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In Vitro Activity of a New Pneumocandin Antifungal, L-743,872, against Azole-Susceptible and -Resistant *Candida* Species

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The in vitro activity of a new pneumocandin, L-743,872, was evaluated with 108 strains of *Candida* and compared with the activities of various antifungals. L-743,872 demonstrated the best activity against azole-susceptible and -resistant strains of *C. albicans*, *C. glabrata*, *C. tropicalis*, *C. parapsilosis*, and *C. kefyr* and less activity against *C. krusei*, *C. lusitaniae*, and *C. guilliermondii*.

In spite of a growing list of antifungal agents, there is still a need for new agents that are fungicidal, have a broader spectrum, have fewer side effects, and can be administered both orally and parenterally.

The pneumocandins are fungicidal, acyl-substituted cyclic hexapeptides that noncompetitively inhibit the synthesis of 1,3-β-glucan, which ultimately results in the lysis of growing fungal cells (4–6, 8). L-743,872 is a new water-soluble pneumocandin with a broad spectrum of antifungal activity (4). We compared the in vitro antifungal activity of L-743,872 against clinical isolates with those of the more commonly utilized antifungal agents.

The organisms included clinical specimens recovered from patients with either candidemia, localized mucosal disease, or asymptomatic colonization. The distribution of species included 50 *Candida albicans* isolates, 21 *Candida glabrata* isolates, 10 *Candida tropicalis* isolates, 7 *Candida parapsilosis* isolates, and 5 isolates each of *Candida krusei*, *Candida lusitaniae*, *Candida kefyr*, and *Candida guilliermondii*. The quality control strains included *C. albicans* ATCC 90028, *C. parapsilosis* ATCC 90018, and *C. glabrata* ATCC 90030.

L-743,872 was obtained as a powder from Merck Research Laboratories, Rahway, N.J. Amphotericin B, fluconazole, ketoconazole, itraconazole, and flucytosine were obtained from their respective manufacturers. The MICs of all antifungal agents for all isolates were determined in accordance with the National Committee for Clinical Laboratory Standards M27-P standards by a broth microdilution method (7). A standard inoculum was diluted to a final concentration of 1×10^2 to 5×10^2 CFU/well in microtiter plates.

The MICs of amphotericin B and L-743,872 were defined as the lowest concentrations that inhibited 100% of the visible growth. The MICs of the azoles and flucytosine were defined as the lowest concentrations that inhibited 80% of visible growth compared with the growth control. All assays were done in duplicate to verify the results. Since there are no definitive MIC breakpoints that separate resistant from susceptible strains, we used an MIC of ≥1.0 μg/ml to define azole resistance, except for fluconazole, for which an MIC of ≥16 μg/ml was used.

The minimal fungicidal concentrations (MFCs) of L-743,872 were determined by subculturing 0.01 ml from the first microtiter well demonstrating complete growth inhibition and from all wells with no visible growth onto Sabouraud dextrose agar plates that were incubated at 30°C for 72 h. Afterwards, colonies were counted, and the MFC was defined as the lowest concentration of drug which yielded negative subcultures or fewer than three colonies (9).

L-743,872 had the lowest MICs at which 50% of organisms were inhibited (MIC_{50}s) and the lowest ranges against *C. albicans* and *C. kefyr*, with ranges of 0.05 to 0.80 μg/ml and 0.02 to 0.40 μg/ml, respectively (Table 1). The *C. albicans* strains included 10 isolates for which the MIC_{50} of fluconazole was 40 μg/ml and the MIC_{90} was 80 μg/ml (Table 2). The MIC_{50}s of L-743,872 for *C. albicans* isolates for which fluconazole MICs were ≥16 μg/ml or ≤8 μg/ml were the same (0.20 μg/ml). The second most susceptible group of yeast isolates included 21 *C. glabrata*, 10 *C. tropicalis*, and 7 *C. parapsilosis* isolates. L-743,872 MIC_{50}s were 0.20 μg/ml for all three *Candida* species, with narrow ranges of 0.10 to 0.40 μg/ml for *C. glabrata*, 0.10 to 0.80 μg/ml for *C. tropicalis*, and 0.10 to 1.6 for *C. parapsilosis* (Tables 1 and 3). In addition, for seven *C. glabrata* isolates the MIC_{50}s of fluconazole, ketoconazole, and itraconazole were 40, 0.80, and 1.6 μg/ml, respectively. As was the case with *C. albicans*, the MIC_{50}s of L-743,872 for the strains of *C. glabrata* for which fluconazole MICs were ≥16 μg/ml or ≤8 μg/ml were similar, 0.20 μg/ml for the susceptible group and 0.4 μg/ml for the fluconazole-resistant *C. glabrata* group of isolates (Table 2). The L-743,872 MIC_{50}s for *C. lusitaniae* and *C. krusei* were both 0.80 μg/ml (Table 3). Overall, the MIC_{50}s and the MIC ranges of L-743,872 were slightly higher for these two species than for *C. albicans*, *C. kefyr*, *C. tropicalis*, *C. glabrata*, and *C. parapsilosis.*

