Synthesis of Glucosides Related to Grape and Wine Aroma Precursors

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Syntheses of the following glucosides are reported: 3′,5′,5′-trimethyl-3′-cyclohexenyl β-D-glucopyranoside, 3′-(1″-cyclohexenyl)-1′-methyl-2′-propynyl β-D-glucopyranoside, (3′*R**,9′*S**)-3′-hydroxy-5′-megastigmen-7′-yn-9′-yl β-D-glucopyranoside, (*E*,3′*R**)-7′-oxo-5′,8′-megastigmadien-3′-yl β-D-glucopyranoside, and 4′-oxo-5′-megastigmen-9′-yl β-D-glucopyranoside.

Keywords: *Damascenone; β-glycosides; precursor; flavor*

INTRODUCTION

In many fruits and fruit products, including wines, there are present free aroma compounds plus complex mixtures of glycosides which, on hydrolysis catalyzed by mild acid or glycosidase enzymes, can also yield important aroma compounds (Williams, 1993; Williams et al., 1993). Research on grapes has shown that most of the known aroma compounds are shikimate- and mevalonate-derived metabolites, with the latter group containing a number of norisoprenoid compounds that are thought to arise from the degradation of carotenoids (Sefton et al., 1989; Williams et al., 1992; Razungles et al., 1993). We have recently shown that the important flavor compound β-damascenone (**1**) [i.e. (*E*)-3,5,8-megastigmatrien-7-one] was formed by acid-catalyzed hydrolysis but not by glycosidase enzyme-catalyzed hydrolysis of grape glycoside fractions (Sefton et al., 1993).

This paper describes the synthesis of some 13-carbon norisoprenoid and related glucosides that were prepared to study the role of such compounds as precursors of β-damascenone. Results of the acid-catalyzed hydrolysis and fungal enzyme studies on some of the synthetic substrates have been published (Sefton et al., 1989; Sefton and Williams, 1991; Skouroumounis et al., 1992, 1993). In addition to enabling the hydrolytic studies to be undertaken, the syntheses have also allowed us to explore methods for the optimal preparation of glycosidic precursors and provided reference material for the characterization of some of the glycosides as natural products.

MATERIALS AND METHODS

Safety Precautions. The preparation of the glycosylating agent 2,3,4,6-tetra-*O*-acetyl-D-glucopyranosyltrichloroacetimidate according to the method of Schmidt and Michel (1980) requires special care. Sodium hydroxide, which can be present in old stocks of sodium hydride, reacts explosively with trichloroacetonitrile after a brief initiation period. Only fresh stocks of sodium hydride should be used. An alternative base is 1,8-diazabicyclo[5.4.0]undecene (DBU) [see Tavecchia et al. (1989)].

General Methods. ^{1}H NMR spectra were recorded with a JEOL JNM-PMX (60 MHz) or a Bruker CXP300 (300 MHz) spectrometer. Chemical shifts and coupling constants for H2 to H6 protons of 2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosides (as well as of 2,3,4,6-tetra-*O*-pivaloyl-β-D-glucopyranosides) were essentially the same as reported elsewhere (Sefton et al., 1992, and references cited therein), except that where mixtures of diastereoisomers were analyzed, a doubling of signals was sometimes observed. Other signals are given below. ^{13}C NMR spectra were recorded with a Bruker CXP300 (75.47 MHz) or WP80 DS (20.1 MHz) instrument. IR spectra were recorded with a Jasco A-102 spectrophotometer, using the 1603 cm^{-1} band of polystyrene as a reference, and are of nujol mulls.

Glycosides were analyzed by GC/MS with a Finnigan 4021 or Finnigan TSQ 70 mass spectrometer, coupled to a Varian 3400 gas chromatograph. The chromatograph was equipped with either a 15 m J&W DB-1701 (column A) or a 30 m J&W DB-5 (column B) fused silica column, each 0.25 mm i.d., 0.25 μm film thickness, with helium carrier gas at a linear velocity of 40 cm/s. Injections were made with a split injector at 200 °C and a split ratio of 1:10. Both columns were held at 100 °C for 5 min, then programmed at 5 °C/min to 320 °C, and held at that temperature for 20 min. Electron impact spectra were taken at 70 eV. They were also analyzed using fast atom bombardment (FAB) and chemical ionization (CI) (using ammonia as ionizing gas) with the Finnigan TSQ 70 mass spectrometer. Accurate mass measurements were performed with a DS90 mass spectrometer.

Glycoside Preparation and Isolation. The following general procedure for glycosylation using the orthoester (**7**) was used (Kunz and Pfrengle, 1986). Under strictly anhydrous conditions and under a nitrogen atmosphere, boron trifluoride etherate (4.8 equiv) was added to a solution of the alcohol (1.0 equiv., freshly distilled or dried at 0.01 mm at room temperature for 8 h) and orthoester (**7**) (1.2 equiv., dried at 0.01 mm at room temperature for 8 h) in dichloromethane (freshly distilled from calcium hydride) at room temperature. Progress of the reaction was monitored by TLC. On completion of the reaction the solution was treated with a saturated solution of sodium hydrogen carbonate, ensuring the aqueous layer was basic after stirring. The organic layer was then separated and washed with water (10 mL) and saturated aqueous sodium chloride solution (10 mL), dried ($MgSO_4$), and evaporated under reduced pressure to give generally clear oils which were chromatographed to isolate the 2,3,4,6-tetra-*O*-pivaloyl-β-D-glucopyranosides.

The general procedure used for depivaloylation was that of Zemplén and Kunz (1923). Under a nitrogen atmosphere, the tetra-*O*-pivaloyl-β-D-glucopyranosides were treated with 0.01 M sodium methoxide/anhydrous methanol for several days (7 days generally). Isolation of the β-D-glucopyranosides involved the following procedure. The solution from the methanolysis

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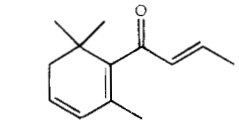
was evaporated to dryness under reduced pressure. The residue was then dissolved in water, and the aqueous layer was extracted with diethyl ether (2 × 10 mL). The aqueous layer was retained and then evaporated to dryness and the residue weighed. The residue was then redissolved in water (1 mL/10 mg of residue) and the pH adjusted by addition of ammonium chloride to neutral pH. The aqueous layer (1 mL, containing approximately 10 mg of residue) was loaded onto a Sep-Pak C_{18} RP cartridge (Waters Associates, part no. 51910), which was eluted with water (20 mL) to remove inorganic salts. The cartridge was then eluted with 2 mL of methanol which was kept separately. This procedure was repeated with the remaining solution. The methanol extract was then evaporated under reduced pressure to yield the β-D-glucopyranoside. The purity of the β-D-glucopyranoside was also monitored by TLC, and further purification was performed by chromatography if required. For larger quantities of product the Bond Elut LRC (C_{18} octadecyl) was used, where up to 100 mg of material can be loaded, and the use of an aspirator ensured rapid separation.

