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Carbohydrate Research 319 (1999) 172–183

CARBOHYDRATE RESEARCH

Vibrational circular dichroism of monosaccharides

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Received 8 March 1999; accepted 18 May 1999

Abstract

Vibrational absorption and circular dichroism spectra of several monosaccharides in the 1500–1180 cm^{-1} region are presented. The spectra are analyzed for similarities and differences among anomeric, homomorphic and epimeric pairs of sugars. Among the anomeric sugars, distinct bands characteristic of anomers are present that can be used to characterize the anomers. The homomorphic sugars are found to give rise to similar vibrational circular dichroism (VCD) sign patterns provided that the substituents do not have interfering VCD bands, or those interfering bands can be identified and properly accounted for. The epimeric sugars have markedly different VCD spectra, indicating that VCD is sensitive to the chirality at individual chiral centers.

Keywords: Monosaccharides; Carbohydrates; Vibrational circular dichroism; Infrared spectra; Circular dichroism; Vibrational spectra; Circularly polarized light

1. Introduction

Vibrational circular dichroism (VCD) [1] measures the differential absorption of left versus right circularly polarized infrared light originating from vibrational transitions of chiral molecules. All $(3N-6)$ vibrational modes of a chiral molecule, N being the number of atoms in the molecule, can give rise to circular dichroism. This is an advantage compared with electronic circular dichroism (ECD) [2], where only a limited number of electronic transitions are usually accessible. Owing to such a large number of available transitions, VCD can lead to much more detailed stereochemical information on the molecule of interest. This becomes all the more important for simple carbohydrates, which do not have electronic transitions in the visible range and for which the ultraviolet region is more difficult to access. Vibrational Raman optical activity (VROA) [3], which is the Raman counterpart of VCD, has been shown [4–11] to be a very useful tool for studying carbohydrates. The complicated nature of vibrational modes and overlapping vibrational bands generally pose problems in the interpretation of vibrational optical activity spectra. Nevertheless, qualitative trends in the observed VROA spectra have provided useful applications ranging from monosaccharides to polysaccharides [4–11].

Previous VCD studies on carbohydrates include two papers in the mid-infrared region [12,13] and two papers in the hydrogen stretching region [14,15]. The smaller VCD signals seen for carbohydrates, compared with those seen for molecules with rigid structures, limited the number of VCD studies in the past on carbohydrates. Improvements in VCD instrumentation have now led to better quality

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0008-6215/99/$ - see front matter
PII: S0008-6215(99)00123-8

VCD measurements with enhanced signal-to-noise ratio, so carbohydrates can now be investigated. In this manuscript we report and analyze the VCD spectra obtained using state-of-the-art instrumentation for several different monosaccharides in the $\sim 1500-1180$ cm^{-1} region.

2. Experimental

The IR and VCD spectra were recorded on a commercial Fourier-transform VCD spectrometer, Chiralir (Bomem-BioTools, Canada) with a ZnSe beamsplitter, BaF_2 polarizer, optical filter (transmitting below 2000 cm^{-1}) and a 2×2 mm HgCdTe detector. One difference from the standard Chiralir instrument is that the photoelastic modulator used was a PEM-80 model (Hinds Instruments) without AR coating on the ZnSe optical element. The VCD spectra were recorded, using the supplied Chiralir software, with 1-h data collection time (two sets of 1247 AC scans and 138 DC scans) at 4 cm^{-1} resolution, except for α- and β-D-glucose, where 3-h data collection time was used. The absorption spectra were obtained from 138 DC scans and the same number of background scans. All spectra were recorded for ~ 0.5 M solutions in dimethyl sulfoxide-d_6 (Me_2SO-d_6) in a variable path length cell with BaF_2 windows. For all of the spectra presented here, solvent absorption and VCD have been subtracted out; the absorption and VCD spectra were scaled to give a maximum absorbance of 1.0 in the region shown.

The monosaccharides were all purchased from Sigma Chemical Co. Me_2SO-d_6 was from Cambridge Isotope Laboratory. Samples that were supplied as α or β anomers are considered, respectively, to be pure α or β anomer samples and were not lypholized. In the case of glucose, fucose, xylose, arabinose, galactose, allose and ribose, the samples were equilibrated in double-distilled water for at least 24 h and then lyophilized. For glucose-$(OD)_5$, the glucose sample was equilibrated in D_2O and then lyophilized. The lyophilized samples appeared to be amorphous. The anomeric compositions for these lyophilized samples were taken to be those for samples equilibrated in aqueous solutions [16]. In the case of L-mannose (74% α, 26% β) and L-lyxose (76% α, 24% β) the anomeric compositions of samples used were provided by the supplier. Upon dissolution in Me_2SO-d_6, the beginning conformer compositions were assumed to remain constant for the duration of VCD measurements, as mutarotation in this solvent is known to be very slow [17].

The transmission properties of optical filter and BaF_2 substrates used in the instrument restrict the range of measurements to 2000–950 cm^{-1}. The strong absorption of Me_2SO-d_6 solvent precludes observing VCD features in the 1100–950 cm^{-1} region. There are no fundamental vibrational absorption bands for monosaccharides in the 2000–1500 cm^{-1} region. When the absorption intensities of monosaccharides are adjusted (via sample path length) to be optimal in the 1500–1180 cm^{-1} range, the absorption of a band at ~ 1150 cm^{-1} becomes excessively high for VCD studies. For these reasons, the presented spectra are limited to the $\sim 1500-1180$ cm^{-1} region.

The VCD spectra of some of the monosaccharides studied here, namely D-glucose, D-galactose, D-allose, D-fucose, D-ribose, D-arabinose, D-xylose and D-lyxose, have been reported before [13]. All of the major VCD features reported in those spectra are verified in the present measurements, except in the case of ribose and lyxose where, while no VCD signals were seen earlier, improved instrumentation now allowed the observation of VCD signals.

3. Results and discussion

For convenience in the presentation and discussion, the monosaccharides considered here are divided into three groups, as was done for Raman optical activity (ROA) analysis of monosaccharides [6].

(1) Anomers of D-sugars, which differ in the orientation of hydroxyl group at the C-1 position between the axial (α anomer) and equatorial (β anomer) positions. Three anomeric pairs of D-sugars are considered here: (a) α-D-

glucose and β-D-glucose; (b) methyl α-D-glucopyranoside and methyl β-D-glucopyranoside; (c) methyl α-D-galactopyranoside and methyl β-D-galactopyranoside.