Similarly, the MFCs for *C. albicans*, *C. glabrata*, *C. kefyr*, *C. tropicalis*, and *C. parapsilosis* were all 0.40 μg/ml, only one tube higher than the MIC_{50}s for these organisms. The MFCs for *C. lusitaniae* and *C. krusei* were slightly higher, at 1.6 μg/ml.

In contrast, the five isolates of *C. guilliermondii* were much less susceptible to L-743,872 than all of the other species evaluated in this study (Table 3). The MIC_{50} of L-743,872 was 1.6 μg/ml. In contrast to the MFCs for the other isolates tested in this study, the MFC of L-743,872 for *C. guilliermondii* was quite high, at 6.25 μg/ml.

The results of this study confirm the in vitro efficacy of a new water-soluble pneumocandin, L-743,872, against numerous species of yeast. L-743,872 is one of the new agents in the pneumocandin group of antifungals and possesses potent in vitro fungicidal activity against a broad spectrum of *Candida* species.

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TABLE 1. In vitro susceptibility of *C. albicans*, *C. tropicalis*, and *C. glabrata* to L-743,872 and other antifungal agents

Organism (no. of isolates)	Antifungal agent	MIC (μg/ml)			MFC_{50} (μg/ml)
		Range	50%	90%	
C. albicans (50)	L-743,872	0.05–0.80	0.20	0.40	0.40
	Amphotericin	0.01–0.8	0.1	0.2	
	Flucytosine	0.04–>40	0.16	0.64	
	Fluconazole	0.08–>80	0.16	40	
	Itraconazole	0.01–3.12	0.01	0.40	
	Ketoconazole	0.01–0.80	0.01	0.01	
C. glabrata (21)	L-743,872	0.10–0.40	0.20	0.40	0.40
	Amphotericin	0.05–0.80	0.20	0.40	
	Flucytosine	0.04–0.16	0.08	0.08	
	Fluconazole	0.64–80	2.5	40	
	Itraconazole	0.10–1.6	0.20	1.6	
	Ketoconazole	0.01–0.40	0.20	0.40	
C. tropicalis (10)	L-743,872	0.10–0.80	0.20	0.40	0.40
	Amphotericin	0.10–0.40	0.20	0.20	
	Flucytosine	0.08–0.64	0.16	0.32	
	Fluconazole	0.64–80	1.25	40	
	Itraconazole	0.02–6.25	0.05	0.80	
	Ketoconazole	0.01–0.80	0.01	0.40	

L-743,872 demonstrates the lowest MICs and is the most active against *C. albicans*, *C. glabrata*, *C. tropicalis*, *C. kefyr*, and *C. parapsilosis*, with similar MIC_{50}s and narrow ranges. On the other hand, L-743,872 demonstrates less activity against *C. lusitaniae* and *C. krusei*, although the MICs are not much higher than they are for the very susceptible strains of *Candida*. In contrast to the relatively low MICs for the *Candida* species given above, L-743,872 demonstrates the least activity with the highest MICs against *C. guilliermondii*, with an MIC_{50} of 1.6 μg/ml and an MFC of 6.25 μg/ml. Although this MIC is higher than the MICs for the other *Candida* species evaluated, the true activity of L-743,872 against *C. guilliermondii* will eventually depend on its pharmacokinetics and the achievable levels in serum and tissue versus its toxicity in humans.

