Chemoenzymatic Synthesis of Naturally Occurring Benzyl 6-*O*-Glycosyl-β-D-glucopyranosides

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Direct β-glucosidation between benzyl alcohol and D-glucose (5) using the immobilized β-glucosidase from almonds with the synthetic prepolymer ENTP-4000 gave a benzyl β-D-glucoside (1) in 53% yield. The coupling of the benzyl β-D-glucopyranoside congener (8) derived from 1 with phenyl 2,3,4-tri-*O*-acetyl-1-thio-β-D-xylopyranoside (9), ethyl 2,3,4-tri-*O*-acetyl-1-thio-α-L-rhamnopyranoside (13), and 2,3,4-tri-*O*-acetyl-α-L-arabinopyranosyl bromide (15) afforded 10, 14, and 16, respectively, as coupled products. Deprotection of 10, 14, and 16 provided the synthetic benzyl β-D-xylopyranosyl-(1→6)-β-D-glucopyranoside (2), benzyl α-L-rhamnopyranosyl-(1→6)-β-D-glucopyranoside (3), and benzyl α-L-arabinopyranosyl-(1→6)-β-D-glucopyranoside (4), respectively.

Key words β-glucosidase; β-glucosidation; natural product synthesis; benzyl β-D-glucopyranoside

Prunus mume SIEB. *et* ZUCC. (Rosaceae) has been used as medicinal food in Japan for a long time and is reported to have many pharmacological properties, such as the inhibitory effects on bradykinin and prostaglandin E_2 production in the abdominal cavities of mice, and the effects on angiotensin-converting enzyme, aldosterone, and corticosterone levels in rat plasma.[1] Moreover, it has been reported that the methanolic extract of *P. mune* exhibited inhibitory effects on rat lens aldose reductose and platelet aggregation.[2] Benzyl β-D-glucopyranoside (**1**) is one of the pharmacologically active constituents of *P. mune*. On the other hand, three kinds of naturally occurring benzyl 6-*O*-glycosyl-β-D-glucopyranoside congeners, benzyl β-D-xylopyranosyl-(1→6)-β-D-glucopyranoside (**2**),[3,4] benzyl α-L-rhamnopyranosyl-(1→6)-β-D-glucopyranoside (**3**)[5] and benzyl α-L-arabinopyranosyl-(1→6)-β-D-glucopyranoside (**4**)[6] were isolated from *Alangium chinense*,[4] *Margyricarpus setosus*,[5] and *Lycopersicon esculentum*,[6] respectively. For the purpose of investigation of pharmacological activity of these β-D-glucopyranoside congeners in comparison to **1**, the synthesis of the above-mentioned β-D-glucopyranoside congeners has aroused our interest. In this paper, we describe the synthesis of benzyl β-D-glucopyranoside (**1**) and its naturally occurring benzyl 6-*O*-glycosyl-β-D-glucopyranoside congeners **2**, **3** and **4**, based on the selective β-glycosidation of benzyl alcohol with D-glucose (**5**) catalyzed by the immobilized β-glucosidase (EC 3.2.1.21) from almonds.

Enzymatic β-Glycosidation In the case of the direct β-glycosidation of primary alcohols with D-glucose (**5**) using β-glucosidase (EC 3.2.1.21) from almonds under thermodynamic conditions, a high concentration of alcohol or a medium with low water activity is reported to be effective,[7] and this method is applied to the synthesis of **1**. Meanwhile, the synthesis of **1** using 4-nitrophenyl β-D-glucopyranoside as a glycosyl donor was reported previously by us.[8] On the other hand, we reported the effectiveness of immobilization of β-glucosidase (EC 3.2.1.21) from almonds with a photocross-linkable resin prepolymer (ENTP-4000) in the direct β-glucosidation of 1,8-octanediol with D-glucose (**5**).[9] Then we examined the direct β-glucosidation of benzyl alcohol with D-glucose (**5**) using the reported immobilized β-glucosidase (EC 3.2.1.21)[9] from almonds. When a large amount of benzyl alcohol (23.4 eq) was used as an acceptor for D-glucose (**5**) in the presence of the immobilized β-glucosidase, benzyl β-D-glucopyranoside (**1**) was obtained in 53% yield. Moreover, the same β-glucosidation using the recovered immobilized enzyme afforded **1** in 52% yield.

