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Note

Preparative route to glucuronyl donors bearing temporary protecting group at O-3 via 6,3-lactonisation by Bz_2O or Piv_2O☆

Andrei V. Kornilov, Elena V. Sukhova, Nikolay E. Nifantiev*

N.D. Zelinsky Institute of Organic Chemistry, Russian Academy of Sciences, Leninsky Prospect 47, 119991 Moscow B-334, Russia

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Abstract

Heating of non-substituted β-D-glucopyranuronic acids in the presence of pivalic or benzoic anhydride in DMF gives 4-*O*-monoacylated or 2,4-di-*O*-acylated 6,3-lactones that can be easily transformed into corresponding methyl uronates bearing free OH-groups at C-2 and C-3 or C-3 only, respectively. This method was used for preparation of selectively 3-*O*-protected glucuronyl donors of thioglycoside and trichloracetimidate types. © 2001 Elsevier Science Ltd. All rights reserved.

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The glucuronyl blocks which carry free or selectively substituted OH-group at C-3 are necessary for the preparation of a variety of oligosaccharides, particularly HNK-1 antigenic structures.[1–4] Such blocks can be prepared via regoiselective *O*-acylation of respective intermediate stannylidene derivatives.[2,3] Another method[1,4,5] consists of treatment of glucopyranuronosides with Ac_2O followed by subsequent methanolysis of the corresponding 6,3-lactone formed to give 3-monohydroxy 2,4-di-*O*-acetylated glucuronosides.

The latter method is effective but gives products which are not often optimal for further synthetic applications, particularly for preparation of glucuronyl donors because the 2-*O*-acetylated ones are less effective reagents as compared with 2-*O*-pivaloylated[6,7] or 2-*O*-benzoylated[8] compounds. In this paper we describe the lactonisation of glucopyranouronosides **1** and **2**[9] in the presence of Bz_2O and Piv_2O as a key step in syntheses of 2,4-di-*O*-benzoylated and pivaloylated glucuronopyranosyl derivatives bearing unsubstituted or temporary protected HO-group at C-3. The starting acid **1** was obtained by treatment of ethyl[methyl(2,3,4-tri-*O*-acetyl-1-thio-β-D-glucopyranosyl)uronate][8] with LiOH in aqueous THF and purified by ion-exchange chromatography.

☆ Synthesis of oligosaccharides related to HNK-1 antigen,[1] Part 4.

* Corresponding author. Fax: +7-095-1358784.
E-mail address: nen@ioc.ac.ru (N.E. Nifantiev).

