STRUCTURE OF SANGGENON G, A NEW DIELS-ALDER ADDUCT FROM THE CHINESE CRUDE DRUG "SĀNG-BÁI-PÍ" (MORUS ROOT BARKS)

Toshio Fukai, Yoshio Hano, Tomoko Fujimoto, and Taro Nomura*

Faculty of Pharmaceutical Sciences, Toho University, 2-2-1,
Miyama, Funabashi-shi, Chiba 274, Japan

Abstract —— From the methanol extract of the Chinese crude drug "Sāng-Bái-Pí" (Japanese name Sōhakuhi), root barks of *Morus* sp. (Moraceae), a new flavanone derivative with a fused dihydrochalcone partial moiety was isolated and named sanggenon G. The structure was shown to be I on the basis of spectral data. Sanggenon G (I) is regarded biogenetically as a Diels-Alder adduct of a chalcone derivative and dehydrogeranylflavanone derivative.

In the previous papers[1], we reported the structure determination of a series of isoprenylated flavonoids obtained from the Chinese crude drug "Sāng-Bái-Pí" (Japanese name "Sōhakuhi") imported from the People's Republic of China. Some of them seemed to be natural Diels-Alder adducts[1c-e]. In this paper, we report the isolation and structure determination of a new flavanone derivative, sanggenon G (I), isolated from the methanol extract of the same crude drug.

The methanol extract[1e] of the crude drug was dissolved in ethyl acetate. The ethyl acetate extract was fractionated sequentially by the column and the preparative thin-layer chromatography over silica gel to give sanggenon G (I) in $6\times10^{-4}\%$ yield from the crude drug.

Sanggenon G (I), amorphous powder, $[\alpha]_D^{16}$ -277° (c=0.093 in methanol), gave the FD-MS spectrum which showed the molecular ion peak at m/z 694, and the ^{13}C nmr spectrum (35°C, CD_3OD) indicated the presence of forty carbons [fourteen aliphatic carbons ($2\times CH_3$, $4\times\text{-}CH_2\text{-}$, $3\times{>}CH\text{-}$, ${>}CH\text{-}O\text{-}$, $2\times{>}C{=}C{<}^{H}$), twenty four aromatic carbons (10xCH, 5xC, 9xC-O) and two carbonyl carbons]. Treatment of I with dimethyl sulfate in acetone gave the octamethyl ether (Ia) as an amorphous powder which was negative to $FeCl_3$ test and showed the molecular ion peak at m/z 806 in the FD-MS spectrum. These results suggest the composition of sanggenon G (I) to be $C_{40}H_{38}O_{11}$.

