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# Synthesis of vicenin-1 and 3, 6,8- and 8,6-di-*C*-β-D-(glucopyranosyl-xylopyranosyl)-4′,5,7-trihydroxyflavones using two direct C-glycosylations of naringenin and phloroacetophenone with unprotected D-glucose and D-xylose in aqueous solution as the key reactions

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ABSTRACT

Vicenin-3 was synthesized from naringenin via a short five-step reaction, which included two regioselective direct C-glycosylations with D-glucose and D-xylose (yields: 22% and 30%, respectively) as the key reactions for a total yield of 4.4%. Vicenin-1 was also synthesized from phloroacetophenone via a 10-step reaction, including the same glycosylation described above, for a total yield of 2.7% with a vicenin-3 yield of 1.7%.



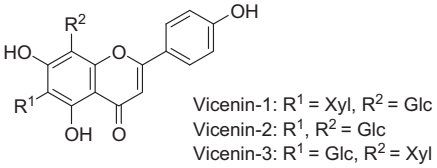
## 1. Introduction

Many flavonoids in plants include glycosides that are mostly present as water-soluble O-glycosides and, rarely, as C-glycosides. The *C*-glycosylflavonoids include a few bis-C-glycosides, mostly flavones. To date, 55 di-*C*-glycosylflavones have been isolated and their structures determined.[1] Many of them include apigenin (4′,5,7-trihydroxyflavone) as the aglycon. Some of these *C*-glycosylflavonoids show bioactivities different from the corresponding *O*-glycosylflavonoids and their aglycons, because of differences in their stability to hydrolysis.[2] Although there are a few reports on the efficient synthesis of mono-*C*-glycosylflavonoids,[3] there are no reports on the synthesis of bis-*C*-glucosylflavonoids except for a recent report.[4]

We have achieved the synthesis of the naturally occurring di-*C*-β-D-glucosylflavone (vicenin-2, see Fig. 1), di-*C*-β-D-glucosyldihydrochalcone, and di-*C*-β-D-glucosylflavanone.[4] In plants, however, it is rare to find a di-*C*-glycosylflavone consisting of two alternative sugars as a bis-C-glycoside, which consists of five kinds of D-sugars, such as glucose, galactose, xylose, arabinose, and rhamnose.[1] Three kinds of 6,8-di-*C*-glycosyl-4′,5,7-trihydroxyflavones have been isolated from plants: vicenin-1 and -3 [6-Xyl-8-Glc (**1**), 6-Glc-8-Xyl (**2**), see Fig. 1], violanthin and isoviolanthin (6-Glc-8-Rha, 6-Rha-8-Glc), and schaftoside and isoschaftoside (6-Glc-8-Ara, 6-Ara-8-Glc).[1] We have not yet attempted the synthesis of bis-C-glycosides that consist of different sugars.

We previously studied an environmentally friendly method for the direct C-glycosylation of acetylpolyphenol with a non-protected sugar in an aqueous solution in the presence of scandium trifluoromethanesulfonate [$Sc(OTf)_3$].[5] The first synthesis of the three di-*C*-glycosylflavonoids listed above was achieved by application of our method.[4] This article describes the total synthesis of **1** and **2** and the application of this direct C-glycosylation method to naringenin and phloroacetophenone.

Vicenin-1 and -3 (**1**, **2**) were isolated from the leaves of *Desmodium styracifolium* MERR (Leguminosae), which has been used as a Chinese folk medicine for cholelithiasis, lithiasis, and inflammation of the liver, among other ailments.[6] Vicenin-1 (**1**) has also been isolated from *Vitex lucens* (Verb.),[7] *Arrhenatherum* sp. (Gram.),[8] *Cymophyllum fraseri* (Cyp.),[9] *Eminium spiculatum* (Acer.),[10] *Ephedra* sp.




Vicenin-1: $R^1$ = Xyl, $R^2$ = Glc
Vicenin-2: $R^1$, $R^2$ = Glc
Vicenin-3: $R^1$ = Glc, $R^2$ = Xyl

**Figure 1.** Structure of vicenin-1 (**1**), -2, and -3 (**2**).

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(Ephed.),[11] and *Rhynchosia jacobii* (Leg.),[12] and **2** has been isolated from *V. lucens* (Verb.),[7] *Camellia sinensis* (Thea.),[13] *Ephedra* sp. (Ephed.),[11] and *Premna integrifolia* (Verb.).[14] However, the bioactivity of these compounds has not yet been reported.

The synthesis of **1** and **2** was examined by two methods: (1) a readily available method of two regioselective direct C-glycosylations of naringenin, followed by oxidation, and (2) two direct C-glycosylations of phloroacetophenone, followed by aldol condensation, acid-cyclization, and then oxidation (Scheme 1).

