ON THE STRUCTURE OF SANGGENON Q, A NEW DIELS-ALDER TYPE ADDUCT FROM MORUS MONGOLICA SCHNEIDER[1]

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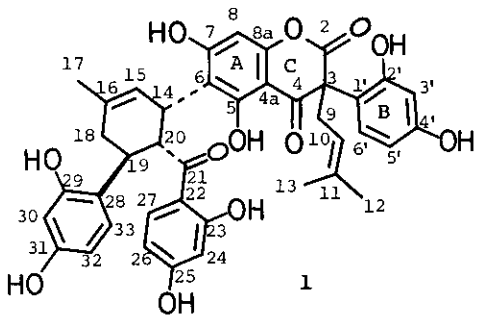
Abstract —— A novel Diels-Alder type adduct, sanggenon Q (1), was isolated from Morus mongolica Schneider (Moraceae) along with fourteen kinds of known phenolic compounds. Structure of sanggenon Q was shown to be 1 on the basis of spectral evidence. Sanggenon Q (1) is regarded biogenetically as a variation of a Diels-Alder type adduct, such as sanggenon C (2).

Previously we reported the structure determination of a series of phenolic compounds isolated from the root bark of the cultivated mulberry tree and from the Chinese crude drug "Sang-Bai-Pi" (Japanese name "Sōhakuhi").[2,3] Some of these phenolic compounds showed interesting biological activities, such as hypotensive effect, antirhinoviral activity, antitumor promoting activity, and so on.[2-4] Further survey for phenolic compounds of Moraceae plants led us to examine the constituents of Morus mongolica Schneider (Moraceae). In this paper, we report the isolation of phenolic compounds and the structure determination of a new compound named sanggenon Q.

From an ethanol extract of the root bark of Morus mongolica Schneider, sanggenon Q (1) was isolated as well as fourteen known phenolic compounds, sanggenon A (3),[2,3] morusin (4),[2,3] kuwanon E (5),[2,3] sanggenon M (6),[5] umbelliferone (7),[2,3] isoliquiritigenin (8),[6] scopoletin (9),[2,3] kuwanon G (10),[2,3] kuwanon J (11),[2,3] mulberrofuran Q (12),[7] sanggenon C (2),[8] sanggenon O (13),[9] oxyresveratrol (14),[2] and albanol B (15).[3]

Sanggenon Q (1), amorphous powder, $[\alpha]_D^{22}$ +111°, gave the FAB-ms spectrum which showed the ion peak at m/z 709 (M^++1), and the ^{13}C nmr spectrum indicated the presence of forty carbons (Table 1). These results suggest that the composition of



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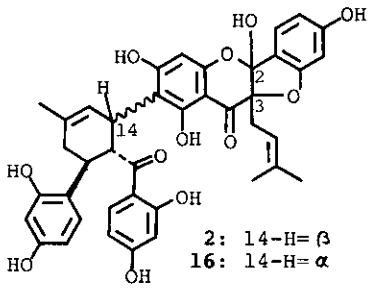

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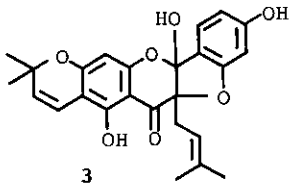
16: 14-H= α



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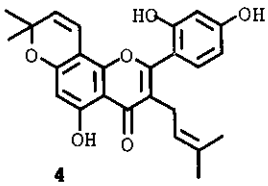

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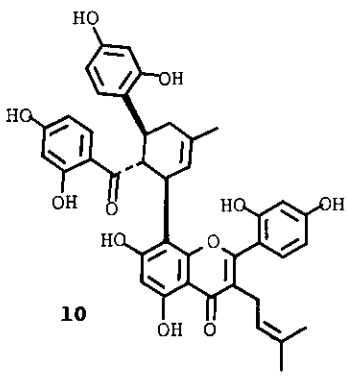

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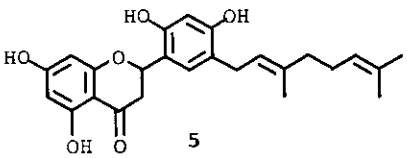