4′-Oxo-5′-megastigmen-9′-yl 2,3,4,6-Tetra-*O*-acetyl-β-D-glucopyranoside (6c). This was prepared according to the method of Ogawa et al. (1981). Under dry conditions and a



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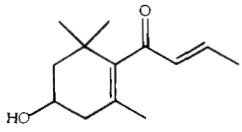

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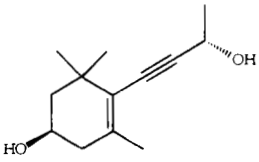

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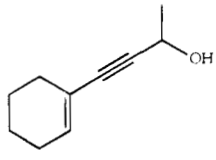

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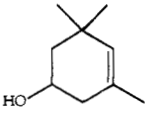

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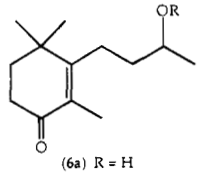
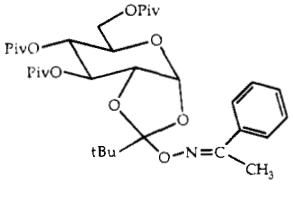


(6a) R = H
(6b) R = Ac
(6c) R = β-D-Glu(OAc)$_4$
(6d) R = α-D-Glu(OAc)$_4$



(7)

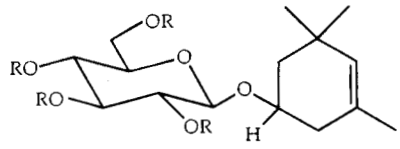
nitrogen atmosphere, 9-hydroxy-5-megastigmen-4-one (**6a**), mp 55–57 °C, from hydrogenation of (*E*)-9-hydroxy-5,7-megastigmadien-4-one (Kaiser and Lamparsky, 1978) using palladium black (90 mg, 0.43 mmol), and 1,2,3,4,6-penta-*O*-acetyl-β-D-glucopyranoside (170 mg, 0.44 mmol) in 1,2-dichloroethane (10 mL) was refluxed over 4 Å molecular sieves for 1 h as described by Nishizawa et al. (1988). The solution was then cooled to 20 °C and trimethylsilyl triflate (99 mg, 86 μL, 0.44 mmol) was added. After 3 h of stirring, dichloromethane (20 mL) was added. The solution was washed with saturated sodium hydrogen carbonate solution (2 × 20 mL), water (10 mL), and saturated aqueous sodium chloride solution (10 mL) and dried ($MgSO_4$), and the solvent was evaporated to yield a clear oil. Chromatography (petroleum ether/ethyl acetate gradient) gave three products. 4-Oxo-5-megastigmen-9-yl acetate (**6b**) as a gum (50%): ^{1}H NMR (60 MHz, $CDCl_3$) δ 1.2 [s, C-1-(Me)$_2$], 1.32 [d, J = 7 Hz, (H-10)$_3$], 1.8 (s, C-5-Me), 2.1 (s, CH_3CO), 2.1–2.7 (m, 4H), 5.0 (m, H-9). 4′-Oxo-5′-megastigmen-9′-yl 2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranoside (**6c**) as a gum (21%): found, $M^{\bullet+}$, 540.2567, $C_{27}H_{40}O_{11}$ requires 540.2570; ν_{max} 1750, 1220, 1650, 1038 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.14 [s, C-1′-(Me)$_2$], 1.30 [d, J = 6.1 Hz, (H-10′)$_3$], 1.63–1.69 (m, 2H), 1.78 (s, C-5′-Me), 1.82–1.99 (m, 2H), 2.00, 2.01 (each s, approximately 3H), 2.02, 2.03, 2.04, 2.06 (each s, approximately 1.5H, CH_3CO), 2.42–2.48 (m, 2H), 3.6–3.9 (m, H-9′, H-5), 4.58 (d, J = 8.0 Hz, H-1); (column A) diastereoisomer 1, m/z 540 (<1), 420 (<1), 360 (3), 331 (8), 271 (4), 193 (21), 169 (38), 152 (16), 109 (100), 81 (23), 55 (20); (column A) diastereoisomer 2, m/z 540 (<1), 420 (<1), 360 (3), 331 (7), 271 (4), 193 (21), 169 (38), 152 (23), 109 (100), 81 (23), 55 (18). 4′-Oxo-5′-megastigmen-9′-yl 2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranoside (**6d**) as a gum (9%): ν_{max} 1750, 1650, 1220, 1038 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.17 [s, C-1′-(Me)$_2$], 1.26 [d, J = 6.2 Hz, (H-10′)$_3$], 1.54–1.74 (m, 2H), 1.78 (s, C-5′-Me), 1.82 (apparent t, J = 6.9 Hz, 2H), 2.02, 2.04, 2.09, 2.10 (each s, CH_3CO); 2.16–2.23 (m, 2H), 2.46 (apparent t, J = 6.9 Hz, 2H), 3.80–3.91 (m, H-9′), 4.0–4.3 [m, H-5, (H-6)$_2$], 4.92 (dd, J = 10.1, 3.5 Hz, H-2), 5.09 (dd, J = 9.8, 9.7 Hz, H-4), 5.47 and 5.49 (each d, J = 3.5 Hz, H-1 for each of the diastereoisomers), 5.54 (dd, J = 10.1, 9.8 Hz, H-3); (column A) diastereoisomer 1, m/z 540 (<1), 485 (<1), 429 (<1), 355 (<1), 331 (10), 271 (5), 207 (10), 193 (18), 169 (33), 152 (18), 109 (100), 81 (30), 55 (33); (column A) diastereoisomer 2, m/z 540 (<1), 484 (<1), 429 (<1), 355 (<1), 331 (10), 207 (6), 193 (22), 169 (28), 152 (20), 109 (100), 81 (30), 55 (28).