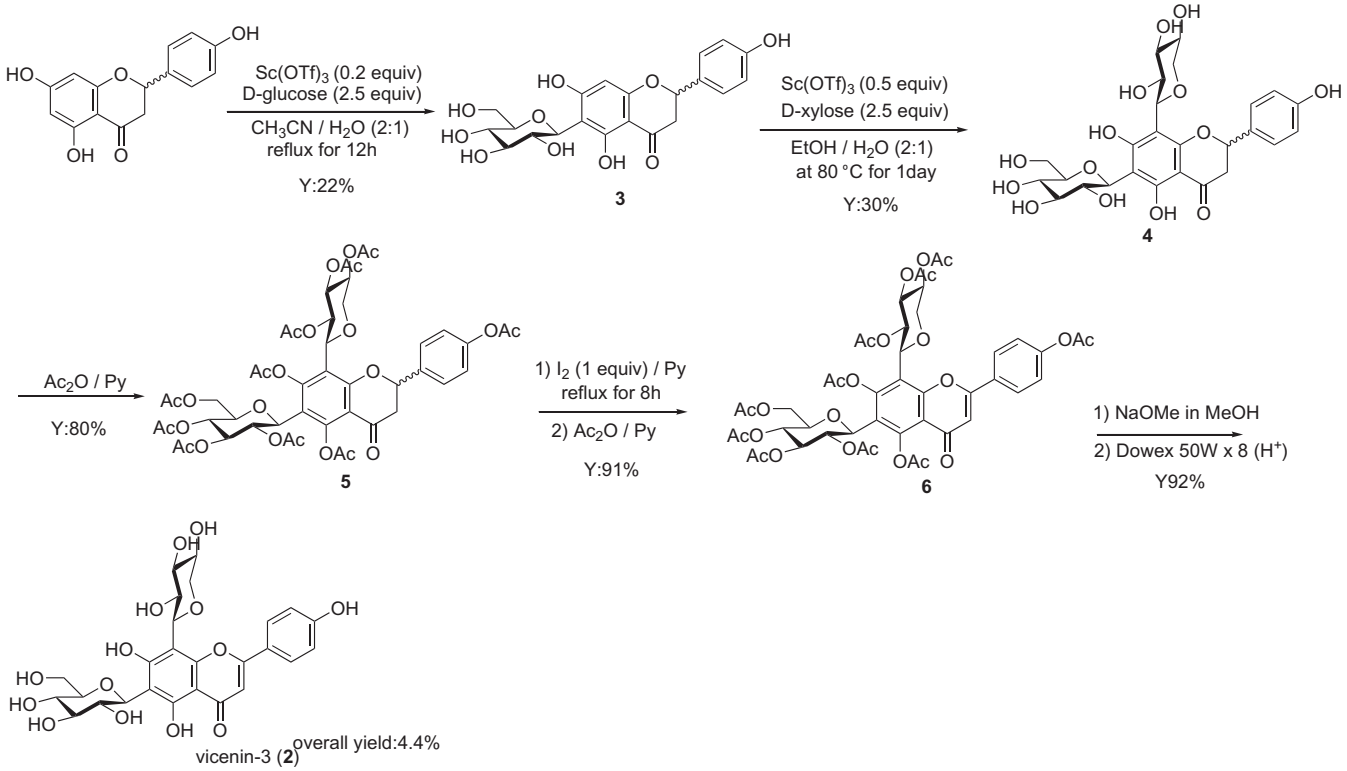
## 2. Results and discussion

Direct C-glycosylation of naringenin, in which the phenolic hydroxyl groups are partially benzyl protected, has been attempted by our group[3c] and by Oyama and Kondo,[3g] whose attempts were unsuccessful. Kondo's group achieved the C-glycosylation of naringenin after reduction of its carbonyl group to a methylene residue. We also tried the direct C-glycosylation of unprotected naringenin with D-glucose in an aqueous solution in the presence of catalytic amounts of $Sc(OTf)_3$ and found that this method afforded 6- or 8-*C*-glucosyl-4′,5,7-trihydroxyflavanone and 6,8-di-*C*-glucosyl-4′,5,7-trihydroxyflavanone in 17.3% and 19.0% yields, respectively.[4] Herein, we applied this method to direct C-glycosylation of naringenin with D-glucose and D-xylose in the synthesis of **1** and **2**.

Direct C-glycosylation of naringenin and isolation of the desired glycoside were performed as follows. An aqueous $CH_3CN$ solution (2:1 $CH_3CN$–$H_2O$) of naringenin (1 equiv) and D-glucose (2.5 equiv) was refluxed in an oil bath (85 °C) for 12 h in the presence of 0.2 equiv of $Sc(OTf)_3$. The reaction mixture was diluted with water (50 mL) and was then placed and absorbed onto a column of Diaion CHP20P resin (ca. 50 mL in water). The resulting resin was washed with water (200 mL) and then eluted with 50% aqueous acetone (100 mL) and 1:1 acetone–MeOH (100 mL). Unabsorbed fractions included unreacted D-glucose and $Sc(OTf)_3$. The eluate, with 50% aqueous acetone and acetone–MeOH, including naringenin and naringenin glycosides, was evaporated to give an amorphous solid, which was subjected to silica-gel column chromatography (15:30:2:0.1 and 30:30:5:0.1 acetone–EtOAc–$H_2O$–AcOH). As a result of this experiment, the naringenin mono-C-glucoside **3** was afforded as a pale-yellow amorphous powder in 22% yield. Since it was unclear whether **3** was 6-*C*-β-D- or 8-*C*-β-D-glucopyranosylnaringenin, **3** was acetylated by $Ac_2O$, pyridine, and *N*,*N*-dimethylaminopyridine (DMAP). Using $^1H$ NMR spectroscopy, the chemical shift of an aromatic proton of the acetate **3′**[4] was compared with that of the acetates of 6-*C*-β-D-glucosylnaringenin (hemiphloin) and 8-*C*-β-D-glucosylnaringenin (isohemiphloin). Since the chemical shift ($\delta$ in $CDCl_3$) of the acetate **3′** was 6.79,[4] and that of hemiphloin and isohemiphloin hepta-acetates was 6.79 and 6.56, respectively,[15] mono-C-glycoside **3** was determined to be a 6-*C*-β-D-glucopyranosyl-4′,5,7-trihydroxyflavanone. This result suggests that the C-glycosylation of naringenin with D-glucose proceeded both regioselectively and stereoselectively.

Next, a second C-glycosylation of **3** with D-xylose was examined. The best conditions were as follows. An aqueous EtOH solution (2:1 EtOH–water) of **3** and 2.5 equiv of D-xylose was refluxed at 80 °C for one day in the presence of 0.5 equiv of $Sc(OTf)_3$. The reaction mixture was worked up and purified in the same manner as described above. The desired 6-*C*-β-D-glucopyranosyl-8-*C*-β-D-xylopyranosylnaringenin (**4**) was obtained in a yield of 30%. Acetylation ($Ac_2O$–pyridine–DMAP) of **4** gave deca-acetate **5** in a yield of 80%, and the structure of **4** was confirmed by a detailed analysis of the $^1H$ and $^1H$–$^1H$ correlation NMR spectroscopy of **5**.

Next, synthesis of flavones via oxidation of flavanone acetate **5** was examined. Dichlorodicyanobenzoquinone (DDQ) was previously used in the oxidation reaction.[3g,4] After oxidation followed by acetylation ($Ac_2O$–pyridine), separation and purification were




**Scheme 1.** Synthesis of vicenin-3 (**2**) via direct C-glucosylation and C-xylosylation of naringenin.

carried out. However, since the separation of products from the resulting hydroquinone acetates was difficult, and the yield was a low 30%, oxidation using iodine was employed.[16] Halogenation of α-H in **5**, followed by elimination of HI due to pyridine, proceeded smoothly. A solution of **5** in pyridine was refluxed for 8 h in the presence of 1 equiv of iodine, followed by re-acetylation ($Ac_2O$–pyridine) to afford flavone deca-acetate **6** as a yellow amorphous solid in a yield of 91%, which was O-de acetylated by sodium methoxide in dry MeOH and neutralized by Dowex 50Wx8 ($H^+$) resin to afford the desired **2** in a yield of 92%. Total synthesis of **2** was achieved via a five-step-reaction, including two direct C-glycosylations from naringenin, in a total yield of 4.4%.

The synthesis of **1** was examined in the same manner as that of **2** (see Scheme 2). Direct C-glycosylation of naringenin with D-xylose gave the desired *C*-β-D-xylosylnaringenin **7** in a 13% yield, along with a 5% yield of 6,8-di-*C*-β-D-xylosylnaringenin **8**. Since the chemical shift of an aromatic proton appeared at 6.75 ppm in the $^1H$ NMR spectrum of the acetate of **7**, the structure of **7** was determined to be 6-*C*-β-D-xylopyranosylnaringenin, as was **3**. Next, a second C-glycosylation of **7** with D-glucose was examined under the same conditions as those for **2**. However, none of the desired 6-*C*-β-D-xylosyl-8-*C*-β-D-glucosylnaringenin **10** was produced. Since this reaction yielded some derivatives of **7**, it was found that **7** was unstable and was converted to spiro-compounds[17] under these harsher reaction conditions. The synthesis of **1** using naringenin as a starting material was, therefore, abandoned.

