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Thermodynamics and transport properties of L-Proline in water and binary aqueous mixtures of acetonitrile at 303.15 K.

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Abstract: The knowledge of thermodynamic properties of amino acids in aqueous medium provides valuable information about the stability of proteins. In this study densities and viscosities of L-Proline in water and aqueous acetonitrile solution (5, 10, 15, 20, 25) % at 303.15 K are reported. From this data the apparent molar volume, partial molar volume and then corresponding partial molar volume of transfer were calculated. The data has further been investigated in terms of relative viscosity and B-coefficients using Jones Dole Equation. The results were used to interpret the concentration dependence of solute-solute and solute-solvent interactions.

Key words: Density, Apparent Molar volume, L-Proline, Acetonitrile.

Introduction

The study of thermodynamic stability of the native structure of proteins have been proved to be quite challenging and still remains a subject of extensive investigation[1]. Due to the complexities arising from the direct thermodynamics study of proteins, investigations on the behaviour of model compounds of proteins like amino acid and peptides in aqueous and mixed aqueous solvents make one to understand the factors governing the stability of the biopolymers[2-3]. The interpretation of behaviour of amino acids is quite helpful in understanding water - protein interactions in solutions. The volumetric, viscometric and acoustic studies of amino acids in aqueous and mixed aqueous solution of organic solvents can provide valuable information for understanding protein unfolding [4-7]. The mixed aqueous solvents are extensively used in chemistry and other fields to control factors like stability, reactivity and solubility of systems [8,9].In this paper, we are presenting the volumetric and viscometric studies of L-Proline in aqueous and mixed aqueous solution containing acetonitile. Proline, abbreviated as Pro or P, is an α-amino acid. It is not an essential amino acid which means that human body can synthesize it. The more common form is L-Proline. L-Proline is biosynthetically derived from the amino acid L-glutamate. It is only one of the twenty DNA-encoded amino acids which have a secondary amino group α to the carboxyl group. It is an essential component of collagen and is important for proper functioning of joints and tendons. It also helps to maintain and strengthen heart muscles. Further, it contributes to healthy immune system.

Additionally, acetonitrile is a widely used solvent which can dissolve a wide range of non-polar and polar chemical compounds. It is an aprotic solvent with a dielectric constant of 37.5. In this paper, an attempt has been made to understand the behaviour of L-Proline in water and water/acetonitrile system using volumetric and viscometric studies.

Experimental

L-Proline, (purity>99%) ,acetonitrile (purity>99%) were procured from S.D. Fine Chemicals. The L-proline was used after drying it under vacuum over calcium chloride in a vacuum desiccators. The triply distilled water obtained by heating it in $KMnO_4$ was used for all density measurements. All the solutions were made by weight. An electronic single pan five digit analytical balance (Mettler; Model AE-240) with a precision of ±0.00001 g was used for weighing. All the solutions were prepared by weight with care and stored in special airtight bottles to avoid the exposure of solution to air and evaporation..The density of solutions was measured using vibrating tube density meter (Model: DMA 4500M, Anton Paar, Austria) with an uncertainty of ±0.00005 g cm^{-3}. Before each series of measurement, it was calibrated using doubly distilled water and dry air at atmospheric pressure. The temperature was automatically kept constant within ± 0.03K with the help of in-built peltier system. The kinematic viscosities, υ (= η/ρ), of the experimental solutions were measured at303.15 K and at atmospheric pressure using suspended level Ubbelohde viscometer (Jain Scientific Glass Works). The viscometer was calibrated so as to determine the two constants A and B in the equation, $\eta/\rho = At - B/t$, obtained by measuring the flow time(t) with triply distilled water, distilled benzene and cyclohexane. The constants A and B have been found to be 0.0117 and –2.2467 corresponding to the time of flow for water as74.80 s, benzene 56.6 s and cyclohexane as 95.2 s at 298.15 K. The viscometer was filled with experimental solutions and the flow time measurements were made using anelectronic stopwatch with an accuracy of ±0.01 s. The measured values of kinematicviscosities were converted into dynamic viscosities after multiplication with the density.

Result and Discussion

The experimental densities of L-Proline in water and aqueous acetonitrile (5, 10,15, 20, 25) % as a function of molality of L-Proline at 303.15 K are given in Table 1.

Table 1: Molal concentration, density (ρ_0), apparent molar volumes (ϕ_v) of L-Proline in water and different compositions of water + acetonitrile at 303.15 K.