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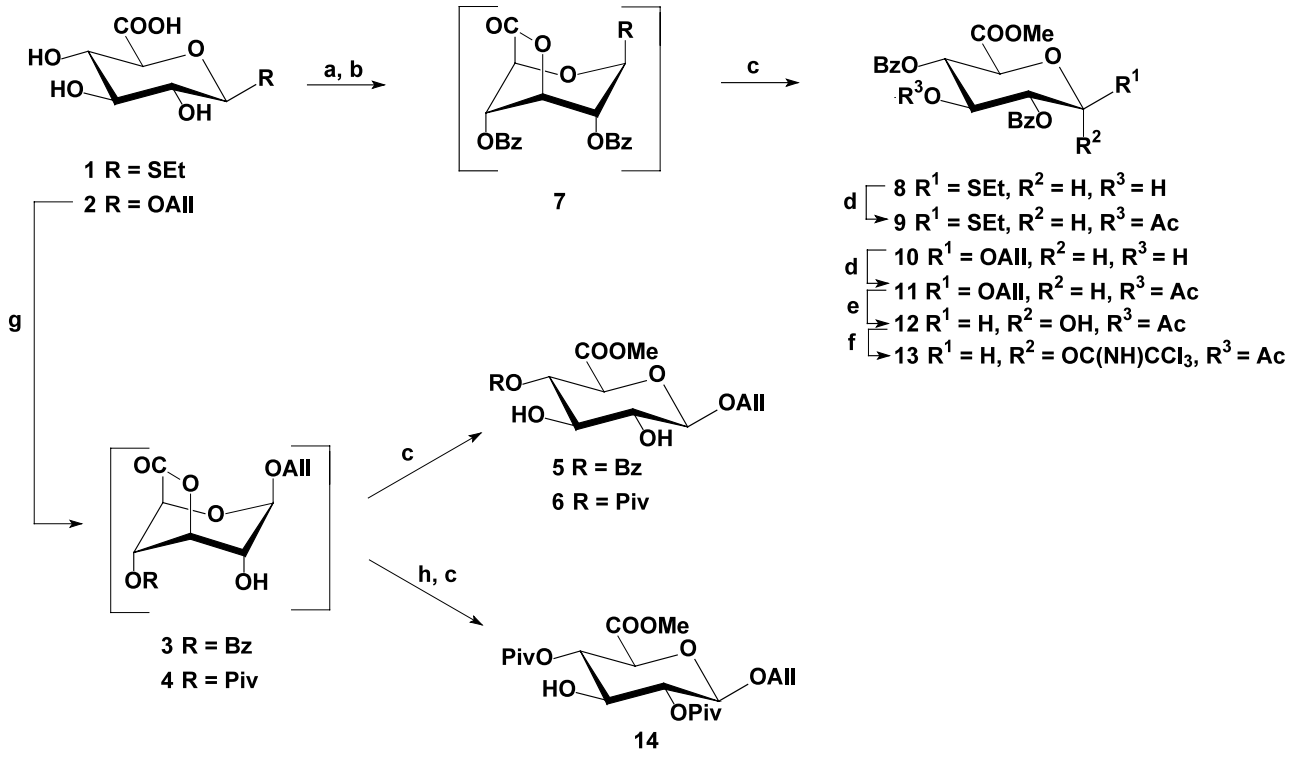
The known conditions for lactonisation of glucuronic acid glycosides in acetic anhydride which was also used as a solvent[5] are not acceptable for treatment with Bz_2O or Piv_2O. We found that the DMF is the optimal solvent for such reactions and their result depends on the amount of acylating agents and reaction conditions used. Particularly, heating of the acids **1** and **2** in presence of 20–25 equivalents of Bz_2O gave the mixture of 4-*O* and 2,4-di-*O*-benzoylated lactones. In contrast, lactonisation of the acid **2** under the same conditions but with six equivalents of Bz_2O resulted in the formation of the monobenzoylated product **3** only. 4-*O*-Monoacylated lactone **4** was also formed exclusively during lactonisation of acid **2** with 39 equivalents of Piv_2O even under heating up to 9 h.

Without purification, the lactones **3** and **4** were subjected to sodium acetate catalysed methanolysis to give the 2,3-diols **5** and **6** in 69 and 59% overall yields, respectively. The presence of ester group at O-4 in diols **5** and **6** was followed from the low-field position of the H-4 signal (δ 5.26 and 4.94 ppm, respectively) in the ^{1}H NMR spectra.

The most effective procedure developed for preparation of 2,4-di-*O*-acylated derivatives includes three steps: (1) Lactonisation of an acid in DMF under heating at 80–85 °C for 3–9 h with either benzoic (20–25 equivalents) or pivalic anhydride (38 equivalents); (2) complete *O*-acylation by addition of Py and DMAP at rt with the formation of peracylated 6,3-lactone of the common structure **7**; (3) methanolysis of the lacton ring in the presence of anhydrous AcONa (Scheme 1).

Such treatment of compounds **1** and **2** gave the 2,4-di-*O*-benzoylated derivatives **8**, **10** and 2,4-di-*O*-pivaloated **14** in 64, 64% and 60% overall yields, respectively, that coincided with the efficiency of the preparation of respective 2,4-di-*O*-acetylated derivatives with the use of Ac_2O.[5] The presence of acyl groups at O-4 and O-2 in the products **8**, **10** and **14** was confirmed by the low-field shift values of H-4 (δ 5.44, 5.50, 5.08 ppm) and H-2 signals (δ 5.32, 5.26, 4.89 ppm) in contrast to the high-field values of H-3 (δ 4.17, 4.18, 3.78 ppm,



Scheme 1. Reagents and conditions: (a) Bz_2O (20–25 equiv), DMF, 85 °C, 4 h; (b) Bz_2O, Py, DMAP, DMF, 36 h, rt; (c) MeOH, anhyd AcONa, DMF; (d) Ac_2O, Py; (e) $PdCl_2$, MeOH, 2 h; (f) CCl_3CN, K_2CO_3, CH_2Cl_2, −5 °C; (g) Bz_2O (6 equiv) or Piv_2O (38 equiv), 85 °C, DMF, 3 h; (h) Piv_2O, Py, DMAP, DMF, 36 h, rt.

respectively) in ^{1}H NMR spectra. Compound **10** contained a small quantity of aromatic impurities difficult to separate by flash chromatography and subsequent crystallisation. Therefore it was characterised as 3-*O*-acetate **11**. The monohydroxy compound **8** was also acetylated to give the compound **9**.

