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Refractive Index, Viscosity, and Solubility at 30 °C, and Density at 25 °C for the System Fructose + Glucose + Ethanol + Water

Adrian E. Flood* and Srisuda Puangsa

School of Chemical Engineering, Suranaree University of Technology, 111 University Avenue, Muang District, Nakhon Ratchasima, 30000, Thailand

Commercial crystalline fructose is currently prepared by chromatographic separation of aqueous solutions of fructose and glucose followed by crystallization in either aqueous or aqueous-ethanol solutions. It may be possible to use one or more crystallization steps instead of the chromatographic process, thus producing crystalline product more directly. In this study the solubility, refractive index, and viscosity of solutions of fructose + glucose + ethanol + water were measured at 30 °C, and the densities of solutions with solvent concentrations of 40, 60, and 80 mass % ethanol were measured at 25 °C. These properties will be useful for crystallization studies for this system.

Introduction

D-Fructose, $C_6H_{12}O_6$, is a monosaccharide widely used as a sweetener, largely due to its high sweetness value, although other physical and chemical properties also make it suitable for a number of products. Fructose is typically produced by hydrolysis of starch into glucose followed by isomerization to an aqueous solution of glucose and fructose. The product of this is a high-fructose syrup (HFS) that is (on a dry mass basis) approximately 42% fructose and 53% glucose with some residual higher carbohydrates (HFS-42). Higher purity syrups (such as HFS-90, which is 90% fructose on a dry mass basis) may be produced by chromatographic separation, while crystalline fructose is currently produced only from the high-purity syrups. Using the sweetness of sucrose as a basis (value 100), the sweetness of the crystalline form of fructose (β -D-fructopyranose) is approximately 180, while that of HFS-90 is only 106.¹ The difference is due to the noncrystallizing tautomers of fructose, which comprise approximately 30% of the fructose in solution, having lower sweetness than β -D-fructopyranose. HFS-42 has a sweetness of 92, which is lower than that of HFS-90, since glucose has low sweetness, approximately 65.

Crystalline fructose is currently prepared using either aqueous or aqueous-ethanolic crystallization of high-purity (90–95%) fructose syrups. Aqueous crystallization is made difficult by the high solubility of fructose in water (approximately 4.3 g of fructose/g of water at 30 °C), which not only affects the yield but also produces very highly viscous solutions. The fructose-water phase diagram is well-known,² and property data suitable for use in crystallization of aqueous fructose solutions by the addition of ethanol have also been determined for the system fructose + ethanol + water.³ Suitable processes for crystallizing fructose using ethanol as a nonsolvent are described in patents,^{4–6} and crystallization data have also been published.^{7,8} Processes crystallizing fructose or glucose directly from lower purity high-fructose syrups (HFS-42 for instance) are not currently used.

Published data on the solubility of sugars in solvents containing alcohols is limited (for example sucrose in

ethanol-water mixtures,⁹ xylose and mannose in ethanol-water mixtures,¹⁰ and glucose in ethanol-water mixtures¹¹). More interest has been shown recently, partly due to an interest in thermodynamic modeling of these systems particularly by the group of Macedo.^{12–14} There are solubility data for a very limited number of multiple-sugar solute systems (fructose and glucose in water¹⁵ and xylose and mannose in water¹⁶ are examples) and essentially no data for the solubility of multiple-sugar solutes in mixed solvents.

Experimental Section

Chemicals. D-(+)-Fructose, D-(+)-glucose anhydrous (both ACS, for analysis), and ethanol anhydrous (99.9% v/v, for analysis) were obtained from Carlo Erba Reagenti (Milan) and used without further purification.

Experimental Procedures. The solubility of fructose and glucose in solutions of ethanol and water was measured at 30 °C, with the solutions maintained at the desired temperature with an uncertainty of ± 0.2 °C. All determinations were made in sealed glass Schott bottles into which a known quantity of ethanol + water (of desired concentration) and a known quantity of anhydrous fructose were added. The ethanol concentration in the ethanol + water solution was known to an accuracy of 0.1 mg/g of solution. An amount of fructose was dissolved in the bottles, with the exact amount varying between bottles such that the experiments covered a range of points between the previously published systems glucose + ethanol + water^{11,12} and fructose + ethanol + water.³ An amount of crystalline anhydrous glucose sufficient to achieve at least 50% excess of glucose over the amount needed for saturation was added to each bottle, and the bottles were then shaken in an orbital shaking bath at 100 rpm and 30 °C until saturation was reached. After 24 h, the refractive index of the liquid was determined every 6 h to determine if saturation was complete. Saturation was complete within 7–10 days for all determinations.

