NOVEL 2,5-DIHYDROFURYL-γ-LACTAM DERIVATIVES FROM HEMEROCALLIS FULVA L. VAR. KWANZO REGEL

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Three novel 2,5-dihydrofuryl-γ-lactam derivatives, fulvanine A, B and C, have been extracted from Hemerocallis fulva L. var. kwanzo Regel along with compound I. Their respective structures have been established as 1-(3-hydroxymethyl-2,5-dihydrofuryl)-azacyclopenta-3,5-dihydroxy-2-one, 1-(3-methyl-2,5-dihydrofuryl)-azacyclopenta-3,5-dihydroxy-2-one and 1-(3-methyl-2,5-dihydrofuryl)-azacyclopenta-3-hydroxy-5-methoxy-2-one. Compound I was found its ^{1}H and ^{13}C-NMR spectra to be identical to oxypinnatanine.

KEYWORDS Hemerocallis fulva var. kwanzo; 2,5-dihydrofuryl-γ-lactam; Liliaceae; Ehrlich reaction; ^{1}H-NMR; NOE

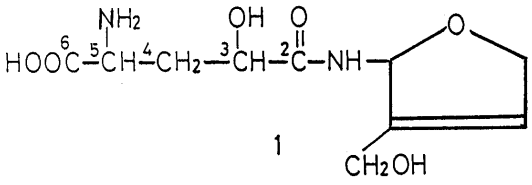
In the course of extensive studies on the distribution of furostanol glycosides,[1] some unidentified compounds from Hemerocallis plants, Liliaceae, were detected on thin-layer chromatographs which showed reddish or bluish purple with Ehrlich reagent. This prompted an investigation of the methanol extract of Hemerocallis fulva L. var. kwanzo Regel, Japanese name "Yabukanzo". We now report the extraction[2] and structures of fulvanine A, B and C along with compound I.

On the IR (KBr) spectrum, compound I (1), was bluish purple with Ehrlich reagent and had absorption maxima due to an amide group: 3350, 1660, 1615 and 1504 cm^{-1}. The ^{1}H-NMR spectrum (D_2O) showed ten non-D_2O exchangeable protons, a pattern essentially identical to that with oxypinnatanine,[3] indicating a vicinal coupling system due to a $-\overset{|}{C}H-CH_2-\overset{|}{C}H-$ structure; and six protons appeared as two sets of nonequivalent methylene proton signals and one-proton multiplet at δ 6.19 and 6.40. 1 was supported by measuring its ^{13}C-NMR spectrum which was assigned according to DEPT and ^{1}H-^{13}C COSY experiments. So compound I was identified as oxypinnatanine, **1**.[3]

Fulvanine A (2), B (3) and C (4), reddish purple with the Ehrlich test, showed a strong absorption maximum at 1700 cm^{-1} due to a γ-lactam ring in the IR (KBr) spectra. The ^{1}H-NMR spectrum (CD_3OD) of 2 showed ten proton signals six of which corresponded to the proton of 1 and was ascribed to each proton of two methylene and two methine groups, which made up 3-hydroxymethyl-2,5-dihydrofuryl moiety. In the double resonance experiments, irradiation of the signal at δ 5.15 transformed the signal at δ 1.98 into a double doublet and sharpened the signal at δ 2.30. Likewise, irradiation at δ 1.98 collapsed the signal at δ 2.30, 4.60 and 5.15. This suggested a $-\overset{|}{C}H-CH_2-\overset{|}{C}H-$ structure in analogy with that of **1**, and the low field shifts of the two methine protons compared with those of 1 apparently resulted from the ring closure between -COOH and >NH groups of 1. Further, ^{1}H-NMR spectrum (DMSO-d_6) exhibited three hydroxyl proton signals at δ 4.96, 5.56 and 5.64 assigned respectively to the positions of $C_{6'}$, C_3 and C_5. These results were indicated by ^{13}C-NMR spectrum in which the 2,5-dihydrofuryl carbon signals were identical with those of 1, and the C_5 carbon indicated a low field shift of about 24 ppm from 1 by the effects of the hydroxylation at C_5 and the formation of the γ-lactam ring. Accordingly, fulvanine A was characterized as 1-(3-hydroxymethyl-2,5-dihydro-2-furyl)-azacyclopenta-3,5-dihydroxy-2-one, **2**.

