Glycosides of Benzyl and Salicyl Alcohols from *Alangium chinense*

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From the water-soluble fraction of the dried leaves of *Alangium chinense*, three new glycosides, benzyl alcohol β-D-glucopyranosyl-(1→2)-[β-D-xylopyranosyl-(1→6)]-β-D-glucopyranoside, 2′-*O*-β-D-glucopyranosylsalicin, and 2′-*O*-β-D-glucopyranosyl-6′-*O*-β-D-xylopyranosylsalicin were isolated along with seven known glycosides. The structures of the new compounds were determined by spectroscopic and chemical means.

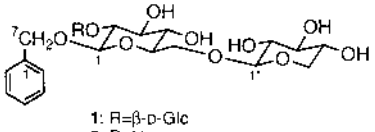
Key words *Alangium chinense;* Alangiaceae; leaf; salicyl glycoside; benzyl glycoside

In the course of our phytochemical studies on *Alangium* plants, we earlier reported new phenolic glycosides from *n*-BuOH soluble fraction of the leaves of *Alangium chinense* (LOUR.) HARMS.[1)] We have further investigated the water soluble fraction of this plant material and isolated three new glycosides **1**—**3** together with salicin (**4**), 6′-*O*-β-D-xylopyranosylsalicin (**5**), 4′,6′-*O*-(*S*)-hexahydroxydiphenoylsalicin, henryoside, 6′-*O*-β-glucopyranosylhenryoside,[2)] benzyl alcohol β-D-xylopyranosyl(1→6)-β-D-glucopyranoside (**6**) and *Z*-hex-3-en-1-ol β-D-xylopyranosyl(1→6)-β-D-glucopyranoside. The known compounds have already been isolated from the *n*-BuOH fraction of *A. chinense*.[1)] This paper deals with the isolation and structural elucidation of the three new compounds.

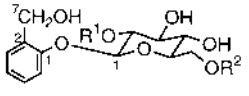
Compound **1**, $C_{24}H_{36}O_{15}$, was obtained as an amorphous powder. It showed UV maxima at 216, 258 and 264 nm and IR bands at 3384, 1637 and 1456 cm^{-1}. Its ^{1}H-NMR spectrum exhibited signals for monosubstituted benzene ring at δ 7.26 (1H, tt, *J*=7.0, 1.5 Hz), 7.33 (2H, br t, *J*=7.0 Hz), 7.45 (2H, br d, *J*=7.0 Hz), benzylic methylene proton signals at δ 4.71 (1H, d, *J*=12.0 Hz) and 4.94 (1H, d, *J*=12.0 Hz) and three anomeric proton signals at δ 4.34 (1H, d, *J*=7.5 Hz), 4.54 (1H, d, *J*=7.5 Hz), 4.62 (1H, d, *J*=8.0 Hz). These spectral data suggested that **1** consisted of benzyl alcohol and three sugar units. Acid hydrolysis of **1** afforded D-glucose and D-xylose which were identified by GLC analysis of the thiazolidine derivatives.[3)] The ^{13}C-NMR spectrum of **1** demonstrated the presence of two β-glucose units and a β-xylose moiety besides a benzyl alcohol moiety. The glycosidic linkages were confirmed by the cross-peaks between H_2-7 and C-1′, between H-1′ and C-7, between H-2′ and C-1″, between H-1″ and C-2′, between H_2-6′ and C-1‴, and between H-1‴ and C-6′ in the ^{1}H-detected heteronuclear multiple-bond connectivity (HMBC), and by the cross-peaks between H_2-7 and H-1′, between H-2′ and H-1″, and between H-6′ and H-1‴ in the nuclear Overhauser enhancement and exchange spectroscopy (NOESY) correlations. This was also supported by comparison of its ^{13}C-NMR spectrum with that of benzyl alcohol xylopyranosyl(1→6)-glucopyranoside (**6**).[4)] Accordingly, compound **1** was deduced to be benzyl alcohol β-D-glucopyranosyl-(1→2)-[β-D-xylopyranosyl-(1→6)]-β-D-glucopyranoside.

