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COMMUNICATIONS TO THE EDITOR

FK463, a Novel Water-soluble Echinocandin Lipopeptide: Synthesis and Antifungal Activity

Sir:

An increasing number of patients with marked immunosuppression as a result of transplantation, cancer and infection with HIV, has led to the increased, and widespread use of systemic antifungal agents.[1] Currently available treatment is limited to 5-fluorocytosine, amphotericin B and azoles such as fluconazole and itraconazole. However, various problems effect the use of these agents. Among the major concerns are toxicity, rapid development of resistance, fungistatic effect and narrow antifungal spectrum, depending on the agent. In the search for new antifungals, fungal cell wall synthesis is an attractive target due to the presence of a number of fungi-specific enzymes,[2] leading to the expectation of selective toxicity to the parasite and not to the host.

Inhibitors of 1,3-β-glucan synthase, a key component of the cell wall of most pathogenic fungi, in particular of the echinocandin and pneumocandin type, have been the subject of a number of investigations.[3,4] Most analogs have functional groups such as amino[5] or phosphonate[6] moieties that were designed to improve the solubility in water, in consideration of their clinical use as injectable agents. Our group have also reported water-soluble echinocandin-like lipopeptides possessing a sulfate ester moiety in the cyclic hexapeptide nucleus.[7~9] To find a more potent antifungal agent, we focused on the lipophilic acyl side chain and synthesized various compounds with novel side chains. As a result, we have discovered the novel echinocandin-like lipopeptide FK463 (**1**). In this paper, we report the synthesis and antifungal activity of FK463.[10]

The synthesis of FK463 is outlined in Scheme 1. The key intermediate for the acyl side chain, methyl 4-(5-(4-pentyloxyphenyl)isoxazol-3-yl)benzoate (**4**), was prepared by regioselective 1,3-dipolar cycloaddition reaction of 4-methoxycarbonylbenzhydroxamic acid chloride (**2**) and 4-pentyloxyphenylacetylene (**3**). Hydrolysis of the ester group of **4**, followed by condensation with 1-hydroxybenzotriazole gave the corresponding activated ester (**5**). The cyclic peptide nucleus FR179642 (**6**), obtained by enzy-

Scheme 1. Synthesis of FK463 (**1**).

