wjert, 2022, Vol. 8, Issue 10, 44-89. *Original Article* **ISSN 2454-695X**

World Journal of Engineering Research and Technology
WJERT
www.wjert.org
SJIF Impact Factor: 5.924

PHYSICOCHEMICAL STUDIES OF BIOLOGICALLY POTENT MOLECULES IN AQUEOUS IONIC LIQUID SOLUTIONS WITH THE MANIFESTATION OF SOLVATION CONSEQUENCES AT DIFFERENT TEMPERATURES OPTIMIZED BY COMPUTATIONAL INVESTIGATIONS

Baishali Saha[a], Sanjoy Barman[a], Sukdev Majumder[a], Niloy Roy[a], Biswajit Ghosh[a], Rinku Chakrabarty[b], Subhankar Choudhury[c], Narendra Nath Ghosh[d], Koushik Baul[a] and Mahendra Nath Roy*[a,e]

[a] Department of Chemistry, University of North Bengal, Darjeeling – 734013, India.
[b]Department of Chemistry, Alipurduar University,Alipurduar736121, India.
[c]Department of Chemistry, Malda College, Malda, 732101, India.
[d]Department of Chemistry, University of Gour Banga, Mokdumpur, Malda, 732103, India.
[e]Vice-Chancellor, Alipurduar University, Alipurduar736121,India, Darjeeling – 734013.

Article Received on 26/07/2022 **Article Revised on 16/08/2022** **Article Accepted on 06/09/2022**

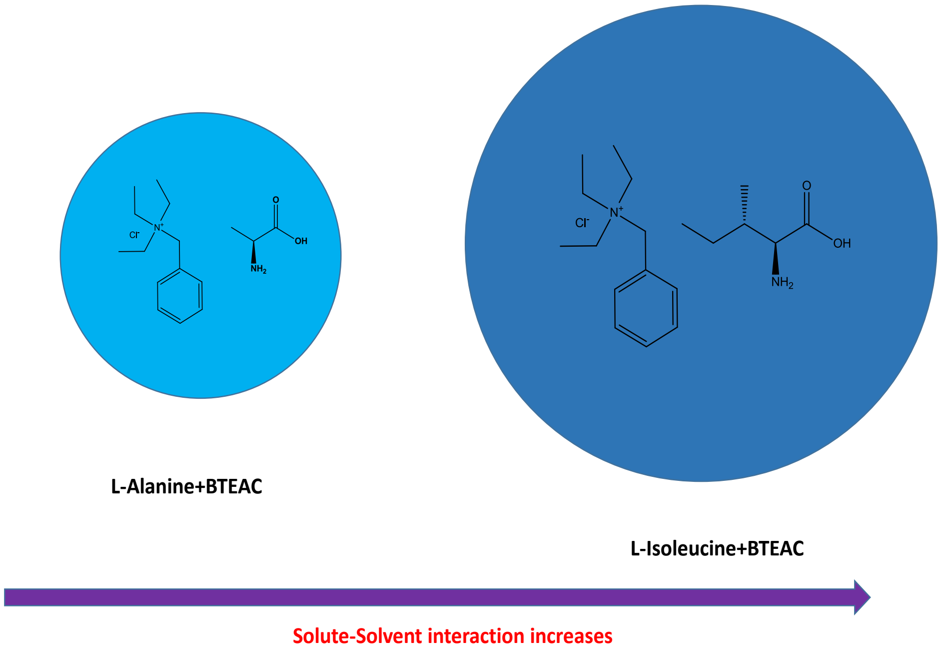
***Corresponding Author**
Mahendra Nath Roy
Department of Chemistry,
University of North Bengal,
Darjeeling – 734013, India.

ABSTRACT

By investigation of different evaluated physico-chemical parameters; density study, viscosity study, refractive index values, conductance study (at four different temperatures) and measurement of surface tension (at 298.15K), the molecular interactions of two different amino acids l-Alanine & L-Isoleucine in an aqueous ionic liquid Benzyltriethylammonium chloride have been studied & explained which is supported by UV-Vis &NMR studies. The strong solute-solvent interaction taking place in the solution have been identified depending on the apparent molar volume (ϕ_V) obtained from Masson equation, viscosity B-coefficient obtained from Jones-Doles equation, molar refraction(R_M), limiting molar refraction (R_M^0) from the Lorentz-Lorenz equation at 298.15K,303.15K,308.15K,313.15K and at 0.001m,0.003m,0.005m concentrations of solutions. Hepler's technique & positive value of (dB/dT) data suggest the structure breaking (chaotropic) nature. Specific conductivity measurement helps to explain the ionic nature of the system. The strong interactions present in the system are also suggested by the different thermodynamic data $\Delta\mu_1^{0\neq}$, $\Delta\mu_2^{0\neq}$, $\Delta H_2^{0\neq}$ and $T\Delta S_2^{0\neq}$. UV-Visible absorption study helps to understand the structural change & the nature of interactions of the solute amino acids with the solvent BTEAC obtained from association constant data. The change in chemical shift value in NMR gives us valuable insight about hydrophobic interactions and obtained a good agreement. The interaction of (L-Isoleucine + BTEAC) is stronger than (L-Alanine + BTEAC) according to the investigation & theoretical result. .Moreover, employing Density functional theory, adsorption energies, electrostatic potential maps (ESP), reduced density gradient plots and different thermodynamic parameters are analyzed to validate experimental observations.

KEYWORDS: Benzytriethylammonium chloride; L-Alanine; L-Isoleucine; apparent molar volume; Nuclear Magnetic Resonance; Computational details.

Graphical Abstract



1. INTRODUCTION

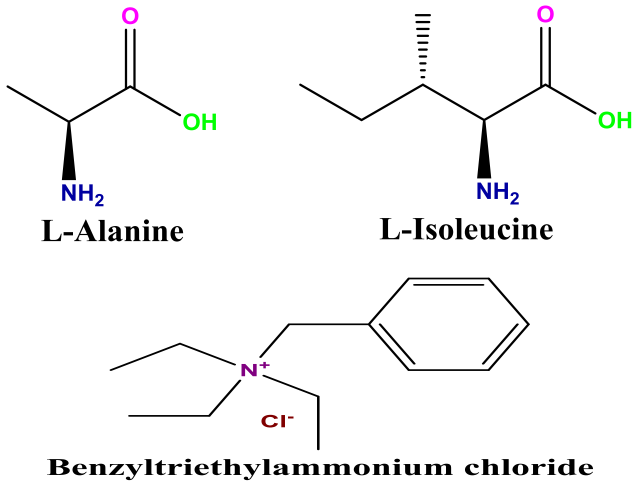
Ionic liquid (IL) defined mainly as organic salt, and they exist in liquid state even at room temperature having a low melting point less than 100°C.[1] It has gain considerable interest in academic as well as industry due to its wide applicability in many areas(lubricants, additives, heat storage).[2,3] Ionic liquids are also known as green solvent because it has ease of reuse, non-volatility, thermal stability and varieties of organic and organometallic compounds can easily dissolve in it.[4] Varieties of ILs can be easily prepared by choosing different combinations of cationic and anionic part which enhances their intrinsic characteristics, such as conductivity, density, viscosity, melting point, polarity, solvation power, hydrophilicity/ hydrophobicity, and acid/base character.[5] The thermophysical and thermochemical properties between ionic liquids and small organic molecules are of high importance in the development of industrial processes and biotechnologies. Benzyltriethylammonium chloride can catalyse poly condensation reactions to prepare polymers having high molecular weight under bi-phasic conditions and also using as liophilic phase-transfer catalyst.[6,7]

Amino acids is an organic compound that contain an amine group (-NH_2), a carboxylic acid group (-COOH) and a side-chain (-R) varying between different amino acids. Amino acids normally obtain as white crystalline solid and the crystalline lattice bind together because of charged interactions.[8] Amino acids being charged molecules are highly soluble in aqueous medium.[9] L-alanine and L-isoleucine which falls under non-essential and essential amino acids category are being used in the present study containing carboxylic acid side chain and generally known as acidic-polar amino acids.[10] The amino acids, the basic building units of proteins, have gain significant interests in multidisciplinary studies. L-Alanine is an example of α- amino acid which is used in biosynthesis of proteins, low blood sugar, in many metabolic processes, provides energy for muscles, brain and central nervous system. L-isoleucine is essential to human for different biological functions such as formation of haemoglobin, muscle growth, immunity boosting, metabolism of protein and glucose transportation.[11]

In our current study, we have chosen the ternary solutions of two different amino acids (L-Alanine and L-Isoleucine) in aqueous Benzyltriethyl ammonium chloride (BTEAC).Though in extensive research work, amino acids having different properties and structures with neutral, acidic, basic group and aliphatic chains, have interactions with this kind of organic quaternary ammonium-based ionic liquid solutions, is still a part of our investigation. Their significant conformations are affected by inter and intra-chain charge-charge interactions as well as hydrophobic interactions through the present side alkyl groups and organic electrolytes (BTEAC).[12] Thermophysical as well as thermodynamic properties of two different amino acids in aqueous ionic liquid provide important information about the structure interpretation (structure breaking) and molecular interactions using solute-solvent interactions.[13] Previously, different studies have already been performed with the same amino acids and ionic liquid.[14,15] However, to our best knowledge, different molecular interactions are yet to be explored with these ternary mixtures [(L-Alanine+BTEAC+H_2O) and (I-Isoleucine+BTEAC+H_2O).

In this work, the systematic measurements of density, viscosity, refractive index, surface tension and conductance of L-Alanine & L-Isoleucine in aqueous Benzyltriethylammonium chloride (shown in Scheme 1) have been investigated in different concentrations of solutions 0.001m, 0.003m and 0.005m at different temperatures 298.15K, 303.15K, 308.15K, 313.15K. From the above data, we have determined different thermodynamic parameters like apparent molar volume (ϕ_V), limiting apparent molar volume ($\phi_V{}^0$), Falkenhagen coefficient (A), viscosity B- coefficients (B), temperature derivative of B, dB/dT, $\Delta\mu 1$ °, $\Delta\mu 2$ °#, ΔH°# and ΔS°# aiming to understand the solute-solvent interaction with deep attention. The significant thermodynamic parameters have been used to explore such better interaction & the experimental datas are well supported by UV-Vis & proton NMR study.

Density functional theory is applied to calculate optimized geometries, molecular electrostatic potential maps and reduced density gradient.Our theoretical parameters also supports the experimental observations.



Scheme1: The molecular structure of L-Alanine, L-Isoleucine and Benzyltriethylammonium chloride.

2. Experimental Section

All the chemicals are purchased from different reputed chemical sources to carry out this work. . The reagents are always kept in the desiccators over P_2O_5 in dry atmosphere. Their chemical name, source, CAS number, mass fraction purity of chemicals used in this work are listed in Table 1.These chemicals were used without further purification.

Table 1: Details of Chemical Name, Attribution, CAS Number and Mass Fraction of Materials Studied.

Chemical Name	Source	CAS Number	Mass fraction purity
Benzyltriethylammonium chloride	Sigma-Aldrich	56-37-1	≥0.99
L-Alanine	LOBA CHEMIEPVT. LTD.	56-41-7	≥0.99
L-Isoleucine	SRL PVT.LTD.	73-32-5	≥0.99

2.2. Apparatus and procedure

Solubility of the taken compounds in deionised and triply distilled water has been checked surely,prior to start of the experimental work and L-alanine & L-isoleucine both are soluble in all portion of aqueous IL solution . Aqueous binary solution of Benzyltriethylammonium chloride that used as solvent is prepared by mass with the help of Mettler Toledo AG-285 having uncertainty ±0.01 mg and then working solutions of six sets are prepared by mass dilution. The uncertainty in molality for all the solutions is estimated to ±0.0001 mol.kg^{-3}.

The conversion of molarity into molality has been evaluated by the help of the following formula.[16]

$$m = \frac{1}{\{(\frac{\rho}{c}) - (\frac{M}{1000})\}} \qquad (1)$$

Where, m is the molality, ρ is density, the molarity (mol/L) of the solution is represented by C and the relative molar mass of the samples is expressed by M.

By the help of Vibrating U-tube Anton Paar digital density meter (DMA 4500 M) having a precision of ±0.00001 g cm−3 we can determine the densities of the solutions (ρ) maintained at ±0.01 K of the desired temperature of 298.15 K to 313.15 K after calibrating with triply distilled water as well as passing dry air earlier each set of experiment.