Compound **2**, $C_{19}H_{28}O_{12}$, was also obtained as an amorphous powder. It showed UV maxima at 213, 269 and 274 nm and IR absorption at 3373, 1605, 1593 and 1493 cm^{-1}. Acid hydrolysis of **2** furnished D-glucose.[3)] The ^{1}H-NMR data of **2** showed signals for four aromatic protons at δ 7.00 (td, *J*=7.5, 1.0 Hz), 7.19 (dd, *J*=8.0, 1.0 Hz), 7.25 (ddd, *J*=8.0, 7.5, 1.5 Hz) and 7.30 (dd, *J*=7.5, 1.5 Hz), benzylic methylene proton signals at δ 4.63 (d, *J*=13.0 Hz) and 4.78 (d, *J*=13.0 Hz) together with the signals arising from a β-glucosyl unit. These spectral feature demonstrated its close similarlity to salicin (**4**) which is the main component of this plant material, but with one more β-glucosyl unit. The attachment of the second glucosyl unit to C-2 of the first glucosyl unit was confirmed by the two-dimensional NMR experiments and the comparison of ^{13}C-NMR data of **2** with those of salicin (**4**).[1)] Thus, compound **2** was characterized as 2′-*O*-β-D-glucopyranosylsalicin.

The molecular formula of compound **3**, $C_{24}H_{36}O_{16}$, obtained from its high resolution secondary ion mass spectrum (HR-SI-MS) was $C_5H_8O_4$ more than that of **2**. Compound **3** liberated D-glucose and D-xylose on acid hydrolysis.[3)] Its spectral features were quite similar to those of **2** except for the signals due to an additional β-xylopyranosyl unit in its NMR spectra (Table 1). The linkage of β-xylopyranose to C-6 of the inner glucosyl unit of **2** was determined by HMBC and NOESY correlations and comparison of its ^{13}C-NMR spectrum with those of **2** and 6′-*O*-β-D-xylopyranosylsalicin (**5**).[1a)] Therefore, **3** was assigned to 2′-*O*-β-D-glucopyranosyl-6′-*O*-β-D-xylopyranosylsalicin.



1: R=β-D-Glc
6: R=H



2: R^1=β-D-Glc, R^2=H
3: R^1=β-D-Glc, R^2=β-D-Xyl
4: R^1=R^2=H
5: R^1=H, R^2=β-D-Xyl

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Table 1. ^{13}C- (125 MHz) and ^{1}H- (500 MHz) NMR Data for Compounds **1**—**3** in CD_3OD

Position	**1**		**2**		**3**	
	C	H	C	H	C	H
1	139.1	—	156.9	—	156.9	—
2	128.9	7.45 br d (7.0)	131.5	—	131.5	—
3	129.3	7.33 br t (7.0)	129.8	7.30 dd (7.5, 1.5)	129.9	7.24—7.31 m
4	128.7	7.26 tt (7.0, 1.5)	123.4	7.00 td (7.5, 1.0)	123.5	7.01 td (7.0, 1.5)
5	129.3	7.33 br t (7.0)	130.0	7.25 ddd (8.0, 7.5, 1.5)	130.3	7.24—7.31 m
6	128.9	7.45 br d (7.0)	116.0	7.19 dd (8.0, 1.0)	116.4	7.24—7.31 m
7	72.2	4.71 d (12.0)	61.4	4.63 d (13.0)	61.5	4.63 d (13.0)
		4.94 d (12.0)		4.78 d (13.0)		4.78 d (13.0)
Glc 1′	102.3	4.54 d (7.5)	100.7	5.02 d (8.0)	100.7	5.01 d (8.0)
2′	83.0	3.53 dd (9.0, 7.5)	82.1	3.82 dd (9.0, 8.0)	82.0	3.83 dd (9.0, 8.0)
3′	77.9[a)]	3.57 t (9.0)	78.4	3.70 t (9.0)	78.3[d)]	3.69 t (9.0)
4′	71.4	3.39 dd (10.0, 9.0)	71.0[b)]	3.44 dd (10.0, 9.0)	71.2	3.43 br t (9.5)
5′	77.0	3.47 ddd (10.0, 5.5, 2.0)	78.1	3.46 ddd (10.0, 5.0, 2.0)	77.3[d)]	3.69 ddd (9.5, 6.0, 2.0)
6′	69.9	3.75 dd (11.5, 5.5)	62.5	3.71[c)] dd (12.0, 5.0)	69.9	3.77 dd (11.5, 6.0)
		4.12 dd (11.5, 2.0)		3.90 dd (12.0, 2.0)		4.12 dd (11.5, 2.0)
Glc 1″	105.2	4.62 d (8.0)	105.0	4.80 d (8.0)	105.0	4.80 d (8.0)
2″	76.1	3.24 dd (9.0, 8.0)	75.9	3.22 dd (9.0, 8.0)	75.9	3.22 dd (9.0, 8.0)
3″	77.8[a)]	3.33—3.36 m	77.9	3.39 t (9.0)	78.0[d)]	3.39 t (9.0)
4″	71.4	3.33—3.36 m	71.1[b)]	3.35 t (9.0)	71.2	3.35 t (9.0)
5″	78.2	3.19 ddd (9.0, 5.0, 2.0)	78.4	3.29 ddd (9.0, 5.0, 2.0)	78.4[d)]	3.29 m
6″	62.6	3.64 dd (12.0, 5.0)	62.3	3.72[c)] dd (12.0, 5.0)	62.3	3.72 dd (12.0, 5.0)
		3.75 dd (12.0, 2.0)		3.81 dd (12.0, 2.0)		3.81 dd (12.0, 2.0)
Xyl 1‴	105.7	4.34 d (7.5)			105.5	4.32 d (7.5)
2‴	75.0	3.22 dd (9.0, 7.5)			75.0	3.20 dd (9.0, 7.5)
3‴	77.8[a)]	3.30 t (9.0)			77.7[d)]	3.28 t (9.0)
4‴	71.2	3.49 ddd (10.0, 9.0, 5.5)			71.2	3.47 ddd (10.5, 9.0, 5.5)
5‴	67.0	3.18 dd (11.5, 10.0)			66.9	3.13 dd (11.5, 10.5)
		3.86 dd (11.5, 5.5)				3.83 dd (11.5, 5.5)