The synthesis of **1** was changed so that two direct C-glycosylations of phloroacetophenone were the key reactions (Scheme 3). This method can employ a first C-glycosylation with D-glucose and a subsequent C-glycosylation with D-xylose, as well as the synthetic method of **2**. The first C-glycosylation of phloroacetophenone, using D-glucose in the manner described above, gave mono-glycoside **11** in a yield of 48%.[5] The successive second C-glycosylation of **11**[7] with D-xylose gave the desired bis-C-glycoside **12** in a yield of 29%. Three phenolic hydroxyl groups in **12** were protected with a benzyl group to give **13** in a yield of 61%. Aldol condensation of **13** with *p*-benzyloxybenzaldehyde in the presence of sodium methoxide in dried MeOH gave chalcone **14** in a yield of 90%. A methanolic solution of **14** was refluxed in the presence of Dowex 50Wx8 ($H^+$) resin to give flavanone, which was successively O-de-benzylated by hydrogenolysis ($H_2$/10% Pd–C) and acetylated by $Ac_2O$–pyridine–DMAP to afford flavanone acetates (a mixture of **5** and its regioisomer). The mixture of flavanone acetates was oxidized by iodine and pyridine followed by acetylation ($Ac_2O$–pyridine–DMAP) to give the desired di-*C*-glycosylflavone deca-acetates (a mixture of **6** and its regioisomer **15**) in a yield of 70%. The mixture of acetates **6** and **15** was separable by preparative HPLC: however, the separation of a mixture of **1** and **2** was much easier. The mixture of acetates was deprotected by NaOMe followed by Dowex 50Wx8 ($H^+$) resin treatment to afford a mixture of **1** and **2**. The resulting mixture was separated by preparative HPLC (ODS column, 40:60 MeOH–5% AcOH aqueous solution) to give **1** and **2** in a ratio of 62:38. Fortunately, the ratio of the desired **1** was higher. Total yield of the synthesis of **1** from phloroacetophenone was 2.75%, together with a yield of 1.68% of **2**. The $^{13}C$ NMR spectral data for the compounds were in agreement with the data obtained for the natural products[6] (see Table 1).

We also examined the Wessely–Moser isomerization of **2**. Compound **2**, which was synthesized from naringenin, was refluxed in 6 N HCl for 2 h. After removal of the solvents, HPLC analysis (monitoring: UV 254 nm) of the residual solid showed that **1** and **2** were included at 31% and 40%, respectively. These results showed that acid isomerization of **2** synthesized from naringenin via the five-step reaction gave **1** in a yield of 31%.

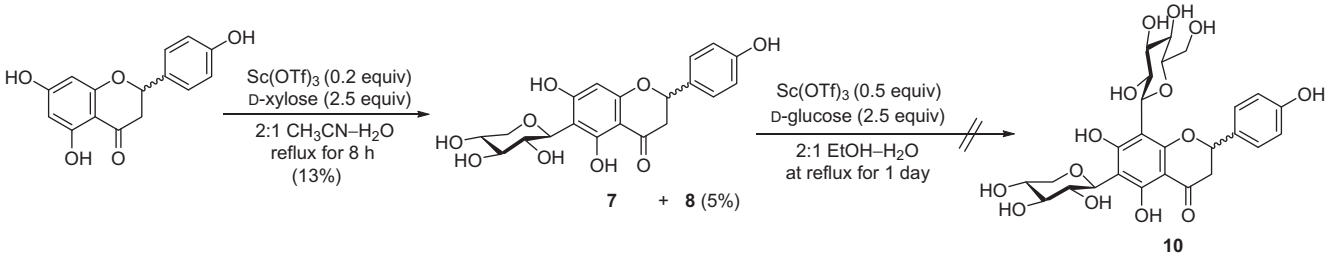
## 3. Conclusions

We accomplished the first total synthesis of 6-*C*-β-D-glucosyl-8-*C*-β-D-xylosyl-4′,5,7-trihydroxyflavone, vicenin-3 (**2**) by a short, five-step reaction involving direct C-glycosylation with D-glucose and D-xylose performed twice, followed by oxidation of the acetate using iodine and pyridine, for a total yield of 4.4%. The first total synthesis of a regioisomer of **2**, vicenin-1 (**1**), was achieved by two direct glycosylations of phloroacetophenone with D-glucose and D-xylose, followed by benzyl protection of the phenolic OH group and aldol condensation, acid-catalyzed cyclization, oxidation of flavanone acetate, deprotection, and HPLC separation, via a total 10-step reaction, in a total yield of 2.75%, along with a vicenin-3 yield of 1.68%.

