Chem. Pharm. Bull. 28(4)1300—1303(1980)

Studies on the Constituents of Aceraceae Plants. III.[1] Strucure of Acerogenin B from *Acer nikoense* Maxim.

Masayoshi Kubo, Takao Inoue, and Masahiro Nagai

Hoshi College of Pharmacy[2]

(Received November 19, 1979)

Acerogenin B (3), $C_{19}H_{22}O_3$, mp 179°, $[\alpha]_D^{23}$ ±0°, a new diarylheptanoid of diphenyl ether type, was isolated from an acid hydrolysate of the glycoside mixture extracted from the bark of *Acer nikoense* Maxim. The structure of acerogenin B (3) was established as formula **3** in Chart 1 on the basis of the spectral data and chemical correlation with acerogenin A (**2**). Acerogenin B (3) was considered to be a racemic compound in view of the optical inactivity of **3** and its derivatives (**4** and **10**).

Keywords——*Acer nikoense*; Aceraceae; acerogenin B; diarylheptanoid; diphenyl ether; ^{1}H- and ^{13}C-NMR

In the previous paper we reported the structure elucidation of aceroside I (**1**),[1] a glucoside of a novel diarylheptanoid acerogenin A (**2**),[3] isolated from the bark of *Acer nikoense* Maxim. During the isolation of the genin (**2**) from an acid hydrolysate of a crude preparation of aceroside I (**1**), we observed the co-occurrence of another aglycone designated acerognin B (**3**).[4] Although no glycoside of **3** has been isolated yet, we obtained acerogenin B (**3**) in a sufficient amount to study its structure from the hydrolysis product of a crude glycoside mixture separated from the ethyl acetate-soluble fraction of the plant extract. The aglycone (**3**) is also detectable on a thin-layer chromatogram (TLC) of the ether-solubles of the plant extract.[4] This report presents the structure elucidation of acerogenin B (**3**), a new diarylheptanoid of diphenyl ether type.

Acerogenin B (**3**), $C_{19}H_{22}O_3$, mp 179°, $[\alpha]_D^{20}$ ±0°, obtained as colorless prisms, is slightly less polar on TLC than acerogenin A (**2**). In the infrared (IR) spectrum, **3** has hydroxy and aromatic ring absorptions, and in the ultraviolet (UV) spectrum, it shows a maximum absorption at 272 nm (log ε=3.35). It gave positive colorations with ferric chloride–potassium ferricyanide and diazo reagents. Acerogenin B (**3**) yielded a monomethyl ether (**4**) on methylation with diazomethane, and a diacetate (**5**) on acetylation. Oxidation of **4** afforded a monoketone (**6**), which showed no hydroxy absorption in its IR spectrum. These chemical and spectral findings suggest that acerogenin B (**3**) is a phenol derivative with a secondary alcoholic function.

In the carbon-13 nuclear magnetic resonance (^{13}C-NMR) spectrum of acerogenin B (**3**), seven doublets and five singlets were observed in the sp^2 carbon signal region, and six triplets and a doublet in the sp^3 carbon region. The chemical shift of the doublet sp^3 carbon at δ_C 70.6 ppm suggested that the carbon bears an oxygen function and that it is assignable to the carbon linked to the secondary alcoholic function. The chemical shifts in the proton nuclear magnetic resonance (^{1}H-NMR) spectrum of the monoketone (**6**) are summarized in Table I together with those of the corresponding monoketone (**7**) derived from acerogenin A (**2**). The ketone (**6**) showed an AA'BB' quartet centered at δ_H 7.11 ppm and an ABX pattern at δ_H 6.81, 6.65 and 5.46 ppm in the chemical shifts of aromatic protons, inicating that **6** has the same substituent pattern on the aromatic rings as acerogenin A (**2**). In particular, the fact that the signal

1) Part II: M. Nagai, M. Kubo, M. Fujita, T. Inoue, and M. Matsuo, *Chem. Pharm. Bull.*, **26**, 2805 (1978).
2) Location: *Ebara 2-4-41, Shinagawa-ku, Tokyo 142, Japan.*
3) M. Nagai, M. Kubo, M. Fujita, T. Inoue, and M. Matsuo, *Chem. Commun.*, **1976**, 338.
4) T. Inoue, Y. Ishidate, M. Fujita, M. Kubo, M. Fukushima, and M. Nagai, *Yakugaku Zasshi*, **98**, 41 (1978).

