METHYL α- AND β-D-IDOPYRANOSIDURONIC ACIDS SYNTHESIS AND CONFORMATIONAL ANALYSIS

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ABSTRACT

Methyl α- and β-D-idopyranosiduronic acid have been synthesized by catalytic oxidation of the corresponding D-idopyranosides, and each has been characterized as a crystalline brucinium salt. Proton and ^{13}C magnetic resonance data indicate that, in solution, the α anomer adopts a conformation represented mainly by the *CI*(D) form and the β anomer favors this conformation almost exclusively. Possible relationships between these findings and the conformation of α-L-idopyranosyluronic acid residues in heparin and dermatan sulfate are discussed, as well as the optical rotations of these polymers.

INTRODUCTION

In conjunction with recent p.m.r.-spectral studies of heparin and other mucopolysaccharides[1,2], it became desirable to examine derivatives of idopyranosyluronic acid as pertinent reference-compounds. The present paper reports the synthesis of the methyl α- and β-glycopyranosides of the D-acid (**1** and **2**, respectively), and considers their conformations in solution.

RESULTS AND DISCUSSION

For the synthesis of **1** and **2**, advantage was taken of the elegant routes devised by Sorkin and Reichstein[3] for conversion of D-galactose into methyl 4,6-*O*-benzylidene-α and β-D-idopyranosides, respectively, and their procedures were used with only slight modification. These acetals were then converted into the corresponding, syrupy glycosides by mild hydrolysis with acid; because the idosides are themselves labile, this step required carefully controlled conditions in order to combine optimal removal of the acetal substituent with minimal hydrolysis of the glycoside[4,5]. Of the two, the β anomer was the less stable. The hydrolysis mixture, containing some 1,6-anhydro-β-D-idopyranose formed as a by-product[4], was then fractionated by preparative chromatography. Catalytic oxidation with platinum and oxygen[6,7] at

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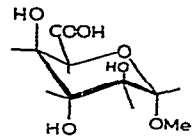
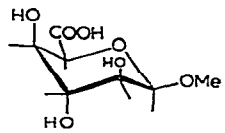


1a



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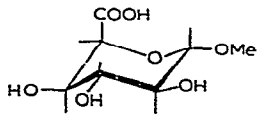

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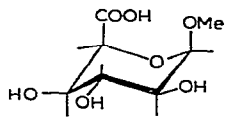

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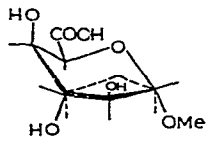
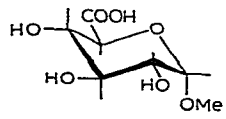


1c



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pH 7–8 was highly effective in converting the glycosides into the uronic acids; these (**1** and **2**) were obtained as sodium salts, and were characterized more fully as their crystalline brucinium salts, m.p. 172° and 167°, respectively.

A study of the n.m.r. spectra of **1** and **2** (as sodium salts in deuterium oxide) has been conducted as a possible source of information about the type of conformation favored by L-idopyranosyluronic acid moieties of heparin and dermatan sulfate. Although both anomers were considered in this context, the α-L anomer is the more relevant in considering dermatan sulfate[8,9] and, as shown recently, heparin[10,11] as well. Additional comments on configuration are made later.

The spectrum of the sodium salt of methyl β-D-idopyranosiduronic acid (**2**) (see Fig. 1B) is the more straightforward. At 100 MHz, it is close to a first-order type. The assignments listed in this Figure have been confirmed by spin-decoupling and, with the good separation of signals obtained, the observed spacings are taken as a reliable measure of the coupling constants (see Table I). In view of their small magnitude, these spacings clearly favor assignment of the *C1*(D) conformation (**2a**) for the β anomer, because *anti* orientation of the ring protons in the alternative conformation **2b** should give rise to coupling constants of 9 to 10 Hz throughout[12,13]. Strongly reinforcing this assignment is the observed long-range coupling of about 2 Hz between H-2 and H-4, which is ascribed to a "W" arrangement of bonds[14,15]. That is, the segment of the molecule encompassing H-2, C-2, C-3, C-4, and H-4 can be represented by a planar, zigzag arrangement, which is inherent in the *C1*(D) conformation (**2a**) but not in the alternative chair conformation **2b**.