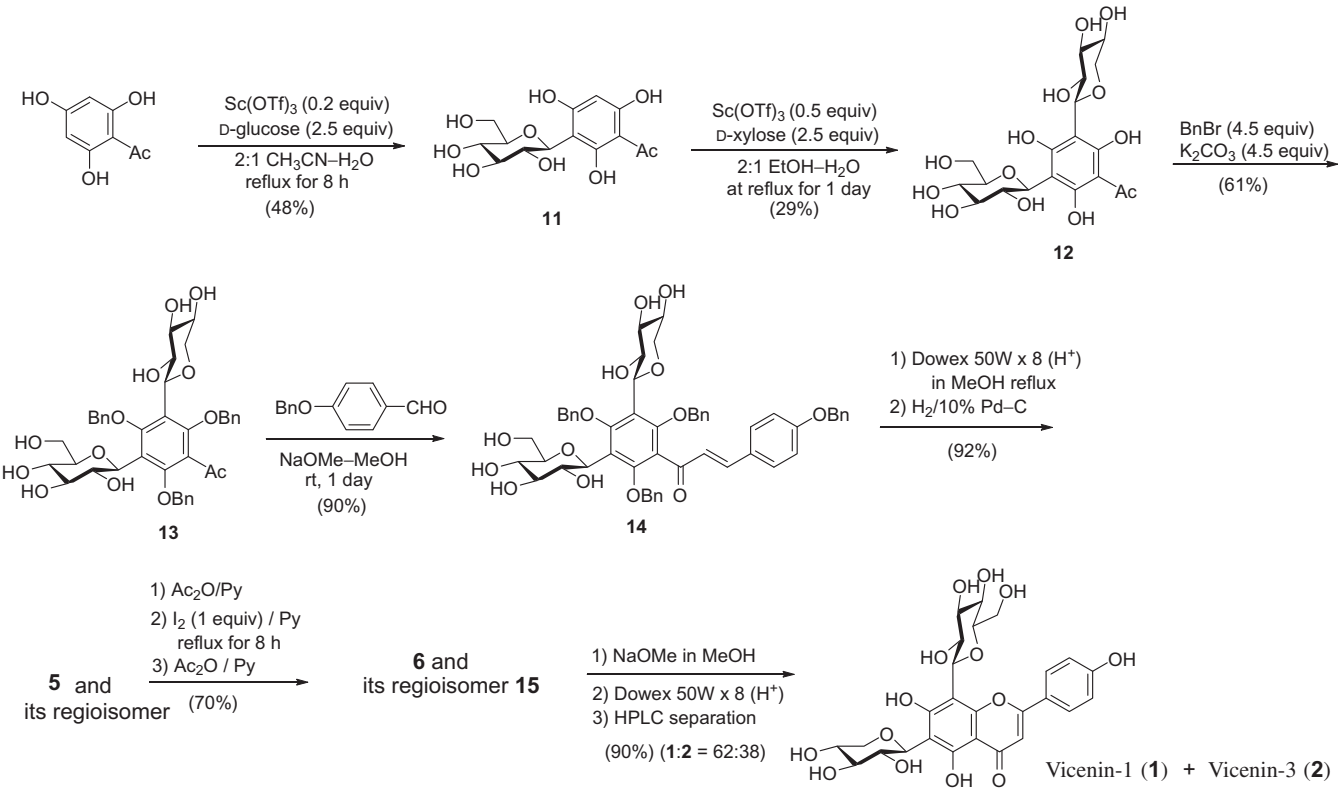
## 4. Experimental

### 4.1. General

$Sc(OTf)_3$ (Taiheiyo Kinzoku Co. Ltd) was purchased and used without any further purification. Reactions were monitored using TLC on 0.25-mm Silica Gel F254 plates (E. Merck), UV light, and a 7% ethanolic solution of phosphomolybdic acid, followed by heat, were used as detection methods. Column chromatography was performed on MCI gel CHP20P® (high porous polymer, 75–150 μm, Mitsubishi Chemical Corp.), and flash column chromatography was performed on silica-gel (40–50 μm, Kanto Reagents Co. Ltd, silica gel 60) to separate and purify reaction products. HPLC analysis and separation were performed using an Inertsil ODS-3 column (GL Science; 5 μm, 4.6 × 250 mm for analytical use, 20 × 250 mm for preparative use; mobile phase: $CH_3CN$ or MeOH–5% AcOH aqueous solution). Optical rotations were recorded on a JASCO DIP-370 polarimeter. IR spectra were recorded on a Horiba FT-720 spectrometer using KBr disks. NMR spectra were recorded on a Varian Inova 500 spectrometer using $Me_4Si$ as the internal standard. Mass spectral data were obtained by fast-atom bombardment (FAB) using *m*-nitrobenzylalcohol (NBA) or glycerol as the matrix on a JEOL JMS-AX505HA instrument. Elemental analyses were performed on a Perkin–Elmer PE 2400 II instrument. After drying at 80–100 °C under reduced pressure for over 2 h, each product was subjected to elemental analysis.




**Scheme 2.** Attempt to synthesize vicenin-3 (**2**) via direct C-xylosylation and C-glucosylation of naringenein.




**Scheme 3.** Synthesis of vicenin-1 (**1**) and -3 (**2**) via two direct C-glycosylations of phloroacetophenone.

**Table 1**
$^{13}$C NMR spectral data for vicenin-1 and -3 (**1** and **2**)

| Compound | 1 | | 2 | |
|---|---|---|---|---|
| Carbon | Natural[a] | Synthetic | Natural[a] | Synthetic |
| 2 | 164.0 | 163.9 | 164.0 | 163.8 |
| 3 | 102.8 | 102.8 | 102.9 | 102.6 |
| 4 | 182.3 | 182.1 | 182.2 | 182.1 |
| 5 | 161.2 | 161.1 | 161.2 | 161.0 |
| 6 | 108.7 | 108.7 | 108.0 | 107.8 |
| 7 | 159.7 | 159.5 | 159.2 | 158.9 |
| 8 | 103.8 | 103.7 | 104.7 | 104.5 |
| 9 | 154.5 | 154.7 | 154.9 | 154.7 |
| 10 | 102.8 | 102.8 | 103.9 | 103.6 |
| 1′ | 121.8 | 121.5 | 121.8 | 121.5 |
| 2′ | 128.7 | 128.5 | 128.5 | 128.3 |
| 3′ | 116.0 | 115.9 | 116.0 | 115.8 |
| 4′ | 161.3 | 160.0 | 161.3 | 161.1 |
| 5′ | 116.0 | 115.9 | 116.1 | 115.8 |
| 6′ | 128.7 | 128.5 | 128.5 | 128.3 |
| G1 | 72.0 | 72.1 | 74.2 | 73.9 |
| G2 | 71.0 | 71.3 | 70.6 | 70.4 |
| G3 | 79.2 | 79.5 | 79.2 | 78.9 |
| G4 | 70.2 | 69.7 | 70.2 | 70.0 |
| G5 | 81.5 | 81.8 | 81.3 | 81.0 |
| G6 | 60.9 | 60.5 | 60.6 | 60.4 |
| X1 | 74.6 | 74.1 | 75.0 | 74.7 |
| X2 | 70.4 | 70.2 | 71.9 | 71.6 |
| X3 | 78.6 | 78.8 | 78.4 | 78.1 |
| X4 | 70.4 | 70.2 | 71.5 | 71.2 |
| X5 | 70.0 | 69.3 | 69.9 | 69.6 |

[a] Measured at 90 °C.

### 4.2. 6-*C*-β-D-Glucopyranosylnaringenin (3)

A solution of naringenin (500 mg, 1.83 mmol) and D-glucose (823 mg, 4.57 mmol) in 2:1 $CH_3CN$–$H_2O$ (50 mL) was refluxed in an oil bath for 12 h in the presence of $Sc(OTf)_3$ (180 mg, 0.36 mmol). The reaction mixture was diluted with $H_2O$ (50 mL) and passed through a column of MCI GEL CHP20P® (2.5 × 100 mm) loaded with water, and the gel was washed with 200 mL of water and eluted with 100 mL of 50% aq acetone and 100 mL of 1:1 acetone–MeOH. The combined eluate was evaporated to give a pale-yellow crude product, which was purified by silica-gel column chromatography (15:30:2:0.1 acetone–AcOEt–$H_2O$–AcOH) to give **3** (175 mg, 22%) as a pale-yellow amorphous powder. Product **3** was an inseparable 1:1 diastereomeric mixture, with rotamers observed by $^1$H NMR spectroscopy.

IR (KBr) $\nu$ 3400, 2898, 1716, 1643, 1616, 1519, 1458 cm$^{-1}$. $^1$H NMR (DMSO-$d_6$) $\delta$ 2.70 (1H, dd, $J$ 17.2, 3.1 Hz, H3a), 3.06 (1H, t, $J$ 9.2 Hz, H3′), 3.09 (1H, ddd, $J$ 1.6, 6.1, 8.5 Hz, H5′), 3.15 (1H, t, $J$ 8.5 Hz, H4′), 3.23 (1H, dd, $J$ 12.6, 17.1 Hz, H3b), 3.36 (1H, dd, $J$ 6.1, 11.7 Hz, H6′a), 3.64 (1H, d, $J$ 11.5 Hz, H6′b), 3.95 (1H, br. t, H2′), 4.46 (1H, $J$ 9.8 Hz, H1′), 4.45 (1H, br s, OH), 4.58 (1H, br t, 6′-OH), 4.83 (2H, br s, OH × 2), 5.40 (1H, dd, $J$ 12.4, 2.3 Hz, H2), 5.93 (1H, s, H8), 6.78 (2H, d, $J$ 8.4, H3′,5′), 7.30 (2H, d, $J$ 8.4 Hz, H2′,6′), 9.58, 10.5, 12.7 (each 3H, s, br s × 2, PhOH × 3). FABMS (negative-ion, $m/z$): 433 $(M-H)^-$. Anal. Calcd for $C_{21}H_{22}O_{10}\cdot 0.75H_2O$: C, 56.31; H, 5.29. Found: C, 56.33; H, 5.52.

