In Vitro Activity of Anidulafungin and Other Agents against Esophageal Candidiasis-Associated Isolates from a Phase 3 Clinical Trial

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The efficacy of anidulafungin, an echinocandin antifungal agent with potent anti-Candida activity, in treating esophageal candidiasis was tested in a double-blind study versus oral fluconazole. Isolates were identified and tested for susceptibility. Candida albicans represented >90% of baseline isolates. The MIC<sub>90</sub> of anidulafungin for all strains was 0.06 mg/liter.

Anidulafungin is an echinocandin antifungal agent with broad-spectrum activity against Candida species (2, 12, 13, 17, 19, 20), including fluconazole-resistant strains (10, 16); concentration-dependent fungicidal activity; and a long postantifungal effect in vitro and in animal infection models (1, 6, 7, 10, 16, 18). It is available in the United States for intravenous treatment of esophageal candidiasis, a debilitating opportunistic infection among persons with HIV infection (9) for which cross-resistance among azoles may limit treatment options (4, 14). In patients treated with the anidulafungin dosage regimen for esophageal candidiasis (100-mg loading dose followed by 50 mg daily, half of the dosage used for invasive candidiasis), the steady-state mean maximum and minimum plasma concentrations were 4.2 and 1.6 μg/ml, respectively (Eraxis US package insert). Thus, anidulafungin may be a useful alternative to both amphotericin B and theazole antifungal agents in treating severe oral and esophageal candidiasis in persons with HIV infection and AIDS. We determined the in vitro activity of anidulafungin against clinical isolates of Candida spp. from esophageal candidiasis patients, most of them HIV infected, enrolled in a large (601 patients) phase 3 randomized, comparative, double-blind, double-dummy clinical study. The comparator was oral fluconazole, 200 mg administered on day 1 followed by 100 mg daily for 14 to 21 days.

Candida isolates obtained from endoscopic biopsy specimens or brushings (11) were sent to a reference laboratory for identification, using standard methods (8), and susceptibility testing. Standard antifungal powders included anidulafungin (Vicuron, Inc., King of Prussia, PA), fluconazole (Pfizer, New York, NY), voriconazole (Pfizer), caspofungin (Merck, Whitehouse Station, PA), flucytosine (Sigma, St. Louis, MO), amphotericin B (Sigma), and itraconazole (Janssen, Beerse, Belgium). Preparation of stock solutions and broth microdilution susceptibility testing were as detailed in CLSI document M27-A2 (5, 15) for all agents except amphotericin B (tested in antibiotic medium 3). Incubation at 35°C was for 24 h (echinocandins) and 48 h (azoles, amphotericin B, and flucytosine). MICs, determined using a reading mirror, were defined as a prominent decrease in turbidity (ca. 50%), except for amphotericin B (complete growth inhibition).

Overall, 96% of patients in both treatment arms were infected with C. albicans at baseline, with or without additional Candida species. A majority of the non-C. albicans species isolated at baseline were present in mixed infection with C. albicans. The predominance of C. albicans is characteristic of esophageal candidiasis (3). A total of 441 unique baseline isolates were received by the reference laboratory, including 411 of Candida albicans, 23 of Candida glabrata, 3 of Candida tropicalis, 2 of Candida krusei, and one isolate each of Candida pelliculosa and Candida lusitaniae.

Anidulafungin had potent activity against these isolates (Table 1). Its MIC<sub>90</sub> was 0.06 μg/ml, and 99% of strains were inhibited by 0.12 μg/ml. The MIC distribution for caspofungin was similar. Micafungin was not available for testing at the time at which the study was conducted. For all of the azoles, susceptibility was greater than 90%. The MIC<sub>30/90</sub> of fluconazole for the 23 C. glabrata isolates was 8/16 μg/ml, respectively. Fluconazole-resistant strains included 3 of C. albicans and 1 of C. glabrata (MIC, ≥64 μg/ml) as well as the 2 of C. krusei (considered resistant irrespective of MIC). The MIC range of anidulafungin for these 6 isolates was 0.015 to 0.06 μg/ml. As noted previously, there is no cross-resistance between azoles and echinocandins (10, 13, 20).

As reported previously, the overall clinical and mycological efficacy of anidulafungin, evaluated at the end of therapy, was noninferior to that of fluconazole (11). Eradication of Candida from the esophagus was either proven by a negative culture at the time of evaluation or presumed on the basis of endoscopic improvement with no culture obtained (e.g., if there were no lesions to be cultured). On a per-patient basis, which requires eradication of all baseline pathogens from a patient, mycological success rates were 87 and 91% for anidulafungin and fluconazole, respectively (11). Among the Candida isolates tested at the reference laboratory, there were too few in the fluconazole treatment arm that were fluconazole resistant or, in the anidulafungin arm, that had anidulafungin MICs of >0.06 μg/ml to permit correlation between eradication of in-
dividual isolates and level of susceptibility. Currently, attempts are under way to rationalize susceptibility breakpoints for echinocandins (21, 22). These analyses are based on the dosage utilized for the treatment of invasive candidiasis, which, in the case of anidulafungin, is twice that used in the treatment of esophageal candidiasis.

In conclusion, characterization of Candida esophageal isolates from a large clinical trial confirmed the potent in vitro activity of anidulafungin against both susceptible and fluconazole-resistant isolates seen in previous nonclinical studies. When evaluated at the end of therapy, anidulafungin and fluconazole had similar efficacies in eradicating infecting organisms from esophageal lesions.

B. P. Goldstein was an employee of Vicuron, Inc., during the development of anidulafungin for treatment of esophageal candidiasis and is currently a consultant for Pfizer Inc.

REFERENCES

5. CLSI. 2006. Quality control MIC ranges for broth microdilution: informational supplement M27-S2. CLSI, Wayne, PA.

TABLE 1. In vitro susceptibilities of 441 esophageal isolates of Candida spp. to anidulafungin and six other systemically active antifungal agents

<table>
<thead>
<tr>
<th>Antifungal agent</th>
<th>Cumulative % inhibited at the following MIC (µg/ml):</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>≤0.007</td>
</tr>
<tr>
<td>Anidulafungin</td>
<td>24 26</td>
</tr>
<tr>
<td>Caspofungin*</td>
<td>6 24 75</td>
</tr>
<tr>
<td>Fluconazole</td>
<td>36 82</td>
</tr>
<tr>
<td>Voriconazole</td>
<td>71 85 91 93 95</td>
</tr>
<tr>
<td>Itraconazole</td>
<td>1 20 78 91</td>
</tr>
<tr>
<td>Fluconytosine</td>
<td>9 59 77 82 91</td>
</tr>
<tr>
<td>Amphotericin B</td>
<td>0.5</td>
</tr>
</tbody>
</table>

* Caspofungin was tested against 404 isolates.