(7) Ambrose, D.; Ellender, J. H.; Gundry, H. A.; Lee, D. A.; Townsend, R. *J. Chem. Thermodyn.* **1981**, *13*, 795.
(8) Ambrose, D.; Ellender, J. H.; Lees, E. B.; Sprake, C. H. S.; Townsend, R. *J. Chem. Thermodyn.* **1975**, *7*, 453.
(9) *TRC-Thermodynamic Tables-Non-Hydrocarbons*. Thermodynamics Research Center, The Texas A&M University System: College Station, TX, extant 1965 (a-5370); 1974 (a-7240); 1984 (a-5450) (loose-leaf data sheets).

Received for review January 17, 1989. Revised September 18, 1989. Accepted October 2, 1989.

Apparent Molar Volumes and Viscosities of Some α- and α,ω-Amino Acids in Aqueous Ammonium Chloride Solutions at 298.15 K

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Apparent molar volumes and viscosity *B* coefficients of some α-, substituted α-, and α,ω-amino acids have been determined in water and aqueous ammonium chloride solutions at 298.15 K with a vibrating-tube digital density meter and a modified Cannon–Ubbelhode suspended-level viscometer. An increase in the volume of transfer of amino acids from water to aqueous ammonium chloride solutions and of viscosity *B* coefficients with increasing electrolyte concentration has been explained due to strong interactions of NH_4^+ and Cl^- with the charged centers of the zwitterions compared to ion–nonpolar-group interactions. The interactions have been rationalized in terms of the cosphere overlap model.

Introduction

Thermodynamic data for amino acids and peptides facilitate interpretation of results of even relatively complex biomolecules where a simple additivity rule is applicable. Volumetric and viscometric properties and changes in enthalpy and free energy give useful information about interactions in solutions and have been used to draw important conclusions regarding protein unfolding (*1*) and the extent of hydrophobic interactions of nonpolar side chains (*2*). Differential affinities of amino acid side chains for water have also been reported (*3*). Salt-induced electrostatic forces are known to play a role in modifying the protein structure (*4*) by affecting properties like solubility, denaturation, and activity of enzymes (*5*, *6*). Amino acids are model compounds for interpretation of the behavior of more complex protein molecules in solution. Their equilibrium properties in solution depend on the relative magnitude of electrostriction caused by the polar end groups, the structure-enforcing influence of the hydrophobic alkyl groups, and the extent of interaction between the hydrophilic and hydrophobic groups. Changes in enthalpy, free energy, and volume have been rationalized in terms of these interactions and have helped in evaluating, interalia, the extent of solvation. For some amino acid–water systems in the presence of salts, the heat capacity and free energy of transfer from aqueous to ionic environment have been reported (*7*, *8*). Recently, Ahluwalia et al. (*9–11*) reported apparent molar volumes of amino acids in aqueous salt and carbohydrate solutions. Ogawa, Mizutani, and Yasuda (*12*) and also Sandhu (*13–15*) have studied the viscosities and apparent molar volumes of amino acids in mixed aqueous solutions. Partial molar volumes of amino acid mixtures in water have also been reported (*16*).

Since ammonium and alkyl-substituted ammonium salts are known to influence the conformational stability of proteins in solution (*17*), a systematic study of their influence on volumetric and viscometric behavior of a series of amino acids is of current interest. Both these properties are sensitive to specific interactive changes in solution. In this paper, apparent molar volumes and viscosities of some α-, substituted α-, and α,ω-amino acids in ammonium chloride solution are reported.

Experimental Section

The α- and α,ω-amino acids chosen for the study were glycine, DL-α-alanine, DL-α-aminobutyric acid, DL-norvaline, DL-norleucine, β-alanine, γ-aminobutyric acid, δ-aminovaleric acid, and ϵ-aminocaproic acid. The α-amino acids with substituted side chain were L-serine, L-threonine, DL-valine, L-glutamine, DL-methionine, and DL-phenylalanine. All amino acids, obtained from Sigma, were of chromatographic purity and were used after drying at ca. 350 K, followed by vacuum desiccation over P_2O_5 for ca. 48 h. Ammonium chloride (B.D.H, AnalaR) was dried for ca. 72 h at ca. 373 K before use. All solutions were prepared afresh by weight with deionized, double-distilled water degassed by boiling.

Solution densities were measured with an Anton Paar (Model DMA 60/601) vibrating-tube digital density metal with precision of $\pm 3 \times 10^{-6}$ g cm^{-3}. The temperature in the measuring cell was controlled to ± 0.01 K and monitored with a Paar Model DT-100-20 digital thermometer. The density meter was calibrated everyday with use of the literature data for densities of dry air (*18*) and water (*19*) at 298.15 K. The partial molar volumes of sodium chloride and ammonium chloride determined with the setup agreed within $\pm 0.5\%$ of the reported values (*20*, *21*). The partial molar volumes of typical amino acids in water also agreed within $\pm 0.5\%$ of the values reported by Jolicoeur (*22*). Density measurements were made on solutions of amino acids in water and also in 0.25, 0.50, 1.25, and 2.00 mol kg^{-1} aqueous NH_4Cl at 298.15 K. The concentration range of amino acids was up to ca. 0.5 mol L^{-1}, except for those with low solubility where it was limited to ca. 0.08 mol L^{-1}.

Viscosities were measured with a modified Cannon–Ubbelhode suspended-level viscometer (efflux time for water, ca. 400 s at 298.15 K) vertically mounted on a brass frame in a water thermostat. The kinetic energy correction term was found to be negligible. Viscosity results of test measurements on 0–2 mol kg^{-1} of ammonium chloride solutions in water agreed within $\pm 0.25\%$ of the literature values (*23*) while the precision of the measurements was $\pm 5 \times 10^{-6}$ g cm^{-1} s^{-1}. Viscosities of so-

