Carbohydrate Research, 81 (1980) 213–223

LASER-RAMAN STUDY OF SOLUTE–SOLVENT INTERACTIONS IN AQUEOUS SOLUTIONS OF D-FRUCTOSE, D-GLUCOSE, AND SUCROSE

MOHAMED MATHLOUTHI,
Institut Universitaire de Technologie, Département "Biologie Appliquée", Université de Dijon, B.P. 510, 21014 Dijon Cédex (France)

CLAUDINE LUU,
Faculté de Pharmacie, 34000 Montpellier (France)

ANNE MARIE MEFFROY-BIGET,
C.N.R.S., Faculté des Sciences, 76130 Mont Saint Aignan (France)

AND DANG VINH LUU
Laboratoire de Spectroscopie Raman, Université des Sciences et Techniques du Languedoc, 34060 Montpellier (France)

(Received October 17th, 1979; accepted for publication, November 26th, 1979)

ABSTRACT

The solute–solvent interactions of D-fructose, D-glucose, and sucrose in aqueous solution were studied by comparison of characteristic, Raman frequencies of the water and the sugar components. Shifts in frequency and intensity were observed in both the bending and the stretching regions of CH_2 and H_2O. The ratios of integrated, Raman intensities $I(CH_2)/I(H_2O)$ of the CH_2 peak and the H_2O bending band, and I(CH)/I(OH) of the C–H stretching line to O–H stretching band were determined. Their evolutions in terms of mass-concentration display discontinuities at specific concentrations for each of the three sugars. These breaks were interpreted as changes in the hydrogen bonding of the various species.

INTRODUCTION

Solvent–solute interactions in aqueous solutions of proteins[1,2] as revealed by Raman spectroscopy have recently been discussed, but, as far as we are aware, this technique has not been applied to the study of structure modifications in aqueous solutions of sugars, except for work by Walrafen[3], who showed the "structure-maker" effect of sucrose by its modification of the Raman spectrum of water below 300 cm^{-1}. We have recently shown that the laser-Raman effect may be applied to the study of the conformational equilibrium of D-fructose[4] in water. However, although Raman spectroscopy had been used frequently during the past few years to study the structure of certain carbohydrates, the aim of these studies was to find the structure of a simple model for such polysaccharides as amylose[5] or cellulose[6,7], to verify the theoretical calculation of the normal modes of vibrations[7-9], or to explain the modification of heteropolysaccharides of biological interest[10,11]. Certain of these investigations were made on H_2O or D_2O solutions[5,7], but the interactions between solute and solvent were not pointed out.

We now compare the characteristic Raman bands of frequencies of water on the one hand to those of carbohydrates on the other, and deduce from this comparison an interpretation of the nature of the solvent–solute interactions. The interactions probably consist of hydrogen bonds which are formed as the concentration is increased.

EXPERIMENTAL

Solutions of D-fructose, D-glucose, and sucrose (Merck products) in distilled water were filtered through a membrane, and the concentrations were checked with an Abbé refractometer. The laser-Raman spectra of these solutions were recorded with a laser-Raman spectrometer previously described[4]. The recording regions were 3700–2700 cm^{-1} and 1700–100 cm^{-1} at a scan speed of 40 $cm^{-1}.min^{-1}$; the wavelength accuracy was within ± 2 cm^{-1}. The integrated intensities (or areas) of peaks were obtained by weighing.

RESULTS AND DISCUSSION

The 1700–100-cm^{-1} *region.* — The general profile of the laser-Raman spectra of the aqueous solutions of D-fructose, D-glucose, and sucrose seems to be independent of concentration; however, it shows differences between D-fructose on the one hand and D-glucose and sucrose on the other. Indeed, the spectrum background of D-fructose solutions exhibits only a maximum of intensity at 1640 cm^{-1}, corresponding to the bending band of water, whereas the spectra of D-glucose and sucrose solutions show, inside the bending band of H_2O, a staggered band between 1000 and 400 cm^{-1}, probably due[12] to the libration movement of H_2O (see Fig. 1). Moreover, we observed that the frequencies which mainly shift are located in the 1500–1200-cm^{-1} region.