This system proved difficult for accurate measurement of fructose and glucose concentrations. In most cases with sugars it has been preferable to determine concentrations using a gravimetric method, such as the total solids

* E-mail: adrianf@ccs.sut.ac.th. Facsimile: +66 44 224220.

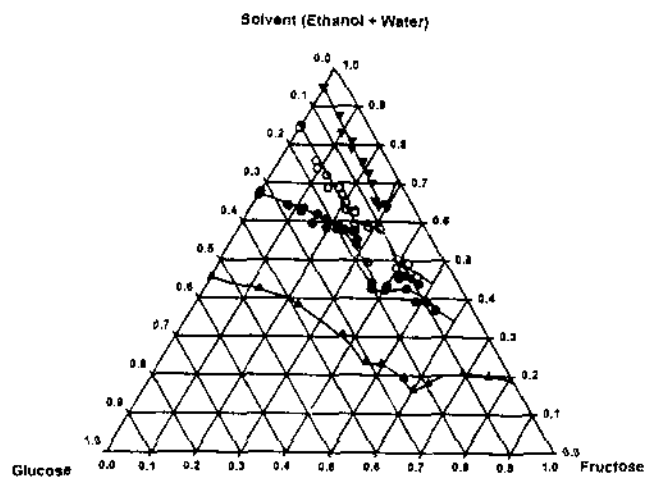


Figure 1. Solubility for the system fructose + glucose + ethanol + water at 30 °C. Solvent compositions: ▽, 80 mass % ethanol; ○, 60 mass % ethanol; ●, 40 mass % ethanol; ▲, 0 mass % ethanol.¹⁵

determination¹⁷ or the method of Peres and Macedo,¹² as these methods have excellent reproducibility. This type of method was not used in the current study, since an accurate determination of two solutes was required, and hence a HPLC method was used. It was also found that if samples containing high concentrations of ethanol but low concentrations of sugar were left to stand over several days, then a detectable (by HPLC) amount of an unknown reaction product formed, whereas this product did not form if the ethanol was removed from solution. The reaction product is not known, although the reaction may involve sugar dehydration. For this reason, saturated liquid samples of approximately 1 mL were taken from the Schott bottles, mass was determined to ± 0.1 mg in sealed weighing bottles, the samples were then partially dried at room temperature (approximately 30 °C) for 17 h to remove the bulk of the ethanol from the sample, and mass was again determined to ± 0.1 mg. Drying at high temperatures was not used because fructose tends to degrade at temperatures higher than 65 °C. After the drying step, the samples were diluted to approximately 1 g of solids/100 mL of solution by the addition of a known amount (± 0.1 mg) of distilled water, which was a suitable concentration for the HPLC method used. It should be noted that the drying process was not used to totally dry the sample but only to remove most of the ethanol so that the reaction between the ethanol and the sugars did not occur. After this sample preparation was carried out, the peak indicating the sugar-ethanol reaction product was not detected for any sample.

The diluted samples were filtered through a 0.45 μ m filter, and then a 2 μ L sample was injected onto a 250 mm \times 4 mm Aminex HPX-87C (Biorad, Bangkok, Thailand) column using a water mobile phase at a flow rate of 0.3 mL/min. The column temperature was 80 °C. Detection was with a diode array detector measuring UV at 192 nm. The uncertainty (95% probable error) in the concentration determinations, including the dilution and HPLC, was 0.002 g of glucose/g of solution and 0.003 g of fructose/g of solution. Duplicate solubility determinations showed that the uncertainty (95% probable error) in the solubility measurements was 0.005 g of sugar/g of solution for both fructose and glucose. Uncertainties in other variables, such as bath temperature, solvent composition, or saturation point, may be responsible for the duplicate bottles having larger uncertainties than were seen in the concentration measurement alone.