The ^{1}H and ^{13}C-NMR spectra of 3 showed that the its γ-lactam moiety is identical with that of 2, while the dihydrofuryl moiety have a methyl group at the C_3 position, showing proton and carbon signals at δ 1.68 and 11.73 ppm, respectively, as a substitute for the hydroxymethyl group of **2**. This exchange effect at C_3 appeared in the ^{13}C-NMR spectrum as the upper field shift of a C_3 carbon signal about 5 ppm from that of 2. Therefore, fulvanine B was characterized as 1-(3-methyl-2,5-dihydrofuryl)-azacyclopenta-3,5-dihydroxy-2-one, 3.

The ^{1}H-NMR spectrum of 4 was also analogous to that of 3, except for one methoxy proton signal at δ



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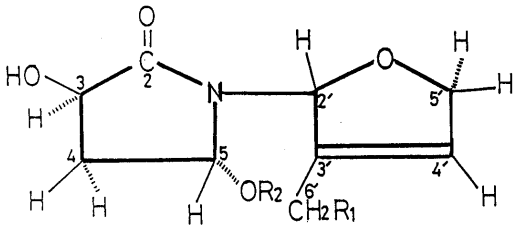

2 : R_1 = OH, R_2 = H
3 : R_1 = R_2 = H
4 : R_1 = H, R_2 = CH_3

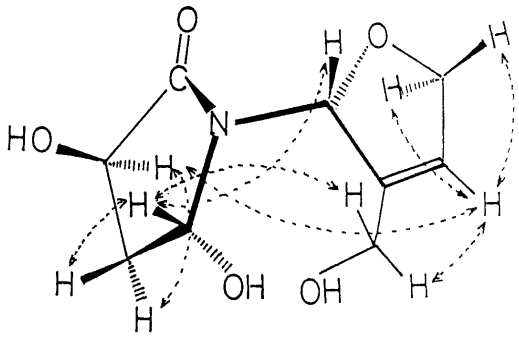

Fig. 1. NOE of **2**

Table I. ^{1}H-NMR Spectral Data for 1, 2, 3, and 4 (J_{H-H} Hz, in Parentheses)

Position	1 (D_2O)	2 (CD_3OD)	3 (CD_3OD)	4 (DMSO-d_6)
3	4.34 dd (9,4)	4.60 dd (9,7)	4.61 dd (9,8)	4.33 ddd (9,8,6) 5.70 d (6,OH)
4	2.22 ddd (15,9,4) 2.34 ddd (15,7,4)	1.98 ddd (13,9,6) 2.30 dd (13,7)	1.98 ddd (13,9,6) 2.30 dd (13,8)	1.81 ddd (13,9,6) 2.31 dd (13,8)
5	3.98 dd (7,4)	5.15 d (6)	5.01 d (6)	4.69 d (6) 3.35 s (CH_3O)
2'	6.40 m	6.45 m	6.29 m	6.15 m
4'	6.19 dddd (4,4,2)	6.22 ddd (4,4,2)	5.96 ddd (5,4,2)	6.02 ddd (4,4,2)
5'	5.63 dddd (14,4,4,2) 4.74 dddd (14,4,4,2)	4.53 dddd (13,4,4,2) 4.76 dddd (13,4,3,2)	4.47 dddd (13,4,4,2) 4.68 dddd (13,5,4,2)	4.37 dddd (14,4,4,2) 4.46 dddd (14,4,4,2)
6'	4.18 dd (14,2) 4.26 dd (14,2)	4.08 m	1.68 m (CH_3)	1.64 m (CH_3)

