DOI: 10.1002/ejoc.200800803

An Efficient Method for the Glycosylation of Isoflavones

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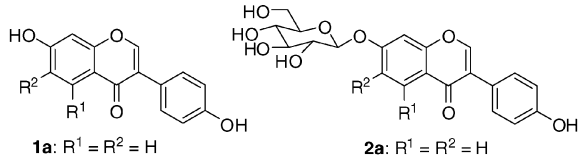
Keywords: Isoflavones / Phytoestrogens / Glucuronides / Glycosylation / Genistein

The isoflavone phytoestrogens are still of current interest for their positive and negative health benefits. However, there are still many unanswered questions regarding their absorption, metabolism and bioavailability. Studies in this area require access to samples of both the isoflavone 7-*O*-glucosides, the form found in plants and the 7-*O*-glucuronides, which are important mammaliam metabolites. A new efficient, high-yielding glycosylation procedure is described for isoflavones, which employs 2,2,2-trifluoro-*N*-(*p*-methoxyphenyl)acetamidates as the glycosyl donors. This methodology was used to prepare the 7-*O*-glycosides of the three main isoflavones, daidzein, genistein and glycitein. The isoflavones were protected with hexanoyl groups which improved their solubility in organic solvents and improved the efficiency of the reaction. The same methodology was then adapted for the synthesis of the analogous 7-*O*-glucuronides. The new synthesis will provide access to large quantities of these compounds for further biological studies.

Introduction

The isoflavone phytoestrogens are a group of polyphenolic compounds with weak estrogenic and antiestrogenic activity, present in the human diet.[1,2] The main dietary sources are soybeans and soy-derived products.[3,4] Epidemiological studies have shown that the consumption of an isoflavone-rich diet is associated with a decrease in the incidence of hormone-related cancers, such as breast[5] and prostate cancer.[6] It has also been suggested that isoflavones may possess other health-promoting activities, including chemoprevention of osteoporosis[7] and cardiovascular disease[8] and lessening of menopausal symptoms.[9,10] However, the biological effects of isoflavones on human health cannot be fully understood until their absorption, metabolism and bioavailability have been fully established.[11–13]

In plants and foodstuffs isoflavones occur predominantly as glycosides (**2a**–**c**),[14] and so these water-soluble derivatives are the forms ingested by humans. The glycosides are then hydrolysed by intestinal glucosidases, both from the host[15] and intestinal bacteria,[16] to give the aglycones, daidzein (**1a**), genistein (**1b**) and glycitein (**1c**). Some studies have reported that the isoflavone aglycones are absorbed more readily than the parent glycosides,[17,18] whereas data from other studies have implied that the bioavailability of genistein and daidzein glycosides is actually better than that of the aglycones.[19] The aglycones can then be metabolised to other biologically active metabolites. For example, daidzein is converted to equol and *O*-demethylangolensin (ODMA),[20] whereas genistein gives 6-hydroxy-ODMA.[21] When the aglycones and their metabolites are absorbed, they are then transported to the liver, where they can undergo conjugation catalysed by hepatic enzymes to give *O*-glucuronides,[22] and to a lesser extent *O*-sulfates.[23]



1a: $R^1 = R^2 = H$
1b: $R^1 = OH$, $R^2 = H$
1c: $R^1 = H$, $R^2 = OCH_3$

2a: $R^1 = R^2 = H$
2b: $R^1 = OH$, $R^2 = H$
2c: $R^1 = H$, $R^2 = OCH_3$

In order to better understand the absorption, metabolism and bioavailability of isoflavones, there is an urgent need for the development of efficient synthetic routes for the synthesis of pure standards of isoflavone *O*-glucosides and *O*-glucuronides to allow their accurate quantification in biological fluids and to study their possible biological roles in vivo. Recently, in our laboratory, Fairley et al. developed an efficient synthesis of isoflavone *O*-sulfates,[24] and now we wish to report herein a new, sophisticated and high-yielding procedure for the synthesis of isoflavone 7-*O*-glucosides and 7-*O*-glucuronides.[25]

Results and Discussion

Previous methods for the glycosidation of polyhydroxyisoflavones are low-yielding (<30%).[26,27] Reaction of daidzein (**1a**) or its protected derivatives 4′-*O*-acetyldaidzein and 4′-*O*-TBDMS-daidzein with α-acetobromoglucose, employing Koenig–Knorr techniques, in CH_2Cl_2 with Ag_2CO_3 as catalyst and in the presence of either lutidine or

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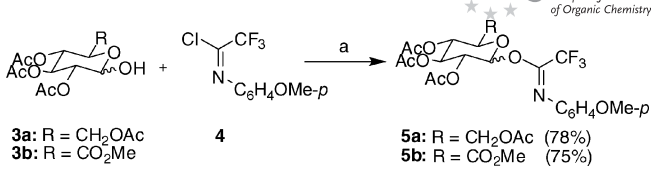
collidine as a base, afforded the glycosidation product in low yields (10–15%). Lewis et al.[26] reported the synthesis of daidzin (**2a**) in 40% yield from α-acetobromoglucose and daidzein (**1a**) by using phase-transfer catalysis. However, in our hands this procedure afforded daidzin (**2a**) in less than 10% yield as indicated by ^{1}H NMR spectroscopy. Thus, it was necessary to search for sophisticated glycosyl donors outside the conventional glycosidation reagents. Alternative glycosyl donors were prepared, allowing glycosidation to be carried out under non-basic conditions, avoiding the use of silver salts. Among these, treatment of phenyl selenoglycoside with daidzein in CH_2Cl_2, employing TMSOTf, Br_2 or I_2 as activator, afforded an intractable mixture of per-acetylated daidzin and per-acetylated daidzein-4′-yl glucoside in low yields. Glycosyl phosphates have emerged as useful glycosyl donors for stereoselective glycosidic bond formation in the synthesis of oligosaccharides and phenolic glycosides.[28] Unfortunately, attempts to adapt this approach for the glucosidation of 4′-*O*-acetyldaidzein proved to be low-yielding and gave irreproducible results.

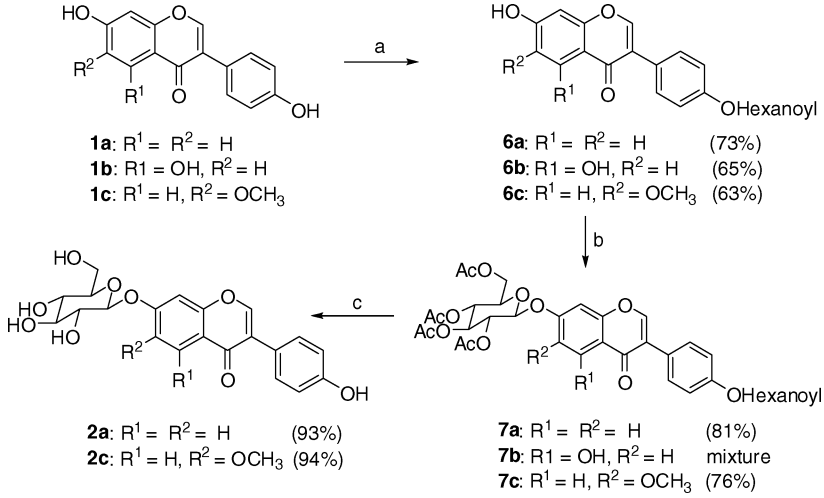
Recently, Li et al. reported an efficient and facile synthesis of flavonoid 7-*O*-glycosides by employing a newly developed benzoyl-protected glycosyl trifluoroacetimidate donor.[29] The method is based on a Lewis acid promoted coupling of a glycosyl trifluoroacetimidate donor with the 7-OH group of flavonoid esters. 2,3,4,6-Tetra-*O*-acetyl-D-glycopyranosyl 2,2,2-trifluoro-*N*-(*p*-methoxyphenyl)acetamidate (**5a**) and methyl 2,3,4-tri-*O*-acetyl-D-glycopyranosiduronyl 2,2,2-trifluoro-*N*-(*p*-methoxyphenyl)acetamidate (**5b**) were readily prepared in 78 and 75% yield, respectively, by treatment of the corresponding 1-hydroxy sugar, **3a** or **3b**, with 2,2,2-trifluoro-*N*-(*p*-methoxyphenyl)acetimidoyl chloride (**4**) in the presence of K_2CO_3 in wet acetone at room temperature for 4 h (Scheme 1). The glucuronyl trifluoroacetimidate **5b** was not stable for long periods at room temperature and had to be stored at –78 °C.



Scheme 1. Reagents and conditions: (a) K_2CO_3, wet acetone, room temp., 4 h.