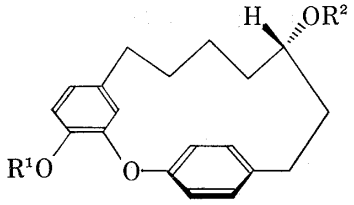
Table I. ^{1}H-NMR Spectral Data of Acerogenin B Methyl Ether Ketone (**6**), Acerogenin A Methyl Ether Ketone (**7**) and the Tetradeuterated Ketone (**6**-d_4) (δ ppm)

	6 ($CDCl_3$)	**6**-d_4 (CCl_4)	**7** ($CDCl_3$)
7-H_2	2.64—2.96(m)	2.76(s)	2.44(m)
8-H_2	2.29(m)	——	1.37(m)
9-H_2	——	——	1.09(m)
10-H_2	1.36—1.92(m)	——	1.90(m)
11-H_2		1.31—1.79(m)	——
12-H_2			2.61(m)
13-H_2	2.64—2.96(m)	2.74(m)	2.96(m)
OCH_3	3.94(s)	3.86(s)	3.93(s)
3-H	6.81 (d, J=8 Hz)	6.66 (d, J=8 Hz)	6.82 (d, J=8 Hz)
4-H	6.65 (d-d, J=2, 8 Hz)	6.49 (d-d, J=2, 8 Hz)	6.63 ((d-d, J=2, 8 Hz)
6-H	5.46 (d, J=2 Hz)	5.36 (d, J=2 Hz)	5.65 (d, J=2 Hz)
16,18-H	7.01 (q, J=9 Hz)	6.94 (q, J=9 Hz)	7.01 (q, J=9 Hz)
15,19-H	7.20	7.25	7.20

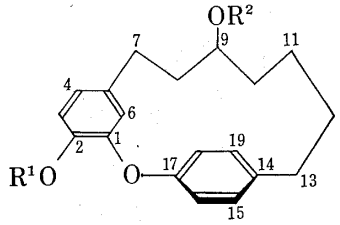
of 6-H of the ketone (**6**) was observed at abnormally high magnetic field (δ_H 5.46 ppm) indicated that the ketone (**6**) is a diarylheptanoid of diphenyl ether type, such as acerogenin A (**2**) and galeon,[5] because this proton is located above the plane of the other benzene ring and is thus subject to the shielding effect of the ring current.[6] The chemical shifts (δ_H 1—3 ppm) due to methylene protons of **6** are obviously different from those of **7**. This finding led us to assume that the ketone (**6**) is a diarylheptanoid isomeric with the ketone (**7**) only in the position of the carbonyl group on the seven-carbon chain.

