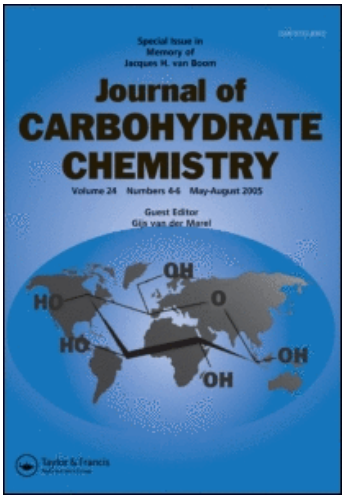

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SYNTHESIS OF L-IDURONIC ACID DERIVATIVES: CRYSTAL STRUCTURE OF METHYL (METHYL 2,3,4-TRI-*O*-ACETYL-β-L-IDOPYRANOSID)URONATE

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ABSTRACT

Several L-iduronic acid derivatives were prepared by chemical synthesis including the reducing sugar, the α– and ß-methyl glycosides and L-iduronolactone. The ß-methyl glycoside, as well as two isomeric orthoesters, were the unexpected products of glycosylation of methyl (2,3,4-tri-*O*-acetyl-α-L-idopyranosyl bromide) uronate. EI-MS was used to distinguish between the orthoesters and the glycosides. The crystal structure of the ß-methyl glycoside was determined by direct methods and refined by full-matrix least squares to a final value of R = 0.067 for 1739 reflections. The pyran ring was found to occur in the $^1C_4$ conformation, with the three acetoxy substituents in axial orientations. In aqueous solution, the α-anomer of the reducing sodium salt is almost entirely in a ^2S_O ring conformation and the α-methyl glycoside is an equilibrium mixture of conformations which is sensitive to pH. The ß-anomers all have spectral data consistent with predominant or only slightly distorted $^1C_4$ chairs.

INTRODUCTION

During our ongoing investigations of metal binding components from biological sources,[1] we required uronic acid standards for monosaccharide analysis by GC and HPLC.[2] In particular we required the lactones, the reducing sugars and the methyl glycosides of D-glucuronic and L-iduronic acids. To our surprise we could find few reports of the synthesis or spectral characterization of the L-iduronic acid derivatives.

The idofuranurono-6,3-lactone has been characterized from acid hydrolysates of dermatan sulfate[3] and by chemical synthesis of 1,2-*O*-isopropylidene-β-L-idofuranurano-6,3-lactone.[4] The phenyl[5] and benzyl[6,7] glycosides have been prepared by oxidation of the corresponding L-idopyranose derivatives. The requisite protected compounds for the direct synthesis of the glycosides and the reducing sugar have been reported by Chiba and Sinay,[8] during their synthesis of oligosaccharides. Recently, the same group has reported the coupling constants of methyl α-L-idopyranosiduronic acid 2e but no other data was given.[9] This group has also reported the synthesis and conformational analysis of methyl 2-*O*-sulfo-α-L-idopyranosiduronic acid.[10]

The conformation of the iduronic acid ring is a matter of controversy.[11] Both calculations[12] and NMR[13] suggest that at least the $^1C_4$, $^4C_1$ and ^2S_O conformations are populated depending on its substituents. During our work a minor modification of the synthetic scheme led to the unanticipated synthesis of the β-anomer. This allowed us to have a series of derivatives of both anomers, which have been fully characterized by ^1H and ^{13}C NMR as well as MS. A remarkable variation in ring conformation is observed for these compounds.

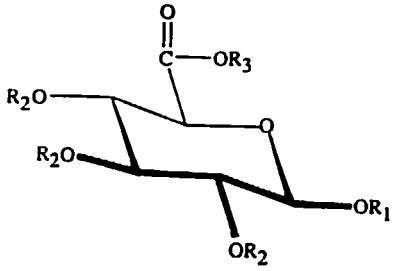
RESULTS AND DISCUSSION

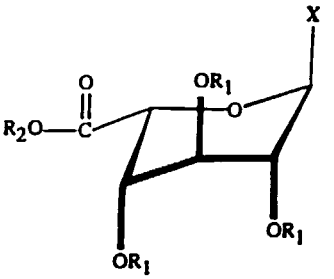
Since both the reducing sugars and the methyl glycosides were required, the synthesis was planned around the known intermediate, 1-*O*-acetate **2a**.[8] Accordingly, D-glucofuranolactone, **5a**, was converted into the well known protected D-glucopyranosiduronic acid derivative, **1a**.[14] This 1-*O*-acetyl derivative was then converted to the corresponding bromide and then glycosylated with methanol to give the protected methyl glycoside, **1b**, using silver zeolite as promoter, in 70% yield.[15] Subsequent base hydrolysis led to the sodium salt, **1c**. The spectral characteristics for this compound[16] are in agreement with those reported in the literature.[17]

The key L-*ido* intermediate **2a** was prepared by epimerization at C5 of the D-*gluco* compound **1a**, using the Ferrier bromination followed by tributyltin hydride

reduction method of Chiba and Sinay.[8] Attempted glycosylation of **2b** (bromide from **2a**) under the conditions used to form **1a**, gave three isomeric solid products. None of these products was the desired α-glycoside **2c**, but all three were isomeric to **2c**. Spectral analysis suggests the following structures: the least polar is endo-orthoester-**3a**, followed by the known exo-orthoester-**3b** and the most polar is the ß-glycoside **4a**. Compound **4a** was subsequently crystallized by slow evaporation of a chloroform/toluene solution and its crystal structure was determined by X-ray analysis.