### 4.3. 6-*C*-β-D-Glucopyranosyl-8-*C*-β-D-xylopyranosylnaringenin (4)

A solution of **3**, D-xylose, and $Sc(OTf)_3$ in 2:1 EtOH–$H_2O$ (4 mL) was stirred at 80 °C for one day. The reaction mixture was separated and purified in the same manner as that used for **3** to give **4** (78 mg, 30%) as a pale-yellow amorphous powder. Product **4** was an inseparable 1:1 diastereomeric mixture, with rotamers observed by $^1$H NMR spectroscopy.

IR (KBr) $\nu$ 3363, 2917, 1626, 1518, 1458 cm$^{-1}$. $^1$H NMR (DMSO-$d_6$) $\delta$ 2.80–2.86 (1H, m, H3a), 2.98–3.14 (3H, m), 3.26 (3H, m), 3.50–3.88 (4H, m), 4.47 (1H, m), 4.60–4.83 (4H, m, OH × 4), 4.90 (1H, m),

4.88–5.02 (3H, OH × 3), 5.36 (0.5H, dd, *J* 2.4, 12.9, H3″), 5.47 (0.5H, dd, *J* 2.8, 12.8, H3″), 6.78 (2H, m, *J* 8.5, H3′,5′), 7.32 (2H, m, *J* 8.5, H2′,6′), 9.25 (1H, br. s, 4′-OH), 9.53 and 9.56 (each 0.5H, s, 7-OH), 12.76 and 12.77 (each 0.5H, s, 5-OH). FABMS (negative-ion, *m*/*z*) 565 $(M-H)^-$. Anal. Calcd for $C_{26}H_{30}O_{14}\cdot 0.5H_2O$: C, 54.25; H, 5.44. Found: C, 54.45; H, 5.66.

### 4.4. 6-*C*-β-D-Glucopyranosyl-8-*C*-β-D-xylopyranosylnaringenin deca-acetate (5)

Flavanone **3** (100 mg, 0.17 mmol) was dissolved in pyridine (0.5 mL) and $Ac_2O$ (0.5 mL), and DMAP (ca. 20 mg) was added to the mixture. The mixture was stirred at room temperature for 16 h. The reaction mixture was poured into ice-cold water (30 mL) and stirred for 0.5 h, and then extracted with AcOEt (10 mL × 2). The combined organic layer was washed with water and brine and dried over $Na_2SO_4$. After removal of the organic solvent, the residual solid was purified by silica-gel column chromatography (2:1 AcOEt–*n*-hexane) to give **5** (139 mg, 89%) as a colorless amorphous powder.

IR (KBr) *ν* 2943, 1766, 1757, 1656, 1604 $cm^{-1}$. $^1H$ NMR ($CDCl_3$) *δ* aglycon moiety: 2.32, 2.44, and 2.50 (each 3H, s, ArOAc × 3), 2.79 (1H, dd, *J* 2.7, 16.8, H3a), 2.95 (1H, dd, *J* 14.4, 16.8, H3b), 5.74 (1H, dd, *J* 2.7, 14.4, H2), 7.23 (2H, d, H3′,5′), 7.58 (2H, d, *J* 8.6, H2′,6′), xylose moiety: 3.27 (1H, t, *J* 10.8, H5a), 3.94 (1H, d, *J* 9.4, H1), 4.25 (1H, t, dd, *J* 5.6, 10.8, H5b), 5.26 (2H, t, *J* 9.3, H3,4), 5.61 (1H, t, *J* 9.4, H2), glucose moiety: 3.77 (1H, dt, *J* 4.5, 9.5, H5), 4.25 (1H, dd, *J* 12.5, 4.5, H6a), 4.43 (1H, dd, *J* 12.5, 4.5, H6b), 4.72 (1H, d, *J* 9.5, H1), 5.61 (1H, t, *J* 9.5, H3), 5.81 (1H, t, *J* 9.5, H2). 1.78, 1.91, 1.99, 2.02, 2.03, 2.04, 2.07 (each 3H, s, OAc × 7). FABMS (positive-ion, *m*/*z*) 987 $(M+H)^+$. Anal. Calcd for $C_{46}H_{50}O_{24}$: C, 55.98; H, 5.11. Found: C, 55.83; H, 5.12.

### 4.5. 6-*C*-β-D-Glucopyranosyl-8-*C*-β-D-xylopyranosyl-4′,5,7-trihydroxyflavone deca-acetate (6)

A solution of **5** (50 mg, 0.05 mmol) and iodine (12.6 mg, 0.05 mmol) in pyridine (2 mL) was refluxed for 8 h. The reaction mixture was allowed to cool to room temperature, and it was then filtered. The filtrate was evaporated in vacuo. The residual solid was re-acetylated in the same manner as for the acetylation of **4**. The acetylated product was purified by silica-gel column chromatography (2:1 AcOEt–*n*-hexane) to give **6** (45 mg, 91%) as a pale-yellow amorphous powder. $[\alpha]_D^{22}$ −5.19 (*c* 0.385, MeOH). IR (KBr) *ν* 2950, 2864, 1782, 1743, 1652, 1600 $cm^{-1}$. $^1H$ NMR ($CDCl_3$) *δ* aglycon moiety: 2.35, 2.49, 2.56 (each 3H, s, ArOAc × 3), 6.60 (1H, s, H3), 7.37 (2H, d, *J* 8.8, H3′,5′), 7.99 (2H, d, H2′,6′), xylose moiety: 3.41 (1H, t, *J* 11.1, H5a), 4.39 (1H, dd, *J* 11.1, 5.6, H5b), 4.50 (1H, d, *J* 9.8, H1), 5.23 (1H, ddd, *J* 9.5, 5.6, 11.1, H4), 5.41 (1H, t, *J* 9.5, H3), 5.68 (1H, t, *J* 9.5, H2), glucose moiety: 3.80 (1H, dd, *J* 9.8, 4.7, H5), 3.95 (1H, d, *J* 12.9, H6a), 4.45 (1H, dd, *J* 4.7, 12.9, H6b), 4.84 (1H, *J* 9.8, H1), 5.16 (1H, dt, *J* 9.8, H4), 5.30 (1H, t, *J* 9.8, H3), 5.71 (1H, t, *J* 9.8, H2), 1.76, 1.88, 2.01, 2.02, 2.05, 2.07, 2.10 (each 3H, s, OAc × 7). FABMS (positive-ion, *m*/*z*) 985 $(M+H)^+$. Anal. Calcd for $C_{46}H_{48}O_{24}$: C, 56.10; H, 4.91. Found: C, 55.89; H, 5.13.

### 4.6. 6-*C*-β-D-Glucopyranosyl-8-*C*-β-D-xylopyranosyl-4′,5,7-trihydroxyflavone, vicenin-3 (2)

To a stirred solution of **6** (40 mg, 0.04 mmol) in dry MeOH (1 mL), a 25% NaOMe methanolic solution (0.2 mL) was added dropwise at room temperature, and the mixture was stirred for 1 h. Dowex 50Wx8 ($H^+$) resin was added to the stirred reaction mixture until the solution pH was neutral. The mixture was filtered and the filtrate was evaporated in vacuo to give **2** (21.1 mg, 91%) as a yellow amorphous powder. $^1H$ NMR spectroscopy of products **2**, **13**, **14**, and **1** was carried out at 80 °C or at 120 °C in DMSO-$d_6$ because rotamers were not observable.