This region is, indeed, the region where the groups which have a local symmetry, such as the CH_2 group, are found. We have previously[12] indicated that most of the modes of vibration of the CH_2 group (bending, wagging, twisting, and rocking) exhibit shifts of frequencies of $\sim$10 cm^{-1} when the concentration of D-glucose and sucrose solutions is changed. From among these different modes of vibration, the bending mode offers the opportunity of an interesting comparison with the bending of H_2O. This comparison was made between the frequencies of δ (HOH) and δ (HCH) in the spectra of each of the solutions of the three sugars, and between the intensities of these vibrations when the concentration was altered. As is shown in Fig. 2, H_2O and CH_2 have the same symmetry (C_{2v}) and the vibrations compared are from the same species (A_1).

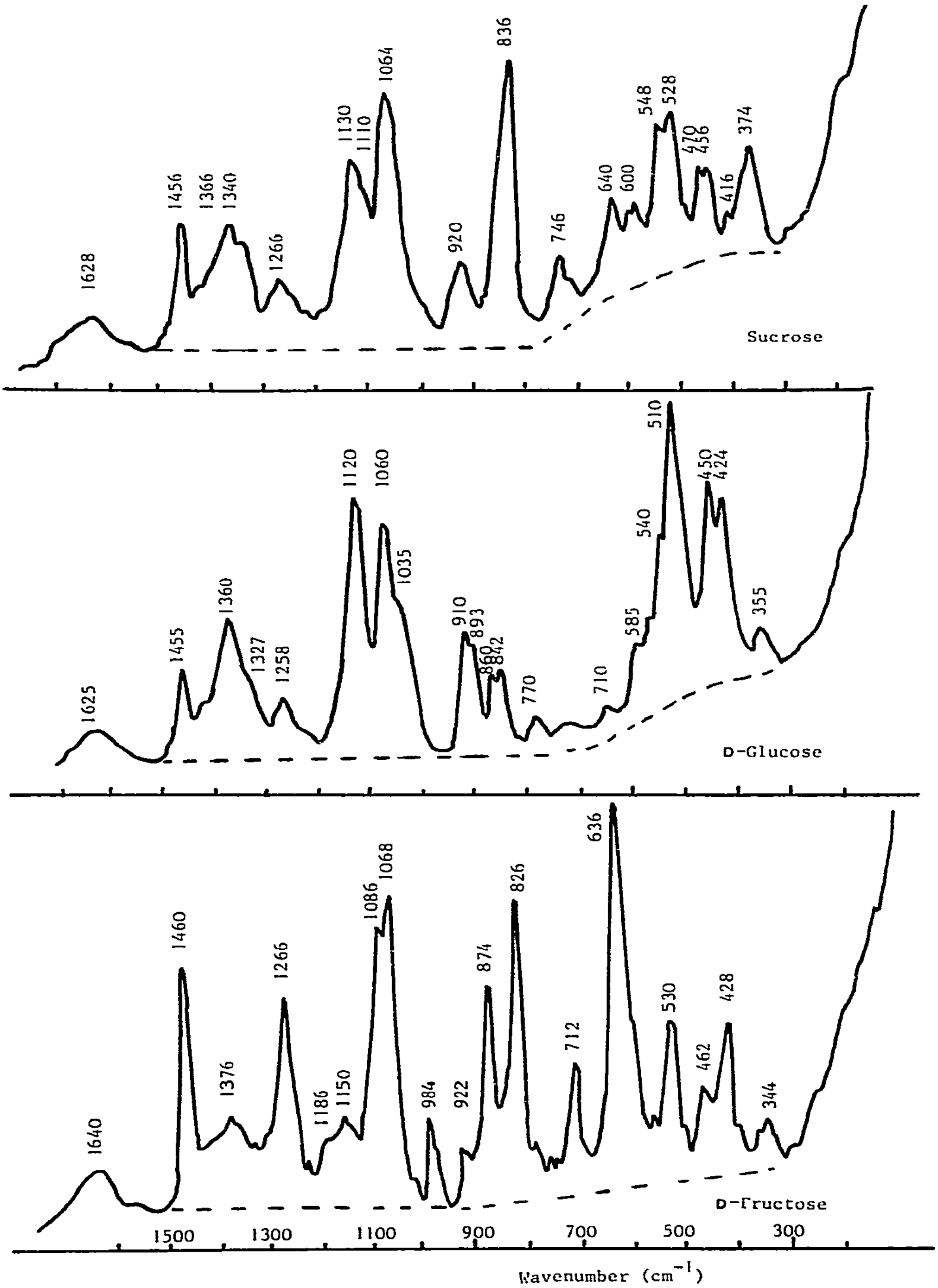

Fig. 1. General aspect of laser-Raman spectra of sucrose (24%, w/w), D-glucose (22%, w/w), and D-fructose (25%, w/w). (The curve ---- indicates the H_2O-libration background.)

