Communications to the Editor

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GANODERIC ACID D, E, F, AND H AND LUCIDENIC ACID D, E, AND F, NEW TRITERPENOIDS FROM GANODERMA LUCIDUM[1)]

Tohru Kikuchi,* Satoko Matsuda, Shigetoshi Kadota, Yoshihiro Murai, and Zenichi Ogita

Reserach Institute for Wakan-Yaku (Oriental Medicines), Toyama Medical and Pharmaceutical University, 2630 Sugitani, Toyama 930-01, Japan

New highly oxidized lanostane-type triterpenoids, ganoderic acid D, E, F, and H and lucidenic acid D, E, and F, were isolated from the gills of *Ganoderma lucidum* and their structures were elucidated on the basis of spectral evidence.

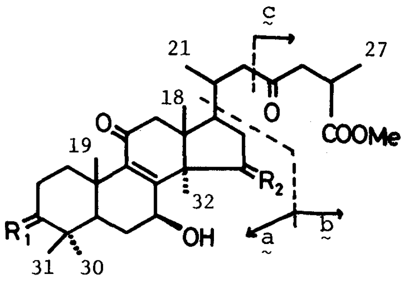
KEYWORDS —— *Ganoderma lucidum*; triterpenoid; ganoderic acid D; ganoderic acid E; ganoderic acid F; ganoderic acid H; lucidenic acid D; lucidenic acid E; lucidenic acid F; NMR

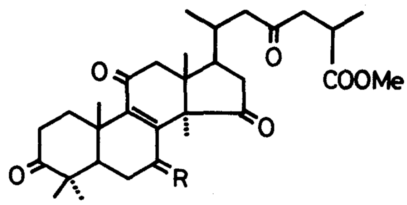
Many bitter triterpenoids have been isolated from the fungus *Ganoderma lucidum* (Polyporaceae) which is widely used as a home remedy (Reishi).[2,3)] Recently we isolated eleven new triterpenoids from the gills of this fungus and determined their structures.[1)] Also Yamazaki et al.[4)] and Furuya et al.[5)] independently obtained several new triterpenoids from the same fungus and some of the compounds obtained by the three groups were found to be identical. This situation prompted us to report our result in this communication.

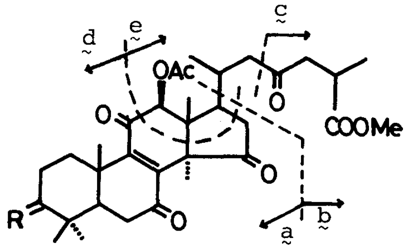
The acidic fraction from the ether extract of the gills[6)] of dried fruit bodies of *Ganoderma lucidum* was methylated with diazomethane and the crude product was separated repeatedly by a combination of silica gel chromatography and preparative layer chromatography (silica gel plates) to give the new triterpenoids, named methyl ganoderate D (1), E (2), F (3), G,[7)] H (4), and I and methyl lucidenate D (5),[7)] E (6), and F (7) and methyl ganolucidate A and B, together with known compounds, methyl ganoderate A (8), B (9),[2)] and C (10) and methyl lucidenate A (11).[3)] In this communication, the stuctures of 1 - 7 will be discussed.

Methyl ganoderate D (1), $C_{31}H_{48}O_7$, colorless prisms, mp 199-202°C, $[\alpha]_D$ +98° ($CHCl_3$), showed the molecular ion peak at m/z 532 together with fragment peaks at m/z 392 (a), 171 (b), 139 (b - CH_3OH), and 129 (c). It showed UV, IR, and ^{1}H-NMR spectra closely similar to those of 8,[2)] except that a new ^{1}H-NMR signal assignable to a carbinol methine proton appeared at δ 3.22 and the 30- and 31-methyl signals showed slight up-field shifts (Table I). Oxidation of 1 with CrO_3 - AcOH afforded a pentaketo ester (2), which was identified with the sample 2 prepared by oxidation of methyl ganoderate A (8). In view of these data, methyl ganoderate D was determined to be methyl 3β,7β,15α-trihydroxy-11,23-dioxo-5α-lanost-8-en-26-oate (1).

