COMPONENTS OF THE ROOT BARK OF *MORUS CATHAYANA*. 1.[1] STRUCTURES OF FIVE NEW ISOPRENYLATED FLAVONOIDS, SANGGENOLS A – E AND A DIPRENYL-2-ARYLBENZOFURAN, MULBERROFURAN V

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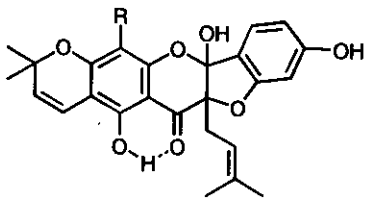
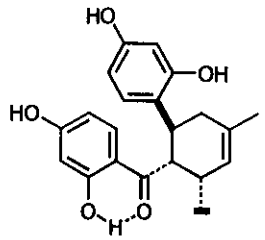
Abstract ——— Five new isoprenylated flavonoids, sanggenols A (**1**), B (**2**), C (**3**), D (**4**), and E (**5**), along with a diprenyl-2-arylbenzofuran, mulberrofuran V (**6**), were isolated from the Chinese moraceous plant, *Morus cathayana*. The structures of sanggenols A – E, and mulberrofuran V were shown to be **1** – **5** and **6**, respectively, on the basis of spectroscopic evidence. Furthermore fourteen kinds of known phenolic compounds were also isolated.

Previously we reported the structure determination of a series of isoprenylated phenols from the moraceous plants.[2,3] Some of these compounds showed interesting biological activities such as hypotensive effect,[2] anti-tumor promoting activity,[4] and inhibitory activities against arachidonate 5-lipoxygenase[5] and testosterone 5α-reductase.[6] In the course of our studies on the constituents of the moraceous plants, we examined the constituents of *Morus cathayana* HEMSLEY, collected in China. This paper deals with the characterization of five new isoprenylated flavonoids, and an isoprenylated 2-arylbenzofuran, along with the isolation of fourteen kinds of known phenolic compounds as follows; sanggenons A (**7**),[7] C (**8**),[8] L (**9**),[9] M (**10**),[9] O (**11**),[10] moracins C (**12**),[11] D (**13**),[11] M (**14**),[12] P (**15**),[12] scopoletin (**16**), umbelliferone (**17**), isoliquiritigenin (**18**), isobavachalcone (**19**),[13] and 2',4',7-trihydroxy-(2*S*)-flavanone (**20**)[14] (Figure 1).

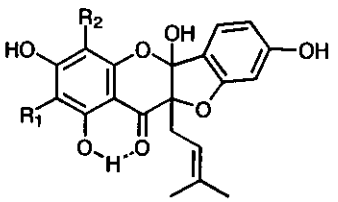
Sanggenol A (**1**), an amorphous powder, $C_{25}H_{28}O_6$, $[\alpha]_D$ + 11°, showed positive to the methanolic ferric chloride test. The uv spectrum of **1** exhibited maxima at 209, 230 (sh), 289, 300 (sh) nm and was similar to those of flavanones.[15] The ^{1}H nmr spectrum of **1** showed the signals of the following protons



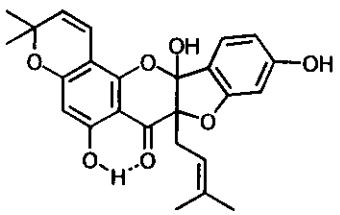
7 : R = H
9 : R = prenyl



A



8 : R_1 = A, R_2 = H
11 : R_1 = H, R_2 = A



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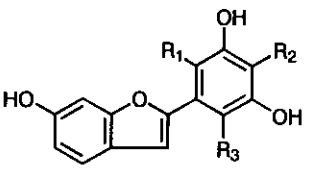
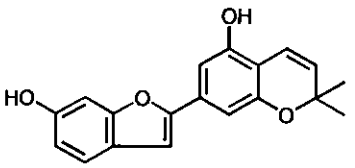


6: R_1 = R_3 = prenyl, R_2 = H
12 : R_1 = R_3 = H, R_2 = prenyl
14 : R_1 = R_2 = R_3 = H



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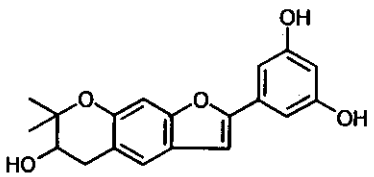

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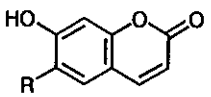
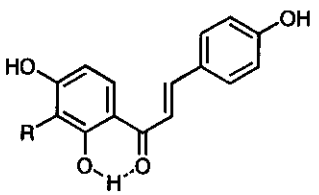
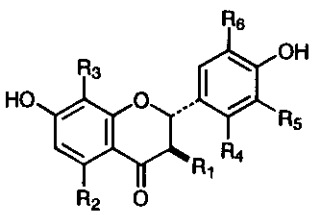


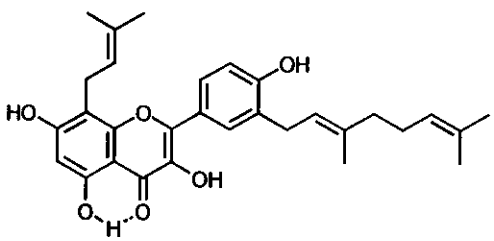
16 : R = OMe
17 : R = H



18 : R = H
19 : R = prenyl



1 : R_1 = R_3 =R_6 = H, R_2 = R_4 = OH, R_5 = geranyl
3 : R_1 = R_2 = OH, R_3 = prenyl, R_4 =R_6 = H, R_5 = geranyl
4 : R_1 = R_2 = R_4 = OH, R_3 = H, R_5 = geranyl, R_6 = prenyl
5 : R_1 = R_2 = R_4 = OH, R_3 = R_6 = prenyl, R_5 = geranyl
20 : R_1 = R_2 = R_3 = R_5 = R_6 = H, R_4 = OH



2

Figure 1.