The glycoside **11** was deallylated with $PdCl_2$ in MeOH[10] to give the hemicetal **12**, which was then treated with CCl_3CN and K_2CO_3 to afford the glucuronyl trichloroacetimidate **13** in 58% overall yield. Compounds **9** and **13** were synthesised for the use as glucuronyl-donor blocks with a temporary acetyl group by O-3 which is selectively removable by mild-acid methanolysis.[11]

In conclusion, lactonisation of the *O*- and *S*-pyranosides of glucuronic acid in the presence of benzoic or pivalic anhydride and subsequent methanolysis leads to the respective 2,4-di-*O*- or 4-*O*-monoacylated derivatives depending on conditions used. The 4-*O*-acylated lactones are the convenient precursors for derivatives of glucopyranuronosides bearing different blocking groups at O-2, O-3 and O-4.

1. Experimental

For description of general experimental procedures, see Ref. 1.

*Ethyl (1-thio-β-*D*-glucopyranosyluronic acid)* (**1**).—To a solution of ethyl[methyl-(2,3,4-tri-*O*-acetyl-1-thio-β-D-glucopyranosyl)-uronate][8] (620 mg, 1.65 mmol) in 16.5 mL (10:1) THF–water, LiOH (2 M, 5 mL) was added at −10 °C, the reaction mixture was allowed to attain rt during 5 h. Cation-exchange resin KU-2 (H^+) (4 mL) and MeOH (10 mL) were added, the mixture was kept for 0.5 h and the resin was filtered off. The resin was washed with MeOH (20 mL) and the filtrate was evaporated. Ion-exchange chromatography and subsequent freeze-drying resulted in amorphous **1** (370 mg, 94%): $[\alpha]_D$ −35° (*c* 1, CH_3OH); ^{1}H NMR (CD_3OD): δ 4.40 (d, 1 H, $J_{1,2}$ 9.1 Hz, H-1), 3.75 (d, 1 H, $J_{4,5}$ 8.8 Hz, H-5), 3.49 (br.t, 1 H, H-4), 3.37 (t, 1 H, $J_{2,3} = J_{3,4}$ 8.5 Hz, H-3), 3.20 (dd, 1 H, H-2), 2.65 (m, SCH_2CH_3), 1.25 (t, SCH_2CH_3). Anal. Calcd for $C_8H_{14}O_6S$: C, 40.33; H, 5.92. Found: C, 40.39; H, 5.96.

*Allyl [methyl(4-*O*-benzoyl-β-*D*-glucopyranosyl)uronate]* (**5**).—The solution of acid **2**[9] (80 mg, 0.34 mmol) was treated with Bz_2O (463 mg, 2.05 mmol) in 3 mL DMF at 75 °C during 3 h and the mixture was diluted with EtOAc (20 mL), washed with satd aq $NaHCO_3$ (40 mL), filtered through cotton wool and concentrated. The residue was dried, then dissolved in anhyd MeOH (5 mL), anhyd NaOAc (16 mg, 0.24 mmol) was added and the mixture was kept for 20 h at rt. The mixture was deionised with cation-exchange resin KU-2 (H^+) during 0.5 h, the resin was filtered off and washed with MeOH (20 mL), filtrates were combined and evaporated. Column chromatography of the residue (1:2 EtOAc–petroleum ether) gave **5** (83 mg, 69%) as a syrup: $[\alpha]_D$ −17° (*c* 1, $CHCl_3$); ^{1}H NMR ($CDCl_3$): δ 5.93 (m, 1 H, $OCH_2CH{=}CH_2$), 5.39–5.14 (m, 2 H, $OCH_2CH{=}CH_2$), 5.26 (dd, 1 H, $J_{3,4}$ 9.5, $J_{4,5}$ 9.9 Hz, H-4), 4.45 (d, 1 H, $J_{1,2}$ 7.4 Hz, H-1), 4.43–4.32 and 4.20–4.13 (2m, 2 H, $OCH_2CH{=}CH_2$), 4.07 (d, 1 H, H-5), 3.86 (dd, 1 H, $J_{2,3}$ 9.2 Hz, H-3), 3.60 (dd, 1 H, H-2), 3.60 (s, 3 H, CO_2CH_3). Anal. Calcd for $C_{17}H_{18}O_8$: C, 58.29; H, 5.18. Found: C, 58.33; H, 5.24.