To overcome the regioselectivity problem and to increase the solubility of the isoflavone in CH_2Cl_2, we sought to acylate the phenolic 4′-hydroxy group. The 7-hydoxy group in isoflavones is more acidic than that at C-4′ because of the electron-withdrawing effect of the *para*-carbonyl group in ring C.[30] Therefore, use of an excess of KO*t*Bu in DMF allows the generation of the 4′,7-diphenolate of either daidzein (**1a**) or glycitein (**1c**), or the 4′,5,7-triphenolate of genistein (**1b**). The electron-withdrawing carbonyl groups *para* to the 7-oxy group and in the *ortho* position to the 5-oxy group significantly diminish their nucleophilicity, thus acylation with 1.0 equiv. of acyl chlorides produces the 4′-acetylated isoflavone selectively. Acylation of daidzein (**1a**), and glycitein (**1c**) with an equivalent amount of hexanoyl chloride in the presence of KO*t*Bu (2.1 equiv.) in dry DMF at room temp. gave the isoflavone 4′-hexanoates **6a**,**c** in 65–78% yield (Scheme 2).

With the isoflavone 4′-hexanoates **6a**,**b**,**c** in hand, we proceeded to investigate their glycosidation with the very reactive trifluoroacetamidate donor **5a**. The glycosyl donor α/β-*O*-acetylglycopyranosyl 2,2,2-trifluoro-*N*-(*p*-methoxyphenyl)acetamidate (**5a**) (1.5 equiv.) was thus treated with the acceptor **6a**,**b**,**c** (1.0 equiv.) by employing $BF_3{\cdot}Et_2O$ as a promoter in dry CH_2Cl_2 and in the presence of freshly dried 4 Å molecular sieves at room temperature. The desired per-acetylated isoflavonoid 7-*O*-glycosides **7a**,**c** were obtained in 81 and 76% yield, respectively (Scheme 2).

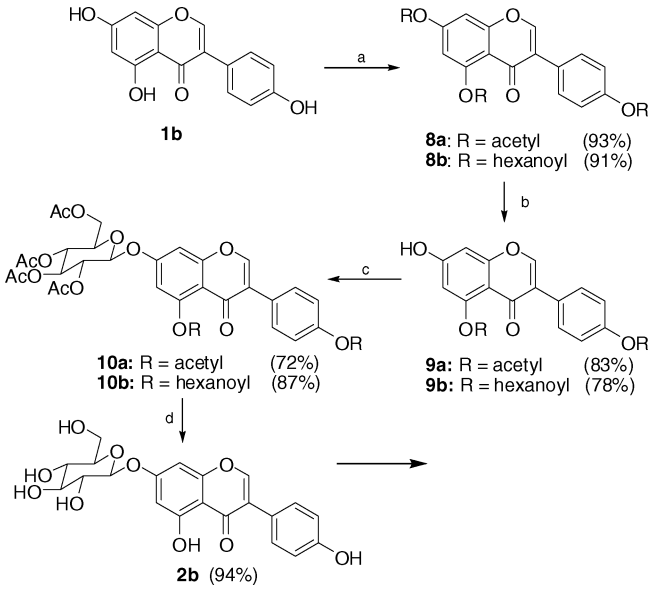


Scheme 2. Reagents and conditions: (a) 2.1 equiv. of KO*t*Bu, DMF, 1 equiv. of hexanoyl chloride, room temp.; (b) **5a**, 0.3 equiv. of $BF_3{\cdot}Et_2O$, CH_2Cl_2, 4 Å MS, room temp., 6 h; (c) K_2CO_3, H_2O/MeOH, THF, 40 °C, 5 h.

Unfortunately, glycosidation of genistein 4′-hexanoate (**6b**) with the glycosyl trifluoroacetimidate donor **5a** under the above conditions afforded an intractable mixture. Therefore, it was concluded that the 5- and 4′-OH groups on genistein (**1b**) both had to be protected. Treatment of a solution of genistein (**1b**) in pyridine with acetic anhydride or hexanoyl chloride (4 equiv.) afforded 4′,5,7-tri-*O*-acylgenistein (**8a**) and **8b** in 93 and 91% yield, respectively (Scheme 3). The 7-*O*-acyl group on the peracylated genistein **8a**,**b**, which is in *para* position to the electron-withdrawing pyrone carbonyl group, is the most electrophilic entity. Taking advantage of this fact, regioselective release of 7-OH was accomplished in good yield by nucleophilic attack of PhSH (1.2 equiv.) on the 7-ester group in the presence of imidazole in *N*-methyl-2-pyrrolidinone (NMP) as solvent at 0 °C (Scheme 3). Glycosylation of both the acceptors **9a**,**b** with the glycosyl donor **5a** by employing the conditions described above resulted in **10a** and **10b** in 72 and 87% yield, respectively.

This attractive synthetic route also allowed us to synthesise the major three soybean isoflavone 7-*O*-glucuronides in good yields. Isoflavon-7-yl-β-D-glucopyranosiduronic methyl esters **11a**–**d** were synthesised in excellent yields by coupling of the acceptors **6a**,**c**, **9a**,**b** with glucuronyl 2,2,2-trifluoro-*N*-(*p*-methoxyphenyl)acetamidate donor **5b** under standard glycosylation conditions described for their corresponding isoflavone 7-*O*-glucosides (Scheme 4).

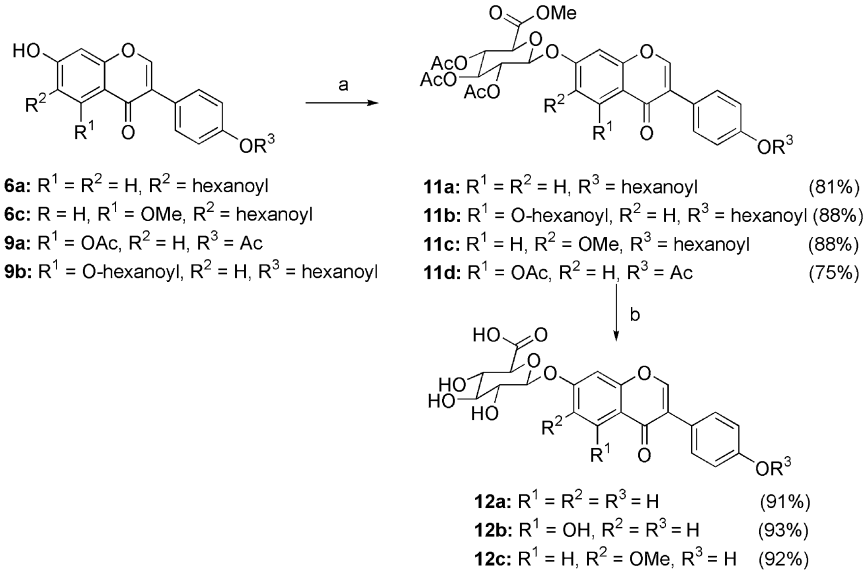
Final removal of the acyl groups to provide the isoflavone 7-*O*-glycosides and 7-*O*-glucuronides was achieved by using K_2CO_3 in MeOH/THF/H_2O (5:5:1). The glycosides were purified by dissolving the crude material in a few drops of water and precipitating with ethanol, whereas the glucuronides were purified by means of C18 reversed-phase chromatography using water/acetonitrile (9:1) as eluant. The purity of both glycosides and glucuronides was confirmed by HPLC using water/acetonitrile (70:30 for the glycoside; 90:20 for the glucuronides) as eluant.



Scheme 3. Reagents and conditions: (a) Ac_2O, Py, room temp., 8 h, or hexanoyl chloride, Py, DMAP, $CHCl_3$, 0 °C; (b) PhsH, NMP, Im; (c) **5a**, $BF_3{\cdot}Et_2O$, CH_2Cl_2, 4 Å MS, room temp., 6 h; (d) K_2CO_3, MeOH, THF, H_2O, 40 °C, 5 h.

Conclusions

A new glycosylation procedure has been developed for isoflavones, which employs 2,2,2-trifluoro-*N*-(*p*-methoxyphenyl)acetamidates as the glycosyl donors. This methodology was used to prepare the 7-*O*-glycosides of the three main isoflavones, daidzein, genistein and glycitein. The isoflavones were protected with hexanoyl groups which had the added bonus of improving their solubility in organic solvents and improved the efficiency of the reaction. The same methodology was then adapted for the synthesis of the analogous 7-*O*-glucuronides, which are mammalian metabolites of the isoflavones. The new synthesis will provide



Scheme 4. (a) **5a**, $BF_3{\cdot}Et_2O$, CH_2Cl_2, 4 Å MS, room temp., 6 h; (d) K_2CO_3 MeOH, THF, H_2O, room temp., 5 h.

access to large quantities of these compounds and enable further study of their biological properties. This efficient, high-yielding methodology is also suitable for the synthesis of ^{13}C-labelled derivatives, and studies on isotopic labelling are currently underway.

Experimental Section

General: NMR spectra were recorded with a Varian Gemini 2000 (^1H 300 MHz, ^{13}C 75.45 MHz) or a Bruker Avance 300 (^1H 300 MHz, ^{13}C 75.45 MHz) spectrometer. Chemical shifts (δ) in ppm are given relative to Me_4Si, coupling constants (J) in Hz. Elemental analyses were carried out in the departmental microanalytical laboratory. IR spectra were recorded with a Perkin–Elmer series 1420 FT IR spectrophotometer. The samples were prepared as Nujol mulls or thin films between sodium chloride discs and recorded in cm^{-1}. EI mass spectra were recorded with a Micromass GC-T. Electrospray mass spectra were recorded with a Micromass LC-T. UV spectra were recorded with a Kontron Uvikon 930 spectrometer. Melting points were recorded with an electrothermal melting point apparatus and are uncorrected. Analytical TLC was carried out on Merck 5785 Kieselgel $60F_{254}$ fluorescent plates. Flash chromatography was performed according to the procedure of Still[31] by using silica gel of 35–70 μm particle size. Dimethylformamide was distilled from magnesium sulfate. Diethyl ether and tetrahydrofuran were distilled from sodium metal and benzophenone.