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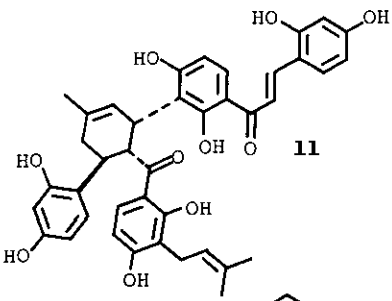

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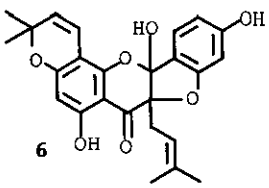

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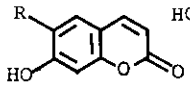
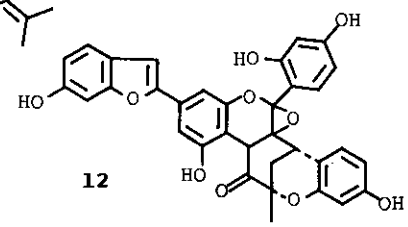


7: R=H
9: R=OCH$_3$



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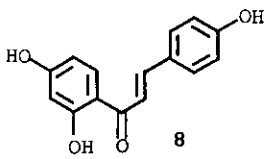

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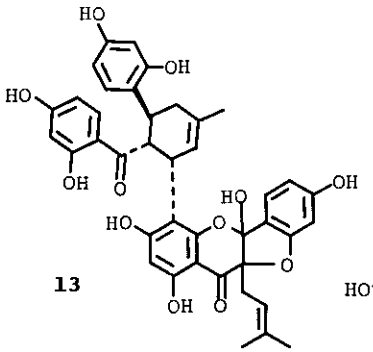

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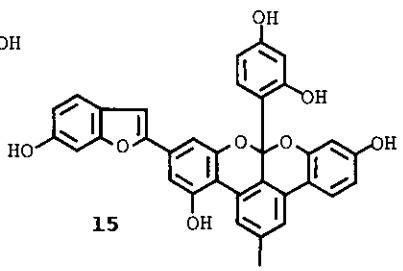

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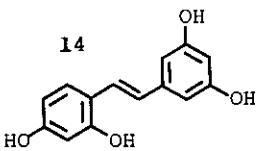

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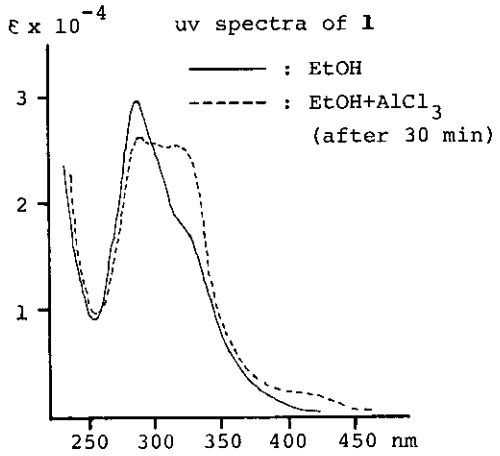
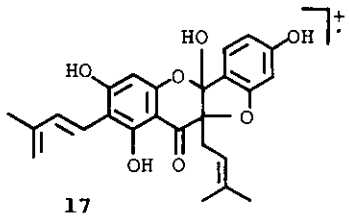
Fig. 1





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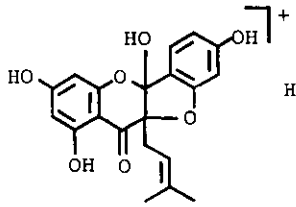

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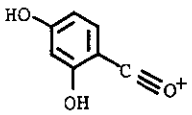

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Fig. 2

Table 1 ^{13}C Nmr chemical shifts (ppm) in acetone-d_6

	1	2		1	2
C-2	170.5	102.4	C-14	32.8	32.8
C-3	59.7	92.0	C-15	122.6	122.8
C-4	199.9	188.4	C-16	135.8	135.0
C-4a	101.9	99.9	C-17	23.8	23.7
C-5	162.2	163.9	C-18	33.4	33.8
C-6	111.4	109.0	C-19	35.6	35.8
C-7	167.0	167.6	C-20	47.7	48.3
C-8	97.0	96.5	C-21	208.9	208.8
C-8a	159.2	162.0	C-22	114.0	114.0
C-1'	122.0	122.2	C-23	165.8	165.9
C-2'	155.1	161.2	C-24	103.6	103.8
C-3'	103.8	99.5	C-25	166.1	166.8
C-4'	155.7	161.2	C-26	107.2	107.6
C-5'	108.8	109.7	C-27	134.9	133.7
C-6'	129.3	125.6	C-28	119.9	121.3
C-9	37.2	32.0	C-29	156.4	156.5
C-10	116.6	118.6	C-30	103.2	103.5
C-11	138.8	136.2	C-31	157.8	157.8
C-12	25.5	25.9	C-32	107.6	108.7
C-13	17.7	18.1	C-33	128.9	129.0