Table 1. ^{13}C Nmr data of **1**, **3** – **5** in acetone-d_6 (100 or 125 MHz)

C	1	3	4	5	C	1	3	4	5	C	3	4	5
2	76.6	84.5	80.0[a]	79.8	(geranyl)					(prenyl)			
3	42.4	73.2	73.0[b]	73.5	1"	23.2	29.0	23.5	23.5	9	22.1		22.1
4	197.9	198.6	198.1	198.0	2"	123.8	123.5	123.7	123.8	10	123.5		123.5
4a	103.5	101.6	102.0	101.5	3"	136.0	136.5	136.1	135.9	11	131.4		131.5
5	164.8	162.7	165.1	162.8	4"	16.5	16.3	16.3	16.3	12	17.7[c]		17.7[c]
6	97.1	96.7	97.2	96.8	5"	40.7	40.6	40.5	40.5	13	25.9[d]		25.9[d]
7	167.5	165.5	167.9	165.6	6"	27.6	27.5	27.4	27.4				
8	96.2	108.6	96.1	108.8	7"	125.3	125.1	125.1	125.1	1"'		29.4	28.3
8a	165.6	161.0	164.4	161.0	8"	132.0	131.8	131.9	131.8	2"'		123.7	123.8
1'	116.8	129.4	116.9	117.4	9"	17.9	17.9[c]	17.8[c]	17.9	3"'		133.9	132.8
2'	154.4	130.1	152.5	152.7	10"	26.1	25.8[d]	26.0[d]	25.8[d]	4"'		18.0[c]	17.9[c]
3'	118.2	128.5	117.4	117.3						5"'		25.9[d]	25.9[d]
4'	157.4	156.3	154.4	154.3									
5'	108.5	115.4	120.8	120.8									
6'	126.0	127.5	126.6	126.1									

[a]br d, J = 148 Hz. [b]br d, J = 145 Hz. [c,d]The signals may be interchanded in each compound.

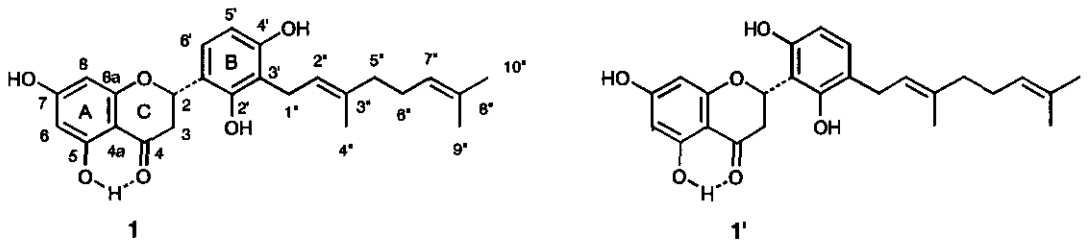
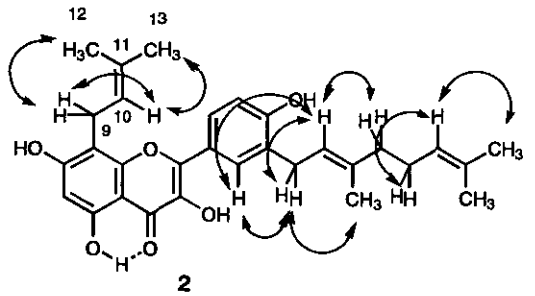


Figure 2.

(acetone-d_6, 400 MHz): protons in a geranyl group, δ 1.55 (3H, br s), 1.61, 1.78 (each 3H, br d, J = 1 Hz), 1.96 (2H, br t, J = 8 Hz), 2.05 (2H, m), 3.44 (2H, br d, J = 6.5 Hz), 5.07 (1H, br t, J = 7 Hz), 5.26 (1H, br t, J = 6.5 Hz), AMX type protons, δ 2.70 (1H, dd, J = 3 and 17 Hz), 3.21 (1H, dd, J = 13 and 17 Hz), 5.74 (1H, dd, J = 3 and 13 Hz), *meta*-coupled aromatic protons, δ 5.94, 5.95 (each 1H, d, J = 2 Hz), *ortho*-coupled aromatic protons, δ 6.52, 7.13 (each 1H, d, J = 8Hz), and protons in three hydroxyl groups, δ 7.39, 8.44 (each 1H, br s), 9.70 (1H, br), and a hydrogen-bonded hydroxyl group, δ 12.19 (1H, s). The ^{13}C nmr spectrum of **1** was analyzed by comparing with the spectra of flavanone derivatives as shown in Table 1. In the spectrum, two oxygenated carbons in the B ring appeared at δ 154.4 and 157.4 indicating that these carbon atoms located at *meta*-positions to each other.[16] From the above results, two possible structures (**1** and **1'**) were suggested (Figure 2). Although, the structure (**1'**) could be eliminated by the following result: Fukai and Nomura have reported that the chemical shift of the

benzylic methylene signal (C-1") of prenyl or geranyl group was depended upon the substituents located at the adjacent positions.[17] In the ^{13}C nmr spectrum of **1**, the benzylic methylene carbon of the geranyl group was observed at δ 23.2. This chemical shift value suggested that the di*ortho*-positions to the geranyl group were replaced by the oxygenated substituents. The absolute configuration of **1** was assigned to be 2*S* by its CD spectrum, in which the positive Cotton effects were exhibited at 315 (sh) and 330 nm.[18] Thus, the structure of sanggenol A was characterized as formula (**1**).

