Platanionosides A−C, Megastigmane Diglycosides from the Leaves of *Alangium platanifolium*

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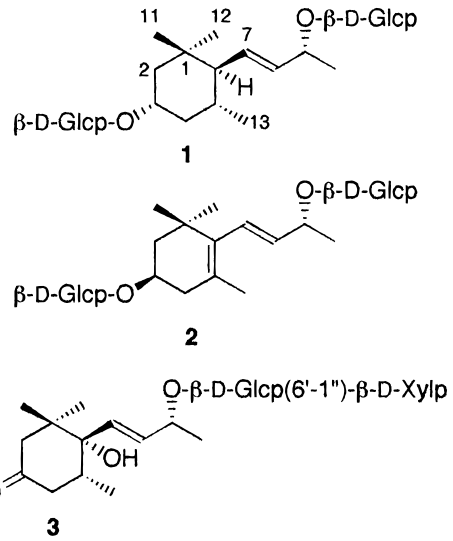
From the leaves of *Alangium platanifolium* var. *platanifolium* collected in Fukuoka Prefecture, Japan, three megastigmane diglycosides (**1**−**3**) were isolated, along with two known compounds, benzyl alcohol 7-*O*-*β*-D-(6′-*O*-*β*-D-xylopyranosyl)glucopyranoside and *Z*-hex-3-en-1-ol 1-*O*-*β*-D-glucopyranoside. The structures of the new compounds, named platanionosides A (**1**), B (**2**), and C (**3**) were elucidated by spectroscopic evidence to be 3*S*,5*R*,6*R*,9*R*,7*E*-megastigma-3,9-diol 3,9-di-*O*-*β*-D-glucopyranoside, 3*R*,9*R*,7*E*-megastigma-5,7-diene-3,9-diol 3,9-di-*O*-*β*-D-glucopyranoside, and 5*R*,6*S*,7*E*-megastigma-3-on-7-en-9-ol 9-*O*-*β*-D-(6′-*O*-*β*-D-xylopyranosyl)glucopyranoside, respectively.

Alangium lamarkii grown in India has been found to contain characteristic ipecac alkaloids in its roots.[1] Due to this observation, we become interested in Alangiaceous plants growing in Japan. Until now, no ipecac alkaloids have been isolated from leaves of Alangiaceous plants in Japan. However, many megastigmane glycosides were isolated from leaves of *Alangium premnifolium* growing in Okinawa Prefecture.[2−4] From the leaves and stem bark of *Alangium platanifolium* var. *trilobum*, collected in Hiroshima Prefecture, iridoid glucoside and phenolic glycosides have been reported.[5,6] In this paper, we report a phytochemical investigation on *A. platanifolium* (Sieb. et Zucc.) Harms var. *platanifolium* Sieb. et Zucc., a closely related species that is endemic plant to western areas of Japan, such as Yamaguchi and Fukuoka Prefectures. From the leaves, three new megastigmane diglycosides (**1**−**3**) along with two known compounds were isolated and identified.



Air-dried leaves of *A. platanifolium* var. *platanifolium* were extracted with MeOH and partitioned between solvents of increasing polarity, namely, *n*-hexane, EtOAc, and *n*-BuOH. The *n*-BuOH-soluble fraction was purified using highly porous synthetic resin, ODS, and silica gel column chromatographies and submitted to droplet counter-current chromatography (DCCC) to give five pure glycosides. Two of them were known glycosides and identified as benzyl alcohol 7-*O*-*β*-D-(6′-*O*-*β*-D-xylopyranosyl)glucopyranoside[2,7] and *Z*-hex-3-en-1-ol 1-*O*-*β*-D-glucopyranoside.[7]