Synthesis of Benzyl β-D-Xylopyranosyl-(1→6)-β-D-glucopyranoside (2) Silylation of **1** gave a silyl ether (**6**; 95% yield), which was subjected to acetylation to afford an acetate (**7**) in 99% yield. Deprotection of silyl group of **7** using 1 M IBr solution in CH_2Cl_2 gave a primary alcohol (**8**) in 76% yield. When 2% I_2 in MeOH solution or *N*-bromosuccinimide (NBS) instead of 1 M IBr solution in CH_2Cl_2 was applied for desilylation of **7**, the yield of **8** was improved to 90% or 98%, respectively. By the following reported procedure,[10] the coupling reaction of benzyl β-D-glucopyranoside congener (**8**) and phenyl 2,3,4-tri-*O*-acetyl-1-thio-β-D-xylopyranoside (**9**)[11] in the presence of *N*-iodosuccinimide (NIS) and trifluoromethanesulfonic acid (TfOH) gave an inseparable mixture (**10** : **11** = 3 : 2) of the coupled products **10** and **11** in 40% yield. Finally, treatment of this mixture with K_2CO_3 in MeOH provided the synthetic benzyl β-D-xylopyranosyl-(1→6)-β-D-glucopyranoside (**2**, 55% yield) and benzyl β-D-xylopyranosyl-(1→4)-β-D-glucopyranoside (**12**, 37% yield). The spectral data (^{1}H- and ^{13}C-NMR) and specific rotation ($[\alpha]_D$ −48.3° (MeOH)) of the synthetic **2** were identical with those (^{1}H- and ^{13}C-NMR and $[\alpha]_D$ −48.0° (MeOH)) of the natural product **2**.[4] Where, **11** was formed by glycosylation of benzyl 2,3,6-tri-*O*-acetyl-β-D-glycopyranoside (**8a**), which was formed from **8** by 4→6 acyl migration in the reaction medium. When **8** was treated with the same coupling reaction medium without **9**, **8a** was partly formed in the reaction mixture. The structure of **12** was determined by the HMBC correlation between H-1″ (δ_H 4.33)/C-4′ (δ_C 80.8). The low yield of the desired compound (**2**) could be explained by the following reason. In the coupling process between **8** and **9**, partial 4→6 acyl migration in the substrate (**8**) under acidic condition (TfOH) might be occurred to afford the undesired coupled product (**11**). Low yield is pre-

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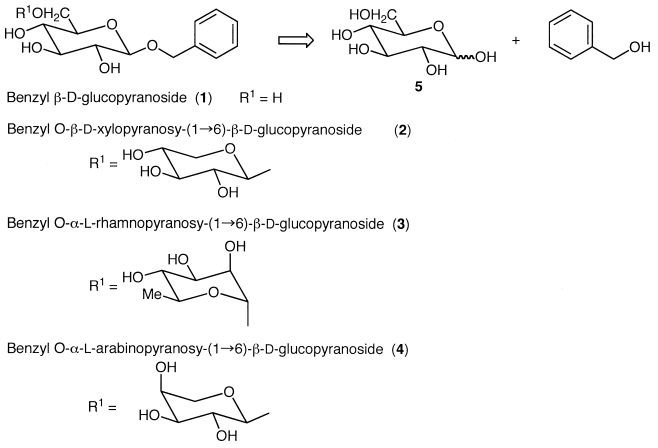

Chart 1

sumably due to low reactivity of **9** as a glycosyl donor. To overcome this low yield process, the usage of 2,3,4-tri-*O*-benzoyl-α-D-xylopyranosyl bromide is presumably effective.

Synthesis of Benzyl α-L-Rhamnopyranosyl-(1→6)-β-D-glucopyranoside (3) Ethyl 2,3,4-tri-*O*-acetyl-1-thio-α-L-rhamnopyranoside (**13**) was synthesized by applying the reported method[12] based on the $BF_3 \cdot Et_2O$ catalyzed reaction of ethane thiol and tetra-*O*-acetyl-α-L-rhamnopyranoside obtained by acetylation of α-L-rhamnose. By the following reported procedure,[10] the coupling reaction of benzyl β-D-glucopyranoside congener (**8**) and ethyl 2,3,4-tri-*O*-acetyl-1-thio-α-L-rhamnopyranoside (**13**) in the presence of *N*-iodosuccinimide (NIS) and trifluoromethanesulfonic acid (TfOH) gave the coupled product (**14**) in 76% yield. Finally, treatment of **14** with K_2CO_3 in MeOH provided the synthetic benzyl β-D-rhamnopyranosyl-(1→6)-β-D-glucopyranoside (**3**) 85% yield. The spectral data (^{1}H- and ^{13}C-NMR) and specific rotation ($[\alpha]_D$ −52.8° (MeOH)) of the synthetic **3** were identical with those (^{1}H- and ^{13}C-NMR and $[\alpha]_D$ −40.0° (MeOH)) of the natural product **3**.[5]

Synthesis of Benzyl α-L-Arabinopyranosyl-(1→6)-β-D-glucopyranoside (4) By following the reported procedure,[13] the coupling reaction of benzyl β-D-glucopyranoside congener (**8**) and 2,3,4-tri-*O*-acetyl-α-L-arabinopyranosyl bromide (**15**) in the presence of silver triflate (AgOTf) and 4A molecular sieves gave the coupled product (**16**) in 40% yield. Finally, treatment of **16** with K_2CO_3 in MeOH provided quantitatively the synthetic benzyl α-L-arabinopyranosyl-(1→6)-β-D-glucopyranoside (**4**). The spectral data (^{1}H- and ^{13}C-NMR) and specific rotation ($[\alpha]_D$ −33.6° (MeOH)) of the synthetic **4** were identical with those (^{1}H- and ^{13}C-NMR and $[\alpha]_D$ −39.0° (MeOH)) of the natural product **4**.[6] The low yield of **16** is presumably overcome by the usage of 2,3,4-tri-*O*-benzoyl-α-L-arabinopyranosyl bromide instead of **15**.