$[\alpha]_D^{22}$ +23.3 (*c* 0.275, MeOH). IR (KBr) *ν* 3392, 2923, 1653, 1575 $cm^{-1}$. $^1H$ NMR (DMSO-$d_6$ + $D_2O$, at 80 °C) *δ* 3.24 (1H, t, *J* 10.7), 3.30 (1H, t, *J* 8.6), 3.61 (1H, dd, *J* 3.8, 12.5), 3.69 (1H, dd, *J* 1.6, 12.2), 3.89 (1H, t, *J* 9.2, 9.4), 3.93 (1H, dd, *J* 5.3, 11.1), 4.77 (1H, d, *J* 9.8, H1′), 4.81 (1H, *J* 9.8, H1), 6.71 (1H, s, H3), 9.17 (1H, br s, 7-OH), 10.15 (1H, br s, 4′-OH), 13.64 (1H, s, 5-OH). FABMS (positive-ion, *m*/*z*) 565 $(M+H)^+$, (negative-ion, *m*/*z*) 563 $(M-H)^-$. Anal. Calcd for $C_{26}H_{28}O_{14}\cdot 2.5H_2O$: C, 51.22; H, 5.47. Found: C, 51.19; H, 5.14.

### 4.7. 6-*C*-β-D-Xylopyranosylnaringenin (7) and 6,8-di-*C*-β-D-xylopyranosylnaringenin (8)

Synthesis and purification were carried out in the same manner as that used for **3** to give **7** (13%) and **8** (5%) as pale-yellow amorphous powders. Products **7** and **8** were an inseparable 1;1 diastereomeric mixtures, with rotamers observed by $^1H$ NMR spectroscopy.

#### 4.7.1. Data for 7

IR (KBr) *ν* 3352, 2914, 1641, 1516, 1462 $cm^{-1}$. $^1H$ NMR (DMSO-$d_6$) *δ* 2.68 (1H, dd, *J* 2.9, 17.1, H3a), 3.01 (1H, t, *J* 10.6, H5″a), 3.09 (1H, t, *J* 8.4, H3″), 3.24 (1H, dd, *J* 12.9, 17.1, H3b), 3.33 (1H, ddd, *J* 4.9, 5.2, 10.6, H4″), 3.70 (1H, dd, *J* 5.2, 10.6, H5″b), 4.37 (1H, d, *J* 9.8, H1″), 4.56 (1H, br s, 2-OH), 4.84 (1H, br s, 3-OH), 4.87 (1H, d, *J* 4.9, 4-OH), 5.39 (1H, dd, *J* 2.9, 12.7, H2), 5.92 (1H, s, H8), 6.78 (2H, d, *J* 8.5, H3′,5′), 7.30 (2H, d, *J* 8.5, H2′,6′), 9.60 (1H, s, 7-OH), 10.69 (1H, br s, 4′-OH), 12.72 (1H, s, 5-OH). FABMS (positive-ion, *m*/*z*) 405 $(M+H)^+$. Anal. Calcd for $C_{20}H_{20}O_9\cdot 0.25H_2O$: C, 58.75; H, 5.05. Found: C, 58.85; H, 4.92.

#### 4.7.2. Data for 8

IR (KBr) *ν* 3396, 2910, 2871, 1628, 1516, 1458 $cm^{-1}$. $^1H$ NMR (DMSO-$d_6$) *δ* 2.82 (1H, dt, *J* 3.0, 17.3, H3a), 3.02–3.16 (5H, m), 3.43 (1H, m), 3.71–3.79 (2H, m), 4.47–4.53 (2H, m), 4.66–4.84 (2H, m, OH × 2), 4.91–4.96 (4H, m), 5.35 (0.5H, dd, *J* 2.9, 12.7, H3″), 5.45 (0.5H, dd, *J* 2.8, 12.8, H3″), 6.77 and 6.78 (each 1H, d, *J* 8.5, H3′,5′), 7.32 (2H, m, *J* 8.5, H2′,6′), 9.07 (1H, br s, 4′-OH), 9.53 and 9.56 (each 0.5H, s, 7-OH), 12.80 and 12.81 (each 0.5H, s, H5). FABMS (positive-ion, *m*/*z*) 537 $(M+H)^+$. Anal. Calcd for $C_{25}H_{28}O_{13}\cdot 2H_2O$: C, 52.44; H, 5.63. Found: C, 52.60; H, 5.83.

### 4.8. 6-*C*-β-D-Xylopyranosylnaringenin hexa-acetate (9)

Naringenin mono-C-glycoside **7** (100 mg) was dissolved in pyridine (2 mL), and $Ac_2O$ (2 mL) and the mixture was stirred at room temperature for 16 h. The reaction mixture was poured into ice-cold water and stirred for 0.5 h and then twice extracted with AcOEt. The organic layer was washed with water and brine, and then dried over anhyd $Na_2SO_4$. After evaporation, the residue was purified by silica-gel column chromatography (60:1 $CHCl_3$–EtOH) to afford acetate **9** (151 mg, 93%) as a pale-yellow amorphous solid. IR (KBr) *ν* 2949, 2860, 1778, 1751, 1693, 1618 $cm^{-1}$. $^1H$ NMR ($CDCl_3$) *δ* 1.84 and 1.86 (3H, each s, OAc), 2.04 and 2.06 (each 3H, s, OAc × 2), 2.31 and 2.38 (each 3H, s, OAc × 2), 2.43 and 2.44 (3H, each s, OAc), 2.75 (1H, dd, *J* 2.6 and 16.6 Hz, H-3a), 3.01 (1H, m, H-3b), 3.36 (1H, m, H-5″a), 4.13(1H, d, *J* 9.5 Hz, H1″), 4.62 (1H, m, H-5″b), 5.02 (1H, t, *J* 9.5 Hz, H-4″), 5.25 (1H, t, *J* 9.5 Hz, H-3″), 5.47 (1H, m, H-2), 5.57 (1H, t, *J* 9.5 Hz, H-2″), 6.77 and 6.78 (1H, s, H-8), 7.15 (2H, d, *J* 8.6 Hz, H-3′,5′), 7.44 (2H, d, *J* 8.6 Hz, H2′,5′). FABMS (positive-ion, *m*/*z*) 657 $(M+H)^+$. Anal. Calcd for $C_{32}H_{32}O_{15}\cdot 0.1CHCl_3$: C, 57.67; H, 4.84. Found: C, 57.75; H, 4.70.

