Short Communication

New Terpenoids, Ganolucidic Acid D, Ganoderic Acid L, Lucidone C and Lucidenic Acid G, from the Fungus *Ganoderma lucidum*

Tsuyoshi NISHITOBA, Hiroji SATO and Sadao SAKAMURA

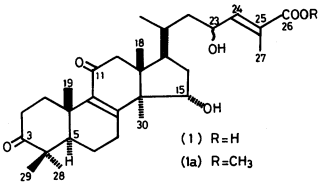
Department of Agricultural Chemistry, Faculty of Agriculture, Hokkaido University, Sapporo 060, Japan

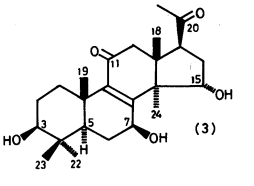
Received November 18, 1985

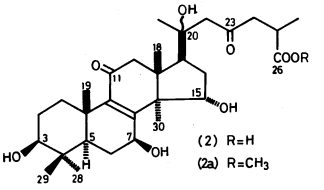
The fruiting body of the fungus *Ganoderma lucidum* (Reishi) has attracted much attention as a folk medicine, and some of its components have been elucidated. We have also reported several bitter terpenoids and related compounds from the fungus.[1～9] The naming of lucidenic acids is a little confused, so we now designate our lucidenic acids D and E[2] as D_1 and E_1, and Kikuchi's[7] as D_2 and E_2, respectively. Recently, we have isolated four new terpenoids, ganolucidic acid D (**1**), methyl ganoderate L (**2a**), lucidone C (**3**) and methyl lucidenate G (**4a**). Ganolucidic acid D (**1**) has a allylic alcohol group in the side chain and can be a possible biogenetic intermediate between the mycelial components[10] and terpenoids of the fruiting body. On the other hand, ganoderic acid L (**2**), which has a hydroxyl group at C-20, can be a possible precursor of lucidone C (**3**). Among the lucidenic acids, lucidenic acid G (**4**) is unique in having a hydroxyl group at C-26.

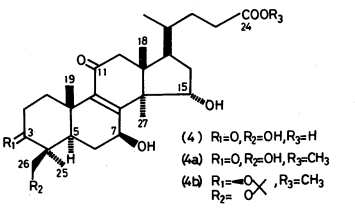
The isolation procedure was the same as that described in our previous papers,[1,3] and the acidic part obtained was separated into thirteen fractions (Fr. 1～13). Fr. 12 was subjected to Lobar column (RP-8, Merck) chromatography and the second fraction was treated with diazomethane. The resulting product was rechromatographed on silica gel and the Lobar (RP-8) column to give methyl ganoderate L (**2a**), lucidone C (**3**) and methyl lucidenate G (**4a**). The fourth fraction in the chromatography of Fr. 12 was purified on a silica gel column, PTLC and HPLC to give ganolucidic acid D (**1**).

Ganolucidic acid D (**1**), crystalline solids, $C_{30}H_{44}O_6$ (M^+ 500.3135). $[\alpha]_D^{23}$ +192° (c= 0.1,EtOH). UV λ_{max}^{EtOH} nm (ε): 257 (7800). IR ν_{max}^{KBr} cm^{-1}: 3400, 1700, 1640. **1** was treated with diazomethane to yield a monomethyl ester **1a**, $C_{31}H_{46}O_6$ (M^+ 514.3304). The ^{1}H-NMR data of **1a** were $\delta_{TMS}^{C_5D_5N}$: 7.17 (1H, dq, J=8.8 and 1.5 Hz), 4.95 (1H, overlapped), 4.62 (1H, ddd, J=8.8, 5.9 and 5.4 Hz), 2.01 (3H, d, J=1.5 Hz), 1.47 (3H, s), 1.26 (3H, s), 1.13 (3H, s), 1.12 (3H, s), 1.09 (3H, d, J= 6.8 Hz), 0.95 (3H, s). The ^{13}C-NMR data of **1a** were $\delta_{TMS}^{C_5D_5N}$: (number of bonded H): 217.1 (0),









198.3 (0), 168.6 (0), 165.5 (0), 146.2 (1), 138.2 (0), 127.3 (0), 72.0 (1), 66.7 (1). These data indicate that **1a** was different from methyl ganolucidate A[8] in the side chain moiety, having a double bond between C-24 and C-25, and a hydroxyl group at C-23. In the ^{1}H-NMR spectrum of **1a**, the signal due to H-24 resonated at δ6.59 in $CDCl_3$, which resembles those of tiglic acid[11] and ganoderic acids U$\sim$Z,[10] so the configuration of the double bond between C-24 and C-25 was assigned as *E*. From these observations, the structure of ganolucidic acid D was concluded to be 15α,23-dihydroxy-3,11-dioxo-5α-lanosta-8,24*E*-dien-26-oic acid (**1**).

