The Constituents of *Cistanche tubulosa* (SCHRENK) HOOK. f. II.[1] Isolation and Structures of a New Phenylethanoid Glycoside and a New Neolignan Glycoside

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A new phenylethanoid glycoside, named tubuloside E (I), and a new neolignan glycoside, dehydrodiconiferyl alcohol γ′-*O*-β-D-glucopyranoside (II), were isolated from the whole plants of *Cistanche tubulosa* (SCHRENK) HOOK. f. (Orobanchaceae), together with dehydrodiconiferyl alcohol 4-*O*-β-D-glucopyranoside (III), syringalide A 3′-α-L-rhamnopyranoside (IV), isosyringalide 3′-α-L-rhamnopyranoside (V), (+)-syringaresinol *O*-β-D-glucopyranoside (VI), (+)-pinoresinol *O*-β-D-glucopyranoside (VII), liriodendrin (VIII), 6-deoxycatalpol (IX), 8-epiloganic acid (X), 20-hydroxyecdysone (XI), 8-hydroxygeraniol 1-β-D-glucopyranoside (XII) and syringin (XIII). The structure of tubuloside E (I) was established as 2-(3,4-dihydroxyphenyl)ethyl *O*-α-L-rhamnopyranosyl-(1→3)-2-*O*-acetyl-4-*O*-*p*-coumaroyl-β-D-glucopyranoside on the basis of chemical evidence and spectral data.

Keywords *Cistanche tubulosa*; Orobanchaceae; phenylethanoid glycoside; iridoid; tubuloside E; dehydrodiconiferyl alcohol glycoside; syringalide rhamnoside; neolignan glycoside; ^{13}C-NMR

In a preceding paper,[1] we reported the isolation and structural determination of new phenylethanoid glycosides, tubulosides A—D from the whole plants of *Cistanche tubulosa* (SCHRENK) HOOK. f. (Orobanchaceae) in Pakistan.

This paper deals with the isolation and structural elucidation of a new phenylethanoid glycoside, named tubuloside E (I), and a new neolignan glycoside, dehydrodiconiferyl alcohol γ′-*O*-β-D-glucopyranoside (II), as well as the isolation of 11 known compounds, dehydrodiconiferyl alcohol 4-*O*-β-D-glucopyranoside (III), syringalide A 3′-α-L-rhamnopyranoside (IV), isosyringalide 3′-α-L-rhamnopyranoside (V), (+)-syringaresinol *O*-β-D-glucopyranoside(VI), (+)-pinoresinol *O*-β-D-glucopyranoside (VII), liriodendrin (VIII), 6-deoxycatalpol (IX), 8-epiloganic acid (X), 20-hydroxyecdysone (XI), 8-hydroxygeraniol 1-β-D-glucopyranoside (XII) and syringin (XIII). Compounds VI, VIII—X, XII and XIII were previously isolated from *Cistanche salsa*.[2]

Tubuloside E (I) was isolated as an amorphous powder, $C_{31}H_{38}O_{15}\cdot 3/2H_2O$, $[\alpha]_D$ −134.0° (MeOH). The infrared (IR) spectrum suggested the presence of hydroxyl groups (3430 cm^{-1}), conjugated ester (1734 cm^{-1}), a double bond (1634 cm^{-1}) and aromatic rings (1608 and 1518 cm^{-1}). The proton nuclear magnetic resonance (^{1}H-NMR) spectrum of I showed signals due to a methyl group of rhamnose [δ 1.07 (3H, d, J=6 Hz)], an acetoxyl group [δ 1.98 (3H, s)], benzylic methylene protons [δ 2.72 (2H, t, J=7 Hz)], a glucose-anomeric proton [δ 4.54 (1H, d, J=8 Hz)], a rhamnose-anomeric proton [δ 5.00 (1H, br s)], two olefinic protons [δ 6.34, 7.66 (1H each, d, J=16 Hz)] and aromatic protons [δ 6.50—7.46 (7H)].

On acetylation, compound I afforded the octaacetate (Ia), whose ^{1}H-NMR spectrum showed five aliphatic acetoxyl groups [δ 1.87, 1.95, 2.02 (3H each, s), 2.10×2 (6H, s)] and three aromatic acetoxyl groups [δ 2.27, 2.28, 2.31 (3H each, s)]. As shown in Table I, the carbon nuclear magnetic resonance (^{13}C-NMR) spectrum of I was almost identical with that of 2′-acetylacteoside (Ib),[1] except for the signals due to the *p*-coumaric acid moiety.

On methanolysis of I with acetyl chloride in methanol, methyl *p*-coumarate and 3,4-dihydroxyphenethyl alcohol were detected on thin layer chromatography (TLC) and high-performance liquid chromatography (HPLC).

Acid hydrolysis of I with 10% sulfuric acid afforded glucose and rhamnose in a ratio of 1 to 1.