Sanggenol B (**2**), yellow needles, mp 138 – 140 °C, $C_{30}H_{34}O_6$, showed positive reaction to methanolic ferric chloride reaction. The uv spectrum exhibited maxima at 205, 230 (sh), 254, 272, 327, 376 nm and was similar to those of flavonol derivatives.[15] The ^1H nmr spectrum of **2** showed the signals of the following protons (acetone-d_6, 500 MHz): protons in a geranyl group, δ 1.57, 1.61, 1.76 (each 3H, br s), 2.08 (2H, m), 2.12 (2H, br t, J = 7 Hz), 3.42 (2H, br d, J = 7 Hz), 5.12, 5.42 (each 1H, br t, J = 7 Hz), protons in a prenyl group, δ 1.65, 1.80 (each 3H, br s), 3.56 (2H, br d, J = 7 Hz), 5.31 (1H, br t, J = 7 Hz), ABX type aromatic protons, δ 7.02 (1H, d, J = 8.5 Hz), 8.02 (1H, d, J = 2 Hz), 8.05 (1H, dd, J = 2 and 8.5 Hz), an aromatic proton, δ 6.34 (1H, s), and protons in three hydroxyl groups, δ 7.92, 9.02, 9.65 (each 1H, br s), and a hydrogen-bonded hydroxyl group, δ 12.11 (1H, s). The location of the geranyl group was indicated by the NOESY spectrum as shown in Figure 3. On the other hand, the location of the prenyl group in the A ring was supported by the following result: Fukai and Nomura reported that the 6- and 8-prenylflavonol could be distinguished by the chemical shifts of the proton signal of the hydrogen-bonded hydroxyl group at the C-5 position and the aromatic proton in the A ring.[19,20] The compound (**2**) showed the proton signals of the hydrogen-bonded hydroxyl group (5-OH) and the aromatic proton at δ 12.11 and 6.34, respectively. These chemical shift values were good agreement with those of the relevant protons of 8-prenylflavonol derivatives.[19,20] From the above results, the structure of sanggenol B was determined as formula (**2**).



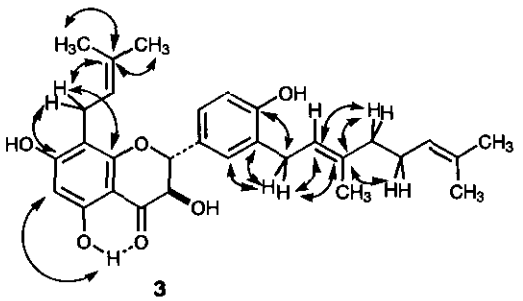
2

C-9-H_2: δ 3.56	C-1"-H_2: 3.42
C-10-H: 5.31	C-2"-H: 5.42
C-12-H_3: 1.80	C-4"-H_3: 1.76
C-13-H_3: 1.65	C-5"-H_2: 2.08
	C-6"-H_2: 2.12
	C-7"-H: 5.12
	C-9"-H_3: 1.57
	C-10"-H_3: 1.61

Figure 3. NOE data of **2** with NOESY spectrum.

Sanggenol C (**3**), an amorphous powder, $C_{30}H_{36}O_6$, $[\alpha]_D$ + 19°, showed positive reaction to methanolic ferric chloride reaction. The uv spectrum exhibited maxima at 207, 230 (sh), 296, 340 nm and was similar

to those of flavanones.[15] The ^{1}H nmr spectrum of **3** showed the signals of the following protons (acetone-d_6, 500 MHz): protons in a geranyl group, δ 1.57, 1.74 (each 3H, br s), 1.62 (3H, br d, J = 1 Hz), 2.05, 2.11 (each 2H, m), 3.39 (2H, br d, J = 7 Hz), 5.13, 5.42 (each 1H, br t, J = 7 Hz), protons in a prenyl group, δ 1.56, 1.60 (each 3H, br d, J = 1 Hz), 3.18 (2H, br d, J = 7 Hz), 5.18 (1H, br t, J = 7 Hz), AXY type protons, δ 4.60 (1H, dd, J = 3.5 and 12 Hz, C-3-H), 4.64 (1H, d, J = 3.5 Hz, C-3-OH), 5.03 (1H, d, J = 12 Hz, C-2-H), ABX type aromatic protons, δ 6.90 (1H, d, J = 8 Hz), 7.26 (1H, dd, J = 2 and 8 Hz), 7.35 (1H, d, J = 2 Hz), an aromatic proton, δ 6.07 (1H, s), and protons in two phenolic hydroxyl groups, δ 8.40, 9.71 (each 1H, br s), and a hydrogen-bonded hydroxyl group, δ 11.64 (1H, s). The ^{13}C nmr spectrum was analyzed by comparing with those of 3-hydroxyflavanones as shown in Table 1.[21] The locations of the geranyl and prenyl groups were confirmed by the HMBC spectrum of **3** as shown in Figure 4. The absolute configuration of **3** was assigned to be 2*R*,3*R* by its CD spectrum, in which the negative and positive Cotton effects were exhibited at 295 and 331 nm, respectively.[18] From the above results, the structure of sanggenol C was elucidated as formula (**3**).



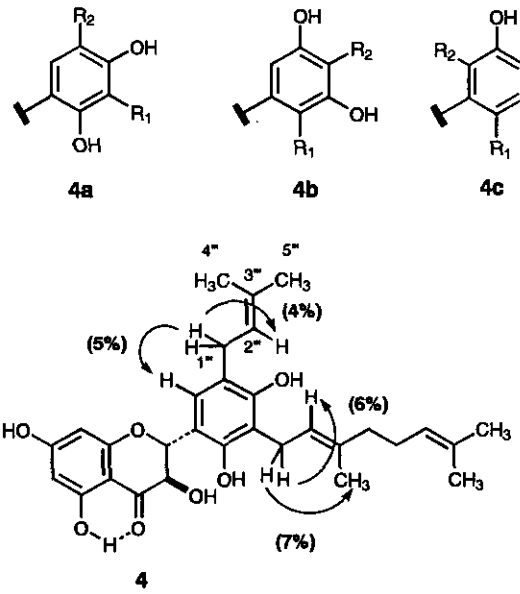
3

C-9-H_2:	δ 3.18	C-1"-H_2:	3.39
C-10-H :	5.18	C-2"-H :	5.42
C-12-H_3:	1.56	C-4"-H_3:	1.74
C-13-H_3:	1.60	C-5"-H_2:	2.05
		C-6"-H_2:	2.11
		C-7"-H :	5.13
		C-9"-H_3:	1.57
		C-10"-H_3:	1.62

Figure. 4 Long-range coupling correlations using the structure determination of **3**.