1,2-Di-*O*-[1′-[[*N*-(1-phenylethylidene)amino]oxy]-2′,2′-dimethylpropylidene]-3,4,6-tri-*O*-pivaloyl-α-D-glucopyranose (7). The orthoester (**7**) was synthesized according to the method of Kunz and Pfrengle (1986), using silver triflate, in 80% yield: mp 112 °C [lit. (Kunz and Pfrengle, 1986) 112–113 °C]; ν_{max} 1730, 1480, 1385, 1360, 1280, 1120 cm^{-1}; ^{13}C NMR δ (20.1 MHz, $CDCl_3$) 12.5, 24.7, 25.6, 26.7, 38.3, 62.2, 67.2, 67.8, 73.0, 77.4, 77.7, 99.0, 99.4, 125.6, 126.7, 128.3, 129.2, 136.2, 156.6, 159.4, 176.6, 177.8. A small amount of an isomer, mp 105–108 °C, was also isolated by chromatography: ν_{max} 1730, 1480, 1385, 1360, 1280, 1120 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.05, 1.08, 1.14, 1.16 (each s, CMe_3), 2.30 (s, =CMe), 3.93 (dd, J = 12.5, 2.9 Hz, H-6), 4.07 (dd, J = 12.5, 1.7 Hz, H-6), 4.67 (ddd, J = 10.0, 2.9, 1.7 Hz, H-5), 5.11 (dd, J = 10.0, 9.8 Hz, H-4), 5.78 (d, J = 6.6 Hz, H-1), 5.97 (dd, J = 9.8, 4.8 Hz, H-3), 7.34–7.40 (m, 3H, ArH), 7.67–7.70 (m, 2H, ArH).

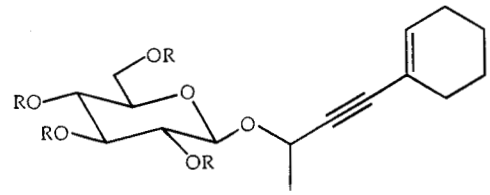
Preparation of 4-(1′-Cyclohexenyl)-3-butyn-2-ol (4). Addition of the dianion from 3-butyn-2-ol to cyclohexanone, following the method for analogous ketones (Loeber et al., 1971), gave 1-(3′-hydroxy-1-butynyl)cyclohexanol in 96% yield, bp 104 °C/0.07 mmHg with ^{1}H NMR data identical with those reported (Saimoto et al., 1983). This diol was acetylated with acetic anhydride in pyridine to yield, after chromatography, 3-(1′-hydroxycyclohexyl)-1-methyl-2-propynyl acetate (78%): ^{1}H NMR (60 MHz, $CDCl_3$) δ 1.55 [d, J = 7 Hz, C(OAc)-Me], 1.2–1.8 (complex CH and CH_2), 2.1 (s, CH_3CO), 3.0 (br, 1H, OH), 5.58 (q, J = 7 Hz, H-1). Phosphoryl chloride (2.8 mL) was added dropwise to this hydroxy acetate (5 g) in triethylamine (10 mL) at room temperature. After 18 h, ether (100 mL) was added, the ether was washed with ice/water (10 mL), saturated sodium hydrogen carbonate solution (30 mL), dried ($MgSO_4$), and evaporated. Distillation yielded 3-(1′-cyclohexenyl)-1-methyl-2-propynyl acetate, bp 85 °C/0.5 mmHg as a yellow oil (4 g, 88%): ^{1}H NMR (60 MHz, $CDCl_3$) δ 1.5 [d, J = 6 Hz, C(OAc)-Me], 1.4–1.8 (m, 4H), 2.0–2.4, (m, 4H), 2.03 (s, CH_3CO), 5.6 (q, J = 6 Hz, H-1), 6.2 (br, H-2′). Reduction of the enyne acetate with lithium aluminum hydride in ether gave the readily autoxidized 4-(1′-cyclohexenyl)-3-butyn-2-ol (**4**) (84%): found, C, 79.6; H, 9.4; $C_{10}H_{14}O$ requires C, 80.0; H, 9.4%. Spectral data were identical with those of **4** prepared by a different route (Delbecq et al., 1979).

3′,5′,5′-Trimethyl-3′-cyclohexenyl 2,3,4,6-Tetra-*O*-pivaloyl-β-D-glucopyranoside (8a). 3,5,5-Trimethyl-3-cyclohexen-1-ol (**5**) (200 mg), using the method of Kunz and Pfrengle (1986), gave, after 10 min and standard workup, the β-D-

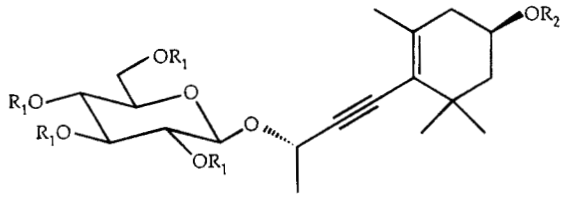
glycoside (**8a**, 90%) as a crystalline solid. Recrystallization



(**8a**) R = Piv
(**8b**) R = H
(**8c**) R = Ac



(**9a**) R = Piv
(**9b**) R = H
(**9c**) R = Ac



(**10a**) R_1 = Piv, R_2 = H
(**10b**) R_1 = H, R_2 = H
(**10c**) R_1 = Ac, R_2 = Ac

from methanol yielded one diastereoisomer: mp 137–139 °C; found, C, 66.0; H, 9.0; $C_{35}H_{58}O_{10}$ requires C, 65.8; H, 9.1%; ν_{max} 1735, 1390, 1280, 1140 cm^{-1}; ^1H NMR (300 MHz/$CDCl_3$) δ 0.93, 0.97 [s, C-5′–$(Me)_2$], 1.11 (s, CMe_3), 1.17 (s, 2 × CMe_3), 1.22 (s, CMe_3), 1.24–1.43 (m, 1H), 1.61 (s, C-3′–Me), 1.64–1.74 (m, 1H), 2.00 (dd, J = 16.8, 9.7 Hz, H-2′ax), 2.24 (dd, J = 16.8, 5.8 Hz, H-2′eq), 3.90 (m, H-1′), 4.66 (d, J = 8.1 Hz, H-1), 5.06 (s, H-4′); ^{13}C NMR (20.1 MHz, $CDCl_3$) δ 23.2, 27.1, 29.4, 31.3, 33.7, 37.8, 38.7, 42.7, 62.4, 68.4, 71.4, 72.3, 74.8, 99.9, 128.7, 131.4, 176.4, 176.6, 177.2, 178.0; second diastereoisomer (data on mixture) δ 0.95, 0.98 (s, 2 × C-5′–Me), 1.11 (s, CMe_3), 1.16 (s, 2 × CMe_3), 1.22 (s, CMe_3), 1.24–1.43 (m, 1H), 1.62 (s, C-3′–Me), 1.64–1.70 (m, 1H), 1.84 (m, 1H), 2.15 (dd, J = 16.5, 5.6 Hz, 1H), 3.90 (m, H-1′), 4.69 (d, J = 8.0 Hz, H-1), 5.06 (m, H-4′).

