Phytochemistry, Vol 24, No 8, pp 1857–1859, 1985.
Printed in Great Britain

0031–9422/85 $3.00+0.00

STRUCTURE OF RENIFOLIN AND RECONFIRMATION OF THE STRUCTURE OF PIROLATIN*

HIROYUKI INOUYE and KENICHIRO INOUE

Faculty of Pharmaceutical Sciences, Kyoto University, Sakyo-ku, Kyoto, 606 Japan

(*Received* 29 *November* 1984)

Key Word Index—*Pyrola renifolia*; *P. japonica*; Pyrolaceae; renifolin; pirolatin; structure; ^{13}C NMR; ^{1}H NMR; NOE.

Abstract—The ^{1}H and ^{13}C NMR spectral analysis of the glucoside renifolin, isolated from *Pyrola renifolia*, demonstrated the so far unknown binding site of the glucose moiety to be at C-8 and hence its structure as 8-β-D-glucosyloxy-2,7-dimethyl-1,4-dihydronaphthalen-5-ol. The earlier reported structure for the glucoside pirolatin, isolated from *Pyrola japonica*, was also reconfirmed by the ^{13}C NMR spectral analysis.

INTRODUCTION

Renifolin (**1**) in *Pyrola renifolia* Maxim. and pirolatin (**2**) in *Pyrola japonica* Klenze (*Pirola japonica* Sieb.) are glucosides characteristic of pyrolaceous plants and are supposed to be biosynthesized via toluquinol. From the biosynthetic point of view, the former seems to be most closely related to the 1,4-naphthoquinone chimaphilin (**3**), which is a widespread constituent in pyrolaceous plants [1]. Although renifolin (**1**) had already been characterized as a β-D-glucopyranoside of 2,7-dimethyl-1,4-dihydronaphthalen-5,8-diol (**4**) by various chemical reactions and spectral data [2], the binding site of glucose to the aglucone moiety remained unsolved. On the other hand, the structure of pirolatin has been shown to be 4-hydroxy-1-β-D-glucopyranosyloxy-5-methyl-2-[*trans*, *cis*-8′-hydroxy-3′,7′-dimethyloctadien-(2′,6′)-yl]-benzene (**2**) also through chemical and spectral analyses [3]. However, the *Z*-configuration of the terminal double bond of its side chain assigned on the basis of an empirical regularity induced from 60 MHz ^{1}H NMR findings [4, 5] seemed to be somewhat unusual and questionable.

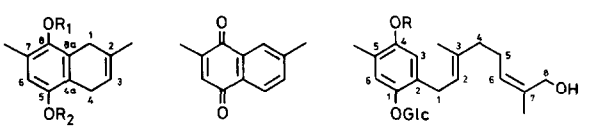
This paper deals with the elucidation of the binding site of sugar to the aglucone of renifolin (**1**) and the further evidence in support of the earlier proposed structure of pirolatin (**2**).

RESULTS AND DISCUSSION

The ^{13}C NMR signals of renifolin (**1**) were assigned on the basis of the chemical shifts, multiplicities and selective proton decoupling experiments (Table 1), providing further evidence for the structure of renifolin (**1**) as a glucoside of 2,7-dimethyl-1,4-dihydronaphthalen-5,8-diol (**4**). The signals of *O*-function bearing aromatic carbons 5 and 8 were assigned on grounds of their multiplicities in the following way. C-5, being long range coupled to C-4 methylene protons and the C-6 aromatic proton, is expected to appear as a double triplet, but in consideration of the small coupling constant of a carbon atom with the proton positioned beyond two-bonds, it may appear as a triplet. On the other hand, C-8 is presumed to appear as a multiplet due to coupling with the C-6 aromatic proton, C-7 aromatic methyl protons and C-1 methylene protons. Therefore, the triplet-like signal at δ_C 151.91 should be assigned to C-5 and the multiplet at δ_C 146.80 to C-8. The collapse of the latter signal to a broad doublet (J = 7 Hz) on irradiation of aromatic methyl protons [δ_H 2.27 (*s*)] (low power selective proton decoupling experiments) further supported the above assignment.

Furthermore, on the basis of the comparison of the ^{13}C NMR signals of **1** and its methyl ether **5** [6] (Table 1), the multiplet at δ147.53 and the triplet-like signal at δ154.75 of the latter compound were assigned to C-8 and C-5, respectively. The large downfield shift (2.84 ppm) of the latter signal as compared to the corresponding signal of **1** can be explained by the methylation of the C-5 hydroxyl group of **1**. The NOE experiments of methyl ether **5** showed further 19% NOE between the methoxy group and the C-6 proton, while 11% effect was observed between the latter and the aromatic methyl group at C-7. This also is in accord with the C-5 position of the methoxy group of **5**, and hence the hydroxy group of **1** is concluded to be at C-5. Consequently, the structure of renifolin (**1**) is 2,7-dimethyl-8-β-D-glucopyranosyloxy-1,4-dihydronaphthalen-5-ol.

