Total Synthesis of Two Isoflavone Bis-*C*-glycosides: Genistein and Orobol 6,8-Di-*C*-β-D-glucopyranosides

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Abstract: This paper describes the first successful synthesis of two isoflavone bis-*C*-glycosides, genistein and orobol 6,8-di-*C*-β-D-glucopyranosides from phloroacetophenone bis-*C*-glycoside in total yields of 27 and 19%, respectively, using a four-step reaction: direct C-glycosylation of phloroacetophenone with unprotected D-glucose; chalcone synthesis by aldol condensation; acetal synthesis by oxidative rearrangement using DIB and *p*-TsOH; and formation of the isoflavone ring using HCl, without protection of the glucose moiety.

Key words: bis-*C*-glycoside, isoflavone, DIB, *p*-toluenesulfonic acid, oxidative rearrangement

Many plant flavonoids are present in their glycoside forms. The most common flavonoid glycosides in plants are the *O*-glycosides, while the *C*-glycosides are rare. J. Maurice wrote in his review of the 1994 publication in 'The Flavonoids' edited by J. B. Harborne:[1] 'Among the *C*-glycosylflavonoids, there are some mono-*C*-glycosides and a few bis-*C*-glycosides. Most of the *C*-glycosylflavonoids are flavones, and 55 di-*C*-glycosylflavones have been identified. Regarding the glycosylisoflavones, 10 mono-*C*-glycosylisoflavones have been isolated. The structures of these compounds were subsequently elucidated, revealing D-glucose or D-rhamnose sugar residues. Further, only 2 di-*C*-glycosylisoflavones have been isolated and their structures elucidated. One was isolated from *Dalbergia paniculata* (Leg.) bark, its structure was determined to be 4′,5,7-trihydroxy-6,8-di-*C*-β-D-glucopyranosylisoflavone, 6,8-di-*C*-β-D-glucosylgenistein (**1**).[2] The other was isolated from *Dalbergia nitidula* (Leg.) bark, and its structure was determined to be 3′,4′,5,7-tetrahydroxy-6,8-di-*C*-β-D-glucopyranosylisoflavone, 6,8-di-*C*-β-D-glucosylorobol (**2**).[3] Both compounds were also isolated from *Dalbergia monetaria* (Leg.).'[1,4] Reportedly, the bioactivity of the mono-*C*-glycosides differs from that of the *O*-glycosides and aglycones.[5] However, there are no reports regarding the bioactivity of the isoflavone bis-*C*-glycosides. Because the flavonoid bis-*C*-glycosides have interesting bioactivities,[6] the isoflavone bis-*C*-glycosides are expected to also possess bioactivity.

Regarding the synthesis of isoflavone *C*-glycosides, the effective total synthesis of mono-*C*-glycoside in a reasonable yield has been described in two previous studies, with the exception of a low-yield synthesis using an acetobromosugar under basic conditions.[1b] Lee et al. reported the synthesis of puerarin,[7] while our group described the synthesis of genistein and orobol *C*-glycosides.[8] In both methods, all the reactions were carried out after protecting all hydroxy groups of both the sugar and phenol residues, and the key oxidative rearrangement reaction employed the highly reactive thallium(III) nitrate (TTN) as an oxidant. Our previous report on the synthesis of flavone bis-*C*-glycosides[9] is the only published study on the topic. In the present study, the total synthesis of two naturally occurring isoflavone (genistein and orobol) bis-*C*-glycosides **1** and **2** is presented (Scheme 1). An environmentally friendly reaction scheme using diacetoxyiodobenzene (DIB) and *p*-toluenesulfonic acid in place of the volatile oxidant TTN[10a,c] and with only benzyl ether as the protecting group at the phenol hydroxy group of the aglycone moiety was developed. Aldol condensation of 2,4,6-tri-*O*-benzyl-3,6-di-*C*-glucosylphloroacetophenone (**3**)[9] with 4-hydroxybenzaldehyde does not proceed without protection of the phenol hydroxy groups, and the subsequent oxidative rearrangement of chalcones **4**[9] and **5** cannot proceed without protection of the 2′,6′-hydroxy groups of chalcone.[10e] Further, deprotection of benzyl ether of the sugar hydroxy groups by hydrogenolysis led to a lowering of the yield owing to a reduction of the alkene.[8] Though Kawamura and co-workers[10e] chose a benzoyl group as the protecting group for the 2′-hydroxy of the chalcone, we chose a benzyl group because it does not deprotect during aldol condensation or upon exposure to oxidative rearrangement conditions. Thus, it readily enables deprotection of the benzyl ether group in an unstable acetal under hydrogenolysis conditions. There are some reports[11] on isoflavone synthesis by methods other than oxidative rearrangement of the chalcone. However, since the existing reports on the synthesis of isoflavones are mostly limited to the synthesis of aglycones, that is, isoflavones without sugar moieties, focus was placed on whether oxidative rearrangement can proceed without protection of the hydroxy groups of the sugar residues.