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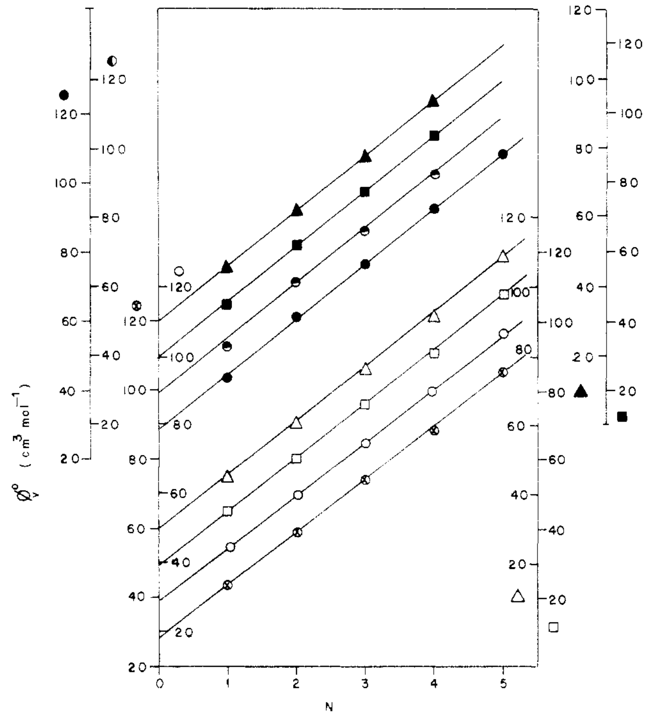

Figure 1. Variation of limiting apparent molar volume of amino acids with N, the number of carbon atoms in the amino acid backbone at 298.15 K: α-amino acids, 0.5 m NH_4Cl (●), 1.0 m NH_4Cl (◐), 1.5 m NH_4Cl (■), 2.0 m NH_4Cl (▲); α,ω-amino acids, 0.5 m NH_4Cl (⊗), 1.0 m NH_4Cl (○), 1.5 m NH_4Cl (□), 2.0 m NH_4Cl (△).

lutions of glycine, DL-α-alanine, DL-α-aminobutyric acid, β-alanine, γ-aminobutyric acid, and L-serine in aqueous solution and 0.25, 0.50, 1.25, and 2.00 mol kg^{-1} of aqueous ammonium chloride were measured at 298.15 K.

Results

The apparent molar volumes (ϕ_v) of amino acids in binary (water + amino acid) and ternary (water + ammonium chloride + amino acid) systems were evaluated from solution densities (ρ) with the relation

$$\phi_v = \frac{1000(\rho_0 - \rho)}{c\rho_0} + \frac{M}{\rho_0} \qquad (1)$$

where M and c are the molar mass and the molarity of the amino acid, respectively, and ρ_0 is the density of the solvent (water or aqueous ammonium chloride solution). ϕ_v varied linearly with c and could be least-squares fitted to the equation

$$\phi_v = \phi_v{}^\circ + S_v c \qquad (2)$$

where $\phi_v{}^\circ$ is the limiting apparent molar volume of the amino acid (which equals its infinite dilution partial molar volume) and S_v is the experimental slope. If the variation of ϕ_v with c showed considerable scatter, $\phi_v{}^\circ$ was determined either graphically or was taken as the average of all the ϕ_v values where slopes were near zero. The regression coefficients of eq 2 for α-, substituted α-, and α,ω-amino acids in water and in aqueous solutions of 0.25, 0.5, 1.25, and 2.00 mol kg^{-1} of NH_4Cl are presented in Table I; volumes of transfer, $\phi_v{}^\circ$(tr) of amino acid from water to aqueous electrolyte solutions are also included. Variations of $\phi_v{}^\circ$ with the number of carbon atoms (N) in the alkyl chain, both for α- and α,ω-amino acids in presence of NH_4Cl are linear (Figure 1). It is seen that, for both groups of amino acids, while the slopes (which are the volume contributions by the CH_2 group) in the presence of 0.5–2.0 mol kg^{-1} of NH_4Cl show almost uniform volume contribution of ca. 16 cm^3 mol^{-1}, which is of the order reported for amino acids in water (*24*), the intercept (which is the volume contribution by polar head groups) increases with an increase of NH_4Cl concentration, albeit to the same extent, both for α- and α,ω-amino acids. The increase is ca. 2.0 cm^3 mol^{-1} as the NH_4Cl concentration increases from 0.5 to 2.0 mol kg^{-1}.

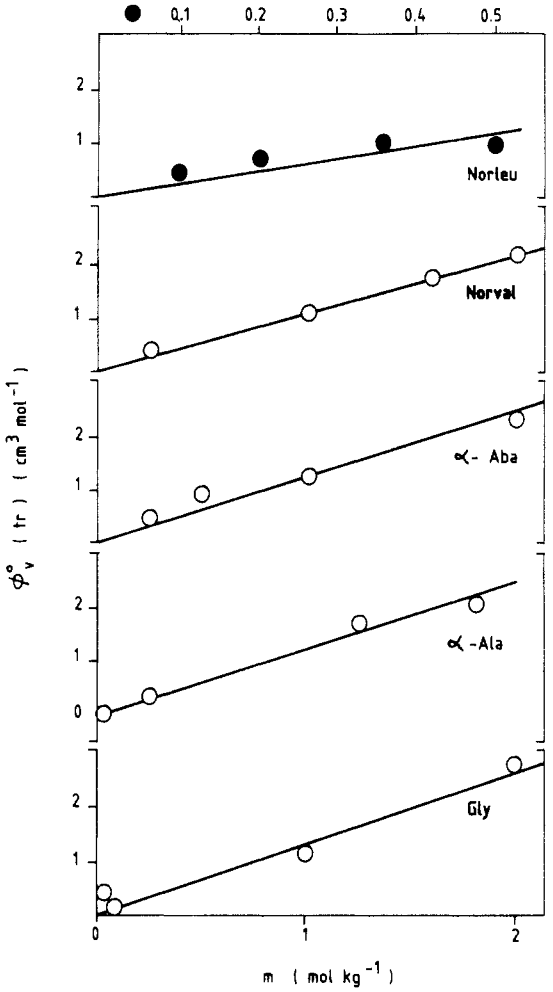

Figure 2. Plot of limiting apparent molar volumes of transfer $\phi_v{}^\circ$(tr) of α-amino acids from water to aqueous NH_4Cl solutions vs m at 298.15 K.

$\phi_v{}^\circ$(tr) for the amino acids calculated from the relation

$$\phi_v{}^\circ(\text{tr}) = \phi_v{}^\circ(\text{aq electrolyte}) - \phi_v{}^\circ(\text{aq})$$

are seen to be positive (increase with an increase in NH_4Cl molality, m). Variation of $\phi_v{}^\circ$(tr) with m is seen to be linear for α-amino acids (Figure 2); it tends to be nonlinear at $m \geq 1.25$ mol kg^{-1} for α,ω-amino acids (Figure 3). $\phi_v{}^\circ$(tr) for α- and α,ω-amino acids as a function of alkyl chain length (N) in 0.5, 1.0, 1.5, and 2.0 mol kg^{-1} of NH_4Cl solutions (Figures 4 and 5) show that while slopes for α,ω-amino acids are positive, those for α-amino acids are negative.