TABLE 2. Comparison of the in vitro susceptibilities of azole-susceptible and -resistant strains of *Candida* to L-743,872 and other antifungal agents

Organism (no. of isolates)[a]	Antifungal agent	MIC (μg/ml)			MFC_{50} (μg/ml)
		Range	50%	90%	
C. albicans S (40)	L-743,872	0.05–0.80	0.20	0.40	0.40
	Amphotericin	0.01–0.8	0.10	0.20	
	Flucytosine	0.04–>40	0.16	0.64	
	Fluconazole	0.08–10	0.16	2.5	
	Itraconazole	0.01–0.40	0.01	0.10	
	Ketoconazole	0.01–0.05	0.01	0.01	
C. albicans R (10)	L-743,872	0.10–0.80	0.20	0.40	0.40
	Amphotericin	0.02–0.40	0.05	0.20	
	Flucytosine	0.08–1.25	0.16	0.64	
	Fluconazole	20–80	40	80	
	Itraconazole	0.02–12.5	0.40	0.80	
	Ketoconazole	0.01–0.05	0.01	0.04	
C. glabrata S (10)	L-743,872	0.10–0.40	0.20	0.20	0.40
	Amphotericin	0.05–0.80	0.20	0.80	
	Flucytosine	0.04–0.16	0.04	0.80	
	Fluconazole	0.64–2.5	1.25	5	
	Itraconazole	0.10–0.80	0.20	0.40	
	Ketoconazole	0.01–0.10	0.01	0.02	
C. glabrata R (7)	L-743,872	0.05–0.40	0.40	0.80	
	Amphotericin	0.20–0.40	0.40		
	Flucytosine	0.04–0.16	0.08		
	Fluconazole	20–80	40		
	Itraconazole	0.80–1.6	1.6		
	Ketoconazole	0.05–0.40	0.40		

[a] S, fluconazole susceptible; R, fluconazole resistant.

TABLE 3. In vitro susceptibilities of *C. kefyr*, *C. krusei*, *C. lusitaniae*, *C. parapsilosis*, and *C. guilliermondii* to L-743,872 and other antifungal agents

Organism (no. of isolates)	Antifungal agent	MIC (μg/ml)		MFC_{50} (μg/ml)
		Range	50%	
C. kefyr (5)	L-743,872	0.05–0.40	0.20	0.40
	Amphotericin	0.05–0.40	0.20	
	Flucytosine	0.08–0.16	0.08	
	Fluconazole	0.32–2.5	1.25	
	Itraconazole	0.10–0.20	0.20	
	Ketoconazole	0.01–0.02	0.01	
C. krusei (5)	L-743,872	0.40–0.80	0.80	1.6
	Amphotericin	0.80	0.80	
	Flucytosine	5–20	10	
	Fluconazole	40–80	40	
	Itraconazole	0.20–0.40	0.40	
	Ketoconazole	0.01–0.02	0.01	
C. lusitaniae (5)	L-743,872	0.40–0.80	0.80	1.6
	Amphotericin	0.05–0.40	0.10	
	Flucytosine	0.08–0.16	0.08	
	Fluconazole	0.32–10	0.32–0.64	
	Itraconazole	0.02–0.10	0.02	
	Ketoconazole	0.01	0.01	
C. parapsilosis (7)	L-743,872	0.10–1.6	0.20	0.40
	Amphotericin	0.10–0.40	0.20	
	Flucytosine	0.08–1.25	0.08	
	Fluconazole	0.16–1.25	0.32–0.64	
	Itraconazole	0.02–0.10	0.02	
	Ketoconazole	0.01	0.01	
C. guilliermondii (5)	L-743,872	1.6	1.6	6.25
	Amphotericin	0.02–0.80	0.40	
	Flucytosine	0.08–0.16	0.08	
	Fluconazole	5.0	5.0	
	Itraconazole	0.20–0.40	0.20	
	Ketoconazole	0.01	0.01	

The most exciting observation was the remarkably good activity L-743,872 demonstrated against the strains of *C. albicans* and *C. glabrata* for which fluconazole MICs were high. Essentially the same L-743,872 MIC_{50} was demonstrated for both the fluconazole-susceptible and the fluconazole-resistant strains of *C. albicans* and *C. glabrata*. Moreover, L-743,872 also demonstrated good activity with low MICs against several *Candida* species for which the MICs of fluconazole, ketoconazole, and itraconazole were high.

In summary, L-743,872 demonstrates great potential as a novel antifungal compound with potent in vitro fungicidal activity against *C. albicans*, *C. glabrata*, *C. tropicalis*, and *C. parapsilosis*, the four most commonly isolated species causing disseminated and mucocutaneous candidiasis in the United States (1–3). L-743,872 should be a valuable addition to the drugs available for the management of fungal infections. In addition,

L-743,872 should be particularly useful for the management of these clinically resistant candidal infections in immunocompromised patients (10, 11).

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