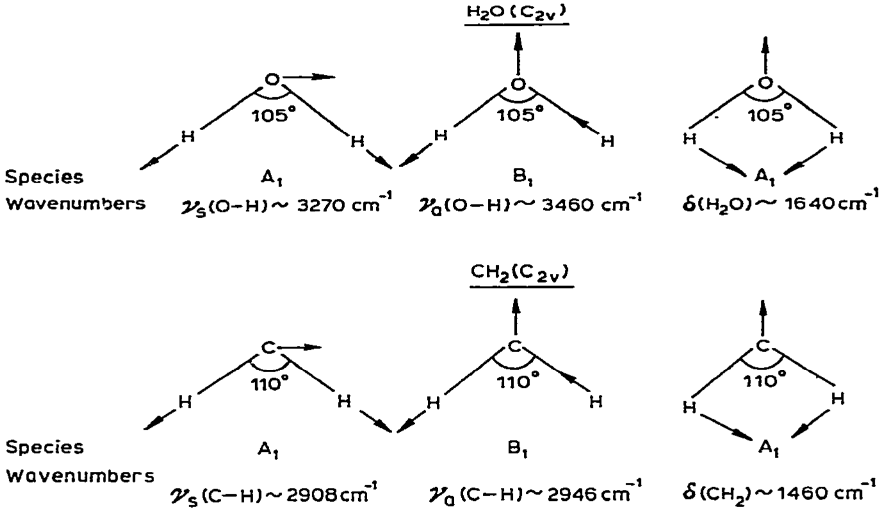

Fig. 2. Fundamental modes of vibration of CH_2 and H_2O.

TABLE I

VIBRATIONS AND BANDS OF D-FRUCTOSE, D-GLUCOSE, AND SUCROSE SOLUTIONS

D-*Fructose*			D-*Glucose*			*Sucrose*		
C (%, w/w)	cm^{-1}	ρ^a	*C (%, w/w)*	cm^{-1}	ρ^a	*C (%, w/w)*	cm^{-1}	ρ^a
$\delta(CH_2)$ vibration								
5	1460	0.69	5	1450	0.55	10.5	1450	0.56
10	1460	0.75	10.7	1452	0.77	18	1454	0.76
20	1460	0.74	19.5	1454	0.70	24.5	1456	0.91
30	1460	0.69	32.7	1455	0.68	33	1456	0.70
40	1460	0.76	41.8	1456	0.64	42.5	1458	0.69
50	1460	0.83	45	1458	0.66	53	1460	0.68
60	1460	0.85	50	1460	0.74	66	1460	0.71
70	1460	0.85	54	1460	0.71			
80	1460	0.90						
$\delta(H_2O)$ band								
5	1640	0.55	5	1614	0.47			
10	1640	0.63	10.7	1618	0.60	10.5	1622	0.55
20	1640	0.56	19.5	1620	0.69	18	1624	0.85
30	1640	0.52	32.7	1627	0.54	24.5	1624	0.56
40	1640	0.67	41.8	1630	0.61	33	1628	0.68
50	1640	0.53	45	1638	0.30	42.5	1634	0.66
60	1640	0.68	50	1640	0.82	53	1640	0.55
70	1640	0.67	54	1640	0.60	66	1640	0.57
80	1640	0.70						

[a] ρ = depolarization ratio.

Shifts of the 1460-cm^{-1} frequency. — The behavior of this vibration is not the same in the solutions of D-fructose (where it remains almost constant when the concentration is increased) as in the solutions of D-glucose and sucrose, where it increases from 1450 to 1460 cm^{-1} when the concentration is increased (see Table I). Inasmuch as the CH_2 groups are strongly individualized, the association of sugar and water by hydrogen bonds might not, as a rule, perturb them, so that there should be no frequency shifts. However, we observed that this frequency shifts with concentration in solutions of D-glucose and sucrose, but not of D-fructose. This frequency shift of $\delta(CH_2)$ in the spectra of D-glucose and sucrose solutions is probably due to the fluctuations of density that occur before the crystallization of the two sugars. Schneider *et al.*[13] concluded from viscosity measurements of sucrose solutions that a possible rearrangement of the molecules occurs at ~30–40% weight concentration and corresponds to the transition from solvated sucrose molecules (sucrose–water association) to the sucrose–sucrose molecular association. Tikhomiroff[14] demonstrated the existence of swarms of sucrose molecules, called protonuclei, in supersaturated sucrose solutions by means of various techniques (density, viscosity, thin-layer chromatography, and interferometry). The sucrose–sucrose association which precedes the crystallization was qualified by Kelly and Mak[15] as a polymerization leading to the critical germ, the hexamer.