Sanggenol D (**4**), an amorphous powder, $C_{30}H_{36}O_7$, $[\alpha]_D$ + 43°, showed positive to the methanolic ferric chloride test. The uv spectrum exhibited maxima at 210, 230 (sh), 291, 330(sh) nm and was similar to that of **3**. The ^{1}H nmr spectrum of **4** showed the signals of the following protons (acetone-d_6, 400 MHz): protons in a geranyl group, δ 1.56, 1.62, 1.78 (each 3H, br s), 1.97 (2H, br t, J = 7 Hz), 2.05 (2H, m), 3.48 (2H, br d, J = 7 Hz), 5.08, 5.24 (each 1H, br t, J = 7 Hz), protons in a prenyl group, δ 1.69 (3H, br s), 1.70 (3H, br d, J = 1 Hz), 3.32 (2H, br d, J = 7 Hz), 5.31 (1H, br t, J = 7 Hz), AMX type protons, δ 4.74 (1H, dd, J = 3 and 11 Hz, C-3-H), 5.20 (1H, br d, J = 3 Hz, C-3-OH), 5.49 (1H, d, J = 11 Hz, C-2-H), *meta*-coupled aromatic protons, δ 5.95, 5.99 (each 1H, d, J = 2 Hz), an aromatic proton, δ 7.13 (1H, br s), and protons in three phenolic hydroxyl groups, δ 7.03, 7.30 (each 1H, s), 9.74 (1H, br), and a hydrogen-bonded hydroxyl group, δ 11.70 (1H, s). The ^{13}C nmr spectrum of **4** was analyzed by comparing with that of **3** as shown in Table 1. In the spectrum, two oxygenated carbons in the

B ring appeared at δ 152.5 and 154.4 indicating that these carbons located at *meta*-positions to each other.[16] These data indicated that the partial structure of the B ring is **4a** – **4c** or **4d**. The benzylic methylene carbons of the geranyl and prenyl groups were observed at δ 23.5 and 29.4. These chemical shifts suggested that the *ortho*-positions to one of the isoprenyl groups were replaced by hydroxyl groups, and the other isoprenyl group is adjacent to a hydroxyl group and the other position is unsubstituted [partial structure (**4a**)]. The locations of the geranyl and prenyl groups were elucidated with NOE experiment as shown in Figure 5. The absolute configuration of **4** was assigned to be 2*R*,3*R* by its CD spectrum, in which the negative and positive Cotton effects were exhibited at 294 and 324 nm, respectively.[18] From the above results, the structure of sanggenol D was assigned as formula (**4**).



C-1"-H_2: δ 3.48
C-2"-H: 5.24
C-4"-H_3: 1.78
C-5"-H_2: 1.97
C-6"-H_2: 2.05
C-7"-H: 5.08
C-9"-H_3: 1.56
C-10"-H_3: 1.62

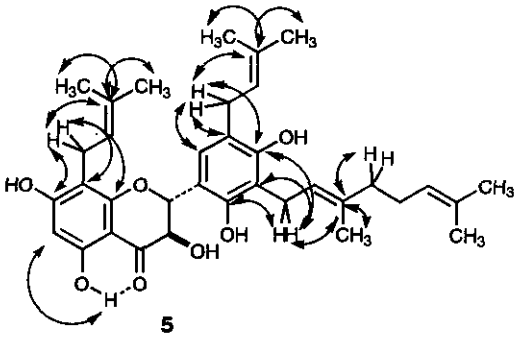
C-1'''-H_2: 3.32
C-2'''-H: 5.31
C-4'''-H_3 } 1.69
C-5'''-H_3 } 1.70

Figure 5. Partial structures (**4a** – **d**) and NOE data of **4**.

(The spin-networks of the geranyl and prenyl groups were confirmed with decoupling experiment.)

Sanggenol E (**5**), an amorphous powder, $[\alpha]_D$ + 34°, showed positive reaction to methanolic ferric chloride reaction and gave the molecular ion peak at *m/z* 576 in the EI mass spectrum. The ^{13}C nmr spectrum of **5** showed the presence of thirty five carbons (Table 1), along with the ^1H nmr spectrum exhibited forty four proton signals. These results suggest the molecular formula to be $C_{35}H_{44}O_7$. The uv spectrum of **5** exhibited maxima at 211, 230 (infl.), 292, 340(sh) nm and was similar to those of **3** and **4**. The ^1H nmr spectrum of **5** showed the signals of the following protons (acetone-d_6, 400 MHz): protons in a geranyl group, δ 1.56 (3H, br s), 1.62, 1.78 (each 3H, br d, J = 1 Hz), 1.98 (2H, br t, J = 8 Hz), 2.08 (2H, m), 3.48 (2H, br d, J = 7 Hz), 5.07, 5.26 (each 1H, br t, J = 7 Hz), protons in two prenyl groups,