The viscosities for all the experimental sets are determined with a Brookfield DV-III Ultra Programmable Rheometer, having a spindle size of 42, by the help of the following equation:

$$\eta = (100/\mathrm{RPM}) \times \mathrm{TK} \times \mathrm{torque} \times \mathrm{SMC} \qquad (2)$$

Where, RPM, TK (0.09373) and SMC (0.327) stand for the speed, viscometer torque constant and spindle multiplier constant respectively. The calibration of instrument is done by using known viscosity samples such as water and aqueous $CaCl_2$ solutions as a reference. Calibration has done to avoid any needless errors before performing any experiment. Temperature of the solution has controlled and monitored at Brookfield Digital TC-500 temperature thermostat bath. The viscosities are determined with an accuracy of ±0.1%. The correctness of each viscosity measurement done is 0.3%, which is determined from the average of triplicate values.

Refractive index is determined by the help of digital refractometer Mettler Toledo bearing light emitting diode (LED) having source of light λ=589.3 nm. The instrument is calibrated with distilled water before and after every few experiments. The accuracy of refractive index measurement is ±0.0002 units.

Systronics-308 Conductivity Bridge and a CD-10 dip-type immersion conductivity cell with cell constant of approximately 0.1±0.001 $cm^{-1}$and a precision of 0.01 percent are used to measure the conductivity of the solutions. By the method suggested by Lind et al.[17] we have calibrated the conductivity cell .Before starting the experiment, the proper cell constant is verified by a freshly prepared 0.001 M aqueous KCl solution.

To measure the surface tensions of the solutions at 298.15K, a Tensiometer (K9, KRUSS; Germany) is used using the platinum ring detachment technique.

JASCO V-530 UV-VIS spectrophotometer is used to determine the UV spectra having uncertainity of wavelength ±2 nm. The temperature is kept constant by using thermostat.

600 MHz Bruker advance machine is used for recording the spectra at T=298.15K in D_2O for obtaining the ^1H NMR spectra as well as chemical shifts for various protons. The differences in chemical shift values are recorded to detect the interaction present between the ionic liquid and studied amino acids. The uncertainity of the chemical shift is measured to be better than 0.0005 ppm.

2.3 Computational details

All density functional theory (DFT) calculations in the present study were accomplished by Gaussian 16 program.[18] Ground state geometry optimizations of the of L-alanine and L-isoleucine, BTEAC and the corresponding complexes were carried out at B3LYP D3/6-31+G(d) level of theory. To account non-covalently bonded interaction energies (hydrogen bonding, π–π stacking) precisely dispersion corrected B3LYP D3 Hybridfunction was used.[19,20] During optimization, solvent effects (THF) were introduced by employing the Polarizable Continuum Model (PCM).[21,22] using the integral equation formalism variant in THF as fund in Gaussian. Furthermore, vibration frequency analysis was carried out to check whether the optimized geometries correspond to global minima on the potential energy surfaces using the same level of theory. Non Covalent Interaction (NCI) index plots of the reduced density gradient (RDG *or* s) vs. molecular density ρ were visualized using the Multiwfn 2.6[23]to interpret weak interactions viz. Vander Waals, H-bonding, staric interactions operating at the optimized geometries. Molecular electrostatic potential (MESP) maps were also analysed to understand the type of charge distributions and interactions operating in the complexes. Finally binding energy oradsorption energies (ΔE_{ads}) in aqueous medium for all complexes were evaluated using the following formula:

$$\Delta \mathbf{E}_{\mathbf{ads}} = \mathbf{E}_{\mathbf{BTEAC\text{-}Amino\ acid}} - \mathbf{E}_{\mathbf{BTEAC}} - \mathbf{E}_{\mathbf{Amino\ acid}}$$

Where $\mathbf{E}_{\mathbf{BTEAC\text{-}Amino\ acid}}$, $\mathbf{E}_{\mathbf{BTEAC}}$, $\mathbf{E}_{\mathbf{Amino\ acid}}$ are the total energy of the geometry optimized complexes, free BTEAC and the amino acids, respectively.

3. RESULTS AND DISCUSSION

The physical parameters, like density (ρ), viscosity (η) and refractive index (n_D) of binary mixtures consist of aqueous Benzyltriethylammonium chloride (BTEAC)solutions in different concentration (0.001m, 0.003m and 0.005m) at four different temperatures (298.15 - 313.15 K with an interval of 5 K) have been stated in Table S1.

The experimental measured values of the above physical parameters of L-Alanine and L-Isoleucine in different concentration (0.001m, 0.003m and 0.005m) of aqueous BTEAC solutions at four mentioned temperatures have been reported in Table S2-Table S5.

3.1 Apparent molar volume

For understanding interactions at the molecular level that take place in the solution phase, the significant parameters are apparent molar volume (ϕ_V) and limiting apparent molar volume ($\phi_V{}^0$). The geometric volume sum of the central solute molecule can represent the ϕ_V and the result changes from its interaction with solute amino acids in the solvent aqueous ionic liquid volume around the co-sphere .The following equation (3) is used to calculate the apparent molar volumes (ϕ_V) from the solutions densities,[24] and the values are given in the Table S6-Table S9.

$$\phi_V = M/\rho - (\rho - \rho_0)/m\rho\rho_0 \qquad (3)$$

Where *M* is the molar mass of the solute present, *m is* the molality of the solution, ρ and ρ_0 are the densities of the solution (Amino acids + BTEAC + H_2O), and solvent (BTEAC + H_2O) respectively.

It is observed that the (ϕ_V) values are positive for all the experimental cases, which represent the strong solute-solvent interactions & the apparent molar volumes (ϕ_V) decrease with increase in concentration (molality, *m*) of the taken amino acids in the same concentration of aq. BTEAC at the fixed temperature. It is further observed that apparent molar volumes (ϕ_V) increase with increasing temperatures and also with the molality of aqueous BTEAC solution and varied linearly with $\sqrt{m}$ using a least-squares treatment by the help of Masson equation.[25] The limiting molar volume, $\phi_V^{\,0}$ (partial molar volume expressed at infinite dilution) values are obtained from the equation-(4), which have been represented in Table 2.

$$\phi_V = \phi_V^{\,0} + S_V^{\,*}\sqrt{m} \qquad (4)$$

Where $\phi_V^{\,0}$ denotes limiting apparent molar volume near infinite dilution and $S_V^{\,*}$ is the experimental slope representing the solute-solute interaction. Only solvent molecules of BTEAC are present in the vicinity of each solute molecule at infinite dilution so $\phi_V^{\,0}$ has no effect for the interaction of solute-solute and it can only measure the solute-solvent interaction.

Table 2: Apparent molar volume ($\phi_V^{\,0}$), viscosity-*B* and viscosity-*A* coefficient and Molar Refraction ($R_M^{\,0}$)of (L-ALANINE + BTEAC + H_2O) and (L-ISOLEUCINE + BTEAC + H_2O) systems in aqueous ionic liquid BTEAC solutions of different concentrations(molality) 0.001m, 0.003m, 0.005m at temperatures 298.15 K, 303.15 K, 308.15 K and 313.15 K.

Temperature T (K)	$\phi_V^{\,0} \times 10^6$ ($m^3\ mol^{-1}$)	$S_V^{\,*} \times 10^6$ ($m^3\ mol^{-3/2}\ kg^{1/2}$)	*B* ($dm^3\ mol^{-1}$)	*A* ($dm^{3/2}\ mol^{-1/2}$)	$R_M^{\,0}$ ($m^3\ mol^{-1}$)
0.001m	(L-ALANINE + BTEAC + H_2O) system				
298.15	62.02	-7.83	0.2671	0.0770	18.31
303.15	63.43	-11.27	0.3880	0.0745	18.33
308.15	64.99	-15.99	0.4754	0.0700	18.35
313.15	66.33	-18.80	0.5788	0.0665	18.37
0.003m	(L- ALANINE + BTEAC + H_2O) system				

298.15	63.38	-11.24	0.3440	0.0757	18.32
303.15	64.92	-15.94	0.4562	0.0720	18.34
308.15	66.48	-20.68	0.5692	0.0688	18.36
313.15	67.88	-23.83	0.6671	0.0642	18.38
0.005m	(L- ALANINE + BTEAC + H_2O) system				
298.15	64.79	-15.31	0.3867	0.0739	18.33
303.15	66.45	-20.94	0.5597	0.0670	18.35
308.15	67.78	-24.89	0.6252	0.0651	18.37
313.15	69.14	-27.73	0.7665	0.0626	18.39
0.001m	(L-ISOLEUCINE + BTEAC + H_2O) system				
298.15	107.52	-10.92	06956	0.0729	26.97
303.15	109.24	-16.35	0.7654	0.0689	27.00
308.15	110.57	-18.54	0.8487	0.0658	27.03
313.15	112.12	-22.08	0.9772	0.0640	27.06
0.003m	(L-ISOLEUCINE + BTEAC + H_2O) system				
298.15	108.93	-14.99	0.8161	0.0677	26.98
303.15	110.72	-21.02	0.8531	0.0663	27.01
308.15	112.24	-24.04	0.9749	0.0632	27.04
313.15	113.62	-26.97	1.0631	0.0598	27.07
0.005m	(L-ISOLEUCINE + BTEAC + H_2O) system				
298.15	110.30	-18.48	0.9177	0.0658	26.99
303.15	112.02	-25.23	0.9611	0.0642	27.02
308.15	113.71	-29.17	1.0753	0.0616	27.05
313.15	114.88	-30.87	1.1708	0.0575	27.08

Combined standard uncertainty in molality according to mass purity of the samples is estimated to be u(m) = ± 0.0001 mol kg^{-1}; Standard uncertainties in temperature u(T) = ± 0.01K.

An inspection of Table 2 and Figure 1 reveals that ϕ_V^0 values are positive & is maximum for L-Isoleucine in 0.005m aqueous BTEAC solution at 313.15 K indicating the presence of stronger solute-solvent interaction between L-Isoleucine and aqueous BTEAC solution ,whereas minimum for L-Alanine in 0.001m aqueous BTEAC solution at 298.15K indicating least solute-solvent interaction between L-Alanine and aqueous BTEAC solution. The ϕ_V^0 has greater magnitude than S_V^* for solutions suggests that the stronger solute-solvent interactions of amino acids & BTEAC predominate over the weakening power of solute-solute interactions for both the cases in all measured temperature. S_V^* Indicates the level of solute-solute interaction and in both the systems (AA+IL), the values are negative and decrease with increasing temperature, which represents that solute-solute interaction is absent for the examined cases.

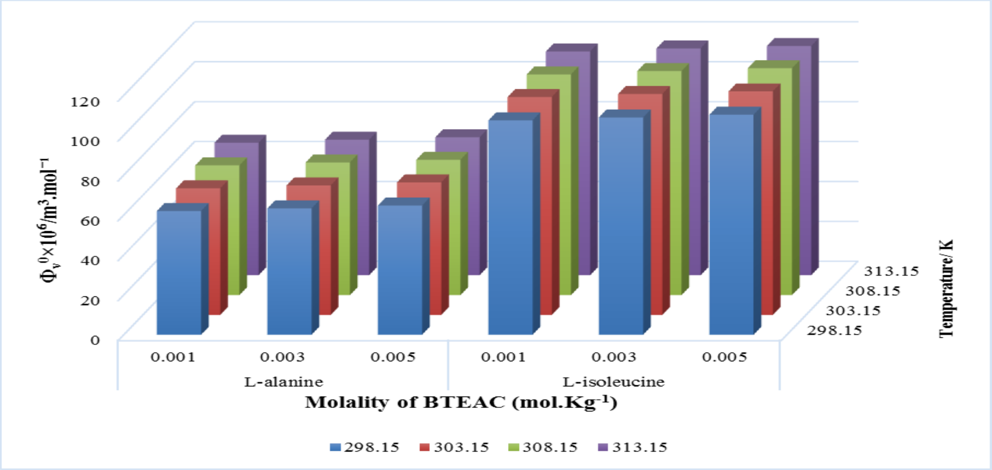


Figure 1: Plot of $\phi_V{}^0$ versus molality (*m*) of aqueous BTEAC solutions and as a function of temperature (T/K).

