Macromolecules

Volume 17, Number 5 May 1984

Synthesis of (1→3)-α-D-Mannopyranan by Stereoregular Cationic Polymerization of Substituted 2,6-Dioxabicyclo[3.1.1]heptanes

Fanzuo Kong and Conrad Schuerch*

Department of Chemistry, College of Environmental Science and Forestry, State University of New York, Syracuse, New York 13210. *Received June 6, 1983*

ABSTRACT: The polymerization of 1,3-anhydro-2,4,6-tri-*O*-(*p*-bromobenzyl)- (I) and 1,3-anhydro-2,4,6-tri-*O*-benzyl-D-mannopyranose (II) was studied under a variety of conditions with different catalysts. Cationic catalysts like trifluoromethanesulfonic anhydride, triphenylcarbenium perchlorate, and the radical cation salt tris(4-bromophenyl)amminium hexachloroantimonate caused stereoregular polymerization to give virtually completely α-linked (1→3)-D-mannopyranan derivatives in high yield and with DP 60–90. Other cationic catalysts caused polymerization with high yields of predominantly α-linked polymers under some conditions. Coordination catalysts and some metal complexes caused polymerization in poor yield, but anionic catalysts failed to polymerize the monomers. The influence of monomer structure, temperature, and solvent on stereoregularity of the polymers was investigated. The substituted polymers were characterized by optical rotation, gel permeation chromatography, osmometry (vapor phase and membrane), intrinsic viscosity, and circular dichroism. The relation of C-1 configuration and molecular volume of the polymers is discussed. Debromobenzylation and debenzylation of the polymers afforded the unsubstituted linear 1,3-D-mannopyranans, which were characterized with optical rotation, IR, ^{13}C NMR, and ^{1}H NMR spectrometry and by complete hydrolysis to mannose by trifluoroacetic acid. Anomeric purity was investigated by selective oxidations of the triacetates. The ^{13}C NMR spectra of the synthetic mannan and fully acetylated mannan were assigned.

For several years, we have been engaged in the development of methods for the synthesis of stereoregular polysaccharides since the polymers obtained are useful model compounds for immunological and allergic reactions and also for studying protein–polysaccharide interactions.[1,2] A number of other laboratories are also involved in research of this area.[3-5] A (1→6)-α-D-mannopyranan of high molecular weight previously synthesized[6] in this laboratory has proven to be cross-reactive with a mannan of similar structure produced by the skin dermatophyte *Trichophytum rubrum*.[7] It is not, however, cross-reactive with most antigenic mannans of yeasts[8] although (1→6)-α-D-mannopyranosyl units constitute the backbone of these polysaccharides. It is therefore, interesting to prepare structures characteristic of the branches which shield the backbone from interaction with antibodies and which, themselves, serve as determinants for yeast mannan. It has recently been shown that the major fraction of polysaccharides extracted from the capsular material of *Cryptococcus bacillisporus* serotypes B and C[9,10] and from *C. clinovii* and *C. uniguttulatus*[11] are D-mannosyl residues linked α-(1→3) in a linear backbone. It is also found that the fruit bodies of *Dictyophora indusiata* Fisch contain a partially acetylated linear (1→3)-α-D-mannan[12] that exhibits antitumor and antiinflammatory activities. Methylation analysis of another heteropolysaccharide isolated from the culture medium of *T. mesenterica* NRRL Y-6158 shows it to be an essentially α-(1→3)-linked mannopyranose backbone with 20% of 2-*O*-β-D-glucopyranosyluronic acid and 2-*O*- and 4-*O*-β-D-xylopyranose as branches.[13] Also, the glycosyl side chains of a number of glycoproteins are known to contain α-D-mannopyranosyl units linked (1→3).[14-18] On the other hand, (1→3)-β-linked mannopyranose residues have been found in cell wall polysaccharides.[11,19,20]

Previously a number of stereoregular polymerizations have been reported for 1,2-anhydro-,[23] 1,4-anhydro-,[24] and a number of 1,6-anhydroglycopyranose derivatives.[2] Recently, the synthesis and polymerization of 1,3-anhydro-2,4,6-tri-*O*-benzyl-β-D-glucopyranose have been reported,[25,26] and the mannopyranose analogues 1,3-anhydro-2,4,6-tri-*O*-(*p*-bromobenzyl)- (I) and 1,3-anhydro-2,4,6-tri-*O*-benzyl-β-D-mannopyranose (II) have also been synthesized by us.[27,28] We now wish to report the stereoregular polymerization of I and II, which has now led to the formation of a number of mannopyranans of different molecular weight and stereoregularity, some of which are completely stereoregular, according to NMR analysis.

Experimental Section

Materials. The monomer I [mp 91–92 °C, $[\alpha]^{23}_{D}$ +31.7° (*c* 0.8, $CHCl_3$)] was synthesized in a large scale (5 g) according to

0024-9297/84/2217-0983$01.50/0

the reported method.[28] Monomer II was synthesized by a ring-closure reaction[28] from 3-*O*-acetyl-2,4,6-tri-*O*-benzyl-α-D-mannopyranosyl chloride in a sufficient quantity (1–1.5 g) for a few polymerizations. Because monomer II did not crystallize, purification by liquid chromatography was repeated and the resulting pure monomer [$[\alpha]^{23}_D$ +53.3° (*c* 1.1, $CHCl_3$)] was dried by distilling dichloromethane from a monomer solution several times on a high-vacuum line.

Dichloromethane, toluene, benzene, xylene, and dimethoxyethane were dried by refluxing on calcium hydride, distilled, and then further dried and kept on calcium hydride under high vacuum.

Phosphorus pentafluoride was generated in situ by pyrolysis of *p*-chlorobenzenediazonium hexafluorophosphate crystallized from water or methanol and dried on a high-vacuum line. Boron trifluoride etherate, antimony pentachloride, and trifluoromethanesulfonic anhydride were distilled three times on a high-vacuum line, with the center cut taken each time, and distributed (by distillation in vacuo) to calibrated capillary tubes connected to a break seal.