Phroloacetophenone bis-*C*-glycoside **3**, which was prepared in 40% yield[12] by direct C-glycosylation of phloroacetophenone with D-glucose in the presence of scandium(III) triflate by refluxing for eight hours in aqueous ethanol, was selectively benzyl-protected with benzyl bromide in the presence of potassium carbonate in *N*,*N*-dimethylformamide in 65% yield[9] (Scheme 1). A cinnamoyl residue was introduced to phroloacetophenone

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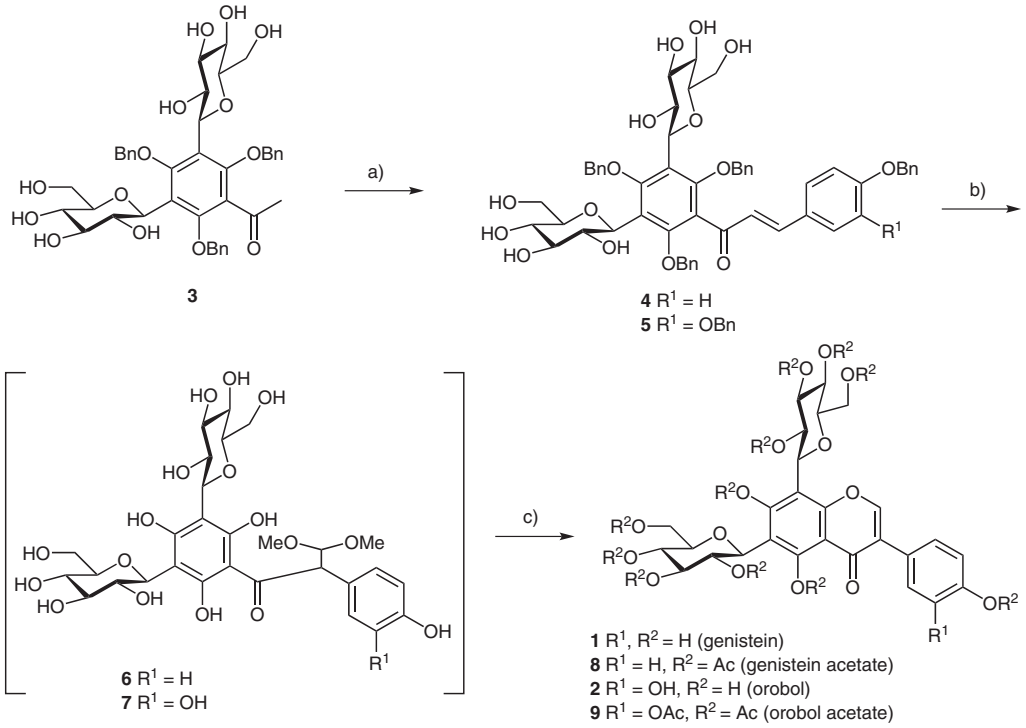

Scheme 1 Total synthesis of genistein and orobol 6,8-bis-*C*-glycosides **1** and **2**. *Reagents and conditions*: a) 4-benzyloxy or 3,4-dibenzyloxybenzaldehyde (2.0 equiv), 28% NaOMe in MeOH, r.t., 19 h; **4**: 88%, **5**: 78%; b) (i) DIB (3.0 equiv), *p*-TsOH (4.0 equiv), anhyd MeOH, r.t., 1 d, (ii) H_2, 10% Pd/C, MeOH, r.t., 1 d; c) aq 6 M HCl–1,4-dioxane–MeOH (0.4:0.3:1), reflux, 1 h; **1**: 31%, **2**: 25% (from **4** and **5**).