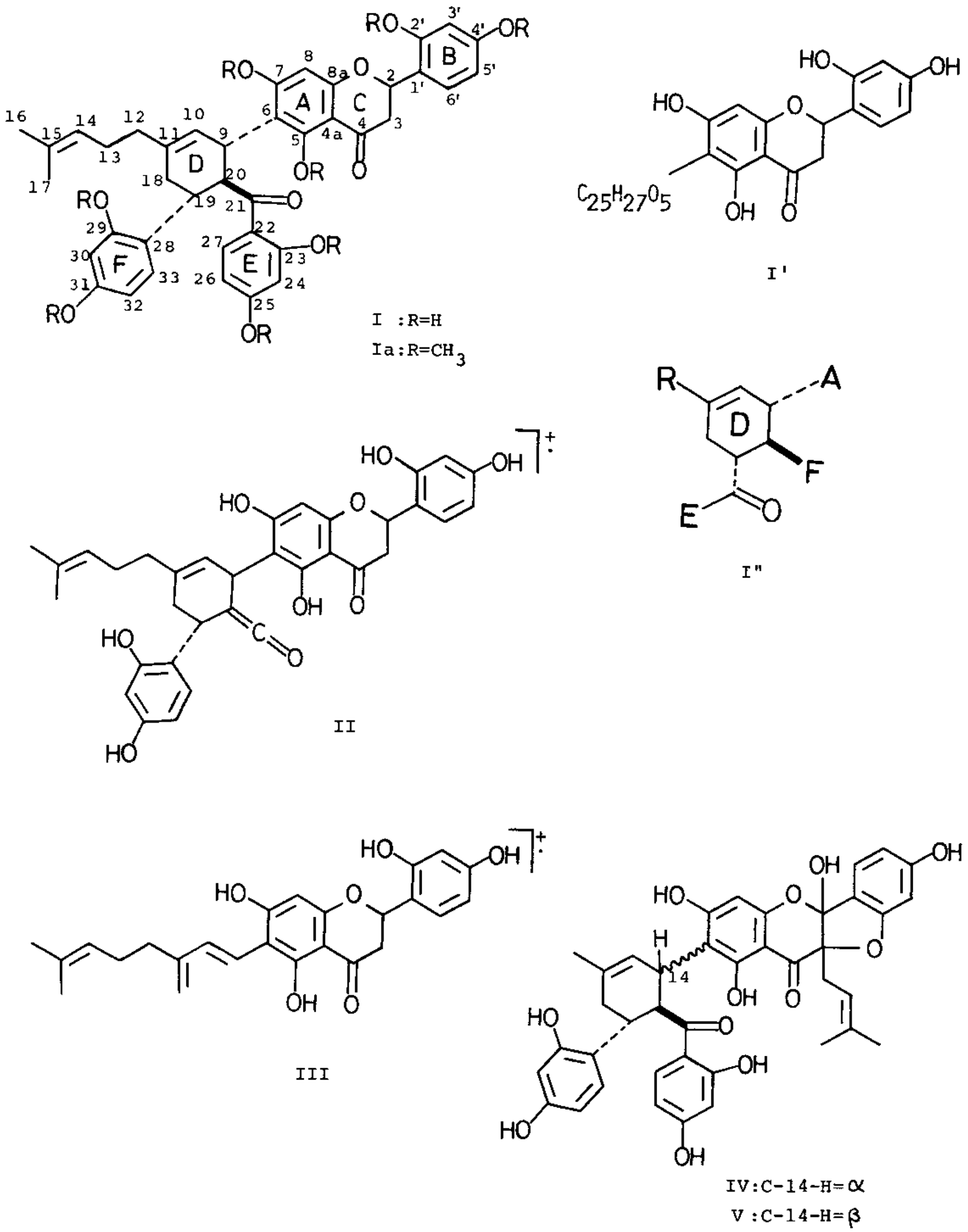
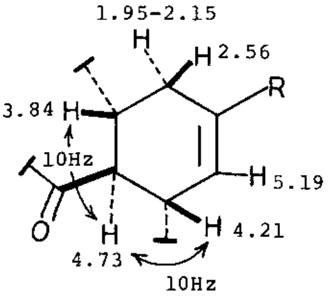

RO
OR
OR
OR
RO
RO
OR
A
B
C
D
E
F
I :R=H
Ia:R=CH3
HO
OH
HO
OH
C25H27O5
I'
R
A
D
F
E
O
I"
HO
OH
HO
OH
HO
HO
II
HO
OH
HO
OH
III
HO
OH
OH
HO
H
14
OH
O
HO
OH
HO
OH
IV:C-14-H=α
V :C-14-H=β

Fig. 1

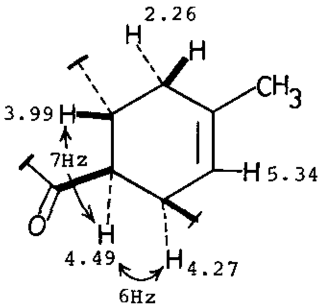
The compound (I) showed the following color reaction: Mg-HCl test (reddish violet), $NaBH_4$ test[2] (reddish brown), $FeCl_3$ test (brown), and showed the following ^1H nmr spectra (acetone-d_6)[3]; δ 2.50-2.70(1H, m, C-3-H-cis)[5], 3.02(1H, dd, J=13 and 16, C-3-H-trans)[4], 5.53(1H, br d, J=13, C-2-H)[4], 12.51(0.5H, OH)[5], 12.91(0.5H, OH)[5], 13.12(1H, s, OH)[5]. These findings show that I is a flavanone derivative having two hydrogen bonded hydroxyl groups. The compound (I) showed the following UV spectra: UV λ_{max}^{EtOH} nm(log ε): 226(sh 4.63), 285(4.43), 290(infl. 4.43), 318(sh 4.10); $\lambda_{max}^{EtOH+AlCl_3}$ nm(log ε): 219(sh 4.72), 290(sh 4.41), 298(4.42), 333(sh 3.95). The UV spectrum of I was similar to those of many known flavanones[6]. In the spectrum of I in the presence of $AlCl_3$, a part of the absorption at 285-290 nm showed a bathochromic shift and the absorption at 290 nm was observed as a shoulder. Taking the ^1H nmr and ^{13}C nmr spectra of I into account, the absorption at 285-290 nm can be ascribed to the two conjugated carbonyl groups which are hydrogen bonded[1d,7]. Sherif et al.[8] reported that $AlCl_3$-induced shift was not observed in the UV spectra when a prenyl group was located ortho to a chelated hydroxyl group. These data led us to presume that one of the ortho positions of the two hydrogen bonded hydroxyl groups is replaced by a prenyl group, and another position is not. Arrangement of substituents in the A and the B ring of the flavanone moiety was assumed by the UV spectra and the ^1H nmr spectrum of I (60°C, acetone-d_6) as follows: δ 5.88(1H, s, C-8-H), 6.39(1H, dd, J=2 and 8, C-5'-H), 6.44(1H, d, J=2, C-3'-H), 7.20(1H, d, J=8, C-6'-H). From these results, the partial structure (I') was suggested.