Table II. ^{13}C-NMR Spectral Data for 1, 2, 3, and 4

Carbon	1 (D_2O)	2 (CD_3OD)	3 (CD_3OD)	4 (DMSO-d_6)
2	178.65	177.92	178.15	177.50
3	71.68	69.50	69.58	67.41
4	35.98	40.14	40.31	34.62
5	55.19	79.09	78.84	84.17
6	175.88			
2'	87.49	88.92	91.16	88.43
3'	138.58	137.63	132.83	131.04
4'	129.08	128.87	127.90	126.72
5'	76.56	75.37	75.43	73.54
6'	58.89	58.04	11.75	11.75
CH_3O				52.29

3.35. Nine protons were assigned on the basis of the data for 3 shown in Table I, and the position bearing the methoxy group appeared to be at C_5 of which the methine proton signal shifted up-field a little from that of 3. This was confirmed by the ^{13}C-NMR spectrum which showed α and ß effects for C_5 and C_4 carbons about 5 ppm, respectively, compared with those of 3. Accordingly, fulvanine C was determined to be 1-(3-methyl-2,5-dihydrofuryl)-azacyclopenta-3-hydroxy-5-methoxy-2-one, 4.

The stereochemistry of 2 was determined by the NOE data as depicted by structure 2 (Fig. 1). The Ehrlich test also seems to be a characteristic of the 2,5-dihydrofuryl moiety in analogy with furost-20(22)-ene or 22-hydroxy-furostane derivatives. Note that fulvanine A, B and C have the unprecedent structure, dihydrofuryl-γ-lactams, indicating the secondary metabolites from the novel amino acid, oxypinnatanine, 1.

REFERENCES AND NOTES

1) S. Kiyosawa, M. Hutoh, T. Komori, T. Nohara, I. Hosokawa, and T. Kawasaki, Chem. Pharm. Bull., 16, 1162 (1968).

2) Extractions and Physical properties of 1-4. The water solution of methanol extract (98g) from a fresh aerial part of Hemerocallis fulva var. kwanzo (6.5Kg) was made to flow through a Diaion (HP 20) column followed by elution with methanol. The MeOH eluate (9.6g) was chromatographed on Sephadex LH-20 (MeOH) to give four fractions (A, 1.20g; B, 1.35g; C, 3.65g; D, 3.8g). Fraction B was subjected in turn to column chromatographies on silica gel ($CHCl_3$-MeOH-H_2O, 8:2:0.2; $CHCl_3$-MeOH, 10:1 or n-hexane-EtOAc, 1:1 V/V) to yield crude 3 and 4, monitoring by TLC with detection by the Ehrlich reaction. The crude fractions was followed by recycle prep. gel partition chromatography (GS-310, MeOH) to give 3 (27mg) and 4 (4mg). From fraction D 2 (80mg) was obtained by the same means as described above. The water solution passed through the Diaion column was evaporated under reduced pressure to yield a viscous residue (98g), which was partitioned three times between n-butanol and water (500 ml each). The water extract (65g) was chromatographed on silica gel (80% EtOH) to yield the positive fraction (4.8g) by the Ehrlich test. Its fraction was subjected to column chromatographed on silica gel ($CHCl_3$-MeOH-H_2O, 3:3:1) and Sephadex G-10 (H_2O), successively, to give a crude 1 (280mg). The crude 1 was further chromatographed by recycle prep. gel partition (GS-310, H_2O), and 1 was obtained as colorless needles (30mg) from H_2O-MeOH solution. Oxypinnatanine (1): colorless needles, mp 152-153°C (dec.).
Fulvanine A (2); colorless needles, mp 118-119°C, $[\alpha]_D$ -0.52° (c=1.0, MeOH). Fulvanine B (3): colorless needles 132-133°C, $[\alpha]_D$ -0.30° (c=0.8, MeOH). Fulvanin C (4): white powder, mp and $[\alpha]_D$ no measured.

3) a) M. D. Grove, M. E. Daxenbichler, D. Weisleder, and C. H. VanEtten, Tetrahedron Letters, 47, 4477 (1971).
b) M. D. Grove, D. Weisleder and M. E. Daxenbichler, Tetrahedron, 29, 2715 (1973).

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