Table 1. Solubilities of D-(-)-Fructose and D-(+)-Glucose in Ethanol + Water at 30 °C

solvent comp (mass % ethanol)	solubility (g of sugar/g of solution)		
	glucose	fructose	total
40.0	0.332	0	0.332
	0.323	0	0.323
	0.280	0.076	0.356
	0.279	0.081	0.360
	0.247	0.118	0.365
	0.257	0.117	0.364
	0.247	0.158	0.405
	0.224	0.157	0.381
	0.210	0.182	0.392
	0.222	0.194	0.416
	0.193	0.217	0.410
	0.200	0.219	0.419
	0.178	0.326	0.504
	0.174	0.332	0.506
	0.176	0.284	0.460
	0.168	0.279	0.447
	0.168	0.259	0.427
	0.162	0.263	0.425
	0.191	0.233	0.424
	0.185	0.237	0.422
	0.203	0.370	0.573
	0.194	0.364	0.558
	0.173	0.404	0.577
	0.165	0.401	0.566
	0.128	0.418	0.546
	0.114	0.426	0.540
	0.124	0.450	0.574
	0.099	0.446	0.545
	0.120	0.486	0.606
	0.091	0.467	0.558
	0.095	0.512	0.607
	0.086	0.543	0.629
	0.000	0.718 ^a	0.718
60.0	0.150	0	0.150
	0.157	0	0.157
	0.161	0.081	0.242
	0.168	0.094	0.262
	0.155	0.124	0.279
	0.169	0.143	0.312
	0.147	0.185	0.332
	0.144	0.169	0.313
	0.149	0.203	0.352
	0.156	0.214	0.370
	0.138	0.234	0.372
	0.141	0.287	0.378
	0.154	0.254	0.408
	0.154	0.252	0.406
	0.126	0.281	0.407
	0.128	0.286	0.414
	0.103	0.312	0.415
	0.105	0.316	0.421
	0.119	0.400	0.519
	0.104	0.391	0.495
80.0	0.090	0.416	0.506
	0.086	0.422	0.508
	0.087	0.453	0.540
	0.084	0.456	0.540
	0	0.603 ^a	0.603
	0.049	0	0.049
	0.050	0	0.050
	0.053	0.071	0.124
	0.070	0.099	0.169
	0.066	0.143	0.209
	0.056	0.133	0.189
	0.060	0.186	0.246
	0.060	0.182	0.242
	0.059	0.214	0.273
	0.081	0.263	0.344
	0.066	0.235	0.301
	0.078	0.281	0.359
	0.061	0.302	0.363
	0.059	0.291	0.350
	0.000	0.266 ^a	0.266

^a From ref 3.

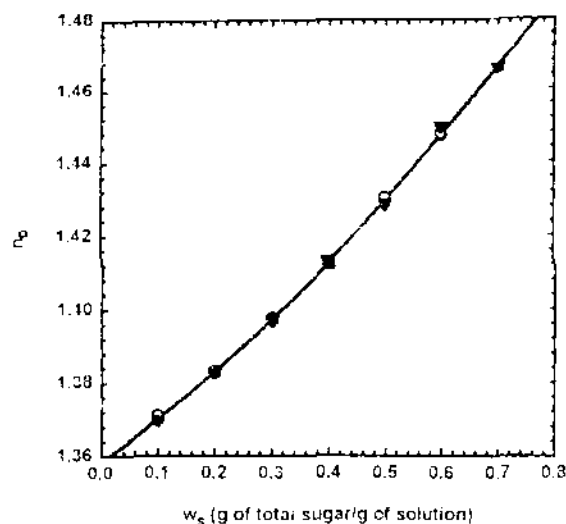


Figure 2. Refractive index for solutions of glucose and fructose in ethanol–water mixtures of 40 mass % ethanol. Ratio of glucose–fructose: ●, 1:1; ○, 2:1; ▼, 1:2.

