

REVISION OF ^1H - AND ^{13}C -NMR ASSIGNMENTS OF LANOSTANOIDS
FROM *GANODERMA LUCIDUM* BY 2D-NMR STUDIES

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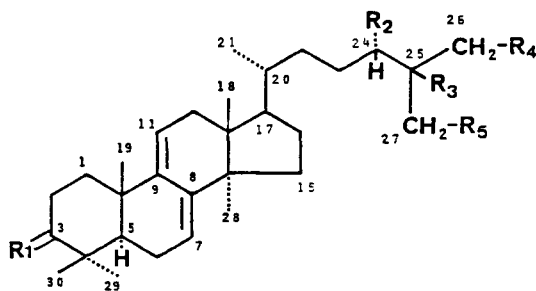
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ABSTRACT.— ^1H - and ^{13}C -nmr assignments of lanostanoids from *Ganoderma lucidum* were revised by 2D nmr, ^1H - ^1H and ^1H - ^{13}C shift correlation spectra, and distortionless enhancement by polarization transfer (DEPT) techniques for ^{13}C -nmr. The assignments of five lanostanoids, ganodermenonol [**1**], ganodermediol [**2**], ganodermatrioniol triacetate [**3a**], ganodermanondiol [**4**], and the 3β -ol of **4** [**6**], were revised by the direct data, and those of ganodermanoniol [**5**] were revised by analogy to the five lanostanoids. The ^1H -nmr assignments for acetate derivatives of **1**, **2**, **4**, and **5** were also revised with the above data.

Many terpenoids have been isolated from a Chinese crude drug "Ganoderma (Lin Zhi Cao)" (1-14). Recently, the Kikuchi group isolated several new lanostanoids, and their structures were elucidated by the experiments of 2D nmr, ^1H - ^1H and ^1H - ^{13}C shift correlation, and DEPT techniques for ^{13}C nmr (15-17). Previously, we also reported several lanostanoids—ganodermenonol [**1**], ganodermediol [**2**], ganodermatrioniol triacetate [**3a**], ganodermanondiol [**4**], and ganodermanoniol [**5**]—from the fresh fruiting body of the fungus, *Ganoderma lucidum* (Fr.) Karst. (Polyporaceae) (18, 19). Although most of the assignments in the ^1H - and ^{13}C -nmr spectra of the previous papers compare favorably with those derived in the earlier literatures (20-24), a number of revisions were found to be necessary by means of 2D nmr together with the DEPT techniques for ^{13}C nmr.



	R ₁	R ₂	R ₃	R ₄	R ₅
1	O	$\Delta^{24(25)}$		OH	H
	----H				
2	—OH	$\Delta^{24(25)}$		OH	H
	----H				
3	—OH	$\Delta^{24(25)}$		OH	OH
	----H				
3a	—OAc	$\Delta^{24(25)}$		OAc	OAc
4	O	OH	OH	H	H
4a	O	OAc	OH	H	H
5	O	OH	OH	OH	H
5a	O	OAc	OH	OAc	H
	----H				
6	—OH	OH	OH	H	H

TABLE 1. Revised ¹H-nmr Assignments for Lanostanoids from *Ganoderma lucidum* and Their Derivatives (in CDCl₃, δ ppm, J=Hz).

