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Preliminary communication

Stereocontrolled, total synthesis of α-D-GalA-[(1→4)-α-D-GalA]$_8$-(1→4)-β-D-GalA-1→OPr, a synthetic model for phytoalexin elicitor-active oligogalacturonic acids*

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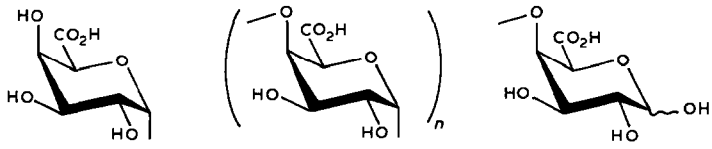
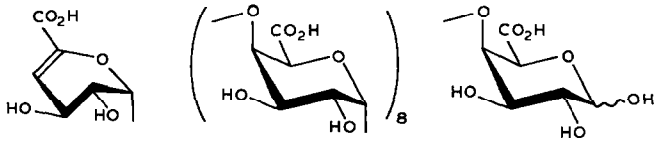
Phytoalexin-elicitor activity was reported for the oligogalactosiduronic acids **1** (n = 7–13) obtained either by partial hydrolysis of soybean cell-walls and citrus pectin[2] with acid, or by digestion of polygalacturonic acid with endopolygalacturonanase of *Rhizopus stolonifer*[3], and also for α-(1→4)-linked decagalactosiduronic acid **2** carrying an unsaturated galacturonic acid residue at the nonreducing end (obtained from soybean cell-walls by treatment with an endo-α-1,4-polygalacturonic acid lyase of the bacterium *Erwinia carotovora*[4]. Oligogalactosiduronic acids of undetermined chain-length were also reported to function as messenger



1

n = 7–13



2

*Part 3 in the series "Synthetic Studies on Plant Cell-Wall Glycans". For Part 2, see ref. 1.
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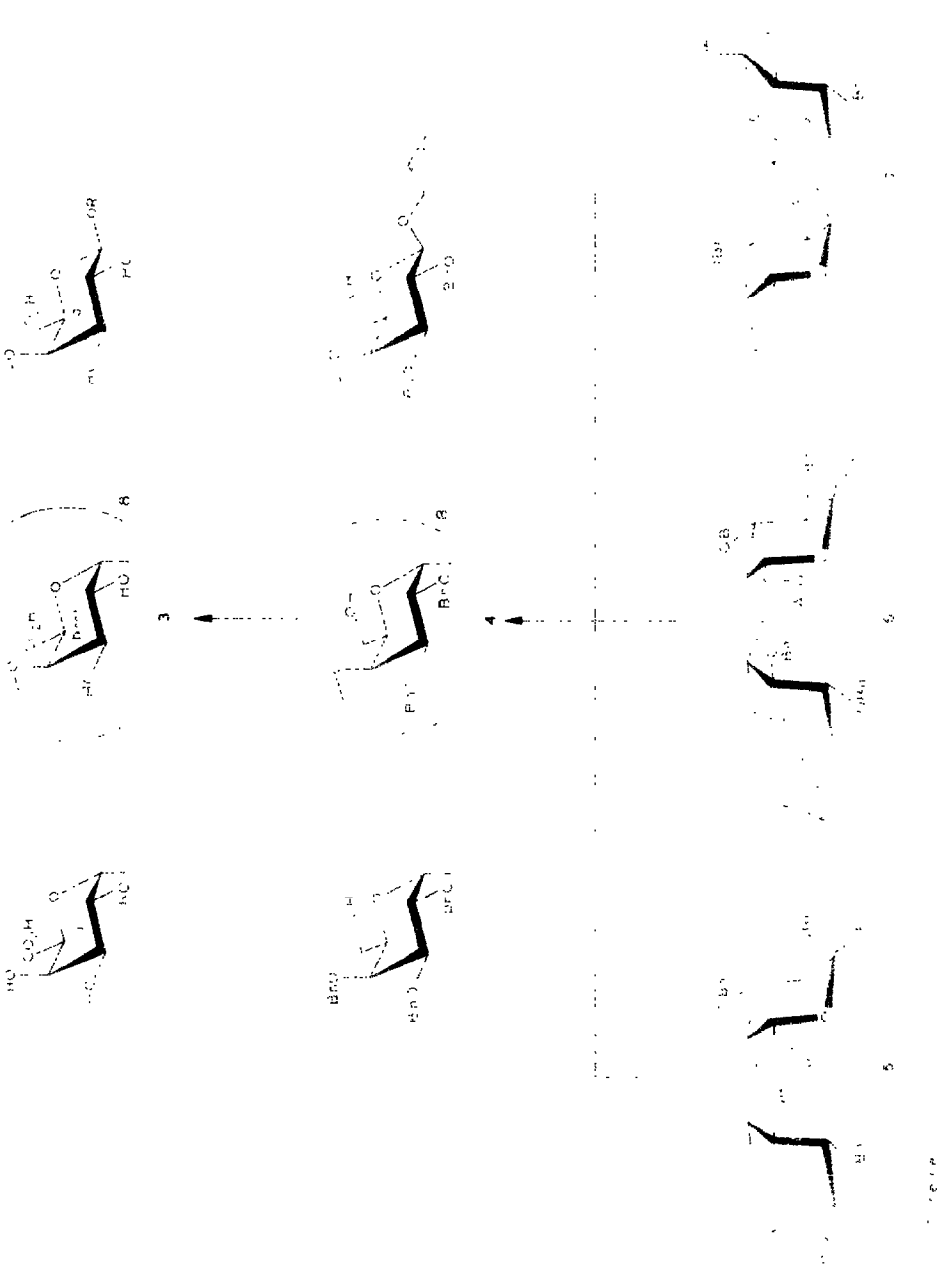