2,3,4,6-Tetra-*O*-acetyl-D-glycopyranosyl 2,2,2-Trifluoro-*N*-(*p*-methoxyphenyl)acetamidate (5a): *N*-(*p*-Methoxyphenyl)acetimidoyl chloride (0.327 g, 1 mmol) was slowly added to a suspension of sugar hemiacetal (1 mmol) and K_2CO_3 (0.276 g, 2 mmol) in wet acetone at room temp. After 4 h of stirring at room temp., the solid was filtered, and the solvent was evaporated under reduced pressure. The residue was subjected to flash chromatography by using petroleum ether/EtOAc (4:1) to give the title compound as an α/β mixture (0.428 g, 78%) as white foam. ^1H NMR (300 MHz, $CDCl_3$): δ = 2.017, 2.027, 2.032, 2.043, 2.051, 2.58, 2.66, 2.083 (8 s, 24 H, 8 Ac), 3.78, 3.79 (2 s, 6 H, OCH_3), 4.07–4.15 (m, 4 H), 4.24 (d, J = 4.8 Hz, 1 H), 4.28 (d, J = 4.8 Hz, 1 H), 5.08–5.25 (m, 6 H), 5.52 (t, J = 9.9 Hz, 1 H), 5.74 (br. s, 1 H, 1-H^β), 5.74 (br. s, 1 H, 1-H^α), 6.76–6.89 (m, 4 H, Ph) ppm. MS (ES): m/z (%) = 572 (100) [M + Na^+], 353 (28). HRMS: calcd. for $C_{23}H_{26}NF_3O_{11}Na$ 572.1356; found 572.1348.

2,3,4-Tri-*O*-acetyl-D-glycopyranosiduronyl 2,2,2-Trifluoro-*N*-(*p*-methoxyphenyl)acetamidate (5b): Compound **5b** (0.40 g, 75%) was obtained as a yellow viscous oil from hemiacetal **3b** and **4** by employing the procedure described for the synthesis of **5a**. ^1H NMR (300 MHz, $CDCl_3$): δ = 2.016, 2.019, 2.027, 2.035, 2.053 (5 s, 18 H, 6 Ac), 3.738, 3.765, 3.773 (3 s, 12 H, 2 CO_2CH_3, 2 OCH_3), 4.42 (d, J = 10.5 Hz, 1 H), 5.14 (dd, J = 10.2, 3.9 Hz, 1 H), 5.21–5.34 (m, 3 H), 5.58 (m, 1 H), 5.85 (br. s, 1 H, 1-H^β), 6.46 (br. s, 1 H, 1-H^α), 6.72–6.85 (m, 8 H, Ph) ppm. MS (ES): m/z (%) = 558 (100) [M + Na]$^+$. HRMS: calcd. for $C_{22}H_{24}NF_3O_{11}Na$ 558.1199; found 558.1203.

General Procedure for Regioselective Acylation of Isoflavones: A solution of isoflavone (2 mmol) and dry *t*BuOK (4.2 mmol) in dry DMF was stirred at room temp. under nitrogen for 10 min. Hexanoyl chloride (0.29 mL, 2.1 mmol) was added dropwise and the mixture stirred for 2 h until TLC showed almost complete disappearance of the substrate. After addition of ice-cold water, the mixture was extracted with EtOAc (3 × 50 mL). The combined organic extracts were dried with $MgSO_4$, concentrated at reduced pressure, and the residue was purified by flash chromatography on silica gel by using DCM/EtOAc (5→15%) as eluant.

4′-*O*-Hexanoyldaidzein (6a): Compound **6a** (0.514 g, 73%) was obtained as a white solid; m.p. 190 °C. ^1H NMR (300 MHz, $CDCl_3$): δ = 0.92 (t, J = 6.9 Hz, 3 H, $CH_2CH_2CH_3$), 1.33–1.47 (m, 4 H, $CH_2CH_2CH_3$), 1.75 (quint, J = 7.2 Hz, 2 H, $O_2CCH_2CH_2CH_2$), 2.56 (t, J = 7.2 Hz, 2 H, O_2CCH_2), 6.84 (d, J = 2.3 Hz, 1 H, 8-H), 6.93 (dd, J = 8.8, 2.3 Hz, 1 H, 6-H), 7.13 (d, J = 8.7 Hz, 2 H, 3′-, 5′-H), 7.54 (d, J = 8.7 Hz, 1 H, 2′-, 6′-H), 7.91 (s, 1 H, 2-H), 8.10 (d, J = 8.8 Hz, 1 H, 5-H) ppm. ^{13}C NMR (75 MHz, $CDCl_3$): δ = 13.9 (C-CH_3), 22.3 (C-CH_2CH_3), 24.6 (C-$CH_2CH_2CH_3$), 31.3 (C-$O_2CCH_2CH_2$), 34.4 (C-$O_2CCH_2CH_2$), 102.7 (C-8), 115.7 (C-6), 117.3 (C-4a), 121.7 (5, C-3), 124.2 (C-3), 127.8 (C-1′), 129.6 (C-5), 130.2 (C-2′, -6′), 150.6 (C-2), 153.0 (C-8a), 158.2 (C-4′), 162.7 (C-7), 172.5 (C-4), 176.2 (C-CO_2) ppm. MS (EI): m/z (%) = 352 (5) $[M]^+$, 254 (100), 137 (11). HRMS: calcd. for $C_{21}H_{20}O_5$ 352.1311; found 352.1317.

4′-*O*-Hexanoylgenistein (6b): Compound **6b** (0.478 g, 65%) was obtained as a white foam. ^1H NMR (300 MHz, $CDCl_3$): δ = 0.99 (t, J = 6.9 Hz, 3 H, $CH_2CH_2CH_3$), 1.38–1.43 (m, 4 H, $CH_2CH_2CH_3$), 1.76–1.83 (m, 2 H, $O_2CCH_2CH_2CH_2$), 2.60 (t, J = 7.5 Hz, 2 H, O_2CCH_2), 6.01 (s, 1 H, 7-OH), 6.23 (d, J = 2.1 Hz, 1 H, 6-H), 6.29 (d, J = 2.1 Hz, 1 H, 8-H), 7.16 (d, J = 8.7 Hz, 2 H, 3′-, 5′-H), 7.54 (d, J = 8.7 Hz, 1 H, 6′- ,2′-H), 7.84 (s, 1 H, 2-H), 12.80 (s, 1 H, 5-H) ppm. ^{13}C NMR (75 MHz, $CDCl_3$): δ = 13.9 (C-CH_3), 22.3 (C-CH_2CH_3), 24.6 (C-$O_2CCH_2CH_2$), 31.3 (C-$O_2CCH_2CH_2$), 34.4 (C-$CH_2CH_2CH_3$), 94.2 (C-8), 99.8 (C-6), 105.9 (C-4a), 121.8 (C-3′, -5′), 123.0 (C-3), 128.4 (C-1′), 130.0 (C-2′, -6′), 150.8 (C-4′), 153.3 (C-2), 157.9 (C-8a), 162.7 (C-5), 167.8 (C-7), 173.0 (C-O_2C), 180.4 (C-4) ppm. MS (EI): m/z (%) = 368 (87) $[M]^+$, 270 (100). HRMS: calcd for $C_{21}H_{20}O_6$ 368.1260; found 368.1256.

4′-Hexanoylglycitein (6c): Compound **6c** (0.481 g, 63%) was obtained as a white foam. ^1H NMR (300 MHz, $CDCl_3$): δ = 0.93 (t, J = 7.0 Hz, 3 H, CH_3), 1.37–1.44 (m, 4 H, $CH_2CH_2CH_3$), 1.74–1.80 (m, 2 H, $O_2CCH_2CH_2$), 2.57 (t, J = 7.5 Hz, 2 H, O_2CCH_2), 4.00 (s, 3 H, OCH_3), 6.46 (s, 1 H, 7-OH), 6.98 (s, 1 H, 8-H), 7.15 (d, J = 8.7 Hz, 2 H, 3′-, 5′-H), 7.58 (d, J = 8.7 Hz, 1 H, 2′-, 6′-H), 7.64 (s, 1 H, 5-H), 7.95 (s, 1 H, 2-H) ppm. ^{13}C NMR (75 MHz, $CDCl_3$): δ = 13.9 (C-CH_3), 22.3 (C-$CH_2CH_2CH_2CH_3$), 24.6 (C-$O_2CCH_2CH_2$), 31.3 (C-$CH_2CH_2CH_3$), 34.4 (C-O_2CCH_2), 56.5 (C-OCH_3), 102.7 (C-8), 104.7 (C-5), 117.8 (C-4a), 121.6 (C-3′, -5′), 123.7 (C-3), 129.6 (C-3), 130.0 (C-2′, -6′), 145.5 (C-6), 150.6 (C-7), 151.4 (C-8a), 152.5 (C-4′), 152.6 (C-2), 172.8 (C-O_2C), 175.8 (C-4) ppm. MS (EI): m/z (%) = 382 (10) $[M]^+$ 284 (100). $C_{22}H_{22}O_6$ (382.41): calcd. C 69.10; H, 5.80; found C 69.58, H 5.36.