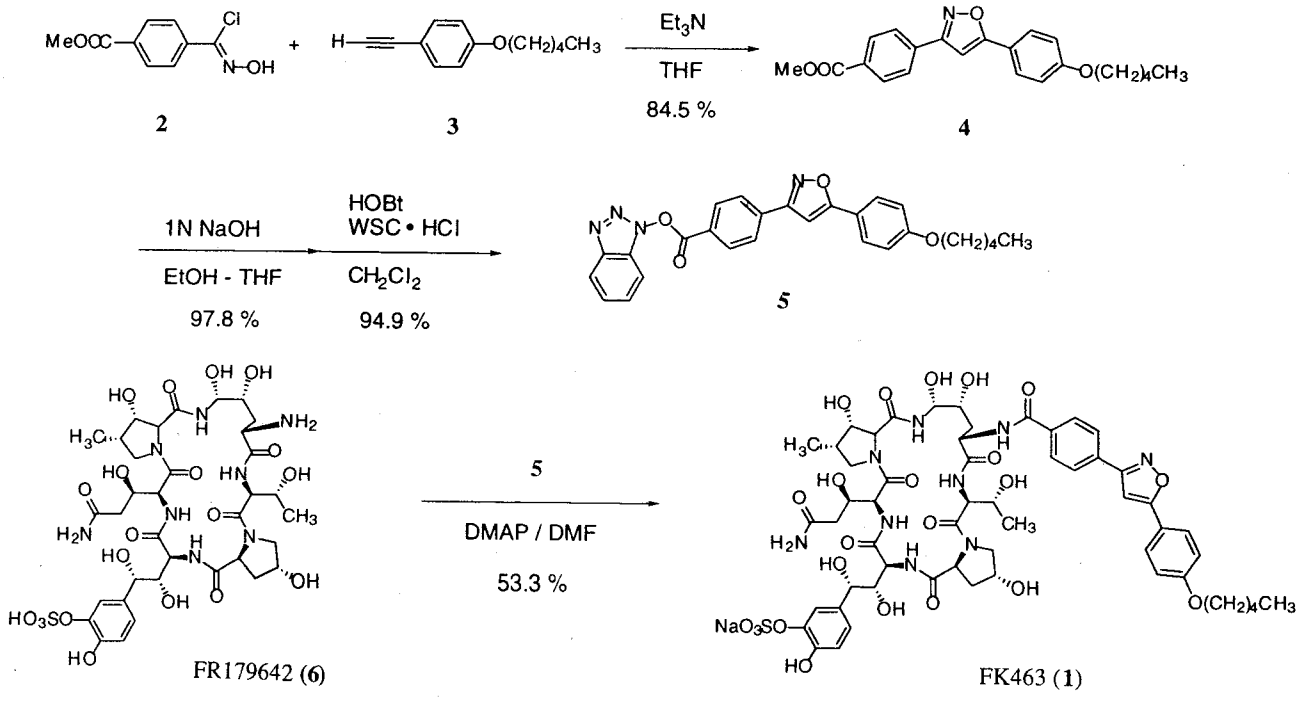

Table 1. Analytical, IR, FAB-MS and ^{1}H-NMR data of **1**.

Elemental Analysis	Calcd for $C_{56}H_{70}N_9NaO_{23}S \cdot 7\ H_2O$ C 47.42, H 5.97, N 8.89 Found C 47.33, H 5.85, N 8.73
FAB-MS	*m/z* 1314 $(M+Na)^+$
IR (KBr) cm^{-1}	3350, 2935, 2873, 1668, 1629, 1538, 1506, 1438, 1257, 1049
^{1}H-NMR (200 MHz, DMSO-d_6) δ	0.9~1.0 (6H, m), 1.08 (3H, d, *J*=5.7 Hz), 1.2~1.6 (4H, m), 1.6~2.0 (5H, m), 2.1~2.4 (3H, m), 2.5~2.6 (1H, m), 3.1~3.2 (1H, m), 3.6~4.6 (15H, m), 4.7~5.2 (10H, m), 5.26 (1H, d, *J*=4.5 Hz), 5.55 (1H, d, *J*=5.9 Hz), 6.7~6.9 (3H, m), 7.0~7.6 (7H, m), 7.85 (2H, d, *J*=8.6 Hz), 7.9~8.2 (4H, m), 8.26 (1H, d, *J*=7.7 Hz), 8.8~9.0 (2H, m)

Table 2. MIC_{90} of FK463 against clinical isolates of fungi.

Organism (No. of strains)	FK463	FLCZ	ITCZ	AMPH-B
C. albicans (37)	0.0156	0.5	0.0313	0.5
C. albicans[a] (4)	0.0313	>64	>8	0.5
C. tropicalis (20)	0.0313	8	0.5	0.125
C. glabrata (20)	0.0156	64	8	1
C. krusei (11)	0.125	64	1	1
C. parapsilosis (17)	1	1	0.5	0.5
C. neoformans[b] (5)	>64	8	0.5	0.5
A. fumigatus (29)	0.0156	>64	0.5	1
A. niger (15)	0.0078	>64	1	0.5

[a] FLCZ-resistant *C. albicans*.

MICs were determined by the microdilution method according to the National Committee for Clinical Laboratory Standard M27-A.

MIC_{90} (μg/ml) : MICs for which growth of 90% of strains are inhibited, respectively.

Culture : 35°C, 2 days ([b] 3days)

matic cleavage of the natural product FR901379,[11] was acylated with **5** to give FK463 (**1**). The structure of **1** was confirmed by elemental analysis, FAB-MS, IR and ^{1}H-NMR (Table 1).

Table 2 shows the antifungal activity of FK463 along with the clinically used antifungal agents itraconazole (ITCZ), fluconazole (FLCZ) and amphotericin B (AMPH-B). FK463 exhibited broad-spectrum activity against clinically important pathogens including *Candida* species and *Aspergillus* species, but displayed no activity against *Cryptococcus neoformans*. FK463 was also effective against azole-resistant *C. albicans* as well as azole-susceptible strains. In comparison with other antifungal agents tested, FK463 showed the most potent activity against *Candida* species and *Aspergillus* species, except *C. parapsilosis*. The further evaluation of FK463 as a clinical candidate and structure-activity relationships (SAR) of a series of FK463-analogues will be published elsewhere.

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