Platanionoside A (**1**) was isolated as colorless crystals, mp 113−116 °C, and the molecular formula of $C_{25}H_{44}O_{12}$ was deduced from HRFABMS. The IR spectrum showed the presence of hydroxyl groups and a double bond (1650 cm^{-1}), while the UV spectrum showed no significant absorption maximum between 220 and 400 nm. The ^1H NMR spectrum showed the presence of two singlet methyls and two doublet methyls, two close doublet signals at δ_H 4.356 (J = 8 Hz) and 4.358 (J = 8 Hz) which were assigned as anomeric protons of sugar moieties, and two olefinic protons at δ_H 5.36 (dd, J = 10, 15 Hz) and 5.54 (dd, J = 7, 15 Hz). Judging from their coupling patterns, the olefinic protons had adjacent methine protons on both sides. The ^{13}C NMR spectrum of **1** indicated the presence of two terminal *β*-glucopyranose units, and the remaining 13 signals were expected to present a megastigmane skeleton. These signals were assigned to one double bond, four methines, two of which bearing an oxygen function (δ_C 75.4 and 78.2), two methylenes, four methyl groups, and a quaternary carbon. These functionalities and the relative arrangement of the ring protons, deduced from the ^1H NMR spectral data, were the same as those of alangionosides G and I, which have been isolated from a related plant, *A. premnifolium*.[4] It is known that on *β*-D-glucosylation to the secondary hydroxyl group, different upfield shifts are observed for adjacent carbon atoms depending on the chirality of the hydroxyl-bearing carbon.[8] Since the ^{13}C NMR spectral data for the ring carbons of **1** were superimposable on those of alangionoside G, and those of the side chain on those of alangionoside I, the structure of **1** was established as 3*S*,5*R*,6*R*,9*R*,7*E*-megastigma-3,9-diol 3,9-di-*O*-*β*-D-glucopyranoside.

Platanionoside B (**2**) was isolated as a white amorphous powder and its elemental composition was determined to be $C_{25}H_{44}O_{12}$. The IR spectrum showed that hydroxyl groups and double bonds (1642 cm^{-1}) were evident in the molecule and the UV absorption maximum at 231 nm indicated the presence of a conjugated diene system. The ^{13}C NMR spectrum showed the presence of two terminal

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β-glucopyranose moieties with the remaining 13 signals comprising di- and tetrasubstituted double bonds, and two secondary carbinols, four methylenes, and one quaternary carbon atom. These functionalities are the same as those of linarionoside C, isolated from *Linaria japonica*,[9] except for the presence of the double bond on the side chain. The absolute stereochemistries of the chiral centers were assigned as 3*R* and 9*R* by comparison of the ^{13}C NMR data with those of linarionoside C for the C-3 position and those of **1** for the C-9 position. Therefore, the structure of platanionoside B (**2**) was elucidated to be 3*R*,9*R*,7*E*-megastigma-5,7-dien-3,9-diol 3,9-di-*O*-β-D-glucopyranoside.

Platanionoside C (**3**) was isolated as a white amorphous powder and its elemental composition was determined to be $C_{24}H_{40}O_{12}$. The IR spectrum showed the presence of an aliphatic ketone (1692 cm^{-1}) and a double bond (1650 cm^{-1}). The ^{13}C NMR spectrum also showed the presence of a ketone function (δ_C 214.9) and a double bond. Together with other NMR spectral data, the planar structure of the aglycon of **3** was expected to be the same as that of ampelopsisionoside, isolated from *Ampelopsis brevipedunculata*[10] and the sugar moiety was found to comprise a terminal β-xylopyranose and a substituted β-glucopyranose. As the chemical shift of the C-6 position of glucopyranose was significantly shifted downfield in the ^{13}C NMR spectrum, the glycosidic linkage was determined to be at this position. The absolute stereochemistry of the chiral center on the side chain was similarly determined to be *R* as for compounds **1** and **2**. By the application of the octant rule, the absolute stereochemistry of the 5-position was confirmed to be *R*, since the CD spectrum of **3** showed a positive Cotton effect at 285 nm ([θ] +572). This value was qualitatively the same as that of ampelopsisionoside. The axial protons at the 2- and 4-positions resulted in downfield shifts from the 1,3-diaxial interaction of the hydroxyl group at the C-6 position. Therefore, the 6-position must have the *S*-configuration. Accordingly, the structure of platanionoside C (**3**) was elucidated to be the 6′-*O*-β-D-xylopyranoside of ampelopsisionoside, namely, 5*R*,6*S*,7*E*-megastigma-3-on-7-en-9-ol 9-*O*-β-D-(6′-*O*-β-D-xylopyranosyl)-glucopyranoside.