$\phi_v{}^\circ$ for substituted α-amino acids at a typical 1.25 mol kg^{-1} of NH_4Cl concentration are given in Table II. The trend of variation of $\phi_v{}^\circ$ as a function of molar mass of the amino acids (Figure 6) follows the order

Gly < α-Ala < Ser < Val < Thr < Glu < Meth < Phe

The solvation number of glycine and some substituted amino acids in aqueous solutions with the partial molar volumes from this study were evaluated (Table III) by the method of Millero (*25*).

Variation of relative viscosity η_r ($=\eta/\eta_0$) with the concentration of amino acid in solution (c) at a given electrolyte (NH_4Cl)

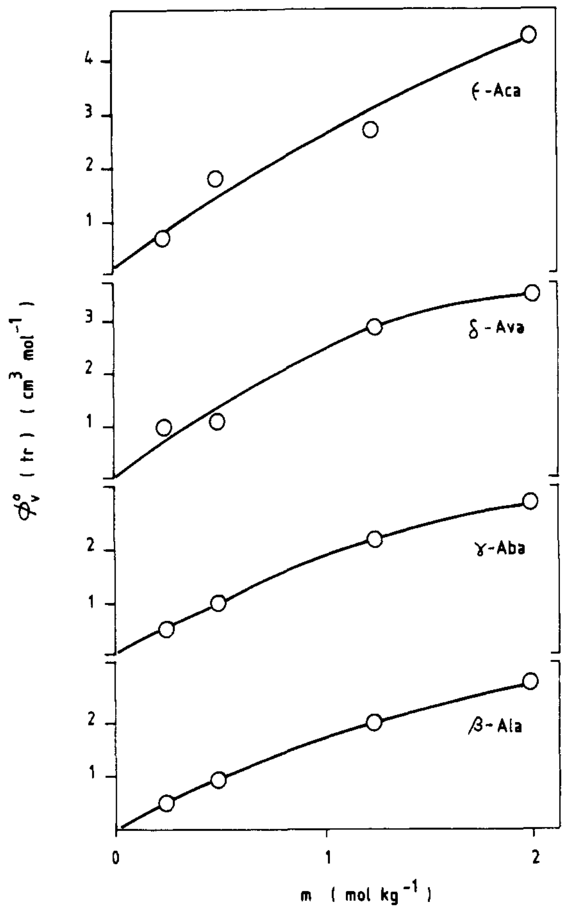

Figure 3. Plot of limiting apparent molar volumes of transfer $\phi_v{}^\circ$(tr) of α,ω-amino acids from water to aqueous NH_4Cl solutions vs m at 298.15 K.

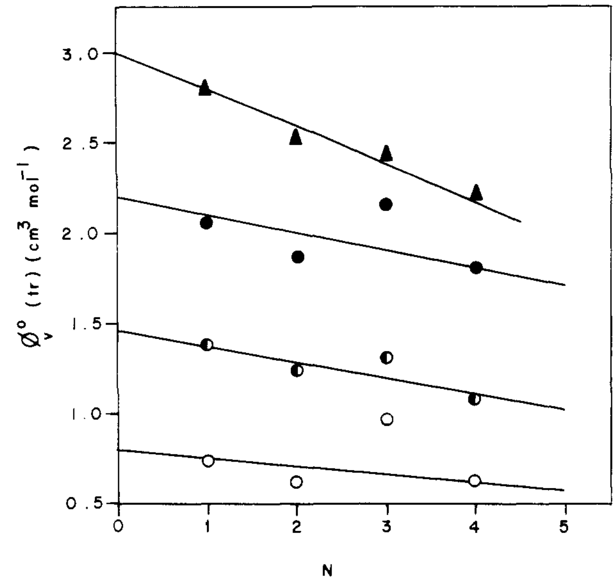

Figure 4. Plot of limiting apparent molar volumes of transfer $\phi_v{}^\circ$(tr) of α-amino acids vs N, the number of carbon atoms in the amino acid backbone, at 298.15 K: 0.5 m NH_4Cl (○), 1.0 m NH_4Cl (◐), 1.5 m NH_4Cl (●), 2.0 m NH_4Cl (▲).

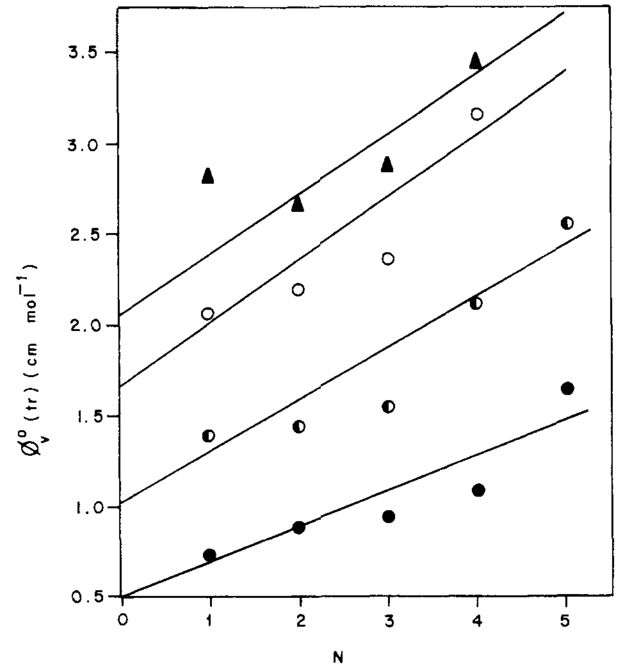

Figure 5. Plot of limiting apparent molar volume of transfer $\phi_v{}^\circ$(tr) of α,ω-amino acids vs N: 0.5 m NH_4Cl (●), 1.0 m NH_4Cl (◐), 1.5 m NH_4Cl (○), 2.0 m NH_4Cl (▲).