Conclusion

Direct β-glucosidation between benzyl alcohol and D-glucose (**5**) using the immobilized β-glucosidase from almonds with the synthetic prepolymer ENTP-4000 gave a benzyl β-D-glucoside (**1**) in 53% yield. The coupling of the benzyl β-D-glucopyranoside congener (**8**) derived from **1** and phenyl 2,3,4-tri-*O*-acetyl-1-thio-β-D-xylopyranoside (**9**), ethyl 2,3,4-tri-*O*-acetyl-1-thio-α-L-rhamnopyranoside (**13**), and 2,3,4-tri-*O*-acetyl-α-L-arabinopyranosyl bromide (**15**) afforded the coupled products **10**, **14**, and **16**, respectively. Deprotection of the coupled products **10**, **14**, and **16** afforded the synthetic benzyl β-D-xylopyranosyl-(1→6)-β-D-glucopyranoside (**2**), benzyl α-L-rhamnopyranosyl-(1→6)-β-D-glucopyranoside (**3**), and benzyl α-L-arabinopyranosyl-(1→6)-β-D-glucopyranoside (**4**), respectively.

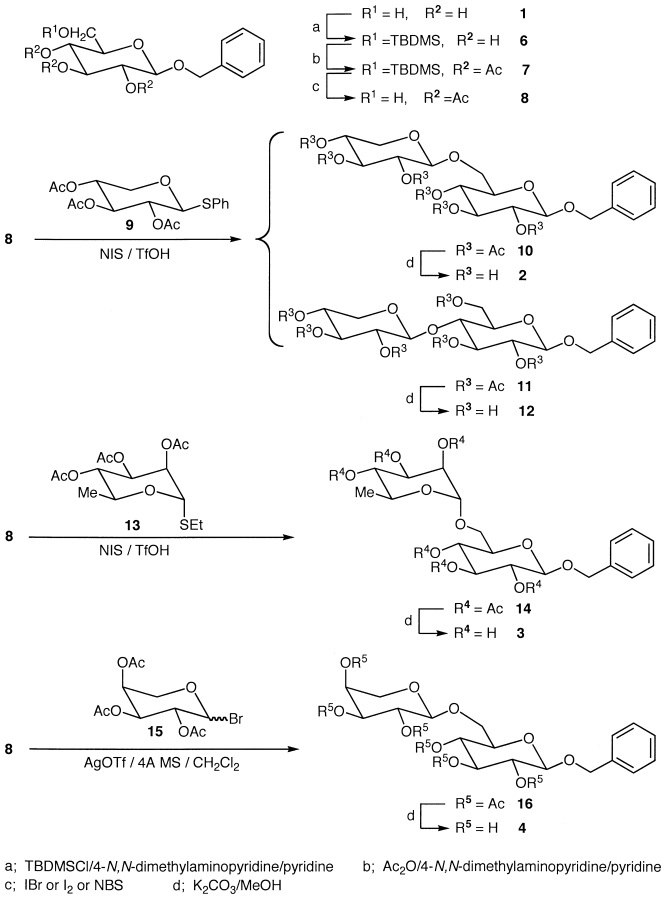

Chart 2

Experimental

^{1}H- and ^{13}C-NMR spectra were recorded on a JEOL JNM-LA 500 spectrometer (Tokyo, Japan). Spectra were recorded with 5—10% (w/v) solution in $CDCl_3$ with Me_4Si as an internal reference. Melting points were determined on a Yanaco MP-3S micromelting point apparatus and are uncorrected. Optical rotations were measured on a JASCO DIP-370 digital polarimeter. The FAB mass spectra were obtained with a JEOL JMS-AX 500 (matrix; *m*-nitrobenzyl alcohol (NBA)) spectrometer. IR spectra were recorded on a JASCO FT/IR-300 spectrometer. All evaporations were performed under reduced pressure. For column chromatography, silica gel (Kieselgel 60) was employed.