A recent paper by Voilley *et al.*[16] showed that the first steps in the crystallization of D-glucose are accelerated by centrifugation, probably due to sedimentation of the aggregates formed in the homogeneous phase. However, even supersaturated solutions of D-fructose do not show heterogeneity of density, or other manifestations

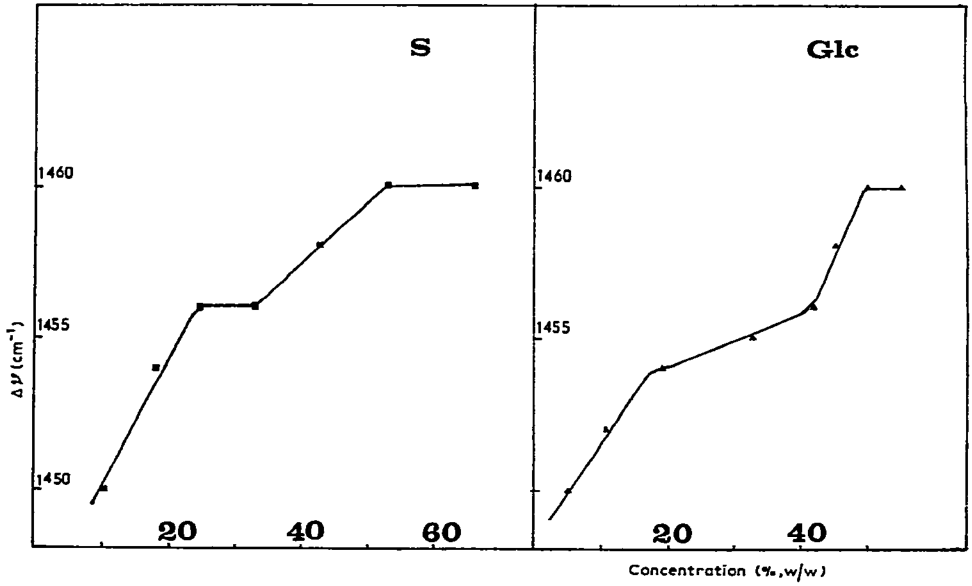

Fig. 3. Shifts of $\delta(CH_2)$ frequency *versus* concentration in aqueous solutions of sucrose (S) and D-glucose (Glc).

of the prenucleation phenomena. Therefore, the frequency shifts of the CH_2 bending-vibration may be assigned to the molecular association between sugar molecules in the D-glucose and sucrose solutions. This association produces aggregates of molecules that have a compact network comparable to crystalline organization, and this leads to the increase of the $\delta(CH_2)$ frequency as the concentration is raised. The evolution of the $\delta(CH_2)$ frequency in terms of mass concentration is shown in Fig. 3. There is a break in the increase of $\Delta\nu$ that probably corresponds to a modification of the kind of association (water–sugar or sugar–sugar). Thus, it seems probable that the prenucleation of D-glucose takes place at a concentration between 20 and 40% (w/w), and that of sucrose at ~35% (w/w). The latter result is in good agreement with that of Schneider *et al.*[13].