According to co-sphere overlap representation.[26,27] three different types of interactions which are possible between AA and BTEAC can be classified as: (a) ion-ion interactions present between $-NH_3^+$ group of AA(L-Alanine & L-Isoleucine) and Cl^-, and between $[BTEA]^+$ and COO^- group of amino acid; (b) ionic-hydrophobic interactions possible between the $[BTEA]^+$ or Cl^- and non-polar alkyl chain of AA and between the hydrophobic groups of BTEAC and $-NH_3^+$ or COO^- groups of amino acids; (c) hydrophobic-hydrophobic interactions present between the non-polar groups (benzyl, phenyl and alkyl group) of BTEAC and the non-polar alkyl part of AA. On comparing the structural context, for both the amino acids first interaction is applicable but types (b) & (c) are more effective in L-Isoleucine comparison to L-Alanine due to the presence of bigger hydrophobic non-polar alkyl group which has been verified later by the help of theoretical simulations.

The variation of $\phi_V{}^0$ with temperature range from 298.15K to 313.15 K is found to follow the following polynomial equation:

$$\phi_V{}^0 = a_0 + a_1 T + a_2 T^2 \qquad (5)$$

Where a_0, a_1 and a_2 represent the empirical coefficients depending on the nature of solute and the molality(m) of aqueous IL solution and T is the temperature expressed in Kelvin unit. Coefficient values so obtained of the above equation for L-Alanine & L-Isoleucine in different concentration of aqueous IL (BTEAC) are reported in Table 3.

Table 3: Values of various coefficients of equation-5 for L-alanine and L-isoleucine in different molalities (*m*) of aqueous IL (BTEAC) solutions.

Conc. of aq. IL solution in molality (mol.Kg^{-1})	$a_0\times10^6$ / m^3 mol^{-1}	$a_1\times10^6$ / m^3 mol^{-1}K^{-1}	$a_2\times10^6$ / m^3 mol^{-1}K^{-2}	$a_0\times10^6$ / m^3 mol^{-1}	$a_1\times10^6$ / m^3 mol^{-1}K^{-1}	$a_2\times10^6$ / m^3 mol^{-1}K^{-2}
	L-alanine + Aq. IL			L-isoleucine + Aq. IL		
0.001	-87.92	0.7056	-0.0007	-141.39	1.3418	-0.0017
0.003	-161.73	1.1873	-0.0015	-366.83	2.8181	-0.0041
0.005	-304.68	2.1457	-0.0030	-495.24	3.6708	-0.0055

Standard uncertainty in molality u(*m*) = ± 0.0001 mol kg^{-1}; Standard uncertainties in temperature u(*T*) = ± 0.01 K

The values of limiting apparent molar expansibilities, $\phi_E{}^0$ can be calculated for various temperature by the help of following equation,

$$\phi_E{}^0 = (\delta\phi_V{}^0 / \delta T)_P = a_1 + 2a_2T \qquad (6)$$

The $\phi_E{}^0$ is obtained by differentiation of eq. 6 with respect to temperature. Hepler recommended whether the solute has in long-range structure-making or breaking capacity in the aqueous mixed system can be assessed by examining the sign of $(\delta\phi_E{}^0/\delta T)_P$ [28].

$$(\delta\phi_E{}^0 / \delta T)_P = (\delta^2\phi_V{}^0 / \delta T^2)_P = 2a_2 \qquad (7)$$

The negative sign of $(\delta\phi_E{}^0 / \delta T)_P$ indicates the molecule is said to be a structure-breaker; otherwise, it is a structure-maker.[29] The inspection of Table 4 indicates that, $(\delta\phi_E{}^0 / \delta T)_P$ data of both amino acids are negative signs under investigation. The results recommend that both the taken amino acids (solutes) are definitely structure-breakers for all concentrations of the aqueous IL solutions under investigation, i.e., the solvent structure is obstructed by the solutes by increasing the solute-solvent interactions.

Table 4: Values of limiting molar expansibilities $(\delta\phi_E{}^0 / \delta T)_P$ for L-alanine and L-isoleucine in different concentration (molality) of aqueous IL (BTEAC) solutions at different temperatures.

Conc of Aq. IL Soln. in molality, m (mol.Kg^{-1})	$(\delta\phi_E{}^0 / \delta T)_P\times10^6$ / m^3 mol^{-1} K^{-2}	
	L-alanine + Aq. IL	**L-isoleucine + Aq. IL**
0.001	-0.0014	-0.0034
0.003	-0.0030	-0.0082
0.005	-0.0060	-0.0110

Standard uncertainty in molality u(*m*) = ± 0.0001 mol kg^{-1}; Standard uncertainties in temperature u(*T*) = ± 0.01 K

3.2 Viscosity

To understand and reveal the molecular interactions in solution, viscometric study is very important. The experimental viscosity data for studied system are given in Table S2-Table S4. The relative viscosity (η_r) has been analyzed with the help of the Jones-Dole equation.[30]

$$(\eta/\eta_0 - 1)/\sqrt{m} = (\eta_r - 1)/\sqrt{m} = A + B\sqrt{m} \qquad (8)$$

Where $\eta_r = \eta/\eta_0$ = the relative viscosity of the solution , η and η_0 represent the viscosities of solutions (amino acid + aqueous BTEAC) and solvent (aqueous BTEAC) respectively and *m* is the molality of amino acids in ternary solutions. The term *A* in equation (8) is known as Falkenhagen coefficient.[31] It is obtained by the ionic attraction theory of Falkenhagen-Vernon and accounts for the interaction of solute- solute and *B* suggests empirical constants also known as viscosity *B*-coefficients, is a measure of structural modifications induced by the solute−solvent interactions that arisen in the mixed medium. By plotting $(\eta_r - 1)/\sqrt{m}$ against $\sqrt{m}$, values of *A*- and *B*- coefficients are evaluated applying least-square technique and the evaluated data are stated in Table 2. The scrutiny of Table 2, it is seen that the *A*-coefficient values decrease with increase in temperature for both the studied amino acids. These results recommend the presence of very weak solute-solute (amino acid-amino acid) interaction exist in solutions and also in excellent agreement with those calculated from S_V^* values.

On the other hand, the viscosity *B*-coefficient.[32] has provided valuable information regarding the solvation and their influence on the structure of the solvent in the surrounding of solute molecules in solutions. It is found from Table 2 and Figure 2 that the values of *B*-coefficient are positive and much higher than the *A*-coefficient values, signifying that the solute-solvent interaction is prevailing over the solute-solute interaction. The positive magnitude of the viscosity *B*-coefficient increases with rising temperature and also with increasing the molality of aqueous BTEAC mixture, meaning that the solute-solvent interaction is enhanced with increasing temperature as well as the molality of aqueous BTEAC mixture. The *B*-values of L-Isoleucine in aqueous BTEAC solution in all respects are much greater than that for L-alanine, representing a greater solute-solvent interaction exists between L-isoleucine and aqueous BTEAC solution. These conclusions are in good agreement with those obtained using limiting apparent molar volume ϕ_V^0 values.

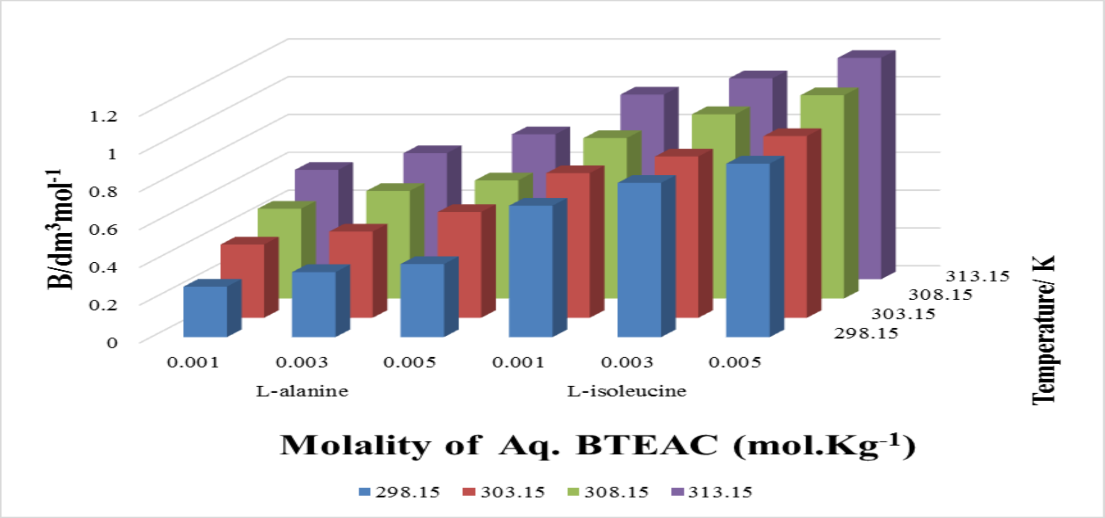


Figure 2: *B* plotted as a function of temperature (T/K) and different concentrations (m) of aqueous BTEAC solutions.

Positive value of *B*-coefficients suggest kosmotropes as strongly and tightly hydrated molecules of solute specify a greater transform in viscosity but negative values of *B*-coefficient show chaotropes for feebly and ineffectively hydrated solute molecules with concentration.[33,34,35] On the other hand, the *B*-coefficients may not be indicative, especially for large hydrophobic solutes. The high B/ϕ_V^0 value describes the primary solvation sheath.[36] The value of B/ϕ_V^0 is between 0 - 2.5 indicates unsolvated spherical species.[37] In place of the sign or magnitude of the *B*-coefficient, the structure-making or breaking capability of solute is better represented by d*B*/d*T* ratio.[38,39,40] The negative and positive sign of d*B*/d*T* recommend structure-making (kosmotropic) and structure-breaking (chaotropic) respectively. The d*B*/d*T* concept is based on Eyring's theory of viscosity,[41] which represents that a negative value of d*B*/d*T* corresponding to the higher activation energy for viscous flow in case of solution comparison to pure solvent.

According to table 5 & table 6, the values of *B*-coefficients are increasing with temperature for both the amino acids. Higher values of B/ϕ_V^0 suggest the formation of primary solvation shell and positive values of d*B*/d*T* indicate both the amino acids are categorized as structure-breaker.

Table 5: Values of B/ϕ_V^0 for L-alanine and L-isoleucine in different concentrations (m) of aqueous IL (BTEAC) mixtures at four different temperatures.

Temperature T (K^b)	Values of B/ϕ_V^0					
	0.001m	**0.003m**	**0.005m**	**0.001m**	**0.003m**	**0.005m**
	L-alanine + Aq. IL			L-isoleucine + Aq. IL		
298.15 K	4.3067	5.4276	5.9676	6.4695	7.4920	8.3200
303.15 K	6.1160	7.0260	8.4216	7.0066	7.7050	8.5797
308.15 K	7.3150	8.5620	9.2240	7.6757	8.6859	9.4565
313.15 K	8.7248	9.8276	11.0862	8.7157	9.3566	10.1915

Standard uncertainty in molality u(*m*) = ± 0.0001 mol kg^{-1}; ^{b} Standard uncertainties in temperature u(*T*) = ± 0.01 K

Table 6: d*B*/d*T* data of amino acids taken in the aqueous BTEAC solutions.

Concentration of Aq. IL Mixture in molality (mol.Kg^{-1})	(d*B*/d*T*) / dm^3 mol^{-1} K^{-1}	
	L-alanine + Aq. IL	**L-isoleucine + Aq. IL**
0.001	0.0204	0.0186
0.003	0.0216	0.0173
0.005	0.0241	0.0175

Standard uncertainty in molality u(*m*) = ± 0.0001 mol kg^{-1}; ^{b} Standard uncertainties in temperature u(*T*) = ± 0.01 K

According to Eyring and co-workers [34], the free activation energy for the viscous flow per mole of solvent, $\Delta\mu_1^{0\neq}$ could be evaluated by using the following equation:

$$\eta_0 = (hN_A/\overline{V}_1^0)\exp(\Delta\mu_1^{0\neq}/RT) \qquad (9)$$

Where *h* represents Planck's constant, N_A denotes Avogadro's number and the partial molar volume of the solvent is signified by $\overline{V}_1^0$. The equation (9) can also be known as follows:

$$\Delta\mu_1^{0\neq} = RT\ln(\eta_0\overline{V}_1^0/hN_A) \qquad (10)$$

Feakins et al.[43,44,45] suggested that if both the equations (8) and (10) are considered, then

$$B = (\overline{V}_1^0 - \overline{V}_2^0) + \overline{V}_1^0[(\Delta\mu_1^{0\neq} - \Delta\mu_2^{0\neq})/RT] \qquad (11)$$

Where $\overline{V}_2^0$ is the limiting partial molar volume (ϕ_V^0) of the solute and $\Delta\mu_2^{0\neq}$ represents ionic activation energy per mole of solute at infinite dilution. After reshuffling the equation (11),we can get equation (12) as,

$$\Delta\mu_2^{0\neq} = \Delta\mu_1^{0\neq} + (RT/\overline{V}_1^0)[B - (\overline{V}_1^0 - \overline{V}_2^0)] \qquad (12)$$

According to Table 7, the values of $\Delta\mu_2^{0\neq}$ are all positive and much greater than $\Delta\mu_1^{0\neq}$, showing that in ground state, the interaction present between the solute (amino acid) and the solvent (aqueous BTEAC mixture) molecules is stronger comparison to the transition state. In terms of free energy, the solvation of the solute is unfavorable in the transition state.