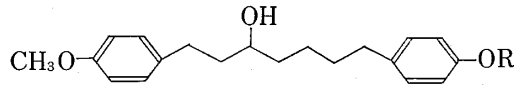
In order to determine the site of the carbonyl group, nuclear magnetic double resonance (NMDR) experiments on the ketone (**6**) were carried out. Irradiation of a six-proton multiplet at δ_H 1.36—1.92 ppm changed the upfield part of a four-proton multiplet at δ_H 2.64—2.96 ppm into a singlet without affecting a two-proton multiplet of δ_H 2.29 ppm, while the downfield part of the four-proton multiplet became a singlet on irradiation at the multiplet at δ_H 2.29 ppm. This result showed that the methylene at δ_H 2.29 ppm and one of two methylenes at δ_H 2.64—2.96 ppm are coupled with each other, but not with any other methylene protons, suggesting that the seven carbon chain is separated by the carbonyl group into two and four methylene groups. This suggestion was confirmed by ^{1}H-NMR experiments on the tetradeuterated ketone (**6**-d_4) prepared by a method similar to that used for the tetradeuterated ketone (**7**-d_4) of acerogenin A (**2**). The chemical shifts of the tetradeuterated ketone (**6**-d_4) are also given in Table I. In the spectrum of **6**-d_4 a multiplet of three methylenes of **6** at δ_H 1.36—1.92 ppm changed into a multiplet of two methylenes at δ_H 1.31—1.79 ppm together with disappearance of the multiplet of **6** at δ_H 2.29 ppm, while a multiplet of two methylenes of **6** at δ_H 2.64—2.96 ppm changed into a two-proton singlet at δ_H 2.76 ppm and a two-proton multiplet centered at δ_H 2.74 ppm. This result showed that the multiplet at δ_H 2.29 ppm and a part of the multiplet at δ_H 1.36—1.92 ppm of **6** are ascribable to two methylenes located alpha to the carbonyl group and that the singlet at δ_H 2.76 ppm of **6**-d_4 is assignable to the methylene (7-H_2) between an aryl group and a deuterated methylene. Since the ketone (**6**) derived from acerogenin B (**3**) is a different compound from the one (**7**) derived from acerogenin A (**2**), the structure of **6** must be proposed as formula **6** in Chart 1, and thus acerogenin B (**3**) corresponds to formula **3**.

5) K.E. Malterud, T. Anthonsen, and J. Hjortås, *Tetrahedron Lett.*, **1976**, 3069.
6) D.G. Farnum and C.F. Wilcox, *J. Am. Chem. Soc.*, **89**, 5379 (1967).



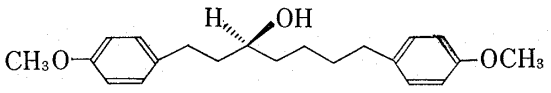
1 : $R^1=\beta$-D-glucopyranosyl, $R^2=H$
2 : $R^1=R^2=H$
7 : $R^1=CH_3$, 11-oxo





9 : R=H
10 : $R=CH_3$

3 : $R^1=R^2=H$, 9 (racemic)
4 : $R^1=CH_3$, $R^2=H$
5 : $R^1=R^2=COCH_3$
6 : $R^1=CH_3$, 9-oxo
8 : $R^1=CH_3$, R^2=tetrahydropyranyl



11

Chart 1

Furthermore, chemical evidence for the above proposal was obtained as follows. The tetrahydropyranyl ether (**8**) of **3** was cleaved with lithium in liquid ammonia, and after removal of the tetrahydropyranyl group by acid treatment, the resulting monomethyl ether (**9**) was methylated to give a dimethyl ether (**10**). Compound **10**, mp 38°, $[\alpha]_D$ ±0°, was identical with (−)-di-O-methylcentrolobol (**11**),[1] mp 58°, $[\alpha]_D$ −7.2°, derived from acerogenin A (**2**), in TLC, gas-liquid chromatography (GLC), mass spectrometry (MS), IR and ^{1}H-NMR, but not in melting point or optical rotation. All the derivatives of acerogenin B (**3**) described above are optically inactive.

On the basis of these spectral and chemical results, acerogenin B (**3**) is a racemic compound, and its structure was established as formula **3** in Chart 1. Acerogenin B (**3**) is a new diarylheptanoid resembling acerogenin A, galeon and hydroxygaleon.[5]

Experimental

All melting points are uncorrected. IR spectra were recorded on Shimadzu IR-400 and Hitachi IR-215 spectrometers. UV spectra were determined with a Shimadzu UV-200 machine. MS was taken on a Hitachi RSM-4 mass spectrometer. NMR spectra were determined with a JEOL FX-100 machine using tetramethylsilane as an internal standard. TLC was performed on Kieselgel 60 F_{254} (Merck) and spots were located by UV detection at 254 nm and by syraying 10% H_2SO_4 followed by heating. GLC was run on a Shimadzu GC-4A chromatograph with a hydrogen flame ionization detector. Circular dichroism (CD) spectra were taken with a JASCO J-40A machine.