molecules that induce synthesis both of proteinase inhibitor I in tomato leaves[5], and ethylene and hydroxyproline-rich glycoprotein in melons[6] in association with their defense against invading microbes.

As part of a project on the synthesis of plant cell-wall glycans of biological interest[7] we now describe a total synthesis of decagalactosiduronic acid **3** which

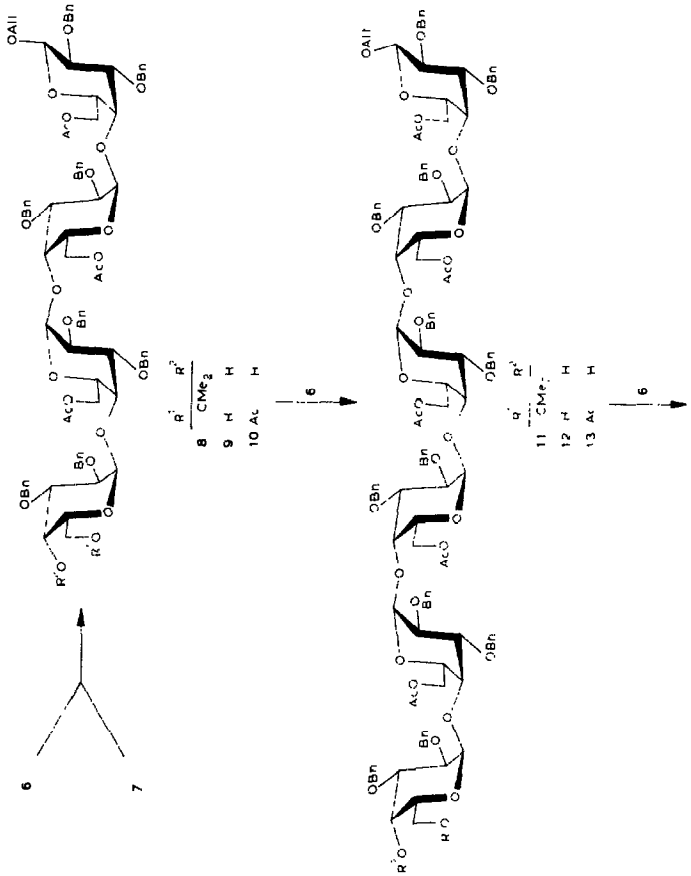
was designed to be functional as a stable model compound in examination of the minimum structural requirements for the maximum elictor activity for both **1** and **2**.

The retrosynthetic analysis of target **3**, based on our previous experiments[1,8], led us to design the suitably protected, α-(1→4)-linked decagalactopyranoside **4** as a key intermediate which, in turn, might be reconstructed by using a stereoselective glycosylation strategy. Thus, three already known galactobiosyl synthons[8], namely, **5, 6**, and **7**, were employed that correspond to (*a*) a nonreducing terminal galactobiosyl unit, (*b*) an oligogalactoside located in the middle of the glycan, and (*c*) a reducing-end galactobiosyl unit, respectively.