Triphenylcarbenium perchlorate was prepared from triphenylcarbinol and perchloric acid according to a reported method.[35] The yellow crystalline triphenylcarbenium perchlorate (mp 145 °C; lit.[35] mp 144 °C) thus obtained could be used for polymerization directly. The perchlorate catalyst could also be prepared from triphenylchloromethane and silver perchlorate. An exactly weighed amount of triphenylchloromethane was put into a tube with a magnetic bar on a high-vacuum line, a certain volume of dichloromethane was distilled into the tube under high vacuum, an equivalent amount of silver perchlorate was added, and the reaction was conducted under high vacuum overnight. The supernatant was transferred quantitatively into break seals used for polymerization. The perchlorate catalysts obtained by using the two different methods had the same catalytic activity and gave polymers with the same properties.

Cobalt carbonyl [$Co_2(CO)_8$], molybdenum carbonyl [$Mo(CO)_6$], and tris(4-bromophenyl)amminium hexachlorantimonate were crystalline compounds from commercial sources (reagent grade) and were used directly.

The coordination catalysts $AlEt_3/H_2O$ (1:1), $AlEt_3/H_2O/(CH_3{-}CO)_2CH_2$ (1:0.5:0.5), and $ZnEt_2/H_2O$ (1:1) were prepared according to the reported method.[26]

Anionic catalyst potassium *tert*-butoxide (mp 256–258 °C) obtained from a commercial source (reagent grade) was used directly for polymerization.

Polymerization. The transfer of solvents and most of the catalysts and all the polymerizations were carried out under high vacuum as previously reported.[26] Some of the polymerization conditions are summarized in Table II. Polymerizations were terminated at the polymerization temperature by adding cold methanol. Reprecipitation was repeated three times into petroleum ether. The solutions from coordination catalyst polymerizations were diluted with chloroform, terminated with methanol, and then poured into petroleum ether. The recovered polymer was dissolved in chloroform and precipitated into petroleum ether twice after filtering out the catalyst residue.

All the substituted polymers were freeze-dried from benzene.

Preparation of (1→3)-D-Mannopyranans. Isolated polymers were debromobenzylated or debenzylated by a published method.[29] The polymer solution was purified by dialysis until ion free using a UM-5 membrane (Diaflo Ultrafilter, Amico Co.) and isolated as white fluffy solids by freeze-drying from distilled water. Elemental analysis indicated that the products after equilibration in the atmosphere contained one molecule of water.

Anal. Calcd for $(C_6H_{10}O_5{\cdot}H_2O)_n$: C, 40.00; H, 6.67. Found: C, 39.89; H, 6.65.

The observed optical rotations were corrected to the values of anhydrous polysaccharides according to the elemental analysis.

Acetylation of (1→3)-D-Mannopyranans. Freeze-dried fluffy homogeneous mannan was used for acetylation.[10] Polysaccharide (25 mg) was thoroughly mixed with formamide (0.5 mL) in a 50-mL flask. To this stirred solution were slowly added dried pyridine (6 mL) and then freshly distilled acetic anhydride (4 mL). The reaction mixture was stirred overnight at room temperature. After the acetylation, the reaction mixture was slowly added to 40 mL of ice water with stirring. Extraction with chloroform was repeated several times and the organic solution obtained was decolorized with activated carbon and then concentrated to 2 mL. Reprecipitation was repeated five times into petroleum ether to remove pyridine and white fluffy polymers were obtained by freeze-drying from benzene.

Oxidation of Acetylated (1→3)-D-Mannopyranans. The oxidation of the acetylated mannan was conducted by a published method.[31,32] The acetylated mannan (40 mg) was dissolved in glacial acetic acid (8 mL) and the solution was treated with chromium trioxide (400 mg) on an ultrasonic bath at 50 °C for 1.5 h. The material was recovered by partitioning between chloroform and water, evaporation of the chloroform phase, and finally freeze-drying from benzene. The polymer thus obtained was subjected to ^{13}C NMR determination directly.

Reduction and Deacetylation of Oxidized Acetylated Mannan. The carefully dried oxidized 2,4,6-tri-*O*-acetyl-(1→3)-D-mannopyranan (40 mg) was dissolved in dry 1,4-dioxane–ethanol (1:1, 8 mL) and treated with sodium borohydride (100 mg) with stirring overnight. Water was added and the solution was kept at room temperature for 3 h. The solution was acidified to pH 3–4 with 0.1 N HCl and then subjected to dialysis. Deacetylated mannan was obtained by freeze-drying the dialyzed solution and characterized by ^{13}C NMR.

Hydrolysis of (1→3)-D-Mannopyranans and Identification of Sugar. (1→3)-D-Mannopyranan (2–4 mg) was placed in a 50-mL flask equipped with a reflux condenser. Trifluoroacetic acid (2 N, 3 mL) was added, and the mixture was refluxed for 1 h according to a reported method.[33] The solution was evaporated in a rotary vacuum evaporator to dryness, and the residue was redissolved in distilled water (10 mL). The evaporation was repeated three times. The sugars thus obtained were dissolved in distilled water (0.1 mL), spotted on Whatman no. 1 paper, and separated by means of a moving phase of ethyl acetate/pyridine/water (10:2:1, upper layer) for 40 h. Sodium hydroxide (5%) in 95% ethyl alcohol and $AgNO_3$ (5%) in acetone were used as spray reagents. Control solutions of mannose and altrose clearly showed both sugars. The hydrolysate showed only mannose, no altrose or disaccharides.