Methyl ganoderate L (**2a**), colorless prisms, mp 228$\sim$230°C. $[\alpha]_D^{23}$ +66° (c=0.2, MeOH). UV λ_{max}^{MeOH} nm (ε): 256 (6750). IR ν_{max}^{KBr} cm^{-1}: 3430, 1720, 1655. ^{1}H-NMR $\delta_{TMS}^{C_5D_5N}$: 5.46 (1H, dd, J=9.2 and 7.3 Hz), 5.0 (1H, overlapped), 3.52 (1H, dd, J=10.6 and 5.1 Hz), 1.64 (3H, s), 1.59 (3H, s), 1.57 (3H, s), 1.54 (3H, s), 1.29 (3H, s), 1.15 (3H, d, J=6.6 Hz), 1.11 (3H, s). The ^{13}C-NMR data showed the presence of thirty-one carbon atoms and the principal signals were $\delta_{TMS}^{C_5D_5N}$: 209.4 (0), 200.2 (0), 176.2 (0), 160.3 (0), 141.7 (0), 77.6 (1), 73.8 (0), 72.4 (1), 69.5 (1). By comparing these data to those of methyl ganoderate D_2[6,7] and I,[8] the structure depicted as **2a** was deduced for methyl ganoderate L. The FD-MS data of **2a** were m/z (%): 548 (M^+, 5.3), 404 (100), 144 (33.4). The very weak intensity of the molecular ion peak is attributable to the McLafferty rearrangement and subsequent easy cleavage between C-20 and C-22. The EI-MS of **2a** did not give the molecular ion peak, but its fragmentation pattern was in good agreement with that of lucidone C (**3**). Thus, the structure of methyl ganoderate L was established to be methyl 3β,7β,15α,20-tetrahydroxy-11,23-dioxo-5α-lanost-8-en-26-oate (**2a**).

Lucidone C (**3**), colorless syrup, $C_{24}H_{36}O_5$ (M^+ 404.2527). $[\alpha]_D^{24}$ +145° (c=0.2, MeOH). UV λ_{max}^{MeOH} nm (ε): 255 (7680). IR ν_{max}^{KBr} cm^{-1}: 3500 (sh), 3430, 1700, 1660. ^{1}H-NMR $\delta_{TMS}^{C_5D_5N}$: 5.34 (1H, dd, J=9.5 and 7.3 Hz), 4.98 (1H, dd, J=9.9 and 7.3 Hz), 3.51 (1H, dd, J=10.8 and 5.3 Hz), 2.11 (3H, s), 1.59 (3H, s), 1.51 (3H, s), 1.29 (3H, s), 1.11 (3H, s), 1.07 (3H, s). ^{13}C-NMR $\delta_{TMS}^{C_5D_5N}$: 207.6 (0), 198.9 (0), 159.9 (0), 141.9 (0), 77.5 (1), 72.4 (1), 69.4 (1). These data are very similar to those of lucidone A,[2] but the presence of a 15α-hydroxyl group is indicated. So the structure of lucidone C was assigned as 3β,7β,15α-trihydroxy-4,4,14α-trimethyl-11,20-dioxo-5α-pregn-8-en (**3**).

Methyl lucidenate G (**4**), colorless syrup, $C_{28}H_{42}O_7$ (M^+ 490.2905). $[\alpha]_D^{21}$ +127° (c=0.2, MeOH). UV λ_{max}^{MeOH} nm (ε): 254 (8040). IR ν_{max}^{KBr} cm^{-1}: 3400, 1730 (sh), 1700, 1660. ^{1}H-NMR $\delta_{TMS}^{C_5D_5N}$: 5.24 (1H, dd, J=9.3 and 7.1 Hz), 4.99 (1H, dd, J=9.9 and 7.3 Hz), 4.37 (1H, d, J=11.0 Hz), 3.99 (1H, d, J=11.0 Hz), 1.72 (3H, s), 1.52 (3H, s), 1.51 (3H, s), 1.07 (3H, s), 0.81 (3H, d, J=5.9 Hz). ^{13}C-NMR $\delta_{TMS}^{C_5D_5N}$: 214.4 (0), 199.8 (0), 174.1 (0), 161.3 (0), 140.6 (0), 72.2 (1), 69.2 (1), 65.1 (2). These data indicate that methyl lucidenate G had the structure depicted as **4a**. The presence of a hydroxyl group at C-26 was confirmed by converting **4a** into **4b**, $C_{31}H_{48}O_7$ (M^+ 532.3392), by treating with $NaBH_4$ and following by $CuSO_4$/acetone. Thus, the structure of methyl lucidenate G was determined to be methyl 7β,15α-dihydroxy-4β-hydroxymethyl-4α,14α-dimethyl-3,11-dioxo-5α-chol-8-en-24-oate (**4a**).

Acknowledgment. We are thankful to Mr. Atsuhiro Honda of Nihon Joyaku Co., Ltd. for supplying the material.

REFERENCES

1) T. Nishitoba, H. Sato, T. Kasai, H. Kawagishi and S. Sakamura, *Agric. Biol. Chem.*, **48**, 2905 (1984).
2) T. Nishitoba, H. Sato and S. Sakamura, *Agric. Biol. Chem.*, **49**, 1547 (1985).
3) T. Nishitoba, H. Sato, T. Kasai, H. Kawagishi and S. Sakamura, *Agric. Biol. Chem.*, **49**, 1793 (1985).
4) T. Nishitoba, H. Sato and S. Sakamura, *Agric. Biol. Chem.*, **49**, 3637 (1985).
5) T. Kubota, Y. Asaka, I. Miura and H. Mori, *Helv. Chim. Acta*, **65**, 611 (1982).
6) H. Kohda, W. Tokumoto, K. Sakamoto, M. Fujii, Y. Hirai, K. Yamasaki, Y. Komoda, H. Nakamura, S. Ishihara and M. Uchida, *Chem. Pharm. Bull.*, **33**, 1367 (1985).

7) T. Kikuchi, S. Matsuda, S. Kadota, Y. Murai and Z. Ogita, *Chem. Pharm. Bull.*, **33**, 2624 (1985).
8) T. Kikuchi, S. Matsuda, Y. Murai and Z. Ogita, *Chem. Pharm. Bull.*, **33**, 2628 (1985).
9) M. Hirotani, T. Furuya and M. Shiro, *Phytochemistry*, **24**, 2055 (1985).
10) J. O. Toth, B. Luu, J. P. Beck and G. Ourisson, *J. Chem. Research* (*M*), 2722 (1983).
11) R. R. Fraser, *Can. J. Chem.*, **38**, 549 (1960).