Shifts of the 1640-cm^{-1} frequency. — The water band situated at 1640 cm^{-1} shows frequency and intensity shifts that differentiate D-fructose from D-glucose and sucrose: these shifts are in the same direction as the modifications of $\delta(CH_2)$ frequency. An increase of $\delta(H_2O)$ frequency with concentration in D-glucose and sucrose solutions is, indeed, observed (see Table I). It has been demonstrated[17] that the bending frequency of water increases with the strength of the hydrogen bond, whereas the stretching frequency decreases. Accordingly, it seems probable that, in D-glucose and sucrose solutions at a certain concentration [~30% (w/w)], the sugar–sugar association is reinforced, and prenucleation occurs. This phenomenon is followed by the water–sugar dissociation. Around the swarms of sugar molecules, the water molecules may be structured, and the hydrogen-bond water–water reinforced. This "structure-maker" effect of sucrose on water in a concentrated solution [58% (w/w)] was noted by Walrafen[3], and Neal and Gorin[18] showed the "structure-breaker" effect of sucrose in dilute sucrose solutions. However, the lack of a $\delta(H_2O)$ frequency shift in D-fructose solutions could be ascribed to the probability that hydrogen bonds have the same energy in both D-fructose–water and water–water association. This assumption is confirmed by some of the physical properties of D-fructose solutions; the crystallization of D-fructose is very difficult to start, even in supersaturated solutions. Moreover, the fact that the temperature of melting of β-D-fructose (95–104°) is almost the same as the ebullition point of water (100°) leads to the conclusion that almost the same energy is needed for the dissociation of a D-fructose–D-fructose and a water–water hydrogen-bond.

Variation of $I(CH_2)/I(H_2O)$. — The Raman spectra shown in Fig. 1 indicate that the Raman line corresponding to the bending of CH_2 is sharp, whereas the H_2O bending-band is broad; this could be due to the likelihood that, under the band at ~1640 cm^{-1}, different species of water association are included. We also observed that the maximum intensity of $\delta(H_2O)$ is sharp at very low concentration (~5%), which supports the hypothesis of the "structure-breaker" effect of low concentrations of the sugar (in solution) on water, and the existence of H_2O monomers. The ratio of intensities, $I(CH_2)/I(H_2O)$, is plotted *versus* mass concentration in Figs. 4, 5, and 6; it shows discontinuities that seem to occur at concentrations characteristic of each of the three carbohydrates. These discontinuities could be assigned to modification

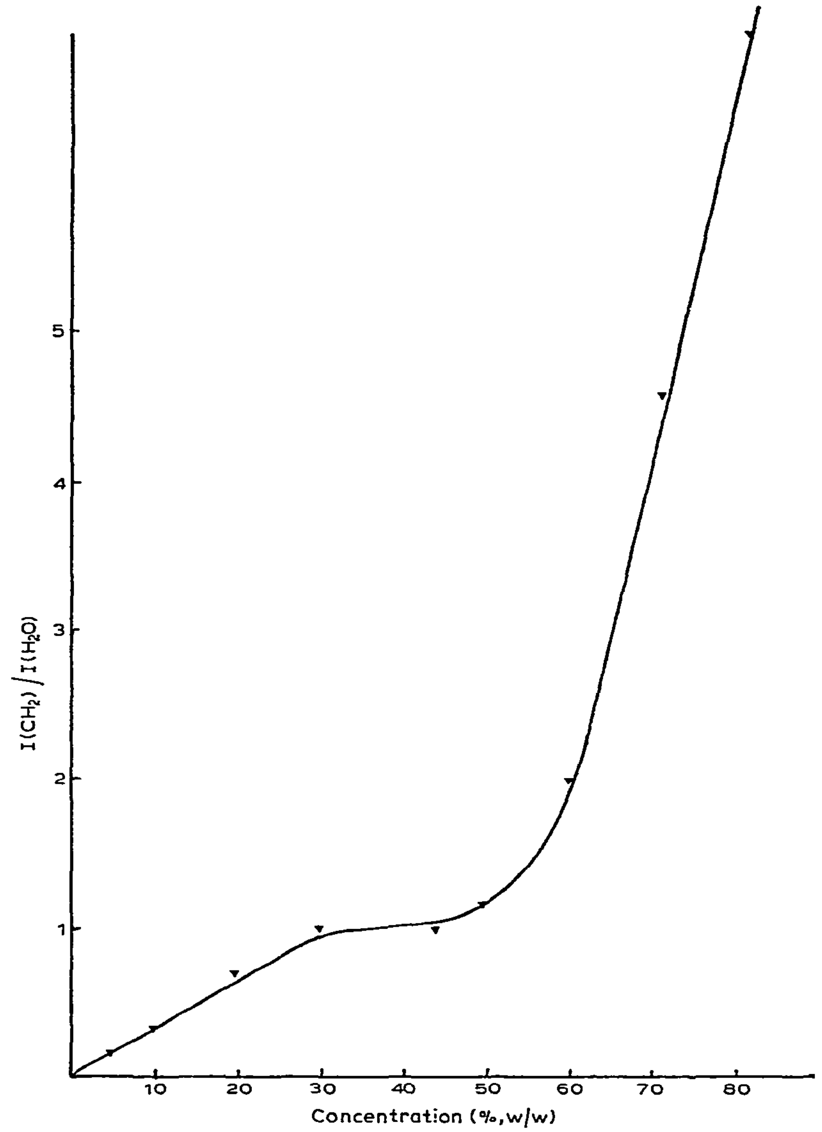

Fig. 4. Integrated, $\delta(CH_2)$: $\delta(H_2O)$ intensity ratio *versus* mass concentration in D-fructose solutions.