### 4.9. 3,5-Di-*C*-β-D-(xylopyranosyl-glucopyranosyl)phloroacetophenone (12)

Synthesis and purification were performed in the same manner as that used for **4**: reaction solvent system, 2:1 EtOH–$H_2O$; refluxing time, one day. $[\alpha]_D^{22}$ +87.6 (*c* 1.005, MeOH). IR (KBr) *v* 3369, 2923, 1701, 1622 $cm^{-1}$. $^1H$ NMR (DMSO-$d_6$) $\delta$ 2.58 (3H, s, Ac), 9.07 (1H, br s, OH), 11.50 (1H, s, OH), 12.0 (1H, br s, OH), (xylose moiety) 3.11 (t, *J* 10.8, H5a), 3.16 (1H, t, *J* 8.9, H3), 3.45 (1H, ddd, *J* 10.8, 8.9, 5.6, H4), 3.59 (1H, t, *J* 8.9, H2), 3.84 (1H, dd, *J* 5.6, 10.8, H5b), 4.72 (1H, d, *J* 9.8, H1), glucose moiety: 3.25 (1H, t, *J* 8.0, H3), 3.26 (1H, m, H6a), 3.32 (1H, t, *J* 9.0, H4), 3.43 (1H, m, H5), 3.59 (1H, m, H6b), 4.56 (1H, d, *J* 9.6, H1), 4.74 (1H, br t, 6-OH), 4.93 (1H, d, OH), 4.99 (2H, br t, OH × 2), 5.04 (1H, d, OH). $^{13}C$ NMR (DMSO-$d_6$) $\delta$ sugar moiety: 60.03, 69.22, 69.73, 70.63, 71.70, 72.57, 75.10, 75.61, 77.76, 78.48, 81.29; 104.03, 104.50, and 105.19 (C2, 4, 6), 161.7 (br), 161.06, and 161.58 (C1, 3, 5), 33.31 and 203.85 (Ac). FABMS (negative-ion, *m*/*z*) 461 $(M-H)^-$. Anal. Calcd for $C_{19}H_{26}O_{13}\cdot H_2O$: C, 47.49; H, 5.89. Found: C, 47.19; H, 5.52.

### 4.10. 2,4,6-Tri-*O*-benzyl-3,5-di-*C*-β-D-(xylopyranosyl-glucopyranosyl)phloroacetophenone (13)

$[\alpha]_D^{22}$ −27.4 (*c* 0.540, MeOH). IR (KBr) *v* 3400, 2921, 2883, 1701, 1577 $cm^{-1}$. $^1H$ NMR (DMSO-$d_6$ + $D_2O$, at 120 °C) $\delta$ 2.47 (3H, s, Ac), 4.75 (2H, d, *J* 10.7, $PhCH_2$), 4.84 and 5.13 (each 1H, d, *J* 10.2 and 10.7, $PhCH_2$), 5.27 (2H, br s, $PhCH_2$), glucose moiety: 3.17 (1H, t, *J* 8.8, H4), 3.20 (1H, t, *J* 8.3 H3), 3.21 (1H, m, H5), 3.49 (1H, dd, *J* 5.8, 11.5, H6a), 3.72 (1H, dd, *J* 2.0, 11.7, H6b), 4.24 (1H, t, *J* 8.8, 9.3, H2), 4.68 (1H, d, *J* 9.7, H1), xylose moiety: 3.08 (1H, t, *J* 11.0, 10.5, H5a), 3.14 (1H, t, *J* 9.0, H3), 3.32 (1H, ddd, *J* 5.3, 9.5, 10.0, H4), 3.87 (1H, dd, *J* 5.4, 11.0, H5b), 4.15 (1H, t, *J* 9.0, 9.2, H2), 4.60 (1H, d, *J* 9.7, H1). FABMS (positive-ion, *m*/*z*) 733 $(M+H)^+$. Anal. Calcd for $C_{40}H_{44}O_{13}\cdot 0.5H_2O$: C, 64.76; H, 6.13. Found: C, 64.71; H, 6.24.

### 4.11. 2,4,6-Tri-*O*-benzyl-3,5-di-*C*-β-D-(xylopyranosyl-glucopyranosyl)-1-[3-(*p*-benzyloxyphenyl)propenoyl]benzene (14)

$[\alpha]_D^{22}$ −22.9 (*c* 0.515, MeOH). IR (KBr) *v* 3400, 2921, 2879, 1624, 1595, 1577, 1508 $cm^{-1}$. $^1H$ NMR (DMSO-$d_6$ + $D_2O$, at 120 °C) $\delta$ 4.77 (2H, d, *J* 9.8, $PhCH_2$), 4.88 and 5.10 (each 1H, d, *J* 10.7, $PhCH_2$), 5.16 (2H, s, 4′-$PhCH_2$), 5.30 (2H, br s, $PhCH_2$), 6.99 (1H, d, *J* 16.1, *trans*-vinyl H), 7.03 (1H, 2H, d, *J* 8.8, H3′,5′), 7.45 (1H, m, *trans*-vinyl H), 7.55 (1H, 2H, d, *J* 8.8, H2′,6′), 7.26–7.57 (20H, m, ArH), glucose moiety: 3.16 (1H, t, *J* 9.2, H4), 3.23 (1H, m, H5), 3.23 (1H, t, *J* 9.5, H3), 3.50 (1H, dd, *J* 5.3, 11.9, H6a), 3.73 (1H, dd, *J* 1.7, 11.9, H6b), 4.25 (1H, t, *J* 9.2, H2), 4.71 (1H, d, *J* 9.7, H1), xylose moiety: 3.10 (1H, t, *J* 10.6, H5a), 3.17 (1H, t, *J* 9.0, H3), 3.32 (1H, ddd, *J* 5.1, 10.0, 10.0, H4), 3.88 (1H, dd, *J* 5.3, 10.9, H5b), 4.16 (1H, t, *J* 9.0, H2), 4.63 (1H, d, *J* 9.7, H1). FABMS (positive-ion, *m*/*z*) 927 $(M+H)^+$. Anal. Calcd for $C_{54}H_{54}O_{14}\cdot 2H_2O$: C, 67.34; H, 6.08. Found: C, 67.58; H, 6.36.

### 4.12. 6-*C*-β-D-Xylopyranosyl-8-*C*-β-D-glucopyranosyl-4′,5,7-trihydroxyflavone deca-acetate (15)

$[\alpha]_D^{19}$ +16.6 (*c* 0.650, MeOH). IR (KBr) *v* 2943, 2866, 1755, 1653, 1604, 1508 $cm^{-1}$. $^1H$ NMR ($CDCl_3$) $\delta$ aglycon moiety: 2.35, 2.48, 2.50 (each 3H, s, ArOAc × 3), 6.66 (1H, s, H3), 7.39 (2H, d, *J* 8.8, H3′,5′), 8.07 (2H, d, H2′,6′), glucose moiety: 3.77 (1H, ddd, *J* 9.5, 4.0, 2.0, H5), 4.20 (1H, dd, *J* 12.7, 2.0, H6a), 4.28 (1H, dd, *J* 4.0, 12.7, H6b), 4.60 (1H, *J* 10.0, H1), 5.42 (1H, t, *J* 9.5, H3), 5.47 (1H, t, *J* 9.5, H4), 5.74 (1H, t, *J* 9.5, H2), xylose moiety: 3.41 (1H, t, *J* 11.0, H5a), 4.18 (1H, dd, *J* 11.0, 5.6, H5b), 4.73 (1H, d, *J* 9.3, H1), 5.04 (1H, ddd, *J* 9.6, 5.6, 11.1, H4), 5.31 (1H, t, *J* 9.6, H3), 5.62 (1H, t, *J* 9.6, H2), 1.76, 1.88, 1.93, 1.99, 2.05, 2.08, 2.10 (each 3H, s, OAc × 7). FABMS (positive-ion, *m*/*z*) 985 $(M+H)^+$. Anal. Calcd for $C_{46}H_{48}O_{24}$: C, 56.10; H, 4.91. Found: C, 55.99; H, 4.83.

