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A NOVEL DISACCHARIDE, WILFORIBIOSE FROM CYNANCHUM WILFORDI HEMSL

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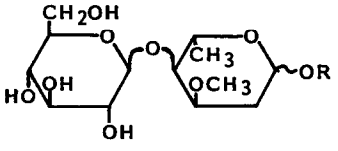
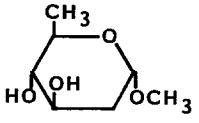
Abstract : The structure of a novel disaccharide, wilforibiose, isolated from the root of Cynanchum wilfordi Hemsl, has been established by spectral data.

The dried root of Cynanchum wilfordi Hemsl (Asclepiadaceae) has been used as a substitute of a tonic, crude drug Ka-shuh-uh (何首烏) which is originated from Poligonaceae plant in Korea. We wish to report the structure elucidation of a novel disachharide named wilforibiose (4) which has a unique 1,4-dioxane linkage in the molecule from a hydrolysate of the crude glycoside mixture of this drug under the following conditions : 0.05 N H_2SO_4-75% MeOH at 60°, 45 min and the subsequent chromatographic separations. Methyl β-(1) and α-glaucobioside (2)[1] was also obtained with 4 (1:2:4=5:3:3).



1 : R=CH_3, β
2 : R=CH_3, α



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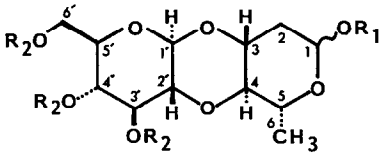


4 : R_1=R_2=H
5 : R_1=CH_3, β ; R_2=H
6 : R_1=CH_3, α ; R_2=H
7 : R_1=R_2=OAc
8 : R_1=CH_3, β ; R_2=OAc
9 : R_1=CH_3, α ; R_2=OAc

Chart 1

Wilforibiose (4), mp 189-191°, $[\alpha]_D$ -23.5° (c=0.98, MeOH), has a molecular formula $C_{12}H_{20}O_8$ by elemental analysis and FD-MS (m/z :293 $(M+H)^+$) and showed a positive Keller-Kiliani reaction suggesting the presence of 2-deoxy sugar moiety. Methyl glycosylation of 4 with 0.5% H_2SO_4 in MeOH at 50° for 30 min gave a mixture of methyl β- (5) and α-wilforibioside (6) accompanied by a small amount of methyl α-D-olivoside (3)[2], $[\alpha]_D$ +78° (c=0.92, H_2O), in one fiftieth of the methyl biosides mixture. This monosaccharide has, however, not been obtained by the other hydrolysis experiments[3].

Table I. ^1H NMR Chemical Shifts for 3 (200 MHz) [4]

1-CH	4.88(1H,dd,J=3.4,1.5 Hz)
2-CHax	2.46(1H,ddd,J=12.7,5.4,1.5 Hz)
2-CHeq	2.01(1H,ddd,J=12.7,11.7,3.4 Hz)
3-CH	4.47(1H,ddd,J=11.7,8.8,5,4 Hz)
4-CH	3.58(1H,t,J=8.8 Hz)
5-CH	4.03(1H,dq,J=8.8,6.4 Hz)
6-CH_3	1.60(3H,d,J=6.4 Hz)
1-OCH_3	3.32(3H,s)

Each of the methyl biosides 5 and 6 was separated by recrystallization from MeOH in the ratio 2:1 as colorless fine needles, 5, mp 186-187°, $[\alpha]_D$ +36.3° (c=0.98, MeOH), and 6, mp 200-203°, $[\alpha]_D$ -70.0° (c=0.96,MeOH) possess the same molecular formulae of $C_{13}H_{22}O_8$ by elemental analysis and FD-MS (m/z : 307 $(M+H)^+$) which suggested the presence of three degrees of unsaturation. The mixture 5 and 6 consumed one mole of periodate indicating the presence of only one glycol structure. On acetylation, this mixture gave triacetates (α-anomer (8) and β-anomer (9)), FD-MS (m/z : 432 (M^+)), which showed no hydroxyl absorption in the IR spectra of it.

Table II. ^1H NMR Chemical Shifts for 5 and 6 (500 MHz) [4]

5		6	
1-CH	4.53(1H,dd,J=9.5,1.8 Hz)	1-CH	4.80(1H,dd,J=3.7,1 Hz)
2-CHax	2.33(1H,ddd,J=11.9,4.3,1.8 Hz)	2-CHax	2.21(1H,ddd,J=12.2,4.9,1 Hz)
2-CHeq	1.89(1H,dt,J=11.9,9.5 Hz)	2-CHeq	1.86(1H,dt,J=12.2,3.7 Hz)
3-CH	3.59(1H,ddd,J=11.9,9.2,4.3 Hz)	3-CH	4.01(1H,ddd,J=12.2,9.2,4.9 Hz)
4-CH	3.29(1H,t,J=9.2 Hz)	4-CH	3.32(1H,t,J=9.2 Hz)
5-CH	3.63(1H,dd,J=9.2,6.4 Hz)	5-CH	3.98(1H,dq,J=9.2,6.4 Hz)
6-CH_3	1.47(3H,d,J=6.4 Hz)	6-CH_3	1.41(3H,d,J=6.4 Hz)
1-OCH_3	3.48(3H,s)	1-OCH_3	3.26(3H,s)
1'-CH	4.76(1H,d,J=7.9 Hz)	1'-CH	4.78(1H,d,J=7.9 Hz)
2'-CH	3.62(1H,dd,J=8.6,7.9 Hz)	2'-CH	3.65(1H,dd,J=9.5,7.9 Hz)
3'-CH	4.21(1H,t,J=8.6 Hz)	3'-CH	4.22(1H,t,J=9.5 Hz)
4'-CH	4.25(1H,t,J=8.6 Hz)	4'-CH	4.21(1H,t,J=9.5 Hz)
5'-CH	4.07(1H,ddd,J=8.6,5.5 1.8 Hz)	5'-CH	4.07(1H,ddd,J=9.5,5.5,1.8 Hz)
6'-CH_2	4.39(1H,dd,J=11.9,5.5 Hz)	6'-CH_2	4.37(1H,dd,J=11.9,5.5 Hz)
	4.58(1H,dd,J=11.9,1.8 Hz)		4.57(1H,dd,J=11.9,1.8 Hz)