The p.m.r. spectrum of the sodium salt of methyl α-D-idopyranosiduronic acid (**1**) was measured at 220 MHz in order to ensure an optimal separation of signals, especially because those for H-3 and H-4 overlap at 100 MHz (see Fig. 1A). As the

splitting patterns are virtually unchanged by this increase in frequency, the spacings measured at 220 MHz were taken to be the coupling constants (see Table I). As also shown in this Table, splitting is affected slightly by temperature.

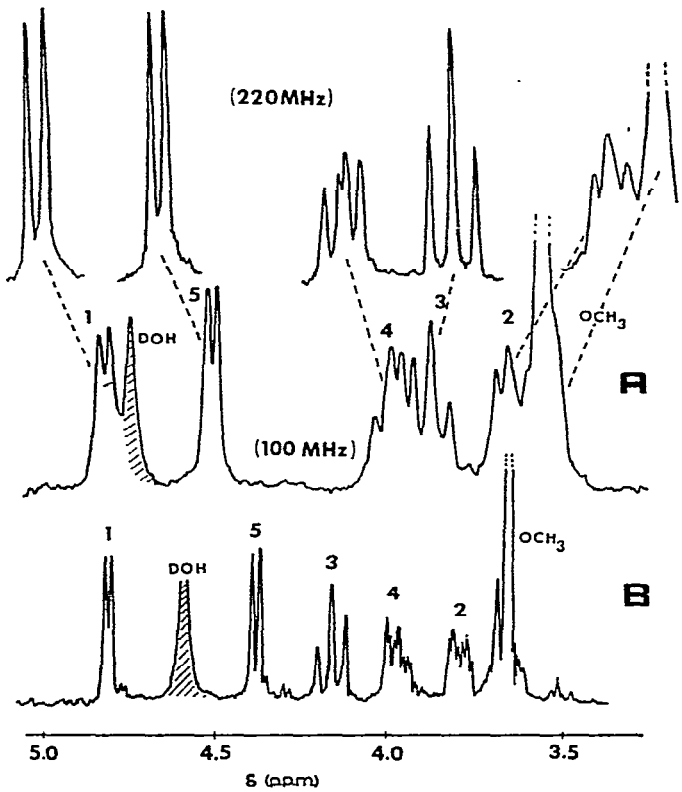

Fig. 1. P.m.r. spectra, for solutions in deuterium oxide, of (A) methyl α-D-idopyranosiduronic acid (1) (sodium salt) at 100 MHz and 25° (lower) and at 220 MHz and 40° (inset), except that signals for H-2 and OCH_3 were recorded at 15°; and (B) methyl β-D-idopyranosiduronic acid (2) (sodium salt) at 100 MHz and 40°.

TABLE I

VICINAL COUPLING-CONSTANTS (Hz) FOR METHYL α- AND β-D-IDOPYRANOSIDURONIC ACIDS (1 AND 2)[a]

Anomer	*Temp. (degrees)*	$J_{1,2}$	$J_{2,3}$	$J_{3,4}$	$J_{4,5}$	$J_{2,4}$
α[b]	15	3.3	5.5	5.5	3.0	
	40	4.0	6.0	6.0	3.5	
	85	4.5	6.0	6.0	5.0	
α[c]	40	3.0	4.8	4.8	2.8	
β[d]	40	1.9	3.5	3.5	2.3	1.2

[a]Sodium salt in deuterium oxide. [b]Measured from the 220-MHz spectrum. [c]Measured from the 100-MHz spectrum, pH <2. [d]Measured from the 100-MHz spectrum.

