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Isolation and Synthesis of β-Miroside An Antifungal Furanone Glucoside from *Prumnopitys ferruginea*.

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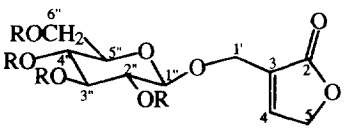
Abstract - The bioactivity-directed isolation and structural determination of β-miroside {3-[(β-D-glucopyranosyloxy)-methyl]-2(5H)-furanone} from the New Zealand gymnosperm *Prumnopitys ferruginea*, is described. Piccin {4-(β-D-glucopyranosyloxy)acetophenone} was also isolated. Syntheses of β-miroside, its α-anomer and an isomer with an exocyclic double bond, are described, along with an unusual reaction of an exocyclic alkoxyalkylidene lactone with N-bromosuccinimide to give a product with an endocyclic double bond. β-Miroside shows antifungal, antibacterial and cytotoxic activities.

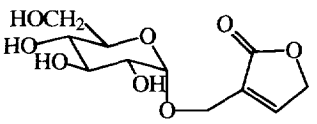
INTRODUCTION

During screening of New Zealand plants and fungi for biologically active natural products[1], a foliage extract of *Prumnopitys ferruginea* (D. Don) Laubenf.[2] (family Podocarpaceae) was found to exhibit antifungal activity.

P. ferruginea (miro) is a forest canopy tree with wide distribution throughout New Zealand, most frequently found in lowland forest. Previous chemical investigations on miro foliage have mainly centred on the essential oil[3-5], which is rich in terpenes. The flavonoid glycosides have also been studied[6]. We recently found mild antimicrobial activity for some essential oil components[5] which might explain the reported antiseptic activity of miro resin[7,8]. We now report the bioactivity-directed isolation of the new glucoside, β-miroside (**1**), as the major antifungal component of *P. ferruginea* foliage, along with the synthesis of β-miroside, its α-anomer (**2**) and an isomer with an exocyclic double bond (**3**).



(1) R = H
(4) R = OAc



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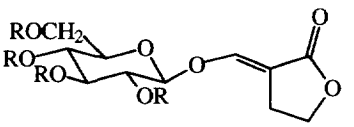


(3) R = H
(11) R = Ac

BIOACTIVITY-DIRECTED ISOLATION OF β-MIROSIDE

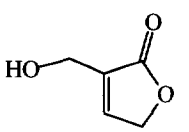
Screening assays showed that a small-scale extract of *P. ferruginea* foliage was active against the dermatophyte fungus *Trichophyton mentagrophytes*, the yeast *Candida albicans*, the Gram-positive bacterium *Bacillus subtilis* and the Gram-negative bacteria *Escherichia coli* and *Pseudomonas aeruginosa*. Cytotoxicity was observed against P-388 leukaemia cells and monkey kidney cells (BSC). The extract did not inhibit the cytopathic effects of either Herpes simplex Type I or Polio Type I viruses[9].

Preliminary chromatographic studies showed good recovery of the antifungal and P-388 activity in polar fractions from octadecyl-bonded reverse-phase (C_{18} rp) and Si gel columns. Activity against *T. mentagrophytes* was used to direct the isolation of the major active component. A larger-scale extract was chromatographed over a C_{18} rp column, with the most active fractions further chromatographed over a diol bonded Si gel column, and finally over another C_{18} rp column to give a single active compound (**1**), as a clear glass. The level of this compound in the plant was about 4 mg/g of dried foliage.

The ^{13}C NMR spectrum revealed that (**1**) contained eleven carbons comprising three methylenes, six methines and two quaternary carbons. Six of the signals were consistent with the presence of a glycoside. This was supported by the ^{1}H NMR spectrum, and by the presence in the mass spectrum of a peak at 163 corresponding to a characteristic $C_6H_{11}O_5$ fragment. No molecular ion was observed in the mass spectrum, under electron impact or chemical ionisation conditions. Treatment of (**1**) with the purified enzyme exoglucanase produced complete hydrolysis of the glucosidic linkage. As the activity of exoglucanase is specific to β-glucosidic linkages[10], compound (**1**) must be a β-glucoside.

Acetylation of glycoside (**1**) with acetic anhydride and pyridine gave a tetraacetate (**4**) whose mass spectrum again showed no parent ion but revealed a M^{+}-CH_2OAc fragment, consistent with an acetylated glucoside. NOE experiments on (**4**) revealed enhancements between the anomeric proton signal and peaks which could be assigned to H-3" and H-5", again consistent with a β-glucoside[11].

Once the signals for the glucose unit had been assigned in the ^{13}C NMR spectrum of (**1**), five signals remained. These were consistent with the aglycone unit, 3-(hydroxymethyl)-2(5H)-furanone (**5**). Four signals (δ 7.80, 1H, quin; 4.99, 2H, q; 4.59, 1H, dq and 4.49, 1H, dq) in the ^{1}H NMR spectrum were of appropriate multiplicity to accommodate this structure, while the presence of a carbonyl resonance in the IR spectrum at 1740 cm^{-1} and a UV λ_{max} of 213 nm (ε 5300) were also consistent with an α,β-unsaturated-γ-lactone system. This was confirmed by hydrolysis of (**1**) with trifluoroacetic acid which gave a sample of (**5**), identical to that described previously[12-15]. Compound (**1**), a new glucoside, has been given the name β-miroside.



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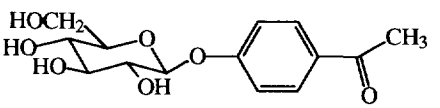
A foliage extract of the only other *Prumnopitys* species found in New Zealand, *P. taxifolia* (D. Don) Laubenf.[2] (matai), also showed cytotoxic and antimicrobial activity in our testing, but we were unable to detect the signals of β-miroside in the ^{1}H NMR spectrum of the D_2O solubles from this extract.

Aglycone (**5**) has been isolated from the moth, *Yponomeuta cagnagellus* (Lepidoptera), and from its host tree, *Euonymus europaeus* (Celastraceae), as well as from six other similar moth species which do not feed on *E. europaeus*[12]. Compound (**5**), known as isosiphonodin, has also been isolated from the roots of *Sedum telephium* (Crassulaceae)[13]. Two syntheses of (**5**) have been reported[14,15].

Picein (**6**) was isolated during the purification of miroside and was readily identified by comparison with literature data[16,17]. The level of picein was 10 mg/g of dried foliage.