Immobilization of β-D-Glucosidase Using a Prepolymer β-D-Glucosidase (EC 3.2.1.21) from almonds was purchased from Sigma Chemical Co. (G-0395, 2.5—3.6 U/mg). Immobilization of β-D-glucosidase from almonds on the photocross-linkable resin prepolymer (ENTP-4000) was carried out using the following procedure. One gram of ENTP-4000 was mixed with 10 mg of a photosensitizer, benzoin ethyl ether, and 110 mg of β-D-glucosidase from almonds (3.4 units/mg). The mixture was layered on a sheet of transparent polyester film (thickness, *ca.* 0.5 mm). The layer was covered with transparent thin film and then illuminated with chemical lamps (wavelength range, 300—400 nm) for 3 min. The gel film thus obtained was cut into small pieces (0.5×5×5 mm) and used for the bioconversion reaction.

Enzymatic Synthesis of Benzyl β-D-Glucopyranoside (1) 1) A mixture of D-glucose **5** (1.1 g, 6.1 mmol), benzyl alcohol (18.8 g, 173.9 mmol), water (2 ml), and the immobilized β-glucosidase was incubated for 4 d at

50 °C. The reaction mixture was filtered off and the filtrate was directly chromatographed on silica gel (35 g) to give benzyl alcohol (16.9 g, 90% recovery) from the $CHCl_3$ eluent and β-glucoside (**1**, 0.875 g, 53% yield) as colorless solid from the $CHCl_3$/MeOH=10 : 1 eluent. The NMR (^{1}H- and ^{13}C-NMR) data of β-glucoside **1** were identical with those of the reported β-glucoside **1**.[7,8]

2) A mixture of D-glucose **5** (1.1 g, 6.1 mmol), benzyl alcohol (18.8 g, 173.9 mmol), water (2 ml), and the recovered immobilized β-glucosidase was incubated for 4 d at 50 °C. The reaction mixture was filtered off and the filtrate was directly chromatographed on silica gel (35 g) to give benzyl alcohol (17.1 g, 91% recovery) from the $CHCl_3$ eluent and β-glucoside (**1**, 0.858 g, 52% yield) as colorless solid from the $CHCl_3$/MeOH=10 : 1 eluent.

Benzyl 6-*O*-*tert*-Butyldimethylsilyl-β-D-glucopyranoside (6) To a solution of **1** (1.00 g, 3.70 mmol) in pyridine (30 ml) was added TBDMSCl (1.10 g, 7.33 mmol) and 4-*N*,*N*-dimethylaminopyridine (DMAP; 10 mg, 0.08 mmol) at 0 °C, and the whole mixture was stirred for 12 h at room temperature. The reaction mixture was condensed and the resulting residue diluted with water and extracted with AcOEt. The organic layer was washed with 10% aqueous HCl, 7% aqueous $NaHCO_3$, and brine. The organic layer was dried over $MgSO_4$ and evaporated to give a residue, which was chromatographed on silica gel (50 g, $CHCl_3$/MeOH (30 : 1)) to afford **6** (1.35 g, 95%) as a colorless amorphous powder. **6**: $[\alpha]_D^{21}$ −58.8° (c=0.873, $CHCl_3$); IR (KBr): 3423 (br), 3032, 2954, 2927, 2881, 2855 cm^{-1}, ^{1}H-NMR ($CDCl_3$): δ 0.00 (6H, s), 0.82 (9H, s), 3.13 (1H, m), 3.25—3.38 (3H, m), 3.66 (1H, dd, J=6.0, 11.2 Hz), 3.81 (1H, dd, J=2.4, 11.2 Hz), 4.17 (1H, d, J=7.2 Hz), 4.25 (1H, br s), 4.44 (1H, br s), 4.47 (1H, d, J=11.6 Hz), 4.73 (1H, d, J=11.6 Hz), 5.20 (1H, br s), 7.12—7.26 (5H, m); ^{13}C-NMR ($CDCl_3$): δ −4.77, −4.75, 18.7, 26.3 [3C], 63.9, 70.7, 71.2, 73.6, 76.1, 76.7, 101.3, 127.9, 128.4 [2C], 128.5 [2C], 137.3; *Anal.* Calcd for $C_{19}H_{32}O_6Si$: C, 59.34; H, 8.39%. Found: C, 58.91; H, 8.86%.

