Volumetric and Ultrasonic Investigation of Glycylglycine in Aqueous FeCl₃ Solutions

M.S. Santosh · D. Krishna Bhat · Aarti S. Bhat

Abstract Viscosity, ultrasonic velocity and density measurements have been carried out for glycylglycine in aqueous FeCl₃ solution as a function of molality at $T = 288.15$ K, 298.15 K and 308.15 K. The experimental data have been used to derive properties such as isentropic compressibility (κ_S), change in isentropic compressibility ($\Delta\kappa_S$), relative change in isentropic compressibility ($\Delta\kappa_S/\kappa_0$), apparent molar compressibility, volume and their limiting apparent molar quantities along with the constants S_K, S_V and viscosity B-coefficient. The obtained thermodynamic properties have been discussed in terms of molecular interactions.

Keywords Viscosity · Ultrasonic velocity · FeCl₃ · Glycylglycine

1 Introduction

Ultrasonic velocity studies on aqueous solutions of amino acids with electrolytes and non-electrolytes provide useful information in understanding the behavior of liquid systems, intramolecular and intermolecular association, complex formation and related structural changes. For the past two decades, a considerable number of studies have been carried out to investigate the hydration of proteins through volumetric and ultrasonic velocity measurements [1–7]. Due to the complex molecular structure of proteins their direct study is somewhat difficult. Therefore, a useful approach is to study simpler model compounds such as the amino acids that are building blocks of proteins. Most studies on amino acids have been carried out in pure and mixed aqueous solution [8–10]. Amino acids, the monomer units of protein molecules, play an important role in all living organisms. Because amino acids are present as zwitterions in aqueous solutions [11], their volumetric and compressibility properties should reflect structural interactions with water molecules as they do in the case of

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electrolytes. Transition metal ions play an important role in plant growth, lipid metabolism and regulation of physiological systems.

To the best of our knowledge, studies on the interactions of glycylglycine with $FeCl_3$ in aqueous solutions at different temperatures and concentrations are very rare. Through this paper, an attempt has been made to understand the behavior of mixtures of a dipeptide and a transition metal salt in aqueous solutions with ultrasonic velocity measurements under diverse conditions. However, ultrasonic velocity data alone do not provide significant information about the nature and relative strengths of various types of intermolecular and interionic interactions occurring among the components. Hence, their derived parameters such as isentropic compressibility (κ_S), change in isentropic compressibility ($\Delta\kappa_S$), relative change in isentropic compressibility ($\Delta\kappa_S/\kappa_0$), apparent molar compressibility (ϕ_K), apparent molar volume (ϕ_V), limiting apparent molar compressibility (ϕ_K^0), limiting apparent molar volume (ϕ_V^0), and the constants (S_K, S_V) along with viscosity B-coefficient, specific acoustic impedance (Z), and relative association (R_A) have been obtained in order to provide clear ideas about such interactions.

2 Experimental

2.1 Materials

Glycylglycine and $FeCl_3\cdot6H_2O$ of 99% purity, used in our studies, were purchased from Sigma-Aldrich, Germany. This commercially available glycylglycine was used without further purification. $FeCl_3\cdot6H_2O$ was used after drying for 72 h in a vacuum desiccator at room temperature. Deionized, doubly-distilled degassed water with a specific conductance less than 1.29×10^{-6} $\Omega^{-1}\cdot cm^{-1}$ was used for the preparation of all solutions. The solutions were prepared on a weight basis by using a Mettler balance having a precision of ±0.01 mg. The temperature of the water bath was controlled within ±0.01 K using a thermostat. In the system studied, the concentration of glycylglycine was varied at (0.0052, 0.0076, 0.0103, 0.0208, 0.0307, 0.0502, 0.0704, 0.0801, 0.0902, and 0.1006) $mol\cdot kg^{-1}$ while that of $FeCl_3$ was kept constant at 0.25 $mol\cdot kg^{-1}$. To prevent formation of air bubbles, all solutions were preheated in sealed Eppendorf tubes at 5 °C above the measurement temperature before filling the ultrasonic and densimetric cells.

2.2 Methods

Viscosities were measured using a Brookfield DV-III Ultra Programmable Rheometer (Brookfield Engineering Laboratories, Inc., USA) that was calibrated using double distilled water and ethanol, and their uncertainty was found to be $\pm0.5\%$ for the studied solutions.