Characterization of Polymers. ^1H and ^{13}C NMR spectra of free mannan were measured in dimethyl-d_6 sulfoxide, which also served as standard, with a Varian XL-100 spectrometer at room temperature or at 70 °C. The latter temperature gave better resolution. ^{13}C NMR spectra of acetylated and benzylated polymers were obtained in chloroform-*d* with tetramethylsilane as internal standard. The ratio of α- and β-linkages of free mannans could be estimated by comparing the average peak area of the anomeric centers, C-5 and C-3. The DP of the mannans could be roughly estimated by comparing the peak area of the C-2 (or C-3) units in the chain with the C-2 (or C-3) units of the nonreducing end group. Optical rotations were determined in chloroform for benzylated and bromobenzylated polysaccharides and in Me_2SO/H_2O (2:1) for free polysaccharides at 23 °C in a Perkin-Elmer Model 141 polarimeter with a jacketed 1-dm cell. The ratio of α- and β-linkages of free mannans could also be estimated from the specific rotation.

The IR spectra of free polysaccharides were obtained with a Perkin-Elmer Model 137 double-beam recording spectrometer on 1% of polymer in potassium bromide pellets.

The CD spectra of substituted polymers were recorded on a JASCO ORD/CD-5 spectropolarimeter equipped with the Sproul Scientific SS-107 modification in the wavelength region from 300 to 200 nm at 23 °C, using a xenon high-pressure lamp as the light source according to the reported method.[21,34]

Molecular weight distributions of 2,4,6-tri-*O*-(*p*-bromobenzyl)- and 2,4,6-tri-*O*-benzyl-(1→3)-D-mannopyranans were obtained by high-pressure gel permeation chromatography in tetrahydrofuran. A Waters Model 200 chromatograph equipped with a UV spectrometer (fixed at 245 nm), a Model 6000 pump, and a Valvco septumless injector (1.0 mL) were used. Four 30-cm-long stainless steel columns packed with μ-Styragel (1×10^4, 1×10^3, 5×10^2, and 1×10^2 Å) were used for the analysis of distribution. Flow rate was 2.0 mL/min. A calibration curve was obtained by using polystyrene (Waters) standard samples. Viscosities of the substituted polymers were determined at 25 °C in chloroform with a Cannon–Ubbelohde viscometer. A Mechrolab Model 301 vapor pressure osmometer was used for most of the measurements of

Table I
Assignments of ^{13}C NMR Spectra of Synthetic (1→3)-D-Mannopyranans and Fully Acetylated (1→3)-D-Mannopyranans (ppm)

	C-1	C-2	C-3	C-4	C-5	C-6	Me	C=O
α-D-mannopyranoside[39]	94.9	71.7	71.3	67.9	73.2	62.0		
β-D-mannopyranoside[39]	94.4	72.0	74.1	67.6	76.7	62.0		
synthetic α-D-mannopyranan	101.5	69.4	78.4	66.1	73.5	61.1		
synthetic α,β-D-mannopyranan (α-linkage)	101.7	69.4	78.6	66.0	73.6	61.1		
synthetic α,β-D-mannopyranan (β-linkage)	98.3	70.2	80.5	68.0	77.0	61.1		
acetylated α,β-D-mannopyranan (α-linkage)	99.5	69.9	75.2	67.2	71.4	62.2	20.8	170.5
acetylated α,β-D-mannopyranan (β-linkage)	95.8	69.9		67.9		62.2	20.8	170.6
nonreducing end group (in Figure 1A)		70.8	70.1	67.4				

Table II
Results of Polymerization[a]

mon no.[b]	cat.[c]	cat./mon mol %	solv	mon/solv, g/100 mL	temp, °C	time, h	yield, %	$[\alpha]^{23}_D$, deg	$M_{GPC} \times 10^{-3}$	$M_{osm} \times 10^{-3}$	DP	η, dL/g	free (1→3)-D-mannan $[\alpha]^{23}_D$, deg	free (1→3)-D-mannan α, %
I29	A	4.0	CH_2Cl_2	37.5	20	2	99	−48.0	41				+100	90
I33	A	4.0	CH_2Cl_2	37.5	−60	6	63	−47.2	20					
I38	A	4.3	benzene	30.0	20	16	90	−31.5	14	45.5	68	0.063		96
I39	A	3.5	toluene	30.0	20	3	96	−44.8				0.086		93
II12	A	2.8	CH_2Cl_2	40.0	20	1.0	85	+0.8	20					92
I17	A	3.0	CH_2Cl_2	40.0	−80	24	90	−6.9	18				+80	80
II20	A	2.8	benzene	30.0	20	16	92	+10.8	11.5	32.0	74	0.080	+120	100
II28	A	2.8	benzene	30.0	−10	16	98.5	−6.3	17.5	26.8	62	0.159	+91	85.5
II22	A	2.8	benzene	30.0	50	1	49	+14.4	6	18	42	0.063		100
II23	A	2.8	xylene	30.0	20	16	85	+6.9	11.7	32.9	76	0.123	+115	97.5
II26	A	2.8	xylene	30.0	50	1	80	+18.1	6.3					100
II24	A	2.8	toluene	30.0	20	16	81	+10.6	11.0	27.7	64	0.126		100
II25	A	2.8	toluene	30.0	50	1	72	+13.5	6.3	18	42	0.073		
I34	B	3.4	CH_2Cl_2	40	20	18	83.9	−47.3	20					95
II17	B	3.4	CH_2Cl_2	40	20	3	90.0	+2.8	18	39	90	0.145	+112	96
II21	B	2.9	benzene	30	20	16	66.0	0	11.5	31.4	73	0.110	+105	92.5
I26	C	2.0	CH_2Cl_2	35	20	20	97.0	−37.5	20	40	60	0.085	+120	100
I27	C	2.0	CH_2Cl_2	35	−25	20	97	−45.5	32	60	90	0.110	+102	91
II27	C	2.0	CH_2Cl_2	35	20	20	85	+16.5				0.055		
I9	D	4.0	CH_2Cl_2	50	−80	18	87.4	−44.3	13	25.1	37	0.062		91
I12	D	4.0	CH_2Cl_2	50	−50	6	94.4	−43.7	16					93
II5	D	4.0	CH_2Cl_2	50	−80	2.5	82.5	−6.2	16.5	24	56	0.153	+90	85
II6	D	1.9	CH_2Cl_2	50	−80	2.5	70.9	−11.1	22	31.8	74	0.176	+80	80
II13	D	1.9	CH_2Cl_2	50	−60	2.5	63.0	−2.2	14	22	51	0.150	+95.6	88
I2	E	1.0	CH_2Cl_2	50	−78	1	70	−46.3	62					
I3	E	1.0	CH_2Cl_2	50	−53	0.8	80	−46.0	80	120.0				92
II2	E	4.2	CH_2Cl_2	50	−60	3	87.4	−3.8	5.4			0.083		
II3	E	1.0	CH_2Cl_2	50	−60	4	63.0	−11.3	4.1	6.0		0.055		