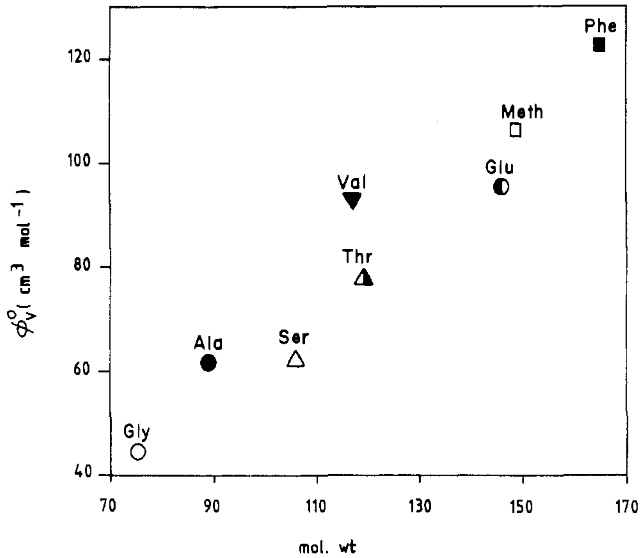

Figure 6. Variation of $\phi_v{}^\circ$ of α-substituted amino acids, $RCH(NH_2)$-COOH, taken in 1.25 m NH_4Cl with their molecular weights at 298.15 K: R = H (○); R = CH_3 (●); R = CH_2OH (△); R = $CH(CH_3)_2$ (▼); R = $CH(OH)CH_3$ (▲); R = $(CH_2)_2CONH_2$ (◐); R = $(CH_2)_2SCH_3$ (□); R = $CH_2C_6H_5$ (■).

molality were least-squares fitted to the Jones–Dole equation in the form

$$\eta_r = 1 + Bc + Dc^2 \qquad (3)$$

where B and D are empirical coefficients. Of these, the B coefficient is known to depend (*26*, *27*) on the size and shape of the solute molecules as well as on the solute–solvent interactions. The values of B and D coefficients of eq 3 are given in Table IV.

Discussion

Changes in partial molar volumes of amino acids in aqueous electrolyte solutions compared to those in water have been interpreted with use of the cosphere overlap model of Friedman and Krishnan (*28*), according to which the effect of overlap of hydration cospheres is destructive. Mishra, Prasad, and Ahluwalia (*29*), using this model, observed that the overlap of cospheres of two ionic species shows an increase in volume, whereas the overlap of hydrophobic–hydrophobic groups and ion–hydrophobic groups results in a net decrease in volume. The increase of $\phi_v{}^\circ$ of amino acids with the molality of NH_4Cl and the increasing positive transfer volumes suggest that the ion–ion and ion–hydrophilic group interactions are stronger than

Table I. Regression Coefficients, $\phi_v°$ and S_v, of the Equation $\phi_v/(\mathrm{cm^3\,mol^{-1}}) = \phi_v° + S_v c$ and the Volumes of Transfer, $\phi_v°(\mathrm{tr})$, of α-, α,ω-, and Substituted α-Amino Acids from Water to Aqueous Ammonium Chloride Solutions at 298.15 K

amino acid	$m(\mathrm{NH_4Cl})$, mol $\mathrm{kg^{-1}}$	no. of data points	$\phi_v°$, $\mathrm{cm^3\,mol^{-1}}$	S_v, L $\mathrm{mol^{-1}}$	$\phi_v°(\mathrm{tr})$, $\mathrm{cm^3\,mol^{-1}}$
α-amino acids					
glycine	0.00	9	42.89 (0.07)[a]	1.58 (0.27)	0.00
	0.02	10	43.28 (0.05)	0.87 (0.18)	0.39
	0.10	10	43.11 (0.03)	1.39 (0.11)	0.22
	1.00	9	43.98 (0.12)	1.78 (0.42)	1.09
	2.00	10	45.71 (0.10)	0.18 (0.32)	2.82
DL-α-alanine	0.00	10	60.23 (0.04)	1.09 (0.16)	0.00
	0.05	9	60.26 (0.04)	1.18 (0.13)	0.03
	0.25	10	60.61 (0.03)	1.23 (0.12)	0.38
	1.25	11	61.95 (0.04)	0.51 (0.11)	1.72
	1.80	11	62.31 (0.03)	0.62 (0.10)	2.08
DL-α-aminobutyric acid	0.00	10	75.24 (0.02)	0.87 (0.08)	0.00
	0.25	9	75.74 (0.04)	0.65 (0.14)	0.50
	0.50	7	76.21 (0.02)	0.44 (0.06)	0.97
	1.00	9	76.55 (0.03)	0.79 (0.12)	1.31
	2.00	8	77.66 (0.03)	0.31 (0.10)	2.42
DL-norvaline	0.00	7	91.61 (0.04)	0.53 (0.19)	0.00
	0.25	7	92.06 (0.03)	0.36 (0.19)	0.45
	1.00	6	92.68 (0.07)	1.08 (0.32)	1.07
	1.60	8	93.43 (0.04)	0.79 (0.18)	1.82
	2.00	8	93.81 (0.04)	0.57 (0.20)	2.20
DL-norleucine	0.00	4	107.13 (0.15)	16.47 (2.85)	0.00
	0.10	5	107.59 (0.10)	14.52 (3.04)	0.46
	0.20	5	107.86 (0.13)	8.40 (4.05)	0.73
	0.35	4	108.15 (0.18)	6.24 (5.12)	1.02
	0.50	5	108.21 (0.29)	3.72 (7.17)	1.08
α,ω-amino acids					
β-alanine	0.00	10	58.20 (0.02)	1.10 (0.09)	0.00
	0.25	9	58.69	1.06	0.49
	0.50	10	59.09 (0.02)	1.05 (0.06)	0.89
	1.25	10	60.16 (0.03)	0.70 (0.10)	1.96
	2.00	7	60.87 (0.01)	0.62 (0.04)	2.67
γ-aminobutyric acid	0.00	10	73.17 (0.03)	0.97 (0.11)	0.00
	0.25	10	73.66 (0.02)	1.08 (0.08)	0.49
	0.50	7	74.13 (0.04)	0.98 (0.20)	0.96
	1.25	7	75.37 (0.03)	0.54 (0.13)	2.20
	2.00	8	76.03 (0.02)	0.73 (0.09)	2.86
α,ω-amino acids					
δ-aminovaleric acid	0.00	6	87.33 (0.09)	5.83 (1.70)	0.00
	0.25	6	88.33 (0.10)	3.46 (1.93)	1.00
	0.50	5	88.43	8.42[b]	1.10
	1.25	5	90.16 (0.05)	1.12 (0.66)	2.83
	2.00	5	90.77 (0.17)	8.41 (3.50)	3.44
ε-aminocaproic acid	0.00	4	104.17	[b]	0.00
	0.25	4	104.75 (0.08)	3.96 (1.81)	0.58
	0.50	4	105.81 (0.04)	−5.84 (1.33)	1.64
	1.25	4	107.46 (0.05)	−7.77 (1.26)	3.29
	2.00	3	109.16 (0.22)	−22.97 (5.90)	4.99
substituted α-amino acids					
L-serine	0.00	6	60.46 (0.12)	2.20 (0.74)	0.00
	0.25	5	60.99 (0.04)	1.54 (0.16)	0.53
	0.50	6	61.54 (0.02)	1.18 (0.09)	1.08
	1.25	6	62.42 (0.02)	0.96 (0.09)	1.96
	2.00	6	63.33 (0.05)	0.50 (0.22)	2.87
L-threonine	0.00	5	76.58 (0.11)	11.82 (2.46)	0.00
	0.25	4	77.44 (0.06)	[b]	0.86
	0.50	4	77.70 (0.07)	[b]	1.12
	1.25	5	78.40 (0.20)	[b]	1.82
	2.00	5	78.43 (0.04)	9.82 (0.98)	1.85
DL-valine	0.00	5	91.48 (0.02)	−7.84 (0.36)	0.00
	0.25	6	92.10 (0.14)	−24.02 (4.39)	0.62
	0.50	5	92.23 (0.13)	−5.15 (2.93)	0.75
	1.25	5	93.24 (0.16)	−11.07 (3.42)	1.76
	2.00	6	93.25 (0.21)	−10.08 (4.27)	1.77
L-glutamine	0.00	4	93.56 (0.13)	8.18	0.00
	0.50	6	93.90 (0.09)	10.48 (1.84)	0.30
	1.25	4	96.16	7.24	2.60
	2.00	7	97.13 (0.05)	−7.75 (0.67)	3.53
DL-methionine	0.00	4	105.32 (0.10)	[b]	0.00
	0.25	3	105.89 (0.04)	7.34 (1.03)	0.57
	0.50	4	105.84 (0.28)	22.66 (8.76)	0.52
	1.25	5	106.51 (0.25)	23.26 (8.21)	1.19
	2.00	2	107.22	[b]	1.90
DL-phenylalanine	0.00	5	122.40	[b]	0.00
	0.25	4	122.42	[b]	0.02
	0.50	5	122.63	[b]	0.23
	1.25	3	123.36	[b]	0.96
	2.00	3	124.09	[b]	1.69