The pyran ring of **4a** is in the $^1C_4$ conformation. This pucker may be preferred to $^4C_1$, because a 1,3-diaxial interaction between two acetoxy groups is less unfavourable than one between a methoxy and a carbomethoxy group. Two of the acetoxy groups are perpendicular to the pyranose ring, the torsion angles H3-C3-O31-C32 and H4-C4-O41-C42 being 2° and -1°, respectively. However, the acetoxy group attached to C2 is twisted away from the one at C4, the corresponding torsion angle being 33° (see Table 6 in Experimental). This distortion results in an acceptable distance (2.930 Å) between O21 and O41. Lichtenthaler and Lindner have recently described similar distortions of some ß-D-xylopyranose derivatives with 1,3 diaxial O-O interactions which have separations of 2.67 to 3.01 Å.[18] In 1,2,3,4,6-penta-*O*-acetyl-α-D-idopyranose the 1,3 diaxial O-O distances are 2.872 and 2.881 Å.[19]

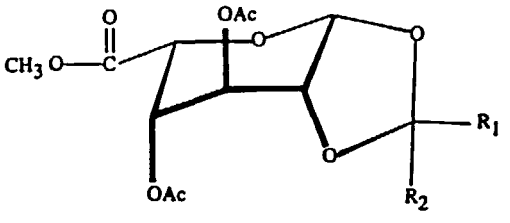


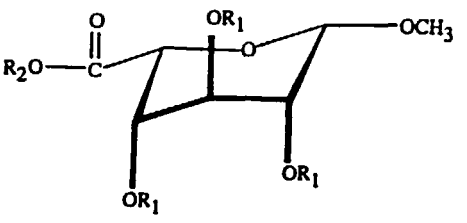


1a	R_1=Ac,	R_2=Ac,	R_3=CH_3
1b	R_1=CH_3,	R_2=Ac,	R_3=CH_3
1c	R_1=CH_3,	R_2=H,	R_3=Na^+

2a	X=OAc,	R_1=Ac,	R_2=CH_3
2b	X=Br,	R_1=Ac,	R_2=CH_3
2c	X=OCH_3,	R_1=Ac,	R_2=CH_3
2d	X=OCH_3,	R_1=H,	R_2=Na^+
2e	X=OCH_3,	R_1=H,	R_2=H^+
2f	X=OH,	R_1=H,	R_2=Na^+

The carbomethoxy substituent in **4a** is orientated such that the carbonyl oxygen is nearly eclipsed and the methoxy oxygen *trans*, the corresponding torsion angles are +15.1° and -164.0° (see Table 6). This coplanar orientation was previously found for the carbomethoxy group in methyl 1,2,3,4-tetra-*O*-acetyl-β-D-galactopyranuronate.[20] As usual, the pyranose ring is somewhat flattened at C3 and more puckered at O5. The endocyclic bond lengths agree within experimental error with "standard" values.[21] The final atomic coordinates are collected in Table 4 (see Experimental). The bond angles and bond lengths are shown in Table 5 and selected torsion angles are shown in Table 6 (see Experimental). A drawing of the final structure, along with the numbering scheme is shown in FIG. 1.





3a	endo (R)	R_1=CH_3,	R_2=OCH_3	**4a**	R_1=Ac,	R_2=CH_3
3b	exo (S)	R_1=OCH_3,	R_2=CH_3	**4b**	R_1=H,	R_2=Na^+
				4c	R_1=H,	R_2=H^+

The stereochemistry of orthoester **3b** was not established in ref. 8. However a 2% ^1H-^1H NOE was observed between the CCH_3 and H2 in **3a** but not in **3b**. In fact, no other NOE's (<0.5%) between the two methyls at the quaternary centre and the ring protons were observed in either isomer. These results are in accord with MM2 calculations of **3a** and **3b** which predict that the only NOE (H-H < 3 Å) should be between the CCH_3 and H2 for the endo (R) isomer, thus establishing the stereochemistry.

The glycosides could be distinguished from the orthoesters by their EI-MS as shown in FIG. 2. The molecular ion was not detected in the EI-MS of these three

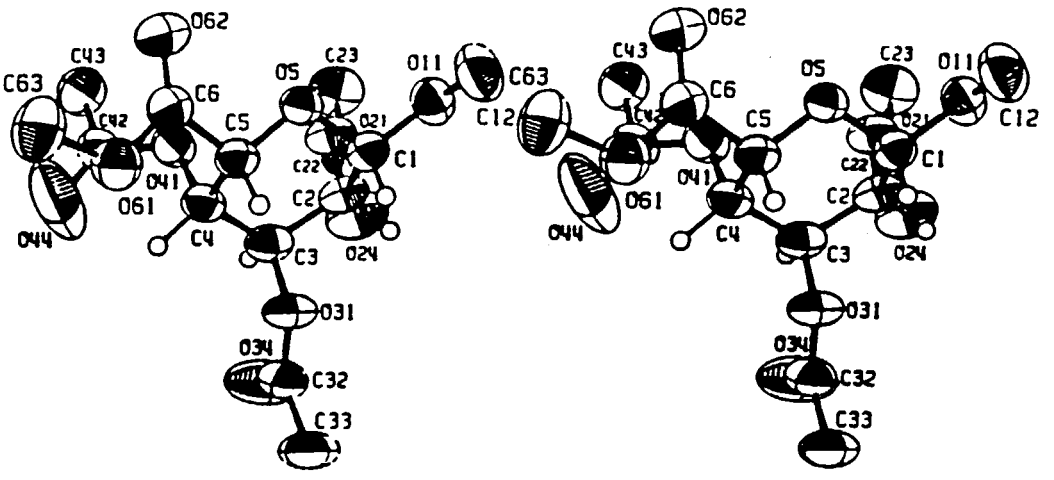


FIG. 1. Stereo ORTEP Drawing of the Crystal Structure of **4a**.

isomers, but the molecular weight was confirmed by the detection of the protonated molecular ion (m/z 349) in the FAB mass spectra of **3a**, **3b** and **4a**. The more intense m/z 317 (M-OCH_3) and the more dominant m/z 155 (further elimination of acetic acid and ketene from m/z 317) ions in the EI-MS of **3a** and **3b** than that of **4a** suggested additional stability of the resulting oxonium ion in compounds **3a** and **3b** after the cleavage of the methoxy radical. The formation of the 5-membered dioxonium ion in the proposed structures of **3a** and **3b** agrees with such a phenomenon. The high resolution data are summarized in Table 1.