[a] Amount of monomers for each polymerization: I, 150 mg; II, 200 mg. [b] Roman numeral I indicates bromobenzylated monomer, II indicated benzylated monomer. [c] Catalyst: A, $(CF_3SO_2)_2O$; B, $BrPh_3N^+SbCl_6^-$; C, $Ph_3C^+ClO_4^-$, D, $SbCl_5$; E, PF_5.

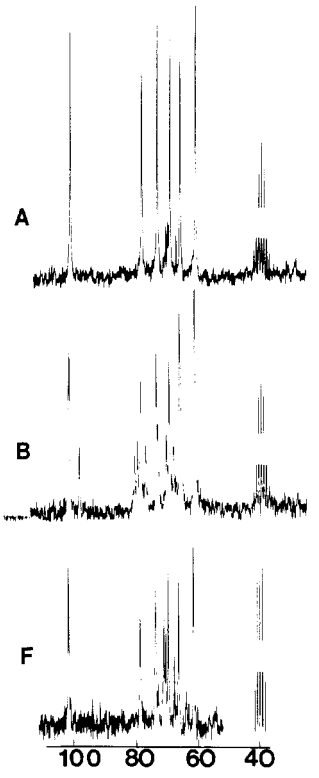
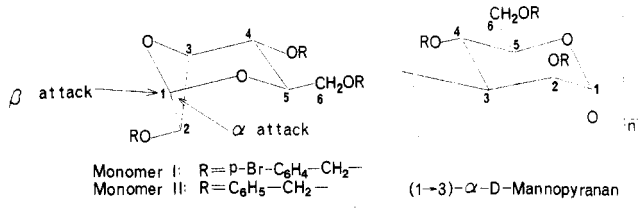


Figure 1. ^{13}C NMR spectra of (1→3)-D-mannopyranans in Me_2SO-d_6 at 70 °C: (A) DI26 (DP ~40, ~100% α); (B) DII6 (DP ~50, ~80% α); (F) DII6 (after acetylation, oxidation, deacetylation, and reduction).

number-average molecular weight, using chloroform as solvent for substituted polymers. A Mechrolab Model 501 high-speed membrane osmometer was also used for the measurements of some polymers of high molecular weight.

Results and Discussion

Monomers I and II were both readily polymerized by cationic catalysts to polysaccharide derivatives. Ten



representative samples with varying molecular weights and specific rotations were debenzylated or debromobenzylated and hydrolyzed to their constituent sugars. In all cases hydrolysis was complete to the monosaccharide stage and the only sugar present was mannose. Its 3-epimer, altrose, was specifically excluded. The only structural problem, therefore, was the assignment of configuration at the anomeric center.

The stereoregularity of free mannans was established by ^{13}C NMR spectra. The reported C-1 signal of (1→3)-α-D-mannopyranan, prepared from a complex heteropolymer of *Candida bogoriensis*,[41] was 103.8 ppm[42] (in D_2O containing 5% NaOD). To assign ^{13}C NMR spectra of the synthetic mannopyranans, α-D-mannopyranose and β-D-mannopyranose[39] served as model compounds. Spectra A and B (Figure 1) are the spectra of typical stereoregular (100% α-linkage) and nonstereoregular (~80% α-linkage) polymers. The assignments are in Table I.

For pure (1→3)-α-D-mannopyranan, as expected, C-3 had a large downfield shift, 7.1 ppm, and C-2 and C-4 had

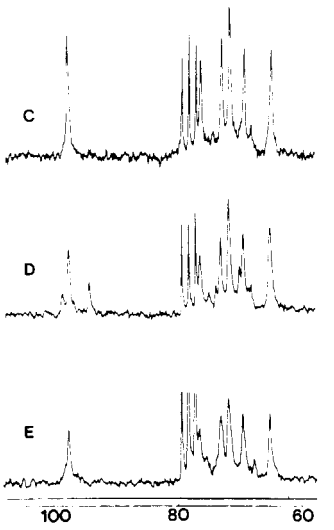


Figure 2. ^{13}C NMR spectra of fully acetylated (1→3)-D-mannopyranans in $CDCl_3$ at 20 °C: (C) DII26 (DP ~20, 100% α); (D) DII6 (~80% α, before oxidation); (E) DII6 (after oxidation).

upfield shifts of 2.3 and 1.8 ppm, respectively, compared with the model compound α-D-mannopyranose.[39] C-1 (101.5 ppm) is upfield of the reported value[42] as our value is determined in dimethyl sulfoxide. The small peaks at 70.8, 70.1, and 67.4 ppm in spectrum A were assigned to C-2, C-3, and C-4 of nonreducing end groups, respectively, by comparison with the ^{13}C spectra of 3-*O*-α-D-mannopyranosyl-D-mannopyranose[40] and methyl-3-*O*-α-D-mannopyranosyl-α-D-mannopyranoside.[37]

The assignment of β-linked units of the synthetic mannopyranan was accomplished by comparing spectra A and B. As expected from the model compounds α- and β-D-mannopyranose, C-3 and C-5 of β-linked polymer had large downfield shifts of 2.1 and 3.5 ppm, respectively, compared with the corresponding C-3 and C-5 of purely α-linked polymer. It was unexpected that C-4 of β-linked units had a downfield shift of 1.9 ppm and C-1 had an upfield shift of 3.2 ppm compared with the corresponding C-4 and C-1 of pure α-linked polymers. The peak at 79.6 ppm was assigned to C-3 (α-linkage) adjacent to a (1→3)-β-linked unit.