*Allyl [methyl(4-*O*-pivaloyl-β-*D*-glucopyranosyl)uronate]* (**6**).—The solution of acid **2** (245 mg, 1.05 mmol) in DMF (8 mL) was treated with Piv_2O (8 mL, 39.43 mmol), and then with anhyd MeOH (10 mL) and anhyd NaOAc (32 mg, 0.48 mmol) as described for **5**. Column chromatography (1:2 EtOAc–petroleum ether) gave **6** (205 mg, 59%) as a syrup: $[\alpha]_D$ −50° (*c* 1, $CHCl_3$); ^{1}H NMR ($CDCl_3$): δ 5.90 (m, 1 H, $OCH_2CH{=}CH_2$), 5.33–5.15 (m, 2 H, $OCH_2CH{=}CH_2$), 4.94 (dd, 1 H, $J_{3,4}$ 9.5, $J_{4,5}$ 9.9 Hz, H-4), 4.38 (d, 1 H, $J_{1,2}$ 7.5 Hz, H-1), 4.36–4.30 and 4.19–4.03 (2m, 2 H, $OCH_2CH{=}CH_2$), 3.94 (d, 1 H, H-5), 3.68 (dd, 1 H, $J_{2,3}$ 9.1 Hz, H-3), 3.66 (s, 3 H, CO_2CH_3), 3.52 (dd, 1 H, H-2). Anal. Calcd for $C_{15}H_{24}O_8$: C, 54.21; H, 7.28. Found: C, 54.19; H, 7.15.

*Ethyl [methyl(2,4-*O*-benzoyl-1-thio-β-*D*-glucopyranosyl)uronate]* (**8**).—To a solution of **1** (65 mg, 0.27 mmol) in DMF (5 mL), Bz_2O (1.53 g, 6.83 mmol) was added and the reaction mixture was kept at 85 °C for 3 h. Pyri-

dine (3 mL) and DMAP (17 mg, 0.34 mmol) were added at rt and the solution was kept for 36 h, diluted with EtOAc (20 mL) and washed with 1 M H_2SO_4 (20 mL), satd aq $NaHCO_3$ (50 mL), and satd aq NaCl (10 mL), filtered through cotton wool, evaporated and coevaporated with toluene (10 mL), dried and dissolved in anhyd MeOH (2 mL). To this solution the anhyd NaOAc (10 mg, 0.15 mmol) was added and the mixture was kept for 12 h at rt. Cation-exchange resin KU-2 (2 mL) was added to a solution for 0.5 h and filtered off. The resin was washed with MeOH (20 mL) and the filtrates were combined and concentrated. Column chromatography of the residue (1:2 EtOAc–petroleum ether) and subsequent crystallisation gave **8** (80 mg, 64%) as needles: mp 164–166 °C (EtOAc–petroleum ether); $[\alpha]_D$ −11° (*c* 1, $CHCl_3$); ^1H NMR ($CDCl_3$): δ 5.44 (t, 1 H, $J_{3,4}$ 9.6 Hz, H-4), 5.32 (t, 1 H, $J_{2,3}$ 9.4 Hz, H-2), 4.71 (d, 1 H, $J_{1,2}$ 9.9 Hz, H-1), 4.20 (d, 1 H, $J_{4,5}$ 9.9 Hz, H-5), 4.17 (t, 1 H, H-3), 3.66 (s, 3 H, CO_2CH_3), 2.75 (m, SCH_2CH_3), 1.23 (t, SCH_2CH_3). MALDI-TOF-MS: Calcd for $[M + Na]^+$: 483.5. Found: 483.1. Anal. Calcd for $C_{23}H_{24}O_8S$: C, 59.92; H, 5.27; S, 6.80. Found: C, 59.99; H, 5.25; S, 6.96.