[a] Values in the parentheses denote the standard deviation of the parameters quoted. [b] Variation of ϕ_v with c showed considerable scatter; in these cases, $\phi_v°$ was determined graphically, and where slopes were near zero, $\phi_v°$ was taken as the average of all the ϕ_v values.

ion–hydrophobic group interactions. In the ternary system, the interaction of NH_4^+ and Cl^- ions with the amino acids is localized at the head groups (COO^- and NH_3^+). Due to these interactions, the electrostriction of water caused by the charge centers of

Table II. Partial Molar Volume, $\phi_v°$, of Substituted α-Amino Acids $RCH(NH_2)COOH$ in 1.25 mol kg^{-1} of Ammonium Chloride at 298.15 K

amino acid	side chain (R)	molar mass	$\phi_v°$(1.25 *m* NH_4Cl), cm^3 mol^{-1}
glycine	$-H$	75.07	44.75
DL-α-alanine	$-CH_3$	89.09	61.95
L-serine	$-CH_2OH$	105.09	62.42
DL-valine	$-CH(CH_3)_2$	117.15	93.24
L-threonine	$-CH(OH)CH_3$	119.12	78.40
L-glutamine	$-(CH_2)_2CONH_2$	146.15	95.55
DL-methionine	$-(CH_2)_2SCH_3$	149.22	106.51
DL-phenylalanine	$-CH_2C_6H_5$	165.19	122.84

Table III. Values of $\bar{V}_e$ and Solvation Number, n_s, for Amino Acids in Aqueous Ammonium Chloride Solutions at 298.15 K

amino acid	$m(NH_4Cl)$, mol kg^{-1}	$\phi_v°$, cm$_3$ mol^{-1}	$\bar{V}_e = \phi_v° - \bar{V}_{int}$,[a] cm^3 mol^{-1}	$n_s = -\bar{V}_e/3.3$
glycine	0.00	42.89	−8.98	2.72 (2.63)[b]
	0.02	43.28	−8.59	2.60
	0.10	43.11	−8.76	2.65
	1.00	43.98	−7.89	2.39
	2.00	45.71	−6.16	1.87
DL-α-alanine	0.00	60.23	−11.51	3.49 (3.41)[c]
	0.05	60.26	−11.48	3.48
	0.25	60.61	−11.13	3.37
	1.25	61.95	−9.79	2.97
	1.80	62.31	−9.43	2.86
β-alanine	0.00	58.20	−13.54	4.10[c]
	0.25	58.69	−13.05	3.95
	0.50	59.09	−12.65	3.83
	1.25	60.16	−11.58	3.51
	2.00	60.87	−10.87	3.29
L-serine	0.00	60.46	−12.89	3.91
	0.25	60.99	−12.36	3.74
	0.50	61.54	−11.81	3.58
	1.25	62.42	−10.93	3.31
	2.00	63.33	−10.02	3.04
L-threonine	0.00	76.58	−11.16	3.38
	0.25	77.44	−10.30	3.12
	0.50	77.70	−10.04	3.04
	1.25	78.40	−9.34	2.83
	2.00	78.43	−9.31	2.82
DL-valine	0.00	91.40	−10.61	3.21
	0.25	92.10	−9.99	3.03
	0.50	92.23	−9.86	2.99
	1.25	93.24	−8.85	2.68
	2.00	93.25	−8.84	2.68
DL-methionine	0.00	105.32	−20.35	6.17 (6.16)
	0.25	105.89	−19.78	5.99
	0.50	105.84	−19.83	6.01
	1.25	106.51	−19.16	5.81
	2.00	107.03	−18.64	5.65
DL-phenylalanine	0.00	122.40	−16.30	4.94 (5.22)
	0.25	122.40	−16.30	4.94
	0.50	122.61	−16.09	4.88
	1.25	122.84	−15.86	4.81
	2.00	124.03	−14.67	4.44

[a] $\bar{V}_{int}$ was estimated from the relationship $\bar{V}_{int} = (0.7/0.634)\bar{V}_{cryst}$ with crystal volume data of Berlin and Pallansch (*36*). [b] Values in parentheses are of Millero, Surdo, and Shin (*25*). [c] β-Alanine has been reported by Ogawa, Yasuda, and Mizutani (*31*) to have a larger number of hydrated water molecules than α-alanine from compressibility studies (n_s values not reported).