By the help of equation (13), the entropy of activation ($\Delta S_2^{0\neq}$)[36] for the solution has been determined.

$$\Delta S_2^{0\neq} = -d(\Delta\mu_2^{0\neq})/dT \qquad (13)$$

By using a least-squares method, $\Delta S_2^{0\neq}$ has been evaluated from the negative slope of the plots of $\Delta\mu_2^{0\neq}$ against *T*.

The enthalpy of activation ($\Delta H_2^{0\neq}$)[36] has been estimated by the help of this relation:

$$\Delta H_2^{0\neq} = \Delta\mu_2^{0\neq} + T\Delta S_2^{0\neq} \qquad (14)$$

As reference, the values of $\Delta S_2^{0\neq}$ and $\Delta H_2^{0\neq}$ are also listed and specified in Table 7.

According to Table 7, it is clear that the value of $\Delta\mu_1^{0\neq}$ is essentially constant for all the concentrations (molality) of the aqueous BTEAC mixture, suggesting that the dependence of $\Delta\mu_2^{0\neq}$ mainly based on the viscosity coefficients and $(\overline{V}_1^0 - \overline{V}_2^0)$ terms. At all the examined temperatures, Positive values of $\Delta\mu_2^{0\neq}$ and compositions of solvent investigated suggest that the process of viscous flow becomes more complex and difficult as the temperature and molality of the aqueous BTEAC mixture increases. As a result, the transition state formation becomes less attractive. From the proposal of Feakins et al.[36] for a substance having positive *B*-coefficients, $\Delta\mu_2^{0\neq} > \Delta\mu_1^{0\neq}$ representing the stronger solute-solvent interactions, accordingly meaning that the transition state formation is followed by breaking and rupturing the intermolecular forces in the solvent structure in the medium as well.[36,38,45] The negative values of $\Delta S_2^{0\neq}$ and $\Delta H_2^{0\neq}$ signifying that the formation of the transition state is related to bond-making and rising in order. Though a precise mechanism is difficult to develop for this, the disordered state of the slip-plane may be recommended.[36,39,46] According to Feakins et al. model, as $\Delta\mu_2^{0\neq} > \Delta\mu_1^{0\neq}$, the solutes (both amino acids) perform as structure-breakers. The solute's d*B*/d*T* behavior in aqueous IL solutions is supported by this.

Table 7: Values of $(\overline{V_1}^0 - \overline{V_2}^0)$, $\Delta\mu_1^{0\neq}$, $\Delta\mu_2^{0\neq}$, $T\Delta S_2^{0\neq}$, and $\Delta H_2^{0\neq}$ for L-Alanine and L-Isoleucine in different molality *(m)* of aqueous IL (BTEAC) solutions at various temperatures.

Parameters	0.001m				0.003m				0.005m			
	T=298.15K	303.15K	308.15K	313.15K	T=298.15K	303.15K	308.15K	313.15K	T=298.15K	303.15K	308.15K	313.15K
	L-Alanine + Aq. IL											
$(\overline{V_1}^0 - \overline{V_2}^0)\cdot 10^6/m^{3\cdot}mol^{-1}$	-43.72	-45.12	-46.64	-47.96	-44.63	-46.15	-47.67	-49.04	-45.59	-47.23	-48.51	-49.84
$\Delta\mu_1^{0\neq}$ /kJ.mol^{-1}	9.41	9.31	9.35	9.30	9.49	9.45	9.41	9.39	9.49	9.45	9.41	9.39
$\Delta\mu_2^{0\neq}$ /kJ.mol^{-1}	51.52	68.89	82.23	98.06	60.86	76.88	93.44	108.33	65.35	89.08	99.10	119.59
$T\Delta S_2^{0\neq}$/ kJ.mol^{-1}	-912.07	-927.37	-942.66	-957.96	-947.88	-963.77	-979.67	-995.57	-1030.08	-1047.35	-1064.63	-1081.90
$\Delta H_2^{0\neq}$ /kJ.mol^{-1}	-860.55	-858.48	-860.43	-887.01	-887.01	-886.90	-886.23	-887.23	-964.73	-958.28	-965.52	-962.31
	L-Isoleucine + Aq. IL											
$(\overline{V_1}^0 - \overline{V_2}^0).10^6/m^{3\cdot}mol^{-1}$	-89.22	-90.92	-92.22	-93.74	-90.18	-91.94	-93.43	-94.78	-91.09	-92.79	-94.44	-95.58
$\Delta\mu_1^{0\neq}$ /kJ.mol^{-1}	9.41	9.31	9.35	9.30	9.49	9.45	9.41	9.39	9.56	9.55	9.51	9.47
$\Delta\mu_2^{0\neq}$ /kJ.mol^{-1}	115.74	127.10	140.71	160.96	129.30	136.30	154.93	169.37	139.75	147.64	165.07	180.30
$T\Delta S_2^{0\neq}$/ kJ.mol^{-1}	-890.07	-904.99	-919.92	-934.85	-827.93	-841.82	-855.70	-869.59	-829.24	-843.15	-857.06	-870.96
$\Delta H_2^{0\neq}$ /kJ.mol^{-1}	-774.33	-777.89	-779.21	-773.89	-698.64	-705.52	-700.77	-700.22	-689.49	-695.51	-691.99	-690.67

Standard uncertainty in molality u(*m*) = ± 0.0001 mol kg^{-1}; Standard uncertainties in temperature u(*T*) = ± 0.01 K

3.3 Refractive Index

The measurement of refractive index (n_D) is also an important aspect to discover the molecular interactions that occur in the solution. The Lorentz-Lorenz relation is used to evaluate the molar refraction (R_M) by the help of the following equation-(15).[40,47]

$$R_M = \{(n_D^2 - 1)/(n_D^2 + 2)\}(M/\rho) \qquad (15)$$

Where R_M, n_D, M and ρ suggest the molar refraction, refractive index, molar mass and density of the solution respectively. The n_D of a material is expressed by the ratio c_o/c , where c and c_o are the speed of light in a particular medium & vacuum respectively. The substance having higher refractive index, shows the efficiency to refract light more easily as the refractive index of a compound depicts its capability to refract light as it starts passing with one medium then into the next [48]. As suggested by Deetlefs et al. [49],the refractive index of a substance is higher when the substance contains more closely packed and denser molecules. According to the inspection of Table S2-Table S5 and Table S6-Table S9,it is proved that the refractive index and molar refraction for the studied L-Isoleucine are greater than L-Alanine in different molality of aqueous BTEAC, signifying that the molecules are more closely packed in the L-Isoleucine + aqueous BTEAC solution.

The following equation (16) is used to evaluate the Limiting molar refraction ($R_M{}^0$) and recorded in Table 2.

$$R_M = R_M{}^0 + R_S\sqrt{m} \qquad (16)$$

Therefore, we observed that (Table 2 and Figure 3) the values of $R_M{}^0$ increase with the increasing temperature, as does the molality of the aqueous BTEAC solution, indicating that the solute-solvent interaction is enhanced with the rise in temperature and molality of aqueous BTEAC solution. The $R_M{}^0$-values of L-Isoleucine in the aqueous BTEAC solution are much greater than that for L-Alanine in all respects, expressing a greater solute-solvent relationship exists in L-Isoleucine. These findings are also in good agreement with those obtained from the apparent molar volume $\phi_V{}^0$ and viscosity *B*-coefficients values as explained earlier.

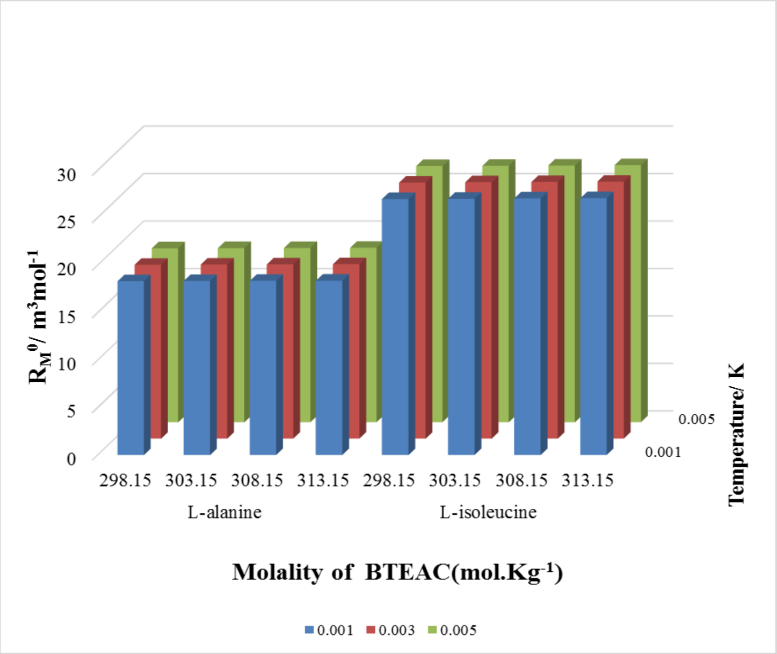


Figure 3: Plot of limiting molar refractive index ($R_M{}^0$) as a function of different molality of aqueous BTEAC solutions and as a function of temperature (*T/K*).

3.4 Conductimetric study

Conductometric study is very much helpful to explore the phenomena occurring in the solution. The conductance study of two aqueous (BTEAC + H_2O) amino acids solutions have been performed at four above mentioned temperatures. The benefits of this study is to discover the interactions (solvent-solvent and solute-solute interactions) and transport phenomena of the L-Alanine + BTEAC + H_2O and L-Isoleucine + BTEAC + H_2O.[50]

The molar conductivities.[51] (*Λ*) of aqueous BTEAC have been seen with increasing the concentration of L-Alanine and L-Isoleucine at four different temperatures and have been reported in Table S10.The molar conductance(*Λ)* values increase with increasing in temperature as well as increasing concentration of IL solution and decrease continuously with increasing concentration of amino acid (Figure 4-7). Regardless of increasing the number of ionic species in solution with the addition of L-Alanine or L-Isoleucine solution in the solution phase, the molar conductivity decreases.[52] It might be attributed because of the enlargement of solute-solvent interactions in solution mixtures between the solute and solvent molecules, which are controlled by ion-ion, ion-hydrophobic, and hydrophobic-hydrophobic interactions. The development of molecular assembly has the potential for ionic species to lose their ability to travel in a solitary state, making them less ambulant and therefore less likely to display conductivity in the solution. Thus, the conductimetric analysis strongly assists the volumetric, viscometric, and refractometric studies and confirms the same perception. Therefore, the free ions are more available in (L-Alanine +Aq.BTEAC) solution showing higher conductance values than (L-Isoleucine +Aq. BTEAC) solution.