The C-1, C-3, C-4, and C-5 peak positions of α-linked mers in (1→3)-α,β-D-mannopyranan were little different from the corresponding peaks in pure (1→3)-α-D-mannopyranan.

The assignments above were generally confirmed by Lindberg's method of selective oxidation of fully acetylated polysaccharides. Spectrum C (Figure 2) is one of an acetylated stereoregular (1→3)-α-D-mannopyranan having the same pattern as spectrum A. After oxidation the ^{13}C NMR spectrum did not change. Spectra D and E (Figure 2) are spectra of acetylated nonstereoregular mannopyranan before and after oxidation, respectively. It can be seen that all of the peaks related to β-linkages disappeared after oxidation. (Three solvent peaks are prominent at 78.5, 77.2, and 76.0 ppm.)

On the basis of spectra C, D, and E, we assigned the ^{13}C NMR spectra of fully acetylated (1→3)-D-mannopyranans as shown in Table I. C-3 and C-5 of β-linked units were not assigned because they overlap the solvent ($CDCl_3$) peaks or the end group peaks. The small peak at 100.6 ppm near C-1 (α-linkage) in spectrum D was assigned to C-1 (α-linkage) adjacent to a β-(1→3)-linked unit.

More clear information was obtained from spectrum F (Figure 1) of the reduced and then deacetylated polymer after oxidation. Comparing spectrum B (before oxidation)

and spectrum F (after oxidation) we confirmed the assignment of ^{13}C NMR spectra of the synthetic mannopyranans as shown already in Table I. The extra peaks near C-6 and the intensity increase of C-2, C-4, and C-3 peaks of end groups were attributed to the degradation and the existence of an open chain unit after the sequence of reactions.

Since both spectra B and D show that the peaks adjacent to β-linked units have almost the same intensity as the peaks of corresponding β-linked units, the β-linkage in nonstereoregular (1→3)-D-mannopyranan is presumably randomly distributed in the polymer rather than in sequence. The product obtained after oxidation, reduction, and deacetylation was a collapsed glassy solid and its ^{13}C NMR spectrum contained enhanced end groups. Both features are characteristic of the expected oligomers and confirm this interpretation.

^1H NMR spectra of the synthetic mannopyranans showed the same pattern at the anomeric center as the ^{13}C NMR spectra. The chemical shifts of H-1 (α-linkage) and H-1 (β-linkage) were 5.0 and 3.4 ppm, respectively.

The fact that no peaks were found in the aromatic region of either the ^{13}C or the ^1H spectra indicated that debenzylation and debromobenzylation were complete.

Knowing the exact ^{13}C NMR peak positions of α- and β-linkages of the synthetic mannopyranans, we estimated the stereoregularity of the polymers by the average ratio of the peak areas of α- and β-linkages at the anomeric centers and at C-3 and C-5. The optical rotation of the unsubstituted mannans varied linearly with our estimate of stereoregularity. Stereoregular mannopyranans (Table II, DI26 and DII20), judged 100% α from ^{13}C and ^1H NMR spectra, had specific rotation of +120°, the same as the reported value.[9] The specific rotation of pure (1→3)-β-D-mannopyranan was then estimated to be −81° by linear extrapolation of the specific rotations of the nonstereoregular mannans. This value is reasonable because the model compounds, methyl-β-D-mannopyranoside[9] and an alternating polymer of (1→3)-β- and (1→4)-β-D-mannopyranan,[20] had optical rotations of −70° and −80°, respectively.

Similarly there was a linear correlation of specific optical rotations between substituted and free mannans except in the case of low molecular weight products. Therefore, the stereoregularity of the synthetic mannans could also be estimated from the specific rotation directly on the polymerization products. High molecular weight bromobenzylated polymers with specific rotation of −38° and benzylated polymers with specific rotation of +10° are stereoregular while more negative values indicate some β-linkages. Low molecular weight stereoregular polymers, judged from ^{13}C NMR spectra, such as II26 (+18.1°), II22 (+14.4°), and II25 (+13.5°) did not follow this relationship.

The absolute number-average molecular weight of the substituted mannans, determined by osmometry (M_{osm}), ranged from 6000 to 39 000 for benzylated mannans and from 15 000 to 120 000 for bromobenzylated mannans.

The number-average molecular weight of substituted mannans was also determined by gel permeation chromatography (GPC) using polystyrene standards (M_{GPC}). It was found that the values of M_{GPC}, which are actually related to the polymer volume, were much smaller than the values of number-average molecular weight (M_{osm}) for the same polymers. The ratio M_{osm}/M_{GPC}, which is inversely related to the volume and independent of the absolute molecular weight of the polymer, changed regularly with stereoregularity: the more stereoregular the polymer, the smaller its molecular volume. Apparently polymers having high stereoregularity can pack more tightly because of their structural regularity, while polymers having structural flaws are relatively loosely packed because of (1→3)-β-linkages. Although the relationship was not quantitative, it still could serve as an ancillary method to judge the stereoregularity of the polymers obtained from same solvent. Figure 3 indicates this relation for some polymers obtained from dichloromethane. Polymers having the same stereoregularity but obtained from polymerizations carried out in different solvents have different ratios of M_{osm}/M_{GPC}. Usually it was much bigger from aromatic solvents than from dichloromethane. The effect of polymerization solvent on the structure of the polymer is an interesting puzzle. It may relate to the randomness of distribution of β-linkages.