Water			
Molal conc. (m) (Mol Kg $^{-1}$)	**Density of Solvent (ρ_0) (kg m^{-3})**	**Density of Sample (ρ) (kg m^{-3})**	**Apparent molar volume ϕ_v x10^{-6} (m^3 mol^{-1})**
0.09994	0.99565	0.997926	92.71
0.19997	0.99565	1.000708	90.24
0.29993	0.99565	1.003773	88.53
0.39994	0.99565	1.007421	86.29
0.49997	0.99565	1.01149	84.17
5% Acetonitrile + Water			
Molal conc. (m) (Mol Kg $^{-1}$)	**Density of Solvent (ρ_0) (kg m^{-3})**	**Density of Sample (ρ) (kg m^{-3})**	**Apparent molar volume ϕ_v x10^{-6} (m^3 mol^{-1})**
0.09996	0.99384	0.996115	92.85
0.19995	0.99384	0.998709	91.30
0.29991	0.99384	1.001778	89.25
0.39996	0.99384	1.005102	87.65
0.49999	0.99384	1.009195	85.22
10% Acetonitrile + Water			
Molal conc. (m) (Mol Kg $^{-1}$)	**Density of Solvent (ρ_0) (kg m^{-3})**	**Density of Sample (ρ) (kg m^{-3})**	**Apparent molar volume ϕ_v x10^{-6} (m^3 mol^{-1})**
0.09998	0.99131	0.993516	93.73
0.19997	0.99131	0.996199	91.38
0.29994	0.99131	0.999128	89.82

0.39997	0.99131	1.002556	87.84
0.49998	0.99131	1.005856	86.96
15% Acetonitrile + Water			
Molal conc. (m) (Mol Kg $^{-1}$)	**Density of Solvent (ρ_0) (kg m^{-3})**	**Density of Sample (ρ) (kg m^{-3})**	**Apparent molar volume ϕ_v x10^{-6} (m^3 mol^{-1})**
0.09998	0.98954	0.991745	93.87
0.19999	0.98954	0.994352	91.89
0.29996	0.98954	0.997281	90.19
0.39999	0.98954	1.000819	87.87
0.49997	0.98954	1.004123	86.99
20% Acetonitrile + Water			
Molal conc. (m) (Mol Kg $^{-1}$)	**Density of Solvent (ρ_0) (kg m^{-3})**	**Density of Sample (ρ) (kg m^{-3})**	**Apparent molar volume ϕ_v x10^{-6} (m^3 mol^{-1})**
0.09996	0.98736	0.989579	93.88
0.19993	0.98736	0.992195	91.91
0.29992	0.98736	0.995097	90.34
0.39999	0.98736	0.998671	87.92
0.49999	0.98736	1.002003	87.00
25% Acetonitrile + Water			
Molal conc. (m) (Mol Kg $^{-1}$)	**Density of Solvent (ρ_0) (kg m^{-3})**	**Density of Sample (ρ) (kg m^{-3})**	**Apparent molar volume ϕ_v x10^{-6} (m^3 mol^{-1})**
0.09998	0.98553	0.987746	94.05
0.19999	0.98553	0.990316	92.30
0.29998	0.98553	0.993136	90.91
0.39999	0.98553	0.996803	88.13
0.49998	0.98553	0.99998	87.49

The apparent molal volumes (Φ_v) for L-Proline in aqueous and mixed aqueous solution of acetonitrile at 303.15 K wer calculated from density using Equation (1).

$$\Phi_v = \frac{M}{\rho_0} + \frac{1000(\rho-\rho_0)}{m\rho\rho_0} \qquad \textbf{(1)}$$

where **M** is the molecular weight of solute, **ρ_0** and **ρ** refer to the densities of solvent and solution, respectively and m is the concentration of L-Proline expressed usually as molality. The Φ_v data has been used to see the effect of acetonitrile concentration on solute-solvent interactions occurring in the ternary mixture of present study. The following linear regression of $\Phi_v$was carried out using the following equation

$$\Phi_v = \Phi_v^0 + S_v m \qquad \textbf{(2)}$$

Where $\Phi_v^0$is the partial molal volume or limiting apparent molal volume which is a measure of solute-solvent interactions, S_v is the experimental slope, also known as volumetric pair wise interaction coefficient [10,11], represents a measure of solute-solute interaction. The observed values of partial molar volume(Φ_v^0) and given in Table 2.