δ 1.70 (3H, br s), 1.71 (3H, br d, J = 1 Hz), 3.33 (2H, br d, J = 7 Hz), 5.33 (1H, br t, J = 7 Hz); 1.60 (6H, br d, J = 1 Hz), 3.19, 3.23 (each 1H, br dd, J = 7 and 14 Hz), 5.18 (1H, br t, J = 7 Hz), ABX type protons, δ 4.65 (1H, dd, J = 3.5 and 11 Hz, C-3-H), 5.41 (1H, br d, J = 3.5 Hz, C-3-OH), 5.45 (1H, d, J = 11 Hz, C-2-H), two aromatic protons, δ 6.08 (1H, s), 7.17 (1H, br s), and protons in three phenolic hydroxyl groups, δ 7.02, 7.32 (each 1H, s), 9.85 (1H, br s), and a hydrogen-bonded hydroxyl group, δ 11.64 (1H, s). The ^{13}C nmr spectrum of **5** was analyzed by comparing with those of **3** and **4** as shown in Table 1. In the spectrum, the chemical shifts of all the carbon atoms of the B ring as well as the geranyl and one of the prenyl groups were good agreement with those of the relevant carbon atoms of **4**. Furthermore the EI-mass spectrum of **5** gave the fragment ion at m/z 221 (**21**). These results suggest that **5** seems to be 6- or 8-prenylsanggenol D. The locations of the geranyl and the two prenyl groups were confirmed by the HMBC spectrum of **5** as shown in Figure 6. The absolute configuration of **5** was assigned to be 2*R*,3*R* by its CD spectrum, in which the negative and positive Cotton effects were exhibited at 296 and 338 nm, respectively.[18] From the above results, the structure of sanggenol E was elucidated as formula (**5**).



C-1"-H_2: δ 3.48 C-1"'-H_2: 3.30
C-2"-H : 5.26 C-2"'-H : 5.33
C-4"-H_3: 1.78 C-4"'-H_3 1.70
C-5"-H_2: 1.98 C-5"'-H_3 1.71
C-6"-H_2: 2.08
C-7"-H : 5.08
C-9"-H_3: 1.56
C-10"-H_3: 1.62

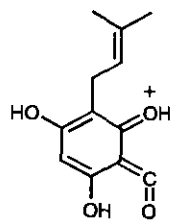


Figure 6. Long-range coupling correlations using the structure determination of **5**.

Mulberrofuran V (**6**), an amorphous powder, $C_{24}H_{26}O_4$, gave a pale brown coloration with the methanolic ferric chloride test. The uv spectrum exhibited maxima at 205, 250 (sh), 394 nm and was similar to those of 2-arylbenzofuran derivatives.[22] The ^{1}H nmr spectrum of **6** showed the signals of the following protons (acetone-d_6, 400 MHz): protons in two prenyl groups, δ 1.34, 1.53 (each 6H, br d, J = 1 Hz), 3.12 (4H, br d, J = 7 Hz), 5.10 (2H, br t, J = 7 Hz), AMNX type protons (three aromatic protons and an olefinic proton), δ 6.54 (1H, d, J = 0.9 Hz), 6.81 (1H, dd, J = 2 and 8 Hz), 6.94 (1H, ddd, J = 0.4, 0.9 and 2 Hz), 7.42 (1H, dd, J = 0.4 and 8 Hz), an aromatic proton, δ 6.59 (1H, s). The ^{13}C nmr spectrum of **6** was analyzed by comparing with those of 2-arylbenzofuran derivatives (Table 2).[23] In the spectrum, the relevant carbons of the two prenyl groups and the carbons of the C-2' and C-6' positions as well as those of the C-3' and C-5' positions were observed as equivalent. From these results, two possible structure (**6**

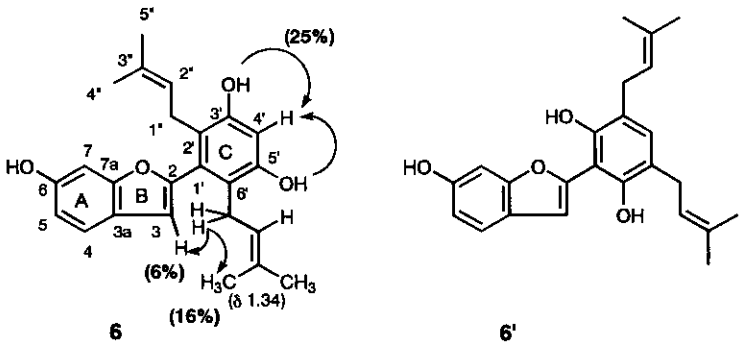


Figure 7. NOE data of **6**.

Table 2. ^{13}C Nmr data of **6**

C	**6**	C	**6**
2	159.5	1'	133.4
3	105.0	2',6'	121.2
3a	122.6	3',5'	156.7
4	121.8	4'	107.0
5	112.9	1"	27.3
6	156.7	2"	125.4
7	98.7	3"	130.1
7a	157.0	4"	18.0
		5"	26.1

(in acetone-d_6, 100 MHz)

and **6'**) could be suggested. The structure (**6**) was supported from the following evidence: The benzylic methylene carbon of the prenyl group was observed at δ 27.3. This chemical shift value supported that one of the *ortho*-positions to the prenyl groups was replaces by a carbon function and another by a hydroxyl group.[17b] Furthermore, the locations of the prenyl groups were confirmed by the NOE experiment as described in Figure 7. From the above results, the structure of mulberrofuran V was characterized as formula (**6**).