Benzyl 2,3,4-Tri-*O*-acetyl-6-*O*-*tert*-butyldimethylsilyl-β-D-glucopyranoside (7) To a solution of **6** (1.35 g, 3.51 mmol) in pyridine (5 ml) was added Ac_2O (4 ml) and 4-*N*,*N*-dimethylaminopyridine (DMAP; 10 mg, 0.08 mmol) at 0 °C, and the whole mixture was stirred for 12 h at room temperature. The reaction mixture was diluted with water and extracted with AcOEt. The organic layer was washed with 0.5 M aqueous HCl, 7% aqueous $NaHCO_3$, and brine. The organic layer was dried over $MgSO_4$ and evaporated to give a residue, which was chromatographed on silica gel (50 g, *n*-hexane/AcOEt (8 : 1)) to afford **7** (1.79 g, 99%) as a colorless amorphous powder. **7**: $[\alpha]_D^{22}$ −33.2° (c=1.08, $CHCl_3$); IR (KBr): 1759 cm^{-1}, ^{1}H-NMR ($CDCl_3$): δ 0.00 (6H, s), 0.83 (9H, s), 1.92 (3H, s), 1.93 (3H, s), 1.93 (3H, s), 3.44 (1H, m), 3.64 (2H, m), 4.44 (1H, d, J=8.0 Hz), 4.55 (1H, d, J=12.4 Hz), 4.80 (1H, d, J=12.4 Hz), 4.95 (2H, m), 5.08 (1H, t, J=9.2 Hz), 7.18—7.29 (5H, m); ^{13}C-NMR ($CDCl_3$): δ −5.17 [2C], 18.5, 20.8, 20.8, 20.8, 25.9 [3C], 62.5, 69.1, 70.3, 71.5, 73.2, 74.8, 98.9, 127.7 [2C], 127.8, 128.3 [2C], 136.7, 169.2, 169.2, 170.2; *Anal.* Calcd for $C_{25}H_{38}O_9Si$: C, 58.80; H, 7.50%. Found: C, 58.87; H, 7.56%.

Benzyl 2,3,4-Tri-*O*-acetyl-β-D-glucopyranoside (8) 1) Method 1: To a solution of **7** (2.11 g, 4.13 mmol) in MeOH (35 ml) was added 1 M IBr in CH_2Cl_2 solution (6.20 g) at room temperature, and the whole mixture was stirred for 3 h at the same temperature. The reaction mixture was diluted with saturated sodium thiosulfate solution and extracted with $CHCl_3$. The organic layer was washed with brine and dried over $MgSO_4$. Evaporation of the organic solvent gave a residue, which was chromatographed on silica gel (50 g, *n*-hexane/AcOEt (2 : 1)) to afford **8** (1.25 g, 76%) as a colorless amorphous powder. **8**: $[\alpha]_D^{23}$ −44.3° (c=0.75, $CHCl_3$); IR (KBr): 3522, 2962, 1734 cm^{-1}, ^{1}H-NMR ($CDCl_3$): δ 1.99 (6H, s), 2.02 (3H, s), 3.51 (1H, m), 3.60 (1H, dd, J=11.6, 4.8 Hz), 3.73 (1H, d, J=11.6 Hz), 4.59 (1H, d, J=8.0 Hz), 4.64 (1H, d, J=12.4 Hz), 4.88 (1H, d, J=12.4 Hz), 5.04 (2H, m), 5.21 (1H, t, J=9.6 Hz), 7.27—7.359 (5H, m); ^{13}C-NMR ($CDCl_3$): δ 20.4 [2C], 20.4, 61.0, 68.5, 70.7, 71.2, 72.6, 73.9, 99.2, 127.3 [2C], 127.6, 128.0 [2C], 136.5, 168.9, 169.5, 169.8; *Anal.* Calcd for $C_{19}H_{24}O_9$: C, 57.57; H, 6.10%. Found: C, 57.37; H, 6.12%.

2) Method 2: To a solution of **7** (0.79 g, 1.55 mmol) in MeOH (10 ml) was added 2% I_2 in MeOH solution (10 ml) at room temperature, and the whole mixture was stirred for 4 d at the same temperature. The reaction mixture was worked up in the same way as for method 1 to give **8** (0.55 g, 90%) as a colorless amorphous powder.

3) Method 3: To a solution of **7** (0.015 g, 0.03 mmol) in 95% aqueous DMSO (1.0 ml)/THF (0.5 ml)/H_2O (28.5 ml) solution was added NBS (0.003 g, 0.033 mmol) at room temperature, and the whole mixture was stirred for 17 h at the same temperature. The reaction mixture was worked up in the same way as for method 1 to give **8** (0.012 g, 98%) as a colorless amorphous powder.