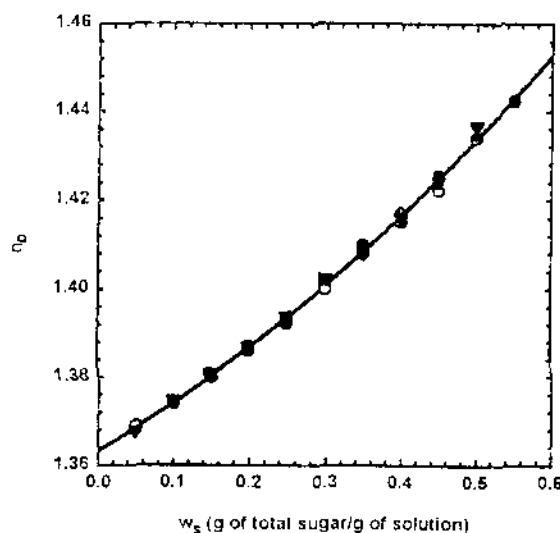


Figure 3. Refractive index for solutions of glucose and fructose in ethanol–water mixtures of 60 mass % ethanol. Ratio of glucose–fructose: ●, 1:1; ○, 2:1; ▼, 1:2.

Refractive index was measured for solvent compositions of 40, 60, and 80 mass % ethanol, and solute compositions of glucose–fructose equal to 1:1, 1:2, and 2:1. The total solute concentrations were chosen so that a range of data points up to approximately the saturation condition were measured. Duplicate refractive index determinations were made on an Abbe refractometer with temperature control to within ± 0.1 °C. The precision of the refractive index was ± 0.0005 refractive index unit.

Solution viscosity was measured in duplicate in a falling ball viscometer (Haake) with the sample viscosity determining which ball was used in the determination. The viscometer was jacketed, and water from a constant-temperature bath kept the viscometer temperature constant to within ± 0.1 °C. The solutions studied had viscosities in the range 1 to 1000 mPa·s, and hence balls 1 (2.4 g·cm⁻³, 15.81 mm) and 4 (8.13 g·cm⁻³, 15.2 mm) were used. These balls were calibrated against sugar solutions of known viscosity. The time period used in the viscosity determination was the average of eight measurements of the time required for the ball to travel the required

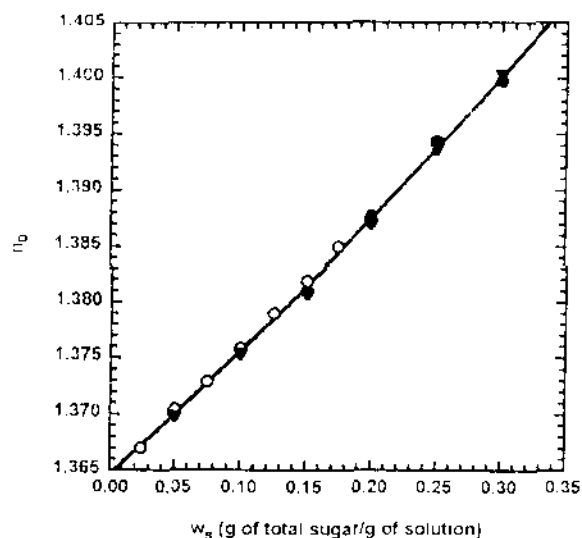


Figure 4. Refractive index for solutions of glucose and fructose in ethanol–water mixtures of 80 mass % ethanol. Ratio of glucose–fructose: ●, 1:1; ○, 2:1; ▼, 1:2.

distance. The error of the viscosity measurements is expected to be within 3%.

Solution density was measured in triplicate at 25 °C using 10 cm³ density determination bottles weighed to ± 0.1 mg. The uncertainty (95% probable error) of the density measurement was 0.0003 g·cm⁻³.