(2) Homomorphic pairs, which differ in the substituent at one of the ring carbons. Several homomorphic pairs of sugars are considered. (a) D-Glucose and D-xylose. Both have approximately 64% β anomer in the equilibrated samples [16]. The CH_2OH group at the C-5 position in glucose is replaced by H in xylose. (b) D-Lyxose and D-mannose. Both have approximately 70% α anomer in the equilibrated samples [16]. The CH_2OH group at the C-5 position in mannose is replaced by H in lyxose. (c) D-Galactose and L-arabinose. The CH_2OH group at the C-5 position in galactose is replaced by H in arabinose. The equilibrium compositions of D-galactose and L-arabinose are [16], respectively, $\sim 68\%$ β and $\sim 63\%$ α anomers (note that the hydroxyl group at the C-1 position has the same equatorial orientation both in the α anomer of L-arabinose and the β anomer of D-galactose). (d) α-D-Fucose and methyl α-D-galactopyranoside. This pair is not strictly homomorphic, since they both differ at the C-5 position (CH_2OH in galactose is replaced by CH_3 in fucose) as well as in the hydroxyl group at the C-1 position (OH group at the C-1 in fucose is replaced by OCH_3 in methyl α-D-galactopyranoside). In the absence of the availability of α-D-galactose, methyl α-D-galactopyranoside is the next closest available example to compare with α-D-fucose. (e) α-D-Glucose and methyl α-D-glucopyranoside. The O–H group at C-1 in α-D-glucose is replaced by OCH_3 in methyl α-D-glucopyranoside. (f) β-D-Glucose and methyl β-D-glucopyranoside. The O–H group at C-1 in β-D-glucose is replaced by OCH_3 in methyl β-D-glucopyranoside. (g) Methyl β-D-glucopyranoside and methyl β-D-xylopyranoside. The CH_2OH group at the C-5 position in the glucoside is replaced by H in the xyloside. (h) D-Allose and D-ribose are homomorphic. However, in D_2O solutions, equilibrated D-allose has 78% β anomer in the pyranose form and 5% β anomer in the furanose form, while equilibrated D-ribose has 58% β anomer in the pyranose from and 13% β anomer in the furanose form [16]; so they are not strictly homomorphic. (i) D-Mannose and methyl α-D-mannoside. Equilibrated D-mannose has 66% α anomer [16], so in the absence of the availability of α-D-mannose, equilibrated D-mannose may be considered to be homomorphic with methyl α-D-mannoside. (j) D-Galactose and methyl β-D-galactoside. Equilibrated D-galactose has 68% β anomer [16], so in the absence of availability of β-D-galactose, equilibrated D-galactose may be considered to be homomorphic with methyl β-D-galactoside. (k) D-Galactose and D-fucose. They differ in the replacement of the CH_2OH group at C-5 in galactose with CH_3 in fucose. The equilibrium anomeric compositions of galactose and fucose are assumed to be similar.

(3) Epimeric pairs, which differ in the orientation of hydroxyl group (between axial and equatorial positions) at one of the ring carbons (other than C-1). These sugars are divided into three groups: (a) C-4-Epimers, which differ in the orientation of hydroxyl group at the C-4 position. The four pairs, D-glucose and D-galactose, methyl α-D-glucopyranoside and methyl α-D-galactopyranoside, methyl β-D-glucopyranoside and methyl β-D-galactopyranoside and D-xylose and L-arabinose fall into this group. (b) C-3-Epimers, which differ in the orientation of hydroxyl group at the C-3 position. The two pairs D-allose and D-glucose, and D-ribose and D-xylose fall into this group. (c) C-2-Epimers, which differ in the orientation of hydroxyl group at the C-2 position. The two pairs α-D-mannose and α-D-glucose, and methyl α-D-glucopyranoside and methyl α-D-mannopyranoside fall into this class. Since α-D-mannose is not commercially available, the equilibrated D-mannose, which has $\sim 66\%$ α anomer, may be used in place of α-D-mannose. D-Ribose and D-arabinose might also be considered as a C-2 epimeric pair, but equilibrated arabinose has 63% α anomer in pyranose form, while equilibrated ribose has 58% β anomer in pyranose form and 13% β anomer in furanose form. In addition, ribose exists in both $^4C_1$ and $^1C_4$ forms while arabinose exists in the $^1C_4$ form, only [16].

Anomeric sugars

(a) α-D-Glucose and β-D-glucose. The VCD spectrum of α-D-glucose (Fig. 1(a)) exhibits a negative (1455 cm^{-1})–weak positive (1416 cm^{-1})–negative (1361 cm^{-1})–positive (1323 cm^{-1})–weak negative (1247 cm^{-1})–weak negative (1196 cm^{-1}) pattern. The corresponding pattern in the VCD spectrum of β-D-glucose (Fig. 1(b)) is weak positive (1476 cm^{-1})–negative (1420 cm^{-1})–negative (1373 cm^{-1})–positive (1355 – 1281 cm^{-1})–negative (1238 cm^{-1}). The main differences seen in going from the α to β anomer are: (a) the appearance of weak positive VCD at 1476 cm^{-1}; (b) disappearance of the 1455 cm^{-1} negative band; (c) sign change for the 1416 cm^{-1} band; (d) sign change for the lower frequency portion (at 1355 cm^{-1}) of the 1361 cm^{-1} band (the higher frequency portion at 1373 cm^{-1} remains negative); (e) the overall VCD intensities seen for the α anomer are greater than those for the β anomer of glucose. For example, the largest positive VCD at 1323 cm^{-1} in α-D-glucose has a dissymmetry factor of 1.7×10^{-4}, while that at 1299 cm^{-1} in β-D-glucose has 0.6×10^{-4}. The absorption

