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Synthesis of an arabinogalactan-type octa- and two isomeric nonasaccharides. Suitable tuning of protecting groups

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Dedicated to Professor Frieder Lichtenthaler on the occasion of his 70th birthday

Abstract—An arabinogalactan-type double-branched octa- and two isomeric nonasaccharides were synthesized using the (2-naphthyl)methyl (NAP) and the acid sensitive but base stable (methoxydimethyl)methyl (MIP) protecting groups. The β-(1→6)-linked hexagalactan skeleton was synthesized having a benzyl and a (2-naphthyl)methyl (NAP) group at positions 2 of the second and the penultimate galactopyranosyl units, and this made possible sequential introduction of α-L-arabinofuranosyl or α-L-arabinofuranosyl-(1→5)-α-L-arabinofuranosyl side chains. © 2003 Elsevier Science Ltd. All rights reserved.

Monoclonal antibodies[1] are used very often for the structure elucidation of complex polysaccharides. Their production or the evaluation of their specificity require well-defined oligosaccharides which are the epitopes of the antigens.

Elucidation of arabinogalactans[2,3] isolated from cell-cultured *Echinacea purpurea* would become possible by using among others the synthetic oligosaccharides **1**–**3**. All of the three target oligosaccharides have the same hexasaccharide-type β-(1→6)-linked galactosyl skeleton in which OH-2 of the second as well as of the penultimate galactopyranosyl units are arabinofuranosylated (Scheme 1).

Our key reaction was the treatment of a free β-D-galactopyranosyl unit with 2,2-dimethoxypropane in the presence of an acid catalyst to obtain the 3,4-*O*-isopropylidene-6-*O*-(methoxydimethyl)methyl-β-D-galactopyranosyl building block.[4] The key disaccharide **4** was prepared starting either from β-D-galactopyranosyl-(1→6)-D-galactose[5] or from β-D-galactopyranosyl-(1→6)-1,2:3,4-di-*O*-isopropylidene-α-D-galactopyranose.[6]

The dioxolane-type isopropylidene protecting groups (PGs) behave as medium acid stable permanent PGs and the more acid labile (methoxydimethyl)methyl ether (MIP) as a temporary PG and compound **4** contains a free OH-2′ functionality. Unfortunately the MIP group is too labile for compound **4** to act as a glycosyl acceptor in a subsequent glycosylation reaction due to hydrolysis or its participation in condensation reactions.[7] For the preparation of the building blocks suitable for the hexagalactan skeleton, OH-2′ of compound **4** was either benzylated[8] to give compound **5**, or (2-naphthyl)methylated (NAP) to afford compound **6**.[9] The benzyl as well as the NAP-groups serve as acid and base stable orthogonal PGs,[10] both can be removed by hydrogenolysis[11] but the NAP group can be cleaved

ß-D-Gal*p*-(1→6)- ß-D-Gal*p*-(1→6)- ß-D-Gal*p*-(1→6)- ß-D-Gal*p*-(1→6)- ß-D-Gal*p*-(1→6)- D-Gal

2 2
↑ ↑
1 1
R' R"

1 R'=R"=α-L-Ara*f*-; **2** R'=α-L-Ara*f* R"= α-L-Ara*f*-(1→5)-α-L-Ara*f*-; **3** R'=α-L-Ara*f*-(1→5)-α-L-Ara*f*- R"= α-L-Ara*f*-

Scheme 1. Structure of the planned branched oligosaccharides (**1**, **2** and **3**).