Accepting that the spectral characteristics of the β-D anomer are representative of the *Cl*(D) conformation, the data for the α-D anomer must signify a significant departure from this chair conformation. That is, the vicinal couplings are larger throughout, and long-range coupling (such as is observed in Fig. 1B) is absent. Possibly, therefore, this spectrum indicates time-averaging between the two chair conformations (**1a** and **1b**), with **1a** preponderating, but becoming relatively less important at elevated temperatures. Another possible contributor to the conformation of **1** is a skew form in which departure from the *Cl*(D) conformation takes place along the C-1, C-2, and C-3 bonds as represented by **1c** [broken lines denote the *Cl*(D) form]. Because a small change in geometry (*e.g.*, of H–C–C bond angles, θ) may have a relatively large effect[13] on the *J* value, these larger coupling-constants of **1** (than of **2**) need not signify substantially different conformations for the anomers. However, chemical-shift data also provide an indication that their conformations differ notably.

Protons of **1** and **2**, aside from H-1, resonate downfield from the protons of methyl α-D-glucopyranosiduronate (**3**). This is seen by comparing the chemical shifts given in Figs. 1A and 1B with the following data for the sodium salt of **3**; such a comparison of **1** with **3** is facilitated by the "stick" diagram presented in Fig. 2.

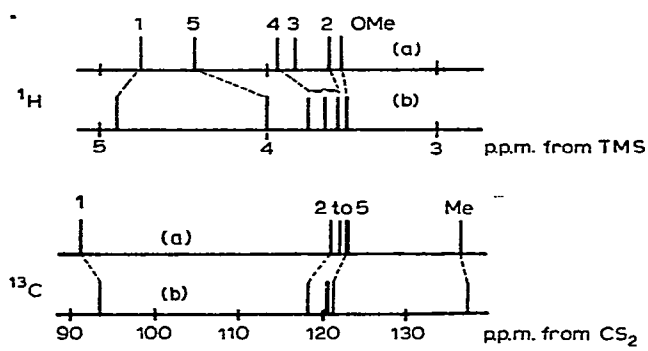

Fig. 2. ^{1}H and ^{13}C chemical-shift data for the sodium salt of (a) methyl α-D-idopyranosiduronic acid (**1**) and (b) methyl α-D-glucopyranosiduronic acid (**3**).

Hence, assuming a *Cl*(D) conformation (or a preponderance thereof) for compounds having the D-*ido* as well as those having the D-*gluco* configuration, this enhanced deshielding may be ascribed[12] to the larger number of equatorially attached protons (on C-2, C-3, and C-4) present in the D-*ido* isomers.

An additional factor to be considered, and one that may also account for the relatively large shifts of H-5 of **1** and **2** (as compared with that of **3**) and of the axial H-1 of **2**, is the multiplicity of destabilizing interactions in **1** and **2**. As noted recently for aldoses and aldosides[16,17], destabilizing interactions appear to cause a change in polarization of the C–H bonds, promoting increased shielding of ^{13}C nuclei[18,19] and concomitant deshielding of the appended protons. Therefore, the unfavorable combination of several axial C–O bonds in the D-*ido* compounds should contribute

to the large chemical-shifts observed for all, or most, of the protons*. However, it is noteworthy that H-1 of the α-D-glucopyranosiduronate **3** is more deshielded than is H-1 of the α-D-*ido* isomer **1**, despite the expectation that steric compression due to the O-1,O-3 diaxial interaction in **1** should outweigh the O-1,O-2 gauche interaction in **3**.

Further information on these points was obtained by comparing the ^{13}C spectra of **1** and **3**. At the anomeric center, as shown in Fig. 2, ^{13}C-1 of **3** is the more shielded, matching the fact that its attached H-1 is the less shielded, and the $O^{13}CH_3$ of **3** is also the more shielded (the *O*-methyl protons are less affected), an interplay of effects that accords with the earlier observations on methyl aldopyranosides[16]. By contrast, and in agreement with what has just been stated, C–H groups 2 to 5 show in general the inverse relationship; *i.e.*, for the α-D-*ido* isomer, the ^{13}C nuclei are more shielded, and the protons less shielded, in comparison with the α-D-*gluco* isomer. We regard these results as further evidence that **1** departs significantly from the *C1*(D) conformation. In particular, it appears that its 1-methoxyl group is so oriented as to minimize interaction with other substituents, and the fact that H-3 of the α anomer **1** is much more shielded than H-3 of the β anomer (see Fig. 1) suggests that O-3, as well as O-1, interacts less severely than it would in a fully puckered, *C1*(D) form.