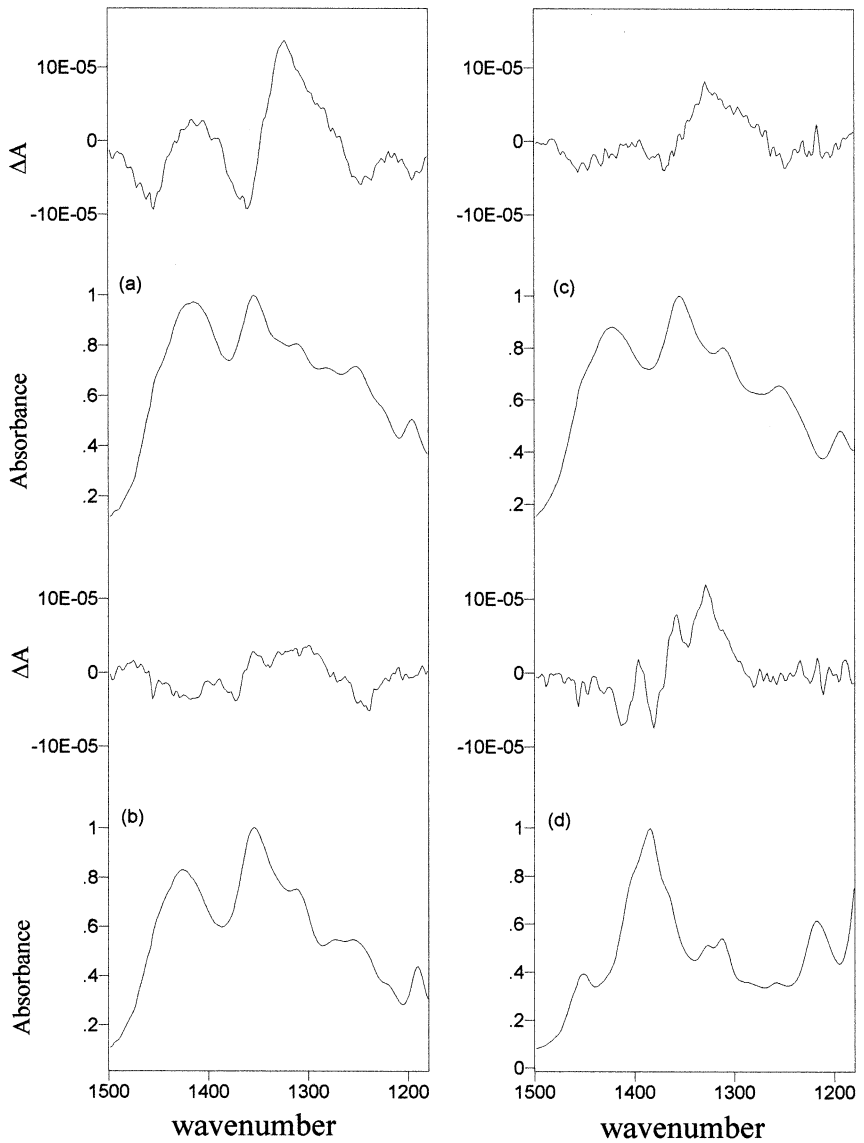

Fig. 1. Vibrational absorption (bottom) and VCD (top) spectra in Me_2SO-d_6 solution of: (a) α-D-glucose (conc, 0.5 M; pathlength, 70 μm); (b) β-D-glucose (conc, 0.5 M; pathlength, 70 μm); (c) D-glucose (conc, 0.5 M; pathlength, 150 μm); (d) D-glucose-$(OD)_5$ (conc, 0.4 M; pathlength, 150 μm). The absorption and VCD spectra are scaled so that maximum absorbance in the region shown is 1.0.

spectral patterns are also different for the two anomers. The VCD spectrum of equilibrated D-glucose (Fig. 1(c)) shows a mixture of α- and β-D-glucose spectra with a negative (1456 cm^{-1})–negative (1369 cm^{-1})–positive (1329 cm^{-1})–negative (1248 cm^{-1}) pattern. Upon deuteration of the hydroxyl groups, both absorption and VCD band shapes become sharper (Fig. 1(d)) and indicate that the hydroxyl groups contribute in a broad range. Specifically, the negative VCD band at 1456 cm^{-1} disappears, and a negative–positive VCD couplet appears at 1414–1396 cm^{-1} in D-glucose-$(OD)_5$. In place of the negative VCD band at 1369 cm^{-1} in D-glucose, a negative–positive couplet in the 1381–1358 cm^{-1} region appears in D-glucose-$(OD)_5$. In addition, the negative VCD band at 1248 cm^{-1} disappears upon deuteration. So the VCD bands in D-glucose at 1456 and 1248 cm^{-1} can be attributed to have dominating contributions from the O–H bending modes, while those seen at 1414 (−), 1396 (+), 1381 (−), 1358 (+) and 1329 (+) in D-glucose-$(OD)_5$ and at 1329 cm^{-1} in D-glucose are to be associated with the methine and CH_2 bending modes. The negative VCD band at 1369 cm^{-1} in D-glucose-$(OH)_5$ may have contributions from the O–H and C–H bending modes, as this band is replaced by a negative–positive couplet in D-glucose-$(OD)_5$.

(b) Methyl α-D-glucopyranoside and methyl β-D-glucopyranoside. The VCD spectrum of methyl α-D-glucopyranoside (Fig. 2(a)) shows a very weak negative–positive couplet (1456–1433 cm^{-1}), weak negative (1371 cm^{-1}), a positive (1332 cm^{-1}) and a large negative (1189 cm^{-1}) band. The positive band at 1332 cm^{-1} with a shoulder on the lower frequency side (1320 cm^{-1}) extends all the way to 1277 cm^{-1}. The VCD spectrum of methyl β-D-glucopyranoside (Fig. 2(b)) shows a weak positive (1475 cm^{-1}), negative (1455–1420 cm^{-1}), positive (1363 cm^{-1}), positive (1314 and 1286 cm^{-1}), and negative (1260–1225 cm^{-1}) bands. Notable differences in the VCD of these two anomers are: (a) The α anomer has a very weak negative–positive couplet in the 1456–1433 cm^{-1} region while the β anomer has positive–negative couplet in the 1475–1450 cm^{-1} region. (b) The negative VCD band at 1371 cm^{-1} in the α anomer is replaced by a positive VCD band at 1363 cm^{-1} in the β anomer. This sign reversal corresponds to a similar sign reversal at 1355 cm^{-1} in the VCD spectra of the α- and β-D-glucose anomers. As seen for the α- and β-D-glucose anomers, the overall VCD intensities seen for the α anomer are greater than those for the β anomer of the glucopyranoside. For example, the largest VCD at 1332 cm^{-1} in methyl α-D-glucopyranoside has a dissymmetry factor of 2.4×10^{-4}, while that at 1286 cm^{-1} in methyl β-D-glucopyranoside has 1.3×10^{-4}.

