TOTAL SYNTHESIS OF (±)-GERICUDRANIN A

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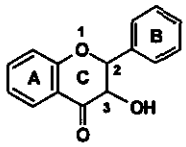
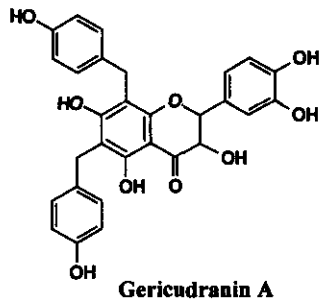
Abstract-An efficient total synthesis of (±)-gericudranin A is described. Trihydroxyacetophenone was converted to (±)-gericudranin A in seven steps *via* sequential protection, aldol condensation, epoxidation, cationic cyclization, regioselective *C*-benzylation and deprotection.

Cudrania tricuspidata Bureau, belongs to Moraceae, is a deciduous tree distributed over Korea, China and Japan. The cortex and root bark of the plant have been used as a traditional medicine for curing neuritis and inflammation in the Orient.[1] Recently, some *C*-benzylated flavonoids named gericudranin A, B, C, E, and J were isolated from the methanol extracts of mulberry tree (*Cudrania tricuspidata* in Moraceae).[2] Among these, gericudranin A has attracted considerable attention with its prominent cytotoxic and antioxidant activity. Although the absolute configuration of natural (+)-gericudranin A has not been determined, the relative stereochemistry at C2/C3 was proved *trans* by ^{1}H-nmr analysis. Its interesting biological activities, unique structure, and low yield production by extraction method prompted us to develop a synthetic method for gericudranin A that makes possible for the large scale production and further molecular modification.



Dihydroflavonol

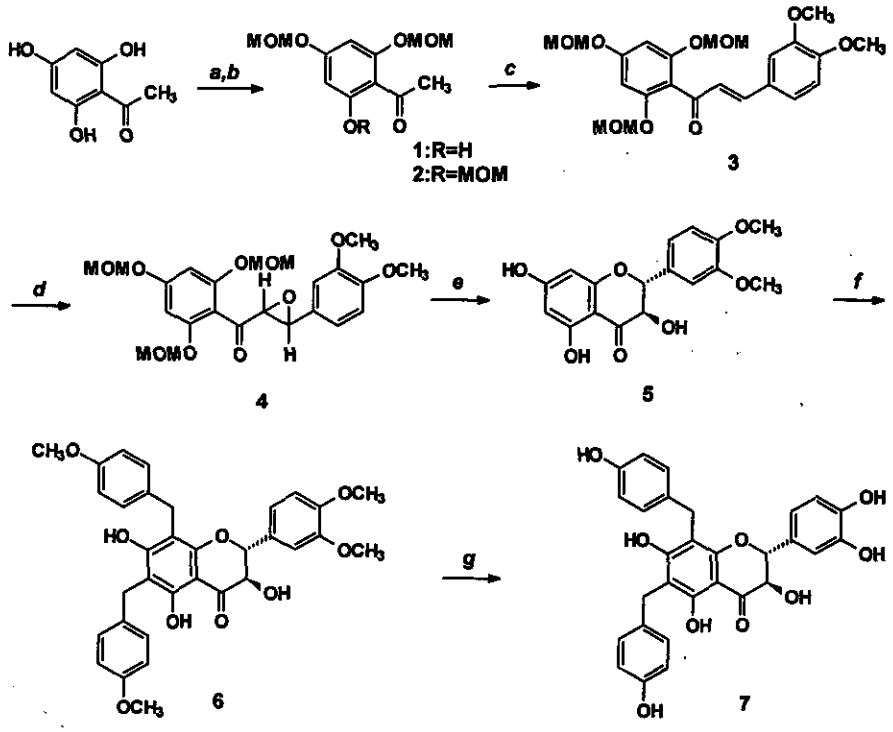


Gericudranin A

Although the numerous types of flavonoids have been reported to date,[3] there are few of *C*-benzylated flavonoids to be isolated.[4] Interestingly, these *C*-benzylated flavonoids share the cytotoxic activities in common. Furthermore,

to our best knowledge, only one report described the partial synthesis of *C*-benzylated flavonone compounds for the purpose of structure determination.[5] The regioselective introduction of benzylic unit into A ring over B ring and the regioselective alkylation at carbon versus oxygen represent the challenging problems for the synthesis of *C*-benzylated flavonoids. We initially attempted *C*-benzylation on the starting material such as phloroglucinol or 2',4',6'-trihydroxyacetophenone for the construction of dibenzylated A ring unit at the early stage of synthesis. However, those methods were found to be unsuccessful for the reasons that they are not suitable for further regioselective functionalization or benzylation, and they require multisteps synthesis. Thus, we decided to carry out the *C*-benzylation after the construction of dihydroflavonol skeleton. To make the A ring more reactive than B ring toward electrophilic aromatic substitution, we plan to protect phenolic OH group in B ring, but not in A ring. Our synthetic route is outlined in the following Scheme.