From the above results, the structure of tubuloside E (I) was established as 2-(3,4-dihydroxyphenyl)ethyl *O*-α-L-rhamnopyranosyl-(1→3)-2-*O*-acetyl-4-*O*-*p*-coumaroyl-β-D-glucopyranoside.

Compound III was isolated as an amorphous powder, $[\alpha]_D$ −56.4° (MeOH). Enzymatic hydrolysis of III afforded an aglycone (IIIb) as an amorphous powder, $[\theta]_{270}$ −11300, $[\alpha]_D$ −31.6° (MeOH) and glucose. Compounds IIIb and III were identified as dehydrodiconiferyl alcohol[3,4] and its 4-*O*-β-D-glucopyranoside,[3,4] respectively, by comparison with authentic samples (IR, ^{1}H- and ^{13}C-NMR spectra).

Compound II was isolated as an amorphous powder, $[\alpha]_D$ −7.4° (MeOH), whose IR spectrum suggested the presence of a hydroxyl group (3430 cm^{-1}) and an aromatic ring (1614, 1522, 1502 cm^{-1}). The ^{1}H-NMR of II showed signals due to two aromatic methoxyl groups [δ 3.74, 3.80 (3H each, s)], a benzylic proton [δ 5.45 (1H, d, J=6 Hz)], two trans olefinic protons [δ 6.17 (1H, dt, J=16, 7 Hz), δ 6.58 (1H, d, J=16 Hz)] and five aromatic protons [δ 6.7—7.1 (5H, m)]. On acetylation, compound II afforded the hexaacetate (IIa), whose ^{1}H-NMR spectrum showed five aliphatic acetyl signals [δ 2.01, 2.03, 2.09, 2.06 (3H, 3H, 3H, 6H)]. Enzymatic hydrolysis of II afforded the aglycone (IIb) and glucose. Compound IIb was isolated as an amorphous powder, $[\alpha]_D$ +34.8° (MeOH), whose IR, ^{1}H- and ^{13}C-NMR spectra were almost identical with those of IIIb, except for $[\alpha]_D$ values and circular dichroism (CD) spectrum.

The absolute configuration of IIb was elucidated by comparison of its CD spectrum with that of IIIb and III, whose stereochemistry were confirmed in published data.[4] The CD spectrum of IIb showed $\Delta\varepsilon$ +14100 (270) and +15200 (283) as positive maxima, on the other hand, that of IIIb showed $\Delta\varepsilon$ −11300 (270) and −10990 (283) as negative maxima. These facts suggest that IIb and IIIb are enantiomers of each other. The stereochemistry of IIb must be that shown in Chart 1.

Thus, compound II was established to be dehydrodiconiferyl alcohol monoglucoside. The position of the glucose