The ultrasonic velocity of the pure components and their mixtures were measured by a variable-path fixed frequency interferometer provided by Mittal Enterprises, New Delhi (Model-83). This apparatus consists of a high frequency generator and a measuring cell. The measurements of ultrasonic velocity were made at a fixed frequency of 2 MHz. The capacity of the measurement cell is 7 mL. Calibration of the ultrasonic interferometer was done by measuring the velocity of AR grade benzene and carbon tetrachloride. The maximum estimated error in the ultrasound measurements is $\pm0.08\%$. The temperature was controlled by circulating water around the liquid-containing cell from a thermostatically-controlled and adequately stirred water bath (precision ±0.1 °C).

Densities were measured using a (Mettler Toledo) Density 30PX digital densitometer having an accuracy of $\pm 3 \times 10^{-3}$ kg·m^{-3}. The densimeter was calibrated using double distilled water. The temperature of the sample and reference resonator cells with minimum volumes of 0.5 cm^3 was controlled with a precision of ± 0.01 K, and a previously described differential technique was employed for all measurements [12].

The physicochemical parameters of solutions of glycylglycine–FeCl$_3$ were measured at the temperatures 288.15, 298.15 and 308.15 K. Based on the above mentioned physico-chemical parameters, derived parameters were calculated for the substrate at different temperatures and molalities. Each measurement was made in triplicate and the average values are reported.

3 Results

The experimental viscosity (η), ultrasonic velocity (u), and density (ρ) data of glycylglycine in aqueous solutions are reported at three different temperatures in Table 1. Table 2 reports data for the above physicochemical parameters and their derived parameters for glycyl-glycine in aqueous FeCl$_3$ solutions. The apparent molar compressibility is calculated by the equation:

$$\phi_K = \frac{1000}{m\rho\rho_0}(\rho_0\kappa_S - \rho\kappa_0) + \left(\frac{\kappa_0 M}{\rho_0}\right) \tag{1}$$

where κ_S, ρ, κ_0, and ρ_0 are the isentropic compressibility and density of solution and solvent, respectively, m is the molal concentration of the solute and M is the molar mass of the solute. ϕ_K is a function of molality as obtained by Gucker [13] from the Debye-Hückel theory [14]:

$$\phi_K = \phi_K^0 + S_K m^{1/2} \tag{2}$$

where ϕ_K^0 is the limiting apparent molar compressibility at infinite dilution and S_K is a constant (limiting slope). Values of ϕ_K^0 and S_K were evaluated by the least-squares method.

The apparent molar volume is obtained from the relation:

$$\phi_V = \frac{1000}{m\rho\rho_0}(\rho_0 - \rho) + \left(\frac{M}{\rho_0}\right) \tag{3}$$

The apparent molar volume was found to vary with molality according to Masson's empirical equation [15]:

$$\phi_V = \phi_V^0 + S_V m^{1/2} \tag{4}$$

where ϕ_V^0 is the limiting apparent molal volume at infinite dilution and S_V is a constant; these values were also determined by the least-squares method.

All the viscosity data were analyzed by means of the Jones-Dole semi-empirical equation [16]:

$$\frac{\eta}{\eta_0} = 1 + Am^{1/2} + Bm \tag{5}$$

where η and η_0 are the viscosities of the solution and solvent, respectively, and m is the molality of the solute system. A and B are constants characteristic for each solute–solvent

Table 1 Molality m, viscosity η, ultrasonic velocity u, and density ρ, for glycylglycine in aqueous solutions at $T = (288.15, 298.15,$ and $308.15)$ K