Scheme



Reagents and conditions:a) CH_3OCH_2Cl, *N,N*-diisopropylethylamine, THF, 0 °C, 73%; b) CH_3OCH_2Cl, NaH, THF, 0 °C, 83%; c) veratraldehyde, KOH, MeOH, room temperature, 90%; d) H_2O_2, 2N-NaOH, MeOH, room temperature, 97%; e) methanolic HCl, reflux, 55%; f) *p*-methoxybenzyl alcohol, $BF_3{\cdot}Et_2O$, 1,4-dioxane, 60 °C, 23%; g) BBr_3-DMS, 1,2-dichloroethane, reflux, 42%

Our synthetic strategy toward gericudranin A is based on the cationic cyclization initiated by acid-catalyzed epoxide ring opening to build up the fully functionalized dihydroflavonol nucleus (**5**), followed by regioselective Friedel-Crafts dibenzylation. Construction of dihydroflavonol ring skeleton begins with sequential protection of

2',4',6'-trihydroxyacetophenone to methoxymethyl ether (**2**) under two different conditions. Aldol condensation of **2** with veratraldehyde followed by dehydration under standard condition gave the chalcone (**3**) in 90% yield. The stereochemistry of **3** was proved *trans* by ^{1}H-nmr spectrum in which the coupling constant between the two vinylic protons appearing at 6.86 and 7.30 ppm is 16 Hz. Subsequent epoxidation of **3** with hydrogen peroxide in alkaline solution gave the epoxide (**4**) in 97% yield. The vicinal coupling constant of two oxirane ring protons appearing at 3.90 and 3.97 ppm as a doublet in its ^{1}H-nmr spectrum is 1.7 Hz. Thus, the relative configuration of the epoxide (**4**) is clearly *trans*.[6] Cationic cyclization, concurrent with deprotection, of the epoxide (**4**) to the dihydroflavonol (**5**) was effected by methanolic hydrochloric acid at reflux temperature in 55% yield. The characteristic ^{1}H-nmr spectrum of C ring in **5** exhibited C2-proton as a doublet (11.0 Hz) at 5.04 ppm, C3-proton as a double of doublet at 4.52 ppm (6.0 and 11.0 Hz) and hydroxyl proton at C3 as a doublet (6.0 Hz) at 5.63 ppm. The large vicinal coupling constant (11 Hz) between two protons at C2 and C3 verifies the *trans* configuration. Regioselective dibenzylation on **5** is now required to construct full carbon skeleton. After numerous experimentations, the yield was improved to an acceptable level by employing 4-hydroxybenzyl alcohol or 4-methoxybenzyl alcohol as an electrophile and $BF_3{\cdot}Et_2O$ as an acid catalyst. The more stabilized the benzylic cation derived from corresponding alcohol is, the lower reaction temperature is required for benzylation of **5**. The lower reaction temperature would guarantee the better regioselective benzylation on A ring over B ring. In this sense, 4-hydroxybenzyl alcohol is better than 4-methoxybenzyl alcohol as an electrophile. It really was, but 4-hydroxybenzylated product was not a suitable substrate for the final step because it is hardly soluble in solvents employed in demethylation process. Best yield was obtained by treatment of **5** with excess 4-methoxybenzyl alcohol and $BF_3{\cdot}Et_2O$ in 1,4-dioxane at 60°C to give the desired product (**6**) in 23% yield. Direct evidence for dibenzylation was provided by its ^{13}C-nmr spectrum in which newly formed two benzylic carbons appeared at 25.23 and 24.76 ppm. High resolution mass(HRms) spectrum also shows a peak at 572.2046 that is exactly consistent with calculated molecular mass. For the final removal of the four methyl protecting groups, we screened the various kinds of demethylating agent commonly used. Boron tribromide-dimethyl sulfide complex was the best choice for our purpose. Thus, synthetic (±)-gericudranin A was obtained by treatment of **6** with BBr_3-DMS complex in 1,2-dichloroethane at refluxing temperature in 42% yield. This synthetic compound proved to be identical with natural gericudranin A in all aspects (^{1}H-nmr, ^{13}C-nmr , ir and tlc analysis).[2a]

As described above, we achieved the first total synthesis of (±)-gericudranin A in seven steps starting from 2',4',6'-trihydroxyacetophenone.