Proton No.	Compounds									
	1	2	2 acetate	3 acetate	4	4 acetate	5	5 acetate	6	
18-H ₁	0.59 (s)	0.57 (s)	0.56 (s)	0.56 (s)	0.58 (s)	0.58 (s)	0.58 (s)	0.58 (s)	0.57 (s)	
19-H ₁	1.20 (s)	0.98 (s)	1.01 (s)	1.01 (s)	1.18 (s)	1.20 (s)	1.20 (s)	1.20 (s)	0.98 (s)	
21-H ₁	0.92 (d)	0.92 (d)	0.91 (s)	0.92 (d)	0.91 (d)	0.91 (d)	0.92 (d)	0.91 (d)	0.93 (d)	
28-H ₁	0.88 (s)	0.88 (s)	0.87 (s)	0.87 (s)	0.86 (s)	0.86 (s)	0.88 (s)	0.86 (s)	0.88 (s)	
29-H ₁	1.09 (s)	1.00 (s)	0.89 (s)	0.89 (s)	1.07 (s)	1.09 (s)	1.09 (s)	1.09 (s)	1.00 (s)	
30-H ₁	1.13 (s)	0.88 (s)	0.96 (s)	0.96 (s)	1.11 (s)	1.13 (s)	1.11 (s)	1.13 (s)	0.88 (s)	
26-H	4.00 (s)	4.00 (s)	4.45 (s)	4.57 (s)	1.15 (s)	1.20 (s)	3.84 (d)	3.90 (d)	1.16 (s)	
27-H	1.67 (s)	1.67 (s)	1.66 (s)	4.66 (s)	(2H) (3H)	(3H) (3H)	(J=11.3)	(J=11.4)	(3H)	
2-H	2.78	(3H)	(3H)	(2H)	1.21 (s)	1.20 (s)	1.21 (s)	1.21 (s)	1.22 (s)	
3α-H	2.35	3.25 (d)	4.51 (dd)	4.50 (m)	2.77 (m)	2.77 (m)	2.80 (m)	2.76 (m)	(3H)	
7-H	5.51 (dd)	(J=5.4, 10.1)	(J=5.2, 10.4)	5.46 (m)	5.52 (dd)	5.51 (dd)	5.51 (dd)	5.51 (m)	3.25 (m)	
11-H	5.41 (m)	5.47 (m)	5.46 (m)	5.46 (m)	(J=2.0, 6.0)	(J=2.0, 6.0)	(J=2.0, 6.1)	5.47 (m)	5.47 (m)	
24-H	5.41 (m)	5.34 (m)	5.34 (m)	5.33 (m)	(J=2.0, 6.0)	(J=2.0, 6.0)	(J=1.5, 5.0)	5.39 (m)	5.32 (m)	
OAc		5.42 (m)	5.46 (m)	5.78 (dd)	3.30 (dd)	4.76 (dd)	3.48 (m)	4.90 (dd)	3.31	
		2.07 (s)	2.06 (3H)	2.06 (3H)	(J=7.3, 7.4)	(J=2.6, 10.0)		(J=2.6, 10.0)		
			2.07 (3H)	2.07 (6H)		2.12 (3H)		2.08 (3H)		
								2.09 (3H)		

We carried out the reexaminations of the ^1H - and ^{13}C -nmr spectra of the reported compounds **1**, **2**, **3a**, **4**, and **6** by means of 2D nmr together with the DEPT and insensitive nuclei enhanced by polarization transfer (INEPT) techniques. The revised assignments of ^1H - and ^{13}C -nmr were performed through measurements of ^1H - ^1H and ^1H - ^{13}C shift correlation, DEPT, and selective INEPT techniques. For example, the ^1H - ^1H shift correlated spectra of ganodermanondiol [**4**] allowed us to assign most of the ^1H signals (Table 1, Figure 1). It is clear that the original assignments for 7-H and 11-H should be inverted by the correlation with the signals for 5-, 6-, and 12-H. The signals due to the 28-Me and 18-Me were precisely assigned on the basis of the presence of re-