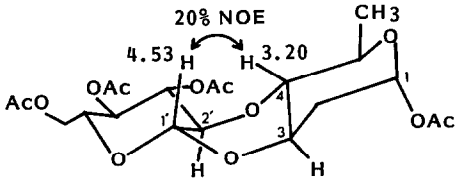
assigned by decoupling experiments

Acetylation of 4 gave α-tetraacetate (7), mp 206-210°, $[\alpha]_D$ -48.4° (c=0.98, $CHCl_3$), $C_{20}H_{28}O_{12}$ (elemental analysis and EI-MS (m/z : 460 (M^+)), whose ^1H NMR spectrum revealed all the proton signals without overlapping showed in Table III. The coupling patterns strongly suggest that this biose consists of glucose and olivose. From the chemical shifts, the acetylated sites are at 3-COH, 4-COH, and 6-COH in the glucose moiety and at 1-COH in the olivose moiety.

Table III. ^1H NMR Chemical Shifts for 7 (500 MHz) [4]

1-CH	6.36(1H,dd,J=3.7,1 Hz)
2-CHax	2.15(1H,ddd,J=11.9,4.6,1 Hz)
2-CHeq	1.91(1H,dt,J=11.9,3.7 Hz)
3-CH	3.95(1H,ddd,J=11.9,9.5,4.6 Hz)
4-CH	3.34(1H,t,J=9.5 Hz)
5-CH	4.08(1H,dq,J=9.5,6.4 Hz)
6-CH_3	1.30(3H,d,J=6.4 Hz)
1'-CH	4.86(1H,d,J=7.9 Hz)
2'-CH	3.74(1H,dd,J=9.8,7.9 Hz)
3'-CH	5.75(1H,t,J=9.8 Hz)
4'-CH	5.50(1H,t,J=9.8 Hz)
5'-CH	4.23(1H,ddd,J=9.8,5.2,2.1 Hz)
6'-CH_2	4.44(1H,dd,J=12.5,2.1 Hz)
	4.53(1H,dd,J=12.5,5.2 Hz)
-OCOCH_3	2.08,2.05x2,2.00(each 3H,s)

These results suggested that this biose has the 1,4-dioxane ring among the hydroxyl groups at C-3 and C-4 of olivose and at C-1 and C-2 of glucose. One of the two structural possibilities was confirmed as the structure (Fig. 1) by NOE measurements of 7 between 4-CH of olivose and 1 -CH of glucose in 20% enhancement. Hence, wilforibiose (4) is assigned to the novel structure : β-D-glucopyranose D-olivopyranose 1',3:2',4-dianhydride.



(δ in $CDCl_3$)

Fig. 1

The ^{13}C NMR (Table IV) for 3, 5, 6, 7, and the mixture of 8 and 9 were measured. The carbon chemical shifts for 3 and 7 were assigned by selective decoupling experiments. Those for 5 and 6 were assigned on the basis of 7, and those for 8 and 9 were on the basis of 5, 6, and selective decoupling experiments.

Table IV. ^{13}C NMR Chemical Shifts for 3, 5, 6, 7, 8, and 9 (50 MHz) [4]

	3	7	5	6	8	9
C-1	99.2	91.8	101.0	98.5	101.2	98.7
2	39.3	34.1	36.6	35.3	36.8	35.5
3	69.3	73.2	75.7	73.7	75.5	73.4
4	79.0	80.9	81.1	81.5	81.1	81.6
5	68.9	68.7	69.6	66.0	70.0	66.3
6	18.7	17.3	17.3	17.3	17.5	17.5
1-OCH_3	54.7		56.2	54.5	56.3	54.6
C-1'		99.0	99.0		99.8	99.9
2'		78.0	78.0		81.5	81.6
3'		72.4	72.4		74.7	74.8
4'		69.7	69.7		71.8	71.9
5'		74.0	73.9		80.4	80.4
6'		62.6	62.5		62.5	62.5
-O$\underline{C}$OCH_3		170.4	170.4			
		170.3	170.2			
		169.9	169.9			
		169.1				
-OCO$\underline{C}H_3$		20.8	20.7			
		20.7	20.5x2			
		20.5x2				

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References and Notes

1) T. Nakagawa, K. Hayashi, and H. Mitsuhashi, Tetrahedron Lett., 1982, 5431.

2) a) Yu. A. Berlin, S. E. Esipov, M. N. Kolosov, M. M. Shemyakin, and M. G. Brazhnikova, Tetrahedron Lett., 1964, 1323 ; b) Idem, Ibid, 1964, 3513.

3) We are not able to account for this process from the point of view of the structure upto the present.

4) ^{1}H, ^{13}C NMR spectra were measured in Py-d_5 with TMS as internal standard.

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