of the nature of the hydrogen bonding. Thus, even if there is no frequency shift in the D-fructose spectra, which means that the different species of hydrogen bond have the same energy, there could probably be a predominance of D-fructose–water bonds in the concentration region below 30%, whereas the probability of having D-fructose–D-fructose bonds seems to be higher at concentrations above 50% (w/w).

However, the boundary between the different species of hydrogen bonds occurs at 30% (w/w) in D-glucose solutions, and 35% (w/w) in sucrose solutions. These limits, apparently different, are comparable when expressed as degrees of under-

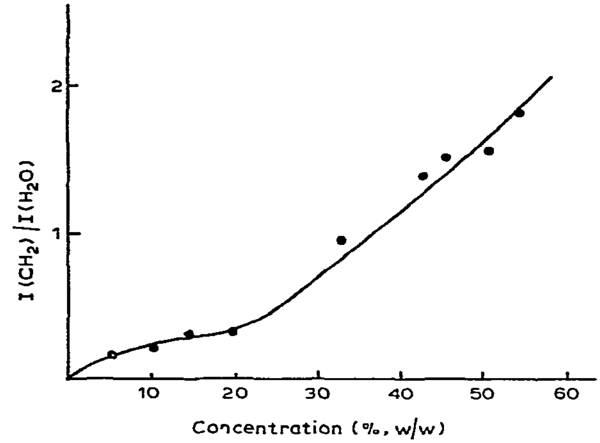

Fig. 5. Integrated, $\delta(CH_2):\delta(H_2O)$ intensity ratio *versus* mass concentration in D-glucose solutions.

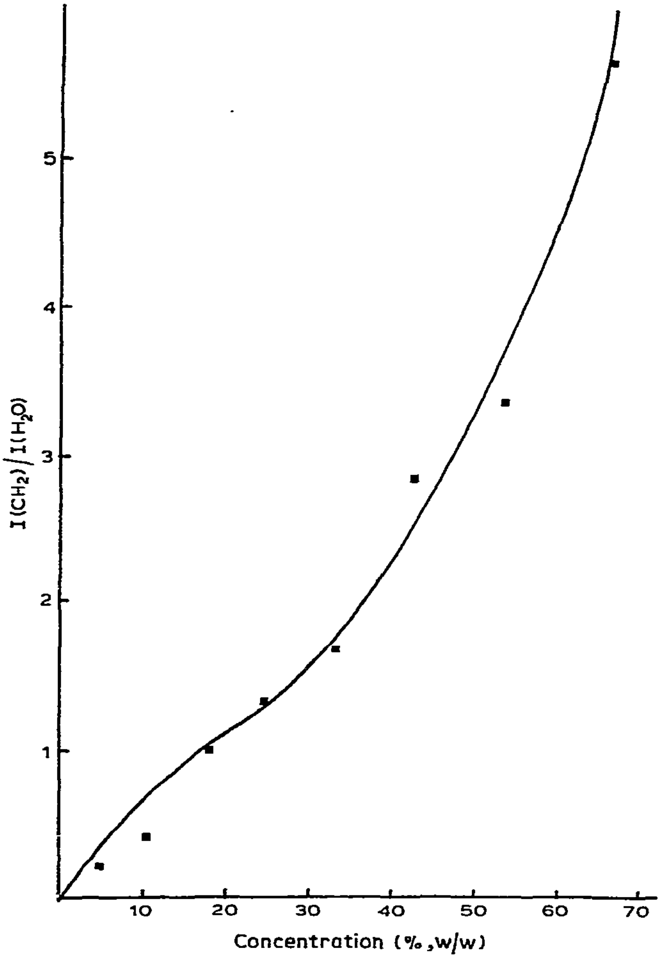

Fig. 6. Integrated, $\delta(CH_2):\delta(H_2O)$ intensity ratio *versus* mass concentration in sucrose solutions.