Subsequently it was shown that **3a**, **3b** and **4a** were stable to prolonged treatment with silver zeolite with or without additional methanol. Furthermore, attempted glycosylation of **2a** using silver triflate with or without the addition of the base, 2,6-di-*tert*-butyl-4-methylpyridine also led to a mixture of **3a**, **3b** and **4a**, along with a trace of **2c**. Silver triflate has been previously reported to promote the exclusive formation of α-linked disaccharides from **2b**,[22] however recently this same group obtained orthoesters using these conditions.[23] Similarly, Helferich conditions[24] using $Hg(CN)_2$ in methanol led to a mixture. An alternate glycosylation procedure using the fluoride analogous to bromide **2b** and BF_3 catalysis, which avoids orthoester formation, has recently been reported.[25]

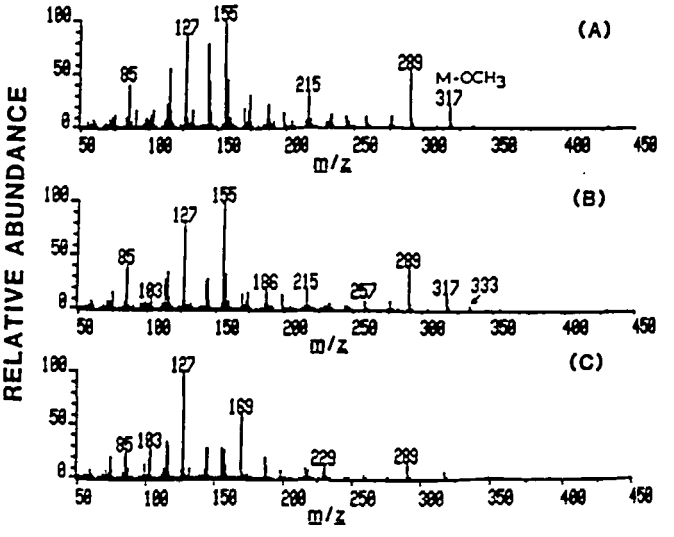


FIG. 2. The EI Mass Spectra of (A) **3a**, (B) **3b** and (C) **4a**.

TABLE. 1. High Resolution Mass Spectral Data For **3a** and **3b**.

m/z OBSERVED (error in mmu)		ASSIGNMENT
3b	**3a**	
317.0873 (0.0)	317.0864 (0.8)	M-OCH_3
289.0925 (-0.1)	289.0918 (0.5)	M-$COOCH_3$
275.0767 (0.0)	275.0786 (-2.0)	M-OCH_3-CH_2CO
257.06434 (1.8)	257.0652 (0.9)	M-OCH_3-HOAc
215.0559 (-0.4)	215.0558 (-0.2)	M-OCH3-HOAc-CH_2CO
197.0444 (0.5)	197.0444 (0.5)	m/z 317-2HOAc
186.0527 (0.1)	186.0522 (0.6)	M-2HOAc-CH_2CO
173.0444 (0.5)	173.0454 (-0.4)	m/z 215-CH_2CO
169.0498 (0.3)	169.0498 (0.3)	m/z 289-2HOAc
155.0318 (2.6)	155.0337 (0.8)	m/z 197-CH_2CO
143.0335 (0.9)	143.0336 (0.8)	$C_6H_7O_4$
127.0390 (0.5)	127.0389 (0.6)	$C_6H_7O_3$

The desired α-glycoside was prepared following Chiba and Sinay by inversion of methyl-ß-D-glucopyranoside, **1b**.[8] The sodium salts, **2f**, **2d** and **4b**, were prepared by base hydrolysis of **2a**, **2c** and **4a** respectively. The data for known compounds **2c** and **3b** are very close to the literature ^{1}H NMR values.[8] The spectra of the remaining compounds have not been reported to our knowledge, although the spectra of the α,ß-methyl glycosides of D-iduronic acid have been reported and the values of the respective enantiomers are similar to those in Table 2.[26] The circular dichroism spectra of these D-iduronic acid derivatives has also been reported.[27]

Also shown in Table 2 are coupling constants calculated from idealized structures as determined by MM2 calculations. Since these calculations do not sample all possible degrees of freedom and they neglect potentially important interactions such as solvation, we do not consider the energies reliable enough for the estimation of conformer populations.[28] However, the calculated constants do allow for tentative assignments of the ring conformations of these compounds. Coupling constants were calculated for all neutral compounds but only the results for the deprotected glycosides **2e** and **4c** are shown in Table 2 (see Experimental).

The glycosides **4a**, **4b** and **4c** all have small (< 4.5 Hz) coupling constants and show long range "W" couplings ($J_{1,3}$ and/or $J_{2,4}$). Such values are consistent with $^1C_4$ chair conformations.[29] The spectra of orthoesters **3a** and **3b** are very similar to each other, except for the substituents at the quaternary centre. The coupling constants are consistent with the $^1C_4$ conformation for both isomers, allowing for the fused ring. The protected α-glycoside **2c** has constants which are consistent with the $^1C_4$ conformation but the free acid **2e** and especially the sodium salt **2d** have constants which are much too large for a pure $^1C_4$ conformation and suggest population of the ^2S_O and/or the $^4C_1$ conformations.

Acidification of reducing **2f** led predominantly to the known α-lactone **6a**.[30] The ^{1}H NMR spectrum of **6a** is deceptively simple for a 5-spin system. In fact, only one coupling constant ($J_{3,4}$) is resolved even at 500 MHz (see Table 3). Furthermore, the intensity of the H1-H3 crosspeak was greater than for the H1-H2 crosspeak in the ^{1}H-^{1}H-COSY spectrum (i.e. $J_{1,3} > J_{1,2}$). This confusing observation was resolved by the NOE spectrum (Irr. H1) that led to the correct assignments (see Table 3). MM2 calculations suggest that the requisite H-H torsion angles of about 90° for H1-H2, H2-H3 and H4-H5 can only be accommodated with the α-anomer, calculated values are 83.6°, -85.4° and
-105.4° respectively. A representation of the proposed structure is shown in FIG. 3.

TABLE. 2. ^{1}H NMR Spectral Data for L-Idopyranosidic Acid Derivatives[a]

