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NOTE

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Revised Structures of Epohelmins A and B Isolated as Lanosterol Synthase Inhibitors from a Fungal Strain FKI-0929

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Abstract The structures of epohelmins A and B isolated as lanosterol synthase inhibitors from a fungal strain FKI-0929 were revised to be 1α-hydroxy-3α-(4′-oxoundec-(5′*E*)-enyl)-pyrrolizidine and 1β-hydroxy-3α-(4′-oxoundec-(5′*E*)-enyl)-pyrrolizidine, respectively, by comparison with spectral data of synthetic compounds.

Keywords epohelmin A, epohelmin B, 1-hydroxy-3α-(4′-oxoundec-(5′*E*)-enyl)-pyrrolizidine, cholesterol biosynthesis, lanosterol synthase

In screening for recombinant human lanosterol synthase inhibitors from microbes, lanopylins [1] and epohelmins [2] were isolated from an actinomycete strain, *Streptomyces* sp. K99-5041, and a fungal strain FKI-0929, respectively.

The structures of lanopylins A_1 and B_1 were assigned as (3*E*)-isohexadecylmethylidene-2-methyl-1-pyrroline and (3*E*)-hexadecylmethylidene-2-methyl-1-pyrroline, respectively, by spectroscopic analyses [1]. In a recent

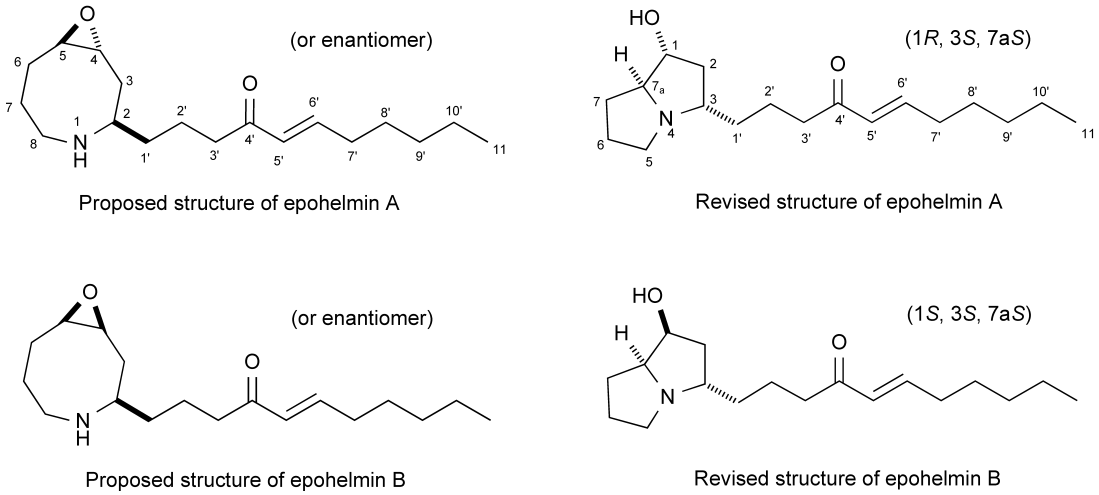

Fig. 1 Structures of epohelmins A and B.

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Table 1 NMR assignment for the acetate salts of epohelmins A and B

	Epohelmin A			Epohelmin B	
Position	^{13}C δ ppm	^{1}H δ ppm (J in Hz)	Position	^{13}C δ ppm	^{1}H δ ppm (J in Hz)
1	74.3	4.17 (1H, td, 6.3, 3.0)	1	70.2	4.38 (1H, br s)
2	40.9	2.06 (1H, m)	2	42.3	2.22 (1H, m)
		2.60 (1H, m)			2.10 (1H, m)
3	67.3	2.99 (1H, m)	3	66.7	3.30 (1H, m)
4			4		
5	51.9	3.43 (1H, dt, 12.0, 7.8)	5	52.9	3.50 (1H, m)
		2.96 (1H, dt, 12.0, 6.0)			2.88 (1H, dt, 11.0, 6.0)
6	24.2	2.04 (2H, m)	6	26.8	2.04 (2H, m)
7	28.4	2.26 (1H, m)	7	24.1	2.21 (1H, m)
		1.72(1H, m)			1.88 (1H, m)
7a	73.9	4.11 (1H, td, 8.3, 3.3)	7a	70.8	4.47 (1H, br dt, 8.5, 4.3)
1′	31.2	1.92 (1H, m)	1′	30.4	1.96 (1H, m)
		1.82 (1H, m)			1.81 (1H, m)
2′	21.1	1.64 (2H, ddt, 7.7, 7.7, 7.7)	2′	21.3	1.65 (2H, ddt, 7.5, 7.5, 7.5)
3′	39.1	2.60 (2H, t, 7.0)	3′	39.3	2.61 (2H, td, 7.1, 2.1)
4′	199.6		4′	199.8	
5′	130.1	6.07 (1H, dt, 16.0, 1.5)	5′	130.2	6.07 (1H, dt, 15.5, 1.5)
6′	148.2	6.83 (1H, dt, 15.7, 6.9)	6′	148.1	6.84 (1H, dt, 16.0, 7.0)
7′	32.5	2.21 (2H, tdd, 7.0, 7.0, 1.5)	7′	32.5	2.21 (2H, tdd, 7.0, 7.0, 1.5)
8′	27.7	1.47 (2H, tt, 7.4, 7.4)	8′	27.7	1.46 (2H, tt, 7.4, 7.4)
9′	31.3	1.31 (2H, m)	9′	31.3	1.30 (2H, m)
10′	22.4	1.29 (2H, m)	10′	22.4	1.32 (2H, m)
11′	13.9	0.90 (3H, t, 7.0)	11′	13.9	0.89 (3H, t, 7.0)