Compound (**20**), an amorphous powder, $[\alpha]_D$ +50°, gave the protonated molecular ion peak at *m/z* 273 in its FAB mass spectrum. The ^{1}H nmr spectrum of **20** showed the signals of the following protons (acetone-d_6, 400 MHz): AMX type protons, δ 2.67 (1H, dd, J = 3 and 17 Hz), 3.03 (1H, dd, J = 13 and 17 Hz), 5.72 (1H, dd, J = 3 and 13 Hz), two sets of AXY type protons, δ 6.44 (1H, d, J = 2 Hz), 6.44 (1H, dd, J = 2 and 8 Hz), 6.48 (1H, d, J = 2 Hz), 6.58 (1H, dd, J = 2 and 8.5 Hz), 7.34 (1H, d, J = 8 Hz), 7.74 (1H, d, J = 8.5 Hz), and protons in three hydroxyl groups, δ 8.39, 8.62, 9.42 (each 1H, br s). From these results, the compound (**20**) was supported to be 2',4',7-trihydroxyflavanone. While the compound (**20**) has been synthesized by Nabaei-Bidhendi and Bannerjee,[14] to our knowledge, this is the first time that it has been identified as a natural product. The absolute configuration of **20** was assigned to be 2*S* by its CD spectrum.[18]

EXPERIMENTAL

Abbreviations: s = singlet, d = doublet, dd = double doublet, ddd = doublet of double doublet, t = triplet, m = multiplet, br = broad, sh = shoulder, infl. = inflection, fr. = fraction. The optical rotations and CD spectra were measured on JASCO DIP-370 and JASCO J-720W instruments, respectively. The other

general procedures and instruments used in our previous papers.[24]

Plant material: Root bark of *Morus cathayana* HEMSLEY was collected in Tian-Zhu Prefecture, Gui-Zhou Province, China, in March 1993 and identified by C.-G. X. The sample was deposited in TOHO herbarium of Toho University.

Isolation of Sanggenols A (1), B (2), C (3), D (4), E (5), and Mulberrofuran V (6)

The dried root bark of *M. cathayana* (3.2 kg) was extracted with methanol (5 *l*) on reflux for 2 h. This procedure was repeated four times. Evaporation of the methanol solution to dryness yielded 262 g of a residue. The residue was extracted with acetone (1 *l*). The acetone solution was concentrated to afforded a residue (57.8 g), which was extracted with ethyl ether (300 m*l*). Evaporation of the solution gave a residue (50 g), which was extracted with *n*-hexane to afforded an oily substance (31 g). The insoluble material (19 g) with *n*-hexane was chromatographed over silica gel (300 g) using benzene-acetone solution (each fraction; 500 m*l*, Column A). The fractions eluted with benzene-acetone (99:1), frs. 20 – 21, 22 – 24, 25 – 26, and 27 – 30, were evaporated to give the residues, 150, 220, 90, and 190 mg, respectively. The frs. 20 – 21, was fractionated by preparative tlc [silica gel, chloroform-ethyl ether (4:1), benzene-methanol (10:1), benzene-ethyl acetate (3:1)] to give sanggenons A (**7**, 20 mg),[7] L (**9**, 15 mg),[9] and M (**10**, 1.5 mg).[9] The frs. 22 – 24 was fractionated by preparative tlc [benzene-acetone (10:1), benzene-methanol (10:1), *n*-hexane-ethyl acetate (1:1), chloroform-methanol (10:1), *n*-hexane-acetone (1:1)] to give sanggenol E (**5**, 4 mg). The frs. 25 – 26 was fractionated by preparative tlc [benzene-acetone (10:1), benzene-methanol (10:1), *n*-hexane-ethyl acetate (1:1), chloroform-methanol (10:1), *n*-hexane-acetone (1:1)] to give sanggenol D (**4**, 2 mg). The frs. 27 – 30 was fractionated by preparative tlc [*n*-hexane-ethyl ether (1:2), benzene-methanol (10:1)] followed by preparative hplc [solvent, *n*-hexane-ethyl acetate (4:1), column, Senshu Pak SSC-Silica 4251-N, 1 cmϕ × 25 cm, detector, uv 280 nm] to give sanggenol B (**2**, 2 mg).

The fractions eluted with benzene-acetone (98:2), frs. 31 – 36 and 37 – 41, were evaporated to give the residues, 100 and 160 mg, respectively. The frs. 31 – 36 was fractionated by preparative tlc [benzene-acetone (4:1), chloroform-methanol (50:1)] to give moracin D (**13**, 2 mg).[11] The frs. 37 – 41 was fractionated by preparative tlc [benzene-acetone (4:1), chloroform-methanol (50:1, 20:1), *n*-hexane-ethyl acetate (1:1), *n*-hexane-acetone (4:1)] to give sanggenol A (**1**, 3 mg), isobavachalcone (**19**, 10 mg),[13] umbelliferone (**17**, 1.5 mg), scopoletin (**16**, 8 g), and moracin C (**12**, 17.5 mg).[11]

The fractions eluted with benzene-acetone (97:3), frs. 46 – 48 and 49 – 54, were evaporated to give the residue, 145 and 180 mg, respectively. The frs. 46–48 was fractionated by preparative tlc [*n*-hexane-ethyl acetate (4:1), *n*-hexane-acetone (4:1), benzene-methanol (95:5)] to give sanggenol C (**3**, 6 mg) and mulberrofuran V (**6**, 2 mg). The frs. 49 – 54 was fractionated by preparative tlc [benzene-acetone (5:1), *n*-hexane-acetone (7:3)] followed by preparative hplc [*n*-hexane-ethyl acetate (4:1), *n*-hexane-acetone (7:1)] to give isoliquiritigenin (**18**, 0.2 mg).