EXPERIMENTAL

Melting points (mp) were determined in open capillaries with Büchi 535 melting point apparatus and are uncorrected. Infrared spectra were recorded on Shimadzu IR-435 instrument. ^{1}H-Nmr spectra were obtained on a Brucker WP-80 SY, Varian Gemini-300 BB, or JEOL JNM-GCX 400 spectrometer. Proton-decoupled ^{13}C-nmr spectra were recorded on a Varian Gemini-300 BB and are reported in part per million (δ) relative to TMS or $CDCl_3$ as an internal standard. EI mass spectra were run on VG Trio-2 GC-MS spectrometer at 70 eV. High

resolution mass and elemental analyses were performed by Korea Basic Science Center, Seoul. Tlc was performed on E. Merck silica gel 60 F_{254} plates(0.25 mm). Liquid column chromatography was performed using forced flow of indicated solvent on Merck Kieselgel 60 (230-400 mesh). Reversed phase column chromatography was performed by using Merck Lichroprep® RP-18 column.

2',4'-Bis(methoxymethoxy)-6'-hydroxyacetophenone (1)

To a stirred solution of 2',4',6'-trihydroxyacetophenone(140 mg, 0.75 mmol) and *N,N*-diisopropylethylamine(1.32 ml, 10.2 mmol) in 30 ml of tetrahydrofuran was added slowly chloromethylmethyl ether(0.23 ml, 2.85 mmol) at 0 °C. After 12 h, the reaction mixture was quenched with 2 ml of water, and diluted with ethyl acetate. The organic phase was washed with 5% aqueous hydrochloric acid ,water and brine, dried over anhydrous sodium sulfate, filtered and concentrated. The crude product was purified by column chromatography(silica gel, eluted with 15% ethyl acetate in hexane) to afford 192.7 mg (73%) of **1** as a white solid: Tlc, R_f 0.58 (1:2 EtOAc/hexane); mp 51 °C(recrystallized from hexane-ethyl acetate); ir(KBr) cm^{-1} 3100, 3000, 1610, 1270, 1210, 1150 ; ^{1}H-nmr(80 MHz, $CDCl_3$) δ 2.50(s, 3H), 3.31(s, 3H), 3.36(s, 3H), 5.01(s, 2H), 5.10(s, 2H), 6.10(s, 2H); EIms, m/z (relative intensity) 256(M^+, 75), 227(7), 211(8), 193(20), 182(100). *Anal.* Calcd for $C_{12}H_{16}O_6$: C, 56.23; H, 6.30. Found: C, 56.27; H, 6.32.

2', 4', 6'-Tris(methoxymethoxy)acetophenone (2)

To a NaH(80% in oil, 0.7 g, 23.3 mmol) suspension in 40 ml of THF was added 2',4'-bis(methoxymethoxy)-6'-hydroxyacetophenone(2.13 g, 8.3 mmol). This solution was stirred for 30 min, and then chloromethylmethyl ether(1.61 ml, 19.9 mmol) was added. After stirred for 30 min, the reaction was quenched by addition of 1 ml of water. The mixture was diluted with benzene, and then the organic phase was washed with 5% aqueous hydrochloric acid, water and brine, dried over sodium sulfate, filtered and concentrated. The crude product was purified by column chromatography(silica gel, eluted with 25% ethyl acetate in hexane) to afford 2.50 g (83%) of **2** as a white solid:Tlc, R_f 0.43(1:2 EtOAc/hexane); mp 40 °C (recrystallized from hexane-ethyl acetate); ir(KBr) cm^{-1} 3000, 1600; ^{1}H-nmr($CDCl_3$, 80 MHz) δ 2.46(s, 3H), 3.44(s, 9H), 5.11(s, 6H), 6.48(s, 2H); EIms, m/z (relative intensity) 300(M^+, 57), 238(36), 226(31), 182(100). *Anal.* Calcd for $C_{14}H_{20}O_7$: C, 55.99; H, 6.71. Found: C, 55.89; H, 6.81.