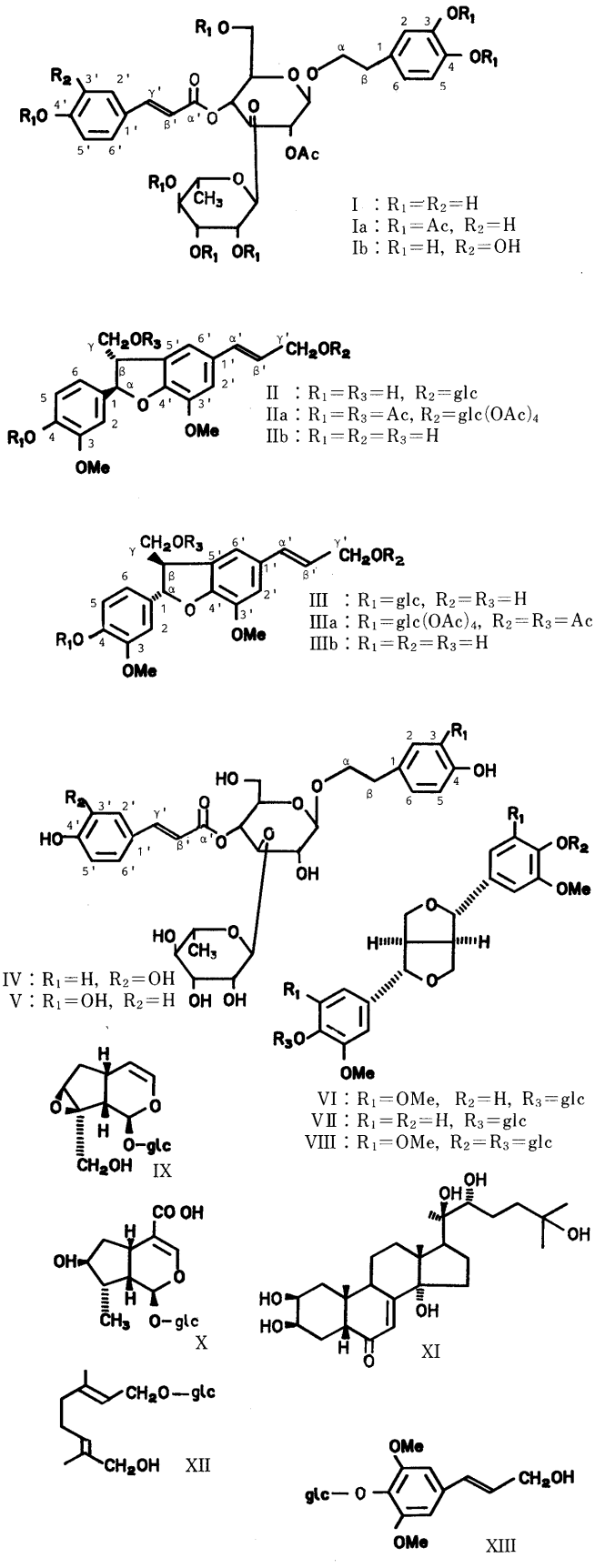

Chart 1

linkage was determined from the ^{13}C-NMR glucosylation shifts of the C-β' and C-γ' atoms. As shown in Table I, the signals at δ 123.5 and 68.8 were assigned to C-β' and C-γ',

Table I. ^{13}C-NMR Chemical Shifts

Carbon No.	I[a]	Ib[a]	II[b]	IIb[b]	III[b]	IIIb[b]
1	131.7	131.9	132.3	132.3	135.3	132.3
2	117.1	117.2	110.4	110.3	110.5	110.4
3	145.9	145.9	147.6	147.4	149.0	147.5
4	144.4	144.4	146.4	146.3	146.2	146.4
5	116.2	116.4	115.3	115.2	115.5	115.3
6	121.3	121.4	118.6	118.4	118.1	118.5
α	72.5	72.5	87.3	87.1	86.8	87.2
β	36.2	36.2	52.9	52.9	53.3	53.0
γ			63.0	62.9	63.1	63.0
1′	126.9	127.7	130.1	129.4	129.2	129.4
2′	131.3	115.5	110.4	110.3	110.5	110.4
3′	116.8	146.6	143.7	143.6	143.7	143.6
4′	161.3	149.6	147.4	147.0	147.1	147.1
5′	116.8	116.6	131.9	130.4	130.7	130.5
6′	131.3	123.2	115.3	114.9	115.0	114.9
α'	168.0	168.1	129.5	128.9	129.0	129.0
β'	114.6	114.7	123.5	127.9	128.1	128.0
γ'	147.7	148.1	68.8	61.5	61.6	
Glc-1	101.6	101.0	102.0		100.2	
2	75.0	75.1	73.6		73.0	
3	80.4	80.3	76.8		77.0	
4	70.7	70.7	70.2		69.7	
5	76.0	76.0	76.8		76.9	
6	62.1	62.2	61.2		60.7	
Rham-1	103.1	103.1				
2	71.8	72.0				
3	71.8	71.7				
4	73.5	73.6				
5	70.7	70.7				
6	18.4	18.4				
OAc	20.9	20.9				
	171.4	171.5				
OMe			55.7	55.6	55.8	55.6

a) In methanol-d_4. *b*) In DMSO-d_6.

respectively. The shifts of the corresponding carbons in going to II from IIb were -4.4 and $+7.3$ ppm, which indicated that a glucosyl moiety in II was linked to the hydroxyl group at C-γ' atom.[5]

From the above results, compound II was established as dehydrodiconiferyl alcohol γ'-*O*-β-D-glucopyranoside.

Compounds IV and V were isolated as amorphous powder (IV, $C_{29}H_{36}O_{14}\cdot 5/2H_2O$; V, $C_{29}H_{36}O_{14}\cdot 1/2H_2O$). The IR spectra suggested the presence of hydroxyl groups (3400 cm^{-1}), a double bond (1634 cm^{-1}), and aromatic rings (1606 and 1518 cm^{-1}).

On acetylation, compounds IV and V afforded the octaacetates (IVa, Va), whose ^{1}H-NMR spectra showed five aliphatic acetoxyl and three aromatic acetoxyl signals, respectively. Compounds IVa and Va were identified as syringalide A 3′-α-L-rhamnopyranoside octaacetate and isosyringalide 3′-α-L-rhamnopyranoside octaacetate, respectively by comparison of (*Rf*, IR and ^{1}H-NMR) with authentic samples.[6] The ^{1}H- and ^{13}C-NMR spectrum of IV and V supported those structures (see Experimental).