Table IV. Viscosity *B* and *D* Coefficients of Amino Acids in Aqueous Ammonium Chloride Solutions at 298.15 K Derived by Regression Analysis of the Equation $\eta_r = 1 + Bc + Dc^2$

amino acid	$m(NH_4Cl)$, mol kg^{-1}	*B*, L mol^{-1}	*D*, L^2 mol^{-1}
glycine	0.000 00	0.149 (0.01)[a]	0.009
	0.019 80	0.140 (0.02)	
	0.101 56	0.150 (0.01)	−0.018
	2.015 70	0.227 (0.01)	−0.081
α-alanine	0.000 00	0.246 (0.01)	0.025
	0.051 32	0.279 (0.01)	−0.033
	0.249 82	0.331 (0.02)	−0.124
	1.259 93	0.358 (0.02)	−0.110
	1.810 01	0.388 (0.03)	−0.230
α-aminobutyric acid	0.000 00	0.299 (0.01)	0.099
	0.504 56	0.329 (0.03)	
	0.998 26	0.395 (0.01)	0.132
	1.999 89	0.421 (0.03)	0.128
β-alanine	0.000 00	0.168 (0.02)	0.180
	0.252 40	0.163 (0.01)	0.157
	0.504 76	0.183 (0.01)	0.145
	1.258 40	0.186 (0.01)	0.183
	2.011 80	0.241 (0.02)	0.163
γ-aminobutyric acid	0.000 00	0.310 (0.03)	0.176
	0.251 85	0.278 (0.02)	0.266
	0.503 15	0.262 (0.01)	0.357
	1.258 05	0.340 (0.01)	0.188
	2.004 20	0.361 (0.01)	0.214
L-serine	0.000 00	0.278 (0.01)	0.053
	0.501 29	0.291 (0.03)	0.081
	1.249 41	0.301 (0.02)	

[a] Values in the parentheses are the standard deviations of the parameters quoted.

the amino acid will be reduced, which results in an increase in volume.

This trend can also be explained with the equation of Shahidi, Farrell, and Edwards (*30*)

$$\phi_v° = V_{vW} + V_v - V_s \qquad (4)$$

in which the partial molar volume of the amino acid may be considered to be made up of the van der Waals volume (V_{vW}), the volume associated with voids (V_v) or empty space, and the volume due to shrinkage (V_s) that arises due to the electrostriction of the solvent caused by the hydrophilic groups present in the amino acid. The presence of NH_4Cl in water decreases the extent of electrostriction caused by the amino acid, which results in a decrease in shrinkage volume. Assuming that V_{vW} and V_v are not significantly affected by the presence of NH_4Cl, an increase in $\phi_v°$ results and consequently a positive value of $\phi_v°$(tr) is shown.

It may be noted (Table I) that $\phi_v°$ of of α,ω-amino acids are less than those of α-amino acids owing to greater electrostriction of the solvent caused by the former. Also, with increasing chain length, $\phi_v°$(tr) of α,ω-amino acids increase while those for α-amino acids decrease. This is because the additional methylene groups provide an increasing structure-enforcing tendency in α-amino acids, and as a result, the water in the overlapping cospheres is more structured than in the bulk. When this water relaxes to the bulk, there is a decrease in volume. But, in α,ω-amino acids, the ion–ion interactions increase with the addition of CH_2 groups, and consequently there is a net increase in volume.

The results on substituted α-amino acids can be rationalized on the basis that the partial molar volume is observed to increase with the increasing molar mass and size of the amino acid (Table II). The increase in $\phi_v°$ from glycine to α-alanine is thus explained. When one H of DL-α-alanine is replaced by a $-OH$ (L-serine), $\phi_v°$ should increase by virtue of its increased size, but because $-OH$ is a hydrophilic group, L-serine behaves as a better structure-breaker than DL-α-alanine. The increase in $\phi_v°$ due to an increase in size and mass is slightly offset by a decrease because of the structure-breaking property of the $-OH$ group in L-serine. Thus, the $\phi_v°$ of serine is seen to be comparable to that of α-alanine. When yet another H atom of DL-α-alanine is replaced by a hydrophobic $-CH_3$ group (L-threonine), $\phi_v°$ increases because of the structure-enhancing behavior of the alkyl group; this also explains the higher partial molar volume of threonine than that of serine. If two H atoms of DL-α-alanine are replaced by two hydrophobic $-CH_3$ groups

Table V. Partial Molar Volumes, $\phi_v°$, Intrinsic Volumes, $\bar{V}_{int}$, Limiting Effective Flow Volumes, $V_f°$, Volumes of the Solvation Sheath, V_{sh}, and $B/\phi_v°$ for Amino Acids in Aqueous NH_4Cl Solutions at 298.15 K

amino acid	$m(NH_4Cl)$, mol kg^{-1}	$\phi_v°$, cm^3 mol^{-1}	$\bar{V}_{int} = \phi_v° + \bar{V}_e$,[a] cm^3 mol^{-1}	$V_f°$,[b] cm^3 mol^{-1}	$V_{sh} = V_f° - \bar{V}_{int}$, cm^3 mol^{-1}	$B/\phi_v°$
glycine	0.019 80	43.28	56.78	56.15	−0.63	3.2
	0.101 56	43.11	56.61	59.55	2.94	3.5
	2.015 70	45.71	59.21	92.14	32.93	5.0
α-alanine	0.051 32	60.26	72.96	112.04	39.08	4.6
	0.249 82	60.61	73.31	133.41	60.10	5.5
	1.259 93	61.95	74.65	143.78	69.13	5.8
	1.810 01	62.31	75.01	160.26	85.25	6.2
α-aminobutyric acid	0.504 56	76.21	89.31	131.71	42.40	4.3
	0.998 26	76.53	89.65	159.10	69.45	5.2
	1.999 89	77.66	90.76	168.75	77.99	5.4
β-alanine	0.252 40	58.69	73.09	67.52	−5.57	2.8
	0.504 76	59.09	73.49	75.63	2.14	3.1
	1.258 40	60.16	74.56	76.84	2.28	3.1
	2.011 80	60.87	75.27	98.47	23.20	4.0
γ-aminobutyric acid	0.251 85	73.66	86.86	114.58	27.72	3.8
	0.503 15	74.13	87.33	110.84	23.51	3.5
	1.258 05	75.37	88.57	137.44	48.87	4.5
	2.004 20	76.03	89.23	145.97	56.74	4.8
L-serine	0.501 29	61.54	74.44	116.44	42.00	4.7
	1.249 41	62.42	75.32	120.49	45.17	4.8

[a] Adopted from Cohn and Edsall (*35*). [b] Obtained using Vand equation (*34*).