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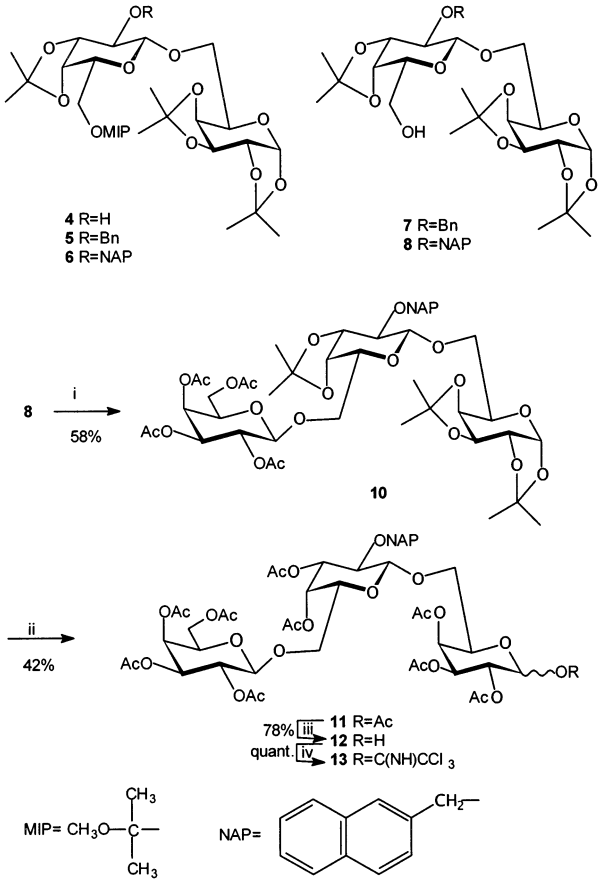
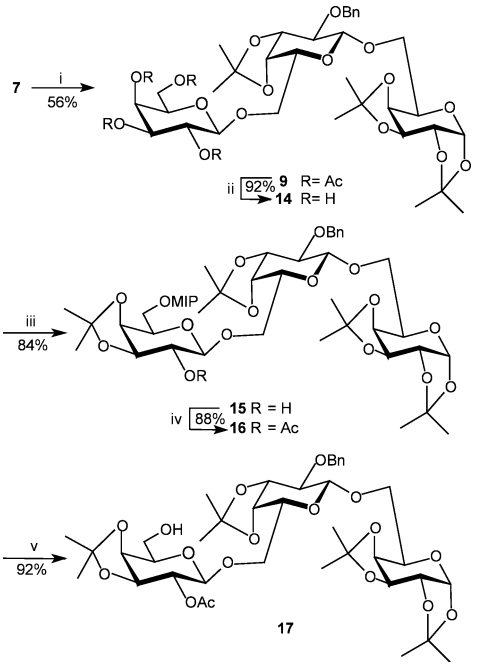
under oxidative[12] (DDQ or CAN) conditions as well. The sensitive MIP group was also removed from compounds **5** and **6** under very mild acid hydrolysis[8] to obtain OH-6′ containing disaccharide-type acceptors (**7** and **8**) (Scheme 2). These were separately glycosylated by tetra-*O*-acetyl-α-D-galactopyranosyl bromide to give two fully protected trisaccharides (**9** and **10**) (Scheme 3).

The trigalactopyranose-type glycosyl donor was prepared as follows: from compound **10** the three isopropylidene groups were removed with 90% aqueous trifluoroacetic acid and the free OH groups were acetylated to give compound **11**. The OAc-1 of **11** was removed by treatment with hydrazine acetate and the resulting **12** was transformed into trichloroacetimidate derivative (**13**) having a readily removable ONAP-2′ group.

To obtain the trisaccharide-type acceptor, compound **9** was saponified using $NaOCH_3$ to give compound **14**. Treatment of **14** with 2,2-dimethoxypropane in the presence of *p*-toluenesulfonic acid introduced isopropylidene and MIP groups into the terminal galactopyranosyl residue to obtain compound **15**. Acetylation resulted in the fully protected trisaccharide derivative (**16**) from which the MIP group was removed upon very mild acidic hydrolysis and the second building block **17** was isolated.



Scheme 2. Digalactosyl building blocks (**7** and **8**) and synthesis of the glycosyl donor trigalactoside (**13**). (i) tetra-*O*-acetyl-α-D-galactopyranosyl bromide, $Hg(CN)_2$, abs. CH_2Cl_2, rt; (ii) 90% aq. CF_3COOH, rt, 15 min; Ac_2O, pyr., 12 h; (iii) hydrazine acetate (1.3 equiv.), DMF, rt, 5 h; (iv) CCl_3CN, K_2CO_3, abs. CH_2Cl_2, 12 h.