TABLE II

δ(OH) BAND OF D-FRUCTOSE, D-GLUCOSE, AND SUCROSE

D-*Fructose*			D-*Glucose*			*Sucrose*		
C (%, w/w)	$\nu_a(OH)$ (cm^{-1})	$\nu_s(OH)$ (cm^{-1})	*C (%, w/w)*	$\nu_a(OH)$ (cm^{-1})	$\nu_s(OH)$ (cm^{-1})	*C (%, w/w)*	$\nu_a(OH)$ (cm^{-1})	$\nu_s(OH)$ (cm^{-1})
0	3469	3268	0	3459	3268	0	3459	3268
5	3463	3274	6	3448	3272	6.2	3455	3290
10	3453	3262	11	3452	3278	12	3458	3284
20	3449	3282	15.5	3448	3276	18.7	3451	3270
30	3443	3271	19.5	3448	3276	29	3451	3272
40	3433	3278	30	3454	3272	38.7	3451	3276
55	3427	3274	40	3460	3260	47.4	3451	3276
80	3420	3282	55	3460	3268	63	3452	3286

saturation. Therefore, the molecular association or prenucleation should probably start at about 2/3rds of the saturation concentration, which is 79% for D-fructose, 46% for D-glucose, and 66% for sucrose at 20°.

The 3700–2700-cm^{-1} region of frequencies. — In this field of frequencies, we would expect to find frequencies of the stretching vibration ν(C–H) and ν(O–H). The CH vibration obviously belongs to the carbohydrate spectra, but the OH band includes H_2O stretching-vibrations, ν_s(O–H) and ν_a(O–H), as well as the OH vibration of each of the three sugars. It may be pointed out that the structure of lines corresponding to the vibrations ν(C–H) situated between 3000 and 2850 cm^{-1} permits differentiation of the three carbohydrates[4]. Indeed, the asymmetrical vibration ν_a(C–H) in CH_2 is stronger than ν_s(C–H) for D-fructose; the opposite is observed for D-glucose, and the spectrum of sucrose exhibits almost the same intensity for the two vibrations. The results are summarized in Table II. As is well known[19], the O–H stretching band of H_2O is an envelope including both isotropic and anisotropic components. Moreover, we observed that the maximum of intensity of the perpendicular spectrum, associated with the asymmetrical stretching-frequency ν_a(O–H), is more distinct than that of the parallel spectrum; therefore, the values reported correspond to the intensity maxima of the perpendicular spectra.

The investigation of this region of frequencies is, indeed, not so easy as that of the preceding region, because ν(O–H) in H_2O is perturbed by the vibration ν(O–H) in the sugars, which is located almost between the frequencies ν_s and ν_a of water; we observed, nevertheless, a shift of ~40 cm^{-1} for the frequency ν_a(O–H) in the spectra of D-fructose, whereas this frequency remains almost constant (~3455 cm^{-1} ±5 cm^{-1}) in the D-glucose and sucrose solution spectra. This shift might be characteristic of the state of association between water and D-fructose. It indicates that D-fructose possesses a greater aptitude than the other two carbohydrates to associate with water. This particular affinity for water hinders, or prevents, the prenucleation of D-fructose that is an essential stage in the formation of a crystalline network.