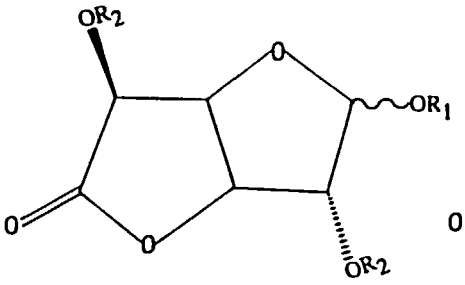
Compound	H1 ($J_{1,2}$)	H2 ($J_{2,3}$)	H3 ($J_{3,4}$)	H4 ($J_{4,5}$)	H5 ($J_{1,3}$), ($J_{2,4}$)	$COOCH_3$	OCH_3	CH_3CO
αMe 2c	4.879 brd (2.7)	4.768 dd (3.6)	5.031 ddd (2.9)	5.106 brt (2.6)	4.796 d (0.7),-	3.768	3.416	2.080 2.059 2.052
βMe 4a	4.685 dd (2.2)	4.843 ddd (4.5)	5.240 brt (4.4)	4.974 ddd (2.8)	4.472 d (0.9), (0.7)	3.682	3.450	2.030 2.008 1.968
exo 3b	5.498 dd (2.7)	3.961 m (1.9)	5.343 dd (2.7)	5.008 m (1.3)	4.391 d -, (1.0)	3.710	3.207	2.083 2.007 1.698
endo 3a	5.226 dd (2.7)	3.727 m (1.8)	5.315 brt (2.5)	5.051 dd (1.5)	4.418 d -, (1.0)	3.699	3.482	2.091 1.995 1.458
αMe 2dNa	4.681 d (4.9)	3.466 dd (6.8)	3.710 brt (6.4)	3.853 dd (4.1)	4.414 d -,-	-	3.492	-
αMe 2eH	4.821 d (3.4)	3.598 ddd (5.6)	3.854 dt (5.0)	4.003 ddd (3.2)	4.705 d (0.8,0.8)	-	3.460	-
βMe 4bNa	4.742 d (1.8)	3.671 dd (4.3)	4.074 brt (4.3)	3.871 brdd (2.6)	4.191 d -, (0.6)	-	3.560	-
βMe 4cH	4.811 d (1.8)	3.721 ddd (4.3)	4.124 brt (4.4)	3.930 ddd (2.7)	4.612 d -, (1.4)	-	3.574	-

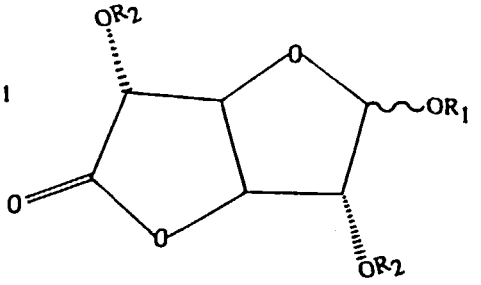
Calculated Coupling Constants in Hz and Corresponding H-H Torsion Angles in Parentheses.

α^1C_4	1.5-3.1 (68.2±10°)	1.5-4.4 (-68.9±10°)	1.7-4.4 (68.3±10°)	0.2-2.1 (57.0±10°)	0.9-2.3[b] (-56.2±10°)
α^4C_1	7.3-8.1 (168.8±10°)	7.9-10.1 (-171.7±10°)	7.9-9.9 (173.1±10°)	4.2-7.1 (-48.5±10°)	2.8-4.7[b] (51.3±10°)
α^2S_O	4.0-5.2 (147.7±10°)	8.6-10.4 (-176.0±10°)	3.9-7.1 (148.1±10°)	2.3-5.4 (35.1±10°)	4.8-6.1[b] (30.7±10°)

a. Chemical shifts in ppm and coupling constants in Hz. Spectra of **2c**, **4a**, **3b** and **3a** were recorded in $CDCl_3$ and spectra of **2d**, **2e**, **4b** and **4c** were recorded in D_2O.

b. $J_{1,2}$ for the ß-Me anomer in Hz.





5a	R_1=H,	R_2=H	**6a**	R_1=H,	R_2=H
5b	R_1=TMS,	R_2=TMS	**6b**	R_1=TMS,	R_2=TMS

The tautomeric equilibrium for the reducing **2f** was very complicated and NMR analysis in D_2O at pH = 7.5 revealed signals for at least 5 tautomers. Kennedy et al.[31] have reported that the tautomeric equilibrium is solvent dependent, but did not report values for aqueous solutions. FIG. 4 shows the anomeric portion of the ^{13}C and the ^1H NMR spectra in which the signals for the five major tautomers are identified. The lactone was assigned from the spectrum (pH = 2.15) of the lactone, **6a**. The pyranoses and furanoses were assigned on the basis of the ^1H-^1H-COSY spectra and the deduced coupling constants. The ^{13}C resonances were assigned by ^1H detected ^{13}C-^1H COSY.

Interestingly, the large coupling constants of the α-pyranose indicate the $^4C_1$ or ^2S_O conformation in contrast to the predominant $^1C_4$ conformation of the ß-anomer (small couplings and long range couplings). In this case MM2 calculations for the ^2S_O conformer indicated a 10° torsion change for H1-H2 to 158±10° and therefore constants ranging from 3.8 - 6.2 Hz. This is in reasonable agreement with the observed value of 6.2 Hz. These conformers were further distinguished by observation of a marked NOE after irradiation of H2 and observation of H5 (Obsd NOE H5/H1 = +0.83 versus Calcd - 0.04 for $^4C_1$ or + 1.07 for ^2S_O) which strongly suggests that the ^2S_O conformation is highly populated.[32] FIG. 5 shows a representation of the MM2 optimized structure of sodium α-L-idopyranosiduronate. The close proximity of H2 to H5 is readily apparent. The ^1H and ^{13}C NMR data are presented in Table 3.

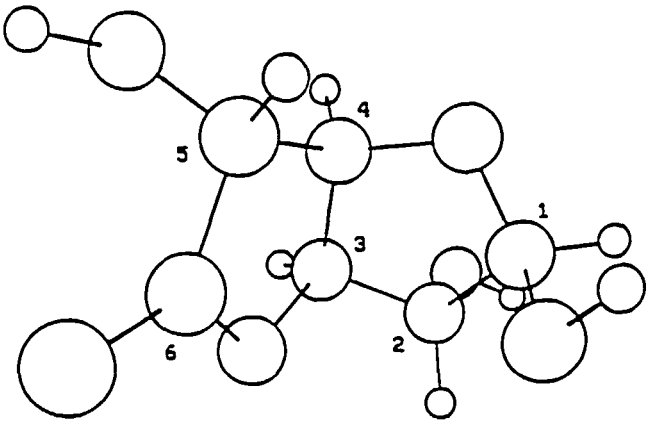


FIG. 3. Ball and Stick Representation of Lactone **6a** in its MM2 Optimized Conformation.