(c) Methyl α-D-galactopyranoside and methyl β-D-galactopyranoside. In the case of galactopyranosides, also, the VCD sign patterns (Fig. 2(c and d)) do not appear similar for the α and β anomers. A very weak negative (1459–1451 cm^{-1}), very weak positive (1443 cm^{-1}), positive (1418 cm^{-1}), positive (1341 cm^{-1}), broad positive (1283 cm^{-1}) and a negative (1188 cm^{-1}) band appear in the VCD spectrum of methyl α-D-galactopyranoside (Fig. 2(c)). Small positive (1471 cm^{-1}), a negative (1454 cm^{-1}), broad positive (1412–1379 cm^{-1}), and positive (1296 cm^{-1}) bands are present in the VCD spectrum of methyl β-D-galactopyranoside (Fig. 2(d)). Except for the difference in band shapes, the main differences in these two spectra are: (a) the β anomer shows a positive–negative VCD couplet in the 1470–1450 cm^{-1} region, while the α anomer shows a very weak negative–positive couplet in the 1460–1440 cm^{-1} region; (b) the presence of a negative VCD band at 1188 cm^{-1} in the α anomer and its absence in the β anomer.

In all of the three pairs of anomers examined, the α anomer exhibited a negative VCD at $\sim$1190 cm^{-1} (with more intensity in methyl D-glucopyranoside and methyl D-galactopyranosides than in D-glucose), while the β anomer did not. Also the β anomers in all three cases exhibit, a sometimes weak, positive–negative VCD couplet in the $\sim$1470–1450 cm^{-1} region, while the α anomers exhibit a weak (or very weak) negative–positive VCD couplet in the $\sim$1460–1440 cm^{-1} region. So these bands may be used as characteristic VCD features of the α and β anomers of glucose and galactose.

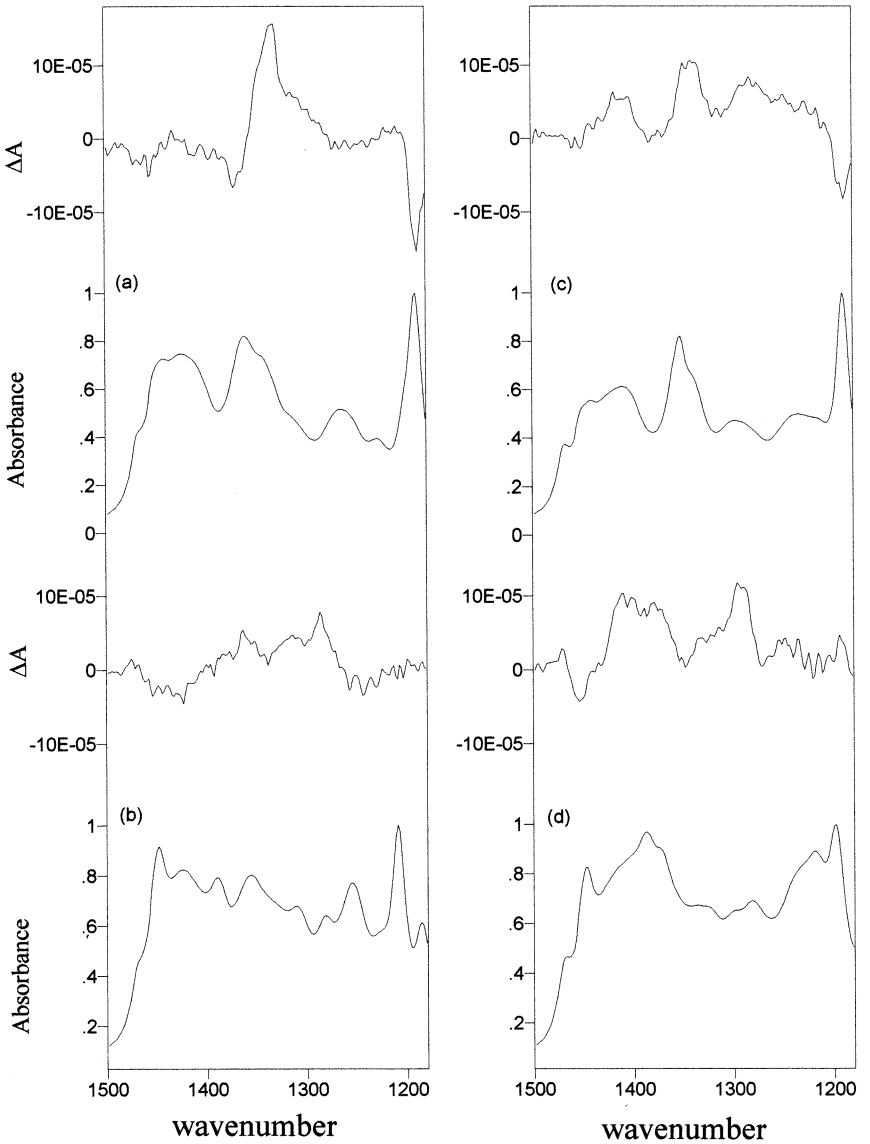

Fig. 2. Vibrational absorption (bottom) and VCD (top) spectra in Me_2SO-d_6 solution of: (a) methyl α-D-glucopyranoside (conc, 0.5 M; pathlength, 70 μm); (b) methyl β-D-glucopyranoside (conc, 0.5 M; pathlength, 120 μm); (c) methyl α-D-galactopyranoside (conc, 0.5 M; pathlength, 120 μm); (d) methyl β-D-galactopyranoside (conc, 0.5 M; pathlength, 150 μm). The absorption and VCD spectra are scaled so that maximum absorbance in the region shown is 1.0.

