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SYNTHESIS OF POLYSACCHARIDE α-(1→3)-L-RHAMNAN

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Summary: The regular polysaccharide α-(1→3)-L-rhamnan (DP ca. 30) was obtained by polycondensation of 4-O-acetyl-3-O-trityl-1,2-O-exo-cyanoethylidene-L-rhamnopyranose catalyzed with triphenylmethylium perchlorate.

Glycosylation of O-trityl ethers by 1,2-O-cyanoethylidene derivatives of sugars[1] results in formation of 1,2-trans-glycosidic linkage practically with absolute stereospecificity[2]. Polycondensation process based on this reaction revealed for the first time a general synthetical route to polysaccharides of regular structure[3,4].

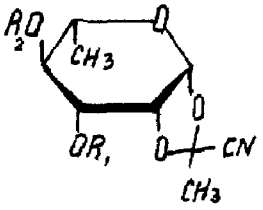
Here we describe the synthesis of α-(1→3)-L-rhamnan, which demonstrates the application of the method for the synthesis of polysaccharides when a secondary hydroxyl group is involved in formation of interglycosidic linkage.

The starting monomer (I) for polycondensation was prepared as follows. Deacetylation of 3,4-di-O-acetyl-1,2-O-exo-cyanoethylidene-β-L-rhamnose[5] (II) 0.005 M CH_3ONa/CH_3OH, 32 ml in 16 ml $CHCl_3$, 30 min, room temp.)[6] gave rise to the free ketal (III) in 98% yield, m.p. 117-118° (ether), $[\alpha]_D$ -12.1° (c 2.0, $CHCl_3$); PMR-spectrum (CD_3OD, δ ppm): 5.44 (d, $J_{1,2}$ 2 Hz, 1H, H1), 4.45 (dd, 1H, H2), 3.79 (m, 1H, H3), 3.32 (m, 2H, H4, H5), 1.83 (s, 3H, C-CH_3), 1.26 (d, $J_{5,6}$ 6 Hz, 3H, C-CH_3 of rhamnose).

III was treated with triphenylmethylium perchlorate[7] (1 mol) in the presence of collidine in CH_2Cl_2 (1h, room temp.). Tritylation of equatorial C_3 hydroxyl group proceeds practically selectively with formation of monotrityl derivative (IV); 84%, m.p. 157-158° (ether - light petroleum), $[\alpha]_D$ +35.6° (c

2.0, $CHCl_3$); PMR-spectrum (CCl_4, δ, ppm): 7.2-7.6 (15H, aromatic), 4.88 (d, $J_{1,2}$ 2 Hz, 1H, H1), 3.8 (m, 1H, H2), 3.6 (m, 2H, H3, H4), 2.93 (m, 1H, H5), 1.89 (s, 3H, C-CH_3), 1.14 (d, $J_{5,6}$ 6 Hz, 3H, C-CH_3 of rhamnose).

Methylation of IV with subsequent standard treatment resulted in formation 1,2,3,5-tetra-O-acetyl-4-O-methyl-rhamnitol as a single product proving unambiguously the position of trityl substituent in IV. Standard acetylation of IV (acetic anhydride in pyridine, 12h, room temp.) gave rise to the monomer I, 90%, m.p. 178-179° (CH_3OH), $[\alpha]_D$ +19.8° (c 2.0, $CHCl_3$); PMR-spectrum (CCl_4, δ, ppm); 7.2-7.6 (15H, aromatic), 5.09 (t, $J_{3,5}$ 9 Hz, 1H, H4), 4.85 (d, $J_{1,2}$ 2 Hz, 1H, H1), 3.75 (m, 2H, H2, H3), 3.09 (m, 1H, H5), 1.94 (s, 3H, OAc), 1.7 (s, 3H, C-CH_3), 1.08 (d, $J_{5,6}$ 6 Hz, 3H, C-CH_3 of rhamnose).



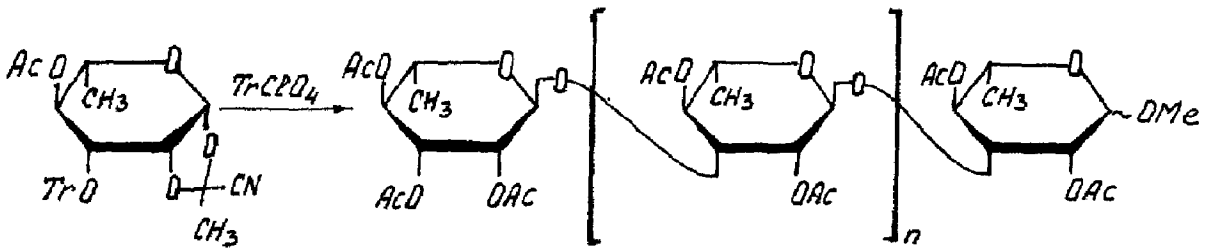
I $R_1 = Ph_3C$, $R_2 = CH_3CO$.

II $R_1 = R_2 = CH_3CO$.

III $R_1 = R_2 = H$.

IV $R_1 = Ph_3C$, $R_2 = H$.

The polycondensation of I was performed as described earlier[3,4]. Solution of I (1 mmol) in CH_2Cl_2 (5 ml) was treated with triphenylmethylium perchlorate (0.1 mmol) at room temperature for 60 hours using vacuum technique.