Table 2: Partial molar volumes (ϕ_v^0), and transfer volumes (ΔV_{tr}^0) of L-Proline in water and acetonitrile + water mixtures at 303.15 K.

Acetonitrile + Water	ϕ_v^0 x 10^6(m^3 mol^{-1})	ΔV_{tr}^0 (m^3 mol^{-1})
0	94.70(±0.187)	-
5	94.93(±0.275)	0.23
10	95.07(±0.455)	0.37
15	95.50(±0.399)	0.80
20	95.54(±0.389)	0.84
25	95.76(±0.530)	1.06

It is observed that the partial molar volume increase with increase in acetonitrile concentration. As ϕ_v^0 reflects the presence of solute-solvent interactions and it can be seen from the table 2 that ϕ_v^0 values are positive for the reported amino acid indicating strong solute-solvent interactions and these interactions further increases with the increase in concentration of acetonitrile. This may be caused by the reduced electrostriction of water due to (NH_3^+) and (COO^-) groups of the amino acid. The same observations have been shown by other authors [12,13] for amino acids in aqueous electrolyte solutions. The sample plot of apparent molar volume vs molality for L-Proline in water and water+acetonitrile at 303.15 K are shown in figures (1-3).

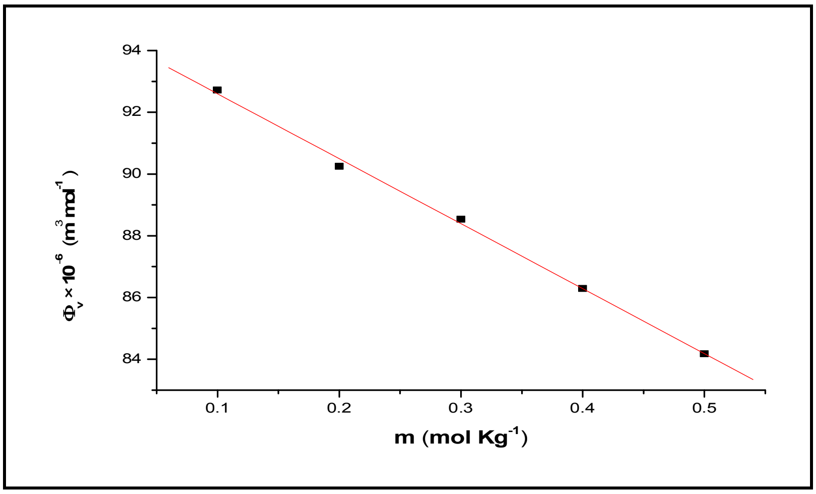

Fig1: Plot of apparent molar volume vs molality for L-Proline in water at 303.15 K.

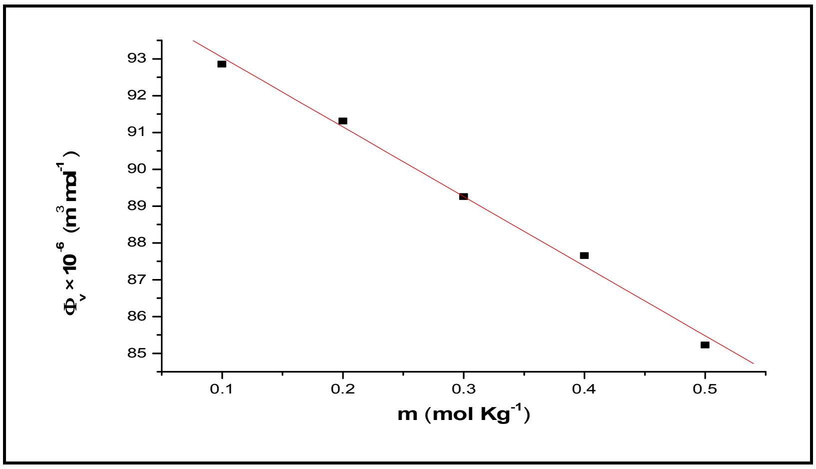

Fig 2: Plot of apparent molar volume vs molality for L-argenine in 5% Proline + water at 303.15 K.