sanggenon Q (1) is $C_{40}H_{36}O_{12}$ and that 1 is a structural isomer of sanggenons C (2) and D (16)[10]. The compound (1) gave an intense green color with methanolic ferric chloride, while exhibited negative to the magnesium-hydrochloric acid test. The ir spectrum disclosed absorption bands for hydroxyl, carbonyl, and conjugated carbonyl groups at 3450, 1750, and 1625 cm^{-1}, respectively. The ^1H nmr spectrum showed the signals of the chelated hydroxyl groups at δ 12.50 and 12.53 ppm. The uv spectrum exhibited maxima at 230 (infl.), 286, 300 (infl.), and 320 (sh) nm,

and was similar to those of sanggenons C (2)[8] and D (16)[10], while the compound (1) was negative to the magnesium-hydrochloric acid test. These results suggest that 1 is a derivative of 2 or 16. In the uv spectrum of 1 in the presence of aluminum chloride, the absorption was observed at 287 nm, and a part of the absorption at 286 nm showed a bathochromic shift (Fig. 2). If the ir and the ^{1}H nmr spectra of 1 are taken into account, the absorption at 286 nm can be ascribed to the two conjugated carbonyl groups which are hydrogen-bonded. Sherif reported that aluminum chloride-induced shift was not observed in the uv spectra when a prenyl group was located ortho to a chelated hydroxyl group.[11] These data led us to a presumption that a prenyl group is substituted for one of the two hydrogen-bonded hydroxyl groups in ortho position, and not for another hydroxyl group. The similar phenomena were observed in the case of 2[8] and 16[10]. The FAB-ms of 1 showed the following fragment ions: m/z 709 (M^++1), 641, 491, 436, 369, 339, 273, 203, 181, 137. In the case of the FAB-ms of 2, the fragment ions were observed at m/z 709 (M^++1), 641, 491, 436, 369, 339, 273, 203, 137.[12] The fragment ions at m/z 436, 369, and 137 were suggestive of the formulae 17, 18, and 19, respectively (Fig. 2). These results suggest that sanggenon Q (1) is a Diels-Alder type adduct such as sanggenon C (2). This suggestion was substantiated by comparing the ^{1}H nmr spectrum of 1 with that of 2. The chemical shifts (δ) and coupling constants (Hz) of protons of the relevant methylcyclohexene ring are shown in Fig. 3, as well as of the 2,4-dihydroxybenzoyl and 2,4-dihydroxyphenyl groups locating on the ring. The remaining protons of 1 are summarized as follows:

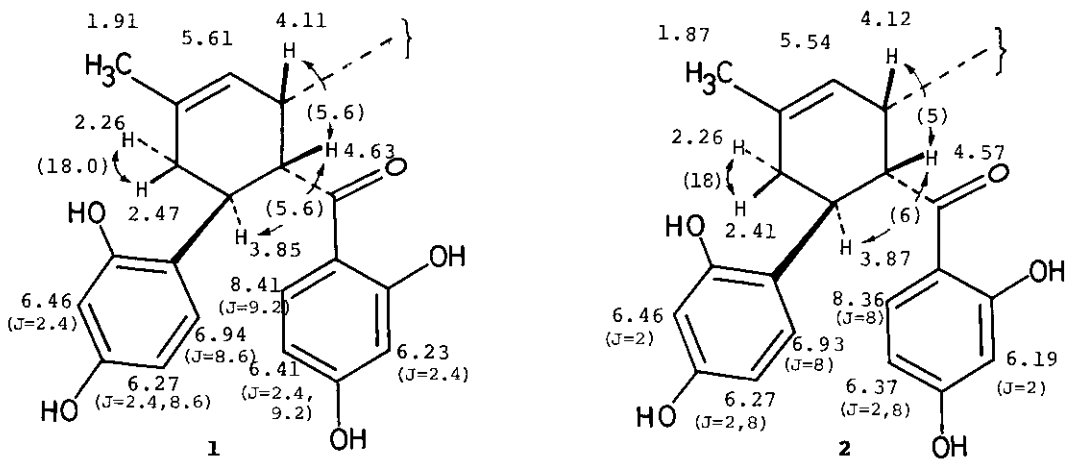


Fig. 3 ^{1}H Nmr chemical shifts and coupling constants in the methylcyclohexene ring and two aromatic rings for **1** and **2** (acetone-d_6).

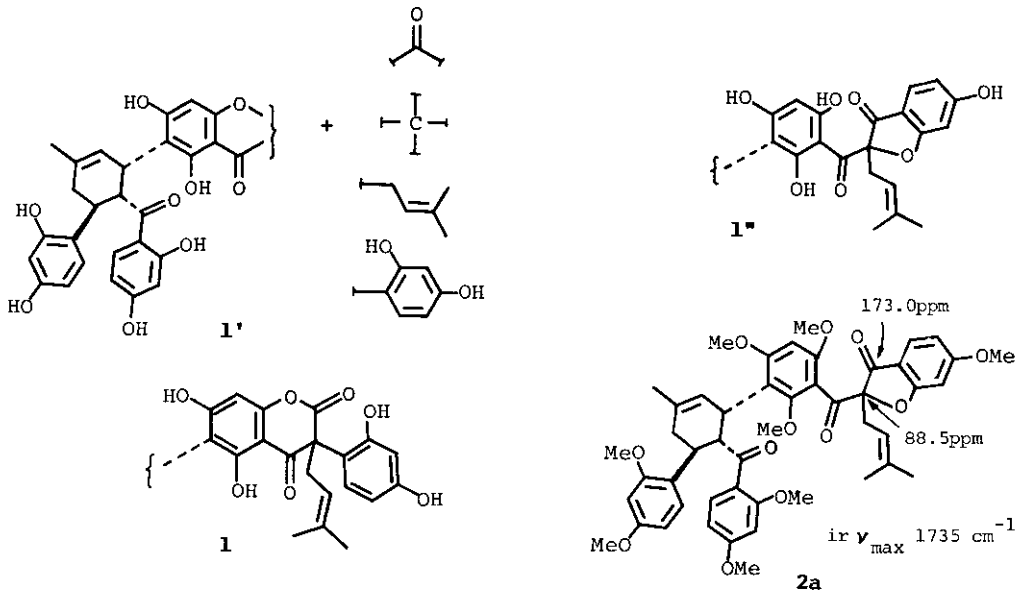
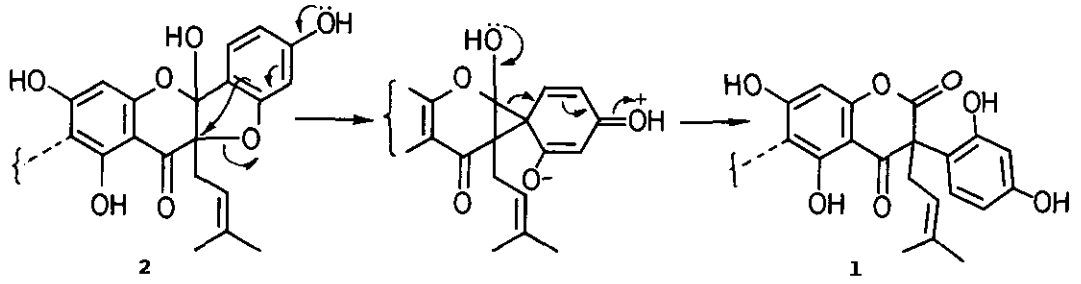