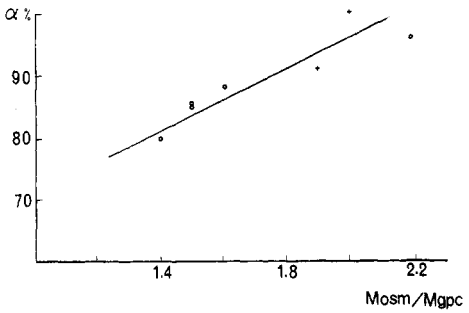


Figure 3. Relation between stereoregularity and M_{osm}/M_{GPC}: (O) benzylated polymer, (+) bromobenzylated polymer. Points in sequence (left to right) are samples II6, II5, II28, II13, I27, I26, and II17.

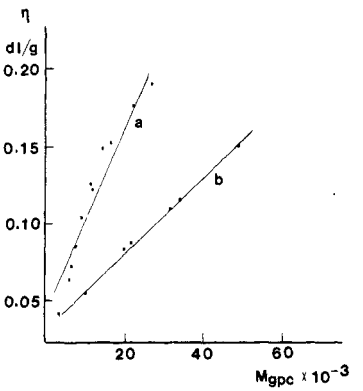


Figure 4. Relation between intrinsic viscosity and M_{GPC}: (a) benzylated polymer; (b) bromobenzylated polymer.

The molecular weight distribution of the substituted polymers was determined by GPC. It was found that all had a relatively narrow and essentially symmetrical molecular weight distribution. The calculated values of M_w/M_n from GPC curves of several representative samples were from 1.4 to 1.6.

The intrinsic viscosity of the substituted polymers was low and ranged from 0.05 to 0.2 dL/g, with bromobenzylated polymers having lower intrinsic viscosities than the corresponding benzylated polymers. Intrinsic viscosity correlated well with the M_{GPC} of both stereoregular and nonstereoregular polymers but not with their M_{osm} because of the variation in molecular volume with stereoregularity. Since all had very similar molecular weight distributions, this correlation holds for either number- or weight-average molecular weights (Figure 4). Infrared and circular dichroism spectra could not be used to distinguish the stereoregularity of free and substituted mannans.

About 60 polymerizations were carried out and some of the results are listed in Table II. It was found that stereoregular polymerization could be achieved with either

monomer under appropriate conditions of catalyst, solvent, and temperature.

Monomers I and II behaved similarly under most conditions although benzylated monomer II usually polymerized faster and gave polymers with higher DP than bromobenzylated monomer I. The opposite was true of polymerizations catalyzed by phosphorus pentafluoride (Table II, I3 and II3). The stereoregularity in most cases was not greatly different between the two monomers. However, all of the polymers obtained from monomer I were more than 90% α-linked, while the stereoregularity of the polymers obtained from monomer II was 80% in some cases.

Solvent used in polymerization had an obvious influence on the stereoregularity and molecular weight of the polymers. Aromatic solvents gave higher stereoregularity than dichloromethane in the polymerizations of both monomers using $(CF_3SO_2)_2O$ as catalyst (Table II, cf. I38 and I29; cf. II20 and II12). The disadvantage of using aromatic solvents was that the polymer molecular weight and the reaction rate decreased (Table II, cf. I29 and I38; cf. II17 and II21) because the polymerizations were carried out at low concentrations due to decreased solubility of the monomers in aromatic solvents. Benzene was the best of the aromatic solvents used (Table II, cf. I38 and I39; cf. II20 and II23).

Temperature had a dramatic influence on both the stereoregularity and the molecular weight of the polymers. Usually ambient temperatures gave good stereoregularity and high molecular weight (Table II, I26, II20, II24, and II27). Lowering temperature usually increased molecular weight but decreased stereoregularity (Table II, cf. II20 and II28; cf. II13 and II6; cf. I26 and I27, etc.). When the polymerization temperature was elevated to 50 °C, however, it caused a serious decrease of molecular weight due to termination and chain-transfer reactions, although the high stereoregularity remained (Table II, cf. II20 and II22; cf. II23 and II26).

Different catalysts have different catalytic activity. Usually the cationic catalysts capable of generating a cation in a single initiation stage, like tris(4-bromophenyl)-amminium hexachloroantimonate and trifluoromethanesulfonic anhydride, gave polymers in higher yield with higher stereoregularity and molecular weight than Lewis acids involving complexation at the initiation stage, like boron trifluoride and antimony pentachloride. The best cationic catalysts for stereoregular polymerization were trifluoromethanesulfonic anhydride, triphenylcarbenium (trityl) perchlorate, and tris(4-bromophenyl)amminium hexachloroantimonate. Trifluoromethanesulfonic anhydride, which had been used as an initiator for tetrahydrofuran,[36] was an effective catalyst for both monomers at room temperature in benzene (II20, I38). Trityl perchlorate, introduced by Bredereck and Hutten,[44] caused completely stereoregular polymerization at room temperature in dichloromethane for both monomers. Tris(4-bromophenyl)amminium hexachloroantimonate, which has been used for the Diels–Alder reaction,[45] proved to be a good catalyst for stereoregular polymerization at room temperature in dichloromethane for both monomers.

Phosphorus pentafluoride, boron trifluoride, and antimony pentachloride, which have been shown to be good initiators for 1,6-anhydro sugar,[6] 1,5-anhydro-β-D-xylofuranose,[43] and 1,5-anhydro-β-D-ribofuranose[24] derivatives, did not cause stereoregular polymerization although phosphorus pentafluoride afforded the polymer of highest molecular weight. Coordination catalysts that have been used for the polymerization of epoxides[38,46] and oxetane[22] were not attractive in the present research because the polymerization rate was too slow. Anionic catalyst potassium *tert*-butoxide did not cause polymerization of either monomer at 60 °C.