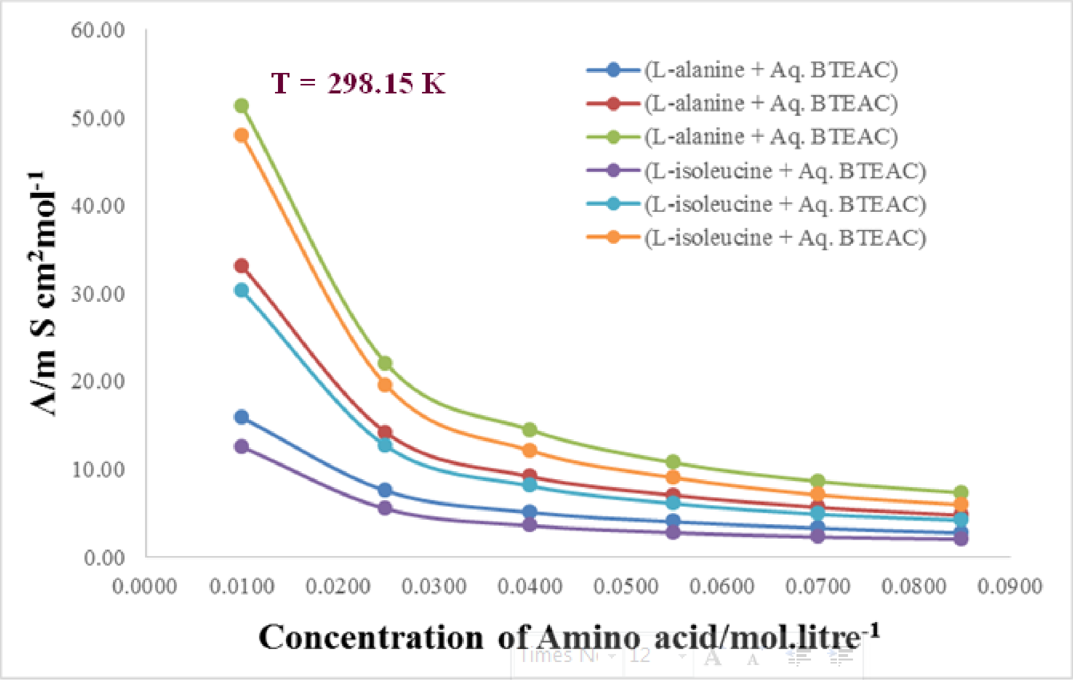


Figure 4: Plot of molar conductance as a function of the concentration of Amino acids taken in various molality of aqueous BTEAC in solutions at 298.15 K.

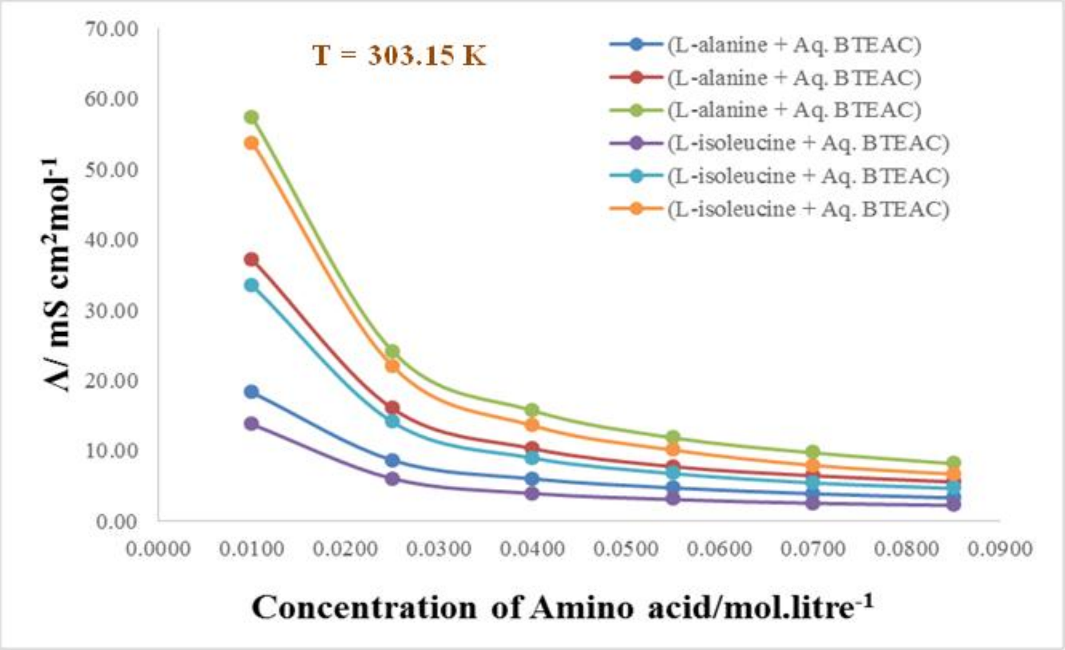


Figure 5: Plot of molar conductance as a function of the concentration of Amino acids taken in various molality of aqueous BTEAC in solutions at 303.15 K.

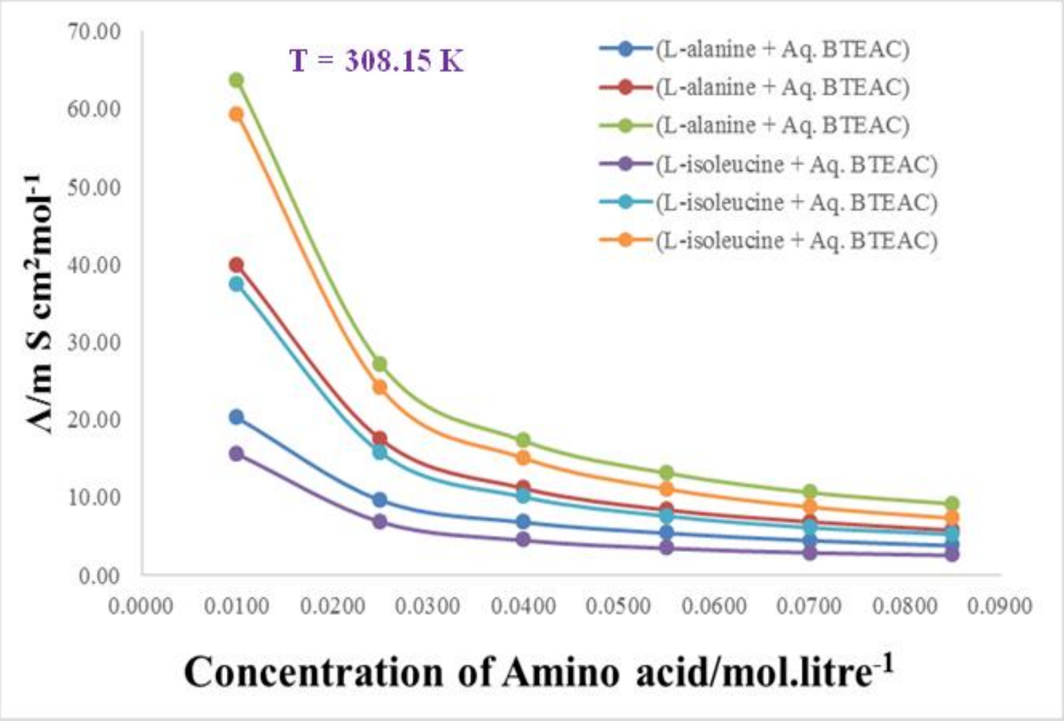


Figure 6: Plot of molar conductance as a function of the concentration of Amino acids taken in various molality of aqueous BTEAC in solutions at 308.15 K.

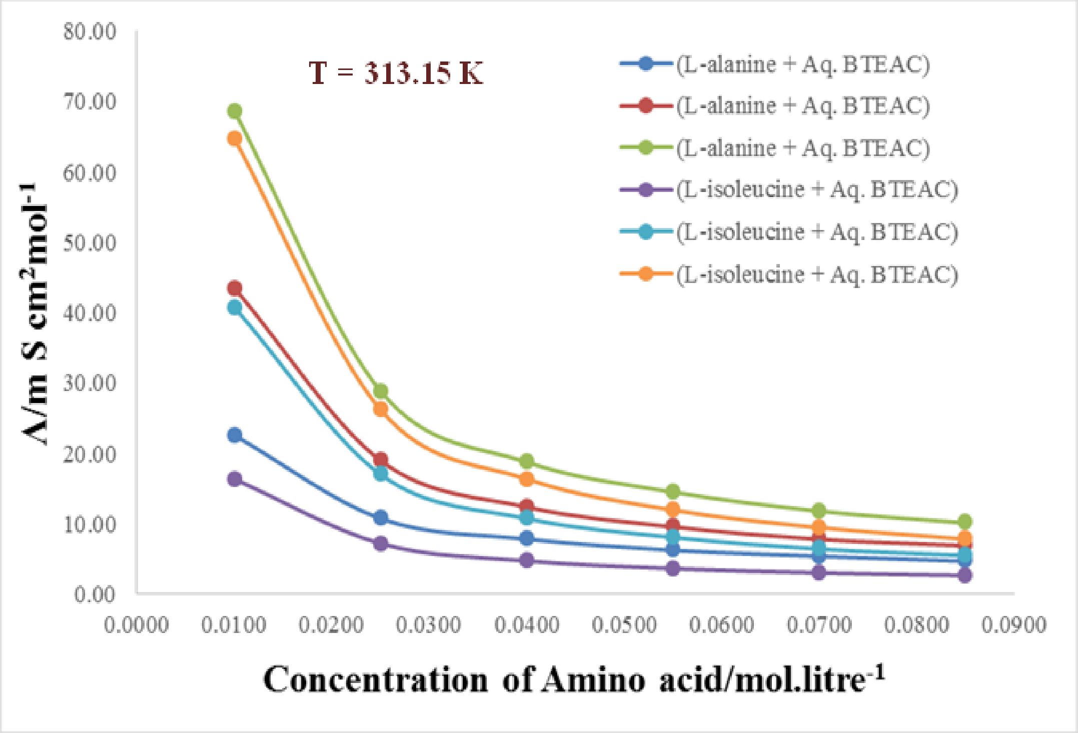


Figure 7: Plot of molar conductance as a function of the concentration of Amino acids taken in various molality of aqueous BTEAC in solutions at 313.15 K.

3.5 Surface Tension

The experimental surface tension values of aqueous solution of BTEAC and amino acid at T= 298.15 K are reported in Table S11. Figure 7 shows the variation of surface tension of aqueous solution of BTEAC and amino acid with the concentration (molality) of amino acid at T= 298.15 K. It is observed that, a rapid increase of surface tension as the concentration of L-Alanine increases. On the other hand, the surface tension decreases with increasing concentration of L-Isoleucine.

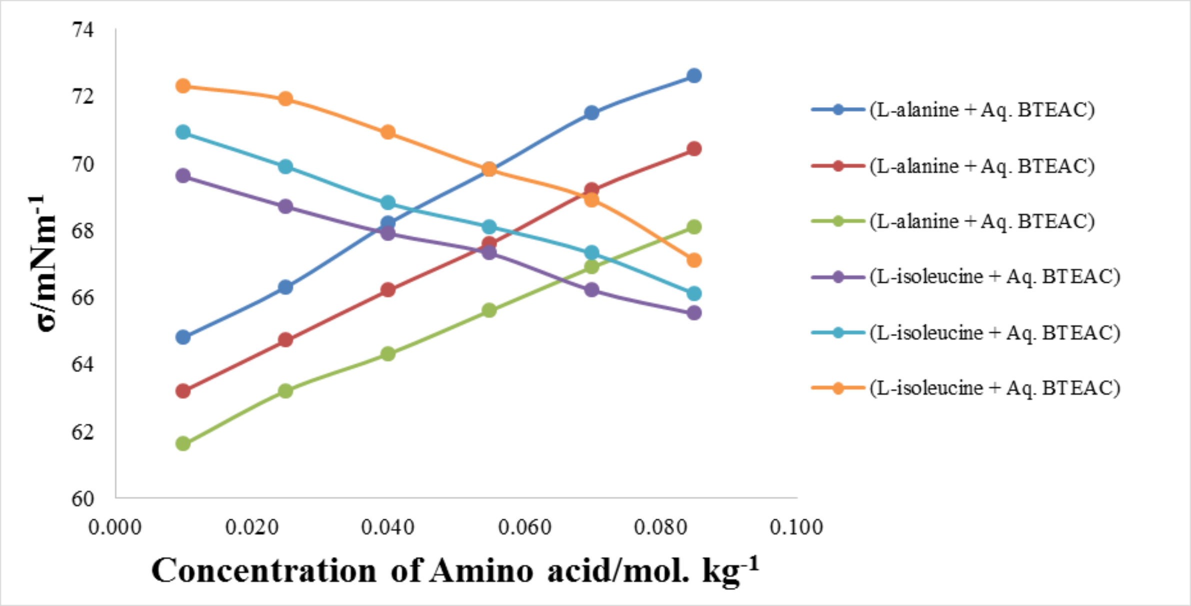


Figure 8: Plot of Surface Tension (*σ)* of L-Alanine and L-Isoleucine as a function of different concentration (molality) of aqueous BTEAC solutions at 298.15 K.

The sign and magnitude of the limiting slopes of surface tension with respect to concentration(molality) are representing the hydrophobic or hydrophilic character of the solute, as it describes the superior form of interaction on the surface[53,54]. From the very dilute range, the limiting slopes were evaluated by means of data, and these are displayed in Table 8.

Table 8: Limiting Slopes (∂σ/∂m) data of the surface tension of amino acids taken in the aqueous solutions.