Methyl ganoderate E (2), $C_{31}H_{42}O_7$, was obtained as yellow needles, mp 206-208°C, $[\alpha]_D$ +167° ($CHCl_3$). This was found to be identical with the oxidation product (2) of methyl ganoderate A.







1 : $R_1 = \lessdot^{OH}_{H}$, $R_2 = \lessdot^{H}_{OH}$

8 : R_1 = O , $R_2 = \lessdot^{H}_{OH}$

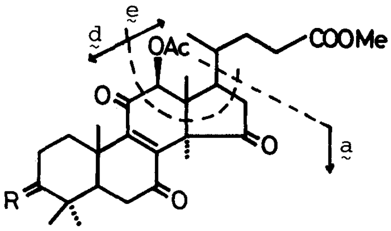
9 : $R_1 = \lessdot^{OH}_{H}$, R_2 = O

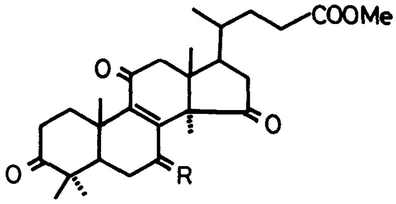
2 : R = O

10 : $R = \lessdot^{OH}_{H}$

3 : R = O

4 : $R = \lessdot^{OH}_{H}$





5 : R = O

6 : $R = \lessdot^{OH}_{H}$

7 : R = O

11 : $R = \lessdot^{OH}_{H}$

Table I. ^{1}H-NMR (200 MHz) Spectral Data of Methyl Ganoderate A (8), D (1), E (2), F (3), and H (4) and Methyl Lucidenate D (5), E (6), and F (7) (δ in $CDCl_3$)

^{1}H (J) \ Compound	8[a)]	1	2	3	4	5	6	7
18-H_3 s	0.98	0.97	0.89	0.85	0.88	0.87	0.89	0.85
19-H_3 s	1.25	1.24	1.29	1.33	1.33	1.33	1.33	1.27
21-H_3 d (6)	0.88	0.87	0.99	0.99	0.98	1.00	1.00	0.96
27-H_3 d (7)	1.18	1.18	1.18	1.18	1.18	—	—	—
30-H_3 s	1.09	1.02	1.12	1.12	1.03	1.12	1.03	1.11
31-H_3 s	1.11	0.84	1.15	1.14	0.82	1.14	0.83	1.14
32-H_3 s	1.27	1.25	1.66	1.80	1.73	1.81	1.74	1.65
COOMe	3.68	3.68	3.70	3.68	3.68	3.68	3.68	3.68
OAc	—	—	—	2.26	2.26	2.24	2.23	—
3-H dd (10, 6)	—	3.22	—	—	3.27	—	3.27	—
7-H br t (8.5)	4.80	4.75	—	—	—	—	—	—
15-H dd (10, 7.5)	4.63	4.57	—	—	—	—	—	—
12-H s	—	—	—	5.68	5.63	5.69	5.64	—

a) See reference 2.

Methyl ganoderate F (3), $C_{33}H_{44}O_9$, amorphous, $[\alpha]_D$ +111° ($CHCl_3$), UV λ: 252 nm (log ε 3.91), IR ν: 1745, 1735, 1710, and 1700 cm^{-1}, showed the M^+ peak at m/z 584 and significant fragment peaks at m/z 542, 524 (M^+-60), 413 (a), 302 (d+1), 283 (e),

Table II. ^{13}C-NMR Spectral Data of Methyl Ganoderate E (2), F (3), and H (4) and Methyl Lucidenate D (5), E (6), and F (7) (ppm in $CDCl_3$)