For some years, there has been strong evidence to the effect that the two chair conformations of idopyranose do not differ greatly in free energy[21–23]. Nevertheless, it has been demonstrated recently that penta-*O*-acetyl-α-D-idopyranose exists in the *C1*(D) conformation, probably exclusively so in chloroform at ambient temperatures[24]. The current findings also deviate in this way from expectation, although **1** exhibits less conformational purity than does the pentaacetate. As ΔF for an ionized carboxyl group is close to that of a CH_2OR group[25], whereas the 1-methoxyl group is associated with a smaller anomeric effect than is the 1-acetoxyl group[26], this latter difference should minimize the tendency of **1** to assume conformation **1a** relative to the adoption by the pentaacetate of the *C1*(D) conformation. On the same basis, conversion of **1** into the free acid should cause[25] a decrease in ΔF for the carboxyl group of ~1 kcal, and hence further lessen the tendency for **1a** to be favored. In fact, however, acidification causes a substantial decrease in the coupling constants for **1** (see Table I), whereas the effect on **2** is negligible, which implies that, as the free acid, **1** is even more truly represented as the *C1*(D) conformer.

Comparing the α-D-idosiduronic acid in turn with its β anomer, it appears that instability due to an O-1,O-3 diaxial interaction in **1a** is greater than that engendered by the absence of an anomeric effect and the presence of an O-1,O-2 gauche interaction in favored conformer **2a**. The greater overall tendency of **1** and **2** to assume the *C1*(D) rather than the alternative chair conformation may also be related to some stabilizing factor in these molecules that has yet to be recognized[24]. Although the conformation of partially substituted sugars can be stabilized by intramolecular hydrogen-bonding between 1,3-diaxial hydroxyl groups, a possibility of this kind in **1** and **2**, caused by

*This also should be anticipated on the basis of the proton-shift relationships deduced by Lemieux and Stevens[20].

the presence of molecules of solvent water appears less likely. However, it is conceivable that water may play a stabilizing role through a "bridge" type of hydrogen-bonding, *viz.*, –O····H–O–H····O–, between the 1,3-diaxially disposed oxygen atoms.

These findings for methyl α-D-idopyranosiduronic acid do not necessarily imply that the α-L-idopyranosyluronic acid residues in heparin or dermatan have the corresponding conformation. In the polymers, polar and nonbonded interactions between groups on adjacent residues along the chain, as well as solvation differences on passing from monomer to polymer, might alter the stability of the conformation favored by the intact monosaccharide derivative. Although it has not proved possible to derive coupling constants for the protons of heparin and dermatan, because of signal broadening associated with polymeric characteristics, the shapes and chemical shifts of signals in the spectra of both mucopolysaccharides suggest that there is a close affinity between the conformation of the uronic acid residues present and that of the α-L-*ido* compound (mirror image of **1**). This applies particularly to signals due to H-1 and H-5 which, especially clearly at 220 MHz, are found to have line widths and chemical shifts close to those of the corresponding signals for **1**, as has already[2] been noted*. Molecular models suggest that an "acceptable" helix having a long pitch to the thread can be constructed to represent heparin built up from alternating[11,28] α-L-idopyranosyluronic acid [*1C*(L) conformation] and 2-amino-2-deoxy-α-D-glucopyranosyl [*C1*(D)] residues. A more drastic, periodic folding of such a helix may be envisaged if the α-D-glucopyranosyluronic acid residues[27], present in heparin in small proportion, occur at infrequent intervals along the polymeric chain.

The anomeric configuration of the L-idopyranosyluronic acid residues of heparin and dermatan sulfate has already been touched on. Previously, the rotatory power of heparin was regarded as anomalously low[9], but this conclusion was based on the assumption that D-glucuronic acid constitutes half of the polymer residues, whereas recent evidence points to a great preponderance of L-iduronic acid residues[2,10,11,28]. With the α-D-glycoside **1** available, an approximate value for the specific rotation of heparin has been calculated, taking into account the admittedly incomplete, compositional information thus far acquired; a value of $\sim +44°$ was obtained, which compares much more favorably with observed values of $+40$ to $+50°$ than values calculated if only α-D-*gluco* isomers[9] are considered. Specific rotations for dermatan sulfate are in the range[9] of $-55°$ to $-60°$. However, if a structure for this polymer is based on equimolecular proportions of α-L-idopyranosyluronate and 2-acetamido-2-deoxy-β-D-galactopyranosyl residues[8,9,29,30] (a small proportion of D-glucosyluronic acid residues is usually also present[9,31]), a calculated specific rotation of only about $-41°$ is obtained. This relatively poor agreement invites further examination.