5,7,4′-Triacetylgenistein (8a): Acetic anhydride (1.4 mL, 14.8 mmol) was added to a solution of genistein (**1b**, 1 g, 3.7 mmol) in pyridine (10 mL) and dry chloroform (5 mL) at room temp. under nitrogen. After stirring at room temp. overnight, the mixture was diluted with CH_2Cl_2 (100 mL) and washed with 1 N HCl (3 × 100 mL), and brine subsequently. The organic layer was dried with $MgSO_4$, and concentrated to give yellow solid, which was recrystallised from MeOH to give **8a** (1.35 g, 93%) as a white solid; m.p. 137 °C (ref.[32] 139–141 °C). ^1H NMR (300 MHz, $CDCl_3$): δ = 2.31, 2.34, 2.42 (3 s, 9 H, O_2CCH_3), 6.86 (d, J = 2.4 Hz, 1 H, 6-H), 7.15 (d, J = 8.7 Hz, 2 H, 3′-, 5′-H), 7.25 (d, J = 2.4 Hz, 1 H, 8-H), 7.49 (d, J = 8.7 Hz, 2 H, 2′-, 6′-H), 7.89 (s, 1 H, 2-H) ppm. ^{13}C NMR (75 MHz, $CDCl_3$): δ = 21.2 (3 C-O_2CCH_3), 109.0 (C-8), 114.0 (C-6), 115.2 (C-4a), 121.7 (C-3′, -5′), 125.7 (C-3), 128.8 (C-1′), 130.2 (C-2′, -6′), 150.64, 150.75 (C-8a, -4′), 152.0 (C-2), 153.8 (C-8a), 157.7 (C-5), 167.9, 169.4 (3 C-O_2C), 174.2 (C-4) ppm.

4′,5,7-Trihexanoylgenistein (8b): To a solution of genistein (**1b**, 1 g, 3.7 mmol) in pyridine (10 mL) and dry chloroform (5 mL) was added DMAP (0.045 g, 0.37 mmol), followed by hexanoyl chloride (1.82 mL, 18.57 mmol) at –20 °C under nitrogen. After 9 h of stirring at room temp., the mixture was diluted with CH_2Cl_2 (100 mL) and washed with 1 N HCl (2×100 mL), and brine subsequently. The organic layer was dried with $MgSO_4$ and concentrated to give a yellow solid, which was recrystallised from MeOH to give **8b** (1.95 g, 91%) as a white solid; m.p. 86 °C. ^{1}H NMR (300 MHz, $CDCl_3$): δ = 0.90–0.96 (m, 9 H, 3 CH_3), 1.38–1.43 (m, 12 H, $CH_2CH_2CH_3$), 1.74–1.80 (m, 6 H, $CO_2CH_2CH_2$), 2.56 (t, J = 7.5 Hz, 2 H, O_2CCH_2), 2.59 (t, J = 7.5 Hz, 2 H, O_2CCH_2), 2.72 (t, J = 7.5 Hz, 2 H, O_2CCH_2), 6.84 (d, J = 2.4 Hz, 1 H, 6-H), 7.13 (d, J = 8.4 Hz, 2 H, 3′-, 5′-H), 7.23 (d, J = 2.4 Hz, 1 H, 8-H), 7.49 (d, J = 8.4 Hz, 1 H, 2′-, 6′-H), 7.88 (s, 1 H, 2-H) ppm. ^{13}C NMR (75 MHz, $CDCl_3$): δ = 13.88, 13.90, 113.94 (3 C-CH_3), 22.3, 22.4, 24.5, 24.1, 24.4, 24.6 (3 C-$CH_2CH_2CH_3$) 31.2, 31.3 (3 C-$O_2CH_2CH_2$), 34.1, 34.4 (3 C-O_2CCH_2), 108.9 (C-8), 114.0 (C-6), 115.2 (C-4a), 121.8 (C-3′, -5′), 125.7 (C-3), 128.8 (C-1′), 130.2 (C-2′, -6′), 150.8 (C-7, -4′), 152.0 (C-2), 154.0 (C-8a), 157.7 (C-5), 170.9, 172.2, 172.3 (3 C-O_2C), 174.2 (C-4) ppm. MS (EI): m/z = 564 (100) $[M]^+$. $C_{33}H_{40}O_8$ (564.68): calcd. C 70.19, H 7.14; found C 70.35, H 6.98.

General Procedure for the Regioselective Deacylation: Thiophenol (0.309 mL, 3.03 mmol) was slowly added to a solution of peracylated isoflavone **7a**,**b** (2.5 mmol) and imidazole (0.06 g, 0.88 mmol) in NMP (3 mL) at 0 °C under nitrogen. The mixture was stirred at room temp. until disappearance of the starting material as monitored by TLC. The reaction mixture was diluted with CH_2Cl_2 (100 mL) and washed with 1 N HCl and brine subsequently. The organic layer was dried with $MgSO_4$, filtered, and the solvent was removed at reduced pressure. The yellow semisolid was subjected to flash chromatography in DCM/EtOAc (5→15%) to give the title compound.

5,4′-Diacetylgenistein (9a): Compound **9a** (0.734 g, 83%) was obtained as a white solid; m.p. 202 °C. ^{1}H NMR (300 MHz, $CDCl_3$): δ = 2.33, 2.34 (2 s, 6 H, 2 O_2CCH_3), 6.60 (d, J = 2.1 Hz, 1 H, 6-H), 6.77 (d, J = 2.1 Hz, 1 H, 8-H), 7.19 (d, J = 8.7 Hz, 2 H, 3′-, 5′-H), 7.55 (d, J = 8.7 Hz, 2 H, 2′-, 6′-H), 7.97 (s, 1 H, 2-H) ppm. ^{13}C NMR (75 MHz, $CDCl_3$): δ = 21.19 (2 C-O_2CCH_3), 101.0 (C-8), 105.7 (C-6), 109.5 (C-4a), 122.0 (C-3′, -5′), 123.7 (C-3), 128.0 (C-1′), 130.0 (C-2′, -6′), 151.0 (C-4′), 153.9 (C-2), 156.1 (C-8a), 156.9 (C-7), 162.4 (C-5), 168.3, 169.4 (2 C-O_2CCH_3), 181.0 (C-4) ppm. MS (EI): m/z (%) = 354 (4) $[M]^+$, 312 (27), 270 (100). HRMS: calcd. for $C_{19}H_{14}O_7$ 354.0740; found 354.0744. $C_{19}H_{14}O_7$ (354.32): calcd. C 64.41, H 3.98; found C 63.93, H 3.78.

5,4′-Dihexanoylgenistein (9b): Compound **9b** (0.909 g, 78%) was obtained as a white solid; m.p. 145 °C. ^{1}H NMR (300 MHz, $CDCl_3$): δ = 0.88–0.97 (m, 6 H, 2 CH_3), 1.31–1.42 (m, 8 H, $CH_2CH_2CH_3$), 1.77 (quint, J = 7.4 Hz, 4 H, $O_2CCH_2CH_2$), 2.56 (t, J = 7.5 Hz, 2 H, O_2CCH_2), 2.71 (t, J = 7.7 Hz, 2 H, O_2CCH_2), 6.40 (d, J = 2.4 Hz, 1 H, 6-H), 6.47 (d, J = 2.4 Hz, 1 H, 8-H), 7.08 (d, J = 8.7 Hz, 2 H, 3′-, 5′-H), 7.44 (d, J = 8.7 Hz, 2 H, 2′-, 6′-H), 7.75 (s, 1 H, 2-H) ppm. ^{13}C NMR (75 MHz, $CDCl_3$): δ = 13.95, 13.99 (2 C-CH_3), 22.3, 22.4, 24.1, 24.6 (2 C-$CH_2CH_2CH_3$) 31.2, 31.3 (2 C-$O_2CCH_2CH_2$), 34.2, 34.4 (2 C-O_2CCH_2), 101.4 (C-8), 109.6 (C-6), 111.2 (C-4a), 121.7 (C-3′, -5′), 125.0 (C-3), 129.1 (C-1), 130.3 (C-2′, -6′), 150.7, 150.8 (C-4′, 8a), 152.0 (C-2), 158.7 (C-7), 161.2 (C-5), 172.9, 173.4 (2 C-O_2C), 174.8 (C-4) ppm. MS (EI): m/z = 466 (100) $[M]^+$. $C_{27}H_{30}O_7$ (466.53): calcd. C 69.51, H 6.48; found C 69.22, H 6.59.