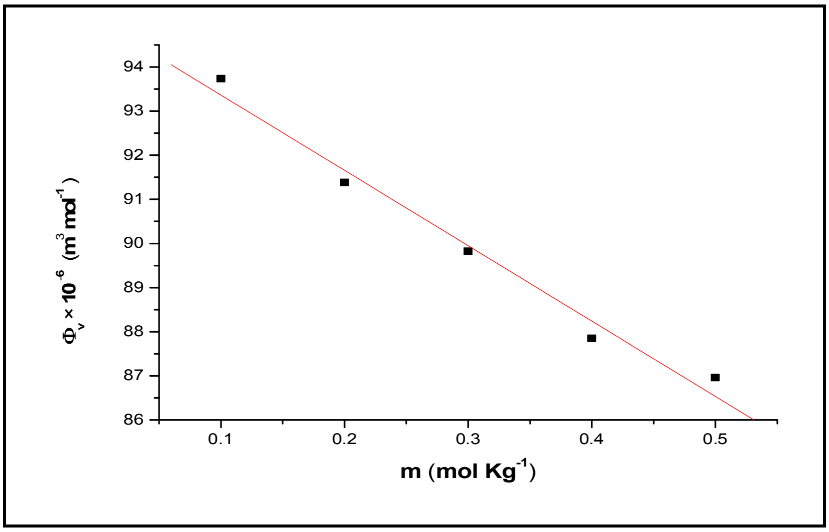

Fig 3: Plot of apparent molar volume vs molality for L-argenine in 10% Proline + water at 303.15 K.

The partial molar volume of transfer from water to aqueous acetonitrile was calculated by using the following equation:

$$\Delta V_{tr}^{0} = \Phi_v^0 \text{ (mixed solvent) -}\square\ \Phi_v^0 \text{ (in water)} \qquad (3)$$

The calculated values of partial molar volume of transfer at infinite dilution are given in Table 2. The partial molar volume at infinite dilution of a non-electrolyte is a combination of two factors and is given by the equation:

$$V_{2,m}^{0} = V_{int} + V_s \qquad (4)$$

Where V_{int} is the intrinsic molar volume of the non-hydrated solute and V_s is the contribution due to the interaction of the solute with water. V_{int} is made up of following type of contribution.

$$V_{int} = V_{vw} + V_{void} \qquad (5)$$

Where V_{vw} is the Vander Waals volume [14] and V_{void} is the volume associated with the voids and empty space present therein[15]. This equation was later modified to evaluate the contribution of a solute molecule to its partial molar volume at infinite dilution as:

$$V_{2,m}^{0} = V_{vw} + V_{void} - n\sigma_s \qquad (6)$$

Where σ_s is the shrinkage in the volume caused by the interaction of hydrogen bonding sites present in the solute with water molecules and n is the number of hydrogen bonding sites in molecule. The $V_{2,m}^{0}$ of amino acids can be written as :

$$V_{2,m}^{0} = V_{vw} + V_{void} - V_{shrinkage} \qquad (7)$$

By assuming that V_{vw} and V_{void} having same magnitude in water and aqueous acetonitrile the positive values of ΔV_{tr}^{0} are due to the increase in $V_{shrinkage}$ values.

Further ΔV_{tr}^{0} values can be explained on the basis of cosphere overlap model [16,17] in terms of solute-co solute interactions. According to this model, ionic-hydrophilic and hydrophilic-hydrophilic group interactions contribute positively, whereas hydrophilic-hydrophobic and hydrophobic -hydrophobic interactions contribute negatively to ΔV_{tr}^{0} values. The positive values of ΔV_{tr}^{0} of L-Proline in aqueous acetonitrile solution indicate that the former types of interactions are predominant over the latter.

The relative viscosity (η_{rel}) is calculated as ratio of viscosity of solution (η) and corresponding solvent (η_0). In order to evaluate viscosity B-coefficient, the value of (η_{rel}) is fitted by the method of least square fit using Jones Dole equation no.(8).

$$\eta_{rel} = \eta/\eta_o = 1 + Ac^{½} + Bc \qquad (8)$$

Where c is the molarity of solution. The value of viscosity B-coefficient depends upon the size of the solute and nature of solute- solvent interactions which is definite for solute-solvent system. The values of relative viscosity and B- coefficients are tabulated in Table 3.

Table3: Molarity, viscosity, relative viscosity and B- coefficients of L-Proline in water and water + Acetonitrile at 303.15 K.