Isolation of Acerogenin B (**3**)——Using the method described in a previous paper,[4] the fraction containing mainly aceroside I (**1**) was obtained from the bark of *Acer nikoense* and then hydrolyzed with 5% HCl to give a mixture of acerogenin A (**2**) and B (**3**). The mixture was chromatographed on silica gel, and elution with $CHCl_3$–MeOH (99: 1) afforded acerogenin B (**3**) and subsequently acerogenin A (**2**). Crude acerogenin B (**3**) was purified by repeated chromatography with silica gel and recrystallization from MeOH to give **3** as colorless prisms, mp 179°, $[\alpha]_D^{20}$ ±0° (c=1.0, EtOH). *Anal.* Calcd for $C_{19}H_{22}O_3$: C, 76.48; H, 7.43. Found: C, 76.50; H, 7.31. MS *m/e*: 298 (M^+). IR ν_{max}^{KBr} cm^{-1}: 3325, 3120, 2930, 2860, 1598, 1519, 1505. UV λ_{max}^{EtOH} nm (log ε): 272 (3.349), 285 (inf.). ^{1}H-NMR (C_5D_5N) δ: 0.72—1.88 (8-H_2, 10-H_2, 11-H_2 and 12-H_2), 2.30—3.12

(7-H_2 and 13-H_2), 3.12—3.42 (9-H) (each m); 5.82 (6-H, d, J=2 Hz), 6.71 (4-H, d-d, J=2 and 8 Hz), 6.85 (3-H, d, J=8 Hz) (1,2,5-trisubstituted benzene); 7.04—7.20 (16,18-H and 15,19-H, complex multiplet, aromatic protons). ^{13}C-NMR(C_5D_5N) δ: 23.0, 28.8, 30.7, 35.2, 37.1, 39.4 (each t) (6×–CH_2–); 70.6 (d, –CH(OH)–); 116.0, 117.1, 122.6, 123.3, 123.5, 130.7, 131.8 (each d) (7×Ar<u>C</u>H); 133.2, 139.3 (each s) (2×Ar<u>C</u>–C); 145.0, 150.7, 156.2 (each s) (3×Ar<u>C</u>–O).

Methyl Ether (4)——Compound 3 (700 mg) was dissolved in a mixture of ether (15 ml) and MeOH (2 ml). After addition of excess diazomethane in ether, the solution was allowed to stand overnight at room temperature. The solvent was evaporated off and the residue was methylated again under the same conditions until methylation was complete. The final product was recrystallized to give **4** as colorless needles (660 mg), mp 118°, $[\alpha]_D^{23}$ ±0° (c=1.8, EtOH). *Anal.* Calcd for $C_{20}H_{24}O_3$: C, 76.82; H, 7.74. Found: C, 77.14; H, 7.84. MS m/e: 312 (M^+). IR ν_{max}^{KBr} cm^{-1}: 3340, 2925, 2850, 1600, 1580, 1510, 1500. ^{1}H-NMR ($CDCl_3$) δ: 0.60—1.32 (11-H_2 and 12-H_2), 1.32—1.92 (8-H_2 and 10-H_2), 2.40—2.91 (7-H_2 and 13-H_2), 2.96—3.28 (9-H) (each m); 3.95 (–OCH_3, s); 5.58 (6-H, d, J=2 Hz), 6.64 (4-H, d-d, J=2 and 8 Hz), 6.80 (3-H, d, J=8 Hz) (1,2,5-trisubstituted benzene); 6.86—7.12 (16,18-H and 15,19-H, complex multiplet, aromatic protons). CD (c=3.3×10^{-3}, MeOH), no Cotton effect.