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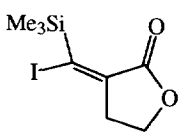
SYNTHESIS OF β-MIROSIDE

Three approaches were made to the synthesis of β-miroside (**1**). Following our experience in the synthesis of the *exo*-methylene-γ-lactones[18] and their subsequent derivatisation[19], we proposed to synthesise the iodomethylene lactone (**7**) by free radical cyclisation of the iodo acetylenic ester (**8**) and introduce the glucose system by nucleophilic replacement of the iodine. Subsequent double bond migration would lead to (**1**).

Iodoalkylidene lactone (**7**) was prepared readily by bubbling ethylene through a solution of N-iodosuccinimide and trimethylsilylpropynoic acid in CH_2Cl_2, followed by dibenzoyl peroxide induced cyclisation of the ensuing iodo ester (**8**)[18]. Our past experience has shown this reaction to give exclusively the (*E*)-isomer in almost all cases. We did not pursue this approach further, however, as trial reactions between glucose tetraacetate and the analogous cyclopentyl fused lactone (**9**) using various neutral, basic and acidic (Lewis acid) conditions gave no glycoside coupling. Although the iodoalkylidene lactones react readily with soft nucleophiles[19], they do not appear to do so with oxygen-centred nucleophiles. In view of these results, an alternative route to β-miroside was undertaken.



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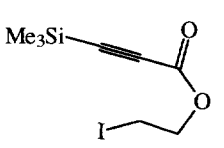

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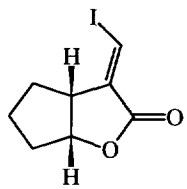

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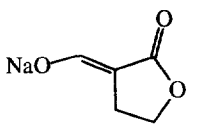
An alternative route to an *exo*-methylene glycoside involved reaction of sodium salt (**10**), with acetobromoglucose to yield the acetylated glycoside (**11**) which has subsequently been reported[20]. This reaction was conducted in DMSO[21] in view of the insolubility of the sodium salt in the non-protic solvents which are typically used for silver triflate activated glycosylations. The yield of (**11**) was 55%, a significant improvement over that reported from the reaction of (**10**) with acetobromoglucose in aqueous acetone[20]. Glucoside (**11**) was effectively de-acetylated with hexabutylditin oxide in methanol to give the free glycoside (**3**) whose ^{1}H NMR spectrum showed the alkoxymethylene proton signal at δ 7.43, in good agreement with data for other (*E*)-alkoxymethylenebutyrolactones[22].



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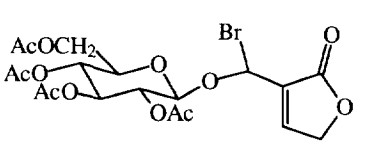
Double bond migrations have been well documented in the literature[23-26] and can be achieved by a number of methods. Catalytic transition metal systems which have been used in a number of instances include Wilkinson's catalyst [Rh(I)$(Ph_3P)_3Cl$][23] and $RhCl_3$ in ethanol[27]. However, treatment of glycoside (**11**) with Wilkinson's catalyst in various solvents and at various temperatures gave no reaction. Similarly, $RhCl_3$ in boiling ethanol also produced no change. Likewise, an attempted ene reaction with N-sulphinyl-*p*-toluenesulfonamide[28,29] left the starting material intact.

The comparative energies of the exocyclic and endocyclic alkoxymethylene lactone isomers were determined by molecular modelling. This predicted the exocyclic isomers to be of lower energy. However, it seemed likely that either an allylic radical or an allylic cation might have more driving force for attaining the endocyclic form where stabilisation from the oxygen might ensue. With this in mind, glycoside (**11**) was reacted with N-bromosuccinimide. NMR analysis of the product showed that double bond migration had indeed occurred to give only one diastereoisomer of the bromo derivative (**12**). Compound (**12**) was found to be a temperature sensitive substance which readily decomposed to give aldehyde (**13**). Analysis by mass spectrometry proved unsuccessful in EI, CI or FAB modes. Under FAB conditions, a peak at 331 Da, attributable to the cleaved glycoside, was consistently seen, but no bromine containing fragments could be detected. Nonetheless, combustion analysis revealed that bromine was present at the expected level. The instability of the compound precluded effective purification, with the result that the microanalytical results were slightly outside the accepted limits, but NMR data (^{1}H, ^{13}C, HETCOR, DEPT) were in complete accord with the proposed structure. In addition, a strong absorbance at 759 cm^{-1} in the IR spectrum provided further evidence for a C-Br bond. The ability to perform an *exo* to *endo* bond rearrangement in systems like the α-alkylidene lactones is a useful one, and applications of this reaction in similar systems are currently under investigation.



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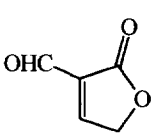


(13)

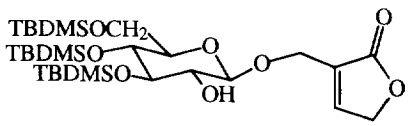
Various methods were investigated in an attempt to reduce the brominated glycoside (**12**) to form (**4**). Radical methods using tributyl tin hydride[30] or tris(trimethylsilyl)silane[31] gave only decomposition

products, as did methods using zinc[32] and magnesium[33]. Sodium borohydride in DMSO[34] appeared to reduce the carbon bromine bond efficiently, but the adjacent double bond was reduced concurrently even in the presence of cerium chloride, which has been used to prevent the reduction of double bonds by borohydride[35].

Direct coupling of the aglycone alcohol (**5**) with acetobromoglucose was investigated as another alternative route to β-miroside. The aglycone was prepared according to published methods[14]. Treatment of acetobromoglucose in CH_2Cl_2 with silver triflate followed by alcohol (**5**) according to the literature procedures for glycoside synthesis[36] gave very low yields of (**4**). This substance proved identical to that prepared previously by acetylation of the naturally occurring β-miroside. By using toluene as a co-solvent to improve the solubility of the silver triflate, yields of (**4**) were improved to 52%.

De-protection of this tetraactetate proved unsuccessful. Treatment with hexabutylditin oxide, which had proved successful with the exo-methylene glucoside tetraacetate (**11**), appeared to cleave the glycoside linkage. Other traditional methods, Amberlite 400 (OH^-)[37] in water and in methanol, 1% NaOH or IR 120 (H^+) in water and methanol all gave a sugar derivative which contained no vinyl proton signals in the NMR spectrum.