Results and Discussion

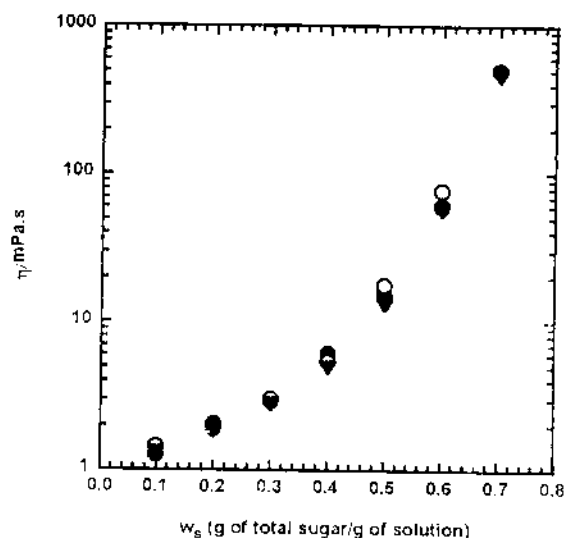
The solubilities of fructose and glucose in ethanol + water are plotted as a ternary diagram in Figure 1. Since the system contains four components, it is not easy to illustrate the data on a two-dimensional plot. The illustration is simplified by having one axis as total solvent (ethanol + water), with the lines on the plot depicting constant solvent composition (in the case of this study 40, 60, and 80 mass % ethanol). The data for the system fructose + glucose + water (equivalent to a 0 mass % ethanol line), which also appear on the plot, were taken from an earlier study at 30 °C.¹⁵ The three data points for pure fructose in ethanol + water (on the glucose axis) have been taken from a recent study by the same author.³ The solubility data from the present study are shown in Table 1.

Two studies^{11,12} have investigated the solubility of glucose in ethanol + water solutions; however, these studies were conducted at different temperatures, 35 °C for the former and 40 and 60 °C for the latter. For this reason, the solubility of glucose in ethanol + water was measured directly in this study. The study of Peres and Macedo¹⁴ gave interaction parameters for a modified UNIQUAC model (optimized using their own experimental results) which could be used to predict these solubility values. At 30 °C, this model predicts a solubility of 0.064 g of glucose/g of solution at 80 mass % ethanol, 0.270 g of glucose/g of solution at 60 mass % ethanol, and 0.459 g of glucose/g of solution at 40 mass % ethanol. These values are significantly higher than the experimental values in this work; however, it should be noted that the temperature used in this study is outside the range of temperatures on which the model is based (40 °C and 60 °C).

The solubilities of both glucose and fructose are decreased as the concentration of ethanol in the solvent is increased over the range of concentrations investigated in the study. The solubilities of glucose and fructose in ethanol

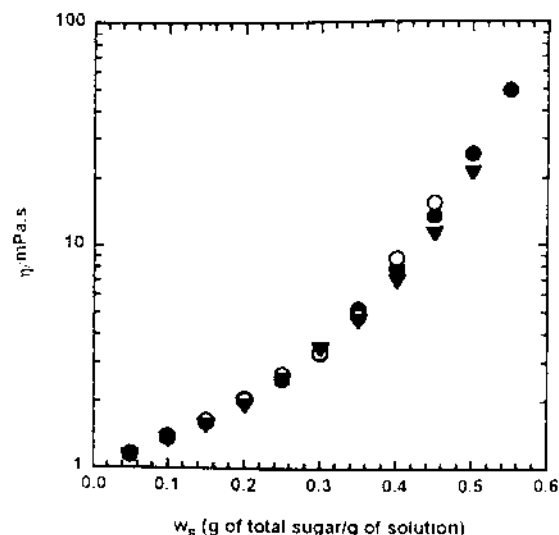
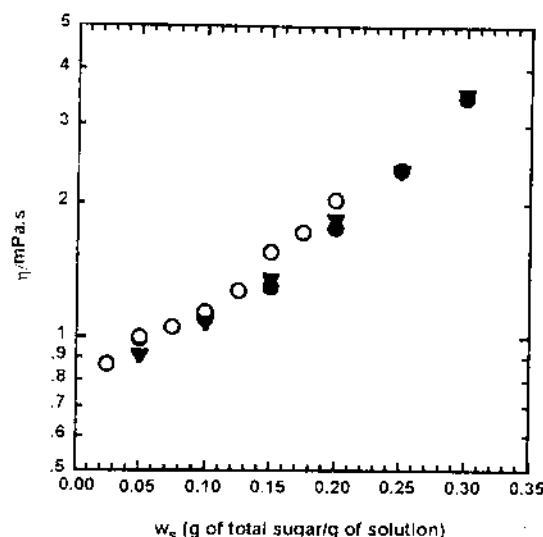
Table 2. Refractive Indexes (n_D) of D-(+)-Fructose + D-(+)-Glucose + Ethanol + Water Solutions at 30 °C