Concentrations of aq. IL Mixture in molality (mol.Kg^{-1})	**(∂σ/∂m)** / mN m^{-1}.Kg. mol^{-1}	
	L-Alanine + Aq. IL	**L-Isoleucine + Aq. IL**
0.001	107.05	-54.48
0.003	96.95	-61.91
0.005	85.52	-68.76

Positive value of (∂σ/∂m) for L-Alanine is characteristic of electrolytes and very polar hydrophilic compounds [55, 56], and they can be described as a consequence of a favourable interaction between zwitterionic groups and cations and anions of the IL (BTEAC).On the other hand, negative sign of (∂σ/∂m) for L-Isoleucine is typical for aqueous solution of surface-active solutes or hydrophobic compounds. This result implies that effect of hydrophobic side chain is more dominating over ionic portion for L-Isoleucine and reverse is true for L-Alanine.

3.6. UV-Visible spectra measurement

UV-Vis absorption study is used to explore the structural change & the nature of interactions of the solute amino acids with the solvent BTEAC by providing information about the stability of the molecular association by calculating the association constants(K_a) or stability constant.[57] The study of such interactions for different mixture of solutions were recorded by varying concentration L-Alanine/L-Isoleucine in absorbance (A) of BTEAC at λmax = 209 nm (Fig.9) at 298.15 K to determine the association constant(Tables S10 & S11). The association constant (K_a) for both the system (BTEAC+ L-Alanine) & (BTEAC+ L-Isoleucine) were obtained from the double reciprocal plots.[58] using the Benesi-Hildebrand equation, were observed to be linear (Fig.10), usually gives the idea of solute- solvent ratio in solution.[59]

$$\frac{1}{\Delta A} = \frac{1}{\Delta\varepsilon[\mathrm{IL}]\mathrm{Ka}}\frac{1}{[\mathrm{AA}]} + \frac{1}{\Delta\varepsilon[\mathrm{IL}]} \tag{17}$$

Where [IL] & [AA] refers to the total concentration of the BTEAC & amino acids respectively(L-Alanine/L-Isoleucine), ΔE denotes the change in molar extinction co- efficient between the absence & presence of amino acids with the ionic liquid & ΔA represents the absorption changes of BTEAC on the addition of amino acids. The values of K_a for each of the system were determined from intercept/slope of the plots using equation (17). The value of K_a for (BTEAC +L-Alanine) is 837.0588 M^{-1} whereas for (BTEAC+ L-Isoleucine) is 3327.299 M^{-1}.[60]

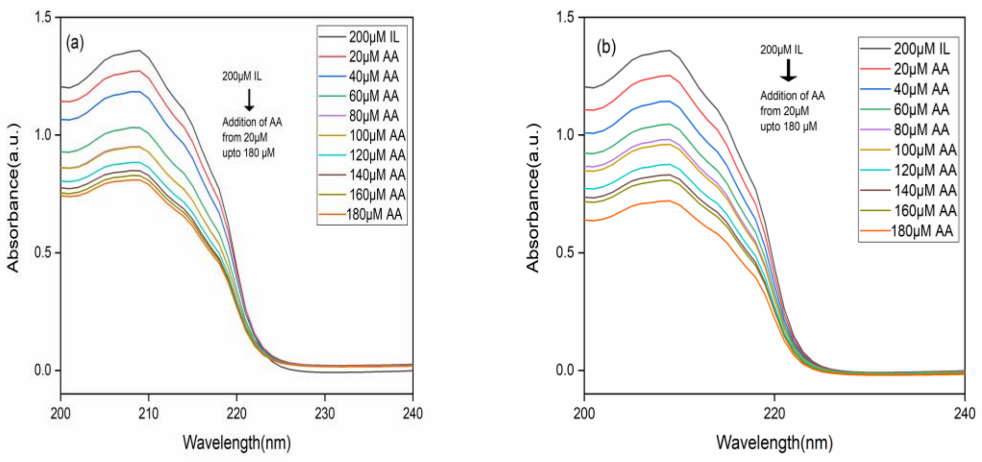
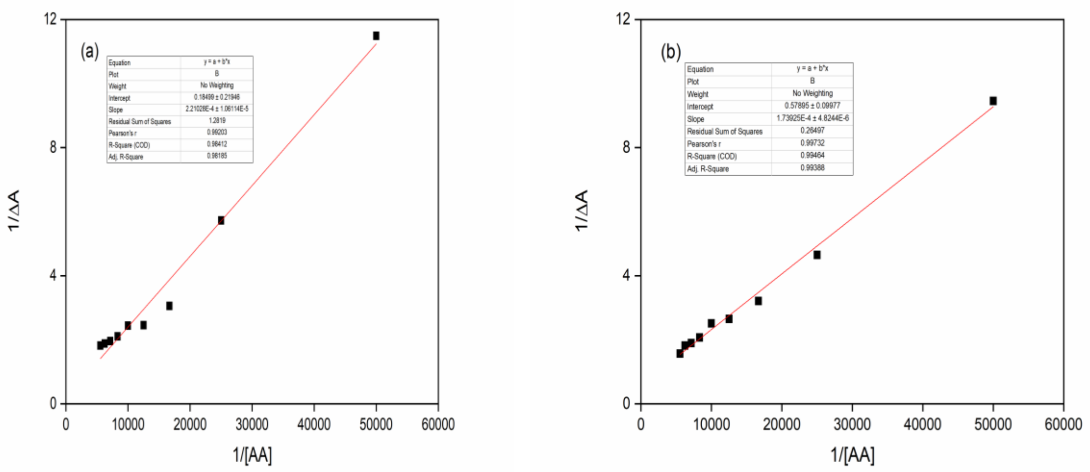


Figure 9: Benesi-Hildebrand double reciprocal plot of (a) BTEAC+ l-Alanine (b) BTEAC+L-Isoleucine system.



(a)

Equation	y = a + b*x
Plot	B
Weight	No Weighting
Intercept	0.18499 ± 0.21946
Slope	2.21028E-4 ± 1.06114E-5
Residual Sum of Squares	1.2819
Pearson's r	0.99203
R-Square (COD)	0.98412
Adj. R-Square	0.98185

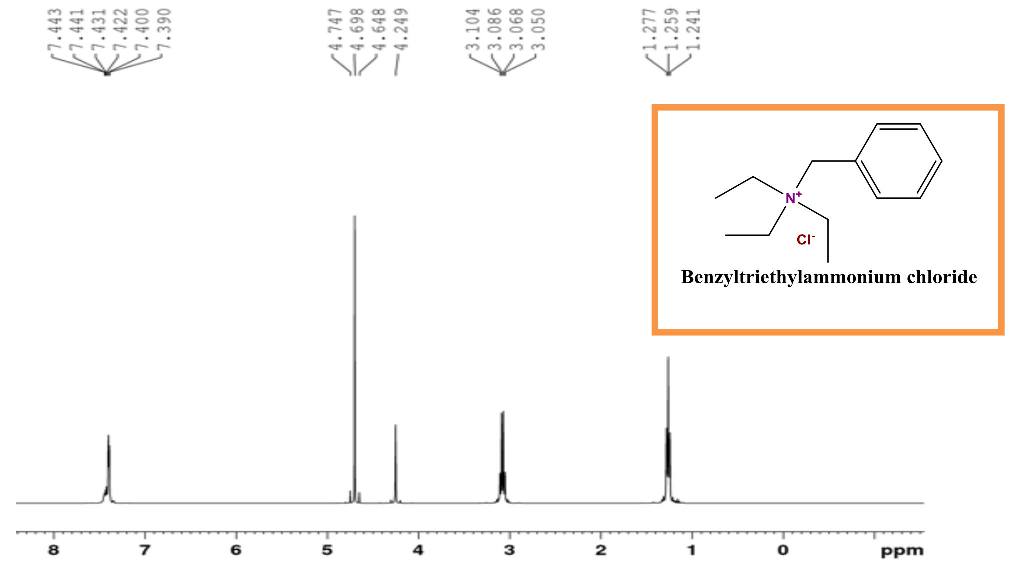
(b)

Equation	y = a + b*x
Plot	B
Weight	No Weighting
Intercept	0.57895 ± 0.09977
Slope	1.73925E-4 ± 4.8244E-6
Residual Sum of Squares	0.26497
Pearson's r	0.99732
R-Square (COD)	0.99464
Adj. R-Square	0.99388

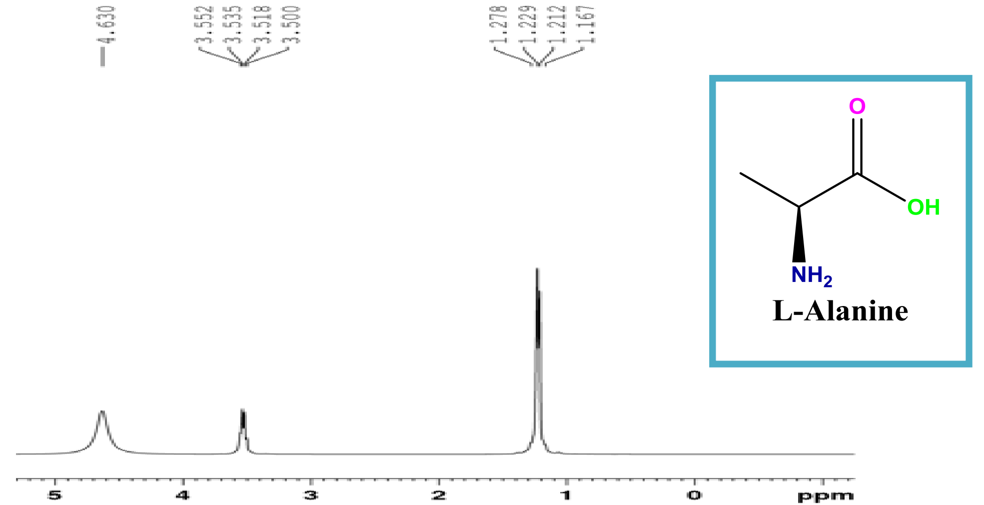
Figure 10: Double reciprocal plot by using Benesi–Hildebrand Method to obtain Slope and intercept of the straight line at 298.15K Temperature for (a) BTEAC + L-Alanine and (b) BTEAC+ L-Isoleucine system.