Compounds VI—XIII were identified as (+)-syringaresinol *O*-β-D-glucopyranoside (VI),[2c] (+)-pinoresinol *O*-β-D-glucopyranoside (VII),[7] liriodendrin (VIII),[2b] 6-deoxycatalpol (IX),[2c] 8-epiloganic acid (X),[2a] 20-hydroxyecdysone (XI),[8] 8-hydroxygeraniol 1-β-D-glucopyranoside (XII),[2a] and syringin (XIII),[2d] respectively, by comparison with authentic samples (see Experimental).

Experimental

Melting points were determined on a Mitamura micro-melting point apparatus and are uncorrected. Optical rotations were measured with a JASCO DIP-140 digital polarimeter. IR spectra were recorded with a Hitachi 279-30 IR spectrophotometer and ultraviolet (UV) spectra with a Hitachi 200-20 spectrophotometer. CD spectra were recorded with a JASCO J-20A CD spectrophotometer and mass spectra (MS) with a JEOL JMS-100. ^{1}H-NMR and ^{13}C-NMR spectra were recorded with a JEOL FX-90Q machine (89.55 and 22.5 MHz, respectively).

Chemical shifts are given on the δ (ppm) scale with tetramethylsilane (TMS) as an internal standard. The abbreviations used for nuclear magnetic resonance (NMR) data are as follows; s, singlet; d, doublet; t, triplet; q, quartet; br, broad. Gas chromatography (GC) was run on a Shimadzu GC-4CM apparatus with a flame ionization detector. HPLC was performed on a Hitachi 655A-11 machine. Silica gel (Wako gel C-300) was used for column chromatography. Silica gel 60 F_{254} (Merck) precoated plates were used for TLC and detection was carried out by spraying 10% H_2SO_4 followed by heating.

Isolation Whole plants of *C. tubulosa* (7 kg), collected in December 1986, in Karachi, Pakistan, were extractd with MeOH (20 l × 2) under reflux. The extract was concentrated under reduced pressure and the residue was suspended in water. This suspension was extracted with EtOAc saturated with water. The water layer was subjected to a Diaion HP-20 (Nippon Rensui Co.) column and washed with H_2O, and then eluted with MeOH. The MeOH eluate (140 g) was chromatographed on a polyamide C-200 (Wako Pure Chemical) column using H_2O and then MeOH to give two fractions (fraction 1, H_2O eluate; fraction 2, MeOH eluate). Fraction 1 was concentrated to give a residue. The residue was chromatographed on a silica gel column using $CHCl_3$–MeOH–H_2O repeatedly. After repeated chromatography of HPLC (column, Develosil ODS-10, 20 × 250 mm; solvent; H_2O–CH_3CN or H_2O–MeOH), 10 compounds were isolated [II (45 mg), III (40 mg), VI (35 mg), VII (43 mg), VIII (12 mg), IX (8 mg), X (10 mg), XI (20 mg), XII (10 mg) and XIII (3.6 mg)]. Fraction 2 was treated in the same way as fraction 1 and gave compounds I (150 mg), IV (10 mg) and V (10 mg).

Tubuloside E (I) Amorphous powder, $[\alpha]_D^{23}$ −134.0° (c = 1.5, MeOH). *Anal.* Calcd for $C_{31}H_{38}O_{15}\cdot 3/2H_2O$: C, 54.98; H, 6.10. Found: C, 55.01; H, 5.98. IR ν_{max}^{KBr} cm^{-1}: 3430, 1732, 1634, 1608, 1518. ^{1}H-NMR (MeOH-d_4) δ: 1.07 (3H, d, J = 6 Hz, CH_3 of rhamnose), 1.98 (3H, s, OAc), 2.72 (2H, t, J = 7 Hz, Ar–CH_2–), 4.54 (1H, d, J = 8 Hz, H-1 of glucose), 5.00 (1H, br s, H-1 of rhamnose), 6.34 (1H, d, J = 16 Hz, Ar–CH = C<u>H</u>′–), 6.5—6.8 (3H, aromatic H), 6.80 (2H, d, J = 9 Hz, H-3′, H-5′), 7.46 (2H, d, J = 9 Hz, H-2′, H-6′), 7.66 (1H, d, J = 16 Hz, Ar–C<u>H</u> = CH–). ^{13}C-NMR: Table I.

Dehydrodiconiferyl Alcohol γ′-*O*-β-D-Glucopyranoside (II) Amorphous powder, $[\alpha]_D^{25}$ −7.4° (c = 1.6, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3430, 1614, 1522, 1502. UV λ_{max}^{MeOH} nm: 227 (sh), 280. ^{1}H-NMR (DMSO-d_6) δ: 3.74, 3.80 (3H each, s, OCH_3), 5.45 (1H, d, J = 6 Hz, H-α), 6.17 (1H, dt, J = 16, 7 Hz, H-β′), 6.58 (1H, d, J = 16 Hz, H-α′), 6.7—7.1 (5H, m, aromatic H). ^{13}C-NMR: Table I.