The ^{13}C NMR signals of pirolatin (**2**) and its methyl



1 R_1 = Glc, R_2 = H
3
2 R = H
4 R_1 = R_2 = H
6 R = Me
5 R_1 = Glc, R_2 = Me

*Part 16 in the series "Constituents of Pyrolaceous Plants". For Part 15 see ref. [3].

Table 1. ^{13}C NMR data of renifolin (1) and its methyl ether (5)

	Renifolin (1)				Renifolin methyl ether (5)		
C	Chemical shift (δ)	Multiplicity*	^1J	Coupling constant (Hz) $>^1J$ (long range)	Chemical shift (δ)	Multiplicity	Coupling constant (Hz) ^1J (Hz)
1	31.21	*Tm*	125		31.21	*Tm*	
2	130.92	*br s*			131.09	*m*	
3	118.66	*Dm*	155		118.71	*Dm*	
4	26.15	*Td*	128	5	26.05	*Td*	
4a	120.87	*m*			122.55	*m*	
5	151.91	*t*-like			154.75	*t*-like	
6	114.98	*Dq*	155	5	110.92	*Dq*	
7	129.87	*q*		6	129.85	*d*	
8	146.80	*m*			147.53	*m*	
8a	132.74	*td*			132.62	*t*	
2-Me	23.55	*Qd*	126	6	23.50	*Qd*	
7-Me	17.42	*Qd*	127	5	17.68	*Qd*	
OMe					55.97	*Q*	144
1′	105.72	*D*	159		105.74	*D*	
2′	75.89	*Dd*	144	4	75.92	*Dd*	
3′	77.94	*D*	140		78.01	*D*	
4′	71.90	*D*	145		71.95	*D*	
5′	78.08	*D*	143		78.03	*D*	
6′	62.98	*T*	143		63.03	*T*	

*Capital letters refer to the pattern resulting from directly bonded protons and lower case letters to long-range ^{13}C–^1H coupling.

Table 2. ^{13}C NMR data of pirolatin (2) and its methyl ether (6)

	Pirolatin (2)					Pirolatin methyl ether (6)		
C	Chemical shift (δ)	Multiplicity	^1J	Coupling constant (Hz) $>^1J$		Chemical shift (δ)	Multiplicity	Coupling constant (Hz) ^1J
1	149.77	*dt*		9	4	150.30	*dt*	
2	131.07	*m*				130.84	*m*	
3	116.56	*Dt*	156	4		112.45	*Dt*	
4	151.62	*dq*		9	4	154.58	*dq*	
5	123.55	*qu*		5		125.69	*qu*	
6	120.28	*Dq*	157	5		120.24	*Dq*	
5-Me	16.08	*Qd*	126	5		16.13	*Qd*	
1′	28.74	*Tdd*	128	5	3	29.12	*Tdd*	
2′	128.61	*Dm*	150			128.46	*Dm*	
3′	136.39	*m*				136.54	*m*	
4′	41.05	*Tm*	126			41.06	*Tm*	
5′	27.27	*Tm*	128			27.32	*Tm*	
6′	124.70	*Dm*	153			124.74	*Dm*	
7′	135.72	*m*				135.83	*m*	
8′	61 52	*Tdq*	141	9	4	61.44	*Tdq*	
3′-Me	16.35	*Qdt*	125	8	4	16.35	*Qdt*	
7′-Me	21.49	*Qdt*	125	7	4	21.53	*Qdt*	
OMe	—					56.29	*Q*	
1″	104.28	*D*	160			104.11	*D*	143
2″	75 22	*Dd*	145	4		75.19	*Dd*	
3″	78.00	*D*	136			78.06	*D*	
4″	71.64	*D*	145			71.56	*D*	
5″	78.30	*D*	136			78.30	*D*	
6″	62.80	*T*	144			62.73	*T*	

ether (**6**) (Table 2) corroborated the structure **2** for pirolatin as proposed previously. Whereas the configuration of the terminal double bond of their side chain seemed still to be somewhat questionable, this was ascertained by the chemical shifts (δ_C 61.52 and 61.44) [7] of C-8′ signals of both compounds, to be the *Z*-configuration as proposed earlier.