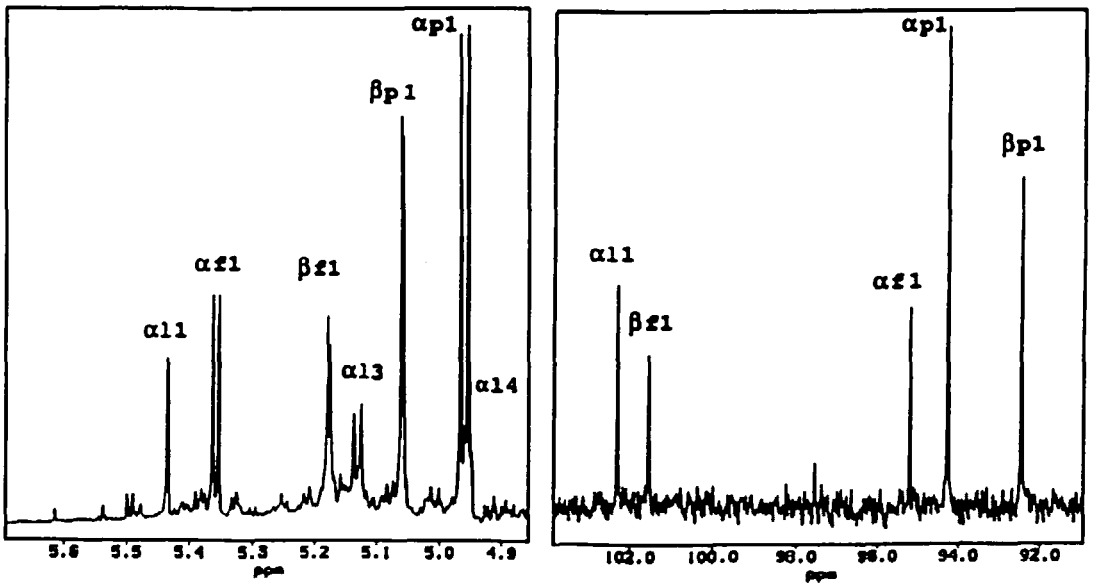


FIG. 4. ^{1}H and ^{13}C NMR Spectra of the Anomeric Protons and Carbons of Reducing Sodium L-Iduronate.

TABLE. 3. ^{1}H and ^{13}C NMR Data for Reducing Sodium L-Iduronate in D_2O.

Tautomer (%)[a]	H1-C1 ($J_{1,2}$)	H2-C2 ($J_{2,3}$)	H3-C3 ($J_{3,4}$)	H4-C4 ($J_{4,5}$)	H5-C5 ($J_{2,4}$)
αp	4.956 d	3.362 dd	3.637 dd	3.802 dd	4.444 d
(28)	94.3	72.8	72.8	71.0	71.6
	(6.2)	(8.0)	(7.8)	(4.9)	-
βp	5.057 d	3.657 ddd	4.120 t	3.920 ddd	4.353 d
(21)	92.5	69.7	70.2	69.4	75.1
	(1.5)	(3.5)	(3.2)	(1.9)	(1.4)
αf	5.358 d	4.228 dd	4.378 dd	4.502 dd	4.185 d
(15)	95.2	73.6	73.7	73.6	75.3
	(4.6)	(6.7)	(7.0)	(2.9)	-
βf	5.175 d	4.146 dd	4.273 dd	4.404 dd	4.168 d
(15)	101.6	72.8	71.6	75.1	73.6
	(2.4)	(3.8)	(4.7)	(3.8)	-
αl	5.433 s	4.359 s	5.129 d	4.951 d	4.492 s
(12)	102.3	76.5	85.9	83.9	73.6
	(<0.5)	(<0.5)	(5.7)	(<0.5)	-
other (9)					

a. Composition estimated from integration of the ^{1}H NMR spectrum and errors are estimated to be ±20% of the reported values.

The negative FAB-MS of **2d** was essentially identical to that of its D-*gluco* isomer **1c**, by observing the ($M-Na^-$) ion at m/z 207. Derivatization of **2f** by hexamethyldisilazane to the more volatile trimethylsilyl (TMS) derivatives for GC/MS analysis produced **6b** and other iduronic isomers (α-ß-furanosyl and pyranosyl structures, cf. NMR results above). The MS of **6b** was identical to that reported in the literature[33] with m/z = 230 as the base peak. Other characteristic ions observed were m/z = 392 (M^+), 377 ($M-CH_3$), 349, 319, 287, 259, 245, 243, 147 and 73. The MS of **5b** was indistinguishable from **6b**, but lactone **5b** had a different GC retention time.

CONCLUSION

A series of α- and ß-methyl glycosides as well as the reducing sugar and the lactone of L-iduronic acid were synthesized and characterized. The ß-anomers all appear to have $^1C_4$ ring conformations in solution. This conformation places the anomeric substituent and the carboxylate equatorial to the ring and the remaining substituents axial. This conformation was also found in the solid state for **4a**. This conformation in the α-anomers would place the anomeric substituent in the axial position. Although in principle this arrangement should be stabilized by the anomeric effect it is also destabilized by 1,3 interactions between O1 and O3. Consequently, other conformers are populated. The spectral data suggest that the α-anomer of the pyranose form is mostly in the ^2S_O conformation. The α-methyl glycoside is also predicted to partially populate this conformer but in a pH sensitive equilibrium with the $^1C_4$ conformer. The effect of added metal ions on this equilibrium is the subject of another communication.[34]

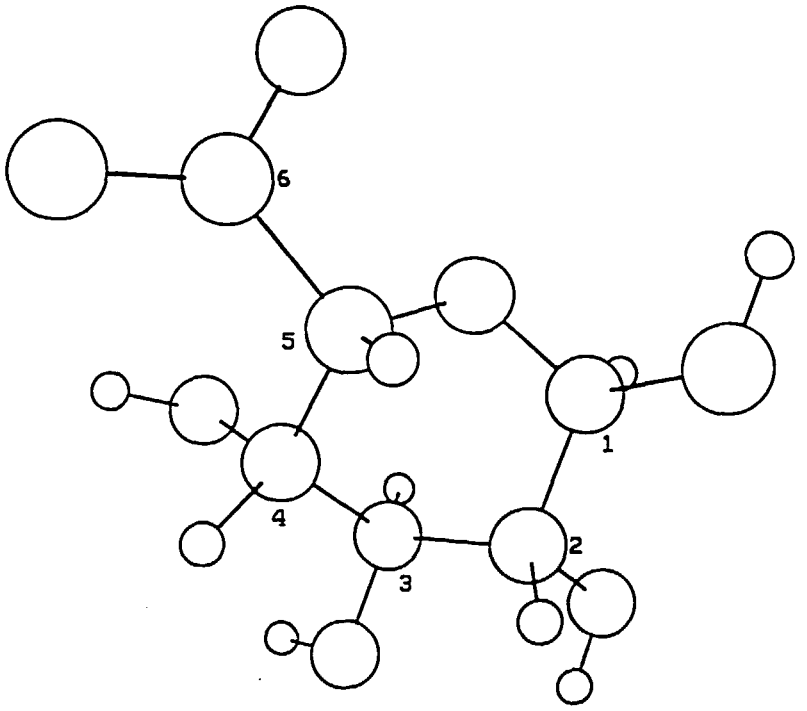


FIG. 5. Ball and Stick Representation of Sodium α-L-idopyranosiduronate in its MM2 Optimized Conformation.