The fractions eluted with benzene-acetone (95:5) was evaporated to give the residue, 540 mg. This residue was rechromatographed over silica gel (50 g) with chloroform-methanol (95:5, each fraction; 300 m*l*, frs. 1' – 37', column B). Moracins P (**15**, 32 mg)[12] and M (**14**, 40 mg)[12] were obtained from the frs. 20' – 21' and 31' – 37' of the column, respectively. The fr. 18' (10 mg) was fractionated by preparative tlc

[*n*-hexane-acetone (3:2), benzene-methyl ethyl ketone (1:1)] to give compound (**20**) (0.5 mg).[14]
The fractions eluted with benzene-acetone (9:1) of column A was evaporated to give the residue, 1.2 g. This residue was rechromatographed over silica gel (50 g), with chloroform-methanol (10:1, each fraction; 300 m*l*, frs, 1" – 5", column C). The fr. 5" of the column (50 mg) was fractionated by preparative hplc [solvent, methanol-H_2O (7:3), column, Shiseido Pak C18, 1cmø×25 cm, detector, uv 280] to give sanggenons C (**8**, 11.5 mg)[8] and O (**11**, 6 mg).[10] The known compounds were identified by direct comparison with respective authentic samples or their spectral data.

Sanggenol A (1)

Compound (**1**) was obtained as a colorless amorphous powder. $FeCl_3$ test on a tlc pate: positive (reddish brown). $[\alpha]_D^{22}$ +11° (*c* 0.16, MeOH). Uv λ_{max}^{MeOH} nm (log ε): 209 (4.62), 230 (sh 4.41), 289 (4.17), 300 (sh). EI-ms (probe): 70 eV, *m/z* (rel. int.) 425 $[M+1]^+$ (10%), 424 $[M]^+$ (33), 407 (7), 406 (13), 381 (6), 368 (5), 355 (6), 339 (17), 337 (15), 284 (17), 202 (14), 165 (14), 153 (100), 123 (27). HR-ms: *m/z* 424.1895 (M^+, $C_{25}H_{28}O_6$ requires: 424.1886). CD ($c = 5.34\times10^{-5}$ g/m*l*, MeOH): $[\theta]_{227}$ 0, $[\theta]_{229}$ −140, $[\theta]_{243}$ 0, $[\theta]_{258}$ +1100, $[\theta]_{290}$ +160 (valley), $[\theta]_{315}$ +950 (sh), $[\theta]_{330}$ +1200.

Sanggenol B (2)

Compound (**2**) was recrystallized from acetone to give yellow needles, mp 138 – 140 °C. $FeCl_3$ test on a tlc plate: positive (reddish brown). Uv λ_{max}^{MeOH} nm (log ε): 205 (4.59), 230 (sh 4.22), 254 (4.12), 272 (4.16), 327 (3.92), 376 (4.07). EI-ms: *m/z* (rel. int.) 491 $[M+1]^+$ (7%), 490 $[M]^+$ (20), 473 (5), 436 (5), 422 (6), 421 (9), 405 (19), 368 (17), 275 (5), 219 (100), 217 (11), 203 (25), 189 (26), 175 (11), 165 (13), 123 (31), 69 (93). HR-ms: *m/z* 490.2367 (M^+, $C_{30}H_{34}O_6$ requires: 490.2355).

Sanggenol C (3)

Compound (**3**) was obtained as a pale yellow amorphous powder. $FeCl_3$ test on a tlc plate: positive (reddish brown). $[\alpha]_D^{22}$ +19° (*c* 0.125, MeOH). Uv λ_{max}^{MeOH} nm (log ε): 207 (4.65), 230 (sh 4.37), 296 (4.19), 340 (3.60). EI-ms: *m/z* (rel. int.) 493 $[M+1]^+$ (5%), 492 $[M]^+$ (12), 463 (4), 436 (4), 394 (7), 368 (5), 339 (7), 331 (8), 270 (34), 221 (18), 219(15), 167 (12), 165 (74), 123 (46), 69 (100). HR-ms: *m/z* 492.2519 (M^+, $C_{30}H_{36}O_6$ requires: 492.2512). CD ($c = 2.5\times10^{-5}$ g/m*l*, MeOH): $[\theta]_{223}$ +26500, $[\theta]_{241}$ +15000, $[\theta]_{258}$ +5000, $[\theta]_{277}$ 0, $[\theta]_{295}$ −37000, $[\theta]_{312}$ 0, $[\theta]_{331}$ +7800, $[\theta]_{400}$ +520.

Sanggenol D (4)

Compound (**4**) was obtained as a pale yellow amorphous powder. $FeCl_3$ test on a tlc plate: positive (reddish brown). $[\alpha]_D^{22}$ +43° (*c* 0.063, MeOH). Uv λ_{max}^{MeOH} nm (log ε): 210 (5.00), 230 (sh 4.75), 291 (4.61), 330 (sh4.12). EI-ms: *m/z* (rel. int.) 509 $[M+1]^+$ (3%), 508 $[M]^+$ (6), 490 (11), 424 (16), 382 (46), 327 (43), 259 (49), 257 (71), 243 (43), 203 (91), 153(65), 126 (56), 123 (62), 69 (100). HR-ms: *m/z* 508.2451 (M^+, $C_{30}H_{36}O_7$ requires: 508.2461). CD ($c = 1.1\times10^{-5}$ g/m*l*, MeOH): $[\theta]_{218}$ 0, $[\theta]_{229}$ +27800, $[\theta]_{243}$ +7800, $[\theta]_{257}$ +10500, $[\theta]_{285}$ 0, $[\theta]_{294}$ −7300, $[\theta]_{305}$ 0.

Sanggenol E (5)

Compound (**5**) was obtained as a pale yellow amorphous powder. $FeCl_3$ test on a tlc plate: positive (reddish brown). $[\alpha]_D^{22}$ +34° (*c* 0.035, MeOH). UV λ_{max}^{MeOH} (log ε): 211 (4.71), 230 (infl. 4.41), 292 (4.18), 340 (sh 3.86). EI-ms: *m/z* (rel. int.) 576 (2), 558 (24), 221 (16), 165 (29), 123 (37), 69 (100). FAB-ms *m/z* : 577 $[M+H]^+$. The determination of the molecular formula by high-resolution ms was failed, because the molecular ion of **5** was unstable in the EI condition. CD ($c = 4.6 \times 10^{-5}$ g/m*l*, MeOH): $[\theta]_{231}$ +6800, $[\theta]_{251}$ +2500 (sh), $[\theta]_{280}$ 0, $[\theta]_{296}$ −6500, $[\theta]_{312}$ 0, $[\theta]_{338}$ +2700.