Silyl ethers provide an excellent alternative to acetate groups as a protection system for alcohols. Deprotection may be achieved under mild conditions by treatment with fluoride ion. Danishefsky[38] has recently described a route to silyl glycosides with stereochemical control at the anomeric carbon to give exclusively the β-anomer. The method described was applied to the synthesis of β-miroside (**1**) via the tri-TBDMS-derivative (**14**). Epoxide (**15**) was prepared by treatment of tri-TBDMS-glucal (**16**) with dimethyldioxirane[39]. Subsequent reaction of (**15**) with alcohol (**5**) was achieved both by mixing the two components neat at room temperature, and by the method described by Danishefsky[38], a low temperature reaction using anhydrous zinc chloride in ether. Both methods produced the desired β-glycoside (**14**) along with the α-anomer (**17**) in a ratio of approximately 2:1.



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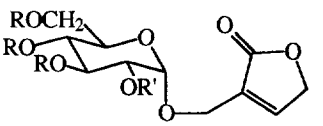
(16)



(17) R = $SiMe_2Bu^t$; R' = H
(18) R = $SiMe_2Bu^t$; R' = Ac
(19) R = R' = Ac

The formation of two isomers is contrary to previous observations using this method. Epoxide (**15**) has been shown to form with exclusively the α-stereochemistry, and to open, presumably in a concerted fashion, to form solely the β-glycoside. As no β-epoxide is present, no mannose derivatives should be formed. ^{1}H NMR analysis of our epoxide (**15**) sample showed only one isomer with data identical to those reported previously[38]. Thus, the epoxide ring opening would seem to have taken place in a non-concerted fashion, thereby leading to (**14**) and its anomer (**17**).

^{1}H NMR coupling constants for (**14**) and (**17**) did not provide convincing proof that these two compounds were anomers. For (**14**) the C-1" signal appeared as a doublet with J = 4.5 Hz, rather than the expected 8-10 Hz for axial-axial coupling, whereas that for (**17**) had *J* = 2.0 Hz, somewhat less than the normal value for axial equatorial coupling (4-6 Hz). Coupling constants for the C-2" signals also provided little assistance [4.0 and 4.5 Hz for (**14**); 2.4 and 5.7 for (**17**)], although the C-2" peak of the C-2" acetate (**18**) derived from (**17**) showed coupling constants more in keeping with an equatorial H-1" and an axial H-2" (*J* = 3.5, 9.0 Hz). The anomalous values obtained for (**14**) and (**17**) tend to suggest that these compounds are distorted from true chair conformations, presumably as a result of the degree of substitution by the bulky TBDMS unit. In support of this, coupling constants of the deprotected glycosides were in accord with the proposed stereochemistry.

Deprotection of the TBDMS ethers was achieved with Dowex 50W-X8 sulfonic acid ion exchange resin in methanol[40]. The use of fluoride in THF, the usual method for removal of TBDMS groupings[41], led to decomposition as indicated by loss of the vinyl resonance of the aglycone in the ^{1}H NMR spectrum. The compound obtained from (**14**) was identical to the natural β-miroside (**1**). The ^{1}H NMR spectrum of the product from (**17**) was consistent with its formulation as α-miroside (**2**). In particular, the 2"-signal appeared as a double-doublet with J = 3.5 and 10.0 Hz as expected for H-2" axial and H-1" equatorial. α-Miroside was characterised as its tetraacetate derivative (**19**).

Glycoside (**3**) undergoes slow methanolysis as shown by the appearance of a methyl ester signal and a shift in the H-5 signal in the ^{1}H NMR spectrum. Mirosides (**1**) and (**2**) also react slowly with methanol with loss of the aglycone to form the methyl glucosides. Some precedent for this has been observed in other systems[42].

BIOLOGICAL SCREENING

Results of biological screening tests on the glycosides (**1**), (**2**) and (**3**), as well as the acetate (**4**) and the aglycone (**5**) are summarised in Table 1.

Biological activities of the naturally occurring β-miroside were very similar to those of the synthetic sample, its anomer (**2**) and tetraacetate (**4**). The exocyclic glucoside (**3**) showed quite different activity, being cytotoxic against BSC cells, but inactive in the other assays. Therefore, activity is unrelated to the glycoside stereochemistry or substitution, but is affected by the position of the double bond. On the other hand, glycosides (**1**), (**2**) and (**4**) showed slightly higher levels of antimicrobial activity than did the aglycone, isosiphonodin (**5**).

Picein (**6**) showed no activity in antifungal, antibacterial or antiviral assays at 300 μg/disk, or in the P388 assay. This is consistent with previous bioassay results[15].

Table 1. Biological Activities of β-Miroside (1) and Related Compounds

Compound	P-388[a] IC_{50}	Cytotoxicity[b]	Antibacterial[c]		Antifungal[c]	
			E. coli 25922[d]	*B. subtilis* 19659[d]	*C. albicans* 14053[d]	*T. mentagrophytes* 28185[d]
(1) (*ex* miro)	19	+++	3	6	1	9
(1) (synthetic)	19	+++	2	3	1	9
(2)	24	+++	2	4	1	9
(3)	>63	ww	0	0	0	0
(4)	13	ww	2	5	0	9
(5)	40	ww	0	2	2	5
Reference[e]	0.02		10	12	10	7

[a] Concentration in μg/ml to give 50% inhibition of growth of P-388 cells.

[b] Measured at 150 μg/disk against monkey kidney cells (BSC); represented as zone sizes +, ++, +++ and ww (whole well).

[c] Activities at 150 μg/disk are represented as width of inhibition zone in mm.

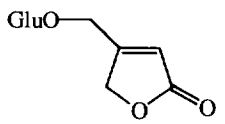
[d] ATCC strain numbers.

[e] Reference compounds and doses: P388, mitomycin C; *E. coli*, gentamycin (10 μg /disk); *B. subtilis*, chloramphenicol (30 μg /disk); antifungal, nystatin (100 units/disk)

CONCLUSION

The previously unreported glycoside, β-miroside (**1**), has been obtained from *P. ferruginea* foliage by isolation techniques directed towards a specific biological activity, inhibition of the fungus *T. mentagrophytes*. β-Miroside also showed activity against other microorganisms and cytotoxicity against two mammalian cell lines.

The discovery of β-miroside completes the family of naturally occuring glucosides of the three isomeric hydroxymethyl-2(5H)-furanones. The 4-substituted compound, siphonoside (**20**) has been reported from *Siphonodon australe* Benth., (family Celastraceae)[43] and shows cytotoxic activity[44]. Ranunculin (**21**), the 5-substituted derivative, which is widespread in the Ranunculaceae[45] also showed cytotoxic activity in our assays (unpublished results). We have recently identified the homologue of ranunculin, glucoside (**22**) in the three species of another gymnosperm genus, *Halocarpus*[46]. However, this compound showed neither antimicrobial nor cytotoxic activity.