Dehydrodiconiferyl Alcohol 4-*O*-β-D-Glucopyranoside (III) Amorphous powder, $[\alpha]_D^{25}$ −56.4° (c = 1.1, MeOH). IR ν_{max}^{KBr} cm^{-1}: 3400, 1604, 1566, 1502. UV λ_{max}^{MeOH} nm: 222, 277. ^{1}H-NMR (DMSO-d_6) δ: 3.76, 3.82 (3H each, s, OCH_3), 5.52 (1H, d, J = 7 Hz, H-α), 6.18 (1H, dt, J = 16, 7 Hz, H-β′), 6.49 (1H, d, J = 16 Hz, H-α′), 6.7—7.2 (5H, m, aromatic H). ^{13}C-NMR: Table I.

Syringalide A 3′-α-L-Rhamnopyranoside (IV) Amorphous powder, *Anal.* Calcd for $C_{29}H_{36}O_{14}\cdot 5/2H_2O$: C, 56.11; H, 5.94. Found: C, 56.40; H, 6.04. IR ν_{max}^{KBr} cm^{-1}: 3400, 1606, 1518. ^{1}H-NMR (MeOH-d_4) δ: 1.15 (3H, d, J = 6 Hz, CH_3 of rhamnose), 2.90 (2H, t, J = 7 Hz, Ar–CH_2–), 4.46 (1H, d, J = 8 Hz, H-1 of glucose), 5.27 (1H, br s, H-1 of rhamnose), 6.35 (1H, d, J = 16 Hz, Ar–CH = C<u>H</u>′–), 6.6—7.3 (3H, aromatic H), 7.68 (1H, d, J = 16 Hz, Ar–C<u>H</u> = CH–). ^{13}C-NMR (CD_3OD) δ: 130.6 (C-1), 116.1 (C-2), 130.8 (C-3, 5), 156.6 (C-4), 116.1 (C-6), 72.1 (C-α, rham-2, 3), 36.2 (C-β), 127.6 (C-1′), 114.6 (C-2′), 149.6 (C-3′), 146.6 (C-4′), 116.4 (C-5′), 123.1 (C-6′), 168.2 (C-α′), 115.2 (C-β′), 147.0 (C-γ′), 104.0 (glu-1), 76.0 (glu-2), 81.5 (glu-3), 70.3 (glu-4), 75.9 (glu-5), 62.3 (glu-6), 102.8 (rham-1), 73.7 (rham-4), 70.5 (rham-5), 18.3 (rham-6).

Isosyringalide 3′-α-L-Rhamnopyranoside (V) Amorphous powder, *Anal.* Calcd for $C_{29}H_{36}O_{14}\cdot 1/2H_2O$: C, 56.31; H, 5.96. Found; C, 56.40; H, 6.04. IR ν_{max}^{KBr} cm^{-1}: 3420, 1608, 1518. ^{1}H-NMR (MeOH-d_4) δ: 1.09 (3H, d, J = 6 Hz, CH_3 of rhamnose), 2.80 (2H, t, J = 6 Hz, Ar–CH_2–), 4.40 (1H, d, J = 8 Hz, H-1 of glucose), 5.23 (1H, br s, H-1 of rhamnose), 6.37 (1H, d, J = 18 Hz, Ar–CH = C<u>H</u>′–), 6.6—7.2 (3H, aromatic H), 7.71 (1H, d, J = 18 Hz, Ar–C<u>H</u> = CH–). ^{13}C-NMR (CD_3OD) δ: 131.4 (C-1), 116.3 (C-2), 144.5 (C-3), 146.0 (C-4), 117.0 (C-5), 121.2 (C-6), 72.2 (C-α), 36.5 (C-β), 127.1 (C-1′), 116.8 (C-2′, 6′), 131.2 (C-3′, 5′), 161.2 (C-4′), 168.2 (C-α′), 114.7 (C-β′), 147.5 (C-γ′), 104.1 (glu-1), 76.1 (glu-2), 81.5 (glu-3), 70.2 (glu-4), 75.9 (glu-5), 62.3 (glu-6), 102.8 (rham-1), 72.1 (rham-2, 3), 73.7 (rham-4), 70.6 (rham-5), 18.4 (rham-6).

(+)-Syringaresinol *O*-β-D-Glucopyranoside (VI) Amorphous powder, $[\alpha]_D^{25}$ −14.4° (c = 0.1, MeOH). *Anal.* Calcd for $C_{28}H_{36}O_{13}$: C, 57.93; H, 6.25. Found: C, 58.01; H, 6.26. IR ν_{max}^{KBr} cm^{-1}: 3430, 1600, 1518. ^{1}H-NMR (DMSO-d_6) δ: 3.78 (12H, s, OCH_3 × 4), 6.60, 6.66 (2H each, s, aromatic H).