m	η	u	$\rho \times 10^{-3}$
$(mol \cdot kg^{-1})$	$(m \cdot Pa \cdot s)$	$(m \cdot s^{-1})$	$(kg \cdot m^{-3})$
		$T = 288.15$ K	
0.0052	1.16	1489.12	1.0086
0.0076	1.23	1496.23	1.0133
0.0103	1.29	1503.97	1.0180
0.0208	1.38	1511.71	1.0227
0.0307	1.46	1519.45	1.0274
0.0502	1.54	1527.19	1.0321
0.0704	1.62	1534.93	1.0368
0.0801	1.69	1542.67	1.0415
0.0902	1.76	1550.41	1.0462
0.1006	1.85	1558.15	1.0509
		$T = 298.15$ K	
0.0052	1.08	1523.42	1.0042
0.0076	1.14	1531.16	1.0085
0.0103	1.20	1538.83	1.0128
0.0208	1.27	1545.50	1.0171
0.0307	1.34	1554.17	1.0214
0.0502	1.41	1561.84	1.0258
0.0704	1.48	1569.51	1.0301
0.0801	1.55	1577.25	1.0344
0.0902	1.62	1584.92	1.0387
0.1006	1.70	1592.59	1.0430
		$T = 308.15$ K	
0.0052	1.02	1575.81	1.0008
0.0076	1.06	1583.67	1.0054
0.0103	1.10	1591.71	1.0100
0.0208	1.16	1599.95	1.0146
0.0307	1.22	1608.09	1.0192
0.0502	1.28	1616.23	1.0239
0.0704	1.34	1624.37	1.0286
0.0801	1.40	1632.51	1.0333
0.0902	1.46	1640.65	1.0380
0.1006	1.54	1648.79	1.0427

system. A is known as the Falkenhagen coefficient [17] that characterizes ionic interactions, and B is the Jones-Dole or viscosity B-coefficient that depends on the size of the solute and the nature of solute–solvent interactions.

The measured ultrasonic velocity values for glycylglycine in aqueous $FeCl_3$ solutions have been fitted by least squares to the following second-order polynomial equation:

$$u = u_0 + u_1 m + u_2 m^2 \tag{6}$$

Table 2 Molality m, viscosity η, ultrasonic velocity u, density ρ, isentropic compressibility κ_S, relative change in the isentropic compressibility $\Delta\kappa_S/\kappa_0$, apparent molar compressibility ϕ_K, and apparent molar volume ϕ_V, for glycylglycine in aqueous $FeCl_3$ solutions at $T = (288.15, 298.15,$ and $308.15)$ K

m (mol·kg⁻¹)	η (m·Pa·s)	u (m·s⁻¹)	$\rho \times 10^{-3}$ (kg·m⁻³)	κ_S (10⁻¹⁰ m²·N⁻¹)	$\Delta\kappa_S/\kappa_0$ (10² m²·N⁻¹)	$-\phi_K$ (10¹³ m⁵·mol⁻¹·N⁻¹)	$-\phi_V$ (10⁻⁶ m³·mol⁻¹)
$T = 288.15$ K							
0.0000	1.27	1652.36	1.0324	3.5476	1.8233	–	–
0.0052	1.35	1660.41	1.0343	3.5068	2.2457	19.6905	74.9154
0.0076	1.42	1667.53	1.0376	3.4659	2.9874	17.6131	74.0420
0.0103	1.50	1674.74	1.0415	3.4233	3.2389	16.4163	72.4128
0.0208	1.61	1681.25	1.0460	3.3822	3.8057	15.1944	70.9588
0.0307	1.73	1688.68	1.0486	3.3442	4.4692	14.0919	69.5975
0.0502	1.82	1695.88	1.0512	3.3076	4.9827	12.9504	68.3761
0.0704	1.94	1702.46	1.0538	3.2740	5.1116	10.1429	68.0708
0.0801	2.05	1709.37	1.0564	3.2339	5.2405	8.4657	67.7655
0.0902	2.17	1717.53	1.0590	3.2010	5.3694	7.0125	67.4602
0.1006	2.29	1725.82	1.0616	3.1626	5.4983	5.9325	67.1549
$T = 298.15$ K							
0.0000	1.20	1690.41	1.0288	3.4016	2.4583	–	–
0.0052	1.28	1698.36	1.0308	3.3633	2.8999	23.0249	78.2275
0.0076	1.34	1705.82	1.0328	3.3274	3.4562	21.9245	76.9742
0.0103	1.41	1712.41	1.0357	3.2926	3.9785	19.7575	75.6531
0.0208	1.53	1720.28	1.0385	3.2538	4.5430	17.6414	74.9523
0.0307	1.64	1728.76	1.0403	3.2164	5.0192	16.5335	73.7383
0.0502	1.72	1736.32	1.0429	3.1805	5.4871	14.3975	71.8640
0.0704	1.78	1744.65	1.0455	3.1423	5.9550	12.2617	69.9897
0.0801	1.83	1752.48	1.0482	3.1063	6.4229	10.1253	69.1154
0.0902	1.92	1760.12	1.0508	3.0718	6.8908	7.9895	68.2411
0.1006	2.02	1769.89	1.0533	3.0307	7.3587	5.9535	67.3668