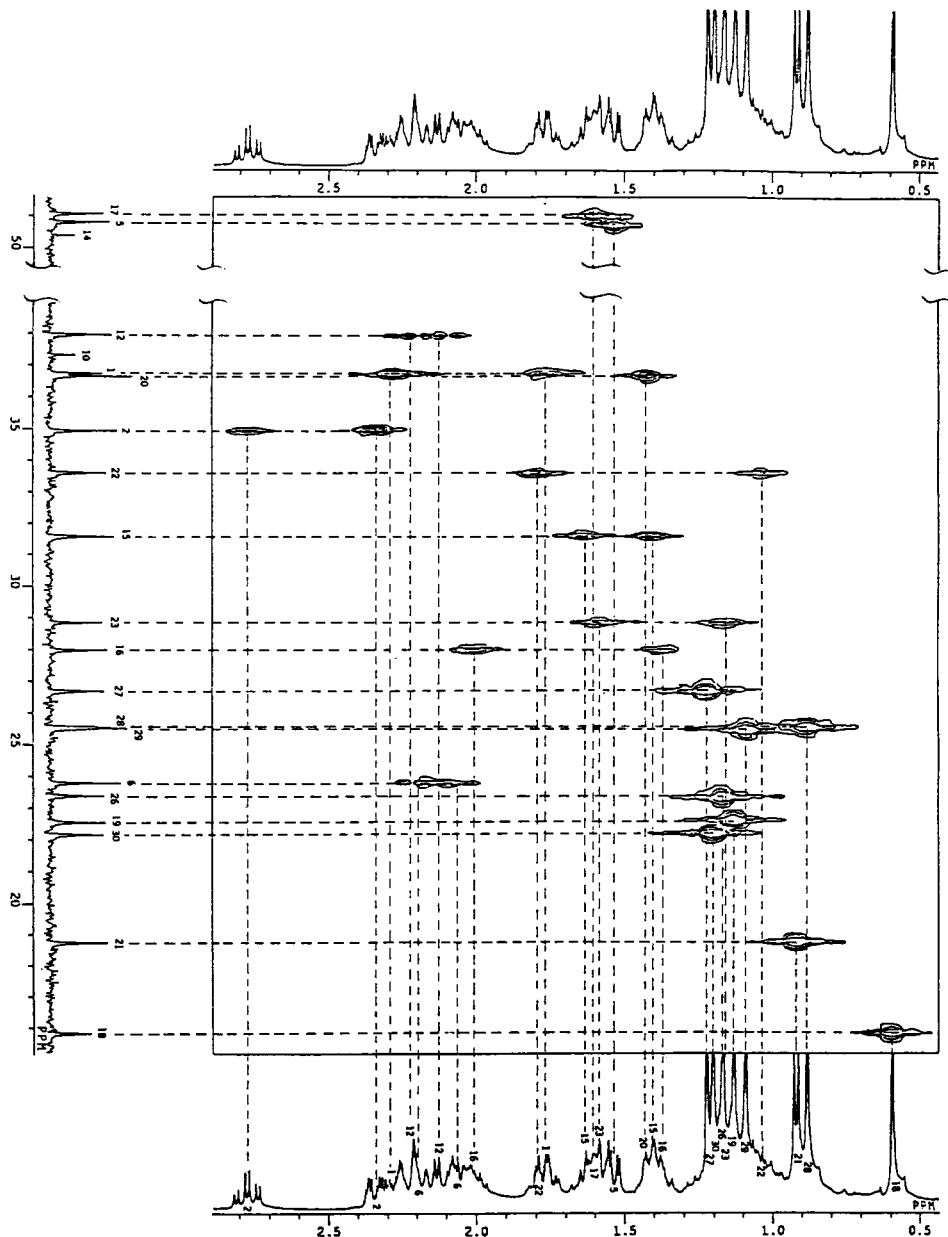


FIGURE 1. Contour map of the ^1H - ^{13}C shift-correlated spectrum of ganodermanondiol [**4**] in the upfield region.

TABLE 2. Revised ^{13}C -nmr Assignments for Lanostanoids from *Ganoderma lucidum* and Their Derivatives (in CDCl_3 , δ ppm).