EXPERIMENTAL

General Procedures. Optical rotations were measured with a Perkin-Elmer polarimeter (model 140) at 25±1 °C. ^{1}H NMR (300.137 MHz or 500.137 MHz) and ^{13}C NMR (75.47 MHz) spectra were recorded with a Bruker AM 300 MHz or a 500 MHz Spectrometer at the Carbohydrate Research Centre, University of Toronto. They were obtained at 21±2 °C either in $CDCl_3$ containing a trace of TMS as the internal standard or in D_2O (99.98%, Merck, Sharpe and Dohme) with a trace of acetone (^{1}H 2.225 ppm relative to internal DSS or ^{13}C 30.5 ppm[13] with Dioxane at 66.8 ppm) as internal standard. Steady state NOE spectra were run in the 1D difference mode. ^{1}H-^{1}H COSY spectra were obtained using conventional Bruker programs and the ^{13}C-^{1}H COSY spectra were obtained in the inverse mode.[35]

MM2 calculations were performed on a 386-20 MHz PC using the software package CHEMCAD (Austin TX). Structures were built by first creating optimized tetrahydropyran rings in each of the three conformers $^1C_4$, $^4C_1$ and ^2S_O and then adding the requisite substituents and reoptimizing. Final structures were corrected for the anomeric effect.[18] Coupling constants were calculated from these H-H torsion angles allowing for a ±10° variation, using equation 8E of Haasnoot et al.[36] The co-ordinates for these structures were then entered into the program CCM using the program TCC, both of which were run on a MicroVAX II computer.[37] This program uses a full distance matrix approach to calculate NOE's.

All mass spectra were recorded using a VG-Analytical ZAB-SE spectrometer and a VG 11-250 data system. The samples were ionized by either Fast Atom Bombardment (FAB) or Electron Impact (EI) depending on the volatilities of the samples or molecular ion information.

FAB-Samples **3a**, **3b** and **4a** were dissolved in methanol with glycerol as the matrix, and positive ion mass spectra were recorded. Samples **2d** and **2f** were dissolved in water with either glycerol or thioglycerol as the matrix and negative ion mass spectra were recorded. For exact masses, 2% polyethylene glycol 300 or 600 was added for calibration. For both positive and negative ion mass the samples were bombarded by neutral Xenon atoms which were generated using the Ion-Tech FAB11N Saddle Field ion gun with 8KeV anode potential and 1mA anode current. The mass spectrometer was calibrated with glycerol at 8KeV accelerating voltage and scanning between m/z 600 to m/z 50, and the mass spectra were recorded with multi-channel analyzing mode.

EI-Samples **3a**, **3b** and **4a** were introduced into the mass spectrometer by direct probe insertion with the ion source temperature set at 220 °C, and ionized by 70

eV electron beam at 100 μA trap current. The resolution was at 750 (10% valley definition) for low resolution mass spectra and at 10,000 for high resolution spectra. The mass spectrometer was calibrated with perfluroalkane and scanning between m/z 500 to m/z 50. For high resolution mass spectra, perfluroalkane was introduced into the mass spectrometer through the septum inlet concurrently with the sample introduction.

Gas Chromatography-Mass Spectrometry (GC-MS) of samples **3a, 3b** and **4a** was performed using a 15m capillary OV1701 column with cold on-column injection. The mass spectrometer conditions were identical to those used in the direct probe insertion experiments. The GC conditions were 40 °C for 2 min, then programmed at 15 °C/min to 200 °C, then 5 °C/min to 260 °C and the temperature was maintained for one additional minute. The temperature of the GC interface was 270 °C . The GC-MS of samples **5b** and **6b** was analyzed using the same capillary column but with a slower program rate.

Methyl 1,2,3,4,-Tetra-*O*-acetyl-α-L-idopyranuronate (2a) ^{13}C NMR ($CDCl_3$) 169.3 - 166.8, (5xCO), 89.8 (C1), 73.3, 67.3, 66.4, 65.5 (C2, C3, C4, C5), 52.6 ($COO\underline{C}H_3$) 20.7-20.4 (4x$\underline{C}H_3CO$); $[\alpha]_D$ -84.2° (*c* 1.1, chloroform), Lit. -88° (*c* 1.0, chloroform) and **Methyl (Methyl 2,3,4-Tri-*O*-acetyl-α-L-idopyranosid)uronate (2c)** ^{13}C NMR ($CDCl_3$): 169.4-168.4 (4x CO), 99.2 (C1), 67.3, 67.0, 66.9, 66.5 (C2, C3, C4, C5), 56.2 (OCH_3), 52.5 ($COO\underline{C}H_3$), 20.7-20.5 (3x $\underline{C}H_3$ CO); $[\alpha]_D$ -54.8° (*c* 1.3, chloroform), Lit. -56.5° (*c* 1.12, chloroform) were prepared from **1a** and **1b**.[8]

Methyl 3,4-Di-*O*-acetyl-1,2-*O*-(1S-methoxymethylidene)-ß-L-idopyranuronate (exo) - (3b), Methyl 3,4-Di-*O*-acetyl-1,2-*O*-(1R-methoxymethylidene)-ß-L-idopyranuronate (endo) - (3a) and Methyl (Methyl 2,3,4-Tri-*O*-acetyl-ß-L-idopyranosid)uronate (4a). The bromide **2b** (0.36 g)[8] dissolved in methylene chloride (10 mL) was added to a suspension of silver zeolite (4 g) in methylene chloride (10 mL) containing methanol (1 mL) at 4 °C. The stirring was continued for 20 h and the temperature allowed to rise to room temperature. The solids were removed by filtration, washed twice with methylene chloride, and the combined washings and filtrates were concentrated to dryness. The residue was purified by flash chromatography on silica gel (hexane:ethyl acetate 6:4) to yield amorphous **3a** (40 mg, 13%); $[\alpha]_D$ + 22 ° (*c* 0.67, chloroform); IR (neat): 1760 cm^{-1} (CO); ^{13}C NMR ($CDCl_3$): 169.3-167.3 (3x CO), 124.1 (C1'), 94.1 (C1), 73.1, 70.3, 66.7, 66.3 (C2, C3, C4, C5), 52.5 ($COO\underline{C}H_3$), 49.9 (OCH_3), 24.0 ($C\underline{C}H_3$), 20.8, 20.5 (2x $\underline{C}H_3CO$) and amorphous **3b** (110 mg, 35%); $[\alpha]_D$ -43.2° (*c* 0.29, chloroform), Lit. -

TABLE. 4. Final Atomic Parameters and their Standard Deviations of 4a.