In summary, from the leaves of *A. platanifolium* var. *platanifoilium*, three new megastigmane diglycosides (**1**−**3**) were isolated. Although the megastigmane glycosides are a currently expanding group, those having two sugars on different hydroxyl groups of the aglycon are relatively rare.

Experimental Section

General Methods. Melting points were determined using a Yamagimoto micro melting point apparatus and are uncorrected. Optical rotations were measured with a Union Giken PM-101 digital polarimeter. The CD spectrum was recorded on a JASCO J-702 spectropolarimeter. FTIR and UV spectra were recorded on Shimadzu FTIR-4200 and UV-160A spectrophotometers, respectively. ^{1}H and ^{13}C NMR spectra were measured on a JEOL JNM α-400 spectrometer at 400 and 100 MHz, respectively, with tetramethylsilane as internal standard. HRFABMS (negative-ion mode) were obtained with a JEOL JMS-SX-102 mass spectrometer with PEG-400 as a matrix.

Reversed-phase gravity column chromatography was performed on Cosmosil 75C_{18}-OPN (Nakarai Tesque, Kyoto, Japan) (Φ = 40 mm, *L* = 25 cm) with a linear gradient solvent system [H_2O−MeOH (9:1, 1 L) → (1:1, 1 L); fractions of 10 g being collected]. DCCC (Tokyo Rikakikai, Tokyo, Japan) was equipped with 500 glass columns (Φ = 2 mm, *L* = 40 cm). The ascending method was used with $CHCl_3$−MeOH−H_2O−*n*-PrOH (9:12:8:2), and 5-g fractions were collected and numbered according to the order of elution of the mobile phase. Preparative HPLC was performed on Inertsil (GL Science, Tokyo, Japan) (ODS; a, Φ = 20 mm, *L* = 25 cm; and b, Φ = 6 mm, *L* = 25 cm), with the flow rate being 6 and 1.6 mL/min, respectively, and detection being performed by UV at 210 nm and/or the refractive index.

Plant Material. Leaves of *A. platanifolium* var. *platanifoilium* (Alangiaceae) were collected in September 1994 in Amagi City, Fukuoka Prefecture, Japan. A voucher specimen (94-APP-Fukuoka-0915) has been deposited in the Herbarium of the Institute of Pharmaceutical Sciences, Hiroshima University Faculty of Medicine.

Extraction and Isolation. The air-dried leaves (3.55 kg) were extracted with MeOH (12 L × 2). To the concentrated MeOH extract (1.5 L) was added 75 mL of H_2O, followed by washing with *n*-hexane (1.5 L). The MeOH layer was concentrated to a dark green mass and then suspended in 1.5 L of H_2O. The suspension was successively extracted with EtOAc (1.5 L) and *n*-BuOH (1.5 L) to give 65.9 g of a *n*-BuOH-soluble fraction. The fraction was subjected to passage over a highly porous synthetic resin (Diaion HP-20, Mitsubishi Kagaku, Tokyo, Japan) column with 20% (6 L), 40% (6 L), 60% (6 L), and 80% (6 L) MeOH in H_2O, and MeOH (6 L) used as eluents. Fractions of 2 L were collected. The residue (14.9 g) from the 40% MeOH eluate was subjected to silica gel (200 g) column chromatography with stepwise increases in the MeOH content in $CHCl_3$ [$CHCl_3$ (1 L), $CHCl_3$−MeOH (99:1, 2 L), (49:1, 2L), (24:1, 4 L), (93:7, 4 L), (9:1, 4 L), (17:3, 4 L), (4:1, 4 L), (3:1, 4 L), and (7:3, 4 L), with fractions of 500 mL being collected. The residue (1.83 g out of 3.75 g in fractions 38−44) of the 15% MeOH eluate was then subjected to reversed-phase column chromatography to give 143 mg of 7-*O*-β-D-(6′-*O*-β-D-xylopyranosyl)glucopyranoside in a crystalline state in fractions 63−81.