^{13}C \ Compd.	2	3	4	5	6	7
1	37.3 t	37.5 t	36.6 t	37.4 t	36.7 t	37.3 t
2	34.7 t	34.1 t	27.3 t	34.0 t	27.4 t	34.6 t
3	217.2 s	214.8 s	77.3 d	214.8 s	77.5 d	215.3 s
4	43.9 s	46.9 s	40.4 s	46.9 s	40.5 s	43.9 s
5	50.9 d	51.0 d	51.4 d	51.0 d	51.4 d	51.0 d
6	33.8 t	33.7 t	33.2 t	33.8 t	33.3 t	33.9 t
7	199.3 s	198.7 s	198.7 s	198.5 s	198.8 s	199.5[a] s
8	149.7 s	149.9 s	151.6 s	149.7 s	151.6 s	149.7 s
9	146.8 s	146.1 s	145.7 s	146.2 s	146.0 s	146.9 s
10	39.4 s	39.3 s	39.1 s	39.3 s	39.2 s	39.4 s
11	199.3 s	194.1 s	193.9 s	194.1 s	194.1 s	199.4[a] s
12	48.9[a] t	79.0 d	79.1 d	79.1 d	79.4 d	49.0 t
13	47.0 s	47.7 s	47.9 s	47.6 s	48.0 s	47.0 s
14	57.2 s	58.6 s	58.4 s	58.6 s	58.5 s	57.2 s
15	206.8 s	205.4 s	205.5 s	205.8 s	206.0 s	207.3 s
16	39.8 t	37.8 t	37.8 t	37.4 t	37.6 t	39.9 t
17	44.5 d	44.5 d	44.7 d	45.2 d	45.5 d	45.2 d
18	16.1 q	12.1 q	12.1 q	12.0 q	12.1 q	16.1 q
19	18.6 q	18.7 q	17.9 q	18.6 q	18.0 q	18.6 q
20	32.0 d	29.4 d	29.3 d	33.0 d	33.0 d	35.4 d
21	19.8 q	21.6 q	21.6 q	20.1 q	20.2 q	18.3 q
22	49.1[a] t	48.5 t	48.4 t	30.1 t	30.2 t	30.8 t
23	207.6 s	207.4 s	207.4 s	31.6 t	31.8 t	31.0 t
24	46.7 t	46.7 t	46.6 t	173.6 s	173.7 s	173.8 s
25	34.6 d	34.7 d	34.6 d	—	—	—
26	176.1 s	176.0 s	176.0 s	—	—	—
27	17.1 q	17.1 q	17.0 q	—	—	—
30	27.6 q	27.6 q	27.9 q	27.6 q	27.9 q	27.7 q
31	20.9[b] q	20.8[a] q	15.5 q	20.8[a] q	15.6 q	20.9[b] q
32	20.3[b] q	20.4[a] q	21.2[a] q	20.4[a] q	21.4[a] q	20.3[b] q
OCH_3	51.9 q	51.9 q	51.8 q	51.4 q	51.6 q	51.7 q
$CO\underline{C}H_3$		20.9[a] q	20.8[a] q	20.8[a] q	20.9[a] q	
$\underline{C}OCH_3$		170.2 s	170.1 s	170.0 s	170.1 s	

a), b) Assignments may be interchanged in each compound.

241, 223 (e - 60), 209, 191 (e - 60 - 32), 139 (b - 32), and 129 (c). The ^{1}H-NMR spectrum showed signals due to an acetoxyl methyl (δ 2.26) and a methine (δ 5.68, s) along with two sec- and five tert-methyl groups (Table I). Inspection of the ^{13}C-NMR spectrum compared with those of 1[4] and 2 (Table II) revealed a marked downfield shift of the C-12 signal, suggesting the presence of an acetoxyl group at the 12-position. This was supported by the upfield shift of the C-18 signal, which may be ascribed to the γ-effect of the acetoxyl group. Furthermore, irradiation of

the 32-methyl group at δ 1.80 induced a 15% NOE increase of the methine signal at δ5.68, indicating the β-orientation of the acetoxyl group. Thus, methyl ganoderate F should be methyl 12β-acetoxy-3,7,11,15,23-pentaoxo-5α-lanost-8en-26-oate (3).