***trans*-3,4-Dimethoxy-2',4',6'-tris(methoxymethoxy)chalcone (3)**

To a mixture of 2',4',6'-tris(methoxymethoxy)acetophenone(1.96 g, 6.52 mmol) and potassium hydroxide(6.91 g, 120 mmol) in 40 ml of absolute methanol was added veratraldehyde(1.08 g, 6.52 mmol). After stirred for 15 h at room temperature, the resulting precipitate was filtered and rinsed with ethanol. The filtrate was concentrated and rediluted with benzene, and washed with 5% aqueous hydrochloric acid, water and brine, dried over sodium sulfate, filtered and concentrated. This residue was purified by column chromatography(silica gel, eluted with 33% ethyl acetate in hexane) and combined with previously obtained filter cake to afford 2.66 g(90%) of chalcone (**3**) as a pale yellow solid: Tlc, R_f 0.4(1:1 EtOAc/hexane); mp 109-110 °C (recrystallized from EtOH-H_2O); ir(KBr) cm^{-1} 3000, 2900, 1640, 1510, 1260, 1220, 1140, 1020, 920; ^{1}H-nmr($CDCl_3$, 400 MHz) δ 3.39(s, 6H), 3.51(s, 3H),

3.90(s, 3H), 3.91(s, 3H), 5.12(s, 4H), 5.19(s, 2H), 6.57(s, 2H), 6.86(d, J=16 Hz, 1H), 6.87(s, 1H), 7.08(m, 2H), 7.30(d, J=16 Hz, 1H); EIms, m/z (relative intensity) 448(M^+, 28), 403(49), 195(100), 177(28), 151(20); *Anal.* Calcd for $C_{23}H_{28}O_9$: C, 61.60; H, 6.29. Found: C, 61.43; H, 6.23.

(2,3)-*trans*-3-[3',4'-Dimethoxyphenyl]-2,3-epoxy-1-[2",4",6"-tris(methoxymethoxy)phenyl]propanone (4)

To a stirred solution of chalcone (**3**) (2.58 g, 5.75 mmol) in 100 ml of methanol at room temperature were added hydrogen peroxide(35% in water, 2.8 ml, 28.8 mmol) and 2N-NaOH(2.9 ml, 5.8 mmol) solution. After 3 h, the reaction was quenched by addition of saturated aqueous Na_2SO_3 solution, concentrated and diluted with benzene. The organic layer was washed with water and brine, dried over sodium sulfate, filtered and evaporated to give a 2.6 g (97%) of epoxide (**4**) as a white solid: Tlc, R_f 0.4 (1:1 EtOAc/hexane); mp 78-79 °C (recrystallized from ethyl acetate-hexane); ir(KBr) cm^{-1} 2950, 1720, 1690, 1600, 1510, 1260, 1230, 1150, 1040, 920; ^{1}H-nmr($CDCl_3$, 300 MHz) δ 3.39(s, 6H), 3.46(s, 3H), 3.85(s,3H), 3.87(s, 3H), 3.90(d, J=1.7 Hz, 1H), 3.96(d, J=1.7 Hz, 1H), 5.10(s, 4H), 5.14(s, 2H), 6.52(s, 2H), 6.7-6.9(m, 3H); EIms, m/z(relative intensity) 464(M^+, 31), 419(14), 285(18), 206(100), 181(40), 165(23), 151(35); *Anal.* Calcd for $C_{23}H_{28}O_{10}$: C, 59.48; H, 6.08. Found: C, 59.31; H, 5.95.

(2,3)-*trans*-3',4'-Dimethoxy-3,5,7-trihydroxyflavanone (5)