3′,5′,5′-Trimethyl-3′-cyclohexenyl β-D-Glucopyranoside (8b). Depivaloylation using the method of Zemplén and Kunz (1923) yielded the diastereoisomeric β-D-glucopyranosides (**8b**) as a crystalline solid in 90% yield. The pure diastereoisomer of **8a** gave **8b** (diastereoisomer 1) as a white solid: mp 118–124 °C; ^{13}C NMR (20.1 MHz, D_2O, dioxane as external standard) δ 23.8, 29.9, 31.8, 34.6, 38.4, 43.1, 62.2, 71.1, 76.4, 77.3, 102.2, 131.3, 133.4; FAB positive ion, m/z 303 ($[M + H]^+$, 35), 163 (40), 145 (42), 124 (66), 107 (100); diastereoisomer 2 (data on mixture) ^{13}C NMR (20.1 MHz, D_2O, dioxane as external standard) δ 23.8, 29.9, 31.8, 34.8, 37.1, 44.5, 62.2, 71.1, 75.8, 76.3, 77.3, 102.1, 130.7, 133.4.

3′,5′,5′-Trimethyl-3′-cyclohexenyl 2,3,4,6-Tetra-*O*-acetyl-β-D-glucopyranoside (8c). *(a) From the Glucoside (8b).* Acetylation of the glucoside (**8b**, the pure diastereoisomer) with acetic anhydride and pyridine gave a single diastereoisomer (**8c**): mp 95–97 °C; ν_{max} 1740, 1430, 1360, 1240, 1160, 1040 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$) δ 0.93, 0.96 [s, C-5′–$(Me)_2$], 1.08–1.29 (m, 2H), 1.59 (s, C-3′–Me), 1.97, 1.99, 2.01, 2.05 (s, CH_3CO), 2.02 (m, 1H), 2.19 (dd, J = 16.6, 5.4 Hz, H-2_{eq}′), 3.88 (m, H-1′), 4.59 (d, J = 8.3 Hz, H-1), 5.05 (s, H-4′); ^{13}C NMR (75.47 MHz, $CDCl_3$) δ 20.6, 20.7, 23.2, 29.5, 31.3, 33.7, 37.7, 42.5, 62.2, 68.7, 71.6, 71.8, 72.9, 75.1, 99.8, 128.8, 131.3, 169.2, 169.4, 170.3, 170.6.

(b) From the Alcohol (5). On the basis of the procedure of Schmidt and Michel (1980), 3,5,5-trimethyl-3-cyclohexenol (**5**, 14 mg, 0.1 mmol) and imidate (70 mg, 0.14 mmol) in dichloromethane (3 mL) were refluxed over 4 Å sieves for 1 h (Nishizawa et al., 1988). The reaction mixture was cooled to 25 °C, boron trifluoride etherate (17 μL, 0.1 mmol) was added, and the mixture was stirred for a further 30 min. Isolation and chromatography (petroleum ether/ethyl acetate) yielded (**8c**) (10 mg, 21%): ^1H NMR (300 MHz, $CDCl_3$) δ 4.63 (d, J = 7.9 Hz), 4.61 (d, J = 8.2 Hz, H-1 of each of the two diastereoisomers).

3′-(1″-Cyclohexenyl)-1′-methyl-2′-propynyl 2,3,4,6-Tetra-*O*-pivaloyl-β-D-glucopyranoside (9a). This glycoside was prepared from the enyne alcohol (**4**) (200 mg) according to the method of Kunz and Pfrengle (1986) under an atmosphere of oxygen-free nitrogen. The reaction mixture was stirred for 10 min at −20 °C, and the β-D-glucopyranoside (**9a**, 60%) was isolated as a gum, as described above. Higher R_f diastereoisomer: ν_{max} 2240, 1740, 1390, 1365, 1280, 1040 cm^{-1}; ^1H NMR (300 MHz, $CDCl_3$) δ 1.04, 1.08, 1.11, 1.15 (each s, CMe_3), 1.33 (d, J = 6.7 Hz, C-1′–Me), 1.5–1.6 (m, 4H), 2.0–2.2 (m, 4H), 4.62 (q, J = 6.7 Hz, H-1′), 4.80 (d, J = 8.3 Hz, H-1), 6.01, (br s, H-2″); ^{13}C NMR (20.1 MHz, $CDCl_3$) δ 21.2, 22.1, 22.3, 25.4, 27.0, 28.9, 38.6, 62.1, 66.8, 68.0, 71.0, 72.3, 84.8, 87.8, 98.6, 120.0, 135.7, 176.6, 177.3, 178.1, 178.3; CI (NH_4^+), m/z 666 ($[M + NH_4]^+$, 9), 634 (4), 535 (4), 534 (13), 533 (11), 532 (30), 500 (24), 499 (56), 265 (24), 211 (39), 195 (10), 194 (21), 181 (22), 160 (7), 149 (8), 146 (7), 136 (14), 134 (36), 133 (92), 132 (78), 131 (13), 121 (7), 120 (29), 119 (19), 118 (26), 117 (15), 110 (7), 109 (9), 106 (12), 105 (35), 104 (9), 103 (16), 102 (13), 97 (13), 92 (7), 91 (30), 86 (9), 85 (85), 81 (12), 79 (9), 78 (9), 77 (18), 69 (13), 65 (7), 58 (19), 57 (100), 56 (15), 55 (20), 53 (9), 51 (8). Diastereoisomer 2 could not be obtained pure, but H-1 and H-2″ appeared at δ 4.76 and 5.95, respectively.

3′-(1″-Cyclohexenyl)-1′-methyl-2′-propynyl β-D-Glucopyranoside (9b). Depivaloylation of **9a** (mixture of diastereoisomers) according to the method of Zemplén and Kunz (1923) yielded the β-D-glucopyranoside (**9b**) as a gum in 41% yield: major diastereoisomer ^{13}C NMR (75.47 MHz, D_2O, *tert*-butyl alcohol as external standard) δ 23.0, 23.5, 23.8, 27.3, 30.6, 62.7, 66.6, 71.5, 74.9, 77.9, 78.0, 87.3, 90.1, 101.4, 121.5, 138.7.

