Chem. Pharm. Bull. 30(1) 341—343 (1982)

The Structure of Rubescensin C: A New Minor Diterpenoid isolated from *Rabdosia rubescens*

SUN HAN-DONG,[a] CHAO JIN-HUA,[b] LIN ZHONG-WEN,[a] TERUYOSHI MARUNAKA,*,[c] YOSHINORI MINAMI,[c] and TETSURO FUJITA[d]

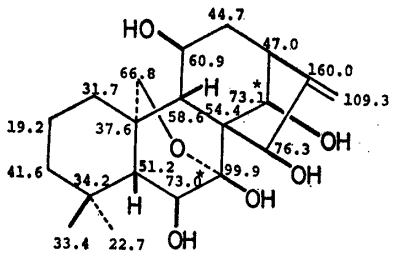
Kunming Institute of Botany, Academia Sinica,[a] Kunming, Yunnan, China, Department of Chemistry, Honan Medical College,[b] Zhengzhon, China, Research Laboratory, Taiho Pharmaceutical Co., Ltd.,[c] Kawauchi-cho, Tokushima, 771-01, Japan, and Faculty of Pharmaceutical Sciences, The University of Tokushima,[d] Tokushima, 770, Japan

(Received June 17, 1981)

A new diterpenoid, rubescensin C, was isolated from the leaves of *Rabdosia rubescens* (HEMSL.) HARA (Labiatae) and its structure was established from spectral and chemical evidence.

Keywords——diterpenoid; *ent*-kaurene; *Rabdosia rubescens*; rubescensin C; structure; ^{1}H-NMR; ^{13}C-NMR

Rabdosia rubescens, which is widely distributed in the Huanghe river valley and the southern district of China, has been used for the treatment of esophageal and cardial carcinoma in Honan. We have reported the isolation of oridonin[1] (=rubescensin A[2]) and ponicidin[3] (=rubescensin B[4]) from the ethereal extract of the dry leaves of this plant. The present paper describes the structural elucidation of a new diterpenoid, rubescensin C, which is a minor component isolated from the same plant.



1

Fig. 1. Rubescensin C (1): ^{13}C-NMR Data (δ/ppm) in C_5D_5N Solution

*: may be reversed.

Rubescensin C [1; $C_{20}H_{30}O_6$; mp 239—241°C; $[\alpha]_D^{25}$ −68.6° (c=0.14, C_5H_5N); IR ν_{max} 1640 cm^{-1} (>C=CH$_2$)] showed the presence of two methyl groups, five methylene groups, seven methine groups, four quaternary carbons, and two olefinic carbons in the ^{13}C nuclear magnetic resonance (NMR) spectrum (Fig. 1).[5] The signals due to an acetal carbon at δ 99.9 (C-7, s), an oxygenated methyl group at δ 66.8 (C-20, t), and an exomethylene group at δ 109.3 (C-17, t) and δ 160.0 (C-16, s) were also observed. On the other hand, the signals due to an oxygenated methyl group at δ 4.14 (20-H$_a$, 1H, dd, J=10 and 2 Hz) and δ 3.87 (20-H$_b$, 1H, d, J=10 Hz), and those of an exomethylene at δ 5.21 and 5.16 (17-2H, each d, J=2Hz) were observed in the ^{1}H-NMR spectrum (Fig. 2). Furthermore, long-lange coupling with δ 4.14 (20-H^a) and δ 2.37 (9β-H, dd, J=9 and 2 Hz) was confirmed by a decoupling experiment. On irradiation at δ 2.37 (9β-H), the doublet-doublet at δ 4.14 collapsed to a doublet (J=10 Hz). On treatment with 15% methanolic hydrochloric acid, rubescensin C was transformed into a saturated ketone [2; $C_{20}H_{30}O_6$; mp 241—243°C; CD (methanol) $\Delta\varepsilon^{25}$: −1.12 (299) (negative maximum)[6]] *via* the garryfoline-cuachichicine rearrangement,[7] indicating the presence of a 15β hydroxyl group in an *ent*-16-kaurene skeleton. The presence of this hydroxyl group was supported by the peaks at δ 5.34 (br s) in the ^{1}H-NMR spectrum and at δ 76.3 (d) in the ^{13}C-NMR spectrum.

Thus, these spectroscopic and chemical findings suggested that rubescensin C has an *ent*-7α, 15α-dihydroxy-7β, 20-epoxykaur-16-ene as the basic structure.