Methyl ganoderate H (4), $C_{33}H_{46}O_9$, yellow needles, mp 155-156°C, $[\alpha]_D$ +55° ($CHCl_3$), UV λ: 256 nm (log ε 3.87), showed the M^+ peak at m/z 586 and fragment ion peaks at m/z 544, 526 (M^+-60), 415 (a), 304 (d+1), 283 (e), 241 (e - 42), 223 (e - 60), 209, 191 (e - 60 - CH_3OH), 139 (b - CH_3OH), and 129 (c) in the mass spectrum. The IR and ^{1}H-NMR spectra of 4 resembled those of 3, except for an additional IR band at 3400 cm^{-1} and the appearance of an NMR signal due to a carbinol methine proton (δ 3.27) and the upfield shift of two tert-Methyl signals (δ 0.82 and 1.03) (Table I), suggesting the 3β-alcohol structure in 4 instead of the 3-keto structure in 3. This was supported by comparison of the ^{13}C-NMR spectra of 4 and 3, which showed a marked upfield shift of the C-3 signal (Table II). Furthermore, oxidation of 4 with CrO_3-AcOH gave 3. Therefore, methyl ganoderate H was determined to be methyl 3β-hydroxy-12β-acetoxy-7,11,15,23-tetraoxo-5α-lanost-8-en-26-oate (4).

Methyl lucidenate D (5), E (6), and F (7) showed the following physical properties: 5, $C_{30}H_{40}O_8$, amorphous, $[\alpha]_D$ +136° ($CHCl_3$); 6, $C_{30}H_{42}O_8$, yellow needles, mp 140-144°C, $[\alpha]_D$ +86° ($CHCl_3$); 7, $C_{28}H_{38}O_6$, yellow needles mp 208-211°C, $[\alpha]_D$ +195° ($CHCl_3$). The UV, IR, and ^{1}H-NMR spectra of 5, 6, and 7 resembled those of 3, 4, and 2, respectively, but the ^{1}H-NMR signal at δ 1.18 due to the 27-methyl group was not observed (Table I). In their ^{13}C-NMR spectra, chemical shifts of the carbons associated with the A - D rings were essentially identical with those of the corresponding carbons of 3, 4, and 2, respectively, but the signals arising from the side chains corresponded to only six carbons, reflecting the γ-substituted methyl pentanoate structure (Table II). Furthermore, the mass spectrum of 5 exhibited the M^+ peak at m/z 528 and fragment ion peaks at m/z 486, 468 (M^+-60), 413 (a), 302 (d+1), 227 (e), 185 (e - 42), 167 (e - 60), 153, and 135 (e - 60 - CH_3OH), while that of 6 showed peaks at m/z 530 (M^+), 488, 470 (M^+-60), 415 (a), and 304 (d+1) along with peaks at m/z 227, 185, 167, 153, and 135. On the basis of these data, the structures of methyl lucidenate D, E, and F were assigned to the formula 5, 6, and 7, respectively.

REFERENCES AND NOTES

1) This work was presented at the 105th Annual Meeting of the Pharmaceutical Society of Japan, Kanazawa, April 1985, Abstr., P. 496.

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5) M. Hirotani and T. Furuya, The 105th Annual Meeting of the Pharmaceutical Society of Japan, Kanazawa 1985, Abstr., p. 496.

6) Extraction of the powdered gills (12.8 g) scraped from dried fruit bodies (2.56 kg) with ether at room temperature gave the ether extract (7 g), which showed an antiandrogenic activity in mice.

7) The trivial names, ganoderic acid D, E, F, and G and lucidenic acid D were suggested by Dr. K. Yamazaki, Hiroshima Univ., to whom we express our gratitude.

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