solvent comp ^a	tot sugar conc ^b	n_D at these ratios of glucose:fructose		
		1:1	2:1	1:2
40.0	0.100	1.3699	1.3709	1.3699
	0.200	1.3824	1.3830	1.3830
	0.300	1.3968	1.3973	1.3970
	0.400	1.4119	1.4124	1.4131
	0.500	1.4293	1.4298	1.4283
	0.600	1.4470	1.4474	1.4495
	0.700	1.4655		1.4660
	0.050	1.3681	1.3691	1.3680
	0.100	1.3740	1.3741	1.3748
60.0	0.150	1.3800	1.3807	1.3808
	0.200	1.3860	1.3870	1.3871
	0.250	1.3893	1.3932	1.3942
	0.300	1.4016	1.4000	1.4025
	0.350	1.4099	1.4080	1.4080
	0.400	1.4170	1.4151	1.4161
	0.450	1.4255	1.4221	1.4243
	0.500	1.4340		1.4360
	0.550	1.4424		
80.0	0.025		1.3670	
	0.050	1.3701	1.3704	1.3699
	0.075		1.3729	
	0.100	1.3758	1.3758	1.3755
	0.125		1.3790	
	0.150	1.3810	1.3819	1.3809
	0.175		1.3850	
	0.200	1.3877	1.3872	1.3871
	0.250	1.3944		1.3938
	0.300	1.3998		1.4005

^a mass % ethanol. ^b g of sugar/g of solution.**Figure 5.** Viscosity for solutions of glucose and fructose in ethanol–water mixtures of 40 mass % ethanol. Ratio of glucose:fructose: ●, 1:1; ○, 2:1; ▼, 1:2.

at 30 °C are 0.0036 g of glucose/g of solution (extrapolated from the data at 40 and 60 °C using the modified UNIQUAC model of Peres and Macedo¹⁴) and 0.035 g of fructose/g of solution,³ respectively, so it is expected that the solubility is monotonically decreasing as the ethanol content of the solvent increases.

When the solvent is 80 mass % ethanol, there is a “salting in” effect whereby the total sugar concentration is higher when both glucose and fructose occur together in solution compared to where only one solute appears. This maximum sugar concentration is 0.36 g of sugar/g of solution (compared to 0.050 g of glucose/g of solution or 0.266 g of fructose/g of solution at saturation for the one-

**Figure 6.** Viscosity for solutions of glucose and fructose in ethanol–water mixtures of 60 mass % ethanol. Ratio of glucose:fructose: ●, 1:1; ○, 2:1; ▼, 1:2.**Figure 7.** Viscosity for solutions of glucose and fructose in ethanol–water mixtures of 80 mass % ethanol. Ratio of glucose:fructose: ●, 1:1; ○, 2:1; ▼, 1:2.

solute mixtures). For both 40 and 60 mass % ethanol, the minimum total sugar concentration is when glucose is the only solute (0.33 and 0.15 g of glucose/g of solution, respectively), while the maximum occurred when fructose was the only solute (0.718 and 0.603 g of fructose/g of solution, respectively). The solubility lines for 60 and 80 mass % ethanol show similar behavior when glucose is the crystallizing form but markedly different behavior when fructose is the crystallizing form. It is noticeable that the solubility of glucose decreases most rapidly between 40 and 60 mass % ethanol, while the solubility of fructose decreases most rapidly between 60 and 80 mass % ethanol. The solubility curve for the system for 40 mass % ethanol is most similar to the behavior of the ternary system fructose + glucose + water.

