
<td>0.26</td>
<td>0.01–1</td>
<td>0.25</td>
<td>0.5</td>
</tr>
<tr>
<td></td>
<td>fluconazole</td>
<td>0.6</td>
<td>0.125–4</td>
<td>0.5</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>posaconazole</td>
<td>0.117</td>
<td>0.03–0.125</td>
<td>0.125</td>
<td>0.125</td>
</tr>
<tr>
<td><em>C. lusitaniae</em> (n = 30)</td>
<td>amphotericin B</td>
<td>0.156</td>
<td>0.125–1</td>
<td>0.125</td>
<td>0.5</td>
</tr>
<tr>
<td></td>
<td>fluconazole</td>
<td>0.43</td>
<td>0.06–0.125</td>
<td>0.125</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>posaconazole</td>
<td>0.12</td>
<td>0.06–1</td>
<td>0.125</td>
<td>0.125</td>
</tr>
<tr>
<td><em>C. glabrata</em> (n = 32)</td>
<td>amphotericin B</td>
<td>0.162</td>
<td>0.01–1</td>
<td>0.125</td>
<td>0.5</td>
</tr>
<tr>
<td></td>
<td>fluconazole</td>
<td>2.52</td>
<td>0.125–128</td>
<td>4</td>
<td>64</td>
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<tr>
<td></td>
<td>posaconazole</td>
<td>0.38</td>
<td>0.125–&gt;16</td>
<td>0.125</td>
<td>16</td>
</tr>
<tr>
<td><em>C. parapsilosis</em> (n = 20)</td>
<td>amphotericin B</td>
<td>0.06</td>
<td>0.01–0.5</td>
<td>0.03</td>
<td>0.25</td>
</tr>
<tr>
<td></td>
<td>fluconazole</td>
<td>0.45</td>
<td>0.125–64</td>
<td>0.25</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>posaconazole</td>
<td>0.13</td>
<td>0.125</td>
<td>0.125</td>
<td>0.125</td>
</tr>
<tr>
<td><em>C. famata</em> (n = 16)</td>
<td>amphotericin B</td>
<td>0.81</td>
<td>0.125–1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>fluconazole</td>
<td>7.03</td>
<td>2–32</td>
<td>8</td>
<td>16</td>
</tr>
<tr>
<td></td>
<td>posaconazole</td>
<td>0.12</td>
<td>0.06–0.25</td>
<td>0.125</td>
<td>0.25</td>
</tr>
</tbody>
</table>

ATCC 200954, ATCC 64125, ATCC 66035, ATCC 42720) were also tested as internal and external control strains, and in the same way the *C. albicans* Mont-R strain was included (provided by the University of Montpellier, France), a fluconazole-resistant isolate. The NCCLS approved quality control strains (*Candida krusei* ATCC 6258 and *C. parapsilosis* ATCC 22019) were also tested. Control strains were tested in each susceptibility test.

All isolates were stored in sterile distilled water. Before testing, the isolates were subcultured on Sabouraud glucose agar (Biolife, Milan, Italy) at 35°C for 24 h to ensure the inoculum’s purity and viability. Antifungal agents were obtained from their respective manufacturers: posaconazole, Schering-Plough Research Institute (Kenilworth, NJ, USA); fluconazole, Pfizer (Sandwich, UK); and amphotericin B, Bristol-Myers Squibb (Princeton, NJ, USA). Amphotericin B and posaconazole were solubilized in DMSO (UVASOL DMSO; Merck, Darmstadt, Germany), and fluconazole was solubilized in water.

Susceptibility testing was carried out according to the NCCLS document M27-A2.27 RPMI 1640 (with glutamine and without bicarbonate) buffered with MOPS (0.165 M) (Sigma Chemical Co., St Louis, MO, USA) was used. The pH of RPMI 1640 was adjusted to pH 7.0 at 25°C using sodium hydroxide (Panreac, Madrid, Spain). The medium was filter-sterilized (Steritop-GP Units, Millipore, Billerica, MA, USA) and stored at 2–8°C. Antifungal drug concentrations ranged from 0.25 to 256 mg/L for fluconazole, and 0.016–16 mg/L for posaconazole and amphotericin B. A strain was considered resistant to fluconazole and amphotericin B if the MICs were ≥64 and >1 mg/L, respectively. A strain was considered to be fluconazole-susceptible-dose-dependent when the MIC was 16–32 mg/L. The MIC$_{50}$ and MIC$_{90}$s were calculated as the concentrations of antifungal that were able to inhibit 50% and 90% of the isolates, respectively.
Antifungal activity of posaconazole, amphotericin B and fluconazole

Ostrosky-Zeichner et al.1 Of particular note is our finding that posaconazole has excellent activity against C. tropicalis; this contrasts with data from Ostrosky-Zeichner et al. who reported an MIC\(_{50}\) of 1 mg/L. In contrast, the posaconazole MIC\(_{50}\) for oropharyngeal C. albicans isolates was 1 mg/L; this is higher than the value (0.13 mg/L) reported by Ostrosky-Zeichner et al.1 These pathogens, together with other Candida, Cryptococcus and Aspergillus species are responsible for most of the cases of disseminated mycoses in humans. Based on the findings of this study, we would predict that posaconazole would be active against the agents responsible for the majority of moderate and severe mycoses detected in Spain. In particular, the majority of the fluconazole-susceptible-dose-dependent and fluconazole-resistant strains were susceptible to posaconazole. Posaconazole was also highly effective against species such as C. parapsilosis and C. famata and other genera not included in our study against which other promising new antifungal drugs, such as the echinocandins, do not seem to be effective in vitro.1 However, further in vivo tests will be required to confirm these findings.

References