3.7 NMR Studies

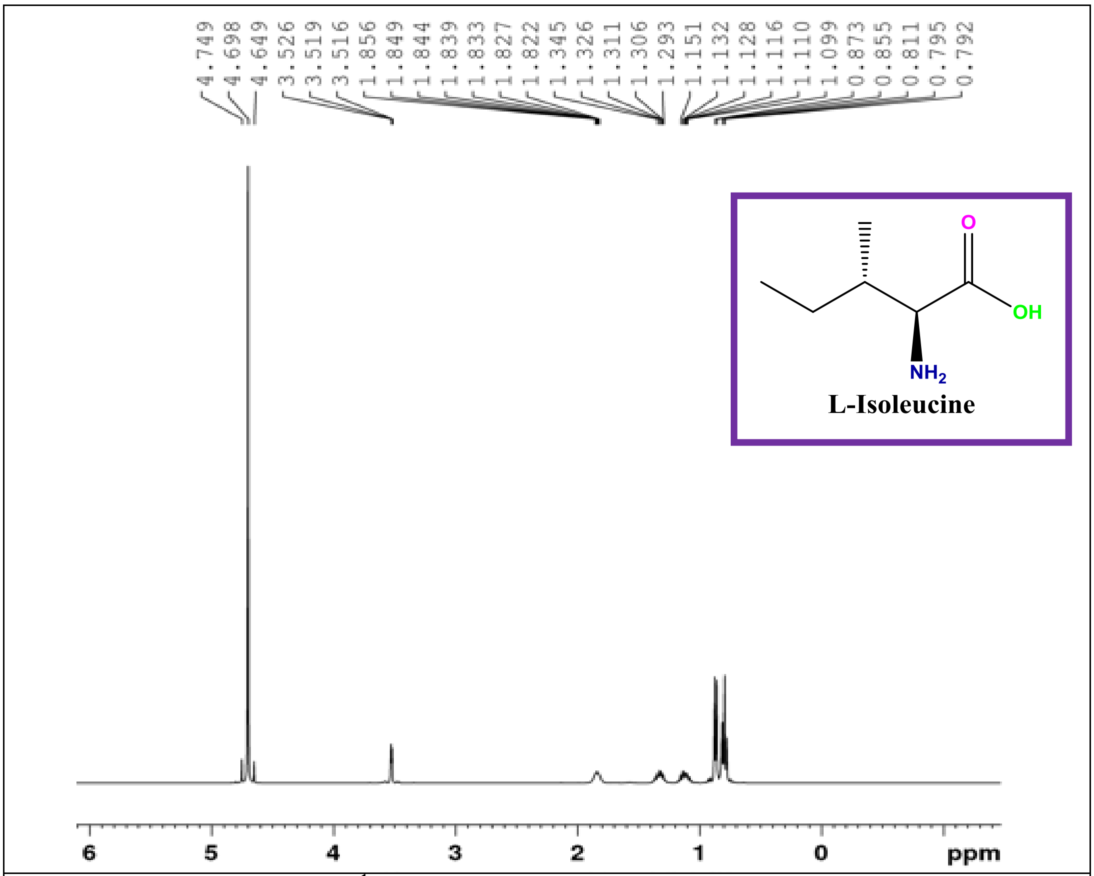
By the help of ^{1}H NMR spectroscopy [55,61], we can explicate the shifts in the electronic location of different protons of BTEAC in the presence of amino acid. The chemical shift of all detectable protons (δ1 to δ4) of BTEAC in D_2O is shown in Picture 1 of Figure 11. The results illustrate that the ^{1}H NMR spectrum of $[BTEA]^+$ ion has four characteristic peaks connected with the terminal $–CH_3$ (δ1) protons of the ethyl chain at 1.241-1.277 ppm, ethyl $–CH_2$ (δ2) protons at 3.050-3.104 ppm, benzyl–CH_2 (δ3) protons at 4.249 ppm, and $-C_6H_5$ protons (δ4) at 7.390-7.443. The chemical shifts of various protons of L-Alanine and L-Isoleucine in D_2O are also shown in Pic. 2 and 3 of Fig. 11. In the ^{1}H NMR spectrum of L-Alanine, there are two characteristic peaks corresponding to $–CH_3$ (δ1) protons at 1.167-1.278 ppm and *α*–CH (δ2) proton at 3.500-3.552 ppm and five characteristic peaks are observed for L-Isoleucine corresponding C-5 protons ($-CH_3$) (δ1) at 0.792-0.873, side-chain CH_3 protons (δ2) at 1.099-1.151 ppm, C-4 protons (δ3) at 1.293-1.345 ppm, C-3 proton (δ4) at 1.822-1.856 ppm and C-2 (*α*–CH) proton (δ5) at 3.516-3.526 ppm.



Picture 1: ^{1}H NMR spectrum of pure Benzyltriethylammonium chloride (BTEAC)



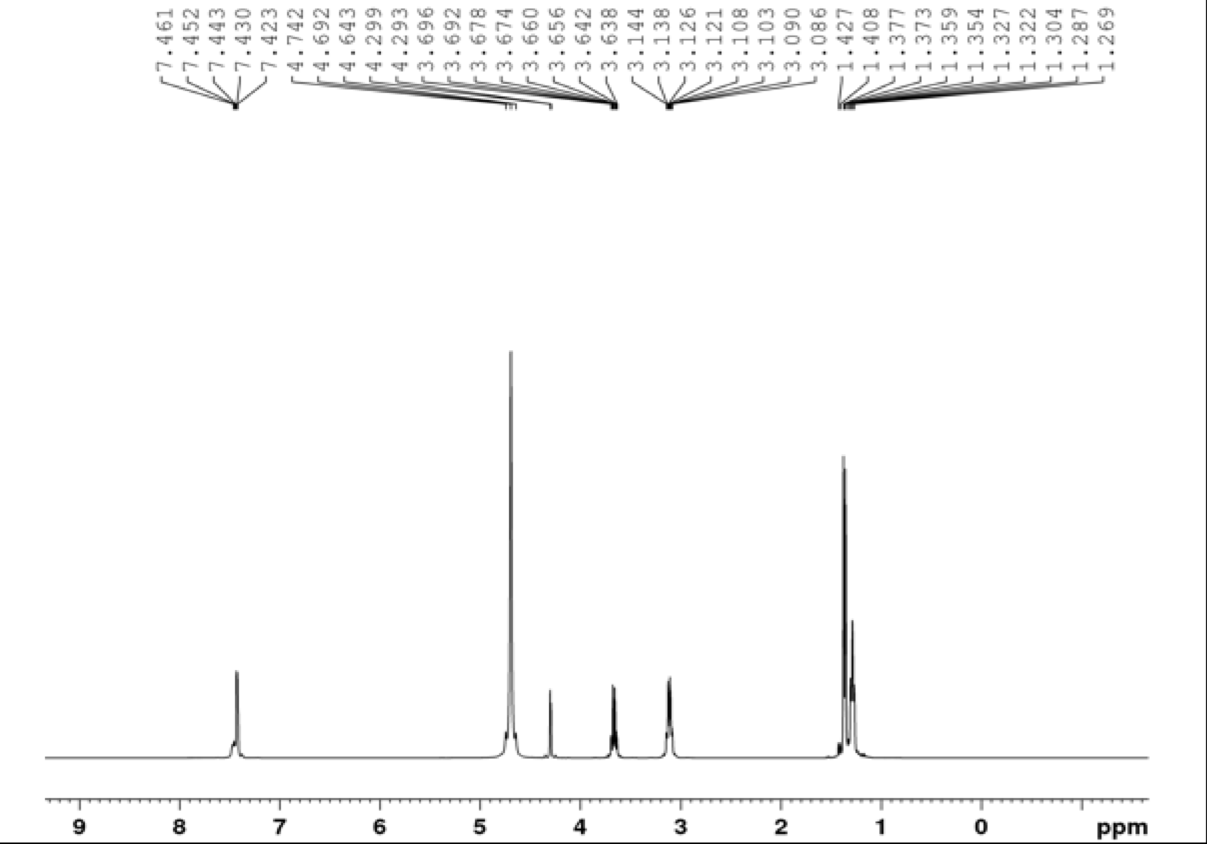
Picture 2: ^{1}H NMR spectrum of pure L-Alanine



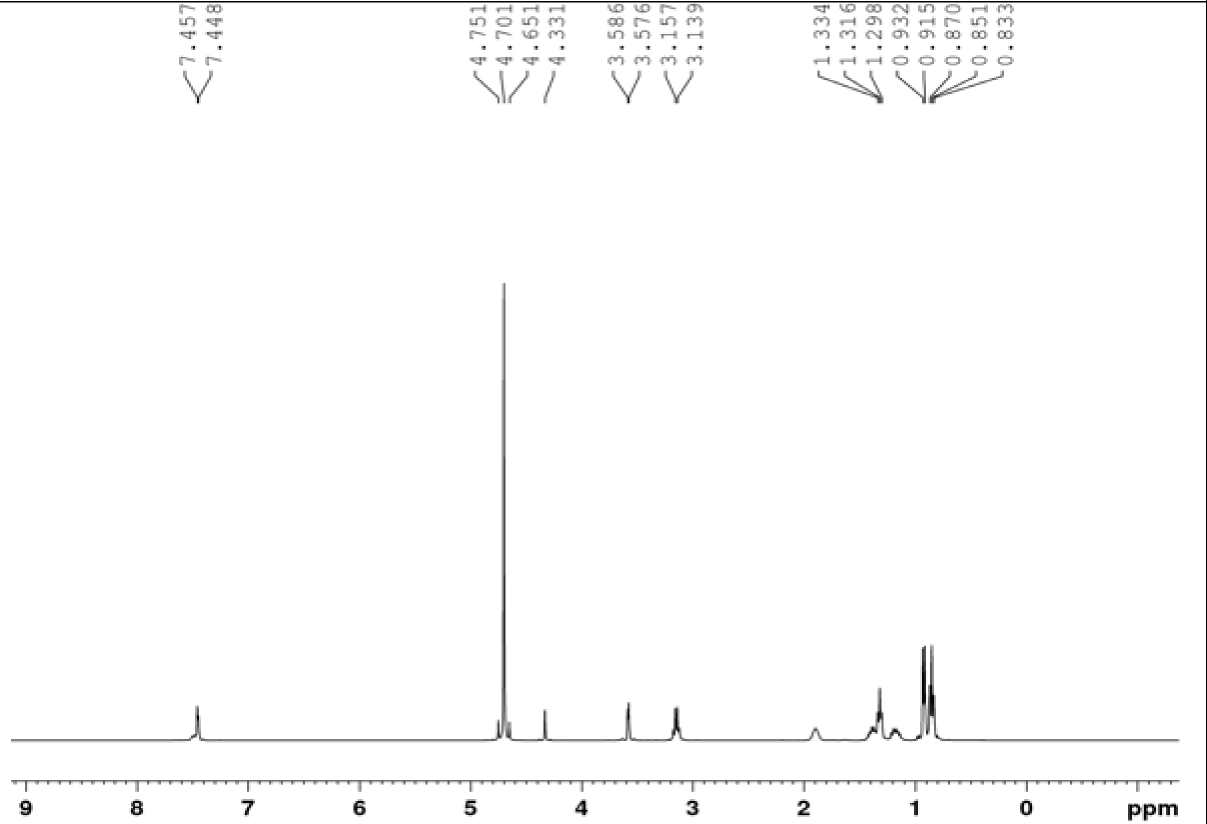
Picture 3:^{1}H NMR spectrum of pure L-Isoleucine

Figure 11: ^{1}H NMR spectra of pure BTEAC (Pic. 1), L-Alanine (Pic. 2) and L-Isoleucine (Pic. 3) in D_2O.

The nature of moieties present in a compound's vicinity used to determine the chemical shift (δ) values of its protons. The change in chemical shifts values of amino acids in presence of BTEAC are shown in Figure 12. The δ values shift downfield (higher frequency) or upfield (lower frequency) occur varying on the deshielding and shielding effects of the surrounding groups or atoms.



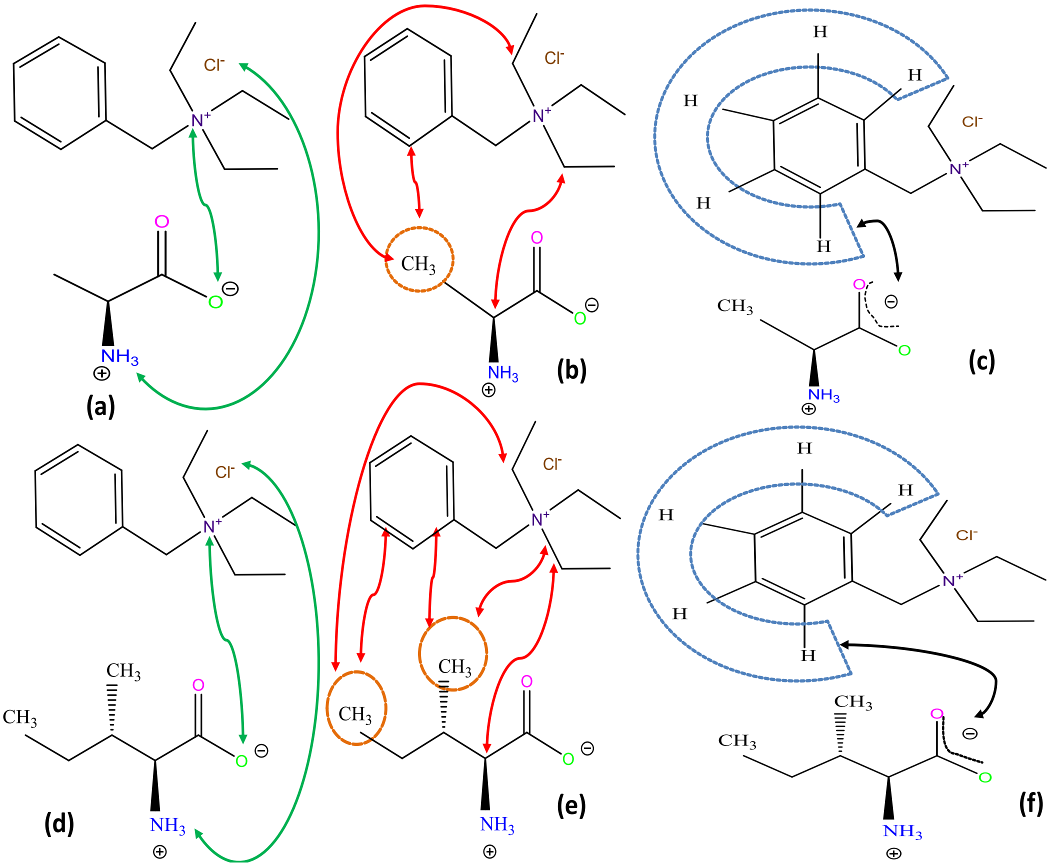
Picture 4: ^{1}H NMR Spectrum of BTEAC + L-Alanine (BTEAC : L-Alanine = 1: 4)



Picture 5: ^{1}H NMR Spectrum of BTEAC + L-Isoleucine (BTEAC : L-Isoleucine = 1: 4)

Figure 12: ^{1}H NMR spectra of BTEAC + LAlanine (Pic. 4) and BTEAC + L-Isoleucine (Pic. 5) in D_2O.