bis-*C*-glycoside **3** by aldol condensation with two equivalents of 4-benzyloxy or 3,4-dibenzyloxybenzaldehyde in the presence of sodium methoxide in anhydrous methanol to yield the chalcone bis-*C*-glycosides **4**[9] and **5** in 88 and 78% yield, respectively. The chalcones **4** and **5** were next subjected to oxidative rearrangement by stirring together with three equivalents of diacetoxyiodosobenzene and 4 equivalents of *p*-toluenesulfonic acid in methanol at room temperature for one day to give a dimethyl acetal, which was successively de-O-benzylated by hydrogenolysis (H_2, 10% Pd/C, r.t., 1 d) to give a deprotected dimethyl acetal [**6**: δ = 3.10 and 3.36 ppm (2 × OCH_3) and **7**: δ = 3.19 and 3.44 ppm (2 × OCH_3)]. In this reaction, hydroxytosyloxyiodobenzene (HTIB), which is an oxidant,[10] was formed in situ from diacetoxyiodobenzene and *p*-toluenesulfonic acid. The acetals **6** and **7** were subjected to acid-catalyzed cyclization after crude purification using silica gel column chromatography (solvent system: acetone–EtOAc–H_2O, 15:15:1).[7,8,13] Next, acid-catalyzed cyclization of **6** and **7** by refluxing in a solution of aqueous 6 M hydrochloric acid–1,4-dioxane–methanol (0.4:0.3:1) afforded the desired genistein and orobol bis-*C*-glycosides **1** and **2**, respectively. Purification of both compounds was carried out after acetylation (Ac_2O–pyridine–DMAP), followed by de-O-acetylation with sodium methoxide in anhyd methanol and neutralization by Dowex® 50WX8-200 (H^+) resin to give pure **1** and **2** in 31 and 25% yield from **4** and **5**, respectively (Scheme 1). NMR analysis of the acetates **8**, **9** of **4** and **5** was used to determine the structures in detail. ^1H and ^{13}C NMR spectroscopy of **1** and **2** were performed at 120 °C to remove the rotamers. ^{13}C NMR spectral data of both natural and synthetic **1** and **2** are shown in Table 1. The chemical shifts of natural and synthetic **1** and **2** were very similar.

Table 1 ^{13}C NMR Spectral Data for Genistein and Orobol 6,8-Bis-*C*-glycosides **1** and **2**

	1		**2**	
Carbon	Natural (DMSO-d_6)[a]	Synthetic (DMSO-d_6 + D_2O)[b]	Natural (DMSO-d_6)[a]	Synthetic (DMSO-d_6 + D_2O)[b]
2	153.6	153.0	153.8	153.0
3	121.2	121.1	122.3	122.2
4	180.5	180.6	180.9	180.6
5	159.8	159.4	160.0	159.8
6	108.6	108.2	108.7	108.2
7	162.8	161.3	161.7	161.4
8	103.8	103.7	103.5	103.5
9	153.6	153.0	154.4	154.7
10	104.1	104.4	104.3	104.3
1′	121.9	122.2	121.5	121.6
2′	130.1	129.7	115.5	115.3
3′	115.0	115.0	144.9	144.6
4′	155.0	154.8	145.6	145.3

Table 1 ^{13}C NMR Spectral Data for Genistein and Orobol 6,8-Bis-*C*-glycosides **1** and **2** (continued)

	1		**2**	
Carbon	Natural (DMSO-d_6)[a]	Synthetic (DMSO-d_6 + D_2O)[b]	Natural (DMSO-d_6)[a]	Synthetic (DMSO-d_6 + D_2O)[b]
5′	115.0	115.0	116.6	116.5
6′	130.1	129.7	120.0	119.8
G1	73.7/73.9	73.68/73.84	73.7	73.65/73.82
G2	70.9/71.1	71.05/71.27	71.3	71.08/71.29
G3	78.5	78.31/78.34	78.5	78.41/78.45
G4	69.9	70.03/70.06	69.9	70.11
G5	81.4/81.6	81.01/81.12	81.6	81.00/81.10
G6	60.6/60.7	60.77/60.86	60.7	60.81/60.89

[a] Measured at 90 °C.[4]
[b] Measured at 120 °C.