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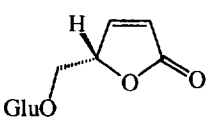

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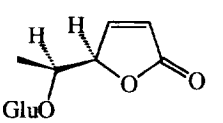

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Our synthesis of β-miroside (**1**) has also resulted in the preparation of the α-anomer (**2**) along with the exocyclic analogue (**3**). This work has revealed the marked reactivity difference between the iodoalkylidene

lactones such as (**7**) with various nucleophiles. Past work[19] has shown that soft nucleophiles react rapidly, but results from this study would suggest that reaction with oxygen centred nucleophiles is not synthetically useful. The synthesis of (**2**) along with (**1**) demonstrates that the previously reported glycoside synthesis based on epoxide (**15**)[38] does not always produce solely the β-glycoside. Nucleophilic opening of the epoxide appears to involve some degree of non-concertedness.

EXPERIMENTAL

General. General experimental details are as described in references 1 and 18. NMR data are for $CDCl_3$ solutions unless otherwise stated. TLC solvent systems were: C_{18} (MeOH-H_2O, 1:1); Si gel (EtOAc-MeOH-H_2O, 81:11:8). Developing reagent: chromatograms were dipped into 2% *w/v* 3,5-dinitrobenzoic acid solution in methanol; after air drying chromatograms were dipped into 20% *v/v* benzyltrimethylammonium hydroxide solution in 3:1 MeOH-H_2O then heated. To detect saccharides, chromatograms were dipped into a solution of naphthoresorcinol (0.2%) and phosphoric acid (10%) in EtOH, then heated.

Collection and Extraction. P. ferruginea foliage (leaves and twigs) was collected from Mt Cargill, Dunedin, New Zealand in November 1992 (voucher specimen 921106-02, kept in the Plant Extracts Research Unit collection). Oven dried (50°) material was frozen with liquid N_2 and ground. Initial screening was carried out using 30 µl/disk of an extract produced by shaking this material (5.0 g) overnight in EtOH (50 ml; 95%).

Bioactivity-Directed Isolation. For the larger scale *P. ferruginea* extraction, ground material (234 g) was extracted with EtOH (3 x 1000 ml) in a Waring blender. The extracts were filtered, combined and evaporated to give a green gum (42 g, *T. mentagrophytes* inhibition zone 3 mm at 300 µg/disk). A first separation was achieved by rp flash chromatography over C_{18} (10 g extract, precoated on 15 g C_{18}, loaded on a 50 g C_{18} column) eluted in steps from H_2O, through MeOH, to $CHCl_3$. Activity was spread over the first three (H_2O) fractions, which were combined (1.89 g, *T. mentagrophytes* inhibition zone 11 mm at 300 µg/disk). The next step was flash chromatography on diol (0.5 g combined actives, precoated onto 2 g diol, loaded on a 10 g diol column) eluted in steps from EtOAc to MeOH. Activity was located in a fraction eluted with 10% MeOH (0.164 g, *T. mentagrophytes* inhibition zone 8 mm at 150 µg/disk). Finally chromatography on C_{18} (0.140 g, loaded on a 10 g C_{18} column) eluting with H_2O:MeOH (3:2) gave β-miroside (**1**) in the first fraction (0.051 g) and picein (**6**) in later fractions (0.040 g). β-Miroside {3-[(β-D-glucopyranosyloxy)methyl]-2(5H)-furanone} (**1**) was a clear glass. TLC C_{18}, R_F 0.90, pink spot, turned purple on heating; TLC Si gel, R_F 0.10; $[\alpha]_D$ -29° (*c* = 0.2, EtOH); UV (EtOH): 213 (ε 5300); IR: 3100-3600 (OH), 1740 (lactone), 1080 (CO); ^{1}H NMR (D_2O): 3.29 (dd, *J* 8, 9 Hz, 1H, H-2"), 3.37 - 3.47 (m, 1H, H-5"), 3.39 (t, *J* 8.5, 8.5 Hz, 1H, H-4"), 3.47 (t, *J* 9, 9 Hz, 1H, H-3"), 3.70 (dd, *J* 5.5, 12.5 Hz, 1H, H-6"), 3.90 (dd, *J* 2, 12.5 Hz, 1H, H-6"), 4.49 (dq, *J* 13.5, 1.5, 1.5, 1.5 Hz, 1H, H-1'), 4.50 (d, *J* 8 Hz, 1H, H-1"), 4.59 (dq, *J* 13.5, 1.5, 1.5, 1.5 Hz, 1H, H-1'), 4.99 (q, *J* 1.5, 1.5, 1.5 Hz, 2H, H-5), 7.80 (quin, *J* 1.5, 1.5, 1.5, 1.5 Hz, 1H, H-4); ^{13}C nmr: 61.7 (C-6"), 63.3 (C-1'), 70.6 (C-4"), 73.3 (C-5), 74.1 (C-2"), 76.7 (C-3"), 77.0 (C-5"), 102.9 (C-1"), 129.7 (C3), 153.7 (C-4), 176.7 (C-2); CIMS (C_4H_{10}) *m/z* (rel. int.) 212 (33), 195 (98), 180 (20), 163 (92), 145 (62), 127 (58), 97 (100), 85 (78). Picein [4-(β-D-glucopyranosyloxy)acetophenone] (**6**) was a white solid, mp 194°C: TLC on C_{18}, R_F 0.6, bluish spot with naphthoresorcinol / phosphoric acid dip; $[\alpha]_D$ -60° (*c*= 1.00, EtOH); UV (EtOH): 213 (ε

12200), 264 (ε 14100); IR: 3600-3100 (OH), 1665 (C=O), 1600, 1500 (benzene) cm^{-1}; ^1H NMR (200 MHz, CD_3OD): 2.71 (s, 3H, H-1), 3.45-3.70 (m, 4H, H-2', 3', 4', 5'), 3.85 (dd, *J* 5, 12 Hz, 1H, H-6'), 4.05 (dd, *J* 2, 12 Hz, 1H, H-6'), 5.18 (d, *J* 8 Hz, 1H, H-1'), 7.29-7.34 (m, 2H, H-5, 7), 8.09-8.18 (m, 2H, H-4, 8); ^{13}C NMR (50.3 MHz, CD_3OD) 26.8 (C-1), 62.8 (C-6'), 71.6 (C-4'), 75.1 (C-2'), 78.3 (C-3'), 78.6 (C-5'), 101.9 (C-1'), 117.6 (C-5, 7), 132.0 (C-4, 8), 133.0 (C-3), 163.4 (C-6), 199.7 (C-2); CIMS (C_4H_{10}) *m/z* 299 (78%, MH^+), 137 (100 %), 121 (64 %), 86 (65%).