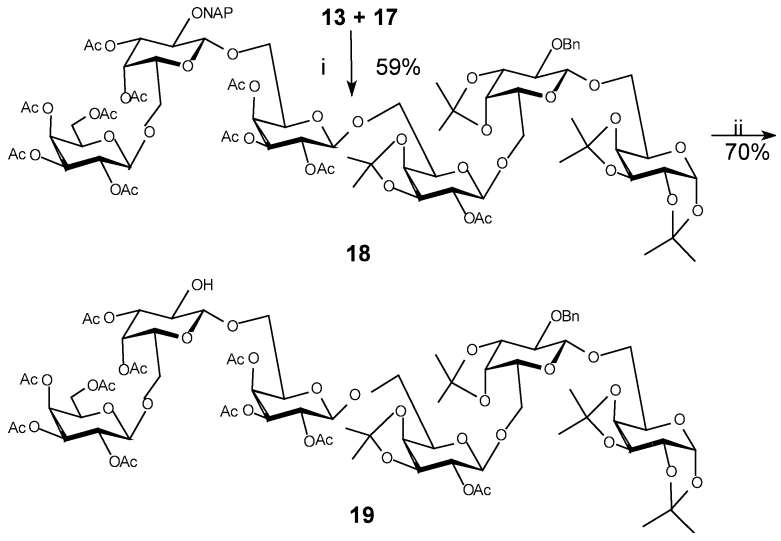


Scheme 3. Synthesis of the glycosyl acceptor trisaccharide. (i) tetra-*O*-acetyl-α-D-galactopyranosyl bromide, $Hg(CN)_2$, abs. CH_2Cl_2, rt, 12 h; (ii) Zemplén-saponification, 3 h; (iii) 2,2-dimethoxypropane, *p*TSA, rt, 4 h; (iv) Ac_2O, pyr., rt; (v) 96% CH_3COOH, H_2O, CH_2Cl_2, reflux, 6 h.

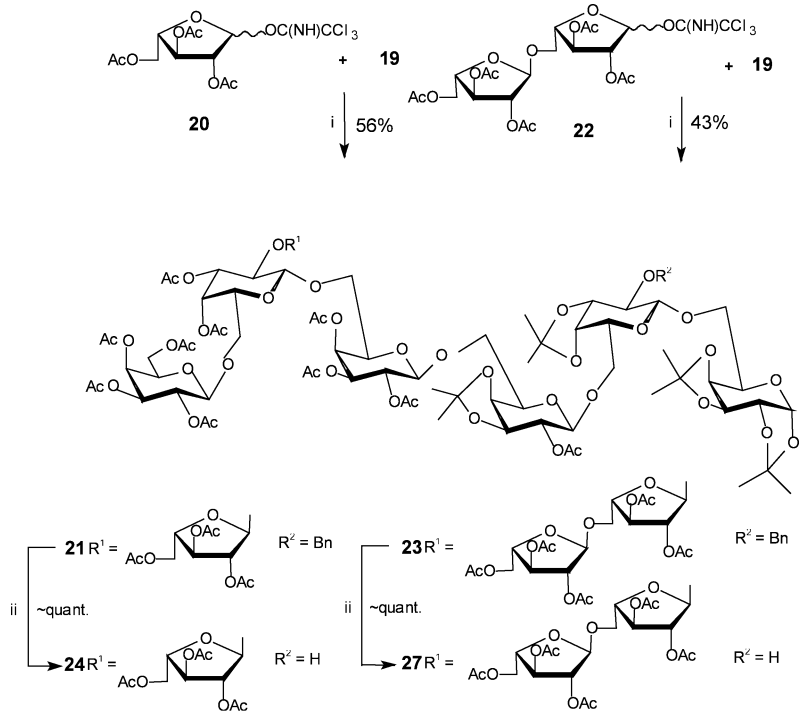
TMSOTf catalyzed coupling of the two trisaccharide-type building blocks (**13** and **17**) afforded the fully protected hexasaccharide (**18**) (Scheme 4) in an acceptable yield (59%). The presence of the two orthogonal protecting groups (NAP and Bn) at positions 2 of the second as well as of the penultimate galactopyranosyl residues makes it possible to use compound **18** for the synthesis of all three (**1**, **2** and **3**) oligosaccharides.

The orthogonal deprotection started with the removal of the NAP group of compound **18** using DDQ[12a] in dichloromethane–methanol (4:1) solvent system and compound **19** was isolated in a yield of 70%.

Compound **19** was glycosylated either with 2,3,5-tri-*O*-acetyl-α,β-L-arabinofuranosyl trichloroacetimidate[9] (**20**) to give the heptasaccharide (**21**), or with 2,3,5-tri-*O*-acetyl-α-L-arabinofuranosyl-(1→5)-2,3-di-*O*-acetyl-α,β-L-arabinofuranosyl trichloroacetimidate[9] (**22**) to afford the octasaccharide **23** (Scheme 5). Removal of the benzyl group of compound **21** by catalytic hydrogenolysis afforded compound **24** which was arabinosylated at position 2 of the second galactopyranosyl unit of the hexameric β-(1→6)-linked galactopyranosyl skeleton furnishing the fully protected octasaccharide **25**. On the



Scheme 4. Synthesis of the fully protected β-(1→6)-linked hexagalactose skeleton (**18**) and its transformation into the aglycon (**19**) ready for arabinosylation. (i) abs. CH_2Cl_2, TMSOTf (0.15 equiv.), –45°C, 2 h; (ii) CH_2Cl_2:MeOH = 4:1, DDQ (1.5 equiv.), rt, 5 h.