Table 2 (*Continued*)

m (mol·kg^{-1})	η (m·Pa·s)	u (m·s^{-1})	$\rho \times 10^{-3}$ (kg·m^{-3})	κ_S (10^{-10} m^2·N^{-1})	$\Delta\kappa_S/\kappa_0$ (10^2 m^2·N^{-1})	$-\phi_K$ (10^{13} m^5·mol^{-1}·N^{-1})	$-\phi_V$ (10^{-6} m^3·mol^{-1})
				$T = 308.15$ K			
0.0000	1.14	1801.32	1.0271	3.0005	2.9316	–	–
0.0052	1.23	1810.28	1.0290	2.9654	3.3998	26.9616	80.1681
0.0076	1.29	1817.63	1.0307	2.9366	3.8762	24.8649	79.2975
0.0103	1.37	1824.05	1.0328	2.9101	4.3754	23.1182	77.5997
0.0208	1.49	1831.27	1.0353	2.8802	4.7858	22.0061	76.3324
0.0307	1.58	1839.72	1.0370	2.8491	5.1247	19.8783	75.4619
0.0502	1.67	1847.51	1.0388	2.8202	5.5672	17.7761	73.5444
0.0704	1.75	1855.63	1.0404	2.7913	6.0097	15.6739	71.6269
0.0801	1.83	1863.92	1.0422	2.7618	6.4122	13.5717	69.7094
0.0902	1.91	1871.85	1.0440	2.7337	6.7947	11.4695	68.7919
0.1006	2.00	1879.29	1.0459	2.7072	7.3372	9.3673	67.8744

Table 3 Least-squares fit coefficients of the ultrasonic velocity equation $u = u_0 + u_1 m + u_2 m^2$ as a function of temperature

Coefficients	$T = 288.15$ K	$T = 298.15$ K	$T = 308.15$ K
u_0 (m·s^{-1})	1435.67	1473.38	1489.42
u_1 (m·s^{-1}·mol^{-1}·kg)	203.51	123.98	48.44
u_2 (m·s^{-1}·mol^{-2}·kg^2)	−398.26	−73.78	213.29
$\sigma_{[u]} \times 10$	2.6	1.5	3.8

Table 4 Limiting apparent molar compressibility ϕ_K^0, limiting apparent molar volume ϕ_V^0, constants S_K and S_V, and the A and B parameters of the Jones-Dole equation for glycylglycine in aqueous FeCl$_3$ solutions at $T = $ (288.15 K, 298.15 K and 308.15) K

Functions	$T = 288.15$ K	$T = 298.15$ K	$T = 308.15$ K
ϕ_K^0 (10^{13} m^5·mol^{-1}·N^{-1})	−32.74	−36.38	−42.91
S_K (10^{-8} N^{-1}·m^{-1}·mol^{-1})	57.63	61.25	69.40
ϕ_V^0 (m^3·mol^{-1})	−127.27	−119.62	−108.58
S_V (N^{-1}·m^{-1}·mol^{-1})	287.17	263.94	248.23
A (dm$^{1/2}$·mol$^{-1/2}$)	0.0289	0.0327	0.0395
B (dm^3·mol^{-1})	0.3487	0.3164	0.2525

where u_0, u_1 and u_2 are the fitted coefficients, and m is the molality of glycylglycine. The fitted coefficients along with the standard deviations are listed in Table 3.

The specific acoustic impedance is the product of the density and ultrasonic velocity of solvent/solution and can be expressed as:
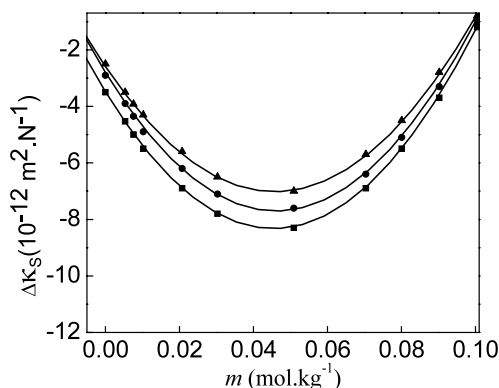
$$Z_0 = \rho_0 u_0 \tag{7}$$

The specific acoustic impedance (Z) and relative association (R_A) was calculated using $Z = \rho u$ and $R_A = (\frac{\rho}{\rho_0})(\frac{u_0}{u})^{1/3}$, where ρ and ρ_0 are the densities of the solution and solvent, respectively, while u and u_0 are their ultrasonic velocities.