Atom	*x*	*y*	*z*	B_{eq}/B
C 1	0.5798 (6)	0.6823 (4)	0.64283 (20)	4.04 (16)
C 2	0.6744 (6)	0.5634 (5)	0.66719 (21)	4.43 (17)
C 3	0.6490 (6)	0.4448 (4)	0.62565 (24)	4.62 (18)
C 4	0.6630 (6)	0.4764 (4)	0.55775 (22)	4.15 (16)
C 5	0.5672 (6)	0.6046 (4)	0.54385 (19)	3.87 (15)
C 6	0.5894 (6)	0.6442 (5)	0.47790 (21)	4.26 (16)
O11	0.6194 (5)	0.7898 (3)	0.67831 (16)	5.02 (14)
C12	0.5007 (11)	0.8946 (6)	0.6711 (3)	6.6 (3)
O21	0.8549 (4)	0.5992 (3)	0.66903 (15)	4.88 (13)
C22	0.9534 (6)	0.5404 (5)	0.71105 (23)	4.80 (19)
C23	1.1311 (8)	0.5971 (7)	0.7119 (3)	6.9 (3)
O24	0.9001 (6)	0.4529 (5)	0.74350 (20)	7.40 (22)
O31	0.4745 (5)	0.4006 (3)	0.63548 (18)	5.13 (15)
C32	0.4531 (8)	0.2808 (5)	0.6584 (3)	5.62 (23)
C33	0.2633 (10)	0.2497 (6)	0.6673 (3)	7.1 (3)
O34	0.5705 (8)	0.2129 (5)	0.6728 (4)	10.0 (4)
O41	0.8432 (4)	0.4961 (3)	0.54479 (17)	4.99 (14)
C42	0.9215 (8)	0.4189 (5)	0.5048 (3)	6.08 (25)
C43	1.1066 (8)	0.4508 (7)	0.4968 (3)	6.2 (3)
O44	0.8432 (10)	0.3358 (10)	0.4799 (5)	17.1 (7)
O61	0.4928 (5)	0.5618 (4)	0.44447 (15)	5.16 (14)
O62	0.6726 (7)	0.7308 (4)	0.45843 (19)	6.82 (20)
C63	0.4925 (10)	0.5789 (7)	0.3791 (3)	6.6 (3)
H1	0.454	0.668	0.651	6.9
H2	0.620	0.532	0.707	3.5
H3	0.755	0.376	0.624	5.6
H4	0.621	0.402	0.534	3.2
H5	0.453	0.586	0.552	3.4

48° (*c* 1.05, chloroform); IR (neat): 1760 cm^{-1} (CO); ^{13}C NMR ($CDCl_3$): 169.3-167.3 (3xCO), 124.6 (C1'), 96.5 (C1), 74.8, 70.1, 66.4, 66.2 (C2,C3,C4,C5), 52.6 ($COO\underline{C}H_3$), 49.2 (OCH_3), 24.9 ($C\underline{C}H_3$), 20.8, 20.5 (2x $\underline{C}H_3CO$) and amorphous 4a (40 mg, 13%); $[\alpha]_D$ +90° (*c* 0.4, chloroform); IR (neat): 1760 cm^{-1} (CO); ^{13}C NMR ($CDCl_3$): 169.6-167.6 (4x CO), 98.9 (C1), 71.8, 67.3, 66.9, 66.9 (C2, C3, C4, C5), 57.2 (OCH_3), 52.3 ($COO\underline{C}H_3$) 20.6-20.4 (3x $\underline{C}H_3CO$). Compound 4a was recrystallized by slow evaporation of a chloroform/toluene solution at room temperature to yield colorless plates mp 120-126 °C. Experimental exact mass value for the MH^+ ion of 4a under FAB ionization was 349.1136 ($C_{14}H_{21}O_{10}$, 0.1 mmu error).

TABLE. 5. Bond Lengths (Å) and Angles (°) of 4a and their Standard Deviations.

Bonds			
C1-C2	1.515 (7)	C6-O62	1.175 (6)
C1-O5	1.425 (5)	O11-C12	1.419 (7)
C1-O11	1.381 (6)	O21-C22	1.340 (6)
C2-C3	1.529 (7)	C22-C23	1.493 (8)
C2-O21	1.447 (5)	C22-O24	1.216 (7)
C3-C4	1.532 (7)	O31-C32	1.332 (6)
C3-O31	1.441 (6)	C32-C33	1.517 (9)
C4-C5	1.535 (6)	C32-O34	1.186 (9)
C4-O41	1.439 (5)	O41-C42	1.328 (6)
C5-O5	1.406 (5)	C42-C43	1.481 (8)
C5-C6	1.516 (6)	C42-O44	1.177 (9)
C6-O61	1.345 (6)	O61-C63	1.450 (7)
Angles			
C2-C1-O5	111.1 (4)	C5-C6-O62	127.7 (5)
C2-C1-O11	109.2 (4)	O61-C6-O62	125.2 (5)
O5-C1-O11	109.9 (3)	C1-O11-C12	113.1 (4)
C1-C2-C3	111.1 (4)	C2-O21-C22	117.1 (4)
C1-C2-O21	105.9 (3)	O21-C22-C23	111.1 (5)
C3-C2-O21	110.0 (4)	O21-C22-O24	122.7 (5)
C2-C3-C4	114.0 (4)	C23-C22-O24	126.2 (5)
C2-C3-O31	106.2 (4)	C3-O31-C32	117.4 (4)
C4-C3-O31	106.2 (4)	O31-C32-C33	111.2 (5)
C3-C4-C5	109.8 (4)	O31-C32-O34	122.8 (6)
C3-C4-O41	107.0 (4)	C33-C32-O34	125.9 (5)
C5-C4-O41	108.1 (3)	C4-O41-C42	119.5 (4)
C4-C5-O5	111.3 (3)	O41-C42-C43	113.0 (5)
C4-C5-C51	111.3 (4)	O41-C42-O44	120.0 (6)
O5-C5-C51	108.9 (3)	C43-C42-O44	127.0 (6)
C1-O5-C5	110.0 (6)	C6-O61-C63	117.9 (5)
C5-C6-O61	107.1 (4)		