A Mixtur of Benzyl 2,3,4,2′,3′,4′-*O*-Hexaacetyl-β-D-xylopyranosyl-(1→6)-β-D-glucopyranoside (10) and Benzyl 2,3,6,2′,3′,4′-*O*-Hexaacetyl-β-D-xylopyranosyl-(1→4)-β-D-glucopyranoside (11) A mixture of **8** (0.067 g, 0.168 mmol), **9** (0.109 g, 0.451 mmol), and NIS (0.120 g, 0.533 mmol) in CH_2Cl_2 (5 ml) was stirred for 30 min at 0 °C. To the above mentioned mixture was added methane sulfonic acid (0.048 g, 0.50 mmol) at 0 °C and the whole mixture was stirred for 3 h at the same temperature. The reaction mixture was filtered with the aid of celite and the filtrate was condensed to afford a residue, which was chromatographed on silica gel (20 g, *n*-hexane/AcOEt (3 : 1)) to give a mixture (**10** : **11**=3 : 2, 0.044 g, 40%) as a colorless oil. Major **10**: IR (KBr): 1751 cm^{-1}, ^{1}H-NMR ($CDCl_3$): δ 1.59—2.14 (18H, m), 3.34 (1H, dd, J=8, 12 Hz), 3.64 (1H, m), 3.85 (1H, d, J=10 Hz), 4.13 (1H, dd, J=4, 12 Hz), 4.51 (1H, d, J=8, 12.0 Hz), 4.55 (1H, d, J=6.4 Hz), 4.63 (1H, d, J=12 Hz), 4.87—5.04 (5H, m), 5.15 (2H, m), 7.25—7.35 (5H, m); ^{13}C-NMR ($CDCl_3$): δ 20.7, 20.7, 20.7, 20.8, 20.8, 20.8, 62.0, 67.7, 68.7, 69.0, 70.4, 70.5, 71.2, 71.3, 72.8, 73.2, 76.7, 99.0, 100.5, 127.4, 127.8, 128.3, 128.3, 136.6, 169.1, 169.1, 169.2, 169.6, 169.8, 170.0; HR-FAB-MS (NBA) *m*/*z*: Calcd for $C_{30}H_{38}O_{16}Na$ 677.2031 $(M+Na)^+$. Found: 677.2058.

Benzyl β-D-Xylopyranosyl-(1→6)-β-D-glucopyranoside (2) and Benzyl β-D-Xylopyranosyl-(1→4)-β-D-glucopyranoside (12) A mixture of the above-mentioned mixture (**10**, **11**, 0.026 g, 0.27 mmol) and K_2CO_3 (0.005 g) in MeOH (2 ml) was stirred for 12 h at room temperature. The reaction mixture was condensed to give a residue, which was chromatographed on silica gel (10 g) to afford **2** (0.009 g, 55%) as a colorless amorphous from $CHCl_3$/MeOH (4 : 1) eluent and by-product **12** (0.006 g, 37%) as a colorless amorphous from $CHCl_3$/MeOH (4 : 1). **2**: $[\alpha]_D^{19}$ −48.3° (c=0.76, MeOH); IR (KBr): 3348 (br), 2925, 2872 cm^{-1}, ^{1}H-NMR (CD_3OD): δ 3.09—3.41 (8H, m), 3.68 (1H, d, J=12 Hz), 3.77 (1H, dd, J=5, 12 Hz), 4.02 (1H, d, J=10 Hz), 4.26 (2H, d, J=8 Hz), 4.56 (1H, d, J=12 Hz), 4.82 (1H, d, J=12 Hz), 7.16—7.34 (5H, m); ^{13}C-NMR (CD_3OD): δ 66.9, 69.8, 71.1, 71.5, 71.9, 74.8, 75.0, 77.0, 77.7, 77.9, 103.2, 105.5, 128.5, 129.1 [2C], 129.1 [2C], 138.8; HR-FAB-MS (NBA) *m*/*z*: Calcd for $C_{18}H_{27}O_{10}$: 403.1604 $(M+1)^+$. Found: 403.1579. By-product **12**: $[\alpha]_D^{19}$ −38.6° (c=0.79, MeOH); IR (KBr): 3348 (br), 2925, 2872 cm^{-1}, ^{1}H-NMR ($CDCl_3$): δ 3.17—3.56 (8H, m), 3.82—3.93 (3H, m), 4.33 (1H, d, J=8 Hz), 4.37 (1H, d, J=7.6 Hz), 4.65 (1H, d, J=12 Hz), 4.90 (1H, d, J=12 Hz), 7.24—7.41 (5H, m); ^{13}C-NMR (CD_3OD): δ 61.8, 67.1, 70.9, 71.8, 74.8, 74.9, 76.1, 76.4, 77.8, 80.8, 103.0, 105.3, 128.5, 129.0 [2C], 129.1 [2C], 138.8; HR-FAB-MS (NBA) *m*/*z*: Calcd for $C_{18}H_{27}O_{10}$: 403.1604 $(M+1)^+$. Found: 403.1558.