a—*d*) Values with the same superscript are interchangeable. Coupling constants (*J* in Hz) are given in parenthesis.

Experimental

UV spectra were recorded on a Shimadzu UV-240 spectrophotometer and IR spectra on a Shimadzu FTIR-8200 spectrophotometer. Optical rotations were measured on a Jasco DIP-370 digital polarimeter. ^{1}H- (500 MHz) and ^{13}C- (125 MHz) NMR spectra were recorded on a Varian VXR-500 spectrometer with tetramethylsilane as an internal standard. SI-MS and HR-SI-MS were obtained with a Hitachi M-4100 mass spectrometer. Glycerol or 3-nitrobenzyl alcohol was used as the matrix. Medium pressure liquid chromatography (MPLC) was carried out with Wakogel FC-40C18. HPLC was performed using a Waters system (600E System Controller, 486 Tunable Absorbance Detector). GLC was carried out on a Shimadzu GC-18A equipped with FID.

Isolation of Glycosides Leaves of *Alangium chinense* were collected in August 1994 in Xishuangbanna, Yunnan, People's Republic of China. A voucher specimen (KPFY-941) is deposited in the laboratory of Kobe Pharmaceutical University. Dried leaves (177.6 g) of *A. chinense* were extracted with hot MeOH and the extracts were fractionated as described previously.[1b)] The residue (24.1 g) from H_2O layers was fractionated on reversed-phase MPLC. Elution with H_2O–MeOH mixtures of the indicated MeOH content gave six fractions, 1 (0%, 15.28 g), 2 (0%, 212.7 mg), 3 (5%, 1.06 g), 4 (10%, 389.2 mg), 5 (20%, 34.4 mg), and 6 (20%, 296.5 mg). Fraction 1 was purified by preparative HPLC (μBondasphere 5μ C18-100 Å, MeOH–H_2O, 1 : 3, 1 : 4) to afford **2** (7.5 mg), **4** (98.6 mg), **5** (41.3 mg) and 6′-*O*-β-glucopyranosylhenryoside (507 mg). Fractions 2—6 were submitted to preparative HPLC (μBondasphere 5μ C18-100 Å, MeOH–H_2O, 2 : 3, 7 : 13, 3 : 7, 1 : 3, 1 : 4), respectively. Fraction 2 yielded **4** (5.5 mg), **5** (9.9 mg) and 6′-*O*-β-glucopyranosylhenryoside (58.5 mg); fr. 3: **3** (2.7 mg) and 6′-*O*-β-glucopyranosylhenryoside (573 mg); fr. 4: **1** (9.1 mg), **4** (11.6 mg), 4′,6′-*O*-(*S*)-hexahydroxydiphenoylsalicin (24.8 mg), henryoside (8.0 mg) and 6′-*O*-β-glucopyranosylhenryoside (27.1 mg); fr. 5: **1** (2.5 mg), 4′,6′-*O*-(*S*)-hexahydroxydiphenoylsalicin (3.3 mg) and henryoside (1.1 mg); fr. 6: 4′,6′-*O*-(*S*)-hexahydroxydiphenoylsalicin (15.3 mg), henryoside (5.2 mg), 6′-*O*-β-glucopyranosylhenryoside (9.6 mg), **6** (12.3 mg) and *Z*-hex-3-en-1-ol β-D-xylopyranosyl(1→6)-β-D-glucopyranoside (1.1 mg).