The stannous chloride–silver perchlorate-promoted glycosylation[9] of the glycosyl acceptor **7** with the glycosyl donor **6** in diethyl ether proceeded in 89% yield with exclusive formation of the α-(1→4)-linked galactotetraose **8**; δ_H (90 MHz): 1.32 (s, 3 H, CCH_3), 1.41 (s, 3 H, CCH_3), 1.92 (s, 3 H), 1.97 (s, 3 H), and 2.02 (s, 3 H) for three acetyl methyl signals; δ_C (22.5 MHz) 99.0, 99.6, and 100.2 for C-1b,c,d, and 102.8 for C-1a. Solvolysis of compound **8** in 4:1 AcOH–H_2O for 30 min at 60° gave a 94% yield of the diol **9**, $[\alpha]_D$ +46.1° (*c* 0.6); δ_H (400 MHz) 1.919, 1.929, and 2.040 for three acetyl methyl signals; δ_C (22.5 MHz) 99.0 ($^1J_{CH}$ 169 Hz), 99.6 ($^1J_{CH}$ 166 Hz), 99.7 ($^1J_{CH}$ 169 Hz) for C-1bcd, and 102.8 ($^1J_{CH}$ 160 Hz) for C-1a. Monoacetylation of the diol **9** with acetyl chloride in pyridine for 1.5 h at −5° gave an 87% yield of tetraacetate **10**, $[\alpha]_D$ +39.1° (*c* 1.1); δ_H (400 MHz) 1.907, 1.915, 1.930, and 2.040 for four acetyl methyl singlets. Glycosylation[9] of the galactotetraosyl acceptor **10** with 1.4 equiv. of the galactobiosyl donor **6** afforded exclusively an 88% yield of galactohexaosyl derivative **11**, which was converted, as already described, in two steps, *via* **12**, $[\alpha]_D$ +51.7° (*c* 0.9); δ_C (22.5 MHz) 99.1 ($^1J_{CH}$ 169 Hz) and 99.6 ($^1J_{CH}$ 171 Hz) for C-1b,c,d,e,f in the ratio of 3:2, and 102.9 ($^1J_{CH}$ 156 Hz) for C-1a, into galactohexaosyl acceptor **13** in 74% overall yield, $[\alpha]_D$ +45.1° (*c* 0.7); δ_H (400 MHz), 1.880, 1.897, 1.915, 1.926, 1.941, and 2.028, for six acetyl methyl singlets.

The third glycosylation, using two equiv. of donor **6**, of the glycosyl acceptor **13** in dichloroethane afforded the galacto-octaosyl derivative **14**, which was hydrolyzed in 4:1 AcOH–H_2O to give, in 48% overall yield, the diol **15**; $[\alpha]_D$ +50.0° (*c* 0.7); δ_H (400 MHz); 1.885 ($COCH_3$), 1.914 (4 $COCH_3$), 1.932 ($COCH_3$), 2.029 ($COCH_3$); δ_C (22.5 MHz): 99.2, 99.5 and 99.7 (4:1:2), for C-1b,c,d,e,f,g,h with $^1J_{CH}$ 169 Hz, and 103.0 ($^1J_{CH}$ 159 Hz) for C-1a. Monoacetylation of compound **15** afforded an 87% yield of octaacetate **16**, $[\alpha]_D$ +41.7° (*c* 0.6); δ_H (400 MHz): 1.875 (s, $COCH_3$), 1.896 (s, $COCH_3$), 1.913 (s, 4 $COCH_3$), 1.933 (s, $COCH_3$), and 2.029 (s, $COCH_3$).

Finally, glycosylation of the galacto-octaosyl acceptor **16** with two equiv. of glycosyl donor **5** in dichloroethane afforded a 76% yield of the desired galactodecaosyl derivative **17**; $[\alpha]_D$ +51.2° (*c* 0.4); δ_H (400 MHz) 1.753 (s, $COCH_3$), 1.859 (s, $COCH_3$), 1.902 (s, 3 $COCH_3$), 1.908 (s, $COCH_3$), 1.913 (s, $COCH_3$), 1.921 (s, $COCH_3$), 1.932 (s, $COCH_3$), and 2.029 (s, $COCH_3$). Deacetylation of compound **17** with 0.1M NaOMe in 15:2 MeOH–THF afforded an 84% yield of a key intermediate, decaol **4**, $[\alpha]_D$ +69.8° (*c* 0.3).