(+)-Pinoresinol *O*-β-D-Glucopyranoside (VII) Amorphous powder, $[\alpha]_D^{25}$ +38.9° (c = 0.6, MeOH). *Anal.* Calcd for $C_{26}H_{32}O_{11}\cdot 1/2H_2O$: C, 58.97; H, 6.28. Found: 58.94; H, 6.29. IR ν_{max}^{KBr} cm^{-1}: 3425, 1606, 1516. ^{1}H-NMR (C_5D_5N) δ: 3.77, 3.80 (3H each, s, OCH_3), 6.9—7.7 (6H, m, aromatic H).

Liriodendrin (VIII) Colorless needles (MeOH), mp 255—257 °C, $[\alpha]_D^{25}$ −9.2° (c = 1.3, pyridine). IR ν_{max}^{KBr} cm^{-1}: 3400, 1598, 1510. ^{1}H-NMR (DMSO-d_6) δ: 3.78 (12H, s, OCH_3 × 4), 6.67 (4H, s, aromatic H).

6-Deoxycatalpol (IX) Colorless needles (MeOH), mp 204—206 °C, $[\alpha]_D^{25}$ −50.1° (c = 0.7, MeOH). *Anal.* Calcd for $C_{15}H_{22}O_9$: C, 52.02; H, 6.40. Found: C, 52.12; H, 6.41. IR ν_{max}^{KBr} cm^{-1}: 3400, 1658. ^{1}H-NMR (D_2O) δ: 1.3—1.9 (1H, m, H-6), 2.1—2.7 (3H, m, H-5, H-6, H-9), 3.65 (1H, s-like, H-7), 3.90, 4.35 (2H, ABsystem, J = 13 Hz, H-10), 4.92 (1H, d, J = 7 Hz, aromeric H), 5.10 (1H, dd, J = 6, 4 Hz, H-4), 5.12 (1H, d, J = 10 Hz, H-1), 6.38 (1H, dd, J = 6, 1 Hz, H-3).

8-Epiloganic Acid (X) Colorless needles (MeOH), mp 147—149 °C, $[\alpha]_D^{25}$ −135.0° (c = 1.0, pyridine). *Anal.* Calcd for $C_{16}H_{24}O_{10}\cdot H_2O$: C, 48.73; H, 6.65. Found: C, 48.60; H, 6.70. IR ν_{max}^{KBr} cm^{-1}: 3400, 1680, 1640, 1430. ^{1}H-NMR (C_5D_5N) δ: 1.18 (3H, d, J = 7 Hz, CH_3), 2.2—2.5 (2H, m, H-6), 2.60 (1H, m, H-8), 3.08 (1H, dt, J = 3, 8.5 Hz, H-9), 3.58 (1H, m, H-5), 5.40 (1H, d, J = 7.5 Hz, anomeric H), 5.91 (1H, d, J = 3 Hz, H-1), 7.92 (1H, s, H-3).

20-Hydroxyecdysone (XI) Crystalline solid, mp 241—242 °C, $[\alpha]_D^{25}$ +70.0° (c = 1.1, MeOH). *Anal.* Calcd for $C_{27}H_{44}O_7$: C, 67.47; H, 9.23. Found: C, 67.28; H, 9.18. IR ν_{max}^{KBr} cm^{-1}: 3440, 1646. ^{1}H-NMR (C_5D_5N) δ: 1.06 (3H, s, H-19), 1.20 (3H, s, H-18), 1.36 (6H, s, H-26, H-27), 1.58 (3H, s, H-21), 6.24 (1H, d, J = 2 Hz, H-7). ^{13}C-NMR (C_5D_5N): 38.0 (C-1), 68.2 (C-2, C-3), 32.5 (C-4), 51.5 (C-5), 203.7 (C-6), 121.8 (C-7), 166.3 (C-8), 34.6 (C-9), 38.8 (C-10), 21.3 (C-11), 31.9 (C-12), 48.2 (C-13), 84.4 (C-14), 32.1 (C-15), 21.6 (C-16), 50.2 (C-17), 18.0 (C-18), 24.6 (C-19), 77.0 (C-20), 21.8 (C-21), 77.7 (C-22), 27.6 (C-23), 42.7 (C-24), 69.8 (C-25), 30.1 (C-26, 27).

8-Hydroxygeraniol 1-β-D-Glucopyranoside (XII) Colorless needles (MeOH), mp 58—60 °C, $[\alpha]_D^{25}$ −40.1° (c = 1.2, MeOH). *Anal.* Calcd for $C_{16}H_{28}O_7\cdot 1/2H_2O$: C, 56.29; H, 8.56. Found: C, 56.40; H, 8.50. IR ν_{max}^{KBr} cm^{-1}: 3350, 1670, 1446. ^{1}H-NMR (C_5D_5N) δ: 1.72, 1.59 (3H each, s, CH_3), 2.04 (4H, br m, H-4, H-5), 4.80 (1H, d, J = 7.5 Hz, anomeric H), 5.56 (2H, br t, J = 7 Hz, H-6).