Acetylation of β-*Miroside*. Treatment of (**1**) (5 mg) with Ac_2O (1.0 ml) and C_5H_5N (1.0 ml) at room temperature overnight followed by chromatography over Si gel (EtOAc/hexanes, 1:1) gave *3-[(2,3,4,6-tetra-O-acetyl-*β*-D-glucopyranosyloxy)methyl]-2(5H)-furanone* (**4**). TLC C_{18}, R_F 0.30, pink spot, turned purple on heating; TLC Si gel, R_F 0.85. $[\alpha]_D$ -27° (*c*= 0.5, EtOH); UV (EtOH): 207 (ε 11500); IR: 1750, 1370, 1220 (ester); ^1H NMR: 2.00 (3H, s, OAc), 2.01 (3H, s, OAc), 2.04 (3H, s, OAc), 2.08 (3H, s, OAc), 3.70 (ddd, *J* 2.5, 5, 10 Hz, 1H, H-5")), 4.11 (dd, *J* 2.5, 12.5 Hz, 1H, H-6"), 4.23 (dd, *J* 5, 12.5 Hz, 1H, H-6"), 4.33 (dq, *J* 14.5, 2, 2, 2 Hz, 1H, H-1'), 4.58 (dq, *J* 14.5, 2, 2, 2 Hz, 1H, H-1'), 4.60 (d, *J* 8 Hz, 1H, H-1"), 4.82 (q, *J* 2, 2, 2 Hz, 2H, H-5), 5.01 (dd, *J* 8, 9.5 Hz, 1H, H-2"), 5.05 (dd, *J* 9.5, 10 Hz, 1H, H-4"), 5.20 (t, *J* 9.5, 9.5 Hz, 1H, H-3"), 7.37 (quin, *J* 2, 2, 2, 2 Hz, 1H, H-4); ^{13}C NMR: 20.5 (2xCH_3, OAc), 20.7 (2xCH_3, OAc), 61.7 (C-6"), 63.5 (C-1'), 68.2 (C-4"), 70.8 (C-5), 71.1 (C-2"), 72.0 (C-5"), 72.5 (C-3"), 100.7 (C-1"), 130.6 (C-3), 147.2 (C-4), 169.4 (2xC, OAc), 170.1 (OAc) , 170.6 (OAc) , 172.3 (C-2); HREIMS *m/z* 371.0933 (3%, M^+ - $C_3H_5O_2$, $C_{16}H_{19}O_{10}$ requires 371.0978), 347.0977 (13%, M^+ - $C_5H_5O_2$, $C_{14}H_{19}O_{10}$ requires 347.0978), 331.0969 (6%, M^+ - $C_5H_5O_3$, $C_{14}H_{19}O_9$ requires 331.1029), 114.0323 (10%, M^+ - $C_{14}H_{18}O_9$, $C_5H_6O_3$ requires 114.0317), 98.0356 (100%, M^+ - $C_{14}H_{18}O_8$, $C_5H_6O_2$ requires 98.0356), 97.0285 (68%, M^+ - $C_{14}H_{17}O_8$, $C_5H_5O_2$ requires 97.0289).

Acid Hydrolysis of β-*Miroside*. A solution of (**1**) (0.092 g) in D_2O (4 ml) and TFA (250 µl) was heated at 70-80° for 24 h. After evaporation, rp flash chromatography over C_{18} (H_2O) gave 3-(hydroxymethyl)-2(5H)-furanone (**5**) (0.020 g, 53%). TLC C_{18}, R_F 0.80, pink spot, turned purple on heating; TLC Si gel, R_F 0.60); UV (EtOH) 210 (ε 8200); IR: 3600-3100 (OH), 1740 (lactone); ^1H NMR: 4.18 (q, *J* 2, 2, 2 Hz, 2H, H-1'), 4.70 (q, *J* 2, 2, 2 Hz, 2H, H-5), 7.30 (quin, *J* 2, 2, 2, 2 Hz, 1H, H-4); ^{13}C NMR: 56.0 (C-1'), 70.9 (C-5), 133.7 (C-3), 146.2 (C-4), 173.6 (C-2).

Enzymatic Hydrolysis of β-*Miroside*. A solution of (**1**) (5 mg) in acetate buffer (pH 5.6, 1.0 ml, 10.0 mmol L^{-1}) was incubated at 37°C for 48 h with a solution of *exo*-β-(1,3)-glucanase (0.07 mg/ml, 120 µl). Aliquots (50 ml) were taken after 12, 24, 36 and 48 h. and frozen for later glucose analysis. Analysis showed hydrolysis was complete after 12 h. TLC (Si gel) of the crude hydrolysate showed the presence of 3-(hydroxymethyl)-2(5H)-furanone (**5**).

2-Iodoethyl trimethylsilylpropynoate (8). Ethylene gas was bubbled through a solution of trimethylsilyl propynoic acid (0.300 g, 2.1 mmol), N-iodosuccinimide (0.474 g, 2.1 mmol) in dry CH_2Cl_2 (30 ml) at 35°C for 1 h. The product was poured into water, washed with $Na_2S_2O_3$ (1M), dried ($MgSO_4$) and evaporated to give (**8**), (0.528 g, 85%). ^1H NMR: 0.19 (s, 9H, CH_3Si), 3.26 (t, *J* 7 Hz, H-1), 4.36 (t, *J*

7 Hz, H-2); ^{13}C NMR: -0.9 (CH_3Si), -1.1 (C-2'), 65.7 (C-1'), 94.0 (C-2), 95.1 (C-3), 152.0 (C-1). The product was cyclised without purification.

(3E)-Dihydro-3-(1-iodo-1-trimethylsilylmethylene)-2(3H)-furanone (7). Dibenzoyl peroxide (0.024 g, 0.10 mmol) was added to a solution of the crude iodoester (**8**) (0.296, 1.0 mmol) in dry benzene (3 ml) and the solution was heated under reflux for 1h. Subsequently, a further portion of dibenzoyl peroxide (0.024 g) was added and heating was continued for 1h. Evaporation of the solvent and chromatography on a silica column (ether/hexanes, 1:3) gave (**7**) (0.282 g, 92%); IR: 1757, 1211 (lactone); ^{1}H NMR: 0.34 (s, 9H, CH_3Si), 2.96 (t, J 7.5 Hz, 2H, H-4), 4.39 (t, *J* 7 Hz, 2H, H-5); ^{13}C NMR: 1.4 (CH_3Si), 40.6 (C-4), 63.8 (C-5), 131.5 (C-1'), 144.9 (C-3), 164.4 (C-2). MS: *m/z* 281 (M^+-15); Anal. Found: C, 32.6; H, 4.6; Calc. for $C_8H_{13}O_2SiI$: C, 32.5; H, 4.4%; HREIMS *m/z* 295.97306 (M^+); Calc for $C_8H_{13}IO_2Si$: 295.97296.