3′-(1″-Cyclohexenyl)-1′-methyl-2′-propynyl 2,3,4,6-Tetra-*O*-acetyl-β-D-glucopyranoside (9c). Acetylation of the β-D-glucopyranoside (**9b**) (5 mg) in acetic anhydride (10 μL) and pyridine (1 mL) yielded (**9c**) as a gum, as an unequal mixture of diastereoisomers (5 mg, 64%): ν_{max} 2920, 2250, 1750, 1700, 1600, 1360, 1230, 1220, 1040 cm^{-1}; major diastereoisomer ^1H NMR (300 MHz, $CDCl_3$) δ 1.4 (d, J = 6.6 Hz, C-1′–Me), 1.59–1.65 (m, 4H), 2.00, 2.01, 2.02, 2.06 (each CH_3CO), 2.05–2.09 (m, 4H), 4.68 (q, J = 6.6 Hz, H-1′), 4.83 (d, J = 7.8 Hz, H-1), 6.09 (br s, H-2″); ^{13}C NMR (75.47 MHz, $CDCl_3$) δ 20.6 (q), 21.4 (q), 22.2 (t), 25.6 (t), 29.2 (t), 62.0 (t), 64.4 (d), 68.7 (d), 71.9 (d), 73.0 (d), 84.9 (s), 87.8 (s), 97.8 (d), 120.0 (s), 135.6 (d), 169.4 (s), 170.3 (s), 170.6 (s); (column B) m/z 480 (3), 439 (5), 420 (6), 331 (3), 245 (4), 176 (5), 169 (28), 139 (23), 132 (100), 105 (30), 91 (28); ν_{max} 2920, 2250, 1750, 1700, 1600, 1360, 1230, 1220, 1040 cm^{-1}.

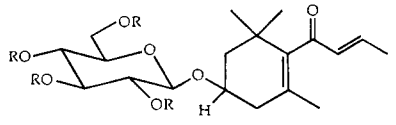
(3′*R, 9′*S**)-3′-Hydroxy-5′-megastigmen-7′-yn-9′-yl 2,3,4,6-Tetra-*O*-pivaloyl-β-D-glucopyranoside (10a).** This compound was prepared according to the method of Kunz and Pfrengle (1986) under an oxygen-free nitrogen atmosphere. The enyne diol diastereoisomer, (3*R**,9*S**)-5-megastigmen-7-yne-3,9-diol (**3**) (Loeber et al., 1971) (90 mg), prepared from the triol, (3′*R**,1*R**,4*S**,6*S**)-1-(3′-hydroxybut-1′-ynyl)-2,2,6-trimethylcyclohexane-1,4-diol (Massy-Westropp et al., 1992), was stirred for 4 h (−10 °C → 25 °C) and yielded **10a** (54%), a gum, as a 1:1 mixture of two diastereoisomers: ^1H NMR (300 MHz, $CDCl_3$) δ 1.0–1.6 (complex), 1.37 [d, J = 6.6 Hz, (H-10′$_3$–Me], 1.88 (s, C-5′–Me), 2.05 (m, 1H), 2.35 (m, 1H), 3.6–4.2 (m, 4H), 4.72 (q, J = 6.7 Hz and 4.83, q, J = 6.6 Hz, H9′ of two diastereoisomers); 4.89, 4.93 (each d, J = 8.2 Hz, H-1 of both diastereoisomers); 5.01–5.16 (m, 2H), 5.31 (t, J = 9.4 Hz,

1H). The β-D-glucopyranoside **10a** was found to readily undergo autoxidation to give a peroxide: FAB positive ion, *m/z* 739 ([M + H + O_2]$^+$, 10), 707 (<1), 539 (2), 499 (26), 415 (3), 345 (46), 313 (3), 223 (43), 211 (64), 191 (100), 185 (85), 150 (100), 133 (60), 115 (20); ν_{max} 3400, 1730, 1390, 1275, 1140 cm^{-1}.

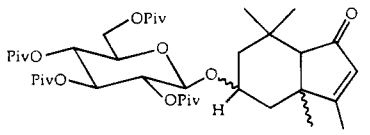
(3′*R,9′*S**)-3′-Hydroxy-5′-megastigmen-7′-yn-9′-yl β-D-Glucopyranoside (10b).** Depivaloylation at room temperature according to the method of Zemplén and Kunz (1923) yielded, after 7 days, **10b** as a gum in 88% yield as a mixture of two diastereoisomers: ^{13}C NMR (75.47 MHz, D_2O, *tert*-butyl alcohol as external standard) δ 23.5, 23.6, 23.8, 29.7, 31.6, 37.8, 41.8, 46.9, 62.6, 62.7, 66.2, 66.7, 69.3, 71.3, 71.7, 74.9, 75.3, 77.7, 77.9, 78.1, 86.5, 94.5, 101.5, 103.2, 124.2, 142.4; FAB, *m/z* 371 (10), 209 (42), 191 (100), 173 (25), 149 (20), 135 (35), 125 (30), 109 (18), 85 (10), 81 (7).

(3′*R,9′*S**)-3′-Acetoxy-5′-megastigmen-7′-yn-9′-yl 2,3,4,6-Tetra-*O*-acetyl-β-D-glucopyranoside (10c).** Acetylation of the glucoside (**10b**) with an excess of acetic anhydride in pyridine yielded the acetylated β-D-glucopyranoside **10c** as a gum: found M$^+$ 598.2851, $C_{29}H_{40}O_{12}NH_4^+$ requires 598.2863; ν_{max} 2900, 2840, 2250, 1750, 1600, 1360, 1250, 1230 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.12, 1.14, 1.15, 1.16 (s, C-1′−Me), 1.46, 1.49 [each d, *J* = 6.6 Hz, (H-10′)$_3$]; 1.52−1.58 (m, 1H), 1.86 (br s, C-5′−Me), 1.95, 1.99, 2.00, 2.01, 2.02, 2.03, 2.04, 2.07 (s, CH_3CO), 2.45 (m, 1H), 4.72, 4.77 (each q, *J* = 6.6 Hz, H-9′), 4.84, 4.87 (each d, *J* = 7.8 Hz, H-1), 5.00 (m, H-3′ and H-2); (column B) diastereoisomer 1 *m/z* 520 (<1), 460 (<1), 358 (<1), 347 (<1), 331 (<1), 188 (17), 175 (30), 173 (36), 172 (20), 159 (100), 157 (42), 131 (18), 109 (17), 91 (8); (column B) diastereoisomer 2 *m/z* 520 (<1), 460 (<1), 358 (<1), 347 (<1), 331 (<1), 188 (8), 175 (25), 173 (42), 172 (38), 159 (100), 157 (50), 131 (20), 109 (17), 91 (10).