(DL-valine), the increase in the partial molar volume should be more, relative to L-threonine owing to greater hydrophobicity of the side chain, as is observed. Glutamine has a hydrophilic amide group (–C(=O)–NH_2) because of which its $\phi_v°$ is less than that of methionine, which has a less hydrophilic –SCH_3 group. Phenylalanine has the maximum value of $\phi_v°$ in the series of amino acids studied, which can be attributed to its largest size and mass.

A decrease in the solvation number on addition of NH_4Cl (Table III) is due to the decrease in the electrostriction of water. β-Alanine with a larger charge separation than α-alanine has a larger value of solvation number, which is consistent with the results of Ogawa, Yasuda, and Mizutani (*31*). DL-Valine has a smaller solvation number than DL-α-alanine, owing to greater hydrophobicity and therefore lesser hydration. Similarly, threonine when compared to serine shows a smaller solvation number. Due to large electrostriction in the case of DL-methionine, solvation numbers are higher than in the other amino acids studied.

The viscosity of the solvent (water + ammonium chloride) decreased with an increase in NH_4Cl concentration over the range employed (up to 2.00 mol kg^{-1}), which can be attributed to the structure-breaking influence of the electrolyte on the hydrogen-bonded structure of water in its vicinity, creating a region of lower viscosity. For the ternary system, at a given concentration of NH_4Cl, the viscosity of the solution increased with the increasing molarity of amino acid. The B coefficients of all the amino acids studied were positive (Table IV) and increased with the concentration of NH_4Cl, which may be considered to arise due to increased solvation. If amino acids in aqueous electrolyte solutions are considered, as a first approximation, as suspensions of spherical particles, as verified (*32*) by the method suggested by Phang (*33*), then it is possible to estimate the limiting effective flow volume $V_f°$ from the Vand equation (*34*), with which the volume of the solvation sheath (V_{sh}) can be approximated relative to the intrinsic volume ($\bar{V}_{int}$) of the amino acid.

$$V_{sh} = V_f° - \bar{V}_{int} \tag{5}$$

$\bar{V}_{int}$ was found from the observed partial molar volume $\phi_v°$ and the decrease in volume ($\bar{V}_e$) due to electrostriction adopted from the data of Cohn and Edsall (*35*) for amino acids taken in water.

$$\bar{V}_{int} = \phi_v° + \bar{V}_e \tag{6}$$

It is assumed that $\bar{V}_e$ values in water are about the same as in aqueous NH_4Cl solution. In fact, $\bar{V}_e$ for amino acids in NH_4Cl solution have also been calculated (Table III) in this study with $\bar{V}_{int}$ obtained from the crystal volume data of Berlin and Pallansch (*36*). It is seen that these $\bar{V}_e$ values can be reasonably approximated to those reported by Cohn and Edsall (glycine, 13.5; α-alanine, 12.7; α-aminobutyric acid, 13.1; β-alanine, 14.4; γ-aminobutyric acid, 13.2; and L-serine, 12.9, which are the amino acids chosen for viscosity measurements). $\bar{V}_{int}$ and thus V_{sh} so evaluated are included in Table V. Because of the various assumptions in assigning $\bar{V}_e$, one may comment only on the relative magnitude of V_{sh}. It can be seen that V_{sh} varies significantly with solvent composition, showing a tendency to increase with an increase in the concentration of NH_4Cl that supports the view that there is some form of solvent structure enforcement (*33*) because of the decrease in electrostriction on adding NH_4Cl. This is consistent with the observed increase in the B coefficients.

It has been reported (*26*) that the value of ratio $B/\phi_v°$ lies between 0 and 2.5 for unsolvated spherical species. It can be noted from Table V that this ratio is greater than 2.5 in most of the cases and increases with an increase in NH_4Cl molality, which is due to the larger increase in B coefficients as compared to that in $\phi_v°$ with the increase in NH_4Cl molality. This can be explained if one assumes that the increase in viscosity is predominantly due to the increased resistance offered by the more structured solvent to the moving amino acid moiety though solvation effects may also contribute.

The significance of the D coefficients is not fully understood, but they increase as the number of CH_2 groups increases. A similar observation has also been reported by Devine and Lowe (*37*) and by Desnoyers et al. (*38*, *39*).

Acknowledgment

We thank the Computer Centre for use of the IBM-360/44 data processing system.

Glossary

B	empirical coefficient
c	molarity of amino acid solution, mol cm^{-3}
D	empirical coefficient
M	molar mass of amino acid
m	molality of amino acid solution, mol kg^{-1}
N	number of carbon atoms
R	a side chain
S_v	empirical constant

$\bar{V}_e$	decrease in volume due to electrostriction
V_f°	limiting effective flow volume
$\bar{V}_{int}$	intrinsic volume of the amino acid
V_s	shrinkage volume
V_v	volume associated with void
V_{vW}	van der Waals volume
V_{sh}	volume of solvation sheath

Greek Letters

ϕ_v	apparent molar volume
ϕ_v°	limiting apparent molar volume or infinite-dilution partial molar volume
$\phi_v^\circ(tr)$	change in partial molar volume when amino acid is transferred from water to aqueous NH_4Cl solution
η	viscosity of solution
η_0	viscosity of pure solvent
η_r	relative viscosity (η/η_0)
ρ	density of solution
ρ_0	density of pure solvent