Mulberrofuran V (6)

Compound (**6**) was obtained as a colorless amorphous powder. $FeCl_3$ test on a tlc plate: positive (pale brown). Uv λ_{max}^{MeOH} nm (log ε): 205 (4.40), 250 (sh 3.74), 394 (3.76). EI-ms: *m/z* (rel. int.) 379 $[M+H]^+$ (32), 378 $[M]^+$ (100), 363 (12), 361 (10), 335 (59), 323 (22), 309 (48), 293 (18), 279 (25), 267 (13), 69 (33). HR-ms: *m/z* 378.1815 (M^+, $C_{24}H_{26}O_4$ requires 378.1831).

2',4',7-Trihydroxy-(2S)-flavanone (20)[14]

Compound (**20**) was obtained as a colorless amorphous powder. $[\alpha]_D^{22}$ +50° (*c* 0.04, MeOH). Uv λ_{max}^{MeOH} nm (log ε): 207 (4.49), 215 (infl. 4.39), 230 (sh 4.21), 277 (4.10), 311 (3.90). FAB-ms: *m/z* 273 $[M+H]^+$. EI-ms: *m/z* 254 $[M-18]^+$, 137. CD ($c = 4.26 \times 10^{-5}$ g/m*l*, MeOH): $[\theta]_{233}$ +1370, $[\theta]_{250}$ +190, $[\theta]_{260}$ +450, $[\theta]_{270}$ +320 (valley), $[\theta]_{302}$ +2160, $[\theta]_{340}$ +640 (sh).

REFERENCES

1. Part 28 in the series 'Constituents of the Moraceae Plants'. Part 27 in the series: Y. Hano, J. Yamanaka, Y. Momose, and T. Nomura, *Heterocycles*, 1995, **41**, No. 12, in press.
2. T. Nomura, in 'Progress in the Chemistry of Organic Natural Products', eds. by W. Herz, H. Grisebach, C. W Kirby, and C.-H. Tamm, Springer, Vienna, New York, 1888, **53**, 87.
3. T. Nomura and Y. Hano, *Nat. Prod. Reports*, 1994, **11**, 205 and references cited therein.
4. Y. Yoshizawa, N. Suganuma, H. Fujiki, T. Fukai, T. Nomura, and T. Sugimura, *Phytotherapy Res.*, 1989, **3**, 193.
5. G. R. Reddy, N. Ueda, T. Hada, A. C. Sackeyfio, S. Yamamoto, Y. Hano, M. Aida, and T. Nomura, *Biochem. Pharmacal.*, 1991, **41**. 115.
6. T. Yanagisawa, T. Sato, M. Chin (Z.-X. Chin), H. Mitsuhashi, T. Fukai, Y. Hano, and T. Nomura, in 'Flavonoids in Biology and Medicine III', ed. by N. P. Das, National University of Singapore, Singapore, 1990, 107.
7. T. Nomura, T. Fukai, and Y. Hano, *Planta Med.*, 1983, **47**, 30.
8. T. Nomura, T. Fukai, Y. Hano, and J. Uzawa, *Heterocycles*, 1981, **16**, 2141.

9. Y. Hano, M. Itoh, N. Koyama, and T. Nomura, *Heterocycles*, 1984, **22**, 1791.
10. Y. Hano and T. Nomura, *Heterocycles*, 1985, **23**, 2499.
11. M. Takasugi, S. Nagao, S. Ueno, T. Masamune, A. Shirata, and K. Takahashi, *Chem. Lett.*, **1978**, 1239.
12. A. Shirata, K. Takahashi, M. Takasugi, S. Nagao, S. Ishikawa, S. Ueno, L. Muñoz, and T. Masamune, *Bull. Sericul. Exp. Sta.*, 1983, **28**, 793.
13. M. Kobayashi, H. Noguchi, and U. Sankawa, *Chem. Pharm. Bull.*, 1985, **33**, 3811.
14. G. Nabaei-Bidhendi and N. R. Bannerjee, *J. Indian Chem. Soc.*, 1990, **67**, 43.
15. T. J. Mabry, K. R. Markham, and M. B. Thomas, 'The systematic Identification of Flavonoids', Springer, New York, 1970.
16. K. R. Markham, V. M. Chari, and T. J. Mabry, in 'The Flavonoids: Advances in Research', eds. by J. B. Harborne and T. J. Mabry, Chapman and Hall, London, 1982, pp. 19–134.
17. a) T. Fukai and T. Nomura, *Heterocycles*, 1989, **29**, 2379; b) T. Fukai and T. Nomura, *Heterocycles*, 1996, **42**, No. 2, in press.
18. W. Gafflield, *Tetrahedron*, 1970, **26**, 4093.
19. T. Fukai and T. Nomura, *Heterocycles*, 1992, **34**, 1213.
20. T. Fukai and T. Nomura, *Heterocycles*, 1993, **36**, 329.
21. P. A. Agrawal, R. S. Thakur, and M. C. Bansal, in 'Carbon-13 NMR of Flavonoids', ed. by P. K. Agrawal, Elsevier, Amsterdam, 1989, Chapter 3.
22. Y. Hano, H. Kohno, M. Itoh, and T. Nomura, *Chem. Pharm. Bull.*, 1985, **33**, 5294.
23. T. Fukai and T. Nomura, *Phytochemistry*, 1995, **38**, 759.
24. T. Fukai, T.-H. Wang, J. Nishizawa, S,-R. Xing, and T. Nomura, *Nat. Med.*, 1994, **48**, 203.

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