In conclusion, we have reported here the first successful synthesis of two isoflavone bis-*C*-glycosides using an environmentally friendly and short process with the following advantages: 1) direct di-C-glycosylation of phloroacetophenone with D-glucose in aqueous solution with a catalytic amount of scandium(III) triflate; 2) the reactions were performed without protection of the glucose hydroxy groups in all processes; and 3) the oxidative rearrangement reaction used diacetoxyiodobenzene and *p*-toluenesulfonic acid. This method is applicable to the synthesis of *C*-glycosyl flavonoids.

The solvents used in these reactions were purified by distillation. Reactions were monitored by TLC, on 0.25 mm silica gel F254 plates (E. Merck) using UV light, and either a 5% ethanolic solution of $FeCl_3$ or a 7% ethanolic solution of phosphomolybdic acid with heat as the coloration agent. 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. Optical rotations were recorded on a JASCO DIP-370 polarimeter. Melting points were determined using an ASONE micro-melting point apparatus and are uncorrected. IR spectra were recorded on a Horiba FT-720 IR spectrometer using a KBr disk. 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 3-nitrobenzyl alcohol (NBA) or glycerol as a matrix on a JEOL JMS-AX505HA instrument. Elemental analyses were performed on a Perkin-Elmer PE 2400 II instrument. Before elemental analysis each product was subjected to drying at 80–100 °C under reduced pressure for more than 2 h.

^1H and ^{13}C NMR spectroscopy of the products **5**, **1**, and **2** were measured at 80 °C in DMSO-d_6 and 120 °C in DMSO-d_6 containing 3–5 drops of D_2O; thus, rotamers were not detectable. NMR data measured at 90 °C[4] could not be assigned due to the occurrence of rotamers.

6,8-Di-*C*-β-D-glucopyranosyl-2,3′,4,4′,6-penta-*O*-benzylchalcone (5)

To a solution of **3** (178 mg, 0.234 mmol) and 3,4-dibenzyloxybenzaldehyde (149.8 mg, 0.468 mmol) in anhyd MeOH (1 mL) was added 28% NaOMe in MeOH (0.5 mL) and the resulting mixture was stirred at r.t. under argon for 1 d. The reaction mixture was added to ice-water (ca. 50 mL) and neutralized with aq 2 M HCl. The resulting yellow precipitate was filtered and washed with H_2O (10 mL) to give the crude product, which was purified by silica gel column chromatography (20:1–5:1, $CHCl_3$–MeOH) to afford **5** (194.1 mg, 78%) as a yellow powder; mp 128–130 °C; $[\alpha]_D^{21}$ –10.3 (*c* 1.03, MeOH).

IR (KBr): 3394, 2929, 2879, 1701, 1637, 1576, 1508 cm^{-1}.

^1H NMR (500 MHz, DMSO-d_6 + D_2O at 120 °C): δ = 4.76 (br s, 2 H, PhCH_2), 4.92 and 5.18 (2 d, *J* = 11.5 Hz, each 1 H, PhCH_2), 5.14 and 5.17 (2 s, each 2 H, 4′, 5′-PhCH_2), 5.32 (br s, 2 H, PhCH_2), 6.99 (d, *J* = 16.1 Hz, 1 H, *trans*-vinyl H), 7.07 (d, *J* = 8.3 Hz, 1 H, H-5′), 7.16 (dd, *J* = 1.7, 8.5 Hz, 1 H, H-6′), 7.28–7.59 (m, 27 H, ArH); δ (glucose moiety) = 3.17 (t, *J* = 8.5 Hz, 2 H, H-4, 4′), 3.22 (m, 4 H, H-3, 3′, 5, 5′), 3.50 (br dd, 2 H, H-6a, 6′a), 3.72 (br dd, *J* = 11.0 Hz, 2 H, H-6b, 6′b), 4.26 (br t, 2 H, H-2, 2′), 4.74 (br d, 2 H, H-1, 1′).

MS (FAB+): *m*/*z* = 1063 $(M + H)^+$.

Anal. Calcd for $C_{62}H_{62}O_{16}·0.5H_2O$: C, 69.45; H, 5.92. Found: C, 69.24; H, 5.80.