EXPERIMENTAL

^{1}H NMR spectra were determined at 200 MHz and ^{13}C NMR spectra at 50.10 MHz in CD_3OD with TMS as internal standard. Renifolin, pirolatin and their methyl ethers obtained previously [2, 3] were used in this study.

Renifolin (**1**). ^{1}H NMR: δ1.78 (3H, *br s*, 2-Me), 2.27 (3H, *s*, 7-Me), 3.17 (4H, *m*, H_2-1 and H_2-4), 3.22–3.57 (4H, *m*, H-2′, H-3′, H-4′, H-5′), 3.65 (1H, *dd*, J = 2.4 and 11.7 Hz, H-6′), 3.78 (1H, *dd*, J = 4.9 and 11.7 Hz, H-6′), 4.60 (1H, *d*, J = 7.3 Hz, H-1′), 5.53 (1H, *m*, H-3), 6.43 (1H, *s*, H-6).

Renifolin methyl ether (**5**). ^{1}H NMR: δ1.78 (3H, *br s*, 2-Me), 2.34 (3H, *s*, 7-Me), 3.15 (4H, *m*, H_2-1 and H_2-4), 3.24–3.56 (4H, *m*, H-2′, H-3′, H-4′, H-5′), 3.65 (1H, *dd*, J = 5.1 and 11.5 Hz, H-6′), 3.75 (3H, *s*, 5-OMe), 3.78 (1H, *dd*, J = 2.7 and 11.5 Hz, H-6′), 4.62 (1H, *d*, J = 7.3 Hz, H-1′), 5.52 (1H, *m*, H-3), 6.55 (1H, *s*, H-6).

Pirolatin (**2**). ^{1}H NMR: δ1.70 (3H, *d*, J = 0.7 Hz, 3′-Me), 1.74 (3H, *d*, J = 1.0 Hz, 7′-Me), 2.13 (3H, *s*, 5-Me), 2.00–2.25 (4H, *m*, H_2-4′, H_2-5′), 3.28–3.49 (6H, *m*, H-2″, H-3″, H-4″, H-5″, H_2-1′), 3.70 (1H, *dd*, J = 4.6 and 12.0 Hz, H-6″), 3.88 (1H, *dd*, J = 1.5 and 12.0 Hz, H-6″), 4.05 (2H, *s*, H_2-8′), 4.72 (1H, *d*, J = 7.6 Hz, H-1″), 5.26 (1H, *br t*, J = 7.1 Hz, H-6′), 5.32 (1H, *qt*, J = 1.0 and 7 6 Hz, H-2′), 6.54 (1H, *s*, H-6), 6.91 (1H, *s*, H-3).

Pirolatin methyl ether (**6**). ^{1}H NMR: δ1.72 (6H, *br s*, 3′-Me, 7′-Me), 2.13 (3H, *s*, 5-Me), 2.01–2.25 (4H, *m*, H_2-4′ and H_2-5′), 3.28–3.47 (6H, *m*, H-2″, H-3″, H-4″, H-5″, H_2-1′), 3.69 (1H, *dd*, J = 3.4 and 12.0 Hz, H-6″), 3.74 (3H, *s*, 4-OMe), 3.88 (1H, *dd*, J = 1.7 and 12.0 Hz, H-6″), 4.04 (2H, *s*, H_2-8′), 4.75 (1H, *d*, J = 7.3 Hz, H-1″), 5.27 (1H, *qt*, J = 1.2 and 6.8 Hz, H-6′), 5.35 (1H, *qt*, J = 1.2 and 7.3 Hz, H-2′), 6.63 (1H, *s*, H-6), 6.97 (1H, *s*, H-3).

Acknowledgement—We thank Professor T. Shingu of Kobe Gakuin University for NOE experiments

REFERENCES

1. Thomson, R. H. (1971) *Naturally Occurring Quinones*, 2nd edn, p. 199. Academic Press, London.
2. Inouye, H. and Arai, T. (1964) *Chem. Pharm. Bull.* **12**, 533.
3. Inouye, H., Tokura, K. and Tobita, S. (1968) *Chem. Ber.* **101**, 4057.
4. Bates, R. B. and Gale, D. M. (1960) *J. Am. Chem. Soc.* **82**, 5749.
5. Bates, R. B., Carnighan, R. O., Rakutis, R. O. and Schauble, J. H. (1962) *Chem. Ind.* 1020.
6. Stothers, J. B. (1972) *Carbon-13 NMR Spectroscopy*, p. 196. Academic Press, New York.
7 Brouwer, H. and Stothers, J. B. (1972) *Can. J. Chem.* **50**, 1361.