Fig. 4

protons in γ,γ-dimethylallyl moiety, δ 1.18, 1.26 (each 3H, s), 2.90 (1H, dd, J= 7.3 and 13.1 Hz), 3.04 (1H, dd, J=8.8 and 13.1 Hz), 4.90 (1H, m); 2,4-dioxygenated phenyl moiety, δ 6.33 (1H, d, J=2.4 Hz), 6.41 (1H, dd, J=2.4 and 8.6 Hz), 7.31 (1H, d, J=8.6 Hz); aromatic proton, δ 6.07 (1H, s). In the ^{13}C nmr spectrum of 1, the chemical shifts of the carbon atoms of the methylcyclohexene ring and of the 2,4-dihydroxybenzoyl and 2,4-dihydroxyphenyl groups locating on the ring were closely resembled to those of the relevant carbon atoms of 2 (Table 1). Furthermore, the chemical shifts of the carbon atoms of the A ring of 1 were similar to those of 2 except the signals of carbon atoms (C-6, 4a, and 8a) which were affected by the additional substituent effect (Table 1). From the above results, the partial structure 1' was suggested for sanggenon Q (Fig. 4). In the ^{13}C nmr spectrum of 1, the signals of three carbonyl carbons were observed at δ 170.5, 199.9 (C-4), and 208.9 (C-21). The presence of carbonyl carbon at C-2 position and γ,γ-dimethylallyl group at C-3 was supported by the following long-range selective ^{1}H decoupling (LSPD) technique. When the signal at δ 2.90 (C-9-H) was weakly irradiated, the signals at the C-4 (δ 199.9) and C-2 (δ 170.5) changed (dd ⟶ d), and the signal at δ 59.7 (C-3) also changed its shape. From the above results, two possible structures (1 and 1") were suggested. The chemical shift values of the carbon atoms at C-3 positions of sanggenon Q (1) and sanggenon C octamethyl ether (2a)[8], and the absorption bands of carbonyl carbons of the ir spectra of 1 and 2a support that the C-3 carbon atom was non-oxygenated tertiary carbon atom and the

carbon atom at the C-2 position was a lactone carbonyl carbon (Fig. 4). From the above results, we propose the formula 1 (except the absolute configuration) for the structure of sanggenon Q. Biogenetically, sanggenon Q seems to be a derivative induced from the Diels-Alder type adduct, such as sanggenon C (2) through the mechanism described in Fig. 5.[13]



2 1

Fig. 5

EXPERIMENTAL

Abbreviations: s=singlet, d=doublet, dd=double doublet, m=multiplet, br=broad, sh=shoulder, infl.=inflection. The general procedures followed are described in our previous papers. The following instruments were used: uv spectra; Shimadzu UV-265 spectrometer, ir spectra; Hitachi 260-30 IR spectrometer, ms; JEOL JMS-DX 303, ^{1}H and ^{13}C nmr spectra; JEOL JNM GX-400 FTNMR spectrometer.

Isolation of Sanggenon Q (1) and Other Phenolic Compounds from the Root Bark of Morus mongolica Schneider

The dried root bark (7 Kg) of Morus mongolica Schneider, collected in An-San, Liaoning, China, in September 1986, was extracted with ethanol at 80 °C for 4 h. Evaporation of the solution to dryness yielded 790 g of residue. This residue was extracted with ether. The ether solution was concentrated to afford the residue (100 g). This residue (100 g) was chromatographed on silica gel (250 g) with benzene-methanol as an eluent, each fraction being monitored by tlc. The fractions eluted with benzene containing 1% methanol was evaporated to leave a residue (10 g), which was rechromatographed on silica gel (200 g) with n-hexane-ether as an eluent. The fractions eluted with n-hexane containing 25% ether was evaporated to leave the residue (1.5 g). This residue (1.5 g) was fractionated by preparative tlc (solvent system, n-hexane:ether=1:1, n-hexane:acetone=2:1) to give sanggenon A (3, 14 mg), morusin (4, 15 mg), kuwanon E (5, 2 mg), sanggenon M (6, 4 mg). The fractions eluted with benzene containing 3% methanol were evaporated to leave a