EXPERIMENTAL

P.m.r. spectra were recorded with a Varian HA-100 or HR-220 spectrometer.

*The effect of an acid pH on these chemical shifts, more particularly the marked deshielding of H-5 of **1** and **2** and of glycosyluronic H-5 protons of heparin and dermatan sulfate, is also described in this earlier paper.

Samples were subjected to a preliminary deuterium-exchange by repeated treatment with fresh deuterium oxide. Tetramethylsilane, contained in a coaxial capillary tube, was utilized as the internal lock signal at 100 MHz; as this procedure causes a downward displacement of signals by about 0.5 p.p.m., the spectra shown have been corrected by −0.5 p.p.m. Proton-decoupled, ^{13}C magnetic resonance spectra were recorded at a frequency of 25.15 MHz with a Varian HA-100 spectrometer, as described previously[16]. Paper chromatography was conducted with Whatman No. 1 paper with 4:1:5 butyl alcohol–ethanol–water as the solvent, and paper electrophoresis was performed on Whatman No. 3 paper in pyridine–acetic acid (pH 6.5) at a potential of 500 V. Solutions were concentrated *in vacuo* at 35–40°.

Methyl 4,6-O-benzylidene-α-D-idopyranoside. — This compound was synthesized by the method of Sorkin and Reichstein[3] involving the following sequence: methyl α-D-galactopyranoside → methyl 4,6-*O*-benzylidene-α-D-galactopyranoside → methyl 2,3-anhydro-4,6-*O*-benzylidene-α-D-gulopyranoside. The yield of the last (20%) was much lower than that (75%) reported by Sorkin and Reichstein, due probably to formation of a methyl ether of **3** as a by-product. Otherwise, the yields were close to those obtained in the earlier study, and all melting points and specific rotations were virtually the same.

Methyl α-D-idopyranoside. — Methyl 4,6-*O*-benzylidene-α-D-idopyranoside (400 mg) was hydrolyzed in 2.5mM sulfuric acid (25 ml) for 2 h at 60°. The solution was made neutral with Dowex 1 X-4 ion-exchange resin, the suspension was filtered, and the filtrate was washed with chloroform (3 × 40 ml) to remove unreacted acetal, and evaporated to yield 255 mg (93%) of methyl α-D-idopyranoside as a colorless syrup, $[\alpha]_D^{20}$ +101° (*c* 2.8, water); lit.[3] $[\alpha]_D^{20}$ +103.3°. Examination by paper chromatography indicated the presence of only one component, having R_{MG} 2.3 (MG, methyl α-D-glucopyranoside).

Methyl α-D-idopyranosiduronic acid (**1**) (*sodium salt*). — An oxidation procedure similar to that described by Marsh and Levvy[32] was used. Platinum-on-carbon catalyst (100 mg) was suspended in a solution of methyl α-D-idopyranoside (250 mg) in water (9 ml) contained in a test tube (2 × 15 cm). The solution was heated to 55° in a water bath, and oxygen was introduced through a capillary tube at a rate sufficient to prevent the catalyst from sinking to the bottom. The pH of the solution was maintained between 7 and 8 by dropwise addition of 0.5M sodium hydrogen carbonate until (after 8 h) an equivalent amount of the alkaline solution (~2.5 ml) had been added. The procedure was continued for an additional hour, during which the pH remained constant. The suspension was then filtered through Celite, and the filtrate was evaporated to a colorless syrup (320 mg); paper chromatography showed the presence of two relatively slow-moving compounds, presumably sodium (methyl α-D-idopyranosid)uronate and an unknown (R_{MG} 0.25 and 0.43, respectively), and a faster-moving component corresponding to the unchanged glycoside. An electrophoretogram showed that the acid and the unreacted idoside travelled at relative rates of 1.4 and 0.7, respectively.