Syringin (XIII) Colorless needles (MeOH), mp 188—189 °C, $[\alpha]_D^{25}$ −15.3° (c = 0.4, MeOH). *Anal.* Calcd for $C_{17}H_{24}O_9$: C, 55.13; H, 6.53. Found: C, 54.83; H, 6.50. IR ν_{max}^{KBr} cm^{-1}: 3400, 1592, 1514. ^{1}H-NMR (DMSO-d_6) δ: 3.78 (6H, s, OCH_3 × 2), 6.3—6.5 (2H, m, –CH = CH–), 6.72 (2H, s, aromatic H).

Acetylation of I Compound I (50 mg) was dissolved in pyridine (1 ml) and acetic anhydride (1 ml) and left at room temperature overnight. The reaction mixture was poured into ice-water, and then extracted with EtOAc. The EtOAc extract was concentrated *in vacuo* and the residue was chromatographed on a silica gel column using benzene–acetone (5 : 1) to give the octaacetate (Ia) (35 mg). IR ν_{max}^{KBr} cm^{-1}: 1760, 1638, 1604, 1508. ^{1}H-NMR ($CDCl_3$) δ: 1.02 (3H, d, J = 6 Hz, CH_3 of rhamnose), 1.87, 1.95, 2.02 (3H each, s, OAc), 2.10 (6H, s, OAc × 2), 2.27, 2.28, 2.31 (3H each, s, Ar–OAc), 2.90 (2H, t, J = 7 Hz, Ar–CH_2–), 6.38 (1H, d, J = 16 Hz, AR–CH = C<u>H</u>–), 6.80—7.15 (3H, m, aromatic H), 7.24 (2H, d, J = 9 Hz, H-3′, H-5′), 7.55 (2H, d, J = 9 Hz, H-2, H-6), 7.72 (1H, d, J = 16 Hz, Ar–C<u>H</u> = CH–).

Acetylation of II and III Compounds II and III (20 mg each) were acetylated in the same way as described for compound I and gave the hexaacetates [IIa (12 mg), IIIa (15 mg)]. IIa: IR ν_{max}^{KBr} cm^{-1}: 1754, 1608, 1506. ^{1}H-NMR ($CDCl_3$) δ: 2.01, 2.03, 2.09 (3H each, s, OAc), 2.06 (6H, s, OAc × 2), 2.30 (3H, s, OAc), 3.84 (3.95 (3H each, s, OCH_3), 5.54 (1H, d, J = 6 Hz, H-α), 6.05 (1H, dt, J = 16, 7 Hz, H-β′), 6.53 (1H, d, J = 16 Hz, H-α′), 6.8—7.1 (5H, m, aromatic H). IIIa: IR ν_{max}^{KBr} cm^{-1}: 1760, 1604, 1516. ^{1}H-NMR ($CDCl_3$) δ: 2.05 (9H, s, OAc × 3), 2.08 (6H, s, OAc × 2), 2.11 (3H, s, OAc), 3.80, 3.92 (3H each, s, OCH_3), 5.51 (1H, d, J = 7 Hz, H-α), 6.14 (1H, dt, J = 16, 7 Hz, H-β′), 6.62 (1H, d, J = 16 Hz, H-α′), 6.8—7.2 (5H, m, aromatic H).

Acetylation of IV and V Compounds IV and V were acetylated in the

same way as described for compound I and gave the octaacetates [IVa, Va]. IVa: IR ν_{max}^{KBr} cm^{-1}: 1754, 1636, 1514. ^{1}H-NMR ($CDCl_3$) δ: 1.03 (3H, d, J=6 Hz, CH_3 of rhamnose), 1.87, 1.94, 1.99 (3H each, s, OAc), 2.09 (6H, s, OAc×2), 2.28 (6H, s, OAc×2), 2.30 (3H, s, OAc), 2.86 (2H, t, J=6.6 Hz, Ar–CH_2–), 3.67 (2H, t, J=7.2 Hz Ar–CH_2–CH_2–), 6.35 (1H, d, J=16 Hz, Ar–CH=CH–), 6.87—7.35 (3H, m, Ar–H), 7.09 (4H, q, J=8.6 Hz, $A_2'B_2'$type), 7.65 (1H, d, J=16 Hz, Ar–CH=CH–). Va: IR ν_{max}^{KBr} cm^{-1}: 1750, 1636, 1608, 1514. ^{1}H-NMR ($CDCl_3$) δ: 1.03 (3H, d, J=6 Hz, CH_3 for rhamnose), 1.87, 1.94, 2.02 (3H each, s, OAc), 2.09 (6H, s, OAc×2), 2.28 (6H, s, OAc×2), 2.31 (3H, s, OAc), 2.86 (2H, t, J=6.6 Hz, Ar–CH_2–), 3.73 (2H, t, J=6.6 Hz, Ar–CH_2–CH_2–), 6.34 (1H, d, J=16 Hz, Ar–CH=CH–), 6.78—7.20 (3H, m, Ar–H), 7.31 (4H, q, J=8.5 Hz, $A_2'B_2'$type), 7.71 (1H, d, J=16 Hz, Ar–CH=CH–).