The FD-MS spectrum of I showed the following fragmentation species: m/z 584 (II), 422 (III). This result suggests that sanggenon G (I) may be a Diels-Alder adduct such as sanggenon C (IV)[1d] and D (V)[1e] regarded as a cycloaddition product with the chalcone and the dehydroprenylflavanone derivative. The presence of the following moieties on the structure of I was supported by a detailed analysis of the ^1H nmr spectrum (60°C, acetone-d_6) using sequential decoupling and by comparison of the ^1H nmr spectra of the prenylflavonoid derivatives obtained from Morus species as follows[1,9]: aromatic protons in a 2,4-dihydroxyphenyl moiety, δ 6.08 (1H, dd, J=2.5 and 8, C-32-H), 6.24(1H, d, J=2.5, C-30-H), 6.90(1H, d, J=8, C-33-H); aromatic protons in a 2,4-dihydroxybenzoyl moiety, δ 6.03(1H, d, J=2.5, C-24-H), 6.12(1H, dd, J=2.5 and 8, C-26-H), 7.67(1H, br d, J=8, C-27-H); protons in a 4-methylpent-3-enyl moiety, δ 1.62, 1.68(each 3H, s, C-15-CH_3), 2.10-2.30(4H, m, C-12-Hx2 and C-13-Hx2), 5.22(1H, m, C-14-H); protons in a cyclohexene ring



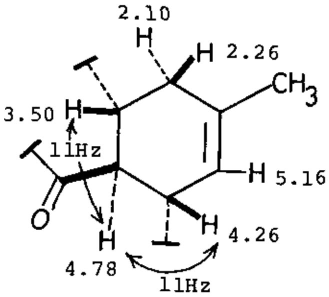
I (dmso-d_6,100°C)

Fig. 2



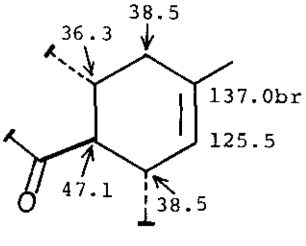
IV (dmso-d_6,120°C)

Fig. 3



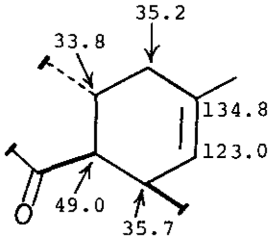
V (dmso-d_6,120°C)

Fig. 4



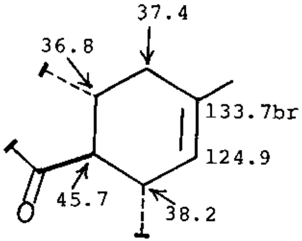
I (CD_3OD,35°C)

Fig. 5



IV (CD_3OD,35°C)

Fig. 6



V (CD_3OD,35°C)