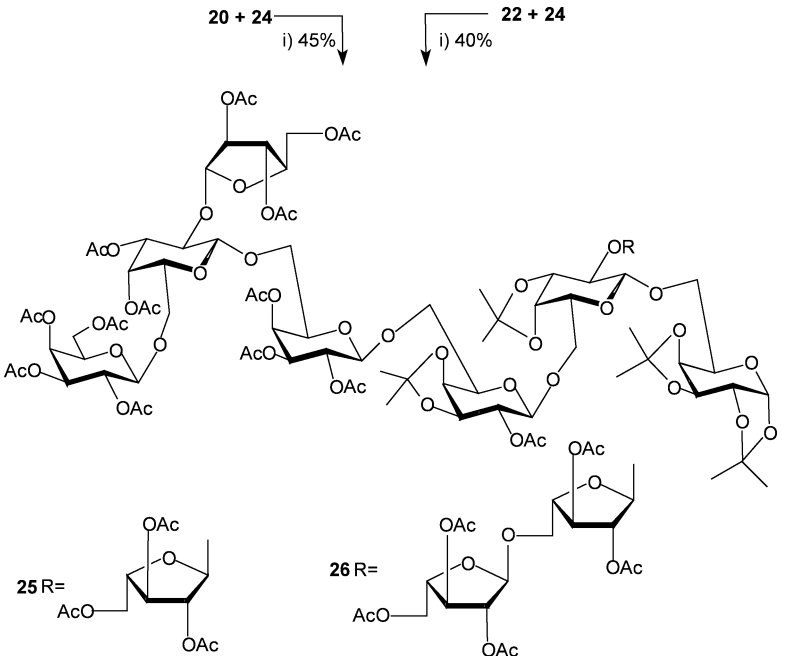
Scheme 5. Synthesis of the protected branched hepta- (**21**) and octasaccharides (**23**) and their hydrogenolysis to the monohydroxy compounds **24** and **27**. (i) abs. CH_2Cl_2, TMSOTf, –45°C; (ii) Pd/C, H_2, EtOAc, rt.

other hand glycosylation of compound **24** with glycosyl donor **22** yielded the nonasaccharide **26** (Scheme 6).

Catalytic hydrogenolysis of the benzyl group of octasaccharide **23** provided the hydroxyl group containing octasaccharide **27** which was treated with the donor **20** and the second nonasaccharide **28** was obtained.

The fully protected arabinogalactans (**25**, **26** and **28**) contained the same set of protecting groups, namely acetyls and dioxolane-type isopropylidene acetals.

Deprotection followed common procedures, involving Zemplén's deacetylation and consecutive mild acid hydrolysis of the isopropylidene groups with tri-