General Procedure for the Glycosylation: To a mixture of the peracetylated glycosyl trichloroacetimidate donor **4** or **11** (3.0 mmol), isoflavonoid acceptor **3** or **9** (.20 mmol) and freshly dried molecular sieves 4 Å (2 g) in dry dichloromethane (20 mL), $BF_3{\cdot}Et_2O$ (0.073 mL, 0.6 mmol) was added under nitrogen. After stirring at room temperature for 5 h, Et_3N (0.5 mL) was added and stirring was continued for 15 min. The solids were removed by filtration through a pad of Celite®, and the filtrate was concentrated. The residue was purified by flash chromatography with petroleum ether/EtOAc (20→40%) to give the title compound.

4′-*O*-Hexanoyldaidzein-7-yl 2′′,3′′,4′′,6′′-Tetra-*O*-acetyl-β-D-glucopyranoside (7a): Compound **7a** (1.10 g, 81%) was obtained as a white solid. m.p. 182 °C. $[\alpha]_D$ = –24.4 (c = 1, $CHCl_3$). ^{1}H NMR (300 MHz, $CDCl_3$): δ = 0.92 (t, J = 7.0 Hz, 3 H, CH_3), 1.37–1.42 (m, 4 H, $CH_2CH_2CH_3$), 1.74–1.80 (m, 2 H, $O_2CCH_2CH_2$), 2.05, 2.07, 2.02, 2.08, 2.10 (4 s, 12 H, 4 O_2CCH_3), 2.57 (t, J = 7.5 Hz, 2 H, O_2CCH_2), 3.96 (ddd, J = 10.0, 5.6, 2.5 Hz, 1 H, 5-H), 4.21 (dd, J = 12.3, 2.5 Hz, 1 H, 6′′-H^a), 4.29 (dd, J = 12.3, 5.6 Hz, 1 H, 6′′-H^b), 5.15–5.35 (m, 4 H, 1′′-, 2′′-, 3′′-, 4′′-H), 7.04 (d, J = 2.4 Hz, 1 H, 8-H), 7.05 (dd, J = 8.4, 2.4 Hz, 1 H, 6-H), 7.16 (d, J = 8.7 Hz, 2 H, 3′-, 5′-H), 7.57 (d, J = 8.7 Hz, 1 H, 2′-, 6′-H), 7.97 (s, 1 H, 2-H), 8.26 (d, J = 8.4 Hz, 1 H, 5-H) ppm. ^{13}C NMR (75 MHz, $CDCl_3$): δ = 13.9 (C-CH_3), 20.69, 20.74 (4 Ac), 22.3 (C-$CH_2CH_2CH_3$), 24.6 (C-$O_2CCH_2CH_2$), 31.2 (C-$CH_2CH_2CH_3$), 34.4 (C-O_2CCH_2), 61.9 (C-6′′), 68.1 (C-4′′), 71.0 (C-2′′), 72.4, 72.5 (C-3′′, -5′′), 98.3 (C-1′′), 104.3 (C-8′), 115.4 (C-6), 120.2 (C-4a), 121.8 (C-3′, -5′), 124.8 (C-3), 128.2 (C-5), 129.1 (C-1′), 130.0 (C-2′, -6′), 150.8 (C-8a), 152.8 (C-2), 157.4 (C-4′), 160.5 (C-7), 169.2, 169.4, 170.2, 170.5 (C-O_2CCH_3), 172.3 (C-O_2CCH_2), 175.4 (C-4) ppm. MS (ES): m/z (%) = 705 (57) $[M + Na]^+$, 703 (68), 661 (5), 413 (17), 371 (100). HRMS: calcd. for $C_{35}H_{38}O_{14}Na$ 705.2159; found 705.2164. $C_{35}H_{38}O_{14}$ (682.68): calcd. C 61.58, H 5.61; found C 61.45, H 5.49.

4′-*O*-Hexanoylglycitein-7-yl 2′′,3′′,4′′,6′′-Tetra-*O*-acetyl-β-D-glucopyranoside (7c): Compound **7c** (1.08 g, 76%) was obtained as a white solid, m.p. 86 °C. $[\alpha]_D$ = –22.4 (c = 1, $CHCl_3$). ^{1}H NMR (300 MHz, $CDCl_3$): δ = 0.87 (t, J = 7.0 Hz, 3 H, CH_3), 1.31–1.36 (m, 4 H, $CH_2CH_2CH_3$), 1.71 (quint, J = 7.5 Hz, 2 H, CH_2CH_2), 1.99, 2.00, 2.03, 2.05 (4 s, 12 H, 4 O_2CCH_3), 2.50 (t, J = 7.5 Hz, 2 H, O_2CCH_2), 3.86 (s, 3 H, OCH_3), 3.81–3.87 (m, 1 H, 5′′-H), 4.14–4.25 (m, 2 H, 6′′-H^a, -H^b), 5.02–5.13 (m, 2 H, 2′′-, 3′′-H), 5.19–5.20 (m, 2 H, 1′′-, 4′′-H), 7.16 (d, J = 8.7 Hz, 2 H, 3′-, 5′-H), 7.21 (s, 1 H, 8-H), 7.58 (d, J = 8.7 Hz, 1 H, 2′-, 6′-H), 7.66 (s, 1 H, 5-H), 7.97 (s, 1 H, 2-H) ppm. ^{13}C NMR (75 MHz, $CDCl_3$): δ = 13.9 (C-CH_3), 20.6, 20.7 (4 Ac), 22.3 (C-$CH_2CH_2CH_3$), 24.6 (C-$O_2CCH_2CH_2$), 31.3 (C-$CH_2CH_2CH_3$), 34.4 (C-O_2CCH_2), 56.4 (C-OCH_3), 61.9 (C-6′′), 68.3 (C-4′′), 70.9 (C-2′′), 72.3, 72.4 (C-3′′, -5′′), 99.9 (C-1′′), 106.3 (C-8), 107.1 (C-5), 120.5 (C-4a), 121.7 (C-3′, -5′), 124.1 (C-3), 129.4 (C-1′), 130.0 (C-2′, -6′), 148.8 (C-6), 150.7 (C-8a), 150.9 (C-7), 151.2 (C-4a), 152.8 (C-2), 169.3, 169.4, 170.2, 170.6 (C-O_2CCH_3), 172.3 (C-O_2CCH_2), 175.2 (C-4) ppm. MS (ES): m/z (%) = 735 (100) $[M + Na]^+$, 713 (17) $[M + H]^+$, 661 (7), 405 (23), 383 (12), 371 (11). HRMS: calcd. for $C_{36}H_{40}O_{15}Na$ 735.2265; found 735.2247. $C_{36}H_{40}O_{15}$ (712.70): calcd. C 60.67, H 5.66; found C 60.50, H 5.22.

5,4′-*O*-Diacetylgenistein-7-yl 2′′,3′′,4′′,6′′-Tetra-*O*-acetyl-β-D-glucopyranoside (10a): Compound **10a** (0.985 g, 72%) was obtained as a white solid, m.p. 169 °C. $[\alpha]_D$ = –24.7 (c = 1, $CHCl_3$). ^{1}H NMR (300 MHz, $CDCl_3$): δ = 2.05, 2.07, 2.08, 2.11 (4 s, 12 H, 4 O_2CCH_3), 2.31 (s, 3 H, 4′-O_2CCH_3), 2.41 (s, 3 H, O_2CCH_3), 3.96 (ddd, J = 12.5, 5.7, 2.7 Hz, 1 H, 5′′-H), 4.20 (dd, J = 12.3, 2.6 Hz, 1 H, 6′′-H^a), 4.29 (dd, J = 12.3, 5.7 Hz, 1 H, 6′′-H^b), 5.13–5.23 (m, 2 H, 3′′-, 2′′-H), 5.31–5.34 (m, 2 H, 1′′-, 4′′-H), 6.69 (d, J = 2.4 Hz, 1 H, 8-H), 6.93 (d, J = 2.4 Hz, 1 H, 6-H), 7.15 (d, J =

8.7 Hz, 2 H, 3′-, 5′-H), 7.49 (d, *J* = 8.7 Hz, 1 H, 2′-, 6′-H), 7.85 (s, 1 H, 2-H) ppm. ^{13}C NMR (75 MHz, $CDCl_3$): δ = 20.6, 21.2 (6 O_2CCH_3), 61.90 (C-6′′), 68.0 (C-4′′), 70.8 (C-2′′), 72.4 (C-3′′, -5′′), 98.0 (C-8), 102.5 (C-6), 109.8 (C-1), 113.7 (C-4a), 121.7 (C-3′, -5′), 125.6 (C-3), 128.8 (C-1′), 130.2 (C-2′, -6′), 150.7 (C-8a), 151.7 (C-4′), 151.8 (C-2), 158.4 (C-7), 159.8 (C-5), 169.2, 169.4, 170.1, 170.5, 172.4 (C-O_2CCH_3), 175.8 (C-4) ppm. MS (ES): *m*/*z* (%) = 707 (100) [M + Na]$^+$, 661 (48), 371 (38). HRMS: calcd. for $C_{33}H_{32}O_{16}Na$ 707.1563; found 707.1588. $C_{33}H_{32}O_{16}$ (684.61): calcd. C 57.90, H 4.51; found C 57.92, H 4.41.