4 Discussion

The derived parameters such as isentropic compressibility, change and relative change in isentropic compressibility, apparent molar compressibility, and apparent molar volume are given in Table 2. The limiting apparent molal compressibility, limiting apparent molar volume, constants S_K and S_V, and viscosity B-coefficient are given in Table 4.

The increase in ultrasonic velocity values with molality suggests that molecular association is important in glycylglycine–FeCl$_3$ aqueous solutions. The factors responsible for such behavior may be the presence of interactions caused by the zwitterionic state of glycylglycine [18] and the hydrophilic nature of aqueous ferric chloride [19]. This may be attributed to the cohesion resulting from ionic hydration. When glycylglycine is dissolved in an aqueous FeCl$_3$ solution, the water molecules are strongly attached to the ions by electrostatic forces [20] that introduce a greater cohesion into the solution. Increased associations

Fig. 1 Variation of the isentropic compressibility, $\Delta \kappa_S$, for glycylglycine in aqueous FeCl$_3$ solutions at: $T = 288.15$ K, ■; 298.15 K, ●; 308.15 K, ▲

observed in the above solutions may also be due to water structure enhancement brought about by an increase in electrostriction in the presence of FeCl$_3$. A shrinkage in the volume of the solvent, due to electrostrictive effect, is caused by the zwitterionic portion of glycyl-glycine. A similar effect was reported by earlier workers [20–22]. The cohesive forces are further enhanced by increasing the solute concentration. The added amount of glycylglycine zwitterions may also occupy the cavities of water clusters, which could lead to the formation of a denser structure for aqueous electrolyte solutions [23]. This process might continue until a concentration of solute is reached at which all cavities are filled. This viewpoint is also validated by Raman studies as a more compact zwitterions–water structure is formed in solutions with the addition of more solute [24].

Density is a measure of solvent–solvent and ion–solvent interactions. Increases in density with concentration indicate an increase in solvent–solvent and solute–solvent interactions and this may be due to shrinkage in volume owing to the presence of solute molecules. This can be attributed to the structure-making ability of the solute in the presence of the solvent. The change in structure of solvent or solution is a result of hydrogen bond formation, dissociation, and the hydrophobic (structure breaking) or hydrophilic (structure making) character of the solute.

The values of κ_S in the studied system show a decreasing trend due to: (i) an increase in number of incompressible molecules/zwitterions in solution, and/or (ii) the formation of a compact structure of zwitterions–water dipoles and a zwitterion–ion structure in solution. This shows that molecular association is greater for glycylglycine molecules, which exists in dipolar form in neutral solution and have stronger interaction with the surrounding water molecules. Water is regarded as an equilibrium mixture of two structures such as an ice-like structure and a closed packed structure [23]. An increase in electrostrictive compression of water around the molecules results in a larger decrease in the compressibility of the solution.

Values of the change in isentropic compressibility ($\Delta \kappa_S$) and relative change in isentropic compressibility ($\Delta \kappa_S / \kappa_0$) of glycylglycine in aqueous FeCl$_3$ solutions at different temperatures are reported in Table 2. The negative values of $\Delta \kappa_S$ increase with increasing molality of the solute. The variations for the change in isentropic compressibility ($\Delta \kappa_S$) are shown in Fig. 1. The negative values of the apparent molar compressibility (ϕ_K) and ϕ_K^0 indicate that electrostriction and hydrophilic interactions occur in this system, thereby suggesting the presence of solute–solvent interactions. The positive S_K values indicate the existence of ion–ion or solute–solute interactions.