(3E)-Dihydro-3-[(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyloxy)methylene]-2(3H)-furanone (11). A mixture of acetobromoglucose (2.00 g, 4.88 mmol) and sodium salt (**10**)[47] (1.00 g, 7.32 mmol) in dry DMSO (40 ml) was stirred at room temperature for 18 h. The solution was diluted with CH_2Cl_2 (100 ml), washed with water (3 x 30 ml), dried ($MgSO_4$), chromatographed on a silica column eluting with EtOAc, then crystallised from Et_2O to give (**11**)[20] (1.19 g, 55%); $[\alpha]_D$ 8.2° (*c*=1.0, $CHCl_3$); IR: 1752, 1368, 1221 (lactone); ^{1}H NMR: 1.99 (s, 3H, OAc), 2.00 (s, 3H, OAc), 2.03 (s, 3H, OAc), 2.06 (s, 3H, OAc), 2.77-2.99 (m, 2H, H-4), 3.83 (ddd, *J* 2, 5, 10 Hz, 1H, H-5"), 4.13 (dd, *J* 2, 12.5 Hz, 1H, H-6"), 4.29 (dd, *J* 5, 12.5 Hz, 1H, H-6"), 4.39 (t, *J* 7.5, 7.5 Hz, 2H, H-5), 4.93 (d, *J* 7.5 Hz, 1H, H-1"), 5.19-5.09 (m, 2H, H-2", 4"), 5.26 (t, *J* 9, 9 Hz, 1H, H-3"), 7.43 (t, *J* 3, 3 Hz, 1H, H-1'); ^{13}C NMR: 20.7 (4 x CH_3, OAc), 23.8 (C-4), 61.5 (C-6"), 66.0 (C-5), 67.7 (C-4"), 70.7 (C-2"), 72.2 (C-3"), 72.8 (C-5"), 101.2 (C-1"), 107.0 (C-3), 150.9 (C-1'), 169.1 (OAc), 169.4 (OAc), 170.1(OAc), 170.6 (OAc), 172.1 (C-2); Anal. Found: C, 51.3; H, 5.4; Calc. for $C_{19}H_{24}O_{12}$: C, 51.4; H, 5.4%.

(3E)-Dihydro-3-[(β-D-glucopyranosyloxy)methylene]-2(3H)-furanone (3). A solution of the tetraacetate (**11**) (0.063 g, 0.142 mmol) and $(Bu_3Sn)_2O$ (0.042 g, 0.07 mmol) in dry methanol (2 ml) was added and stirred at 55°C for 72 h. A further portion of $(Bu_3Sn)_2O$ (0.021 g) was added and stirring was continued for 24 h. Evaporation of the solvent, followed by chromatography on C-18 silica (H_2O) gave (**3**) (0.026 g, 65%); $[\alpha]_D$ 66.3° (*c*=0.15, EtOH); IR 3378 (OH), 1680, 1070 (lactone); ^{1}H NMR (D_2O, referenced $(CH_3)_2CO$, δ 2.05): 2.80 (td, *J* 7.5, 7.5, 2.5 Hz, 2H, H-4), 3.44 - 3.27 (m, 4H, H-2", 3", 4", 5"), 3.57 (dd, *J* 4, 8 Hz, 1H, H-6"), 3.74 (br. d, *J* 8 Hz, 1H, H-6"), 4.30 (t, 2H, *J* 7.5, 7.5 Hz, H-5), 4.81 (d, *J* 8 Hz, 1H, H-1"), 7.45 (t, *J* 2, 2 Hz, 1H, H-1'); ^{13}C NMR (D_2O, referenced $(CH_3)_2CO$, δ 30.2): 23.2 (C-4), 60.2 (C-6"), 67.6 (C-5), 68.9, 72.4, 75.0 and 76.5 (C-2",3",4",5"), 103.1 (C-1"), 106.3 (C-3), 153.0 (C-1'), 175.8 (C-2). Reacetylation with Ac_2O/C_5H_5N gave tetraacetate identical to (**11**).

3-[Bromo(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyloxy)methyl]-2(5H)-furanone (12). A mixture of tetraacetate (**11**) (1.19 g, 2.69 mmol) and N-bromosuccinimide (0.50 g, 2.83 mmol) in $CHCl_3$ (4 ml) and CCl_4 (40 ml) was heated under reflux for 30 min. The succinimide was removed by filtration and the filtrate was washed with $NaHCO_3$ (10%, 2x10 ml), then water. Drying ($MgSO_4$), and evaporation of the

solvent *in vacuo*, gave (**12**) (1.26 g, 89%); IR 1731, 1245, 1045 (ester), 759 (C-Br); ^{1}H NMR: 2.00 (s, 3H, OAc), 2.02 (s, 3H, OAc), 2.06 (s, 3H, OAc), 2.07 (s, 3H, OAc), 3.84 (ddd, *J* 2, 5, 10 Hz, 1H, H-5"), 4.10 (dd, *J* 2, 12.5 Hz, 1H, H-6"), 4.28 (dd, *J* 5, 12.5 Hz, 1H, H-6"), 4.87 (m, $W_{h/2}$ 10 Hz, 2H, H-5), 4.93 (d, *J* 8 Hz, 1H, H-1"), 5.15-5.01 (m, 2H, H-2",4"), 5.30 (t, *J* 9, 9 Hz, 1H, H-3"), 6.69 (q, *J* 1.5, 1.5, 1.5 Hz, 1H, H-1'), 7.62 (q, *J* 1.5, 1.5, 1.5 Hz, 1H, H-4); ^{13}C NMR: 20.6 (2 x CH_3, OAc), 20.7 (2 x CH_3, OAc), 61.5 (C-6"), 67.9 (C-4"), 70.3 (C-2"), 70.6 (C-5), 72.1 (C-3"), 72.6 (C-5"), 75.4 (C-1'), 97.5 (C-1"), 132.8 (C-3), 148.1 (C-4), 169.4 (OAc), 169.5 (OAc), 169.5 (OAc), 170.0 (OAc), 170.5 (C-2); Anal. Found: C, 42.9; H, 4.5; Br, 16.3; Calc. for $C_{19}H_{23}O_{12}Br$: C, 43.6; H, 4.4; Br, 15.3%.