5,4′-*O*-Dihexanoylgenistein-7-yl 2′′,3′′,4′′,6′′-Tetra-*O*-acetyl-β-D-glucopyranoside (10b): Compound **10b** (1.38 g, 87%) was obtained as a white solid, m.p. 174 °C. $[\alpha]_D$ = –26.3 (*c* = 1, $CHCl_3$). ^{1}H NMR (300 MHz, $CDCl_3$): δ = 0.85–0.90 (m, 6 H, 2 CH_3), 1.35–1.42 (m, 8 H, 2 $CH_2CH_2CH_3$), 1.72–1.82 (m, 4 H, 2 $O_2CCH_2CH_2$), 2.05, 2.07, 2.08, 2.12 (4 s, 12 H, 4 O_2CCH_3), 2.56 (t, *J* = 7.5 Hz, 2 H, O_2CCH_2), 2.72 (t, *J* = 7.8 Hz, 2 H, O_2CCH_2), 3.96 (ddd, *J* = 12.3, 5.7, 2.6 Hz, 1 H, 5′′-H), 4.20 (dd, *J* = 12.3, 2.6 Hz, 1 H, 6′′-H^a), 4.29 (dd, *J* = 12.3, 5.6 Hz, 1 H, 6′′-H^b), 5.13–5.23 (m, 2 H, 4′′-, 2′′-H), 5.31–5.34 (m, 2 H, 1′′-, 3′′-H), 6.67 (d, *J* = 2.4 Hz, 1 H, 8-H), 6.92 (d, *J* = 2.4 Hz, 1 H, 6-H), 7.13 (d, *J* = 8.7 Hz, 2 H, 3′-, 5′-H), 7.48 (d, *J* = 8.7 Hz, 2 H, 2′-, 6′-H), 7.84 (s, 1 H, 2-H) ppm. ^{13}C NMR (75 MHz, $CDCl_3$): δ = 13.9, 14.0 (C-CH_3), 20.59, 20.63 (4 O_2CCH_3), 22.3, 22.4 (C-$CH_2CH_2CH_3$), 24.1, 24.6 (C-$O_2CCH_2CH_2$), 31.27, 31.31 (C-$CH_2CH_2CH_3$), 34.1, 34.4 (C-O_2CCH_2), 61.90 (C-6′′), 68.1 (C-4′′), 70.9 (C-2′′), 72.5 (C-3′′, -5′′), 98.1 (C-1′′), 102.3 (C-8), 109.8 (C-6), 113.7 (C-4a), 121.7 (C-3′, -5′), 125.6 (C-3), 128.8 (C-1′), 130.2 (C-2′,6′), 150.9 (C-8a), 151.4 (C-4′), 151.7 (C-2), 158.4 (C-7), 159.8 (C-5), 169.2, 169.4, 170.1, 170.5 (C-O_2CCH_3), 172.4 (C-O_2CCH_2), 175.8 (C-4) ppm. MS (ES): *m*/*z* (%) = 819 (100) [M + Na]$^+$, 803 (23). HRMS: calcd. for $C_{41}H_{48}O_{16}Na$ 819.2840; found 819.2854. $C_{41}H_{48}O_{16}$ (796.82): calcd. C 61.60, H 6.07; found C 61.22, H 5.69.

Methyl (4′-*O*-Hexanoyldaidzein-7-yl β-D-2′′,3′′,4′′-triacetylglucopyranosid)urinate (11a): Compound **11a** (1.08 g, 81%) was obtained as a white solid, m.p. 205 °C. $[\alpha]_D^{20}$ = –24.2 (0.001 g mL^{-1} in $CHCl_3$). ^{1}H NMR (300 MHz, $CDCl_3$): δ = 0.94 (t, *J* = 6.9 Hz, 3 H, CH_2CH_3), 1.37–1.42 (m, 4 H, $CH_2CH_2CH_3$), 1.72–1.82 (m, 2 H, CH_2CH_2), 2.04, 2.06, 2.08 (3s, 3×3 H, 3 $COCH_3$), 2.57 (t, *J* = 7.5 Hz, 2 H, $O_2CCH_2CH_2$), 3.73 (s, 3 H, CO_2CH_3), 4.28 (d, *J* = 9 Hz, 1 H, 5′′-H), 5.31–5.40 (m, 4 H, 1′′,2′′3′′,4′′-H), 7.04 (d, *J* = 2.4 Hz, 1 H, 8-H), 7.06 (dd, *J* = 9.3, 2.4 Hz, 1 H, 6-H), 7.15 (d, *J* = 9.0 Hz, 2 H, 3′,5′-H), 7.57 (d, *J* = 9.0 Hz, 2 H, 2′,6′-H), 7.97 (s, 1 H, 2-H), 7.25 (d, *J* = 9.3 Hz, 1 H, 5-H) ppm. MS (ES): *m*/*z* (%) = 691 (87) [M + Na]$^+$, 669 (14) [M + H]$^+$, 357 (100). $C_{34}H_{36}O_{14}$ (668.65): calcd. C 61.07; H, 5.43; found C 60.70; H, 5.58.

Methyl (5,4′-*O*-Dihexanoylgenistein-7-yl β-D-2′′,3′′,4′′-triacetylglucopyranosid)urinate (11b): Compound **9b** (1.38 g, 88%) was obtained as a white solid; m.p. 94 °C. $[\alpha]_D$ = –25.6 (*c* = 1, $CHCl_3$). ^{1}H NMR (300 MHz, $CDCl_3$): δ = 0.89–0.95 (m, 6 H, 3 CH_3), 1.37–1.45 (m, 8 H, 2 $CH_2CH_2CH_3$), 1.72–1.82 (m, 4 H, 2 $O_2CCH_2CH_2$), 2.06, 2.07, 2.08 (3 s, 9 H, 3 O_2CCH_3), 2.56 (t, *J* = 7.5 Hz, 2 H, O_2CCH_2), 2.72 (t, *J* = 7.8 Hz, 2 H, O_2CCH_2), 3.73 (s, 3 H, -CO_2CH_3), 4.29 (m, 1 H, 5′′-H), 5.28–5.43 (m, 4 H, 1′′-, 2′′-, 3′′-, 4′′-H), 6.67 (d, *J* = 2.4 Hz, 1 H, 8-H), 6.92 (d, *J* = 2.4 Hz, 1 H, 6-H), 7.13 (d, *J* = 8.7 Hz, 2 H, 3′-, 5′-H), 7.48 (d, *J* = 8.7 Hz, 1 H, 2′-, 6′-H), 7.84 (s, 1 H, 2-H) ppm. ^{13}C NMR (75 MHz, $CDCl_3$): δ = 13.95, 14.0 (C-CH_3), 20.5, 20.6 (3 O_2CCH_3), 22.3, 22.4 (C-$CH_2CH_2CH_3$), 24.1, 24.6 (C-$O_2CCH_2CH_2$), 31.28, 31.32 (C-$CH_2CH_2CH_3$), 34.1, 34.4 (C-O_2CCH_2), 53.2 (C-CO_2CH_3), 68.7 (C-4′′), 70.8 (C-5′′), 71.4 (C-3′′), 72.8 (C-2′′), 98.0 (C-1′′), 102.3 (C-6), 109.8 (C-8), 113.8 (C-4a), 121.7 (C-3′, -5′), 125.6 (C-3), 128.8 (C-1′), 130.2 (C-2′, -6′), 150.9 (C-4′), 151.4 (C-8a), 151.8 (C-2), 158.4 (C-7), 159.6 (C-5), 166.7 (C-CO_2Me), 169.2, 169.3, 170.0, 172.3, 172.4 (3 O_2CCH_3, 3 O_2CCH_2), 174.1 (C-4) ppm. MS (ES): *m*/*z* (%) = 805 (100) [M + Na]$^+$, 783 (12). HRMS: calcd. for $C_{40}H_{46}O_{16}Na$ 805.2684; found 805.2665. $C_{40}H_{46}O_{16}$ (782.79): calcd. C 61.37, H 5.92; found C 60.89, H 5.50.