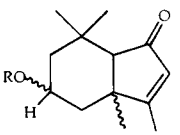
(*E*)-7′-Oxo-5′,8′-megastigmadien-3′-yl 2,3,4,6-Tetra-*O*-pivaloyl-β-D-glucopyranoside (11a). The synthesis of the β-D-glucopyranoside **11a** according to the method of Kunz and Pfrengle (1986) yielded several products. After 15 min at room temperature two β-D-glucopyranosides were obtained, **11a** and **12**, together with Nazarov cyclization product (**13a**) and



(**11a**) R = Piv
(**11b**) R = H
(**11c**) R = Ac



(**12**)



(**13a**) R = H
(**13b**) R = Ac

starting material. After 50 min, only a trace of starting material was recovered, but the β-D-glucopyranoside **12** and its aglycon **13a** were both obtained as the major products. Compounds were isolated using the chromatotron (Furniss et al., 1989) and a solvent gradient of petroleum ether/diethyl ether. Aglycon (**13a**): mp 85−90 °C [lit. (Ohloff et al., 1973) 109−110 °C for *cis* and 95−96 °C for *trans*]; ^{1}H NMR (60 MHz, $CDCl_3$) δ 0.8, 1.01, 1.2, 1.3 (each s, CH_3), 1.4−1.9 (m, 4H), 2.1 (s, vinyl-Me), 2.8 (br s, 1H), 3.8−4.4 (m, 1H), 5.8 (br, 1H); (column A) diastereoisomer 1, *m/z* 208 (21), 193 (14), 176 (7), 175 (67), 147 (7), 123 (41), 111 (31), 110 (100), 109 (55), 108 (17), 105 (12), 95 (34), 91 (18), 81 (24), 79 (24), 77 (13), 67 (9); diastereoisomer 2, *m/z* 208 (16), 193 (10), 175 (42), 147 (8), 123 (33), 111 (24), 110 (100), 109 (50), 108 (18), 105 (12), 95 (26), 91 (19), 82 (18), 81 (39), 79 (22), 77 (13), 67 (10); ν_{max} 3350, 1675, 1620 cm^{-1}. β-D-Glucopyranoside (**11a**), a crystalline solid, as a mixture of two diastereoisomers: ν_{max} 2960, 1740, 1680, 1620, 1480, 1395, 1365, 1280, 1140 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.95, 0.97, 1.12, 1.13, 1.16, 1.17, 1.22 (s, CH_3), 1.54−1.85 (m, 2H), 1.91 [d, *J* = 6.7 Hz, (H-10′)$_3$], 1.85−2.35 (m, 2H), 3.96−4.04 (m, H-3′ and H-6$_a$), 4.66, 4.70 (each d, *J* = 7.5 Hz, H-1 of both diastereoisomers), 6.14 (d, *J* 16.2 Hz, H-8′), 6.70 (dq, *J* = 16.2, 6.7 Hz, H-9′); solid probe *m/z* 706 (<1), 550 (1), 500 (6), 499 (20), 210 (33), 192 (11), 191 (53), 190 (39), 149 (13), 137 (6), 134 (7), 133 (8), 126 (8), 122 (7), 121 (40), 120 (13), 119 (9), 118 (22), 108 (13), 107 (9), 105 (10), 104 (7), 103 (11), 98 (7), 97 (11), 95 (6), 91 (11), 86 (10), 85 (100), 83 (10), 82 (6), 81 (16), 79 (6), 77 (15), 71 (7), 70 (12), 69 (75), 59 (6), 58 (22). β-D-Glucopyranoside **12**: mp 135−138 °C; found M$^{+\cdot}$ 706.4286, $C_{39}H_{62}O_{11}$ requires 706.4292; ^{1}H NMR (300 MHz, $CDCl_3$) δ (mixture of several diastereoisomers) <2.1 (complex), 4.1−4.2 (m, H-3′), 4.61, 4.62 (each d, *J* = 8.0 Hz, H-1), 5.76 (s, vinyl-H).

(*E*)-7′-Oxo-5′,8′-megastigmadien-3′-yl 2,3,4,6-Tetra-*O*-acetyl-β-D-glucopyranoside (11c). (*a*) Attempts to prepare this compound according to the method of Schmidt and Michel (1980) were unsatisfactory and gave only 6% of the product (**11c**). 3-Hydroxy-β-damascone (**2**) (130 mg, 0.63 mmol) in dichloromethane (1 mL) was added slowly over 1 h to a solution of imidate (370 mg, 0.75 mmol) and boron trifluoride etherate (79 μL) in dichloromethane (2 mL) at −10 °C. After addition, the reaction mixture was washed with a saturated solution of sodium hydrogen carbonate (2 × 20 mL), water (20 mL), and saturated aqueous sodium chloride solution (10 mL) and dried ($MgSO_4$). After solvent removal, chromatography (diethyl ether) gave the acetate of 3-hydroxy-β-damascone (**2**) and the cyclized compound (**13b**) together with some α- and β-glycosides (**11c**) (6%). When the order of addition was reversed, the result was similar.

(*b*) The procedure of Ackermann et al. (1989) gave, after chromatography and starting with 3-hydroxy-β-damascone (**2**) (600 mg), the β-glycoside (**11c**, 15%), a crystalline solid, as a mixture of diastereoisomers: found M$^{+\cdot}$ 538.2403, $C_{27}H_{38}O_{11}$ requires 538.2414; ν_{max} 2950, 2930, 2850, 1750, 1640, 1430, 1360, 1220, 1040 cm^{-1}; ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.95, 1.13 (s, C-1′−Me), 1.35−1.48 (m, 2H), 1.51 (s, C-5′−Me), 1.90 [dd, *J* = 6.9, 1.5 Hz, (H-10′)$_3$], 1.97, 1.99, 2.01, 2.02, 2.04, 2.05 (each s, CH_3CO), 2.3 (m, 2H), 3.9−4.05 (m, H-3′), 4.60, 4.62 (each d, *J* = 7.9 Hz, H-1 of two diastereoisomers), 6.12 (dq, *J* = 15.7, 1.5 Hz, H-8′), 6.69 (dq, *J* = 15.7, 6.9 Hz, H-9′); ^{13}C NMR (75.47 MHz, $CDCl_3$) δ 18.3, 20.5, 20.9, 28.9, 29.5, 35.8, 36.0, 37.5, 38.8, 44.2, 45.2, 61.4, 62.1, 62.3, 68.6, 71.5, 71.8, 72.8, 73.3, 73.4, 99.7, 127.1, 128.1, 134.3, 139.6, 140.5, 146.1, 169.1, 169.3, 170.2, 170.5, 201.1; (column A) diastereoisomer 1, *m/z* 538 (<1), 331 (1), 190 (42), 175 (15), 121 (98), 107 (39), 69 (100); diastereoisomer 2, *m/z* 538 (<1), 331 (1), 190 (38), 175 (13), 169 (18), 121 (82), 107 (50), 69 (100). Some diastereoisomeric α-glucoside was obtained as a mixture with 2,3,4,6-tetra-*O*-acetyl-β-D-glucose: ^{1}H NMR (300 MHz, $CDCl_3$) δ 0.93, 1.10 (s, C-1′−Me), 1.89 (dd, *J* = 6.9, 1.6 Hz, C-5′−Me), 3.90−4.05 (m, H-3′), 5.30, 5.32 (each d, *J* = 3.5 Hz, H-1 of two diastereoisomers), 6.10 (dq, *J* = 15.7, 1.6 Hz, H-8′), 6.69 (dq, *J* = 15.7, 6.9 Hz, H-9′); (column A) diastereoisomer 1, *m/z* 538 (<1), 497 (<1), 331 (1), 190 (36), 149 (18), 121 (100), 110 (38), 69 (78); diastereoisomer 2, *m/z* 538 (<1), 497 (<1), 331 (2), 190 (38), 149 (18), 121 (100), 110 (50), 69 (100). A further component, presumed to be **13b** on the basis of retention time and mass spectral data, was isolated as a mixture of diastereoisomers: (column A) Diastereoisomer 1, *m/z* 235 (1), 190 (12), 175 (62), 161 (10), 149 (10), 147 (18), 135 (8), 123 (12), 110 (100), 109 (33), 81 (75), 55 (8); diastereoisomer 2, *m/z* 235 (1), 190 (12), 175 (48), 161 (8), 147 (19), 133 (5), 122 (16), 110 (100), 109 (32), 82 (15), 81 (78), 55 (5).