Molar conc. ($C*10^2$) (Mol lit $^{-1}$)	**η (Pa s)**	**η_r**	**B - Coefficient (lit mol^{-1})**
Water			
9.86	0.82029	1.02831	0.018
19.56	0.84289	1.05665	
29.10	0.86548	1.08497	
38.52	0.88809	1.11331	
47.82	0.91069	1.14165	
5% Acetonitrile + Water			
9.84	0.89713	1.02901	0.022
19.52	0.92243	1.05802	
29.04	0.94772	1.08703	
38.43	0.97303	1.11606	
47.71	0.99834	1.14509	
10% Acetonitrile + Water			
9.82	0.92499	1.01729	0.030
19.47	0.92499	1.03437	
28.97	0.92499	1.05156	
38.33	0.92499	1.06875	
47.55	0.92499	1.08594	
15% Acetonitrile + Water			
9.80	1.10249	1.02083	0.031
19.44	1.12499	1.04166	
28.92	1.14748	1.06249	
38.27	1.16999	1.08333	
47.47	1.19248	1.10416	
20% Acetonitrile + Water			
9.78	1.15930	1.01710	0.032
19.39	1.17880	1.03420	
28.85	1.19829	1.05131	
38.19	1.21781	1.06843	
47.37	1.23731	1.08554	
25% Acetonitrile + Water			
9.76	1.26871	1.03087	0.033
19.36	1.30672	1.06175	
28.80	1.34471	1.09262	
38.12	1.38272	1.12350	
47.28	1.42071	1.15438	

The B- coefficient values are positive which shows strong alignment of acetonitrile with L-Proline solute molecules.The strong solute- solvent interaction immobilizes the neighbouring solvent molecules and presents large obstruction to viscous flow of solution thereby increasing its viscosity. The plot of relative viscosity and concentration is presented in Figures (4-6).

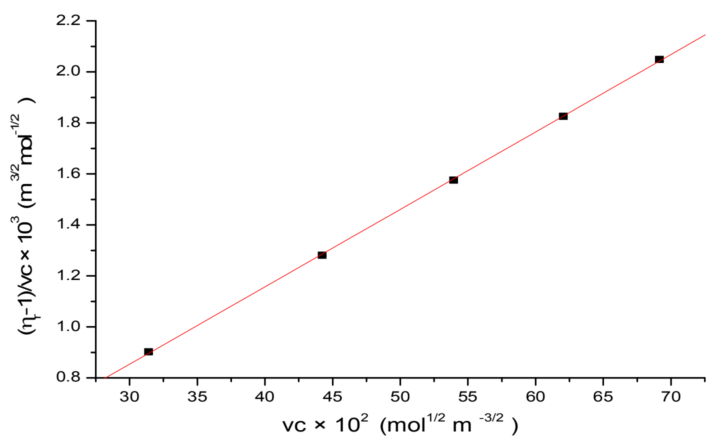

Fig4: Plot of (η_{r}-1)/√c VS √c for L-Proline in water at 303.15 K.

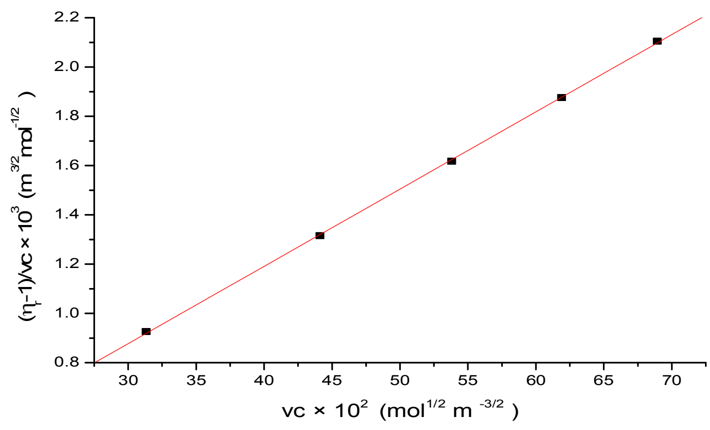

Fig 5: Plot of (η_{r}-1)/√c VS √c for L-argenine in 5% Proline + water at 303.15 K.

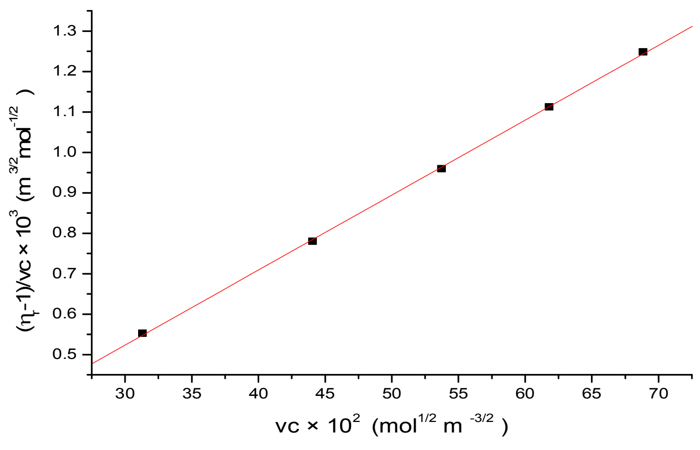

Fig 6: Plot of (η_{r}-1)/√c VS √c for L-argenine in 10% Proline + water at 303.15 K.