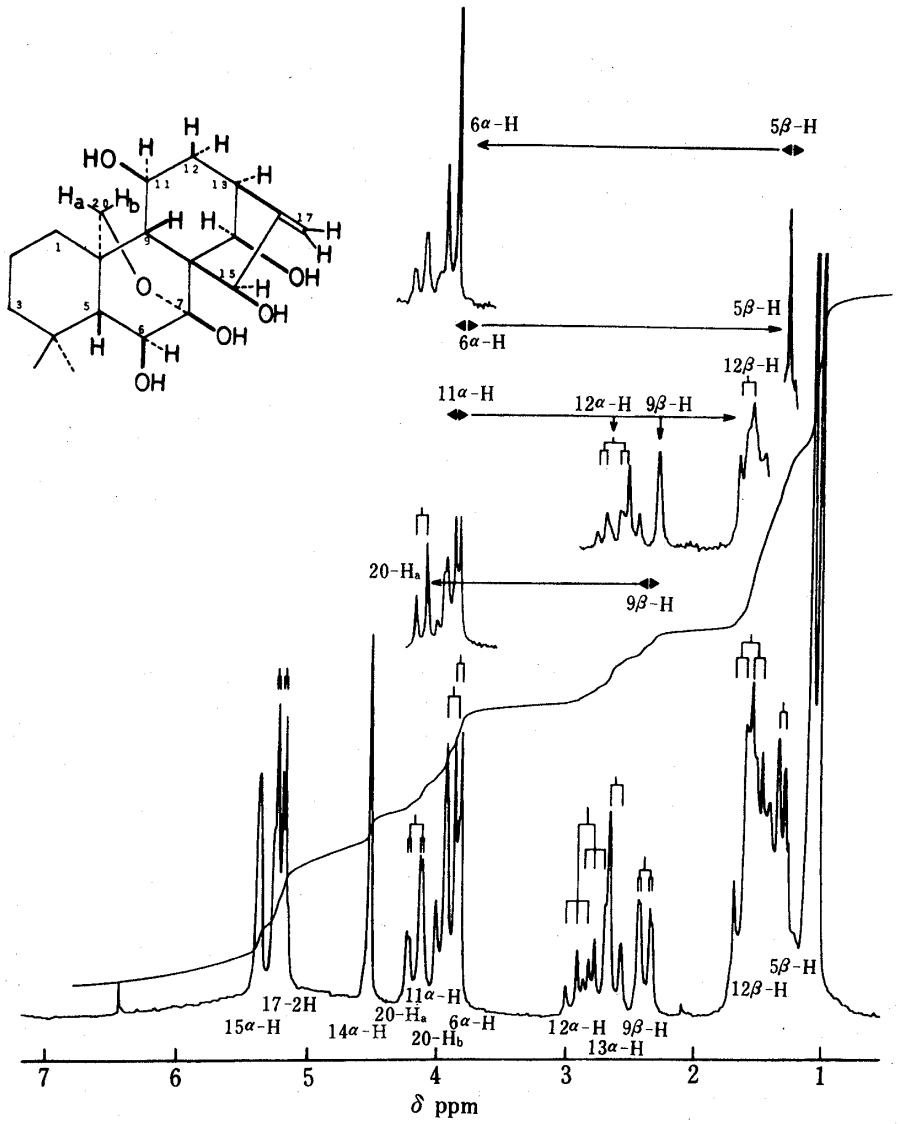
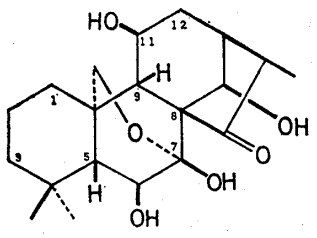


Fig. 2. The ^{1}H-NMR Spectrum of Rubescensin C in $CDCl_3+C_5D_5N$ Solution

Rubescensin C was found to have another three hydroxyl groups: IR ν_{max} 3300 cm^{-1}; ^{1}H-NMR δ 3.82 (1H, d, J=5 Hz), 3.91 (1H, m), and 4.50 (1H, s); ^{13}C-NMR δ 73.1, 73.0, and 60.9 (each d). The location of one hydroxyl group was elucidated as 14β from the chemical shift (δ 4.50) and coupling pattern (sharp s)[1] of the 14α proton signal.



2

The locations of the other two hydroxyl groups at 6β and 11β were supported by the following decoupling experiments. On irradiation at δ 1.32 (5β-H, d, J=5 Hz), the doublet at δ 3.82 (6α-H) changed into a singlet. On the other hand, on irradiation at δ 3.82, the doublet at δ 1.32 changed into a singlet. Further, on irradiation at δ 3.91 (11α-H), the doublet-doublet at δ 2.37 (9β-H, J=9 and 2 Hz) collapsed to a broad singlet and the sextet at δ 2.83 (12α-H, J=9, 9, and 14 Hz) changed into a doublet-doublet (J=9 and 14 Hz); further, the doublet-doublet at δ 1.57 (12β-H, J=9 and 14 Hz) changed into a doublet (J=14 Hz). These results indicate that the signal of 11α-H couples with 9β-H, 12α-H, and 12β-H, having

a J value of 9 Hz. In addition, the signal pattern at δ 2.71 (13α-H) indicates that 13α-H only couples with 12α-H.

Consequently, the structure of rubescensin C can be represented as **1**.