### 4.13. 6-*C*-β-D-Xylopyranosyl-8-*C*-β-D-glucopyranosyl-4′,5,7-trihydroxyflavone, vicenin-1 (1)

$[\alpha]_D^{22}$ +41.3 (*c* 0.450, MeOH). IR (KBr) *v* 3367, 2921, 1653, 1575 $cm^{-1}$. $^1H$ NMR (DMSO-$d_6$ + $D_2O$, at 80 °C) $\delta$ 3.64 (1H, br d, *J* 12.0), 3.91 (1H, dd, *J* 5.3, 10.9), 4.65 (1H, d, *J* 8.8), 4.78 (1H, dd, *J* 9.8, 14.7), 6.69 (1H, s, H3), 6.93 (2H, d, *J* 8.5, 3′,5′-H), 7.93 (2H, d, *J* 8.5 Hz, H-2′,6′), 9.19 (1H, br s, 7-OH), 10.12 (1H, br s, 4′-OH), 13.67 (1H, s, 5′-OH). FABMS (positive-ion, *m*/*z*) 565 $(M+H)^+$. Anal. Calcd for $C_{26}H_{28}O_{14}\cdot 1.2H_2O$: C, 53.27; H, 5.24. Found: C, 53.04; H, 5.26.

## References


1. Maurice, J. *C*-Glycosylflavonoids. In *The Flavonoids*; Harborne, J. B., Ed.; Chapman and Hall: London, 1994; pp 57–93.
2. (a) Zavodnik, L. B. *Radiat. Biol. Radiol.* **2003**, *43*, 432–438; (b) Zavodnik, L. B.; Zavodnik, I. B.; Lapshina, E. A.; Shkodich, A. P.; Bryszewska, M.; Buko, V. V. *Biochemistry (Moscow)* **2000**, *65*, 946–951; (c) Lin, R. C.; Li, T. K. *Am. J. Clin. Nutr.* **1998**, *68*, 1512S; (d) Kawaguchi, K.; Melloalves, S.; Watanabe, T.; Kikuchi, S.; Satake, M.; Kumazawa, Y. *Planta Med.* **1998**, *329*, 855–859; (e) Matsubara, Y.; Suekuni, H.; Honda, S.; Kakehi, K.; Murakami, T.; Okamoto, K.; Miyake, H. *Jpn. Heart J.* **1980**, *21*, 583; (f) Iizuka, Y.; Murakami, T.; Matsubara, Y.; Yokoi, K.; Okamoto, K.; Yokoi, K. *Agric. Biol. Chem.* **1986**, *50*, 781; (g) Matsubara, Y.; Sawabe, A. *J. Synth. Org. Chem. Jpn.* **1994**, *52*, 318–327; (h) Kawasaki, M.; Hayashi, T.; Arisawa, M.; Morita, N.; Berganza, L. H. *Phytochemistry* **1988**, *27*, 3709–3711; (i) Ohsugi, T.; Nishida, R.; Fukami, H. *Agric. Biol. Chem.* **1985**, *49*, 1897–1900.
3. (a) Frick, W.; Schmidt, R. R. *Liebigs Ann. Chem.* **1989**, 565–570; (b) Mahling, J.-A.; Jung, K.-H.; Schmidt, R. R. *Liebigs Ann.* **1995**, 461–466; (c) Kumazawa, T.; Ohki, K.; Ishida, M.; Sato, S.; Onodera, J.-I.; Matsuba, S. *Bull. Chem. Soc. Jpn.* **1995**, *68*, 1379–1384; (d) Kumazawa, T.; Minatogawa, T.; Matsuba, S.; Sato, S.; Onodera, J.-i. *Carbohydr. Res.* **2000**, *329*, 507–513; (e) Kumazawa, T.; Kimura, T.; Matsuba, S.; Sato, S.; Onodera, J.-i. *Carbohydr. Res.* **2001**, *334*, 183–193; (f) Lee, D. Y. W.; Zhang, W.-Y.; Karnati, V. V. R. *Tetrahedron Lett.* **2003**, *44*, 6857–6859; (g) Oyama, K.-i.; Kondo, T. *J. Org. Chem.* **2004**, *69*, 5240–5246; (h) Sato, S.; Hiroe, K.; Kumazawa, T.; Onodera, J.-i. *Carbohydr. Res.* **2006**, *341*, 1091–1095.
4. Sato, S.; Akiya, T.; Nishizawa, H.; Suzuki, T. *Carbohydr. Res.* **2006**, *341*, 964–970.
5. Sato, S.; Akiya, T.; Suzuki, T.; Onodera, J.-i. *Carbohydr. Res.* **2004**, *339*, 2611–2614.
6. Yasukawa, K.; Kaneko, T.; Yamanouchi, S.; Takido, M. *Yakugaku Zasshi* **1986**, *106*, 517–519.
7. Seikel, M. K.; Chow, J. H. S.; Felman, L. *Phytochemistry* **1966**, *5*, 439–455.
8. Jay, M.; Ismaili, A. *Phytochemistry* **1989**, *28*, 3035–3037.
9. Johnson, R. H.; Wallace, J. W. *Biochem. Syst. Ecol.* **1988**, *16*, 521–523.
10. Shammas, G.; Couladi, M. *Sci. Pharm.* **1988**, *56*, 277–281.
11. Porter, P. L.; Wallace, J. W. *Biochem. Syst. Ecol.* **1988**, *16*, 261–262.
12. Seetharamamma, B.; Rao, C. V.; Gunasekar, D. *Indian J. Nat. Prod.* **1989**, *5*, 22.
13. Chaboud, A.; Raynaud, J.; Dellamonica, G. *Pharm. Acta Helv.* **1989**, *64*, 16–18.
14. Purushothaman, K. K.; Vasanth, S. *Indian Drugs* **1986**, *23*, 482.
15. Hillis, W. E.; Horn, D. H. S. *Aust. J. Chem.* **1965**, *18*, 531–542.
16. (a) Voigtlaender, H.-W.; Haertner, H. *Arch. Pharm.* **1983**, *316*, 3, 219–222; (b) Lee, Y.-J.; Wu, T.-D. *J. Chin. Chem. Soc.* **2001**, *48*, 201–206.
17. (a) Kumazawa, T.; Assahi, N.; Matsuba, S.; Sato, S.; Furuhata, K.; Onodera, J.-i. *Carbohydr. Res.* **1998**, *308*, 213–216; (b) Kumazawa, T.; Chiba, M.; Matsuba, S.; Sato, S.; Onodera, J.-i. *Carbohydr. Res.* **2000**, *328*, 599–603; (c) Sato, S.; Kumazawa, T.; Watanabe, K.-i.; Matsuba, S.; Onodera, J.-i. *Carbohydr. Res.* **2004**, *339*, 429–433; (d) Sato, S.; Miura, M.; Sekito, T.; Kumazawa, T. *J. Carbohydr. Chem.* **2008**, *27*, 86–102.