In summary, the best conditions for stereoregular polymerization were those of runs I26, II20, and II17 as indicated in Table II.

The synthetic (1→3)-D-mannopyranans were fluffy white solids after freeze-drying from water. They absorbed water from the atmosphere avidly, reaching equilibrium quickly. The solubility of the synthetic mannans depended on their stereoregularity. Pure (1→3)-α-D-mannopyranan was almost insoluble in water after precipitating during dialysis, while less stereoregular mannans were somewhat soluble in water because of the existence of a more disordered structure. All of the synthetic mannans were soluble in dimethyl sulfoxide. We estimated the degree of polymerization of the synthetic mannans (DI26, DII17, DII7, and DII20, etc.) to be about 30–60 from ^{13}C spectra of the polymers.

The preceding evidence indicates that they are appropriate models of naturally occurring substances and can serve in biological and immunological research.

Acknowledgment. This research is completed with financial support from Research Grant GM-06168 from the Division of General Medical Sciences, National Institutes of Health. We gratefully acknowledge the assistance of Dr. R. Eby in the determination of ^{13}C and ^{1}H NMR spectra. Preliminary observations of polymerization by A. J. Varma and disaccharide spectra from F. Good and L. F. Awad are also acknowledged.

References and Notes

(1) Schuerch, C. *Acc. Chem. Res.* **1973**, *6*, 184–191.
(2) C. Schuerch "Advances in Carbohydrate Chemistry and Biochemistry"; Tipson, E. S., Horton, D., Eds.; Academic Press: New York, 1982; Vol. 39, p 157–212.
(3) Sumitomo, H.; Okada, M. *Adv. Polym. Sci.* **1978**, *28*, 47–82.
(4) Micheel, F.; Brodde, O. E.; Reinking, K. *Justus Leibigs Ann. Chem.* **1974**, 124–136.
(5) Uryu, T.; Koyama, Y.; Matsuzuki, K. *J. Polym. Sci., Polym. Lett. Ed.* **1979**, *17*, 673–678.
(6) Fréchet, J.; Schuerch, C. *J. Am. Chem. Soc.* **1969**, *91*, 1161–1164.
(7) Sondheimer, S.; Yamaguchi, H.; Schuerch, C. *Carbohydr. Res.* **1979**, *74*, 327–332.
(8) (a) Mitchell, W. O.; Hasenclever, H. F. *Infec. Immun.* **1970**, *1*, 61–63. (b) Okubo, Y.; Shibata, N.; Matsumoto, T.; Suzuki, M.; Schuerch, C.; Suzuki, S. *J. Bacteriol.* **1980**, *144*, 92–96.
(9) Bhattacharjee, A. K.; R-Chung, K. J.; Glaudemans, C. P. T. *Carbohydr. Res.* **1980**, *82*, 103–111.
(10) Bhattacharjee, A. K.; R-Chung, K. J.; Glaudemans, C. P. T. *Carbohydr. Res.* **1979**, *73*, 183–192.
(11) Elinov, N. P.; Abelian, V. A.; Rustova, N. V. *Mikol. Fitopatol.* **1981**, *15*(5), 394–398.
(12) Hara, C.; Kiho, T.; Ukai, S. *Carbohydr. Res.* **1982**, *111*, 143–150.
(13) Fraser, C. G.; Jennings, H. J.; Moyna, P. *Can. J. Biochem.* **1973**, *51*, 219–224.
(14) Cifonelli, J. A.; Smith, F. *J. Am. Chem. Soc.* **1955**, *77*, 5682–5684.
(15) Bishop, C. T.; Blank, F.; Yardner, P. E. *Can. J. Chem.* **1960**, *38*, 869–880.
(16) Haworth, W. N.; Hirst, E. L.; Isherwood, F. A. *J. Chem. Soc.* **1937**, 784–791.
(17) Haworth, W. N.; Heath, R. L.; Peat, S. *J. Chem. Soc.* **1941**, 833–842.
(18) Y. Fukazawa Presented at the U.S.–Japan Seminar on Synthetic Polysaccharides and their Biochemical Functions, Syracuse, New York, Oct. 8–12, 1979.
(19) Naccache, V. M.; Dietrich, S. M. C. *Rev. Microbiol.* **1981**, *12*, 61–64.
(20) Gorin, P. A. J.; Horitsu, K.; Spencer, J. F. T. *Can. J. Chem.* **1965**, *43*, 950–954.
(21) Merle, J. P.; Sarko, A. *Carbohydr. Res.* **1973**, *30*, 390–394.
(22) Saegusa, T.; Imai, H.; Furukawa, J. *Makromol. Chem.* **1963**, *65*, 70–73.