6
7
8 CMe2
9 H H
10 Ac H
6
11 CMe2
12 H H
13 Ac H
6

R¹ R²
14 CMe₂
15 H H
16 Ac H
5
17 R = CH₂OAc
4 R = CH₂OH
18 R = CHO
19 R = COOH
20 R = CO₂Me

Scheme 2

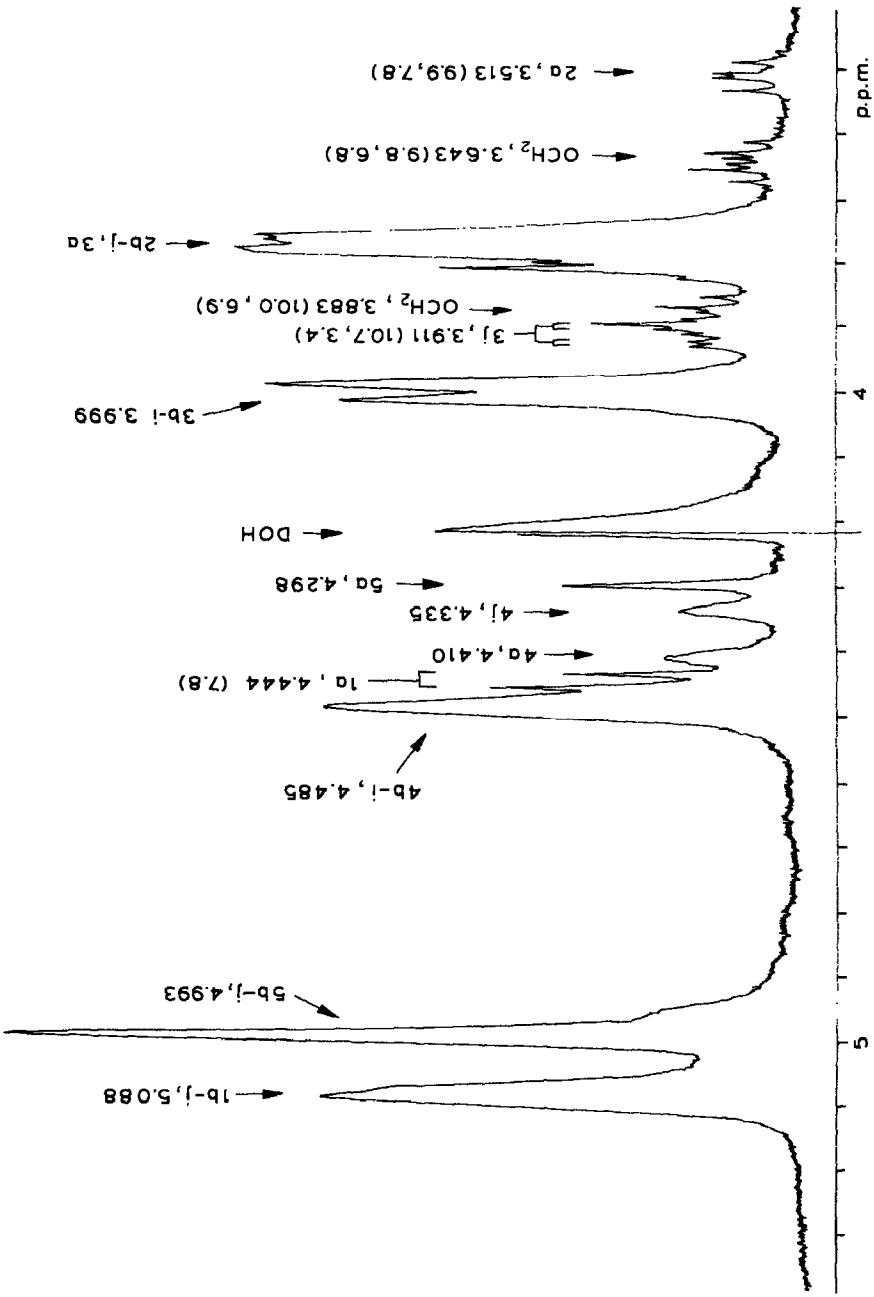


Fig. 1. 400-MHz ^{1}H-n.m.r. spectrum of compound **3** (R = propyl) recorded for a solution in D_2O at 80°. The values of δ_H are expressed in p.p.m. downward from Me_4Si by reference to an internal standard of *tert*-BuOH (1.230). The values in parentheses are $^3J_{HH}$ values in Hz.

Crucial oxidation of compound **4** into uronic acid **19** was examined in two steps. Swern oxidation[10] of **4** afforded decaaldehyde **18** which, without purification, was oxidized with a freshly prepared solution of $NaClO_2$ in water according to the procedure recently reported by Dalcanale and Montanari[11], to give the expected decacarboxylic acid **19** in 62% overall yield; $[\alpha]_D$ +134.4° (*c* 0.3).

Treatment of acid **19** with diazomethane afforded decacarboxylic acid methyl ester **20**, the ^{1}H-n.m.r. spectrum of which clearly revealed the signals for methyl protons as ten singlets at δ_H (400 MHz) 3.077, 3.178, 3.197, 3.199, 3.203, 3.219, 3.233, 3.327, 3.408, and 3.654, confirming that the crucial transformation of deca-alcohol **4** into decacarboxylic acid **19** had been successfully excecuted.

Catalytic hydrogenolysis of compound **19** in the presence of 10% Pd–C in aq. methanol afforded the target decagalactosiduronic acid **3** (R = propyl). The structure of compound **3** was assigned by the unambiguous synthetic sequence and was further confirmed by the ^{1}H-n.m.r. data (see Fig. 1).

In conclusion, the stable, α-(1→4)-linked, decagalactosiduronic acid **3** was synthesized in a regio- and stereo-controlled way by employing two galactobiosyl glycosyl donors, **5** and **6**, and a galactobiosyl glycosyl acceptor **7** as key synthons.

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