The solubility curve for the system D-(+)-fructose + D-(+)-glucose + water¹⁵ shows two distinct eutectics: one where the crystal form of glucose changes from glucose monohydrate to anhydrous glucose and one where fructose becomes the preferred crystalline phase. The first of these points is not clearly evident in the four-component system, although it may still exist. The second eutectic point is

Table 3. Viscosities of D-(+)-Fructose + D-(+)-Glucose + Ethanol + Water Solutions at 30 °C

solvent comp ^a	tot sugar conc ^b	η /(mPa·s) at these ratios of glucose-fructose		
		1:1	2:1	1:2
40.0	0.100	1.26	1.43	1.35
	0.200	2.04	1.90	1.92
	0.300	2.92	3.00	2.89
	0.400	6.12	5.37	5.12
	0.500	15.4	17.6	13.7
	0.600	60.7	77.2	59.6
	0.700	502		485
60.0	0.050	1.16	1.15	1.16
	0.100	1.39	1.36	1.37
	0.150	1.61	1.64	1.60
	0.200	2.06	2.02	1.98
	0.250	2.54	2.66	2.58
	0.300	3.33	3.36	3.59
	0.350	5.24	4.89	4.79
	0.400	8.00	8.85	7.18
	0.450	13.6	15.7	11.7
	0.500	25.6		21.7
	0.550	49.2		
80.0	0.025		0.86	
	0.050	0.99	1.00	0.91
	0.075		1.05	
	0.100	1.10	1.14	1.08
	0.125		1.27	
	0.150	1.30	1.56	1.36
	0.175		1.72	
	0.200	1.76	2.03	1.85
	0.250	2.38		2.39
	0.300	3.46		3.55

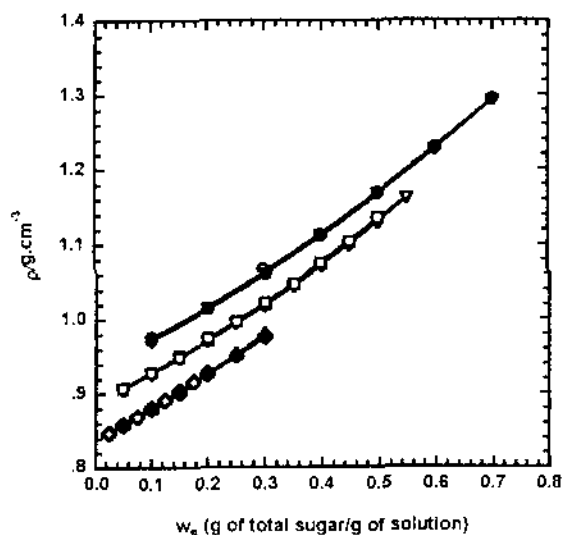
^a mass % ethanol. ^b g of sugar/g of solution.

clearly evident. Work is in progress to determine the preferred crystalline phase for a range of temperatures and concentrations in this system.

In general, it is preferable to measure solubility through both dissolution and crystallization experiments, which will bracket the solubility by approach from both above and below. In the present study, measurement through crystallization was not attempted, since it is possible that glucose monohydrate would crystallize under certain conditions and thus the water content of the solvent would be reduced as crystallization progresses. The present study uses only dissolution of anhydrous sugars, which will not alter the solvent composition.

The refractive indexes for solutions of D-(+)-fructose + D-(+)-glucose + ethanol + water with solvent compositions of 40, 60, and 80 mass % ethanol are shown in Figures 2, 3, and 4, respectively. It is clear from these diagrams that the proportion of glucose to fructose in the solution does not have a significant effect on the refractive index at any of the solvent compositions studied. This is significant in that it shows that refractive index will give no information on the solute ratio in solution, although it is still useful as a measure of total solute for this system. As the solvent ratio increases, the refractive index of infinitely dilute solutions increases slightly, probably as a result of the differences in the refractive indexes of ethanol ($n_D = 1.3594$ at 25 °C) and water ($n_D = 1.3325$ at 25 °C);¹⁸ however, the change in refractive index due to changes in solvent composition is not as significant at higher sugar concentrations. The refractive index data for the system are shown in Table 2.