Increasing of the reaction time up to 120h does not change the yield and molecular weight of the polymer formed. After 60 h CH_3OH and pyridine were added to the reaction mixture to stop the polymerization process. The solution was evaporated, the residue was washed with hexane to remove TrCN and treated with 1 M CH_3ONa in CH_3OH at room temperature monitoring the deacetylation by IR-spectroscopy (1750 cm^{-1}-. Only repeated treatment with 1 M CH_3ONa/CH_3OH completely removed the acetyl groups. Two fractions of free polysaccharide were obtained. Water-insoluble fraction (70 mg) was removed by filtration, column chromatography of the water-soluble fraction on Biogel P-10 (80 x 1.3

cm, V_o 29 ml, elution with 0.1 M AcOH) gave rise to the high molecular weight fraction, 10 mg, $[\alpha]_D$ -63.3° (c 0.76, H_2O). The total yield of free polysaccharide was 69% (starting on I). Both fractions had the identical structure and differed only by molecular weight.

Formolysis (85% HCOOH, 100, 2h) and hydrolysis (0.13 M H_2SO_4, 100°, 15 h) of both fractions gave rise to L-rhamnose as a single product identified by GLC as L-rhamnitol pentaacetate (GLC and GLC-MS). Methylation of both fractions with subsequent standard procedure[8] gave rise to 1.5-di-O-acetyl-2,3,4-tri-O-methyl-rhamnitol (V) and 1,3,5-tri-O-acetyl-2,4-di-O-methyl-rhamnitol (VI) as the only products (identification by GLC and GLC-MS). These data demonstrate complete regiospecificity of polycondensation process and prove unambiguously the structure of both fractions of synthetic polysaccharide as 1,3-L-rhamnan. The V : VI ratio for water-soluble fraction is 1 : 16, that for water-insoluble fraction - 1 : 30, that gives degree of polymerization 16 and 30, and molecular weight of about 2500 and 4500 respectively.

The proof of stereospecificity of glycosidic linkages presents some problems due to insolubility of synthesis rhamnan, although the insolubility itself demonstrates a high degree of regularity of polymer (cf.[9]). To this end the total product of polycondensation after careful washing off the low-molecularweight impurities was oxidized with CrO_3 in Ac_2O/AcOH[10] at 50° for 1.5 and 4 hours. Products of the reaction were reduced with $NaBH_4$ and subjected to methylation analysis according to the standard procedure[8] followed by GLC analysis, the ratio V : VI in an 1.5- hour experiment being 1 : 15 and that in 4-hour experiment 1:16. It indicates that not more than one glycosidic linkage was cleaved and increase of the oxidation time does not change the result. Taking into account the known lack of absolute specificity of CrO_3 - oxidation[10] and possibility of glycosidic linkage acetolysis under conditions used these data demonstrate practically full stereospecificity of the synthetic rhamnan, containing only α-(1→3)- glycosidic linkages.

The ^{13}C NMR spectrum of the permethylated rhamnan (VII) prepared by Hakomori methylation is in complete agreement with this conclusion. The data for polysaccharide and two reference compounds 2,3,4-tri-O-methyl-β-L-rhamnoside (VIII) and its α-anomer (IX) are listed in Table 1.

Synthetic rhamnan exhibits the only signal at 99.2 ppm in the region of anomeric carbon resonance and more diagnostic signal at 68.7 ppm in the C_5-resonance region which are characteristic for α-rhamnosidic linkage; in the region near 102 ppm (C_1, β-anomer) and 72 ppm (C_5, β-anomer) no signals are present.

Table 1

^{13}C data of the permethylated compounds VII - IX
(solution in $CDCl_3$, TMS, δ scale)

Compounds	C_1	C_2	C_3	C_4	C_5	C_6	OCH_3 at C_1	OCH_3 at C_2	OCH_3 at C_3	OCH_3 at C_4
α-(1→3)-L-rhamnan	99.2	80.9	79.5*	82.4*	68.7	18		58.8		61.1
Rhap→Me	102.5	77.35	84.3	82.1	71.9	17.8	56.9	61.55	57.45	60.8
Rhap→Me	98.35	77.55	81.65	82.35	67.9	17.9	54.6	58.95	57.6	60.7

* attribution may be reversed.

The synthesis of α-(1→3)-L-rhamnan presented above is a good demonstration for possible extention and more general application of the new method of synthesis of polysaccharides.

References

1. A.F.Bochkov, N.K.Kochetkov, Carbohydrate Res., 39, 355 (1975)
2. V.I.Betaneli, M.V.Ovchinnikov, L.V.Backinowsky, N.K.Kochetkov, Carbohydrate Res., 76, 252 (1979).
3. A.F.Bochkov, I.V.Obruchnikov, V.M.Kalinevich, N.K.Kochetkov, Tetrahedron Lett., (1975), 3403.
4. N.K.Kochetkov, I.V.Obruchnikov, Tetrahedron Lett., (1977), 57.
5. V.I.Betaneli, M.V.Ovchinnikov, L.V.Backinowsky, N.K.Kochetkov, Carbohydrate Res., 68, C11 (1979).
6. V.I.Betaneli, M.V.Ovchinnikov, L.V.Backinowsky, N.K.Kochetkov, Dokl. Akad. Nauk SSSR, in press.
7. H.J.Dauben, Jr., L.R.Honnen, K.M.Harmon. J.Org.Chem., 25, 1442 (1960).
8. C.G.Hellerqvist, B.Lindberg, S.Svensson, Carbohydrate Res., 8, 43 (1968).
9. A.I.Usov, K.S.Adamyats, S.V.Yarotsky, A.A.Anoshina, N.K.Kochetkov, Zh. Obshch. Khim., 44, 416 (1974).
10. J.Hoffman, B.Lindberg, S.Svensson, Acta Chem. Scand., 26, 661 (1972).

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