Literature Cited

(1) Enea, O.; Jolicoeur, C. *J. Phys. Chem.* **1982**, *86*, 3870.
(2) Kauzmann, W. *Adv. Protein Chem.* **1979**, *14*, 1.
(3) Edsall, J. T.; McKenzie, H. A. *Adv. Biophys.* **1983**, *16*, 53.
(4) Perutz, M. F. *Science* **1978**, *201*, 1187.
(5) Nandi, P. K.; Robinson, D. R. *J. Am. Chem. Soc.* **1972**, *94*, 1229-1315.
(6) Bull, H. B.; Breese, K. *Arch. Biochem. Biophys.* **1970**, *137*, 299.
(7) Kelley, B. P.; Lilley, T. H. *J. Chem. Soc., Faraday Trans. 1* **1978**, *74*, 2771.
(8) Larson, J. W.; Morrison, D. G. *J. Phys. Chem.* **1976**, *80*, 1449.
(9) Bhat, R.; Ahluwalia, J. C. *J. Phys. Chem.* **1985**, *89*, 1099.
(10) Bhat, R.; Ahluwalia, J. C. *Int. J. Pept. Protein Res.* **1987**, *30*, 145.
(11) Rajiv Bhat; Nand Kishore; Ahluwalia, J. C. *J. Chem. Soc., Faraday Trans. 1* **1988**, *84*, 2651.
(12) Ogawa, T.; Mizutani, K.; Yasuda, M. *Bull. Chem. Soc. Jpn.* **1984**, *57*, 2064.
(13) Sandhu, J. S.; Urmila Kashyap. *J. Electrochem. Soc. India* **1987**, *36*, 267.
(14) Sandhu, J. S.; Urmila Kashyap. *Ind. J. Chem., Sect. A* **1987**, *26*, 952.
(15) Sandhu, J. S.; Gurbir Singh. *J. Ind. Chem. Soc.* **1988**, *65*, 173.
(16) Rao, Mandapati, V. R.; Atreyi, Mahalakshmi; Rajeswari, Moganty, R. *Can. J. Chem.* **1988**, *66*, 487.
(17) Badley, R. A.; Carruthers, L.; Philips, M. C. *Biochim. Biophys. Acta* **1977**, *110*, 495.
(18) Weast, R. C., Ed. *Handbook of Chemistry and Physics*; Chemical Rubber: Cleveland, OH, 1969.
(19) Kell, G. S. *J. Chem. Eng. Data* **1967**, 12, 66.
(20) Ellis, A. J. *J. Chem. Soc. A* **1966**, 1579.
(21) Stokes, R. H. *Aust. J. Chem.* **1975**, *28*, 2109.
(22) Jolicoeur, C.; Riedl, B.; Desrochers, D.; Lemelin, L.; Zamojska, R.; Enea, O. *J. Solution Chem.* **1986**, *15*, 109.
(23) Goldsack, D. E.; Franchetto, R. *Can. J. Chem.* **1978**, *56*, 1442.
(24) Lilley, T. H. *The Chemistry and Biochemistry of Amino Acids*; Barrett, G. C., Ed.; Chapman & Hall: London, 1985; Chapter 23.
(25) Millero, F. J.; Lo Surdo, A.; Shin, C. *J. Phys. Chem.* **1978**, *82*, 784.
(26) Stokes, R. H.; Mills, R. Viscosity of Electrolytes and Related Properties. *International Encyclopedia of Physical Chemistry and Chemical Physics*; Pergamon: London, 1965.
(27) Tyrrell, H. J. V.; Kennerley, M. *J. Chem. Soc. A* **1968**, 2724.
(28) Friedman, H. L.; Krishnan, C. V. *Water, A Comprehensive Treatise*; Franks, F., Ed.; Plenum Press: New York, 1973; Vol. 3, Chapter 1.
(29) Mishra, A. K.; Prasad, K. P.; Ahluwalia, J. C. *Biopolymers* **1983**, *22*, 2397.
(30) Shahidi, F.; Farrell, P. G.; Edwards, J. T. *J. Solution Chem.* **1976**, *5*, 807.
(31) Ogawa, T.; Yasuda, M.; Mizutani, K. *Bull. Chem. Soc. Jpn.* **1984**, *57*, 662.
(32) Meena, N. Volumetric and Transport Studies of Some Amino Acids in Aqueous Electrolyte Solutions. Ph.D. Thesis, University of Delhi, 1986.
(33) Phang, S. *Aust. J. Chem.* **1977**, *30*, 1605.
(34) Vand, V. *J. Phys. Chem.* **1948**, *52*, 277.
(35) Cohn, E. J.; Edsall, J. T. *Proteins, Amino Acids and Peptides*; Reinhold: New York, 1965.
(36) Berlin, E.; Pallansch, M. J. *J. Phys. Chem.* **1968**, *72*, 1887.
(37) Devine, W.; Lowe, B. M. *J. Chem. Soc. A* **1971**, 2113.
(38) Desnoyers, J. E.; Arel, M.; Leduc, P. A. *Can. J. Chem.* **1969**, *47*, 547.
(39) Desnoyers, J. E.; Perron, G. *J. Solution Chem.* **1972**, *1*, 199.

Received for review June 19, 1989. Accepted September 27, 1989. The University Grants Commission, New Delhi, is thanked for a Research Fellowship to M.N. This paper was abstracted from the Ph.D. thesis of M.N.

Supplementary Material Available: Tables of detailed density and viscosity data for different amino acids as a function of concentration in aqueous ammonium chloride solutions (15 pages). Ordering information is given on any current masthead page.

Viscosity and Density of Ternary Mixtures of Toluene, Bromobenzene, 1-Hexanol, and 1-Octanol

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Mixture viscosities and densities of the ternary mixtures of toluene, bromobenzene, 1-hexanol, and 1-octanol were measured at 30, 40, 50, and 60 °C. The nonidealities reflected in mixture viscosities are expressed and discussed in terms of excess viscosities, which were negative in most of the cases. The ternary η-X-T data were fitted in a Redlich-Kister-type equation along with a ternary contribution term.

Introduction

Extending our earlier work (*1-8*) on viscosities and dielectric constants of liquid mixtures, the present paper reports the viscosities and the densities for the ternary mixtures of toluene, bromobenzene, 1-hexanol, and 1-octanol in the temperature range from 30 to 60 °C.

Experimental Section

Materials. Toluene (BDH), 1-hexanol (BDH), and 1-octanol (Ferak Berlin) were fractionally distilled and dried while bromobenzene (E. Merck) after repeated fractional distillation was collected at 156 ± 0.5 °C and retained for use. The mean values of repeat density, viscosity, and refractive index measurements of the liquids so purified did not deviate from the corresponding literature values beyond allowable limits (Table IV). Redistilled deionised and degassed water (electrical conductivity $< 7.0 \times 10^{-7}\ \Omega^{-1}\ cm^{-1}$) was used in each case for checking the instruments and calibrating the pycnometers.

Experimental Measurements. Ternary mixtures were prepared by weight with an accuracy of 0.0001 g, taking care that the resulting ternary compositions represent the data points suitably distributed away from the vertices and also located in

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0021-9568/90/1735-0093$02.50/0