Viscosities for the system D-(+)-fructose + D-(+)-glucose + ethanol + water for solvent compositions of 40, 60, and 80 mass % ethanol are shown in Figures 5, 6, and 7, respectively. The viscosity for the systems is strongly dependent on the total sugar content, with increasing

**Figure 8.** Density for solutions of fructose and glucose in ethanol-water mixtures. Curves are 40 mass % ethanol with the following ratios of glucose-fructose (●, 1:1; ○, 2:1; ▼, 1:2), 60 mass % ethanol with the following ratios of glucose-fructose (▽, 1:1; ■, 2:1; □, 1:2), and 80 mass % ethanol with the following ratios of glucose-fructose (◆, 1:1; ◇, 2:1; ▲, 1:2).**Table 4. Densities of D-(+)-Fructose + D-(+)-Glucose + Ethanol + Water Solutions at 25 °C**

solvent comp ^a	tot sugar conc ^b	ρ /(g·cm ⁻³) at these ratios of glucose-fructose		
		1:1	2:1	1:2
40.0	0.100	0.9726	0.9757	0.9724
	0.200	1.0140	1.0143	1.0162
	0.300	1.0604	1.0661	1.0625
	0.400	1.1115	1.1123	1.1141
	0.500	1.1691	1.1686	1.1700
	0.600	1.2285	1.2280	1.2287
	0.700	1.2944		1.2948
	0.800	1.3603		1.3607
60.0	0.050	0.9070	0.9071	0.9074
	0.100	0.9285	0.9282	0.9283
	0.150	0.9497	0.9502	0.9489
	0.200	0.9722	0.9717	0.9740
	0.250	0.9957	0.9963	0.9964
	0.300	1.0198	1.0200	1.0221
	0.350	1.0459	1.0466	1.0470
	0.400	1.0724	1.0739	1.0755
	0.450	1.1006	1.1030	1.1044
	0.500	1.1310		1.1356
	0.550	1.1640		
80.0	0.025		0.8473	
	0.050	0.8583	0.8587	0.8573
	0.075		0.8694	
	0.100	0.8804	0.8806	0.8793
	0.125		0.8918	
	0.150	0.9019	0.9038	0.9011
	0.175		0.9158	
	0.200	0.9259	0.9280	0.9250
	0.250	0.9503		0.9520
	0.300	0.9763		0.9793

^a mass % ethanol. ^b g of sugar/g of solution.

solute concentration giving very strongly increasing viscosity. The highest viscosities recorded (around 500 mPa·s) were for 40 mass % ethanol, where the high solubility allows for high solute concentrations. The solutions measured were all undersaturated: since sugar solutions may be held at solute concentrations substantially higher than saturation without crystallization, the viscosities for the system are potentially very high. The viscosity is weakly dependent on the ethanol content of the solvent, with higher ethanol contents giving slightly lower viscosities,

although this dependence is much weaker than the dependence on solute concentration. The viscosity of pure ethanol at 30 °C (0.964 mPa·s) is greater than that of water (0.815 mPa·s),¹⁸ so the behavior in this system is unusual, although it is probably due to differences in the solution structures. The solute (glucose-fructose) ratio has no significant effect on the viscosity over the range of values in this study. Data for the viscosity of the system are given in Table 3.

The densities of solutions of D-(+)-fructose + D-(+)-glucose + ethanol + water are shown in Figure 8. The density of the solution is not significantly affected by the solute (glucose-fructose) ratio in solution, although this is likely due to the two solutes having very similar densities. The densities of solid anhydrous glucose and fructose are 1.562 g·cm⁻³ (at 18 °C) and 1.600 g·cm⁻³ (at 20 °C), respectively.¹⁹ The densities of the solutions are strongly (and nonlinearly) dependent on the total sugar content, with the density increasing with increasing sugar content. If a power law was fitted to the data, the exponent would be of the order 1.15 to 1.30 for all solvent compositions studied, with higher ethanol contents giving smaller exponents. The ethanol content of the solvent has a significant effect on the density, with solutions having higher ethanol content displaying lower density for the same sugar content, as would be expected from the differences in density between the two solvents. The densities of pure ethanol and water at 30 °C are 0.783 g·cm⁻³ and 1.023 g·cm⁻³, respectively.¹⁸ Density data for the system are given in Table 4.

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