(23) Yamaguchi, H.; Schuerch, C. *Biopolymers* **1980**, *19*, 297-309.
(24) Uryu, T.; Kitano, K.; Ito, K.; Yamanouchi, J.; Matsuzaki, K. *Macromolecules* **1981**, *14*(1), 1-9.
(25) Ito, H.; Eby, R.; Kramer, S.; Schuerch, C. *Carbohydr. Res.* **1980**, *86*, 193-202.
(26) Ito, H.; Schuerch, C. *Macromolecules* **1981**, *14*, 246-249.
(27) Varma, A. J.; Schuerch, C. *J. Org. Chem.* **1981**, *46*, 798-803.
(28) Kong, F.; Schuerch, C. *Carbohydr. Res.* **1983**, *112*, 141-147.
(29) Uryu, T.; Schuerch, C. *Macromolecules* **1971**, *4*, 342-347.
(30) Lin, J. W. P.; Schuerch, C. *J. Polym. Sci., Part A-1*, **1972**, *10*, 2045-2060.
(31) Hoffman, J.; Lindberg, B.; Svensson, S. *Acta. Chem. Scand.* **1972**, *26*, 661-666.
(32) Lindberg, B.; Lindgvist, B.; Lonngren, J.; Nimmich, W. *Carbohydr. Res.* **1977**, *58*, 443-451.
(33) Fengel, D.; Wegener, G. Spring meeting of the cellulose paper, and textile division, American Chemical Society, paper no. 17, Appleton, Wisconsin, May 17-19, 1978.
(34) Mukherjee, S.; Marchessault, R. H.; Sarko, A. *Biopolymers* **1972**, *11*, 291-301.
(35) Dauben, H. J.; Honnen, L. R.; Harmon, K. M. *J. Org. Chem.* **1960**, *25*, 1442-1445.
(36) Smith, S.; Hubin, A. J. *J. Macromol. Sci., Chem.* **1973** *A7*, 1399.
(37) Good, F.; Eby, R.; Sarko, A., unpublished results from this laboratory.
(38) Vandenberg, E. J. *J. Polym. Sci.* **1960**, *47*, 486-489; *J. Polym. Sci., Part A-1* **1969**, *7*, 525-567.
(39) Stothers, J. B. "Organic Chemistry"; Blomquist, A. T., Wasserman, H., Eds.; Academic Press: New York and London, 1972; Vol. 24, p 461.
(40) Awad, L. F.; Schuerch, C., unpublished results from this laboratory.
(41) Gorin, P. A. J.; Spencer, J. F. T. *Can. J. Chem.* **1968**, *46*, 3407-3411.
(42) Gorin, P. A. J. *Can. J. Chem.* **1973**, *51*, 2375-2378.
(43) Uryu, T.; Yamanouchi, J.; Hayashi, S.; Tamaki, H.; Matsuzaki, K., in press.
(44) Bredereck, H.; Hutten, V., unpublished data cited in: Klar, J. *Chem.-Ztg.* **1963**, *87*, 731-740.
(45) Bellville, D. J.; Wirth, D. D.; Bauld, N. L. *J. Am. Chem. Soc.* **1981**, *103*, 718-720.
(46) Hirano, T.; Nakayama, S.; Tsuruta, T. *Makromol. Chem.* **1975**, *176*, 1897-1800.

Synthesis and Free Radical Polymerization of Methyl α-Benzylacrylate

Enrique L. Madruga,*† Julio San Román,† M. Angeles Lavia,† and M. Carmen Fernandez Monreal‡

Instituto de Plasticos y Caucho, Juan de la Cierva 3, Madrid-6, Spain, and Departamento de Quimica Orgânica, Universidad Complutense, Madrid, Spain. *Received March 30, 1983*

ABSTRACT: Methyl α-benzylacrylate was synthesized from dimethyl malonate following well-known organic reactions. The purified monomer was polymerized by a free radical mechanism in benzene, using 2,2′-azobis(isobutyronitrile) as initiator, at several temperatures ranging from 40 to 80 °C. The absence of polymeric species at polymerization temperatures of 70 and 80 °C, together with the low molecular weight of the polymers obtained, seems to indicate that a ceiling temperature between 60 and 70 °C may be expected. The application of results obtained to known kinetic treatments gives a ceiling temperature of 67 ± 2 °C under the experimental conditions used in this work.

Introduction

Polymerization of methyl and other alkyl methacrylates has been reported by many authors; however, little attention has been paid to the free radical polymerization of α-alkyl- or α-arylacrylates. Attempts to initiate the polymerization of α-alkyl- or α-arylacrylic esters resulted in either low molecular weight polymers or no polymer at all,[1,2] which was attributed to the strong interfering effect exerted on free-radical polymerization by the α-substituent group.

From a thermodynamic point of view, the free energy of polymerization mainly depends on the enthalpy of polymerization, since the entropy of polymerization presents values relatively constant or with little variation in a narrow range and is less sensitive than the enthalpy of polymerization to the chemical structure of monomers. As it is well-known, the enthalpy of polymerization is low for α,α-disubstituted monomers and therefore free energy of polymerization is less negative and the polymerization reaction more difficult.[3] Thus, free-radical polymerization of methacrylate monomers containing phenoxy groups as substituents on the α-methyl position with relative low yield and molecular weights has been reported by Lenz et al.[4] and the results have been attributed to the low ceiling temperatures of polymerization expected for these sterically hindered monomers. However, in our knowledge, no ceiling temperatures for this family of monomers have been reported and it may be interesting to know this parameter in order to gain more information about the reactivity of this kind of vinyl monomers. The present report is concerned with the synthesis, free-radical polymerization, and ceiling temperature determination of methyl α-benzylacrylate.

Experimental Section

Synthesis of Monomer. The synthesis of monomer involved three steps and well-characterized organic reactions were used in all of them.[5,6]

Step 1. Dimethyl benzylmalonate was prepared from dimethyl malonate (0.95 mol) and sodium methoxide (0.87 mol) in 1450 mL of methanol and benzyl chloride (0.91 mol). The reaction mixture was warmed until neutralization. The product was fractionated under vacuum and collected at 118-122 °C at 2 mmHg. A 42.2% yield was obtained.

Step 2. Monomethyl Ester of Benzylmalonic Acid. To a solution of potassium hydroxide (0.425 mol) in 145 mL of methanol was added 0.425 mol of dimethyl benzylmalonate in 560 mL of dried methanol. After standing for about 24 h, the solution was acidified with 25 mL of HCl (35%) and the monoester was extracted with ether. A 91.3% yield was obtained.

Step 3. Methyl α-Benzylacrylate. Diethylamine (0.37 mol) was added to monomethyl ester of benzylmalonic acid (0.37 mol) and the mixture was kept in an ice bath with stirring while 38 mL of formaldehyde (35% in methanol–water solution) was slowly

† Instituto de Plasticos y Caucho.
‡ Universidad Complutense.

0024-9297/84/2217-0989$01.50/0