6,8-Di-*C*-β-D-glucopyranosyl-2,4,4′-trihydroxyisoflavone (1)

To a solution of DIB (238.6 mg, 0.741 mmol) and *p*-TsOH·H_2O (189.4 mg, 0.996 mmol) in anhyd MeOH (0.5 mL) was added dropwise a solution of **4** (238.1 mg, 0.249 mmol) in anhyd MeOH (0.7 mL) and the mixture was stirred at r.t. for 1 d. To the reaction mixture was added aq 10% $Na_2S_2O_3$ (50 mL). The resulting mixture was passed through an MCI gel® CHP20P column (ca. 50 mL in H_2O). After washing the gel column with H_2O (ca.150 mL) and removing the nonabsorbent, the absorbent was eluted with 50% aq acetone (100 mL) and then with acetone–MeOH (4:1, 50 mL). After evaporating the solvent from the eluate in vacuo, the residual crude product was purified by silica gel column chromatography (15:15:1 acetone–EtOAc–H_2O) to afford the crude benzyl-protected acetal (211.5 mg) as a yellow oil, which was dissolved in MeOH (2 mL). To the methanolic solution of the benzyl-protected acetal was added 10% Pd/C (29.9 mg), and the mixture was vigorously stirred at r.t. under a H_2 atmosphere (balloon) for 24 h. The mixture was filtered through a Celite pad, and the filtrate was evaporated in vacuo to afford **6** (117.9 mg, 86% from **4**) as a colorless solid. Crude **6** (117.9 mg) was dissolved in MeOH (1.0 mL), and then 1,4-dioxane (0.3 mL) and aq 6 M HCl (0.4 mL) were added. The mixture was refluxed for 1 h. After removing the solvent in vacuo, EtOH (7 mL) was added to the residue and then evaporated to remove the HCl. This procedure was repeated twice. The residual crude **1** was acetylated with Ac_2O (0.5 mL) in pyridine (0.5 mL) in the presence of DMAP (ca. 20 mg) at r.t. for 1 d. The mixture was added to ice-water (ca. 30 mL) and extracted with EtOAc (3 × 7 mL). The combined EtOAc extracts were washed with aq 0.5 M HCl (7 mL) and brine (10 mL), and dried (Na_2SO_4). After evaporation of the solvent in vacuo, the residue was purified by silica gel column chromatography (1:1 *n*-hexane–EtOAc) to afford **8** (110.8 mg, 42% from **4**). To a solution of **8** (55.7 mg) in anhyd MeOH (1 mL) was added 28% NaOMe in MeOH (0.05 mL, ca. 50 mg) and stirred at r.t. for 1 h. After monitoring the disappearance of the substrate by TLC, Dowex® 50WX8-200 (H^+) resin was added to the mixture until the pH of the solution became neutral. The mixture was filtered and washed with MeOH (5 mL). The filtrate was evaporated in vacuo to afford **1** (23.8 mg, 31% from **4**) as a yellow solid.

1

Mp 228–230 °C (Lit.[3] mp 225 °C, Lit.[4] mp 222–224 °C, Lit.[2] mp 225–227 °C); $[\alpha]_D^{21}$ +72 (*c* 0.54, MeOH).

IR (KBr): 3346, 2925, 2893, 1709, 1647, 1585, 1516 1458 cm^{-1}.

^1H NMR (500 MHz, DMSO-d_6 + D_2O at 90 °C): δ = 3.30–3.34 (m, 6 H, H-3, 3′, 4, 4′, 5, 5′), 3.37 (dd, *J* = 11.3, 4.1 Hz, 2 H, H-6a, 6′a), 3.70 (dd, *J* = 11.3, 1.9 Hz, 2 H, H-6b, 6′b), 3.80 and 3.81 (2 t, *J* = 8.3 Hz, each 1 H, H-2, 2′), 4.80 and 4.84 (2 d, *J* = 9.8 Hz, each 1 H, H-1, 1′), 6.83 (dd, *J* = 9.0, 1.9 Hz, 2 H, H-3′, 5′), 7.37 (dd, *J* = 2.1, 9.0 Hz, 2 H, H-2′, 6′), 8.27 (s, 1 H, H-2), 9.29 (br s, 2 H, 2 × OH), 9.60 (br s, 1 H, OH), 13.64 (s, 1 H, OH).

MS (FAB+): *m*/*z* = 595 $(M + H)^+$.

Anal. Calcd for $C_{27}H_{30}O_{15}·1.5H_2O$: C, 52.17; H, 5.35. Found: C, 52.34; H, 5.31.