To a stirred solution of epoxide (**4**) (2.63 g, 5.66 mmol) in 50 ml of methanol was added 30 ml of methanolic HCl. After 12 h, the reaction was concentrated and diluted with ethyl acetate, washed with water and brine, dried over sodium sulfate, filtered and evaporated to give the crude product as a pale yellow solid. This crude solid was purified by column chromatography(silica gel, eluted with 40% ethyl acetate in hexane) to give 1.03 g (55%) of flavanone (**5**) as a white solid: Tlc, R_f 0.21(1:1 EtOAc/hexane); mp 201-202 °C (recrystallized from methanol-ethyl acetate); ir(KBr) cm^{-1} 3400, 1640, 1520, 1475, 1260, 1160, 1140, 1020; ^{1}H-nmr(DMSO-d_6 /$CDCl_3$, 400 MHz) δ 11.79(s, 1H), 10.49(s, 1H), 7.07(d, J=2.0 Hz, 1H), 7.06(dd, J=2.0, 8.0 Hz, 1H), 6.93(d, J=8.0 Hz, 1H), 5.94(d, J=2.0 Hz, 1H), 5.90(d, J=2.0 Hz, 1H), 5.63(d, J=6.0 Hz, 1H), 5.04(d, J=11.0 Hz, 1H), 4.52(dd, J= 6.0, 11.0 Hz, 1H), 3.88(s, 3H), 3.86(s, 3H); ^{13}C-nmr(DMSO-d_6 /$CDCl_3$, 75 MHz) δ 195.74, 166.09, 162.59, 161.48, 148.20, 147.62, 128.47, 119.46, 110.02, 109.92, 109.87, 99.36, 95.46, 94.35, 82.08, 71.02, 54.68; EIms, m/z(relative intensity) 332(M^+, 100), 303(70), 180(87), 165(59), 153(100). HRms(EI) exact mass calcd for $C_{17}H_{16}O_7$ (M^+) 332.0896, found 332.0860.

(2,3)-*trans*-6,8-Bis(*p*-methoxybenzyl)-3',4'-dimethoxy-3,5,7-trihydroxyflavanone (6)

To a stirred mixture of flavanone (**5**) (0.30 g, 0.90 mmol) and *p*-methoxybenzyl alcohol(0.45 ml, 3.24 mmol) in 20 ml of 1,4-dioxane at 60 °C was added $BF_3{\cdot}Et_2O$ (0.8 ml, 6.5 mmol) dropwisely. After stirred at 60 °C for 12 h, the reaction was quenched with 2 ml of water, and diluted with ethyl acetate. The organic layer was washed with water and brine, dried over sodium sulfate, filtered and evaporated to give the crude product as a hard syrup. This crude product was purified first by normal phase column chromatography(silica gel, 25% → 50% ethyl acetate in hexane), and finally by reversed phase column chromatography(C18-ODS column, 30% → 50% methanol in water) to give 120 mg (23%) of dibenzylated product (**6**) as a white solid: Tlc, R_f 0.35 (1:1 EtOAc/hexane); mp 171-172 °C (recrystallized from ethanol-water); ir(KBr) cm^{-1} 3450, 3000, 1615, 1505, 1240, 1105; ^{1}H-nmr

(DMSO-d_6 /$CDCl_3$, 400 MHz) δ 12.21(s, 1H), 8.21(s, 1H), 6.92-7.12(m, 7H), 6.72-6.77(m, 4H), 5.77(d, J=6.0 Hz, 1H), 5.07(d, J= 11.0 Hz, 1H), 4.54(dd, J= 11.0, 6.0 Hz, 1H), 3.83-3.59(m, 4H), 3.78(s, 3H), 3.74(s, 3H), 3.70(s, 6H); ^{13}C-nmr(DMSO-d_6 /$CDCl_3$, 75 MHz) δ 196.25, 160.60, 157.35, 156.02, 155.62, 147.32, 146.83, 131.10, 131.05, 128.27, 127.29, 118.67, 111.61, 111.57, 109.47, 109.35, 106.71, 105.74, 98.76, 88.11, 70.30, 53.87, 53.80, 53.76, 53.16, 25.23, 24.76; EIms, m/z(relative intensity) 572(M^+, 23), 393(14), 178(77), 151(100), 121(20), 108(16); HRms(EI) exact mass calcd for $C_{33}H_{32}O_9$ (M^+) 572.2046, found 572.2046.

(±)-Gericudranin A (7)

To a stirred solution of boron tribromide-dimethyl sulfide complex(2.40 g, 7.75 mmol) in 50 ml of 1,2-dichloroethane was slowly added the 20 ml of 1,2-dichloromethane solution of flavanone (**6**) (180 mg, 0.32 mmol). After stirred at reflux for 12 h, the reaction mixture was concentrated, rediluted with ether, and extracted with aqueous 1N- NaOH (10 ml x 4) solution. The combined water extracts were acidified with 5% aqueous HCl, and then extracted with ether. The ether layer was washed with water and brine, dried over sodium sulfate, filtered, evaporated, and the residue was purified by column chromatography, first on silica gel (10:1 dichloromethane/ methanol) and lastly on C18-ODS column (1:1 acetonitrile/water), to give 71 mg (42%) of (±)-gericudranin A (7) as a pale yellow solid. This synthetic material proved to be identical with natural gericudranin A in all aspects (^{1}H-nmr, ^{13}C-nmr , ir and tlc analysis).

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