Methyl (4′-*O*-Hexanoylglycitein-7-yl β-D-2′′,3′′,4′′-triacetylglucopyranosid)urinate (11c): Compound **11c** (1.23 g, 88%) was obtained as a white solid; m.p. 129 °C. $[\alpha]_D$ = –39.1 (*c* = 1, $CHCl_3$). ^{1}H NMR (300 MHz, $CDCl_3$): δ = 0.92 (t, *J* = 7.0 Hz, 3 H, CH_3), 1.37–1.51 (m, 4 H, $CH_2CH_2CH_3$), 1.72–1.85 (quint, *J* = 7.5 Hz, 2 H, CH_2CH_2), 2.06, 2.07, 2.1 (3 s, 9 H, 3 O_2CCH_3), 2.60 (t, *J* = 7.5 Hz, 2 H, O_2CCH_2), 3.93 (s, 3 H, OCH_3), 4.16–4.24 (m, 1 H, 5′′-H), 5.28–5.43 (m, 4 H, 1′′-, 2′′-, 3′′-, 4′′-H), 7.16 (d, *J* = 8.7 Hz, 2 H, 3′-, 5′-H), 7.21 (s, 1 H, 8-H), 7.58 (d, *J* = 8.7 Hz, 2 H, 2′-, 6′-H), 7.66 (s, 1 H, 5-H), 7.97 (s, 1 H, 2-H) ppm. ^{13}C NMR (75 MHz, $CDCl_3$): δ = 13.9 (C-CH_3), 20.5, 20.6, 20.08 (3 O_2CCH_3), 22.3 (C-$CH_2CH_2CH_3$), 24.6 (C-$O_2CCH_2CH_2$), 31.2 (C-$CH_2CH_2CH_3$), 34.4 (C-O_2CCH_2), 53.0, (C-CO_2CH_3), 56.4 (C-OCH_3), 68.9 (C-4′′), 70.3 (C-5′′), 71.7, 72.7 (C-2′′, -3′′), 99.6 (C-1′′), 106.3 (C-8), 107.7 (C-5), 120.7 (C-4a), 121.7 (C-3′, -5′), 124.0 (C-3), 129.4 (C-1′), 130.0 (C-2′, -6′), 148.9 (C-5), 150.5 (C-8a), 150.7 (C-7), 151.1 (C-4a), 152.8 (C-2), 166.8, 169.2, 169.3, 170.0, 170.6 (C-O_2CCH_3), 172.3 (C-O_2CCH_2), 175.2 (C-4) ppm. MS (ES): *m*/*z* (%) = 735 (100) [M + Na]$^+$, 713 (17) [M + H]$^+$, 661 (7), 405 (23), 383 (12), 371 (11). HRMS: calcd. for $C_{36}H_{40}O_{15}Na$ 735.2265; found 735.2247. $C_{35}H_{38}O_{15}$ (698.68): calcd. C 60.17, H 5.48; found C 60.50, H 5.05.

Methyl (5,4′-*O*-Diacetylgenistein-7-yl β-D-2′′,3′′,4′′-triacetylglucopyranosid)urinate (11d): Compound **11d** (1.00 g, 75%) was obtained as a white foam; m.p. 205 °C. $[\alpha]_D$ = –19.4 (*c* = 1, $CHCl_3$). ^{1}H NMR (500 MHz, $CDCl_3$): δ = 2.07 (s, 6 H, 2 O_2CCH_3), 2.08, 2.31, 2.42 (3 s, 9 H, 3 O_2CCH_3), 3.74 (s, 3 H, CO_2CH_3), 4.28 (d, *J* = 9.2 Hz, 1 H, 5′′-H), 5.31–5.33 (m, 2 H, 2′′-, 3′′-H), 5.37–5.39 (m, 2 H, 1′′-, 4′′-H), 6.70 (d, *J* = 2.4 Hz, 1 H, 8-H), 6.94 (d, *J* = 2.4 Hz, 1 H, 6-H), 7.15 (d, *J* = 8.6 Hz, 2 H, 3′-, 5′-H), 7.49 (d, *J* = 8.6 Hz, 1 H, 2′-, 6′-H), 7.86 (s, 1 H, 2-H) ppm. ^{13}C NMR (75 MHz, $CDCl_3$): δ = 20.55, 20.64, 20.65, 21.2 (5 O_2CCH_3), 53.2 (C-OCH_3), 68.7 (C-5′′), 70.7 (C-2′′), 71.4 (C-3′′), 72.8 (C-4′′), 98.0 (C-1′′), 102.5 (C-6), 109.8 (C-8), 113.7 (C-4a), 121.8 (C-3′, -5′), 125.6 (C-3), 128.9 (C-1′), 130.3 (C-2′, -6′), 150.8 (C-8a), 151.2 (C-7), 151.9 (C-2), 158.4 (C-4′), 159.7 (C-5), 166.3 (C-O_2CCH_3), 169.2, 169.3, 169.4, 169.6, 170.0 (5 O_2CCH_3), 174.2 (C-4) ppm. MS (ES): *m*/*z* (%) = 693 (100) [M + Na]$^+$, 671 (12). HRMS: calcd. for $C_{32}H_{30}O_{16}Na$ 693.1432; found 693.1436. $C_{32}H_{30}O_{16}$ (670.58): calcd. C 57.32, H 4.51; found C 56.91, H 4.92.

General Procedure for Deprotection of the Peracylated Isoflavone Glycosides: A mixture of peracylated isoflavone glycoside and K_2CO_3 (2 equiv.) was dissolved in a solution of MeOH/THF/H_2O at room temp. under nitrogen. After stirring at 40 °C for 5 h, the mixture was cooled to room temp., neutralised with Dowex-50 H$^+$, and then filtered and concentrated. The pale yellow solid was purified by preparative HPLC using H_2O/CH_3CN (7:3) for the glycosides and H_2O/CH_3CN (9:1) for the glucuronides.

Daidzein-7-yl β-D-Glucopyranoside (Daidzin) (2a): Compound **2a** (0.387 g, 93%) was obtained as a white solid; m.p. 236–237 °C (ref.[33] 235–237 °C). $[\alpha]_D$ = –24.6 (*c* = 1, DMSO) {ref.[20] $[\alpha]_D$ = –24.6 (*c* = 1, DMSO)}. ^{1}H NMR (300 MHz, [D$_6$]DMSO): δ = 3.17–3.21 (m, 1 H, 4′′-H), 3.28–3.36 (m, 2 H, 2′′-, 3′′-H), 3.45–3.50 (m, 2 H, 5′′-, 6b′′-H), 3.71–3.74 (m, 1 H, 6a′′-H), 4.65 (t, *J* = 5.5 Hz, 1 H, 6′′-OH), 5.115 (d, *J* = 7.0 Hz, 1 H, 1′′-H), 5.12 (d, *J* = 5.2 Hz, 1 H, 4′′-OH), 5.19 (d, *J* = 4.6 Hz, 1 H, 3′′-OH), 5.48 (d, *J* = 4.7 Hz, 1 H, 2′′-OH), 6.83 (d, *J* = 8.6 Hz, 2 H, 3′-, 5′-H),

7.15 (dd, *J* = 8.9, 2.2 Hz, 1 H, 6-H), 7.24 (d, *J* = 2.2 Hz, 2 H, 8-H), 7.42 (d, *J* = 8.6 Hz, 2 H, 2′-, 6′-H), 8.06 (d, *J* = 8.9 Hz, 1 H, 5-H), 8.58 (s, 1 H, 2-H), 9.58 (s, 1 H, 4′-OH) ppm. ^{13}C NMR (75 MHz, [D$_6$]DMSO): δ = 60.6 (C-6′′), 69.6 (C-4′′), 73.1 (C-2′′), 76.5 (C-3′′), 77.2 (C-5′′), 99.9 (C-1′′), 103.3 (C-8), 115.0 (C-3′, -5′), 115.6 (C-6), 118.4 (C-4a), 122.3 (C-1′), 123.7 (C-3), 127.0 (C-5), 130.1 (C-2′, -6′), 153.4 (C-2), 157.0 (C-8a), 157.3 (C-4′), 161.4 (C-7), 174.7 (C-4) ppm. MS (ES): *m*/*z* (%) = 439 (100) [M + Na]$^+$. HRMS: calcd. for $C_{21}H_{20}O_9Na$ 439.1005; found 439.1010.

Genistein-7-yl β-D-Glucopyranoside (Genistin) (2b): Compound **2b** (0.406 g, 94%) was obtained as a white solid; m.p. 256 °C (ref.[27] 254–255 °C). $[\alpha]_D$ = –28.7 (*c* = 1, DMSO) {ref.[26] $[\alpha]_D$ = –32.0 (*c* = 1, DMSO)}. ^{1}H NMR (500 MHz, [D$_6$]DMSO): δ = 3.13–3.17 (m, 1 H, 4′′-H), 3.26–3.32 (m, 2 H, 2′′-, 3′′-H), 3.43–3.48 (m, 2 H, 5′′-, 6′′-H^b), 3.67–3.72 (m, 2 H, 6′′-H^a), 4.63 (t, *J* = 5.5 Hz, 1 H, 6′′-OH), 5.07 (d, *J* = 7.0 Hz, 1 H, 1′′-H), 5.10 (d, *J* = 5.0 Hz, 1 H, 4′′-OH), 5.17 (d, *J* = 5.0 Hz, 1 H, 3′′-OH), 5.44 (d, *J* = 5.0 Hz, 1 H, 2′′-OH), 6.47 (d, *J* = 2.2 Hz, 1 H, 6-H), 6.72 (d, *J* = 2.2 Hz, 1 H, 6-H), 6.83 (d, *J* = 8.5 Hz, 2 H, 3′-, 5′-H), 7.40 (d, *J* = 8.5 Hz, 2 H, 2′-, 6′-H), 8.43 (s, 1 H, 2-H), 9.62 (s, 1 H, 4′-OH), 12.97 (s, 1 H, 5-OH) ppm. ^{13}C NMR (125 MHz, [D$_6$]DMSO): δ = 60.6 (C-6′′), 69.6 (C-4′′), 73.1 (C-2′′), 76.4 (C-3′′), 77.2 (C-5′′), 94.5 (C-8), 99.6 (C-6), 99.8 (C-1′′), 106.1 (C-4a), 115.1 (C-3′, -5′), 121.0 (C-1′), 122.5 (C-3), 130.2 (C-2′, -6′), 154.6 (C-2), 157.2 (C-8a), 157.5 (C-4′), 161.6 (C-5), 163.0 (C-7), 180.5 (C-4) ppm. MS (ES): *m*/*z* (%) = 455 (100) [M + Na]$^+$. HRMS: calcd. for $C_{21}H_{20}O_{10}Na$ 455.0954; found 455.0949.