3-[(2,3,4,6-Tetra-O-acetyl-β-D-glucopyranosyloxy)methyl]-2(5H)-furanone (4). A solution of silver triflate (0.92 g, 3.6 mmol) in dry CH_2Cl_2 (40 ml) and dry toluene (15 ml) was cooled to -30°C. A solution of alcohol (**5**)[14] (0.205 g, 1.8 mmol) in CH_2Cl_2 (3 ml) was added, followed by a solution of acetobromoglucose (0.738 g, 1.8 mmol) in CH_2Cl_2 (2 ml) and toluene (0.5 ml). The mixture was stirred at -20°C for 2.5 h. The reaction was neutralised by the addition of Et_3N (10 drops), the product washed with $Na_2S_2O_3$, 1M HCl, $NaHCO_3$, water, dried ($MgSO_4$), and the solvents evaporated *in vacuo* to give crude product (1.71 g). Chromatography on a silica column (EtOAc/hexanes, 2:1) gave (**4**) (0.415 g, 52%), identical to the sample prepared by acetylation of β-miroside.

3-[(3, 4, 6-Tri-O-tert-butyldimethylsilyl-α-D-glucopyranosyloxy)methyl]-2(5H)-furanone (17) and 3-[(3, 4, 6-tri-O-tert-butyldimethylsilyl-β-D-glucopyranosyloxy)methyl]-2(5H)-furanone (14).

Method A. Epoxide (**15**)[38] (0.037 g, 0.073 mmol) was added to alcohol (**5**)[14] (0.25 ml, 2.2 mmol) and the mixture was stirred for 18 h. Chromatography on silica (EtOAc/hexanes, 1:3) gave a mixture of two glycosides in a 1:2 ratio (0.031 g, 68%). A second silica column gave the pure β-glycoside (**14**) (0.006 g, 14%). $[\alpha]_D$ -22° (*c* = 0.62, $CHCl_3$); IR: 3500 (OH), 1748 (C=O), ^{1}H NMR: 0.03, 0.04 and 0.12 (each s, 3H, CH_3Si), 0.13 (s, 9H, 3 x CH_3Si), 0.89 (s, 9H, $(CH_3)_3CSi$), 0.91 (s, 18H, 2x$(CH_3)_3CSi$), 3.13 (d, *J* 8 Hz, 1H, OH), 3.48 (quin, *J* 4, 4, 4, 4 Hz, 1H, H-2"), 3.62-3.68 (m, 2H, H-5"), 3.73 (dd, *J* 6, 10 Hz, 1H, H-6"), 3.80-3.86 (m, 2H, H-3", 4"), 4.04 (dd, *J* 7, 10 Hz, 1H, H-6"), 4.31 (dq, *J* 15, 2, 2, 2 Hz, 1H, H-1'), 4.62 (dq, *J* 15, 2, 2, 2 Hz, 1H, H-1'), 4.68 (d, *J* 4 Hz, 1H, H-1"), 4.83 (q, *J* 2, 2, 2 Hz, 2H, H-5), 7.43 (quin, *J* 1.5, 1.5, 1.5, 1.5 Hz, 1H, H-4); ^{13}C NMR: -5.2, -5.1, -4.5, -4.2, -4.1, -3.9 (each CH_3Si), 18.0 ($(CH_3)_3CSi$), 18.4 (2x$(CH_3)_3$**C**Si) , 26.0 (2x(**C**$H_3)_3$CSi) , 26.1 ($(CH_3)_3$**C**Si) , 62.6 (C-1'), 63.2 (C-6") , 70.1 (C-4"), 70.7 (C-5), 72.5 (C-2"), 73.7 (C-3"), 78.9 (C-5"), 101.8 (C-1"), 131.5 (C-3), 146.6 (C-4), 172.5 (C-2); MS (low res.): small *m/z* 618 (M^+); MS (high res.): *m/z* 561.2748 (M^+-57); Calc for $C_{29}H_{58}Si_3O_8$: 561.27351; Anal. Found: C, 56.6; H, 8.9; Calc. for $C_{29}H_{58}Si_3O_8$: C, 56.3; H, 9.4%.

Method B. A solution of alcohol (**5**) (0.165 g, 1.45 mmol) in THF (1 ml) was stirred with freshly activated 4A molecular sieves for 0.5 h, after which time a solution of epoxide (**15**) (0.365 g, 0.72 mmol) in THF (1 ml) was added and the reaction mixture was cooled to -78C. A solution of $ZnCl_2$ in Et_2O (0.605 M, 2.4 ml, 1.45 mmol) was added dropwise, and the mixture was stirred at -78°C for 1 h, then allowed to warm to room temperature and stirred for a further 18 h. The reaction mixture was diluted with EtOAc (10 ml), washed with sat. $NaHCO_3$, dried ($MgSO_4$) and the solvents evaporated *in*

vacuo. The crude product was chromatographed on silica (EtOAc/hexanes, 1:3) to give three fractions: (i) β-glucoside (**14**) (0.040 g, 15%); (ii) a mixture (2:1 / β:α) of (**14**) and (**17**) (0.097g, 28%); (iii) α-glucoside (**17**) (0.032 g, 12%); $[\alpha]_D$ 24° (*c* = 0.71, $CHCl_3$); IR: 3502 (OH), 1760 (C=O); ^{1}H NMR: 0.03, 0.04 and 0.12 (each s, 3H, CH_3Si), 0.13 (s, 9H, 3xCH_3Si), 0.89 (s, 9H, $(CH_3)_3CSi$), 0.91 (s, 18H, 2x$(CH_3)_3CSi$), 3.00 (br. d, *J* 11 Hz, 1H, OH), 3.52 (m, $W_{h/2}$ 10 Hz, 1H, H-2"), 3.64-3.96 (m, 5H, H-3", 4", 5", 6", 6"), 4.34 (dq, *J* 14, 2, 2, 2 Hz, 1H, H-1'), 4.65 (dq, *J* 14, 2, 2, 2 Hz, 1H, H-1'), 4.85 (q, *J* 2, 2, 2 Hz, 2H, H-5), 4.89 (d, *J* 2.5 Hz, 1H, H-1"), 7.49 (ddd, *J* 2, 2, 2 Hz, 1H, H-4); ^{13}C NMR: -5.2, -5.1, -4.4, -4.1, -3.9 and -3.6 (each CH_3Si), 18.1 ($(CH_3)_3CSi$), 18.3 (2x$(CH_3)_3CSi$), 25.9, 26.0 and 26.2 (each $(CH_3)_3CSi$), 61.9 (C-6"), 62.3 (C-1'), 70.2 (C-4"), 70.8 (C-5), 71.7 (C-2"), 74.3 (C-3"), 78.2 (C-5"), 96.2 (C-1"), 131.3 (C-3), 146.8 (C-4), 172.5 (C-2); MS: *m/z* 618 (M^+), 561 (M^+ - 57); Anal. Found: C, 56.2; H, 9.6; Calc. for $C_{29}H_{58}Si_3O_8$: C, 56.3; H, 9.4%.