(*E*)-7′-Oxo-5′,8′-megastigmadien-3′-yl β-D-Glucopyranoside (11b). Sodium hydroxide (0.1 M, 2.6 mL, 0.26 mmol) was added to the glycoside **11c** (37.8 mg, 0.07 mmol) in tetrahydrofuran (6 mL) at 25 °C. After 2 h, the solvent was removed under vacuum, and standard workup, as described for isolation of the β-glucosides, yielded the β-glucoside **11b** (40%) as a crystalline solid: ^{13}C NMR (75.47 MHz, D_2O, *tert*-butyl alcohol as external standard) δ 18.4, 21.0, 29.2, 29.7, 36.0, 37.6, 39.2, 44.3, 45.5, 61.7, 69.8, 72.5, 73.5, 73.3, 75.6, 76.3, 101.2, 101.6, 127.4, 130.5, 134.3, 140.4, 142.8, 146.4, 201.3; FAB positive ion, *m/z* 371 ([M + H]$^+$, 4), 222 (4), 209 (100), 191 (57), 173

(16), 149 (7), 135 (10), 125 (7), 107 (5); negative ion, m/z 369 ($[M - H]^-$, 100), 324 (6), 221 (10), 179 (25), 161 (15), 99 (9), 89 (10), 33 (5). Attempted deacetylation using the method of Fiandor et al. (1985), which uses ammonia, led to conjugate addition.

RESULTS AND DISCUSSION

In the course of this work, four methods of β-D-glycosylation were considered. Three, the methods of Kunz and Pfrengle (1986), Schmidt and Michel (1980), and Ogawa et al. (1981), are homogeneous glycosylation reactions and have been reported to proceed with high stereoselectivity. Homogeneous glycosylations are known to give high yields but, because they employ Lewis acids, may not be suitable for acid-sensitive substrates.

The method of Ackermann et al. (1989), a heterogeneous reaction utilizing a silver salt under relatively neutral conditions, was investigated here for glycosylation of the acid-sensitive aglycon 3-hydroxydamascone (**2**).

In this study, glucosides were prepared from 3-hydroxy-β-damascone (**2**), enyne diol (**3**), and the related models 4-(1-cyclohexenyl)-3-butyn-2-ol (**4**) and 3,5,5-trimethyl-3-cyclohexen-1-ol (**5**). Two of the glycosides **10b** (with undefined stereochemistry) and **11b** were previously synthesized by Anderson et al. (1978). The latter was formed in 13% yield, and only ^{13}C NMR spectral data were given. No yields or spectral data for the glycoside **10b** were reported.

The synthesis of the glycoside **8a** was carried out using the method of Kunz and Pfrengle (1986) which employs the orthoester (**7**). It was envisaged that the bulky pivaloyl protecting group would reduce the problems of both transesterification and α-glycosidation encountered during preliminary experiments on 9-hydroxy-5-megastigmen-4-one (**6a**) using the method of Ogawa et al. (1981). In that case a substantial amount of 4-oxo-5-megastigmen-9-yl acetate (**6b**) was obtained with an approximately 2:1 mixture of β- and α- glycosides (**6c** and **6d**). The orthoester (**7**), required for the method of Kunz and Pfrengle (1986), was made from 2,3,4,6-tetra-*O*-pivaloyl-α-D-glucopyranosyl bromide (Kunz and Harreus, 1982) with silver triflate. In addition to the reported product (**7**), about 8% of an isomer was isolated by chromatography. Its spectral data were consistent with it being the *Z* isomer of **7**. In particular it gave the same molecular ion at m/z 633 and the anomeric proton had similar chemical shift and coupling constant (δ 5.9, J = 6.0 Hz; *cf.* δ 6.2, J = 6.0 Hz for **7**].

The β-glycoside **8a** was obtained in 90% yield as a mixture of diastereoisomers. Recrystallization from methanol gave one diastereoisomer which gave a molecular ion at 638. The anomeric proton resonated as a doublet of 8.1 Hz at δ 4.66. The corresponding proton for the other diastereoisomer also appeared as a doublet (J = 8.0 Hz) at δ 4.69. Methanolysis of (**8a**) with sodium methoxide in methanol, according to the procedure of Zemplén and Kunz (1923), gave the glucoside **8b** in high yield. This was characterized as its acetate (**8c**).

4-(1′-Cyclohexenyl)-3-butyn-2-ol (**4**), using the method of Kunz and Pfrengle (1986), yielded the glucopyranoside tetrapivaloate (**9a**). The reaction was done under dry oxygen-free nitrogen at −20 °C with 1.5 equiv of orthoester (**7**). The β-glycoside tetrapivaloate (**9a**) was isolated by chromatography in 60% yield (2,6-di-*tert*-butyl-*p*-cresol was added to avoid autoxidation), but the two diastereoisomers were only partially separated by HPLC. In the ^1H NMR spectrum of the high R_f diastereoisomer, the anomeric proton appeared at δ 4.8 (J = 8.3 Hz), compared to δ 4.7 (J = 8.1 Hz) for the lower R_f diastereoisomer. The molecular ion (m/z 648) was obtained with FAB mass spectrometry.

Treatment of the β-glycoside **9a** (both diastereoisomers) with sodium methoxide in methanol yielded 3-(1′-cyclohexenyl)-1-methyl-2-propynyl β-D-glucopyranoside (**9b**) in 41% yield. Neither electron impact nor chemical ionization gave the molecular ion. The glucoside **9b** was acetylated and characterized as its tetraacetate (**9c**).