Diacetate (5)——Ac_2O (0.5 ml) was added to a solution of **3** (32 mg) in pyridine (0.5 ml), and the solution was allowed to stand for one day at room temperature. The reaction mixture was worked up as usual and the crude acetate was recrystallized from MeOH to give **5** as colorless needles (37 mg), mp 119°. *Anal.* Calcd for $C_{23}H_{26}O_5$: C, 72.23; H, 6.85. Found: C, 72.12; H, 6.78. MS m/e: 382 (M^+). IR ν_{max}^{KBr} cm^{-1}: 2930, 2850, 1760, 1722, 1587, 1500. ^{1}H-NMR (CCl_4) δ: 1.05 (2×–CH_2–, m), 1.55 (2×–CH_2–, m), 1.89 (–$OCOCH_3$, s), 2.32 (–$OCOCH_3$, s), 2.60 (–CH_2–, m), 4.27 (AcO–C<u>H</u>, m), 5.54 (6-H, d, J=2 Hz), 6.52 (4-H, d-d, J=2 and 8 Hz), 6.77 (3-H, d, J=8 Hz), 6.90—7.32 (16,18-H and 15,19-H, complex multiplet, aromatic protons).

Methyl Ether Ketone (6)——Jones' reagent was added to a solution of **4** (100 mg) in acetone until an orange color persisted, then the mixture was left to stand for 10 min at room temperature. The reaction mixture was poured into water. Crude crystals were obtained by the salting-out technique. The precipitates collected were washed with water and recrystallized from MeOH to give **6** as colorless needles (84 mg), mp 118—120°, $[\alpha]_D^{22}$ ±0° (c=2.0, EtOH). *Anal.* Calcd for $C_{20}H_{22}O_3$: C, 77.39; H, 7.14. Found: C, 77.66; H, 7.25. MS m/e: 310 (M^+). IR ν_{max}^{KBr} cm^{-1}: 2940, 2900, 2850, 1702, 1600, 1580, 1510, 1498. The ^{1}H-NMR spectral data are given in Table I. CD (c=1.9×10^{-4}, EtOH), no Cotton effect.

8,10-Tetradeuterated Ketone (6-d_4)——A solution of sodium methoxide (prepared from Na (50 mg) and MeOD (0.5 ml)) (0.5 ml) was added to a solution of **6** (50 mg) in methanol-d_1 (5 ml) and the mixture was treated as described previously.[1] The crude product was recrystallized from methanol-d_1 to give **6**-d_4 as colorless needles (37 mg), mp 119—120°. MS m/e: 314 (M^+). The ^{1}H-NMR spectral data are given in Table I.

Reduction of 4 in Liquid Ammonia——Conversion of **4** (550 mg) to the pyranyl ether (**8**), reduction with lithium in liquid ammonia and removal of the pyranyl group with *p*-toluenesulfonic acid were performed as described previously.[1] The resulting product was purified by silica gel chromatography using C_6H_6–AcOEt (9:1) as an eluent to give **9** as an oil (68.3 mg). MS m/e: 314 (M^+).

Methylation of 9——An excess of dimethylsulfate was added to a solution of **9** (50 mg) in 10% NaOH. After stirring for 5 min, the reaction mixture was extracted with ether. The ether layer was washed with water, dried over Na_2SO_4 and evaporated to dryness. The residue was chromatographed on silica gel and elution with C_6H_6–AcOEt (20:1) afforded a crude product, which was recrystallized from ether–hexane (3:1) to give **10** as colorless needles (23 mg), mp 37—38°, MS m/e: 328 (M^+). $[\alpha]_D^{28}$ ±0° (c=1.6, EtOH). IR ν_{max}^{KBr} cm^{-1}: 3325, 3240, 2940, 2855, 1615, 1587, 1518. ^{1}H-NMR ($CDCl_3$) δ: 1.20—1.82 (4×–CH_2–, m), 2.56 (2×Ar–CH_2–, m), 3.58 (–CH(OH)–, m), 3.78 (2×Ar–O<u>CH</u>$_3$, s), 6.77—7.12 (2×AA'BB' systems of aromatic protons, m). TLC: solvent, C_6H_6–AcOEt (6:1); Rf, 0.41. GLC: column temp. 225°; carrier gas N_2 (1.3 kg/cm^2); t_R (min) 22.2.

Acknowledgement This work was supported in part by a Grant-in-aid for Scientific Research from the Ministry of Education, Science and Culture of Japan, which is gratefully acknowledged.