X-Ray Analysis of 4a. Precession photographs indicated the orthorhombic space group $P2_12_12_1$ with four molecules in the unit cell. The following cell dimensions were determined from 25 reflections in the range $70 < 2\theta < 91°$: a = 7.748 (1), b = 10.203 (1), c = 22.000 (1) Å. Intensity data were measured on a CAD-4 diffractometer with Cu Kα radiation. Of the 2035 unique reflections with $2\theta \leq 148°$, 1739 had intensities $I \geq 3\sigma(I)$. The structure was determined by direct methods.[38] Atomic parameters of all non-hydrogen atoms were refined by full-matrix least squares with anisotropic temperature factors. The five hydrogen atoms attached to the pyran ring were placed in calculated positions. The refinement converged at R = 0.067. A list of structure factors is available from the second author.

TABLE. 6. Torsion Angles (°) of **4a**.

O5-C1-C2-C3	-52.9	O5-C1-C2-O21	66.6
O5-C1-C2-H2	-174	O11-C1-C2-C3	-174.3
O11-C1-C2-O21	-54.8	O11-C1-C2-H2	65
H1-C1-C2-H2	-55	C2-C1-O5-C5	65.9
O11-C1-O5-C5	-173.1	O5-C1-O11-C12	75.8
C2-C1-O11-C12	-162.1	C1-C2-C3-C4	42.8
H1-C1-O11-C12	-44	O21-C2-C3-O31	169.2
C1-C2-C3-O31	-73.8	H2-C2-C3-H3	-73
O21-C2-C3-C4	-74.2	C3-C2-O21-C22	-88.3
O21-C2-C3-H3	49	C2-C3-C4-C5	-42.8
C1-C2-O21-C22	151.5	O31-C3-C4-O41	-169.1
H2-C2-O21-C22	33	H3-C3-C4-H4	72
C2-C3-C4-O41	74.4	C4-C3-O31-C32	121.2
O31-C3-C4-C5	73.8	C3-C4-C5-O5	54.2
C2-C3-O31-C32	-117.1	O41-C4-C5-C6	59.3
H3-C3-O31-C32	2	H4-C4-C5-H5	56
C3-C4-C5-C6	175.8	C5-C4-O41-C42	-122.2
O41-C4-C5-O5	-62.3	C4-C5-O5-C1	-66.6
C3-C4-O41-C42	119.4	C4-C5-C6-O61	73.0
H4-C4-O41-C42	-1	O5-C5-C6-O61	-164.0
C6-C5-O5-C1	170.3	O62-C6-O61-C63	1.2
C4-C5-C6-O62	-107.9	C2-O21-C22-O24	5.8
O5-C5-C6-O62	15.1	C3-O31-C32-O34	3.0
C5-C6-O61-C63	-179.7	C4-O41-C42-O44	0.7
C2-O21-C22-C23	-174.0	C4-O41-C42-C43	-179.5
C3-O31-C32-C33	178.8	H5-C5-C6-O62	134

Sodium (Methyl-α-L-idopyranosid)uronate (2d). Fully protected 2c (200 mg) was dissolved in THF (4 mL) and cooled to 4 °C and cold 1M NaOH (2 mL, Fisher Titration grade) was added dropwise via syringe. The mixture was left to stir at 4° C for 16 h then excess Dowex 50W-H^+ (pH=2.5) was added and left to stir for 30 min. The solids were removed by filtration and rinsed with water (10 mL) and methanol (40 mL). The combined filtrates were repeatedly concentrated to dryness with the addition of toluene to ensure removal of acetic acid. The residue was taken up in methanol, filtered and then precipitated by addition of toluene (2 vol.) then excess hexane to yield after filtration the free acid **2e** IR (neat): 1735 cm^{-1} (CO); ^{13}C NMR (D_2O): 173.7 (CO), 102.5 (C1), 69.8 (C2), 70.9 (C3), 70.3 (C4), 69.5 (C5), 56.5 (OCH_3). This amorphous solid was redissolved in a small volume of water and passed through a short column (2.5x20 cm) of SP-Sephadex Na^+ form to yield an amorphous solid **2d** . (115 mg, 87%); $[\alpha]_D$ -49.7° (*c* 0.21 methanol); IR (neat): 1605

cm^{-1} (CO); ^{13}C NMR (D_2O): 176.4 (CO), 101.8 (C1), 70.9 (C2), 72.2 (C3), 71.0 (C4), 70.8 (C5), 56.5 (OCH_3); Exact mass for MNa^+ ion was 231.0472 ($C_7H_{11}O_7Na$, error 0.9 mmu).

Sodium (Methyl-ß-L-idopyranosiduron)ate (4b). Compound **4b** was prepared from **4a** (20 mg) as described for **2d** above, to yield the free acid **4c** IR (neat): 1740 cm^{-1} (CO); ^{13}C NMR (D_2O): 173.3 (CO), 100.6 (C1), 69.5 (C2), 69.7 (C3), 69.5 (C4), 73.4 (C5), 57.4 (OCH_3) ; Exact mass for MH^+ ion was 209.0660 ($C_7H_{12}O_7$) and after cation exchange **4b**. (12 mg, 61%); $[\alpha]_D$ +8.0° (*c* 0.1, water); IR (neat): 1610 cm^{-1} (CO). ^{13}C NMR (D_2O): (CO), 100.3 (C1), 69.5 (C2), 70.3 (C3), 70.0 (C4), 74.7 (C5), 57.2 (OCH_3).

Sodium α,ß-L-iduronate (2f). Compound **2f** was prepared from **2a** (20 mg) as described for **2e** above, to yield from the acidic resin the amorphous reducing lactone **6a** IR (neat) 1775 and 1730 m^{-1} (CO) and after anion exchange resin treatment amorphous **2f** (10 mg, 87%); IR (neat): 1610 cm^{-1} (CO). NMR spectra are presented in Table 3.

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