6,8-Di-*C*-β-D-glucopyranosyl-2,4,4′-trihydroxyisoflavone Undecaacetate (8)

Mp 164–166 °C (Lit.[3] 160 °C); $[\alpha]_D^{21}$ –33 (*c* 0.505, $CHCl_3$).

IR (KBr): 2943, 1759, 1655, 1604, 1508 cm^{-1}.

^1H NMR (500 MHz, $CDCl_3$): δ (isoflavone moiety) = 2.32, 2.46, 2.53 (3 s, each 3 H, 3 × ArOCOCH_3), 7.16 and 7.50 (2 d, *J* = 8.5 Hz, each 2 H, H-3′, 5′ and H-2′, 6′), 8.03 (s, 1 H, H-2); δ (glucose moiety) = 1.751, 1.918, 2.024, 2.045, 2.048, 2.051, 2.067, 2.078 (8 s, each 3 H, 8 × OCOCH_3), 3.72 and 3.80 (2 m, each 1 H, H-5′, 5), 3.95 and 4.15 (2 dd, *J* = 1.2, 12.6 Hz and 2.2, 12.4 Hz, each 1 H, H-6a, 6′a), 4.21 and 4.45 (2 dd, *J* = 5.4, 12.2 Hz and 4.9, 12.9 Hz, each 1 H, H-6′b, 6b), 4.54 and 4.81 (2 d, *J* = 10.2 and 9.0 Hz, each 1 H, H-1′, 1), 5.15 and 5.24 (2 t, *J* = 9.8 Hz, each 1 H, H-4′, 4), 5.30 and 5.37 (2 dd, *J* = 9.3, 9.5 Hz, each 1 H, H-3′, 3), 5.69 and 5.96 (2 br t and t, *J* = 9.6 Hz, each 1 H, H-2′, 2).

MS (FAB+): *m*/*z* = 1057 $(M + H)^+$.

Anal. Calcd for $C_{49}H_{52}O_{26}$: C, 55.68; H, 4.96. Found: C, 55.42; H, 4.90.

6,8-Di-*C*-β-D-glucopyranosyl-2,3′,4,4′-tetrahydroxyisoflavone (2)

To a solution of DIB (177.8 mg, 0.552 mmol) and *p*-TsOH·H_2O (143.1 mg, 0.752 mmol) in anhyd MeOH (0.5 mL) was added dropwise a solution of **5** (194.1 mg, 0.183 mmol) in anhyd MeOH (0.9 mL) and the mixture stirred at r.t. for 1 d. To the stirred reaction mixture was added aq 10% $Na_2S_2O_3$ (40 mL). The resulting mixture was passed through an MCI gel® CHP20P column (ca. 50 mL in H_2O). After washing the gel with H_2O (150 mL) and removing the nonabsorbent, the absorbent was eluted with 50% aq acetone (100 mL) and then with acetone–MeOH (4:1, 50 mL). After evaporating the solvents from the eluate, the residual crude product was purified by silica gel column chromatography (20:1–5:1 $CHCl_3$–MeOH) to afford the crude benzyl-protected acetal (127.8 mg) as a yellow oil, which was dissolved in MeOH (2 mL). To the methanolic solution was added 10% Pd/C (28 mg) and the mixture was vigorously stirred at r.t. under a H_2 atmosphere (balloon) for 15 h. The mixture was filtered through a Celite pad and the filtrate was evaporated in vacuo to afford **7** (67.2 mg, 88% from **5**) as a colorless solid. Crude **7** (67.2 mg) was dissolved in MeOH (1.0 mL), and then 1,4-dioxane (0.3 mL) and aq 6 M HCl (0.4 mL) were added to the methanolic solution. The mixture was refluxed for 1 h. After removing the solvent in vacuo, EtOH (7 mL) was added to the residue, and then the mixture was evaporated in vacuo to remove the HCl. This procedure was repeated twice. The residual crude **2** was acetylated with Ac_2O (0.5 mL) in pyridine (0.5 mL) in the presence of DMAP (ca. 20 mg) at r.t. for 1 d. The mixture was added to ice-water (ca. 30mL) and extracted with EtOAc (3 × 7 mL). The combined EtOAc extracts were washed with aq 0.5 M HCl (7 mL) and brine (10 mL), and dried (Na_2SO_4). After evaporation of the solvent in vacuo, the residue was purified by silica gel column chromatography (1:1 *n*-hexane–EtOAc) to afford **9** (58.2 mg, 29% from **5**). To a solution of **9** (58.2 mg) in anhyd MeOH (1 mL) was added 28% NaOMe in MeOH (0.05 mL, ca. 50 mg), followed by stirring at r.t. for 1 h. After monitoring by TLC, Dowex® 50WX8-200 (H^+) resin was added to the mixture until the pH was neutral. The mixture was filtered, and then washed with MeOH (5 mL). The filtrate was evaporated in vacuo to afford **2** (27.5 mg, 25% from **5**) as a yellow solid.