Fig. 7

moiety were shown in Fig. 2. All these results indicate that the structure of sanggenon G is possibly represented by I or I" (except stereochemistry). The location of the 2,4-dihydroxyphenyl and the 2,4-dihydroxybenzoyl moiety on the cyclohexene ring of I was supported by comparison of the ^1H nmr spectrum of I with those of sanggenon C (IV) (Fig. 3) and D (V) (Fig. 4). In respect to the chemical shifts and coupling constants of the relevant cyclohexene ring, I was more similar to V than to IV. In the ^{13}C nmr spectrum of I (35°C, in CD_3OD)[10], the chemical shift values of the carbon atoms of the cyclohexene ring (Fig. 5) were more similar to those of V (Fig. 7) than to those of IV (Fig. 6). These results suggest that sanggenon G (I) and D (V) have the same disposition concerning the location of the 2,4-dihydroxyphenyl and the 2,4-dihydroxybenzoyl moiety on the cyclohexene ring, and also have the same relative configuration. The chemical shift values of the carbon atoms of I, except those of the cyclohexene ring, are summarized as follows: carbon atoms in flavanone moiety, δ 43.1(C-3), 75.8(C-2), 97.0(C-8), 102.9 (C-3'), 103.8(C-4a), 107.8(C-6), 107.9(C-5'), 110.4(C-1'), 156.6(C-2'), 159.5(C-4'), 163.3(C-8a), 165.7(C-5), 165.9(C-7), 198.4(C-4); carbon atoms in a 2,4-dihydroxyphenyl moiety, δ 103.4(C-30), 107.8(C-32), 122.9(C-28), 134.1(C-33), 157.0(C-29 and C-31); carbon atoms in a 2,4-dihydroxybenzoyl moiety, δ 103.8(C-24), 107.4(C-26), 116.2(C-22), 128.8(C-27), 165.5(C-23 and C-25), 210.8(C-21); carbon atoms in 4-methylpent-3-enyl moiety, δ 17.8(C-17), 25.9(C-16), 27.3(C-13), 38.5(C-12), 125.2 (C-14), 130.4(C-15).

From these results, we propose the formula (I) for the structure of sanggenon G. Sanggenon G (I) is optically active and is the first example of a natural product which is considered to be formed by a Diels-Alder type of enzymatic reaction process of a chalcone derivative and a dehydrogeranylflavanone derivative.

ACKNOWLEDGEMENT We are grateful to Dr. M. Takase, Research Laboratory of Zen-Yaku Kogyo Co., Ltd. for ^{1}H- and ^{13}C-nmr spectrum measurement, and to Dr. K. Fukushima, Research Institute for Chemobiodynamics, Chiba University, for FD-MS measurement.

REFERENCES AND FOOTNOTES

1a T. Nomura, T. Fukai, Y. Hano, Y. Sugaya, and T. Hosoya, *Heterocycles*, 1980, **14**, 1785; b T. Nomura, T. Fukai, and Y. Hano, *Planta medica*, in press; c T. Nomura, T. Fukai, Y. Hano, and S. Urano, *Planta medica*, 1983, **47**, 95; d T. Nomura, T. Fukai, Y. Hano, and J. Uzawa, *Heterocycles*, 1981, **16**, 2141; e T. Nomura, T. Fukai, Y. Hano, and J. Uzawa, *Heterocycles*, 1982, **17**, 381; f T. Nomura, T. Fukai, Y. Hano, and K. Tsukamoto, *Heterocycles*, 1983, **20**, No.4, in press.

2 R.M. Horowitz, *J. Org. Chem.*, 1957, **22**, 1733.

3 The ^{1}H nmr spectrum of I observed at room temperature showed the complex patterns with the broad signals. At higher temperature, the ^{1}H nmr spectrum of I showed the simple and clear pattern. The similar phenomenon was observed on sanggenon D (V)[1e].

4 The spectrum was measured at 60°C.

5 The spectrum was measured at 23°C.

6 T.J. Mabry, K.R. Markham, and M.B. Thomas, "The Systematic Identification of Flavonoids", Spring-Verlag, Berlin, 1970, Part II.

7 Y. Oshima, C. Konno, H. Hikino, and M. Matsushita, *Tetrahedron Lett.*, 1980, **21**, 3381.

8 C.A. Sherif, R.K. Gupta, and M. Krishnamurti, *Tetrahedron Lett.*, 1980, **21**, 641.

9 T. Nomura and T. Fukai, *Heterocycles*, 1981, **15**, 1531, and references cited therein.

10 Assignments of the carbon atoms in I were performed by off-resonance decoupling technique as well as by comparison of the ^{13}C nmr spectra of the model compounds[1,9].

Received, 21st January, 1983