The residue (1.71 g out of 3.91 g in fractions 45−58) of the 20% MeOH eluate obtained by silica gel column chromatography was subjected to reversed-phase column chromatography and then the residue (307 mg in fractions 109−126) was purified by DCCC. The residue (125 mg in fractions 21−27) was subjected to repeated preparative HPLC [H_2O−MeOH (13:7 and then 7:3)] to give 32 mg of **1** and 29 mg of **2**. The residue (33 mg in fractions 39−45) was finally purified by preparative HPLC [H_2O−MeOH (13:1) to furnish 19 mg of **3**. *Z*-Hex-3-en-1-ol 1-*O*-β-D-glucopyranoside (61 mg) was isolated from fractions 46−54 of the DCCC separation.

Known Compounds Isolated. Benzyl alcohol 7-*O*-β-D-(6′-*O*-β-D-xylopyranosyl)glucopyranoside, mp 192−195 °C; $[\alpha]^{29}_D$ −63.9° (*c* 0.83, MeOH).[2] *Z*-Hex-3-en-1-ol 1-*O*-β-D-glucopyranoside, $[\alpha]^{29}_D$ −43.8° (*c* 0.73, MeOH).[7]

Platanionoside A (1): colorless crystals (MeOH): mp 113−116 °C; $[\alpha]^{25}_D$ −25.0° (*c* 0.72, MeOH); IR (KBr) ν_{max} 3420, 2950, 1650, 1462, 1370, 1080, 1044, 1022 cm^{-1}; ^{1}H NMR (CD_3OD) δ 0.83 (3H, d, *J* = 7 Hz, H_3-13), 0.87 (3H, s, H_3-11), 0.92 (3H, s, H_3-12), 1.03 (1H, q, *J* = 12 Hz, H-4ax), 1.17 (1H, t, *J* = 12 Hz, H-2ax), 1.28 (3H, d, *J* = 6 Hz, H_3-10), 1.35 (1H, t, *J* = 10 Hz, H-6), 1.56 (1H, m, H-5), 1.85 (1H, ddd, *J* = 2, 4, 12 Hz, H-2eq), 2.12 (1H, br d, *J* = 12 Hz, H-4eq), 3.12 and 3.17 (each 1H, dd, *J* = 8, 9 Hz, H-2′ and 2″), 3.20 (1H, ddd, *J* = 2, 5, 9 Hz, H-5′ or 5″), 3.30−3.45 (5H, m, H-3′,3″,4′,4″, and 5′ or 5″), 3.66 and 3.68 (each 1H, dd, *J* = 5, 12 Hz, H-6′a and 6″a), 3.79 and 3.86 (each 1H, dd, *J* = 2, 12 Hz, H-6′b and 6″b), 3.88 (1H, tt, *J* = 4, 12 Hz, H-3), 4.35 (1H, qd, *J* = 6, 7, H-9), 4.356 and 4.358 (each 1H, each d, *J* = 8 Hz, H-1′ and 1″), 5.36 (1H, dd, *J* = 10, 15 Hz, H-8), 5.54 (1H, dd, *J* = 7, 15 Hz, H-7); ^{13}C NMR (CD_3OD) see Table 1; HRFABMS (negative-ion mode) *m*/*z* 535.2727 [M − H]$^-$ (calcd for $C_{25}H_{43}O_{12}$, 535.2755).

Platanionoside B (2): white amorphous powder; $[\alpha]^{25}_D$ −42.9° (*c* 0.56, MeOH); IR (KBr) ν_{max} 3400, 2926, 1642, 1385, 1076, 1040 cm^{-1}; UV (MeOH) λ_{max} (log ε) 231 (3.71) nm; ^{1}H NMR (CD_3OD) δ 1.03 (3H, s, H_3-11), 1.07 (3H, s, H_3-12), 1.32 (3H, d, *J* = 6 Hz, H_3-10), 1.51 (1H, t, *J* = 12 Hz, H-2ax), 1.71 (3H, s, H_3-10), 1.90 (1H, ddd, *J* = 2, 4, 12 Hz, H-2eq), 2.06 (1H, br dd, *J* = 10, 17 Hz, H-4ax), 2.45 (1H, br dd, *J* = 6, 17 Hz, H-4eq), 3.16 (1H, dd, *J* = 8, 9 Hz, H-2′ or 2″), 3.20−3.45 (7H, m, H-2′ or 2″, 3′,3″,4′,4″, 5′, and 5″), 3.68 (2H, dd, *J* = 5,