Glycitein-7-yl β-D-Glucopyranoside (Glycitin) (2c): Compound **2c** (0.419 g, 94%) was obtained as a white solid; m.p. 203–204 °C (ref.[34] = 192–195 °C). $[\alpha]_D$ = –39.4 (*c* = 1, DMSO) {ref.[34] $[\alpha]_D$ = –13.4 (*c* = 3.65, DMF)}. ^{1}H NMR (500 MHz, [D$_6$]DMSO): δ = 3.17–3.20 (m, 1 H, 4′′-H), 3.26–3.33 (m, 2 H, 2′′-, 3′′-H), 3.44–3.49 (m, 2 H, 5′′-H, 6′′-H^b), 3.68–3.72 (m, 2 H, 6′′-H^a), 3.88 (s, 3 H, OCH$_3$), 4.56 (br. s, 1 H, 6′′-OH), 5.05 (br. s, 2 H, 3′′-, 4′′-OH), 5.17 (d, *J* = 7.5 Hz, 1 H, 1′′-H), 5.36 (br. s, 1 H, 4′′-OH), 6.82 (d, *J* = 8.5 Hz, 2 H, 3′-, 5′-H), 7.32 (s, 1 H, 8-H), 7.41 (d, *J* = 8.5 Hz, 2 H, 2′-, 6′-H), 7.49 (s, 1 H, 5-H), 8.37 (s, 1 H, 2-H), 9.62 (s, 1 H, 4′-OH) ppm. ^{13}C NMR (125 MHz, [D$_6$]DMSO): δ = 55.8 (C-O*C*H$_3$), 60.6 (C-6′′), 69.6 (C-4′′), 73.0 (C-2′′), 76.7 (C-3′′), 77.2 (C-5′′), 99.6 (C-1′′), 103.4 (C-8), 104.7 (C-5), 114.9 (C-3′, -5′), 117.8 (C-4a), 122.5 (C-1′), 123.1 (C-3), 130.0 (C-2′, -6′), 147.4 (C-6), 151.2 (C-8a), 151.5 (C-7), 153.0 (C-2), 157.2 (C-4), 174.3 (C-4) ppm. MS (ES): *m*/*z* (%) = 469 (100) [M + Na]$^+$. HRMS: calcd. for $C_{42}H_{22}O_{10}Na$ 469.1111; found 469.1116.

(Daidzein-7-yl β-D-glucopyranosid)uronic Acid (12a): Compound **12a** (0.39 g, 91%) was obtained as a white solid. m.p. 186–187 °C. $[\alpha]_D$ = –70.7 (*c* = 1, H$_2$O). ^{1}H NMR (500 MHz, D$_2$O): δ = 3.48–3.55 (m, 3 H, 2′′-, 3′′-, 4′′-H), 3.76 (d, *J* = 9.0 Hz, 1 H, 5′′-H), 4.86 (d, *J* = 7.5 Hz, 1 H, 1′′-H), 6.75 (d, *J* = 8.5, Hz, 2 H, 3′-, 5′-H), 6.90 (d, *J* = 2.5 Hz, 1 H, 8-H), 6.94 (dd, *J* = 9.0, 2.5 Hz, 1 H, 6-H), 7.12 (d, *J* = 8.5 Hz, 2 H, 2′-, 6′-H), 7.72 (d, *J* = 9.0 Hz, 1 H, 5-H), 7.86 (s, 1 H, 2-H) ppm. ^{13}C NMR (125 MHz, D$_2$O): δ = 71.5 (C-4′′), 72.4 (C-2′′), 75.0 (C-3′′), 76.1 (C-4′′), 99.4 (C-1′′), 103.5 (C-8), 115.3 (C-3′, -5′), 115.8 (C-6), 118.1 (C-4a), 122.8 (C-1′), 123.4 (C-3), 126.8 (C-5), 130.3 (C-2′, -6′), 154.7 (C-2), 155.7 (C-4′), 157.1 (C-8a), 161.0 (C-7), 175.1 (C-6′′), 177.8 (C-4) ppm. MS (ES): *m*/*z* (%) = 429 (100) [M – H]$^-$. HRMS: calcd. for $C_{21}H_{17}O_{10}$ 429.0822; found 429.0829.

(Genistein-7-yl β-D-glucopyranosid)uronic Acid (12b): Compound **12b** (0.414 g, 93%) was obtained as a white solid; m.p. 212–213 °C. $[\alpha]_D$ = –27.6 (*c* = 1, H$_2$O). ^{1}H NMR (500 MHz, [D$_6$]DMSO): δ = 3.12 (dd, *J* = 10.0, 9.0 Hz, 1 H, 4′′-H), 3.23–3.30 (m, 2 H, 2′′-, 3′′-H), 3.50 (d, *J* = 10.0 Hz, 1 H, 5′′-H), 5.03 (d, *J* = 7.5 Hz, 1 H, 1′′-H), 5.06 (d, *J* = 4.5 Hz, 1 H, 4′′-OH), 5.35 (d, *J* = 4.5 Hz, 1 H, 3′′-OH), 6.47 (d, *J* = 2.2 Hz, 1 H, 8-H), 6.72 (d, *J* = 2.2 Hz, 1 H, 6-H), 6.83 (d, *J* = 8.4 Hz, 2 H, 3′-, 5′-H), 7.38 (br. s, 1 H, COOH), 7.38 (d, *J* = 8.4 Hz, 2 H, 2′-, 6′-H), 8.42 (s, 1 H, 2-H), 9.77 (s, 1 H, 4′-OH), 12.94 (s, 1 H, 5-OH) ppm. ^{13}C NMR (125 MHz, [D$_6$]-DMSO): δ = 72.0 (C-4′′), 72.9 (C-2′′), 73.5 (C-3′′), 76.5 (C-5′′), 94.5 (C-8), 99.51, 99.54 (C-1′′, -6), 105.9 (C-4a), 115.1 (C-3′, -5′), 120.7 (C-1′), 122.4 (C-3), 130.0 (C-2′, -6′), 154.5 (C-2), 157.1 (C-8a), 157.7 (C-4′), 161.5 (C-5), 163.1 (C-7), 171.7 (C-*C*O$_2$H), 180.5 (C-4). MS (ES): *m*/*z* (%) = 445 (100) [M – H]$^-$. HRMS: calcd. for $C_{21}H_{17}O_{11}$ 445.0771; found 445.0768.

(Glycitein-7-yl β-D-glucopyranosid)uronic Acid (12c): Compound **12c** (0.42 g, 92%) was obtained as a white solid; m.p. 204–206 °C. $[\alpha]_D$ = –47.6 (*c* = 1, H$_2$O). ^{1}H NMR (500 MHz, D$_2$O): δ = 3.57 (quint, *J* = 9.1, 7.2 Hz, 2 H, 3′′-, 4′′-H), 3.65 (t, *J* = 8.1 Hz, 1 H, 2′′-H), 3.70 (s, 3 H, OCH$_3$), 3.83 (d, *J* = 11.5 Hz, 1 H, 5′′-H), 4.87 (d, *J* = 8.1 Hz, 1 H, 1′′-H), 6.83 (d, *J* = 8.2 Hz, 2 H, 3′-, 5′-H), 6.94 (s, 1 H, 8-H), 7.02 (s, 1 H, 5-H), 7.20 (d, *J* = 8.2 Hz, 2 H, 2′-, 6′-H), 7.87 (s, 1 H, 2-H) ppm. ^{13}C NMR (125 MHz, D$_2$O): δ = 58.3 (C-O*C*H$_3$), 74.1 (C-4′′), 74.8 (C-2′′), 77.6 (C-3′′), 78.9 (C-5′′), 102.1 (C-1′′), 106.0 (C-8), 107.0 (C-4a), 117.8 (C-3′, -5′), 120.3 (C-1′), 125.2 (C-5), 125.6 (C-3), 132.7 (C-2′, -6′), 149.3 (C-6), 153.3 (C-8a), 154.0 (C-7), 156.8 (C-2), 158.1 (C-4), 177.6 (CO$_2$H), 179.1 (C-4) ppm. MS (ES): *m*/*z* (%) = 459 (100) [M – H]$^-$. HRMS: calcd. for $C_{22}H_{19}O_{11}$ 459.0927; found 459.0933.

Acknowledgments

This work was funded by the Food Standards Agency (FSA) under contract T05028.

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Received: August 18, 2008
Published Online: October 13, 2008