residue (19.1 g), which was rechromatographed on silica gel (250 g) with benzene-ether as an eluent. The fractions eluted with benzene containing 30-50% ether were evaporated to leave the residue (3.8 g). The residue (3.8 g) was fractionated by preparative tlc (solvent system, chloroform:methanol=19:1, benzene:acetone=3:1, to give umbelliferone (7, 34 mg), isoliquiritigenin (8, 3 mg), scopoletin (9, 2 mg). A part of the fractions eluted with benzene containing 5% methanol were evaporated to give a residue (580 mg). This residue (580 mg) was fractionated by preparative tlc (solvent system, ether only, chloroform:methanol=4:1) and by HPLC analysis [solvent: 50% methanol-H_2O, flow rate: 0.6 ml/min, detector: UV 280 nm, column: Capcell pak C18 (4.6ø x 250 mm)] to give kuwanons G (10, 5 mg), J (11, 4 mg), mulberrofuran Q (12, 11 mg), sanggenons Q (1, 15 mg), C (2, 16 mg), O (13, 8 mg), oxyresveratrol (14, 30 mg). A part of the fractions containing 10% methanol was evaporated to give a residue (2.5 g), which was fractionated by preparative tlc (solvent system, benzene:acetone=8:5, chloroform:acetone=1:1, chloroform:methanol=6:1, ether only) to give sanggenons C (2, 40 mg), O (13, 29 mg), and albanol B (15, 16 mg). The identification of these known compounds was carried out by comparison with the spectral data of authentic samples.

Sanggenon Q (1)

Compound 1 was obtained as pale yellow amorphous powder. $[\alpha]_D^{22}$ +111° (c=0.158, methanol). $FeCl_3$ test: green. Mg-HCl test: negative. Ir ν_{max}^{KBr} cm^{-1}: 3450 (br), 1750, 1625, 1600, 1510, 1450. Uv λ_{max}^{EtOH} nm (log ε): 230 (infl. 4.58), 286 (4.47), 300 (infl. 4.40), 320 (sh 4.26). $\lambda_{max}^{EtOH+AlCl_3}$: 287 (4.43), 300 (4.42), 320 (4.41). ^1H Nmr (acetone-d_6): δ 1.18, 1.26 (each 3H, s, C-11-CH_3), 1.91 (3H, s, C-16-CH_3), 2.26 (1H, br d, J=18.0, C-18-H), 2.47 (1H, br d, J=18.0, C-18-H), 2.90 (1H, dd, J=7.3 and 13.1, C-9-H), 3.04 (1H, dd, J=8.8 and 13.1, C-9-H), 3.85 (1H, m, C-19-H), 4.11 (1H, br, C-14-H), 4.63 (1H, t, J=5.6, C-20-H), 4.90 (1H, m, C-10-H), 5.61 (1H, br s, C-15-H), 6.07 (1H, s, C-8-H), 6.23 (1H, d, J=2.4, C-24-H), 6.27 (1H, dd, J=2.4 and 8.6, C-32-H), 6.33 (1H, d, J=2.4, C-3'-H), 6.41 (1H, dd, J=2.4 and 9.2, C-26-H), 6.41 (1H, dd, J=2.4 and 8.6, C-5'-H), 6.46 (1H, d, J=2.4, C-30-H), 6.94 (1H, d, J=8.6, C-33-H), 7.31 (1H, d, J=8.6, C-6'-H), 8.41 (1H, d, J=9.2, C-27-H), 12.50, 12.53 (each 1H, s, C-5- and C-23-OH). FAB-ms m/z: 709 (M^++1), 641, 491, 436, 369, 339, 273, 203, 181, 137.

ACKNOWLEDGEMENT

We are grateful to Dr. J-Y. Lo, the Third Plant of Medicine, An-San, Liaoning,

China, for collection of the material and preparation of the ethanol extract of Morus mongolica Schneider.

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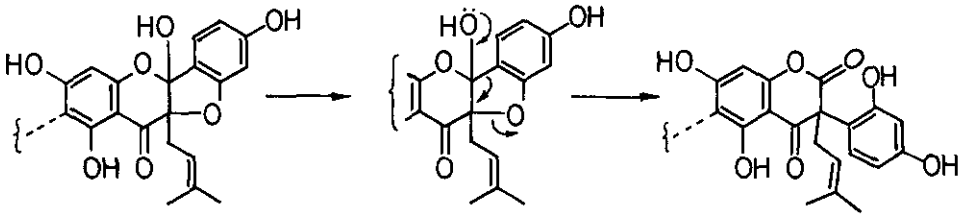
13. The following pinacol-pinacolone rearrangement like mechanism also seems to be possible. To confirm this point, the chemical correlation between **2** and **1** is now progress.



2 **1**

Received, 7th September, 1988