Table 5 Specific acoustic impedance values (Z, 10^{-3} kg^{-2}·s^{-1}) for glycylglycine in aqueous FeCl$_3$ solutions at T = (288.15, 298.15 and 308.15) K

m (mol·kg^{-1})	$T = 288.15$ K	$T = 298.15$ K	$T = 308.15$ K
0.0000	1705.896	1739.093	1850.135
0.0052	1717.362	1750.669	1862.778
0.0076	1730.229	1761.770	1873.431
0.0103	1744.241	1773.543	1883.878
0.0208	1758.587	1786.510	1895.913
0.0307	1770.749	1798.429	1907.789
0.0502	1782.709	1810.808	1919.193
0.0704	1794.052	1824.031	1930.597
0.0801	1805.778	1836.949	1942.577
0.0902	1818.864	1849.534	1954.211
0.1006	1832.130	1864.225	1965.549

Values of the apparent molar volume (ϕ_V) are negative over the entire range of molalities due to solute–solvent interactions and electrostrictive solvation of the ions (Table 2). In the present system, it may be presumed that the interactions may take place as ion–dipolar/hydrophilic group interactions between ions of FeCl$_3$ and the (NH$_3^+$, COO$^-$), ($-$OH) groups of glycylglycine, and ions of FeCl$_3$ with the non-polar parts of glycylglycine. The values of limiting apparent molal volume (ϕ_V^0) have a non-linear rise with increasing temperature. This enhances the electrostriction of water molecules and also indicates smaller solute–solvent interaction in this system. The constant S_V exhibits positive values suggesting the presence of ion–ion interactions.

Viscosity is an important parameter for understanding the structure as well as molecular interactions occurring in solutions. The structural changes influence the viscosity to a certain extent as compared to density and compressibility. It is observed from Tables 1 and 2 that the viscosity increases with solute molality and the increasing trend indicates the existence of molecular interactions. In order to have a clear idea of this, the viscosity B-coefficient was also obtained. From Table 4 it is observed that the values of both A- and B-coefficient are positive. Because the A-coefficient is a measure of ionic interactions, it is evident that there are weak ion–ion interactions occurring in this system. The B-coefficient is known to be a measure of order or disorder introduced by the solute to the solvent. It is also a measure of solute–solvent interactions. The behavior of the B-coefficient in the above system suggests the existence of strong solute–solvent interactions. The magnitude of the B-coefficient values confirms that glycylglycine acts as an effective structure maker in aqueous FeCl$_3$ solution.

The specific acoustic impedance values increase with increase in molality as well as with temperature (Table 5). The trends of variation of Z with solute molality and temperature are similar to those exhibited by the variation of ultrasonic velocity. The increase in Z suggests greater interaction between solute and solvent through hydrogen bonding.

The values of R_A are presented in Table 6. The R_A values show an increasing trend with increasing concentration. A close examination of Table 6 reveals that the R_A values are slightly greater than one. This indicates that the system under investigation is essentially ideal in nature. Also, the variation in R_A values with temperature is insignificant over the studied temperatures. Consequently, the variation in temperature over the studied range does not seem to significantly affect the nature of intermolecular/interionic interactions operating in these solutions.

Table 6 Relative association values (R_A) for glycylglycine in aqueous FeCl$_3$ solutions at $T = (288.15, 298.15$ and $308.15)$ K

m (mol·kg^{-1})	$T = 288.15$ K	$T = 298.15$ K	$T = 308.15$ K
0.0000	1.0292	1.0269	1.0266
0.0052	1.0306	1.0277	1.0274
0.0076	1.0320	1.0285	1.0282
0.0103	1.0334	1.0294	1.0290
0.0208	1.0348	1.0303	1.0298
0.0307	1.0362	1.0311	1.0307
0.0502	1.0376	1.0320	1.0316
0.0704	1.0390	1.0329	1.0325
0.0801	1.0404	1.0338	1.0334
0.0902	1.0418	1.0347	1.0343
0.1006	1.0432	1.0356	1.0352

5 Conclusions

Hydrophilic intermolecular interactions and electrostriction occur in the studied system. The existence of ion–solvent or solute–solvent interactions results in attractive forces that promote the structure-making tendency, while ion–ion or solute–solute interactions result in dipole–dipole, dipole–induced–dipole and electrostrictive forces that enhance the structure-breaking properties of glycylglycine. The dipolar (zwitterions) characteristic of glycylglycine molecules give a clear picture of solute–solvent interactions and has proved to be most interesting owing to the strong H-bond interactions with water. Hence, it is evident from the above study that volumetric and ultrasonic velocity studies serve as a powerful probe in understanding the molecular interactions present in these solutions.

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