The preparation of the glucoside of the enyne diol (**3**) (Loeber et al., 1971) was first attempted via the Koenigs–Knorr method because the product (**10c**) could be analyzed by gas chromatography (in the pivaloate series the glycosides could not be analyzed by GLC). Such analysis was required for comparison with the mixture of glycosides obtained from grape juice by C_{18} reversed phase chromatography to determine if the glucoside was present as a natural product. However, the Koenigs–Knorr approach was unsuccessful, giving mixtures from which the glycosides could not be isolated.

Use of the method of Kunz and Pfrengle (1986), with 1.2 equiv of the orthoester (**7**), was successful and gave the β-glycoside **10a** in 54% yield. It was considered that glycosidation would occur at the less hindered C-9 hydroxyl group, and the ^1H NMR spectrum supported this assumption. The anomeric protons of the two diastereoisomers of **10a** had chemical shifts similar to those of the model enyne derivative **9a** and appeared at δ 4.89 and 4.93 (appearing as a triplet) with J approximately 8.2 Hz for each of the diastereoisomers.

To establish which of the two hydroxyl groups of **3** had been glycosylated, the product was depivaloated (sodium methoxide/methanol, 88% yield) and acetylated to obtain further ^1H NMR data. The ^1H NMR spectrum of **10c** showed 10 acetate groups (the signals for 2 of these were superimposed on others, giving 8 singlets in total), consistent with a mixture of two diastereoisomeric β-glycosides. The signals for the anomeric protons at δ 4.84 and 4.87 appeared as a triplet, and an obscured multiplet for H3′ was observed at δ 5.0. The latter was shown by a decoupling experiment to be coupled to multiplets at δ 1.55 and 2.45, assigned to two of the alicyclic protons H2′ and H4′. This result confirms that the downfield signal at δ 5.0 is due to H3′ in the pentaacetate. In contrast, the signals for H9′ in **10c** had essentially the same chemical shift as the corresponding signals for the glycoside **10a.** These data showed that glycosylation occurred at the C9′ hydroxyl group.

Glycosylation of 3-hydroxy-β-damascone (**2**) with the orthoester (**7**) (1.2 equiv) and boron trifluoride etherate for 15 min gave only 10% of the required products (**11a**). In addition, the β-glycoside **12**, the cyclized product **13a**, and starting material were recovered. The two β-glycosides, **11a** and **12**, were each mixtures of diastereoisomers which were obtained as solids after chromatography. Longer reaction times gave more of the β-glycoside **12** and less of **11a**. The ^1H NMR spectrum of **11a** showed the anomeric protons at δ 4.66 and 4.70 (appearing as an apparent triplet) with J approximately 7.5 Hz. The spectrum was generally complex, but many of the protons could be assigned from the COSY spectrum. The anomeric protons of the bicyclic compound **12a** appeared at δ 4.62 and 4.61 with couplings of 8.0 Hz. The bicyclic compounds **12** and **13** are Nazarov cyclization products (Ohloff et al., 1973; Shiloff and Hunter, 1979).

In an attempt to prepare the glucoside **11b**, compound **11a** was treated with sodium methoxide in methanol. However, the product so obtained was consistent with having arisen by conjugate addition of methanol to the side chain double bond, a reaction which is also favored with the aglycon **2**. Synthesis of the acetylated analogue **11c** was therefore attempted. It was anticipated that deacetylation of this substrate could be achieved without competing conjugate addition to the side chain.

The method of Schmidt and Michel (1980), using 2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyltrichloroacetimidate as the glycosidation agent, and with 1 equiv of boron trifluoride to minimize Nazarov cyclization, proceeded only in low yield (6%) with 3-hydroxy-β-damascone (**2**). In addition, the α-glycoside and the acetate of **2** were isolated. Under similar conditions, it had been found that 3,5,5-trimethyl-3-cyclohexen-1-ol gave 21% of the β-glycoside **8c**. Milder conditions (0.2 equiv of boron trifluoride etherate) gave only trace amounts of the desired glycosides (**11c**). However, the modification by Ackermann et al. (1989) of the Koenigs–Knorr reaction, using the soluble silver trifluoroacetate, was found to give the β-glycoside **11c** in better yield (15%) with only a trace of the α-glycoside. The anomeric protons of the diastereoisomeric β-glycosides **11c** were observed at δ 4.62 and 4.60, each as a doublet with J = 7.9 Hz. The anomeric protons of the α-glycoside, also a mixture of two diastereoisomers, appeared at δ 5.38 and 5.40, each with J = 3.5 Hz.

By using the β-glycoside **11c** as reference, GC/MS analysis confirmed the presence of **11c** in acetylated glycosidic fractions isolated from Riesling wine. The glycoside **10c** has not so far been similarly identified.

Deacetylation of **11c** with sodium methoxide in methanol or with ammonia was unsatisfactory due to conjugate addition to the 8,9 double bond. However, satisfactory hydrolysis with less than 5% of conjugate addition was achieved with sodium hydroxide in aqueous tetrahydrofuran. The mass and ^{13}C NMR spectra confirmed the structure of the glucoside **11b**.

CONCLUSION

This work shows the application of modern methods of glycosylation, using homogeneous reaction systems, to several norterpenoid and model compounds. It is evident that the synthetic methods described here will find use in the preparation of a wide range of terpenoid glycosides and thus greatly assist research on fruit flavor precursors.

The results obtained with 3-hydroxy-β-damascone (**2**) demonstrate that, in spite of the mild reaction conditions employed in the glycosylations and subsequent deprotection steps, such bifunctional molecules can still present difficulties. Nevertheless, synthetic strategies to circumvent these difficulties are available.

For aglycons stable to Lewis acids in organic solvents, the method of Kunz and Pfrengle (in a homogeneous reaction system) was preferred, giving β-glycosylation with high stereospecificity. Although the preparation of glucose pentaacetate, used in the method of Ogawa et al. (1981), was facile in comparison to the preparation of reagents used in other methods, glycosylation of the aglycon **6a** with this reagent gave poor stereoselectivity and competing transesterification. It was not utilized further. The method of Schmidt and Michel (1980) gave both α- and β-anomers and, although employing a milder Lewis acid medium than the method of Kunz and Pfrengle (1986), was not suitable for the acid sensitive aglycon **2**.

Where an aglycon is sensitive to Lewis acids, then the Koenigs–Knorr reaction as modified by Ackermann et al. (utilizing a heterogeneous reaction system) is the method of choice. However, some competing transacetylation and α-glycosylation can be expected.

Although the glycosides prepared here were generally mixtures of diastereoisomers, with complex spectra expected for such mixtures, the products were suitable for hydrolytic studies reported elsewhere.

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