*Ethyl [methyl(3-*O*-acetyl-2,4-*O*-benzoyl-1-thio-β-*D*-glucopyranosyl)uronate]* (**9**).—Glucuronide **8** (60 mg, 0.13 mmol) was acetylated in a mixture of Py (2 mL) and Ac_2O (2 mL) at 20 °C during 24 h. The mixture was coevaporated with toluene (10 mL) and dried. Column chromatography of the residue (1:2 EtOAc–petroleum ether) and subsequent crystallisation gave 3-*O*-acetate **9** (63 mg, 94%) as needles: mp 207–209 °C (EtOAc–petroleum ether); $[\alpha]_D$ −9° (*c* 0.6, $CHCl_3$); ^1H NMR: δ 5.70 (t, 1 H, $J_{3,4}$ 9.4 Hz, H-3), 5.55 (t, 1 H, $J_{4,5}$ 9.6 Hz, H-4), 5.42 (t, 1 H, $J_{2,3}$ 9.0 Hz, H-2), 4.78 (d, 1 H, $J_{1,2}$ 9.0 Hz, H-1), 4.26 (d, 1 H, H-5), 3.70 (s, 3 H, CO_2CH_3), 2.78 (m, SCH_2CH_3), 1.25 (t, SCH_2CH_3). MALDI-TOF-MS: Calcd for $[M + Na]^+$: 525.5. Found: 524.9. Anal. Calcd for $C_{25}H_{26}O_9S$: C, 59.75; H, 5.21. Found: C, 59.82; H, 5.21.

*Allyl [methyl(3-*O*-acetyl-2,4-*O*-benzoyl-β-*D*-glucopyranosyl)uronate]* (**11**).—The solution of acid **2** (320 mg, 1.37 mmol) in DMF (15 mL) was treated with Bz_2O (6.19 g, 27.4 mmol), then with pyridine (5 mL), DMAP (84 mg, 0.69 mmol), and finally with MeOH (2 mL) and anhyd NaOAc (10 mg, 0.15 mmol) as described for **8**. Column chromatography (1:2 EtOAc–petroleum ether) gave **10** (381 mg), which was then acetylated with Ac_2O (2 mL) in Py (2 mL) at 20 °C for 24 h. The solution was coevaporated with toluene (2 × 10 mL) and the residue was subjected to column chromatography (1:2 EtOAc–petroleum ether) and subsequent crystallisation gave **11** (415 mg, 61%) as needles: mp 176–177 °C (1:2 EtOAc–petroleum ether); $[\alpha]_D$ −3° (*c* 1, $CHCl_3$); ^1H NMR (CD_3OD): δ 5.78 (m, 1 H, $OCH_2CH{=}CH_2$), 5.68 (dd, 1 H, $J_{2,3}$ 9.1, $J_{3,4}$ 9.3 Hz, H-3) 5.59 (t, 1 H, $J_{4,5}$ 9.3 Hz, H-4), 5.43 (dd, 1 H, $J_{1,2}$ 7.4 Hz, H-2), 5.29–5.01 (m, 2 H, $OCH_2CH{=}CH_2$), 4.85 (d, 1 H, H-1), 4.45–4.33 and 4.20–4.05 (2m, 2 H, $OCH_2CH{=}CH_2$), 4.29 (d, 1 H, H-5), 3.70 (s, 3 H, CO_2CH_3), 1.85 (s, 3 H, $COCH_3$). MALDI-TOF-MS: Calcd for $[M + Na]^+$: 521.5. Found: 521.1. Anal. Calcd for $C_{26}H_{26}O_{10}$: C, 62.65; H, 5.26. Found: C, 62.59; H, 4.98.

*Methyl (3-*O*-acetyl-2,4-*O*-benzoyl-α-*D*-glucopyranosyl trichloroacetimidate)uronate* (**13**).—Palladium chloride (25.8 mg, 0.15 mmol) was added to a solution of **11** (207 mg, 0.42 mmol) in anhyd MeOH (10 mL). The reaction mixture was stirred for 2 h at rt and filtered through a Celite layer, and evaporated. The residue was diluted with of EtOAc (20 mL), washed with satd aq $NaHCO_3$ (50 mL), filtered through cotton wool, concentrated and then filtered through a pad of silica gel to give 135 mg of **12** as amorphous foam. Hemicetal **12** was treated with trichloroacetonitrile (295 μL, 2.95 mmol) and K_2CO_3 (200 mg, 1.43 mmol) in anhyd CH_2Cl_2 (2 mL) under dry argon at −5 °C for 2.5 h and the mixture was concentrated. Column chromatography of the residue (1:3 EtOAc–petroleum ether + 1% Et_3N) gave amorphous **13** (151 mg, 58%): $[\alpha]_D$ +61° (*c* 0.5, $CHCl_3$); ^1H NMR ($CDCl_3$): δ 8.70 (s, 1 H, N*H*), 6.85 (d, 1 H, $J_{1,2}$ 3.6 Hz, H-1), 6.04 (t, 1 H, $J_{2,3} = J_{3,4}$ 10.0 Hz, H-3), 5.58 (dd, 1 H, $J_{4,5}$ 10.2 Hz, H-4), 5.46 (dd, 1 H, H-2), 4.67 (d, 1 H, H-5), 3.68 (s, 3 H, CO_2CH_3), 1.90 (s, 3 H, $COCH_3$).