The hydrated solutes can be classified in two types; namely, kosmotropes i.e. structure makers while weakly hydrated ones are chaotropes i.e structure breakers. In general, positive B coefficients suggest kosmotropic behavior since strongly hydrated solutes exhibit a large change in viscosity with concentration.

Conclusions

Partial molar volumes of L-Proline have been determined in water and binary acetonitrile-water solutions at different concentrations of acetonitrile at 303.15 K . From this study it was observed that partial molar volumes increase with increase in acetonitrile concentrations. All these parameters support strong solute-solvent interactions in the ternary mixture of present study. Further the increase in B values with increase in acetonitrile concentrations indicates increase in obstruction to the free flow of water because of increase in co-solute concentrations..

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References:

1. Timasheff S N., Fasman G D., ‘Structure and stability of Biological Macromolecules; Marcel Dekker: New York, 1969, Vol 2(a) Chapter 2, p 65; (b) Chapter 3, p 213.
2. Raildi G., Biltonen R., Int. rev. sci: physical chemistry, Ed. HA, Skinner; Butterworth, 1975,10,147-189.
3. Kauzman W., Adv. Protein chem., 1959, 14,1.
4. Chalikian T.V, Volumetric Properties of Proteins, Ann. Rev. Biophys. Biomol. Struct., 2003, 32, 207-235 .
5. Enea O., Jolicoeur C., Heat capacities and volumes of several oligopeptides in urea-water mixtures at 25.degree.C. Some implications for protein unfolding.,J. Phys. Chem., 1982, 86, 3870-3881.
6. Taulier N., Chalikian T. V., Compressibility of protein transitions. Biochim. et Biophys. Acta - Protein Structure, 2002, 1595, 48-70.
7. Zhao H., Viscosity B-coefficients and standard partial molar volumes of amino acids, and their roles in interpreting the protein (enzyme) stabilization.,Biophys. Chem., 2006, 122, 157-183.
8. Wadi R.K., Ramasami P., Partial molal volumes and adiabatic compressibilities of transfer of glycine and DL-alanine from water to aqueous sodium sulfate at 288.15, 298.15 and 308.15 K., J. Chem. Soc. Faraday Trans., 1997, 93, 243-247.
9. Banipal T.S., Sehgal G., Partial molal adiabatic compressibilities of transfer of some amino acids and peptides from water to aqueous sodium chloride and aqueous glucose solutions.,Thermochim. Acta, 1995, 262, 175-183.
10. Desnover J.E., Structural effects in aqueous solutions- A thermodynamic approach., Pure Appl Chem.,1982, 54, 1469-1478.
11. Hedwig G.R., Reading J.F., Lilley T.H., Partial molar heat capacities and partial molar volumes of some N-acetyl amino acid amides, some N-acetyl peptide amides and two peptides at 25 °C , J Chem.Soc.Faraday Trans,1991,87,1751-1758.
12. Wen W.Y., Saito S., Apparent and Partial Molal Volumes of Five Symmetrical Tetraalkylammonium Bromides in Aqueous Solutions J. Phys. Chem., 1964, 68, 2639-44.
13. Yan Z., Wang J., Lu J., Effect of temperature on volumetric and viscosity properties of some α-amino acids in aqueous calcium chloride solutions Fluid Phase Equilib.,2004, 215, 143-150.
14. Bondi A., vander Waals Volumes and Radii. J Phys. Chem 1964, 68,441.
15. Bondi A., Free Volumes and Free Rotation in Simple Liquids and Liquid Saturated Hydrocarbons., J. Phys. Chem., 1954, 58, 929-939.
16. Gurney R.W., Ionic Process in solution; McGraw-Hill, New York, 1953, Vol.3, Chapter 1.
17. Bhat R., Kishore N., Ahluwalia J.C., Thermodynamic studies of transfer of some amino acids and peptides from water to aqueous glucose and sucrose solutions at 298.15 K, J. Chem. Soc. Faraday Trans., 1988, 84, 2651-2665.

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