Benzyl Alcohol β-D-Glucopyranosyl-(1→2)-[β-D-xylopyranosyl-(1→6)]-β-D-glucopyranoside (**1**): $[\alpha]_D^{22}$ −41° (*c*=0.6, MeOH). UV λ_{max}^{MeOH} nm (log ε): 216 sh (3.72), 258 (3.87), 264 sh (2.85). IR ν_{max}^{KBr} cm^{-1}: 3384, 1637, 1456. ^{1}H-NMR: Table 1. ^{13}C-NMR: Table 1. Negative ion SI-MS *m*/*z*: 563 $(M-H)^-$, 431, 401. Negative ion HR-SI-MS *m*/*z*: 563.1993 (Calcd for $C_{24}H_{35}O_{15}$: 563.1977). NOESY: H-2′/H-1″, H-6′/H-1‴, H_2-7/H-1′, H-1″/H-5″, H-1‴/H-5‴, H-1‴/H-3‴, H-1″/H-3″, H-1′/H-5′, H-2, 6/H_2-7. HMBC: H-1‴→C-6′, H-1′→C-7, H-2′→C-1″, H_2-6′→C-1‴, H-1″→C-2′, H_2-7→C-1′.

2′-*O*-β-D-Glucopyranosylsalicin (**2**): $[\alpha]_D^{22}$ −33° (*c*=0.35, MeOH). UV λ_{max}^{MeOH} nm (log ε): 213 sh (3.87), 269 (3.08), 274 (3.06). IR ν_{max}^{KBr} cm^{-1}: 3373, 1605, 1593, 1493. ^{1}H-NMR: Table 1. ^{13}C-NMR: Table 1. Negative ion SI-MS *m*/*z*: 447 $(M-H)^-$, 123. Negative ion HR-SI-MS *m*/*z*: 447.1498 (Calcd for $C_{19}H_{27}O_{12}$: 447.1503). NOESY: H-3/H_2-7, H-6/H-1′, H-1′/ H-3′, H-1′/H-5′, H-2′/H-4′, H-2′/H-1″, H-1″/ H-5″. HMBC: H-1′→C-1, H-2′→C-1″, H-1″→C-2′.

2′-*O*-β-D-Glucopyranosyl-6′-*O*-β-D-xylopyranosylsalicin (**3**): $[\alpha]_D^{19}$ −31° (*c*=0.27, MeOH). UV λ_{max}^{MeOH} nm (log ε): 213 sh (3.92), 270 sh (3.35), 275 (3.37). IR ν_{max}^{KBr} cm^{-1}: 3368, 1606, 1493. ^{1}H-NMR: Table 1. ^{13}C-NMR: Table 1. Negative ion SI-MS *m*/*z*: 579 $(M-H)^-$, 447, 417, 123. Negative ion HR-SI-MS *m*/*z*: 579.1942 (Calcd for $C_{24}H_{35}O_{16}$: 579.1926). NOESY: H-3/H_2-7, H-6/H-1′, H-1′/H-3′, H-1′/H-5′, H-1″/H-2′, H_2-6′/H-1‴, H-1‴/ H-5‴. HMBC: H-1′→C-1, H-2′→C-1″, H-1″→C-2′, H_2-6′→C-1‴, H-1‴→C-6′.

Acid Hydrolysis of Glycosides 1—3 Each glycoside (1 mg) was heated at 95 °C with dioxane (0.5 ml) and 5% H_2SO_4 (0.5 ml) for 1 h. After neutralization with Amberlite IRA-400 (OH^- form), the reaction mixture was concentrated and the residue was passed through a Sep-Pak C_{18} cartridge with H_2O. The eluate was concentrated and the residue was treated with L-cysteine methyl ester hydrochloride (1 mg) in pyridine (0.125 ml) at 60 °C for 1 h. The solution was then treated with *N*,*O*-bis(trimethylsilyl)trifluoroacetamide (0.05 ml) at 60 °C for 1 h. The supernatant was applied to GLC; GLC conditions: column, Supelco SPB^{TM}-1, 30 m×0.25 mm; column temperature, 230 °C; N_2 flow rate, 0.8 ml/min; t_R of derivatives, D-glucose 13.0 min, L-glucose 13.6 min, D-xylose 7.5 min, L-xylose 8.0 min. D-Glucose was detected from **1**—**3** and D-xylose was detected from **1** and **3**.

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