Carbon No.	Compound									
	1	2	3a	4	4a	5	5a	6 ^a		
1	36.59(t)	35.76(t)	35.40(t)	35.63(t)	36.40(t)	36.55(t)	36.41(t)	36.35(t)		
2	34.82(t)	27.82(t)	24.33(t)	34.87(t)	34.83(t)	34.84(t)	34.82(t)	28.73(t)		
3	216.95(s)	78.98(d)	80.82(d)	216.92(s)	216.83(s)	217.00(s)	216.76(s)	78.06(d)		
4	47.43(s)	38.73(s)	37.22(s)	47.50(s)	47.46(s)	47.47(s)	47.42(s)	39.36(s)		
5	50.66(d)	49.15(d)	49.27(d)	50.70(d)	50.68(d)	50.29(d)	50.27(d)	49.75(d)		
6	23.62(t)	23.13(t)	22.90(t)	23.67(t)	23.62(t)	23.61(t)	23.62(t)	23.54(t)		
7	119.83(d)	120.25(d)	120.01(d)	119.91(d)	119.88(d)	119.87(d)	119.89(d)	121.00(d)		
8	142.81(s)	142.69(d)	142.63(s)	142.81(s)	142.77(s)	142.77(s)	142.79(s)	142.93(s)		
9	144.47(s)	145.93(s)	145.64(s)	144.46(s)	144.42(s)	144.43(s)	144.47(s)	146.52(s)		
10	37.17(s)	37.36(s)	37.82(s)	37.17(s)	37.17(s)	37.17(s)	37.18(s)	37.81(s)		
11	117.24(d)	116.26(d)	116.50(d)	117.23(d)	117.15(d)	117.22(d)	117.17(d)	116.54(d)		
12	37.76(t)	37.85(t)	37.95(t)	37.79(t)	37.74(t)	37.76(t)	37.74(t)	38.07(t)		
13	43.68(s)	43.84(s)	43.81(s)	43.75(s)	43.68(s)	43.67(s)	43.68(s)	44.09(s)		
14	50.27(s)	50.35(s)	50.32(s)	50.31(s)	50.28(s)	50.66(s)	50.68(s)	50.64(s)		
15	31.44(t)	31.53(t)	31.50(t)	31.45(t)	31.44(t)	31.45(t)	31.40(t)	31.88(t)		
16	27.86(t)	27.95(t)	27.91(t)	27.85(t)	27.76(t)	27.87(t)	27.75(t)	28.18(t)		
17	50.86(d)	50.94(d)	50.83(d)	50.94(d)	50.68(d)	50.96(d)	50.68(d)	51.47(d)		
18	15.65(q)	15.71(q)	15.69(q)	15.71(q)	15.65(q)	15.73(q)	15.65(q)	16.04(q)		
19	22.40(q)	22.79(q)	22.87(q)	22.06(q)	22.03(q)	22.04(q)	22.04(q)	25.98(q)		
20	36.01(d)	36.12(d)	36.07(d)	36.53(d)	36.59(d)	36.55(d)	36.58(d)	37.13(d)		
21	18.39(q)	18.44(q)	18.41(q)	18.62(q)	18.55(q)	18.64(q)	18.52(q)	19.03(q)		
22	35.87(t)	35.98(t)	35.78(t)	33.47(t)	32.61(t)	33.53(t)	32.56(t)	34.46(t)		
23	24.50(t)	24.97(t)	24.81(t)	28.71(t)	26.37(t)	28.83(t)	25.90(t)	29.32(t)		
24	126.86(d)	126.99(d)	137.83(d)	79.58(d)	80.75(d)	79.10(d)	76.34(d)	79.93(d)		
25	134.34(s)	134.33(s)	128.64(s)	73.26(s)	72.49(s)	74.07(s)	73.19(s)	72.86(s)		
26	69.01(t)	69.11(t)	66.83(t)	23.18(q)	24.95(q)	67.59(t)	68.40(t)	23.11(q)		
27	13.62(q)	13.65(q)	59.84(t)	26.57(q)	26.76(q)	20.88(q)	20.98(q)	26.11(q)		
28	25.38(q)	25.60(q)	25.56(q)	25.45(q)	25.36(q)	25.42(q)	25.30(q)	25.87(q)		
29	25.28(q)	28.17(q)	28.10(q)	25.32(q)	25.36(q)	25.42(q)	25.40(q)	28.83(q)		
30	22.43(q)	15.80(q)	16.97(q)	22.46(q)	22.43(q)	22.42(q)	22.44(q)	16.67(q)		

^aIn pyridine-*d*₅.

spective long-range coupling to 15 α - and 12 α -H and the selective INEPT techniques. The assignments of the 19-, 26-, 27-, 29-, and 30-Me signals were performed on the INEPT and the reported shift values (20–24). The revised ^1H -nmr assignments for lanostanoids **1–6** are shown in Table 1. The ^1H - ^{13}C shift correlation spectra of **4** lead readily to precise assignments of the ^{13}C signals associated with the DEPT and INEPT spectra. The assignments of quaternary carbon signals C-13 and C-14 were accorded the reported values reported by INEPT techniques (25,26). Revision in the assignments of ^{13}C signals was also necessary; thus, the signals for C-1 and -20 and C-10 and -12 should be exchanged, respectively, by the indications of DEPT and INEPT spectra, and the assignments for C-15, -16, -22, and -23 also should be revised. The revised ^{13}C -nmr assignments for lanostanoids **1–6** are also shown in Table 2.

EXPERIMENTAL

GENERAL EXPERIMENTAL PROCEDURES.— ^1H - and ^{13}C -nmr spectra were taken on a JEOL JNM-GX 400 spectrometer with TMS as an internal standard, and chemical shifts are recorded in δ values. ^1H - ^1H homonuclear shift correlated nmr spectra were measured by the use of a ^1H - ^1H shift correlation sequence with a 45 $^\circ$ mixing pulse and N-type peak selection. Data processing was carried out with the standard JEOL software. The sample concentration was 5–10 mg in 0.5 ml of the solvent, total acquisition time about 12 h, and processing time about 20 min. ^1H - ^{13}C heteronuclear shift correlated nmr spectra were obtained with the usual pulse sequence, and data processing was performed with the standard JEOL software. The sample concentration was 5–20 mg in 0.5 ml of the solvent. Acquisition of 16 transients for each of the 256 FIDs required about 13 h and processing took about 1 h.

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