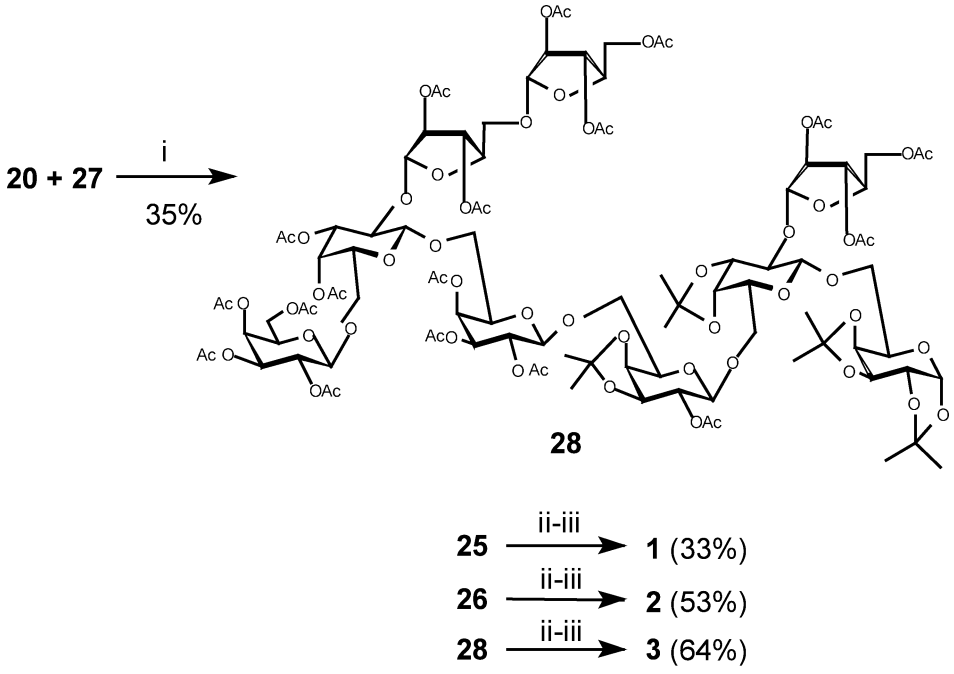


Scheme 6. Introduction of the arabinofuranosyl- and the diarabinofuranosyl units into position OH-2′. (i) abs. CH_2Cl_2, TMSOTf (1 equiv.), –45°C.

fluoroacetic acid resulting in the free octa-(**1**) and the two nonasaccharides (**2** and **3**) (Scheme 7).

The identity and stereochemical integrity of all compounds described here were confirmed by HPLC and by ^1H and ^{13}C NMR spectra. MALDI-TOF mass spectra[13] provided further support for the declared structures.

In summary we have demonstrated an efficient synthesis of a branched octa- and two nonasaccharides. Oligosaccharides remain challenging synthetic targets, the introduction and the use of the MIP acetal and its combination with NAP and benzyl ethers hopefully will gain application in the synthesis of other complex oligosaccharides.



Scheme 7. Synthesis of the fully protected nonasaccharide **28** and deprotection of compounds **25**, **26** and **28** into free oligosaccharides **1**, **2**, and **3**. (i) abs. CH_2Cl_2, TMSOTf (1 equiv.), –45°C, 2 h, (ii) $NaOCH_3$, MeOH, rt. quant., (iii) 90% aq. CF_3COOH, rt, 15 min.

Acknowledgements

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References

1. Hoson, T. *Int. Rev. Cytol.* **1991**, *130*, 233.
2. Wagner, H.; Proksch, A. *Angew. Phytother.* **1981**, *2*, 166.
3. Proksch, A.; Wagner, H. *Phytochemistry* **1987**, *26*, 1989.
4. Lipták, A.; Fügedi, P.; Kerékgyártó, J.; Nánási, P. *Carbohydr. Res.* **1983**, *113*, 225.
5. Gent, P. A.; Gigg, R. *J. Chem. Soc., Perkin Trans. 1* **1974**, 1835.
6. Glaudemans, C. P. J.; Zissis, E.; Jolley, M. E. *Carbohydr. Res.* **1975**, *40*, 129.
7. Borbás, A.; Jánossy, L.; Lipták, A. *Carbohydr. Res.* **1999**, *318*, 98.
8. Csávás, M.; Borbás, A.; Jánossy, L.; Batta, Gy.; Lipták, A. *Carbohydr. Res.* **2001**, *336*, 107.
9. Csávás, M.; Borbás, A.; Szilágyi, L.; Lipták, A. *Synlett* **2002**, *6*, 887.
10. Kanie, O.; Ito, Y.; Ogawa, T. *J. Am. Chem. Soc.* **1997**, *116*, 12073.
11. Gaunt, M. J.; Yu, J.; Spencer, J. B. *J. Org. Chem.* **1998**, *63*, 4172.
12. For some recent examples, see: (a) Xia, J.; Abbas, S. A.; Locke, R. D.; Piskorz, C. F.; Alderfer, J. L.; Matta, K. L. *Tetrahedron Lett.* **2000**, *41*, 169; (b) Liao, W.; Locke, R. D.; Matta, K. L. *Chem. Commun.* **2000**, 369; (c) Lipták, A.; Borbás, A.; Jánossy, L.; Szilágyi, L. *Tetrahedron Lett.* **2000**, *41*, 4949; (d) Borbás, A.; Szabó, Z.; Szilágyi, L.; Bényei, A.; Lipták, A. *Tetrahedron* **2002**, *58*, 5723.
13. MALDI-TOF measurements: Compound **18**: 1884.37, **19**: 1684.26, **21**: 1942.62, **23**: 2159.11, **25**: 2110.05, **26**: 2325.79, **28**: 2325.95, **1**: 1277.27, **2**: 1410.74, **3**: 1411.01 [M+Na^+].