Table 1. ^{13}C NMR Spectral Data for Platanionosides A (**1**), B (**2**), and C (**3**) (400 MHz, CD_3OD)

carbon	**1**	**2**	**3**
1	35.9	37.7	44.0
2	47.9	47.2	52.5
3	75.7	73.1	214.9
4	43.9	39.8	46.2
5	32.2	126.9	37.8
6	58.7	138.2	78.1
7	133.2	129.2	134.6
8	136.5	137.9	134.2
9	78.2	78.3	78.0
10	21.8	21.4	21.5
11	21.8	21.7	25.1
12	32.2	28.8	25.4
13	21.4	30.7	16.6
1′,1″	102.8, 102.3	102.5, 102.4	102.7
2′,2″	75.4, 75.2	75.4, 75.2	75.3
3′,3″	78.1, 78.1	78.2, 78.2	77.8
4′,4″	71.8, 71.4	71.7, 71.4	71.2
5′,5″	77.9, 77.9	77.9, 77.9	76.9
6′,6″	62.9, 62.6	62.8, 62.6	69.6
1′			105.5
2″			74.9
3″			77.7
4″			71.3
5″			67.0

12 Hz, H-6′a and 6″a), 3.76 and 3.86 (each 1H, dd, J = 2, 12 Hz, H-6′b and 6″b), 4.10 (1H, m, H-3), 4.38 and 4.43 (each 1H, d, J = 8 Hz, H-1′ and 1″), 4.41 (1H, m, H-9), 5.51 (1H, dd, J = 7, 16 Hz, H-8), 6.05 (1H, br d, J = 16 Hz, H-7); ^{13}C NMR (CD_3OD) see Table 1; HRFABMS (negative-ion mode) m/z 533.2577 $[M - H]^-$ (calcd for $C_{25}H_{41}O_{12}$, 533.2598).

Platanionoside C (3): white amorphous powder, $[\alpha]^{25}_D$ −26.0° (*c* 1.23, MeOH); CD (*c* 0.0664, MeOH) [θ] (nm) +572 (285); IR (KBr) ν_{max} 3400, 2970, 1692, 1650, 1042 cm^{-1}; ^1H NMR (CD_3OD) δ 0.90 (3H, d, J = 6 Hz, H_3-13), 0.93 (3H, s, H_3-12), 0.99 (3H, s, H_3-11), 1.32 (3H, d, J = 6 Hz, H_3-10), 1.82 (1H, dd, J = 2, 14 Hz, H-2eq), 2.12 (1H, ddd, J = 2, 4, 14, H-4eq), 2.28 (1H, dqd, J = 2, 4, 14 Hz, H-5), 2.45 (1H, t, J = 14 Hz, H-4ax), 2.83 (1H, d, J = 14 Hz, H-2ax), 3.18 (1H, dd, J = 10, 11, H-5″a), 3.20 (1H, dd, J = 8, 9 Hz, H-2′), 3.22 (1H, dd, J = 7, 9 Hz, H-2″), 3.25−3.40 (5H, m, H-3′,3″,4′, 5′, and 5″a), 3.47 (1H, ddd, J = 5, 9, 10 Hz, H-4″), 3.70 (1H, dd, J = 4, 12 Hz, H-6′a), 3.84 (1H, dd, J = 5, 11 Hz, H-5″b), 4.06 (1H, br d, J = 12 Hz, H-6′b), 4.28 (1H, d, J = 7 Hz, H-1″), 4.35 (1H, d, J = 8 Hz, H-1′), 4.44 (1H, dquintet, J = 1, 6 Hz, H-9), 5.74 (1H, dd, J = 1, 16 Hz, H-7), 5.89 (1H, dd, J = 6, 16 Hz, H-8); ^{13}C NMR (CD_3OD) see Table 1; HRFABMS (negative-ion mode) m/z 519.2439 $[M - H]^-$ (calcd for $C_{24}H_{39}O_{12}$, 519.2441).

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