Experimental

Melting points were determined on a Yanagimoto micro-melting point apparatus and are uncorrected. Infrared (IR) absorption spectra were taken on a Hitachi 285 spectrometer, circular dichroism (CD) spectra on a Jasco J-500 instrument, and specific rotations on a Jasco DIP-180 automatic polarimeter. ^{1}H- and ^{13}C-NMR spectra were measured with a JEOL JNM-FX-100 machine and chemical shifts are given in δ (ppm) values with TMS as the internal standard. Mass spectra were recorded on a JEOL JMS-D-300 spectrometer.

Rubescensin C (**1**)——Extraction procedures have been reported[8] previously. Recrystallization from MeOH afforded plates. mp 239—241°C. $[\alpha]_D^{25}$ −68.6° (c=0.14, pyridine). *Anal.* Calcd for $C_{20}H_{30}O_6$: C, 65.55; H, 8.25. Found: C, 65.29; H, 8.23. MS m/e: 366 (M^+). IR ν_{max}^{KBr} cm^{-1}: 3300, 1640. NMR ($CDCl_3$+C_5D_5N) δ: 1.03, 1.09 (each 3H, C-4Me$_2$), 1.32 (1H, d, J=5 Hz, C-5-H), 1.57 (1H, dd, J=9 and 14 Hz, C-12β-H), 2.37 (1H, dd, J=9 and 2 Hz, C-9-H), 2.71 (1H, d, J=9 Hz, C-13-H), 2.83 (1H, ddd, J=9, 9, and 14 Hz, C-12α-H), 3.82 (1H, d, J=5 Hz, C-6-H), 3.87 (1H, d, J=10 Hz, C-20-H$_a$), 3.91 (1H, m, C-11-H), 4.14 (1H, dd, J=10 and 2 Hz, C-20-H$_b$), 4.50 (1H, s, C-14-H), 5.16 and 5.21 (each 1H, br s, C-17-H$_2$), 5.34 (1H, br s, C-15-H).

Preparation of a Saturated Ketone (**2**)——A solution of rubescensin C (14.4 mg) in MeOH (1 ml) and 20% HCl (3 ml) was stirred for 67 h at room temperature. The reaction mixture was extracted with AcOEt and the organic layer was washed with water and dried over $MgSO_4$. After evaporation it gave a crystalline residue (13 mg). Recrystallization from MeOH afforded a ketone (**2**). mp 241—243°C. MS (high resolution) Calcd for $C_{20}H_{30}O_6$: M^+ 366.204. Found: 366.208. CD (methanol) $\Delta\varepsilon^{25}$: −1.12 (299) (negative maximum). IR ν_{max}^{KBr} cm^{-1}: 3350, 1710. NMR (C_5D_5N): 1.09, 1.21 (each 3H, s, C-4-Me$_2$), 1.16 (3H, d, J=8 Hz, C-16-H$_3$).

Acknowledgement The authors wish to express their thanks to Mr. T. Suzue and Mr. H. Minami, Taiho Pharmaceutical Co., Ltd., for encouragement throughout this work, and also to Mr. Y. Umeno of the same company, for mass spectral measurement.

References and Notes

1) E. Fujita, T. Fujita, H. Katayama, and M. Shibuya, *J. Chem. Soc.* (*C*), **1970**, 1674.
2) Honan Institute of Medical Sciences, Honan Medical College, Yunnan Institute of Botany, Chengchow Chemicopharmaceutical Plant (Honan, Oeop. R. China), *K'o Hsueh T'ung Pao*, **23**, 53 (1978) [*Chem. Abstr.*, **88**, 115378*p* (1978)].
3) E. Fujita, M. Taoka, M. Shibuya, and T. Fujita, *J. Chem. Soc. Perkin I*, **1973**, 2277.
4) T.-M. Chang, Z.-Y. Chen, J.-H. Chao, Q.-Z. Zhao, H.-D. Sun, and Z.-W. Lin, *K'o Hsueh T'ung Pao*, **25**, 1051 (1980) [*Chem. Abstr.*, **94**, 145226*d* (1981)].
5) The assignments are based on a combination of PND, off-resonance spectra, and comparison with related compounds; T. Fujita, Y. Takeda, and T. Shingu, *Heterocycles*, **16**, 227 (1981); M. Ochi, M. Okamura, H. Ozuki, I. Miura, I. Kubo, and T. Kubota, Abstracts of the 24th Symposium on Essential Oils and Terpenes, Kooriyama, September, 1980, p. 229.
6) T. Kubota, T. Matsuura, T. Tsutsui, S. Uyeo, H. Irie, A. Numata, T. Fujita, and T. Suzuki, *Tetrahedron*, **22**, 1659 (1966).
7) E. Fujita, T. Fujita, M. Shibuya, and T. Shingu, *Tetrahedron*, **25**, 2517 (1969).
8) H.-D. Sun, Z.-W. Lin, C.-Q. Qin, J.-H. Chao, and Q.-Z. Zhao, *Acta Botanica Yunnanica*, **3**(1), 95 (1981).