Synthetic β-Miroside, 3-[(β-D-glucopyranosyloxy)methyl]-2(5H)-furanone (1). A solution of (**14**) (0.064 g, 0.10 mmol) in methanol (10 ml) was stirred with DOWEX 50W-X8 ion exchange resin (1.5 g) at 45°C for 12 h. The resin was removed by filtration, the solvent evaporated and the crude product was chromatographed on C-18 silica (EtOH/H_2O, 4:1). Evaporation of the solvent *in vacuo*, followed by repeated trituration with 5 ml portions of dry methanol gave pure β-miroside (**1**) {0.024 g, 86%), identical with the isolated sample; $[\alpha]_D$ -22° (*c* = 0.25, EtOH).

Synthetic α-Miroside, 3-[(α-D-glucopyranosyloxy)methyl]-2(5H)-furanone (2). A solution of (**17**) (0.025 g, 0.04 mmol) in methanol (10 ml) was stirred with DOWEX 50W-X8 ion exchange resin (1.5 g) at 45°C for 12 h. The resin was removed by filtration, the solvent evaporated and the crude product was chromatographed on C-18 silica (EtOH/H_2O, 4:1). Evaporation of the solvent *in vacuo*, followed by repeated trituration with 5 ml portions of dry methanol gave pure α-miroside (**2**) (0.010 g, 89%). $[\alpha]_D$ 71° (*c* = 0.48, EtOH); IR: 3320 (OH), 1755, 1080 (lactone); ^{1}H NMR (D_2O, referenced $(CH_3)_2CO$, δ 2.05): 3.18-3.28 (m, 2H, H-2", 3"), 3.42 (dd, *J* 4, 10 Hz, 1H, H-6"), 3.55-3.63 (m, 2H, H-4", 6"), 3.73 (ddd, *J* 2, 11, 11.5 Hz, 1H, H-5"), 4.20 (dq, *J* 14, 2, 2, 2 Hz, 1H, H-1'), 4.34 (dq, *J* 14, 2, 2, 2 Hz, 1H, H-1'), 4.87 (m, $W_{h/2}$ 3.5 Hz, 3H, H-1", 5), 7.67 (quin, *J* 2, 2, 2, 2 Hz, 1H, H-4); ^{13}C NMR (D_2O, referenced $(CH_3)_2CO$, δ 30.5): 61.5 (C-1'), 61.6 (C-6"), 70.5 (C-4"), 72.2 (C-5"), 73.1 (C-3"), 73.4 (C-5), 74.0 (C-2"), 99.0 (C-1"), 129.7 (C-3), 153.5 (C-4), 175.4 (C-2).

3-[(2,3,4,6-Tetra-O-acetyl-α-D-glucopyranosyloxy)methyl]-2(5H)-furanone (19). Acetylation of α-miroside (**2**) (0.018 g, 0.0065 mmol) as described previously for the acetylation of β-miroside, gave, after purification on silica, eluting with 2:1 EtOAc/hexanes, the tetraacetate derivative (**19**) (0.022 g, 76%); $[\alpha]_D$ 80° (*c* = 0.15, $CHCl_3$); IR: 1746, 1367,1221, 1042 (lactone); ^{1}H NMR: 2.03 (s, 3H, OAc), 2.04 (s, 3H, OAc), 2.08 (s, 3H, OAc), 2.11 (s, 3H, OAc), 4.11-4.16 (m, 2H, H-5", 6"), 4.23-4.32 (m, 2H, H-1', 6"), 4.51 (dq, *J* 14, 2, 2, 2 Hz, 1H, H-1'), 4.90 (q, *J* 2, 2, 2 Hz, 2H, H-5), 4.92 (dd, *J* 4, 10 Hz, 1H, H-2"), 5.09 (t, *J* 10, 10 Hz, 1H, H-4"), 5.18 (d, *J* 4 Hz, 1H, H-1"), 5.50 (t, *J* 10, 10 Hz, 1H, H-3"), 7.49 (quin, *J* 2, 2, 2, 2 Hz, 1H, H-4); ^{13}C NMR: 20.7 (OAc), 20.8 (2 x CH_3, OAc), 20.8 (OAc), 61.5 and 61.9 (C-1', 6"), 67.7, 68.5, 70.0 and 70.7 (C-2", 3", 4", 5"), 70.7 (C-5), 95.7 (C-1"), 130.4 (C-3), 147.7

(C-4), 169.6 (2xC), 170.2 (2xC) and 170.7 (OAc and C-2); HREIMS *m/z* 371.09706 (M^+ - CH_2OAc); Calc for $C_{16}H_{19}O_{10}$: 371.09782.

3-[(2-O-Acetyl-3, 4, 6-tri-O-tert-butyldimethylsilyl-α-D-glucopyranosyloxy)methyl]-2(5H)-furanone (18). A solution of (**17**) (0.014 g, 0.023 mmol) in dry CH_2Cl_2 (1 ml) was treated with DMAP (0.010 g, 0.082 mmol) and Ac_2O (2 drops) for 1.5 h at R.T. The mixture was diluted with CH_2Cl_2 (5 ml), washed with 5% HCl (2x10 ml), sat. $NaHCO_3$, dried ($MgSO_4$) and the solvent evaporated *in vacuo* to give (**18**) (0.013 g, 85%); $[\alpha]_D$ 55.5° (*c* = 0.88, $CHCl_3$); IR: 1751 (lactone); ^{1}H NMR: 0.10 (m, 18H, 6xCH_3Si), 1.90 (m, 27H, 3x$(CH_3)_3$CSi), 2.1 (s, 3H, OAc), 3.90-3.45 (m, 4H, H-4", 5", 6", 6"), 4.03 (dd, *J* 7, 9 Hz, 1H, H-3"), 4.20 (dq, *J* 14, 2, 2, 2 Hz, 1H, H-1'), 4.49 (dq, *J* 14, 2, 2, 2 Hz, 1H, H-1'), 4.62 (dd, *J* 3.5, 9 Hz, 1H, H-2"), 4.84 (q, *J* 2, 2, 2 Hz, 2H, H-5), 5.05 (d, *J* 3.5 Hz, 1H, H-1"), 7.38 (quin, *J* 2, 2, 2, 2 Hz, 1H, H-4); HREIMS *m/z* 603.28386 (M^+ - C_4H_9); Calc for $C_{27}H_{51}O_9Si$: 603.28410.

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