Ethyl 2,3,4-*O*-Triacetyl-1-thio-α-L-rhamnopyranoside (13) To a solution of 1,2,3,4-*O*-tetraacetyl-α-L-rhamnopyranose (10.0 g, 30.1 mmol) in $CHCl_3$ (50 ml) was added ethane thiol (2.35 g, 39.1 mmol) and 47% $BF_3 \cdot Et_2O$ complex (22.4 g, 74 mmol) at 0 °C and the whole mixture was stirred for 6 h at room temperature. The reaction mixture was diluted with water and the organic layer was washed with 7% aqueous $NaHCO_3$, and brine. The organic layer was dried over $MgSO_4$ and evaporated to give a residue, which was chromatographed on silica gel (50 g, *n*-hexane/AcOEt (5 : 1)) to afford a mixture of α- and β-isomers **13** (8.82 g, 88%). Crystallization of this mixture from ether gave a isomers **13** (4.05 g, 40%) as a colorless needles. **13**: mp 135 °C; $[\alpha]_D^{23}$ +69.1° (c=1.06, $CHCl_3$); IR (KBr): 1749 cm^{-1}, ^{1}H-NMR ($CDCl_3$): δ 1.29 (3H, d, J=6 Hz), 1.99 (3H, s), 2.06 (3H, s), 2.16 (3H, s), 2.58—2.69 (2H, m), 4.24 (1H, m), 5.10 (1H, t, J=10 Hz), 5.20 (1H, d, J=1.6 Hz), 5.24 (1H, dd, J=10, 3.2 Hz), 5.34 (1H, dd, J=1.6, 3.2 Hz); ^{13}C-NMR ($CDCl_3$): δ 14.9, 17.7, 20.6, 20.6, 20.8, 25.6, 70.4, 70.8, 71.9, 74.9, 81.9, 169.8, 170.2, 170.3; *Anal.* Calcd for $C_{14}H_{22}O_7S$; C, 50.29; H, 6.63%. Found: C, 50.43; H, 6.64%.

Benzyl 2,3,4,2′,3′,4′-*O*-Hexaacetyl-α-L-rhamnopyranosyl-(1→6)-β-D-glucopyranoside (14) A mixture of **8** (0.155 g, 0.39 mmol), **13** (0.13 g, 0.39 mmol), molecular sieves 4A (0.5 g) and NIS (0.187 g, 0.83 mmol) in CH_2Cl_2 (5 ml) was stirred for 30 min at 0 °C. To the above mentioned mixture was added methane sulfonic acid (0.098 g, 1.02 mmol) at 0 °C and the whole mixture was stirred for 3 h at the same temperature. The reaction mixture was filtered with the aid of celite and the filtrate was condensed to afford a residue, which was chromatographed on silica gel (20 g, *n*-hexane/AcOEt (3 : 1)) to give **14** (0.20 g, 76%) as a colorless amorphous powder. **14**: $[\alpha]_D^{21}$ −71.5° (c=1.08, $CHCl_3$); ^{1}H-NMR ($CDCl_3$): δ 1.22 (3H, d, J=4.4 Hz), 2.00 (3H, s), 2.00 (3H, s), 2.01 (3H, s), 2.05 (3H, s), 2.05 (3H, s), 2.12 (3H, s), 3.65—3.75 (3H, m), 3.85—3.93 (1H, m), 4.56 (2H, d, J=8 Hz), 4.63 (2H, d, J=12 Hz), 4.86 (1H, d, J=0.8 Hz), 4.87 (2H, d, J=12 Hz), 4.96 (1H, t, J=9.6 Hz), 5.04 (1H, dd, J=9.6, 8.0 Hz), 5.05—5.10 (1H, m), 5.28—5.31 (2H, m), 5.78 (1H, t, J=9.6 Hz), 7.26—7.37 (5H, m); ^{13}C-NMR ($CDCl_3$): δ 17.2, 20.4, 20.4, 20.45, 20.5, 20.6, 20.65, 66.4, 66.7, 68.8, 69.2, 69.4, 70.4, 70.8, 71.1, 72.6, 73.3, 90.0, 99.0, 127.5 [2C], 127.7,

128.3 [2C], 136.6, 169.1, 169.3, 169.7, 169.75, 169.8, 170.0; *Anal.* Calcd for $C_{31}H_{40}O_{16}$; C, 55.68; H, 6.03%. Found: C, 55.44; H, 5.92%.

Benzyl α-L-Rhamnopyranosyl-(1→6)-β-D-glucopyranoside (3) A mixture of **13** (0.493 g, 0.73 mmol) and K_2CO_3 (0.005 g) in MeOH (2 ml) was stirred for 12 h at room temperature. The reaction mixture was condensed to give a residue, which was chromatographed on silica gel (10 g, $CHCl_3$/MeOH (4 : 1)) to afford **3** (0.26 g, 85%) as a colorless oil **3**: $[\alpha]_D^{19}$ −52.8° (c=1.37, MeOH); IR (KBr): 3375 (br), 2926 cm^{-1}, ^{1}H-NMR (CD_3OD): δ 1.24 (3H, d, J=8 Hz), 3.22—3.40 (3H, m), 3.55—3.71 (3H, m), 3.87 (1H, dd, J=1, 3 Hz), 3.94 (1H, d, J=7.6 Hz), 4.29 (1H, d, J=8 Hz), 4.58 (1H, d, J=12 Hz), 4.83 (1H, d, J=12 Hz), 7.20—7.38 (5H, m); ^{13}C-NMR (CD_3OD): δ 18.11, 67.95, 69.60, 71.42, 71.61, 71.95, 72.17, 73.76, 74.78, 76.59, 77.08, 101.92, 102.81, 128.50, 129.03 [2C], 129.04 [2C], 138.41; HR-FAB-MS (NBA) *m/z*: Calcd for $C_{19}H_{28}O_{10}Na$; 439.1580 $(M+Na)^+$. Found: 439.1601.