Methanolysis of I Compound I (*ca.* 1 mg) was refluxed with methanolic 5% CH_3COCl (2 ml) for 30 min, and then the reagent was evaporated off *in vacuo*. The presence of methyl *p*-coumarate and 3,4-dihydroxyphenethyl alcohol in the residue was detected by TLC [$CHCl_3$–MeOH (10:1)] and HPLC [column, Develosil ODS-7 (4.6×250 mm); solvent, 55% MeOH; detector (UV), 220 nm; flow rate, 1.0 ml/min]. Methyl *p*-coumarate; *Rf* 0.82, t_R (min) 8.8, 3,4-dihydroxyphenethyl alcohol; *Rf* 0.31, t_R (min) 3.2.

Acid Hydrolysis of I A solution of compound I (2 mg) in 10% H_2SO_4 (1 ml) was heated in a boiling water bath for 30 min. The solution was passed through an Amberlite IR-45 column and concentrated to give a residue, which was reduced with sodium borohydride (*ca.* 3 mg) for 1 h. The reaction mixture was passed through an Amberlite IR-120 column and concentrated to dryness. Boric acid was removed by distillation with MeOH and the residue was acetylated with acetic anhydride (1 drop) and pyridine (1 drop) at 100 °C for 1 h. The reagents were evaporated off *in vacuo*. Glucitol acetate and rhamnitol acetate were detected in a ratio of 1 to 1 from compound I by GC. Condition: column, 1.5% OV-17, 3 mm×1.5 m; column temp., 180 °C; carrier gas, N_2 (30 ml/min). t_R (min): 2.0 (rhamnitol acetate), 5.5 (glucitol acetate).

Enzymatic Hydrolysis of II and III Compound II (13 mg) or III (40 mg) was hydrolyzed with cellulase (40 mg) in water (1 ml) for 2 h at 37 °C. The reaction mixture was extracted with EtOAc. The EtOAc extract was concentrated *in vacuo* to give the residue, which was chromatographed on the HPLC [column, Develosil ODS-10 (20×250 mm); solvent. II: H_2O–CH_3CN (77.5:22.5); III: H_2O–CH_3CN (80:20); detector (UV), 205 nm; flow rate, 6.0 ml/min] to give IIb (3.3 mg), IIIb (5.7 mg). IIb: $[\alpha]_D^{25}$ +34.8° (c=0.3, MeOH). CD (c=3.3 mg/10 ml, MeOH) $[\theta]^{25}$ (nm): 1740 (330), 15200 (283), 14100 (270), 0 (247), −16700 (232). IR ν_{max}^{KBr} cm^{-1}: 3380, 1606, 1520, 1500. UV λ_{max}^{MeOH} nm: 276. MS m/z (%): 358 (M^+, 53), 340 (M^+-H_2O, 100), 338 (58), 325 (47), 310 (23), 162 (41), 151 (36), 137 (59). ^{1}H-NMR (acetone-d_6) δ: 3.55 (1H, br t, J=6 Hz, H-β), 3.84, 3.87 (3H each, s, OCH_3), 4.19 (2H, br d, J=5 Hz, H-γ'), 5.56 (1H, d, J=6, 5 Hz, H-α), 6.23 (1H, dt, J=16, 5 Hz, H-β'), 6.56 (1H, br d, J=16 Hz, H-α'), 6.7—7.1 (5H, m, aromatic H). ^{13}C-NMR: Table I. IIIb: $[\alpha]_D^{25}$ −31.6° (c=0.6, MeOH). CD (c=2.28 mg/10 ml, MeOH) $[\theta]^{25}$ (nm): 2200 (330), 0 (310), −10990 (283), −11300 (270), 0 (242), 5650 (232). ^{1}H-NMR (acetone-d_6) δ: 3.84, 3.87 (3H each, s, OCH_3×2), 4.19 (2H, br d, J=5 Hz, H-γ'), 5.56 (1H, d, J=6.5 Hz, H-α), 6.23 (1H, dt, J=16, 5 Hz, H-β'), 6.52 (1H, d, J=16 Hz, H-α'), 6.7—7.1 (5H, m, aromatic H). ^{13}C-NMR: Table I. The IR and UV spectra were similar with that of IIb.

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