A solution of syrupy oxidation product in the minimal volume of water was

applied to a column of DEAE-Sephadex (20 ml, formate form). After 1 h, the column was washed with water, yielding methyl α-D-idopyranoside (80 mg), and then with 50mM formic acid. The second eluate was washed with ether (3 × 400 ml), concentrated to 50 ml, and then washed continuously with ether. The aqueous solution was carefully made neutral with 0.5M sodium hydrogen carbonate and freeze-dried, to yield 190 mg (65.3%) of chromatographically pure **1**; $[\alpha]_D^{20}$ +70.8° (*c* 1.53, water). The p.m.r. spectrum is described in Fig. 1 and Table I.

Methyl α-D-idopyranosiduronic acid (**1**) (*brucinium salt*). — A solution of compound **1** (30 mg) in 10 ml of water was de-ionized with Amberlite IR-120 (H^+) ion-exchange resin, and the suspension filtered. The pH of the filtrate was then adjusted to 8 with a solution of brucine in ethanol. The solution was concentrated to 0.5 ml, diluted with water (15 ml), and decolorized with carbon; the suspension was filtered, and the filtrate was successively washed with chloroform (3 × 30 ml) and ether (3 × 30 ml), and concentrated to 0.5 ml, and acetone was added to incipient turbidity. The crystals obtained on storage in the cold were filtered off, and washed successively with 60% aqueous acetone and acetone; yield 30 mg (39%). Recrystallization from aqueous acetone gave a product having m.p. 171–172° and $[\alpha]_D^{20}$ +26.9° (*c* 1.48, water).

Anal. Calc. for $C_{30}H_{38}N_2O_{11}$: C, 59.8; H, 6.4; N, 4.7. Found: C, 59.8; H, 6.5; N, 4.6.

Methyl 4,6-O-benzylidene-β-D-idopyranoside. — This compound was synthesized essentially according to Sorkin and Reichstein[3] by a sequence analogous to that for **3**, but starting with methyl β-D-galactopyranoside. As in the α series, the melting points, specific rotations, and yields were closely similar to those obtained in the earlier study, except that our yield of the intermediate 2,3-anhydride was much lower (20%) than theirs (65%).

Methyl β-D-idopyranoside. — Methyl 4,6-*O*-benzylidene-β-D-idopyranoside (500 mg) was hydrolyzed in 2.5mM sulfuric acid (30 ml) for 1.5 h at 60°. Subsequent processing as for the α anomer yielded 304 mg (89%) of the chromatographically pure compound as a colorless syrup, $[\alpha]_D^{20}$ −49.2° (*c* 2.5, water). On a paper chromatogram, the compound showed R_{MG} 2.7.

Methyl β-D-idopyranosiduronic acid (**2**) (*sodium salt*). — Methyl β-D-idopyranoside (150 mg) was dissolved in water (6 ml), and platinum-on-carbon catalyst (70 mg) was added to the solution in a test tube. The oxidation was performed, as described for methyl α-D-idopyranoside, for 7 h at 55°; ~1 equivalent of 0.5M sodium hydrogen carbonate (~1.7 ml) had by then been added. The mixture was filtered, and the filtrate was evaporated to a colorless syrup (165 mg). An electrophoretogram showed the presence of 3 components, having relative mobilities of 0.7 (unreacted methyl β-D-idopyranoside), 1.5 (major product, presumably compound **2**), and 1.9 (unidentified).

A solution of the mixture in a small volume of water was applied to a column of neutral Sephadex G-10 (300 ml), because DEAE-Sephadex proved unsatisfactory. After 1 h, the column was washed with water, the eluate being collected in 5-ml fractions. Analysis of the fractions by electrophoresis showed that a partial separation

of components had been achieved. By combining appropriate fractions, the following were obtained: (*a*) 40 mg of a colorless syrup (unidentified); (*b*) a mixture of **2** and the unidentified product (53 mg); (*c*) pure **2** (43 mg); (*d*) a mixture of **2** and methyl β-D-idopyranoside (10 mg); and (*e*) pure methyl β-D-idopyranoside (10 mg).