^{1}H (500.00 MHz) and ^{13}C (125.65 MHz) NMR spectra were obtained in chloroform-*d*. Epohelmin A contains 0.85~0.90 equiv of HOAC, while epohelmin B contains 1 equiv of HOAc. The acetate absorbs at δ 2.00~2.01 in the ^{1}H NMR spectra and δ 176.5~177 and δ 22.5~23 in the ^{13}C NMR spectra.

report, the structure of lanopylin B_1 was confirmed by synthesis [3].

Epohelmins A and B were proposed to be two of the diastereomers of 4,5-epoxy-2-(4′-oxoundec-(5′*E*)-enyl)-heptamethylenamine, and their relative stereochemical configurations to be (2*R*, 4*R*, 5*R*) or (2*S*, 4*S*, 5*S*) for epohelmin A, and (2*R*, 4*S*, 5*R*) or (2*S*, 4*R*, 5*S*) for epohelmin B, respectively, by spectroscopic analyses [2]. However, in the process of structural determination, we did not place much importance on the evidence that 4,5-epoxy-heptamethylenamines readily cyclize to 1-hydroxypyrrolizidines [4, 5], and that the methine hydrogens and carbons of *trans* and *cis*-epoxycyclooctanes absorb at δ 2.8~2.9 and δ 59.6~55.6, respectively [6]. Recently, 1α- and 1β-hydroxy-3α-(4′-oxoundec-(5′*E*)-enyl)-pyrrolizidines were synthesized by Snider and Gao [7], and all of the spectral data were identical to those of epohelmins A and B. Revised NMR assignments for epohelmins, which were isolated as the acetate salts, are shown in Table 1. We thus here report the revised structures of epohelmins A and B, 1α-hydroxy-3α-(4′-oxoundec-(5′*E*)-enyl)-pyrrolizidine and 1β-hydroxy-3α-(4′-oxoundec-(5′*E*)-enyl)-pyrrolizidine, respectively, and their absolute configurations as (1*R*, 3*S*, 7a*S*) and (1*S*, 3*S*, 7a*S*), respectively.

References

1. Sakano Y, Shibuya M, Matsumoto A, Takahashi Y, Tomoda H, Ōmura S, Ebizuka Y. Lanopylins A_1, B_1, A_2 and B_2, novel lanosterol synthase inhibitors from *Streptomyces* sp. K99-5041. J Antibiot 56: 817–826 (2003)
2. Sakano Y, Shibuya M, Yamaguchi Y, Masuma R, Tomoda H, Ōmura S, Ebizuka Y. Epohelmins A and B, novel lanosterol synthase inhibitors from a fungal strain FKI-0929. J Antibiot 57: 564–568 (2004)
3. Snider BB, Zhou J. Synthesis of lanopylin B_1. J Org Chem 70: 1087–1088 (2005)

4. Glass RS, Deardorff DR, Gains LH. Pyrrolizidine synthesis by intramolecular cyclization of a substituted azacyclooctane-4,5-oxide. Tetrahedron Lett 33: 2965–2968 (1978)
5. Subramanian T, Lin CC, Lin CC. Synthesis of oxazolidinyl azacycles via ring-closing olefin metathesis: a practical entry to the synthesis of deoxy-azasugars and hydroxypyrrolizidines. Tetrahedron Lett 42: 4079–4082 (2001)
6. Poon THW, Pringle K, Foote CS. Reaction of cyclooctenes with singlet oxygen. Trapping of a perepoxide intermediate. J Am Chem Soc 117: 7611–7618 (1995)
7. Snider BB, Gao X. Structure revision and syntheses of epohelmins A and B. Org Lett 7: 4419–4422 (2005)