Homomorphic sugars

(a) D-*Glucose and* D-*xylose*. The VCD spectrum of D-xylose (Fig. 3(a)) is rich in VCD bands and appears different from that of D-glucose (Fig. 1(c)) at first sight. But a closer examination reveals that the two have a similar pattern of VCD signs, as expected for molecules with the same sense of chirality at individual chiral centers. The VCD spectrum of D-xylose exhibits negative (1455 cm^{-1}), positive (1405 cm^{-1}), negative (1366 cm^{-1}), positive (1346–1280 cm^{-1}), negative (1254 cm^{-1}), weak positive (1229 cm^{-1}) and negative (1199 cm^{-1}) bands. These features are similar to those in D-glucose, except that the positive bands at 1405 and 1229 cm^{-1} and the negative band at 1199 cm^{-1} seen for D-xylose are not present for D-glucose. The rich structure of VCD bands and enhanced VCD intensities seen for D-xylose are attributable to the absence in D-xylose of conformational mobility and vibrational modes arising from the CH_2OH group at the C-5 position.

(b) L-*Lyxose and* L-*mannose*. The VCD spectrum of L-lyxose (Fig. 3(b)) exhibits negative bands at 1465 and 1435 cm^{-1} and posi-

tive features at 1398, 1373, 1342, 1315, 1272 and 1254 cm^{-1}. The VCD spectrum of L-mannose (Fig. 3(c)) shows weak positive VCD in the 1470–1440 cm^{-1} region, and predominantly positive VCD features in the 1420–1200 cm^{-1} region, except for a negative band at 1319 cm^{-1}. The presence of the CH_2OH group in mannose, and its absence in lyxose, seems to affect the VCD in the regions at 1465–1435 and 1319 cm^{-1}, suggesting the contribution of CH_2OH group at these locations. If we ignore the negative VCD band at 1319 cm^{-1} in L-mannose and negative VCD bands at 1465 and 1435 cm^{-1} in L-lyxose, overall VCD in these two cases is the same (positive) as expected for molecules with same sense of chirality at individual chiral centers. The VCD magnitudes in lyxose and mannose are not very different from each other, so the conformational mobility resulting from the CH_2OH group in mannose is either restricted or not influencing VCD as much as it does in glucose.

(c) D-*Galactose and* L-*arabinose*. The VCD spectrum of D-galactose (Fig. 3(d)) shows a negative band in the 1460–1450 cm^{-1} region,

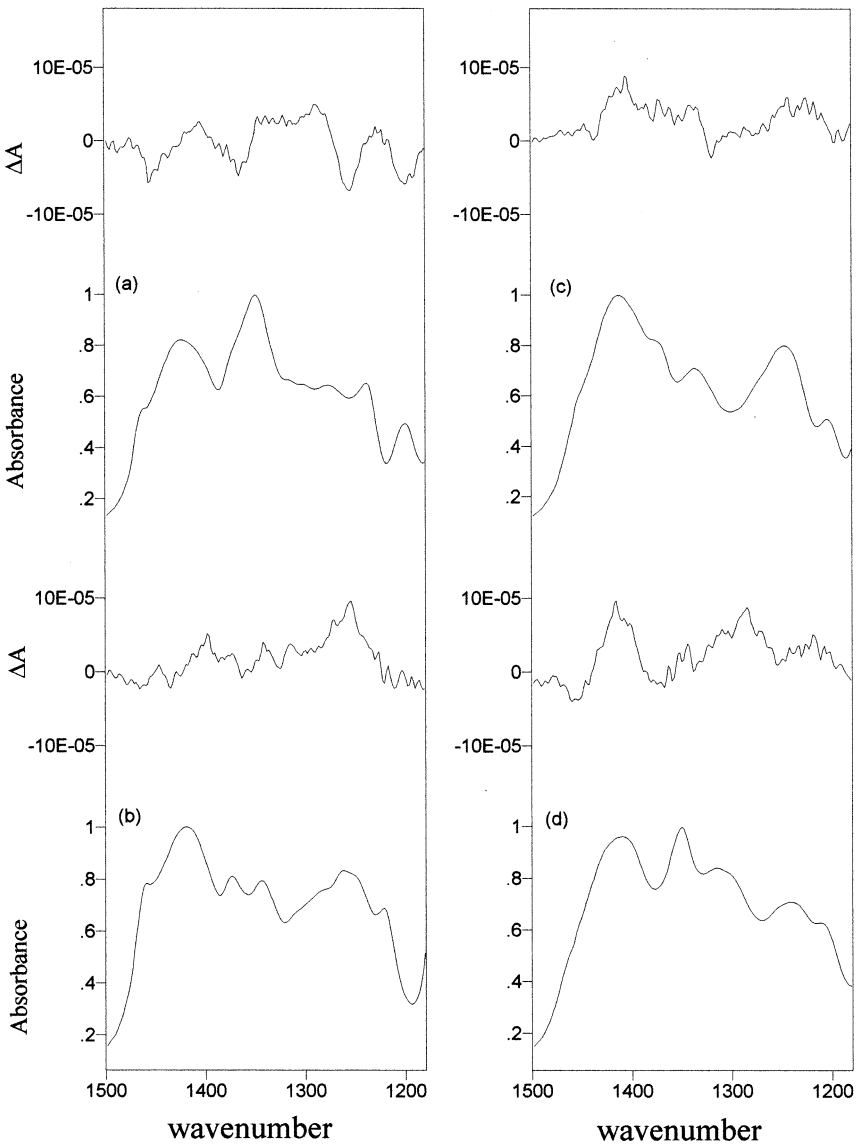

Fig. 3. Vibrational absorption (bottom) and VCD (top) spectra in Me_2SO-d_6 solution of (a) D-xylose (conc, 0.5 M; pathlength, 175 μm); (b) L-lyxose (76% α, 24% β; conc, 0.5 M; pathlength, 130 μm); (c) L-mannose (74% α, 26% β; conc, 0.5 M; pathlength, 130 μm); (d) D-galactose (conc, 0.5 M; pathlength, 150 μm). The absorption and VCD spectra are scaled so that maximum absorbance in the region shown is 1.0.