The sodium (methyl β-D-idopyranosid)uronate obtained had $[\alpha]_D^{20}$ −59.4° (*c* 1.43, water). Its p.m.r. spectrum is described in Fig. 1 and Table I.

Methyl β-D-idopyranosiduronic acid (**2**) (*brucinium salt*). — An aqueous solution of sodium (methyl β-D-idopyranosid)uronate (60 mg) was de-ionized with Amberlite IR-120 (H^+) ion-exchange resin, and the acid was converted into the brucinium salt as for its α anomer (**1**). The crude salt crystallized from aqueous acetone, to yield 40 mg (27%) of product which, on recrystallization (yield, 25 mg.), had m.p. 165–167°, $[\alpha]_D^{20}$ −37° (*c* 1.42, water).

Anal. Calc. for $C_{30}H_{38}N_2O_{11}$: C, 59.8; H, 6.4; N, 4.7. Found: C, 59.6; H, 6.3; N, 4.8.

Methyl α-D-glucopyranosiduronic acid (**3**) (*sodium salt*). — Methyl α-D-glucopyranoside (500 mg) in 15 ml of water was oxidized, as described for methyl α-D-idopyranoside, for 6 h at 55°. The resulting mixture was separated on a column of DEAE-Sephadex (formate, 20 ml) as already described, yielding chromatographically and electrophoretically pure material (82%) as a colorless syrup; $[\alpha]_D^{20}$ +105° (*c* 1.4, water). Its p.m.r. spectral characteristics are described under "Results and Discussion".

Calculated optical rotations of heparin and dermatan sulfate. — (*a*) *Heparin.* This calculation was based on the assumption that there is an ~1:1 ratio of aminodeoxyhexose:uronic acid, the latter being comprised of about 4:1 *ido:gluco*. Rotatory contributions[33] were obtained by use of those for the following reference compounds, as *O*-sulfation appears to have little effect on optical rotation[34]: *from* methyl α-L-idopyranosiduronic acid (Na salt) (m.w. 230), $[\alpha]_D$ −70.8°; M_D −16,300°, *i.e.* (×4/10) −6,520°; *from* methyl α-D-glucopyranosiduronic acid (Na salt) (m.w. 230), $[\alpha]_D$ +105°; M_D +24,200°, *i.e.* (×1/10) +2,420°; *from* methyl 2-deoxy-2-sulfamino-α-D-glucopyranoside[35] (Na salt) (m.w. 313), $[\alpha]_D$ +103°; M_D +32,240°, *i.e.* (×5/10) +16,120°. Hence, $[\alpha]_D = -6{,}520° + 2{,}420° + 16{,}120° = (+12{,}020°)/271$ (av.) = +44°.

(*b*) *Dermatan sulfate.* This calculation was based on a ratio of ~1:1 of aminodeoxyhexose:uronic acid. Rotatory contributions[33] were obtained with the following reference compounds: *from* methyl α-L-idopyranosiduronic acid (Na salt) (m.w. 230), $[\alpha]_D$ −70.8°; M_D −16,300°; *from* methyl 2-acetamido-2-deoxy-β-D-galactopyranoside[36] (m.w. 235), $[\alpha]_D$ −12°; M_D −2,820°. Hence, $[\alpha]_D = -16{,}300° + (-2{,}820°)$ $(= -19{,}120°)/465 = -41°$.