2

Mp 234–237 °C (dec.) (Lit.[3] 183–186 °C, Lit.[4] mp 240-241 °C); $[\alpha]_D^{21}$ +56 (*c* 0.275, MeOH).

IR (KBr): 3367, 2925, 1716, 1701, 1635, 1522, 1458 cm^{-1}.

^1H NMR (500 MHz, DMSO-d_6 + D_2O at 100 °C): δ = 3.28–3.35 (m, 6 H, H-3, 3′, 4, 4′, 5, 5′), 3.59 (dd, *J* = 11.8, 4.5 Hz, 2 H, H-6a, 6′a), 3.72 (dd, *J* = 11.6, 5.8 Hz, 2 H, H-6b, 6′b), 3.82 and 3.84 (2 m, each 1 H, H-2, 2′), 4.81 and 4.84 (2 d, *J* = 9.6 Hz, each 1 H, H-1, 1′), 6.80 (d, *J* = 8.1 Hz, 1 H, H-5′), 6.83 (dd, *J* = 2.0, 8.2 Hz, 1 H, H-6′), 7.03 (d, *J* = 2.1 Hz, 1 H, H-2′), 8.23 (s, 1 H, H-2), 9.05 (br s, 2 H, 2 × OH), 9.29 (br s, 1 H, OH), 13.79 (s, 1 H, OH).

MS (FAB+): *m*/*z* = 611 $(M + H)^+$.

Anal. Calcd for $C_{27}H_{30}O_{16}·2.5H_2O$: C, 49.47; H, 5.38. Found: C, 49.62; H, 5.35.

6,8-Di-*C*-β-D-glucopyranosyl-2,3′,4,4′-tetrahydroxyisoflavone Dodecaacetate (9)

Mp 157–159 °C (Lit.[3] mp 150 °C); $[\alpha]_D^{21}$ –31 (*c* 0.53, $CHCl_3$,).

IR (KBr): 2943, 1755, 1655, 1604, 1506 cm^{-1}.

^1H NMR (500 MHz, $CDCl_3$): δ (isoflavone moiety) = 2.312, 2.465, 2.529 (2 s, each 3 H, 3 × ArOCOCH_3), 7.26 and 7.38 (2 d, *J* = 8.5 Hz, each 2 H, H-3′, 5′ and H-2′, 6′), 8.06 (s, 1 H, H-2); δ (glucose moiety) = 1.748, 1.914, 2.024, 2.050, 2.052, 2.068, 2.073, 2.078 (8 s, each 3 H, 8 × OCOCH_3), 3.72 (ddd, *J* = 2.1, 5.3, 9.8 Hz, 1 H, H-5′), 3.95 (m, 1 H, H-6a), 4.15 (dd, *J* = 2.1, 12.3 Hz, 1 H, H-6′a), 4.21 (dd, *J* = 5.3, 12.3 Hz, 1 H, H-6′b), 4.46 (dd, *J* = 4.1, 12.3 Hz, 1 H, H-6b), 4.53 (d, *J* = 10.0 Hz, 1 H, H-1′), 4.80 (d, *J* = 8.5 Hz, 1 H, H-1), 5.15 and 5.24 (2 t, *J* = 9.8 Hz, each 1 H, H-4′, 4), 5.30 and 5.37 (2 t, *J* = 9.4 Hz, each 1 H, H-3′, 3), 5.69 (br t, 1 H, H-2′), 5.95 (dd, *J* = 9.4, 10.0 Hz, 1 H, H-2).

MS (FAB+): *m*/*z* = 1115 $(M + H)^+$.

Anal. Calcd for $C_{51}H_{54}O_{28}$: C, 54.94; H, 4.88. Found: C, 54.84; H, 4.93.

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