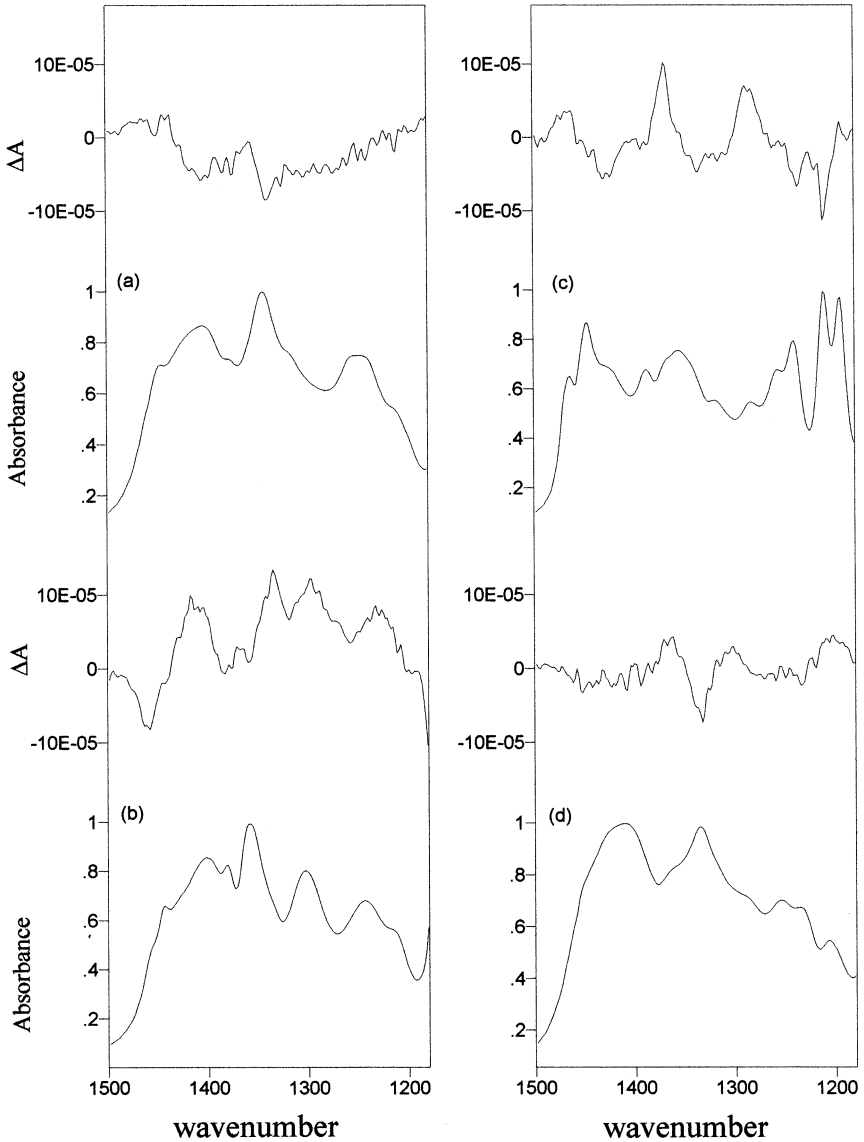

Fig. 4. Vibrational absorption (bottom) and VCD (top) spectra in Me_2SO-d_6 solution of: (a) D-arabinose (conc, 0.5 M; pathlength, 150 μm); (b) α-D-fucose (conc, 0.5 M; pathlength, 130 μm); (c) methyl β-D-xylopyranoside (conc, 0.5 M; pathlength, 120 μm); (d) D-allose (conc, 0.5 M; pathlength, 150 μm). The absorption and VCD spectra are scaled so that maximum absorbance in the region shown is 1.0.

and positive bands in the rest of the region shown. This pattern is similar to that seen (Fig. 2(d)) for methyl β-D-galactopyranoside (the weak positive VCD seen for the latter at ~ 1471 cm^{-1} is not seen for D-galactose, probably because of the lower percentage of β anomer in equilibrated D-galactose). Since L-arabinose is homomorphic with D-galactose, the VCD spectrum of D-arabinose should have opposite signs to those of D-galactose if the homomorphic pairs are to exhibit the same VCD sign patterns. This in fact appears to be true, as the VCD spectrum of D-arabinose (Fig. 4(a)) shows positive VCD in the 1480–1440 cm^{-1} region and negative VCD features in the rest of the region shown. Unlike in the case of xylose and glucose, the VCD intensities in arabinose are generally smaller than those in galactose, so the presence of CH_2OH group in galactose seems to enhance VCD, suggesting that the conformational mobility of CH_2OH group in galactose is probably restricted.

(d) α-D-Fucose and methyl α-D-galactopyranoside. The VCD spectrum of α-D-fucose (Fig. 4(b)) is very similar to that of D-galactose

(Fig. 3(d)), with negative VCD at 1458 cm^{-1} and positive VCD bands in the rest of the region. The same sign pattern is also seen for methyl α-D-galactopyranoside (Fig. 2(c)), except that the first negative VCD feature (corresponding to the one at 1458 cm^{-1} in α-D-fucose) is very weak and the positive feature at 1232 cm^{-1} is not as well resolved as in α-D-fucose. The VCD magnitudes are somewhat larger in α-D-fucose than in methyl α-D-galactopyranoside. Since fucose has CH_3 group in place of CH_2OH at the C-5 position, and galactoside has OCH_3 group in place of OH at the C-1 position, differences at both the C-5 and C-1 positions might be responsible for this difference in VCD magnitudes. However, since the magnitudes of VCD intensities in D-galactose and D-fucose are similar (vide infra), the reduced VCD magnitudes in methyl α-D-galactopyranoside might be attributed to the methoxy group at C-1 position.

(e) α-D-Glucose and methyl α-D-glucopyranoside. The negative (1455 cm^{-1})–positive (1416 cm^{-1}) couplet seen in α-D-glucose (Fig. 1(a)) has become much weaker in methyl α-D-glucopyranoside (Fig. 2(a)), so this couplet may have a dominant contribution from the hydroxyl group at C-1. The weak negative VCD band seen at 1247 cm^{-1} in α-D-glucose is not apparent in glucoside, but the weak negative VCD band seen at 1196 cm^{-1} in α-D-glucose appears with enhanced VCD intensity in glucoside at 1189 cm^{-1}.

(f) β-D-Glucose and methyl β-D-glucopyranoside. Both compounds (Fig. 1(b) and Fig. 2(b)) have common VCD features at 1476–1468 cm^{-1} (positive), ~1423 cm^{-1} (negative), ~1363 cm^{-1} (positive), ~1315 cm^{-1} (positive), and 1286 cm^{-1} (positive). The negative VCD band seen at 1238 cm^{-1} in β-D-glucose appears at 1244 cm^{-1} as a weak negative VCD band in methyl β-D-glucopyranoside. However, the negative VCD band seen at 1373 cm^{-1} in β-D-glucose is not present in methyl β-D-glucopyranoside, which may be attributable to the fact that the O–H bending vibration at C-1 in β-D-glucose is replaced by the O–CH_3 vibrations. It is possible that VCD from the symmetric bending mode of methyl group might be changing the appearance of VCD features here.