Benzyl 2,3,4,2′,3′,4′-*O*-Hexaacetyl-α-L-arabinopyranosyl-(1→6)-β-D-glucopyranoside (16) To a solution of **8** (0.6 g, 1.52 mmol) in CH_2Cl_2 (10 ml) was added 2,3,4-tri-*O*-acetyl-α-L-arabinopyranosyl bromide (**15**, 1.03 g, 3.03 mmol) and tetramethylurea (TMU, 0.704 g, 6.6 mmol) and AgOTf (0.778 g, 3.03 mmol) at 0 °C under argon atmosphere. The whole was covered with aluminum foil and stirred for 3.5 h at room temperature. The reaction mixture was cooled at 0 °C and quenched with AcOEt (100 ml) and 7% aqueous $NaHCO_3$ solution (20 ml). The organic layer was washed with brine and dried over Na_2SO_4. Evaporation of the organic solvent gave a residue, which was chromatographed on silicagel (20 g, *n*-hexane/AcOEt (4 : 1)) to afford **16** (0.4 g, 40% yield) as a colorless amorphous. **16**: $[\alpha]_D^{26}$ −32.1° (c=1.01, $CHCl_3$); IR (KBr): 1739 cm^{-1}, ^{1}H-NMR ($CDCl_3$): δ 1.99 (3H, s), 2.01 (3H, s), 2.02 (3H, s), 2.03—2.04 (6H, m), 2.14 (3H, s), 3.57—3.62 (2H, m), 3.65—3.71 (1H, m), 3.90 (1H, dd, J=1.5, 10.6 Hz), 4.31 (1H, dd, J=3.5, 12.1 Hz), 4.48 (1H, d, J=7.0 Hz), 4.52 (1H, d, J=8.0 Hz), 4.61 (1H, d, J=12 Hz), 4.89 (1H, d, J=12 Hz), 4.95 (1H, dd, J=9.6, 9.6 Hz), 5.03 (1H, dd, J=9.6, 9.6 Hz), 5.02—5.05 (1H, m), 5.15 (1H, d, J=9.6 Hz), 5.17—5.26 (2H, m), 7.28—7.38 (5H, m); ^{13}C-NMR ($CDCl_3$): δ 20.69, 20.73 [2C], 20.75, 20.89, 21.01, 63.18, 67.61, 67.99, 69.13, 69.22, 70.12, 70.54, 71.43, 72.97, 73.28, 99.17, 100.91, 127.71 [2C], 128.05, 128.56 [2C], 136.85, 169.45, 169.53, 169.68, 170.21, 170.32, 170.36; HR-FAB-MS (NBA) *m/z*: Calcd for $C_{30}H_{39}O_{16}$ 655.2238 $(M+1)^+$. Found: 655.2241.

Benzyl β-D-Arabinopyranosyl-(1→6)-β-D-glucopyranoside (4) A mixture of **16** (0.15 g, 0.23 mmol) and K_2CO_3 (0.032 g) in a mixed solvent (MeOH : CH_2Cl_2=5 : 1; 12 ml) was stirred for 15 min at room temperature. The reaction mixture was condensed to give a residue, which was chromatographed on silica gel (10 g, CH_2Cl_2/EtOH (5 : 1)) to afford **4** (0.062 g, 69%). **4**: $[\alpha]_D^{24}$ −33.6° (c=0.025, H_2O); IR (KBr): 3600—3100 cm^{-1}, ^{1}H-NMR (CD_3OD+D_2O): δ 3.17—3.22 (1H, m), 3.30—3.39 (2H, m), 3.40—3.52 (4H, m), 3.72 (1H, dd, J=5.0, 11.6 Hz), 3.76—3.84 (2H, m), 4.04 (1H, dd, J=2.0, 11.6 Hz), 4.26 (1H, d, J=7.0 Hz), 4.35 (1H, d, J=7.6 Hz), 4.60 (1H, d, J=12 Hz), 4.83 (1H, d, J=12 Hz), 7.21—7.37 (5H, m); ^{13}C-NMR (CD_3OD+D_2O): δ 66.31, 66.99, 69.38, 70.83, 72.01, 72.23, 73.70, 74.52, 76.36, 77.24, 102.88, 104.88, 129.02, 129.32 [2C], 129.46 [2C], 138.26; HR-FAB-MS (NBA) *m/z*: Calcd for $C_{18}H_{27}O_{10}$ 403.1604 $(M+1)^+$. Found: 403.1594.

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