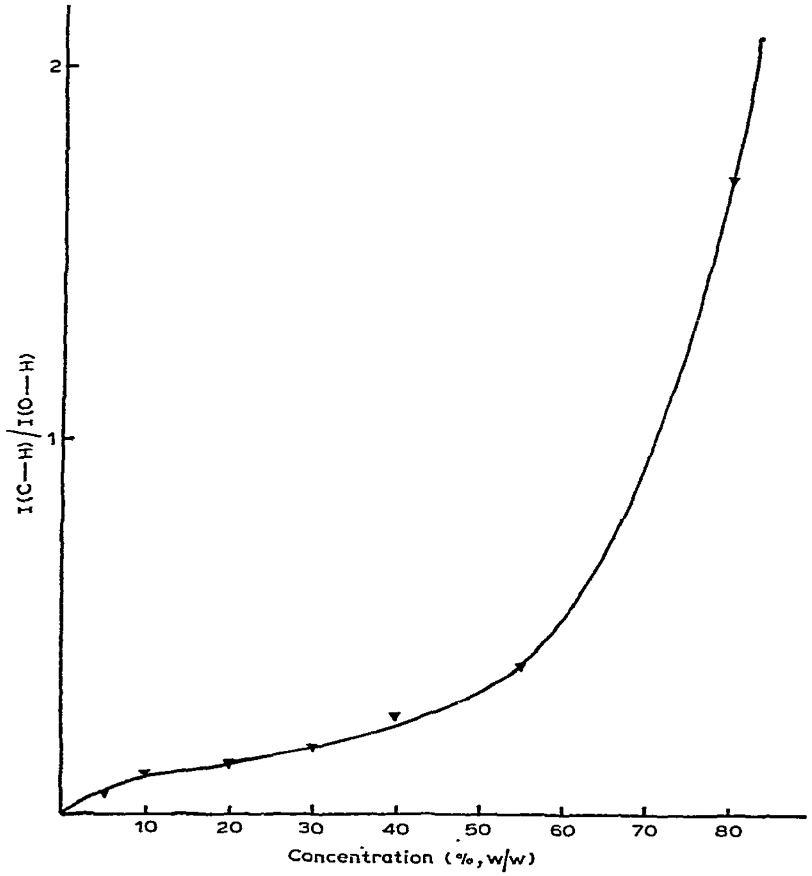

Fig. 7. Integrated, ν(CH):ν(OH) ratio *versus* mass concentration of D-fructose.

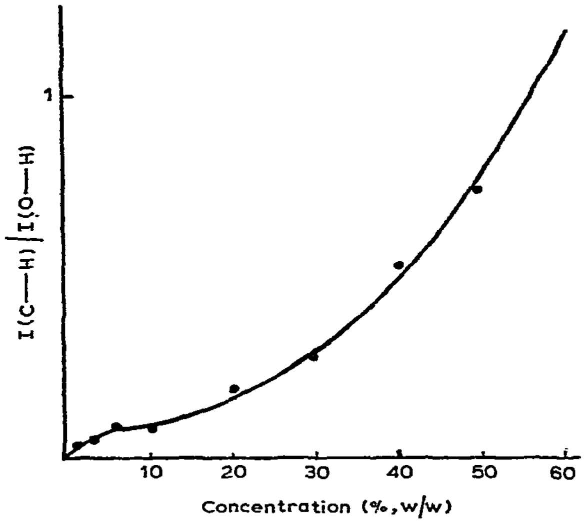

Fig. 8. Integrated, ν(CH):ν(OH) ratio *versus* mass concentration of D-glucose.

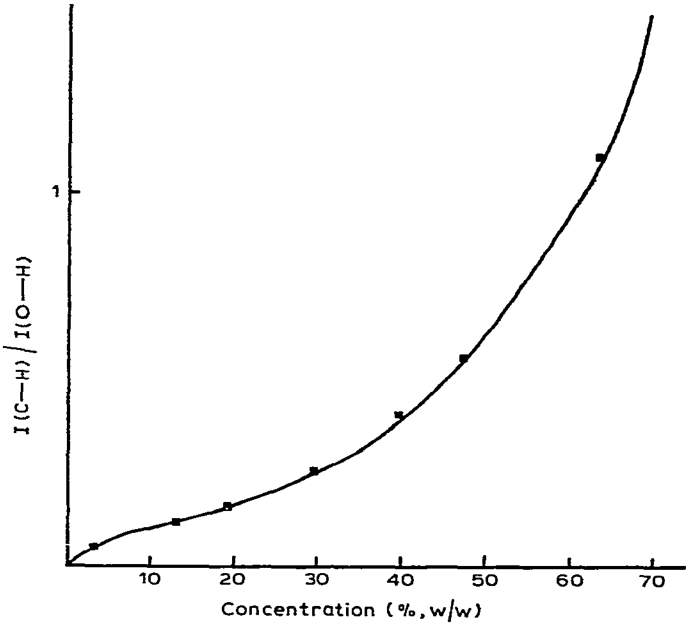

Fig. 9. Integrated, ν(CH) : ν(OH) ratio *versus* mass concentration of sucrose.

The profile of the curves representing I(CH)/I(OH) in terms of the mass concentration (Figs. 7–9) is comparable to that of the preceding curves. The discontinuities are less evident, probably due to the fact that I(OH) comprises three components, two of which come from water and the third from the carbohydrates.

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