(g) Methyl β-D-xylopyranoside and methyl β-D-glucopyranoside. Both compounds (Fig. 4(c) and Fig. 2(b)) exhibit a positive–negative couplet in the ~1470–1450 cm^{-1} region, positive VCD bands at ~1365 and 1285 cm^{-1} and negative VCD in the ~1250–1200 cm^{-1} region. However, methyl β-D-xylopyranoside exhibits a weak negative VCD band at 1335 cm^{-1}, which is not apparent in methyl β-D-glucopyranoside. In general, the absorption and VCD bands of methyl β-D-xylopyranoside appear sharper and better resolved than those of methyl β-D-glucopyranoside. This is attributable to spectral broadening resulting from the conformational mobility introduced by the CH_2OH group in methyl β-D-glucopyranoside.

(h) D-Allose and D-ribose. In the 1500–1400 cm^{-1} region D-allose (Fig. 4(d)) has weak negative VCD, while D-ribose (Fig. 5(a)) has a negative (1470–1450 cm^{-1})–positive (1450–1420 cm^{-1}) couplet. In the 1400–1300 cm^{-1} region, D-allose has positive (~1365 cm^{-1})–negative (~1335 cm^{-1})–positive (1303 cm^{-1}) triplet, while D-ribose has a weak positive (1347 cm^{-1})–negative (1337 cm^{-1})–negative (1311 cm^{-1}) pattern. In the 1300–1200 cm^{-1} region, D-allose has one positive VCD feature at ~1202 cm^{-1}, while D-ribose has weak negative bands (1287 and 1263 cm^{-1}) and a broad positive VCD band ranging from 1250 to 1190 cm^{-1}. These differences are larger than those seen for any other homomorphic pair considered above, and they occur probably because, as mentioned earlier, D-ribose exists in both pyranose and furanose forms, while allose exists predominantly in the pyranose form. In addition, D-ribopyranose exists in both $^4C_1$ and $^1C_4$ forms, while D-allopyranose exists only in the $^4C_1$ form [16].

(i) D-Mannose and methyl α-D-mannopyranoside. Except for one negative VCD band at 1319 cm^{-1}, the entire VCD spectrum of L-mannose (Fig. 3(c)) has positive VCD. The VCD spectrum of methyl α-D-mannopyranoside (Fig. 5(b)) has negative VCD in the entire region, except for weak positive VCD in the 1460–1450 cm^{-1} region. Thus, this pair supports the general observation that the homomorphic pair of sugars have similar VCD spectra.

(j) D-*Galactose and methyl β-*D-*galactopyranoside*. The VCD spectra of both compounds (Fig. 3(d) and Fig. 2(d)) are similar in that negative ($\sim$1454 cm^{-1})–positive ($\sim$1410 cm^{-1})–positive ($\sim$1296 cm^{-1}) features dominate both spectra. The weak positive VCD at 1471 cm^{-1} seen in methyl β-D-galactopyranoside is not apparent in the VCD spectrum of D-galactose, which may be attributable to the fact that the amount of β anomer is reduced in D-galactose. The positive VCD at 1379 cm^{-1} seen in methyl β-D-galactopyranoside, and not apparent in the VCD spectrum of D-galactose, might be attributable to the methoxy group in the former.

(k) D-*Galactose and* D-*fucose*. The VCD spectra of both D-fucose (Fig. 5(c)) and D-galactose (Fig. 3(d)) show a negative–positive doublet in the $\sim$1460–1418 cm^{-1} region, so the contribution of CH_2OH group vibrations to VCD in this region is ruled out. A negative (1381 cm^{-1})–positive (1367 cm^{-1}) couplet present in D-fucose may have a dominating contribution from the CH_3 group vibrations, since this couplet is not apparent in D-galactose. In the 1350–1280 cm^{-1} region, and in the 1250–1200 cm^{-1} region, the VCD of D-fucose is positive, as it is for D-galactose. The weak negative band at 1256 cm^{-1} seen in