Allyl [methyl(2,4-O-pivaloyl-β-D-glucopyranosyl)uronate] (**14**).—Acid **2** (123 mg, 0.52 mmol) was treated with Piv_2O (4 mL, 19.72 mmol) in DMF (4 mL) for 9 h, then with Py (4 mL) and DMAP (16.7 mg, 0.34 mmol) for 36 h and finally with anhyd MeOH (2 mL) and anhyd NaOAc (10 mg, 0.15 mmol) as described for **8** and subjected to column chromatography (1:3 EtOAc–petroleum ether) and subsequent crystallisation to give **14** (123 mg, 60%) as needles: mp 120–121 °C (1:3 EtOAc–petroleum ether); $[\alpha]_D$ −48° (*c* 1, $CHCl_3$); ^1H NMR ($CDCl_3$): δ 5.81 (m, 1 H, $OCH_2CH{=}CH_2$), 5.29–5.14 (m, 2 H, $OCH_2CH{=}CH_2$), 5.08 (dd, 1 H, $J_{3,4}$ 9.2, $J_{4,5}$ 9.8 Hz, H-4), 4.89 (dd, 1 H, $J_{1,2}$ 7.7, $J_{2,3}$ 9.2 Hz, H-2), 4.55 (d, 1 H, H-1), 4.38–4.28 and 4.10–4.00 (2m, 2 H, $OCH_2CH{=}CH_2$), 3.97 (d, 1 H, H-5), 3.78 (t, 1 H, H-3), 3.72 (s, 3 H, CO_2CH_3). Anal. Calcd for $C_{20}H_{32}O_9$: C, 57.68; H, 7.74. Found: C, 57.43; H, 7.68.

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References

1. Kornilov, A. V.; Sherman, A. A.; Kononov, L. O.; Shashkov, A. S.; Nivant'ev, N. E. *Carbohydr. Res.* **2000**, *329*, 717–730.
2. Nakano, T.; Ito, Y.; Ogawa, T. *Carbohydr. Res.* **1993**, *243*, 43–69.
3. Isogai, Y.; Kawase, T.; Ishida, H.; Kiso, M.; Hasegawa, A. *J. Carbohydr. Chem.* **1996**, *15*, 1001–1023.
4. Kononov, L. O.; Kornilov, A. V.; Sherman, A. A.; Zyryanov, E. V.; Zatonsky, G. V.; Shashkov, A. S.; Nifant'ev, N. E. *Bioorg. Khim.* **1998**, *24*, 608–622.
5. Chernyak, A. Ya.; Kononov, L. O.; Kochetkov, N. K. *Carbohydr. Res.* **1991**, *216*, 381–398 and references cited therein.
6. Vlahov, J.; Snatzke, G. *Liebigs Ann. Chem.* **1983**, 570–574.
7. Kornilov, A. V.; Kononov, L. O.; Zatonsky, G. V.; Shashkov, A. S.; Nifant'ev, N. E. *Bioorg. Khim.* **1997**, *23*, 655–666.
8. Krog-Jensen, C.; Oscarson, S. *Carbohydr. Res.* **1998**, *308*, 287–296.
9. Chernyak, A. Ya.; Antonov, K. V.; Kochetkov, N. K. *Bioorg. Khim.* **1987**, *13*, 958–966.
10. Ogawa, T.; Yamamoto, H. *Agric. Biol. Chem.* **1985**, *49*, 475–482.
11. Byramova, N. E.; Ovchinnikov, M. V.; Backinowsky, L. V.; Kochetkov, N. K. *Carbohydr. Res.* **1983**, *124*, c8–c11.