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REFERENCES

1 A. S. PERLIN, M. MAZUREK, L. B. JAQUES, AND L. W. KAVANAGH, *Carbohyd. Res.*, 7 (1968) 369.
2 A. S. PERLIN, B. CASU, G. R. SANDERSON, AND L. F. JOHNSON, *Can. J. Chem.*, 48 (1970) 2260.
3 E. SORKIN AND T. REICHSTEIN, *Helv. Chim. Acta*, 28 (1945) 1.
4 L. F. WIGGINS, *J. Chem. Soc.*, (1949) 1590.
5 R. BENTLEY, *J. Amer. Chem. Soc.*, 82 (1960) 8211.
6 K. HEYNS, *Ann.*, 588 (1947) 177.
7 K. HEYNS AND H. PAULSEN, *Angew. Chem.*, 69 (1967) 600.
8 P. J. STOFFYN AND R. W. JEANLOZ, *J. Biol. Chem.*, 235 (1960) 2507.
9 J. S. BRIMACOMBE AND J. M. WEBBER, *Mucopolysaccharides*, Elsevier, Amsterdam, 1964, p. 82.
10 A. S. PERLIN AND G. R. SANDERSON, *Carbohyd. Res.*, 12 (1970) 183.
11 A. S. PERLIN, D. M. MACKIE, AND C. P. DIETRICH, *Carbohyd. Res.*, 18 (1971) 185.
12 R. U. LEMIEUX, R. K. KULLNIG, H. J. BERNSTEIN, AND W. G. SCHNEIDER, *J. Amer. Chem. Soc.*, 80 (1958) 6098.
13 M. KARPLUS, *J. Chem. Phys.*, 30 (1959) 11; *J. Amer. Chem. Soc.*, 85 (1963) 2870.
14 F. A. L. ANET, *Can. J. Chem.*, 39 (1961) 789.
15 S. STERNHELL, *Quart. Rev.* (London), (1969) 236.
16 A. S. PERLIN, B. CASU, AND H. J. KOCH, *Can. J. Chem.*, 48 (1970) 2596.
17 H. J. KOCH AND A. S. PERLIN, *Carbohyd. Res.*, 15 (1970) 403.
18 D. M. GRANT AND B. V. CHENEY, *J. Amer. Chem. Soc.*, 89 (1967) 5315.
19 G. W. BUCHANAN, J. B. STOTHERS, AND S. WU., *Can. J. Chem.*, 47 (1969) 3113.
20 R. U. LEMIEUX AND J. D. STEVENS, *Can. J. Chem.*, 44 (1966) 249.
21 R. E. REEVES, *Advan. Carbohyd. Chem.*, 6 (1951) 107.
22 S. J. ANGYAL, *Angew. Chem. Int. Ed. Engl.*, 8 (1969) 157.
23 For a recent discussion, see V. S. R. RAO, K. S. VIJAYALAKSHMI, AND P. R. SUNDARARAJAN, *Carbohyd. Res.*, 17 (1971) 341.
24 N. S. BHACCA, D. HORTON, AND H. PAULSEN, *J. Org. Chem.*, 33 (1968) 2484.
25 E. L. ELIEL, N. L. ALLINGER, S. J. ANGYAL AND G. A. MORRISON. *Conformational Analysis*. Interscience, New York, 1967, p. 441.
26 R. U. LEMIEUX, in P. DE MAYO (Ed.), *Molecular Rearrangements*, Part II, Interscience, New York, 1964, p. 739.
27 M. L. WOLFROM, J. R. VERCELLOTTI, AND G. H. S. THOMAS, *J. Org. Chem.*, 29 (1964) 536.
28 V. LINDAHL AND O. AXELSSON, *J. Biol. Chem.*, 246 (1971) 74.
29 K. MEYER AND E. CHAFFEE, *J. Biol. Chem.*, 138 (1941) 491.
30 P. HOFFMAN, A. LINKER, AND K. MEYER, *Science*, 124 (1956) 1252.
31 L. A. FRANSSON, *J. Biol. Chem.*, 243 (1968) 1504.
32 C. A. MARSH AND G. A. LEVVY, *Biochem. J.*, 68 (1958) 617.
33 C. S. HUDSON, *Sci. Papers Bur. Stand.*, 21 (1926) 245, 272.
34 M. J. HARRIS AND J. R. TURVEY, *Carbohyd. Res.*, 15 (1970) 51.
35 M. L. WOLFROM, L. A. GIBBONS, AND A. J. HUGGARD, *J. Amer. Chem. Soc.*, 79 (1957) 5043.
36 Z. YARASIEJSKA AND R. W. JEANLOZ, *J. Amer. Chem. Soc.*, 80 (1958) 6325.