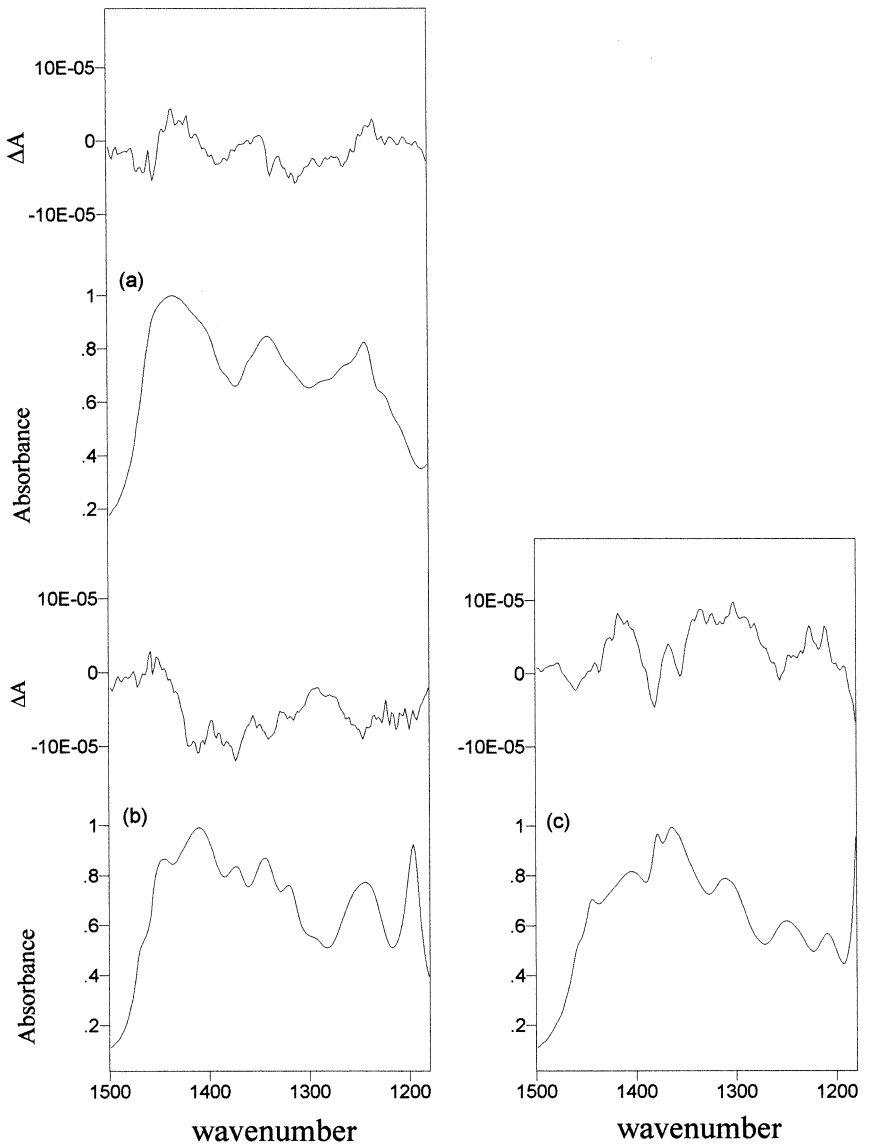

Fig. 5. Vibrational absorption (bottom) and VCD (top) spectra in Me_2SO-d_6 solution of: (a) D-ribose (conc, 0.5 M; pathlength, 150 μm); (b) methyl α-D-mannopyranoside (conc, 0.5 M; pathlength, 120 μm); (c) D-fucose (conc, 0.5 M; pathlength, 130 μm). The absorption and VCD spectra are scaled so that maximum absorbance in the region shown is 1.0.

D-fucose is not apparent in D-galactose. The magnitudes of the VCD intensities observed for these two molecules are not very much different, so the conformational mobility of the CH_2OH group in galactose may either be restricted or not influencing VCD (as concluded in the comparison of galactose and arabinose).

Epimeric sugars

(a) C-4 Epimers. The VCD spectra of D-glucose (Fig. 1(c)) and D-galactose (Fig. 3(d)) differ in the following aspects: the large positive VCD seen at 1416 cm^{-1} in D-galactose is not present in the spectrum of D-glucose; negative VCD seen at 1369 cm^{-1} in D-glucose is not present in D-galactose; negative VCD seen at 1248 cm^{-1} in D-glucose appears as positive at 1218 cm^{-1} in D-galactose. Similarly, the VCD spectra of methyl α-D-glucopyranoside (Fig. 2(a)) and methyl α-D-galactopyranoside (Fig. 2(c)) differ in the following aspects: the positive VCD seen at 1418 cm^{-1} in galactoside is not present in glucoside; the negative VCD seen at 1371 cm^{-1} in glucoside is not present in galactoside; the positive VCD seen in galactoside at $\sim$1283 cm^{-1} is absent in glucoside. The major differences seen among the VCD spectra of methyl β-D-glucopyranoside (Fig. 2(b)) and methyl β-D-galactopyranoside (Fig. 2(d)) are limited to the observation that the positive VCD seen at 1412 cm^{-1} in galactoside is absent in glucoside and appears as negative VCD at $\sim$1423 cm^{-1}. The differences in the VCD spectra of another epimeric pair, D-xylose and L-arabinose, are more striking. While D-xylose (Fig. 3(a)) exhibits several VCD couplets in the 1400–1200 cm^{-1} region, D-arabinose (Fig. 4(a)) exhibits all negative VCD features in this region. The only common feature between their VCD spectra is that D-xylose shows a negative–positive couplet in the 1460–1400 cm^{-1} region and D-arabinose shows a positive–negative couplet (which will be negative–positive for L-arabinose) in the same region. Thus, the VCD spectra of these epimeric sugars suggest that a change in the configuration at C-4 chiral center has a profound effect on the appearance of VCD spectra.

(b) C-3 Epimers. The VCD spectra of D-glucose (Fig. 1(c)) and D-allose (Fig. 4(d)) show entirely different sign patterns in the 1400–1200 cm^{-1} region. While D-glucose has a negative–broad positive–negative sign pattern, D-allose has a positive–negative–positive–weak negative–positive sign pattern. One common feature between their VCD spectra is that both show a negative VCD at $\sim$1452 cm^{-1}. The second epimeric pair, D-xylose (Fig. 3(a)) and D-ribose (Fig. 5(a)), also show significantly different VCD sign patterns in the 1400–1200 cm^{-1} region. The negative–positive VCD couplet in the 1500–1400 cm^{-1} region is a common feature in their spectra. Thus, the VCD spectra of the C-3 epimeric sugars suggest that a change in the configuration at the C-3 chiral center also has a profound effect on the appearance of VCD spectra.

(c) C-2 Epimers. To assess the C-2 epimeric pairs, the VCD spectra of α-D-glucose and L-mannose (noting that VCD of D-mannose will be opposite to that of L-mannose) may be compared (Fig. 1(a and c)). The VCD sign patterns seen for D-mannose and α-D-glucose are different in the 1500–1400 cm^{-1} region. While D-mannose has negative VCD, α-D-glucose has a negative–positive couplet in this region. However, since equilibrated D-mannose contains a significant amount of β anomer ($\sim$30%), a better comparison of C-2 epimeric pairs may be obtained by considering methyl α-D-glucopyranoside (Fig. 2(a)) and methyl α-D-mannopyranoside (Fig. 5(b)). The VCD spectra of this pair appear to be entirely different from each other in the entire 1500–1200 cm^{-1} region. Thus, the VCD spectra of the C-2 epimeric sugars also suggest that a change in the configuration at a chiral center has a profound effect on the appearance of VCD spectra.

4. Conclusions

VCD spectra obtained for several different monosaccharides indicate the following: (a) among the anomeric sugars, distinct bands characteristic of anomers are present, which can be used to characterize the anomers; (b) homomorphic sugars give rise to similar VCD sign patterns provided that the substituents do

not have interfering VCD bands or those interfering bands can be identified and properly accounted for. This is justifiable on the grounds that the VCD signs are determined by the absolute configuration at a given chiral center; (c) epimeric sugars have totally different VCD spectra. This is again justifiable because a change in the absolute configuration at one chiral center will have a dramatic effect on the overall stereochemistry of a given carbohydrate.

Acknowledgements

Grants from NSF (CHE9707773) and Vanderbilt University are gratefully acknowledged.

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