The results show that the chemical shifts of BTEAC protons move to a higher frequency with the addition of amino acids. It is also observed that there are downfield shift (δ) values of protons for both amino acids. A downfield change indicates the decrease in electron density around the protons of hydrophobic groups of BTEAC and amino acids, due to shift of electron density from the vicinity of hydrophobic groups towards positive end of ionic groups, signifying the ion-hydrophobic interactions. It is also observed that compared with protons of the phenyl group, the protons of ethyl groups and benzyl group of BTEAC show a downfield shift with much higher magnitude. On the other hand, the chemical shift of protons in the phenyl group gives the smallest changes. The results suggest that the alkyl chains and benzyl group of BTEAC and the hydrophobic part (terminal-CH_3 for L-Alanine, terminal-CH_3, β-CH_3, β-CH, δ–CH2 for L-Isoleucine) of amino acids are the main contributors for hydrophobic interactions. According to NMR data, the hydrophobic interaction is stronger incase of (L-Isoleucine + BTEAC) system comparison to (L-Alanine + BTEAC) system. All type of interactions are shown in scheme 2.



Scheme 2: Plausible molecular interactions between Ionic Liquid and Amino acids, (a) & (d) Ion-Ion interaction. (⟷) (b) & (e) Hydrophobic-Hydrophobic interaction. (⟷) (c) & (f) Ion-Hydrophobic interaction. (⟷).

3.8 Theoretical study

The ground state optimized geometries of the BTEAC + L-Alanine and BTEAC + L-Isoleucine composites are illustrated in Figure. 13 (a-b). In their ground state geometries strong H-bonding interaction exists for both BTEAC + L-Alanine and BTEAC + L-Isoleucine composites. The much stronger H bonding in BTEAC + LAlanine has been revealed from its shorter H-bonding (2.17-2.25Å) with respect to BTEAC + L-Isoleucine (2.27-2.35Å). Contrastingly the adsorption energy for BTEAC + L-Isoleucine(E_{ads} = -211.33 KJ/mole) is quite high compared to BTEAC + L-Alanine(E_{ads} = -180.51KJ/mole). In BTEAC + L-Isoleucine (2.28 Å -2.50 Å) composite the alkyl-alkyl distance is much smaller than in BTEAC + L-Alanine (2.61 Å). Here we note that strong hydrophobic interaction functioning in BTEAC + L-Isoleucine composites explains quite high adsorption energy involving L-Isoleucine than L-Alanine.

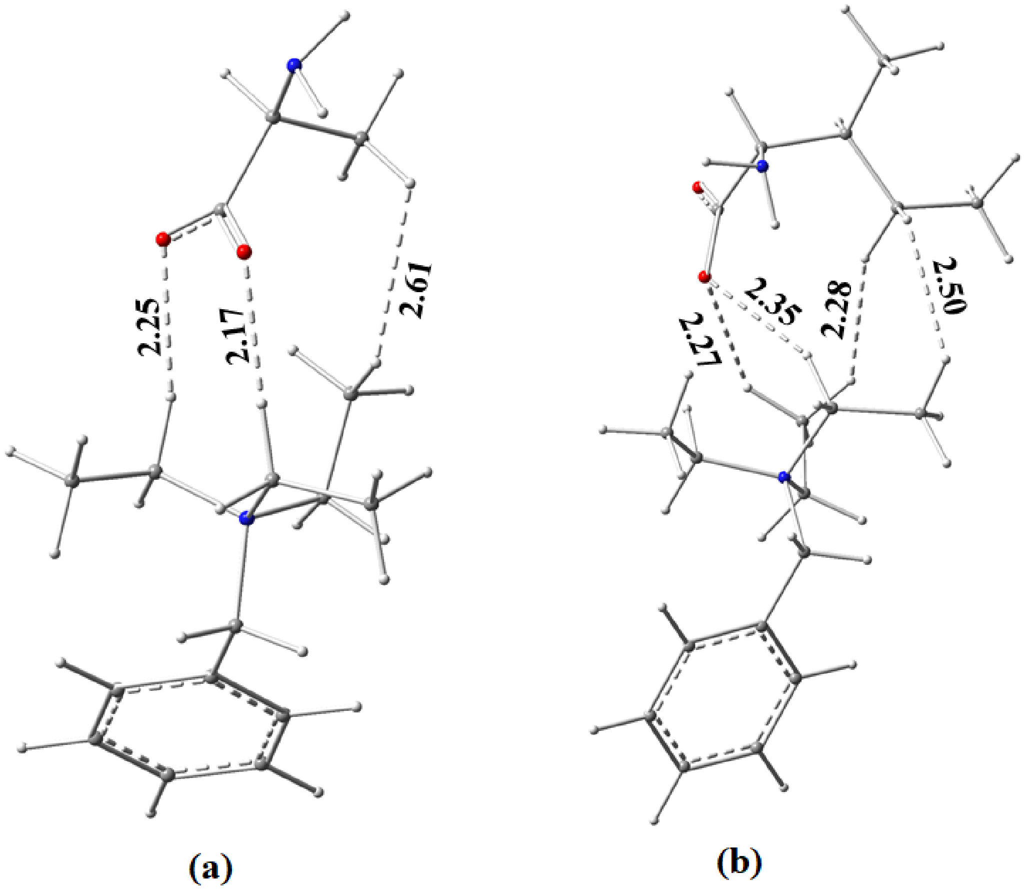


Figure 13: Optimized geometries for the (a) BTEAC + L-Alanine (b) BTEAC + L-Isoleucine.

This additional hydrophobic interaction of alkyl groups of L-Isoleucine and BTEAC moiety also supports the experimental NMR shifting.

Table 9: ΔH° (Kcal); ΔG° (Kcal) and ΔS°(cal/K) for (a) L- Alanine + BTEAC (b) L-Isoleucine+ BTEAC complexes in water medium.

Complex	**L-Alanine+BTEAC**	**L-Isoleucine+BTEAC**
ΔH°(kJ/mol)	-40.7906	-47.7692
ΔG°(kJ/mol)	-31.64028946	-38.3847627
ΔS°(cal/K)	-30.69	-31.475

Different thermodynamic parameters viz. enthalpy change (ΔH°), Gibbs free energy (ΔG°) and entropy contribution (ΔS°) have been analyzed and listed in Table 9. Huge negative ΔH° and ΔG° clearly signify that the mixing of the amino acid and BTEAC is exothermic and spontaneous nature at room temperature and 1 atm pressure. Moreover for L-Isoleucine+BTEAC ΔH° and ΔG° are quite high than L-Alanine+ BTEAC. Here we note that high negative enthalpy change for composites revealed that these interactions is an enthalpy-driven exothermic process. Furthermore, distributions of charges within the composites and electrostatic interactions are analyzed by molecular electrostatic maps as illustrated in the figure 14.Thered region and yellow region of ESP maps demonstrate that stronger electrostatic interactions between amino acids and BTEAC operating in these composites.

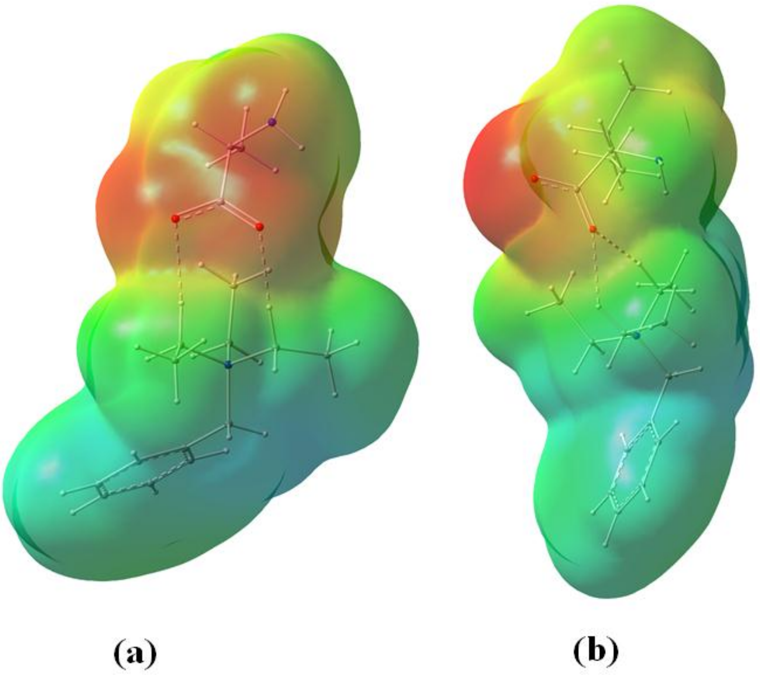
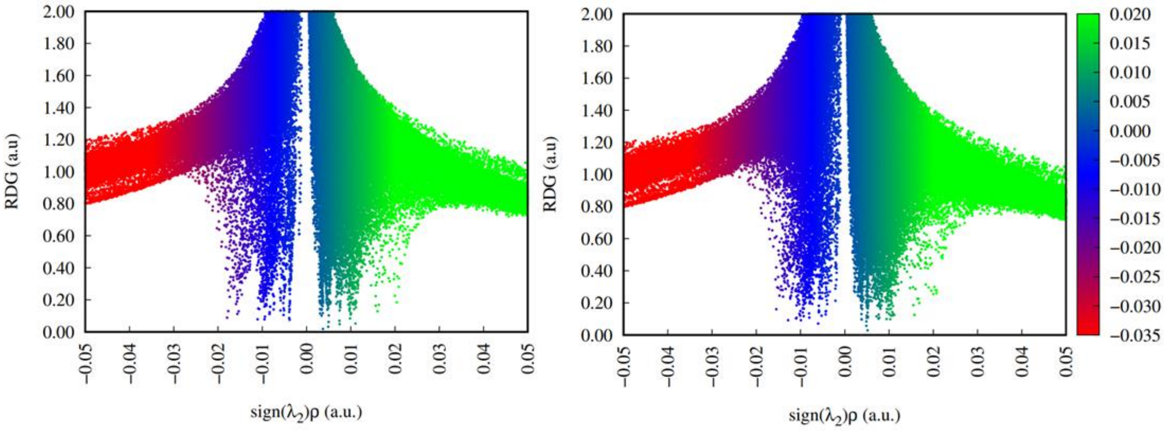

(a) (b)

Figure 14: Electrostatic potential maps for (a) L- Alanine + BTEAC (b) L-Isoleucine+ BTEAC complexes.

To predict the non-covalent interactions like Vander Waals , H-bonding , staric interactions we have analyzed reduced density gradient (RDG) using Non-covalent index (NCI) technique as illustrated in Figure 15.Here the negative values indicate stabilizing interaction while the repulsive interaction are experienced by positive values of sign(λ2)ρ. Systematic analysis of Reduced density gradient (RDG) graphs clearly demonstrates the presence of strong H-bonding (light blue point at -0.01-0.02 region). Furthermore, stabilizing Vander Waals interaction (blue point at -0.00-0.01 region) is more prominent in L-Isoleucine+ BTEAC than L- Alanine + BTEAC.



(a) (b)

Figure 15: Plots of reduced density gradient (RDG) against electron density multiplied by the sign of the second Hessian eigen value (sign (λ2)ρ(r)) for (a) L- Alanine + BTEAC (b) L-Isoleucine+ BTEAC.

4. CONCLUSION

Some physico-chemical methodologies such as density, viscosity, refractive index and conductance study, and surface tension at four different temperatures provides the idea of interaction between the amino acid and ILs in solution. The comprehensive full calculation of different thermodynamic parameters such as apparent molar volume, limiting apparent molar volume, viscosity *B*-coefficient, molar refraction, limiting molar refraction, molar conductivities, and measurement of surface tension helps to identify the solute-solvent interaction depends on the concentration of solute and temperature. The positive value of (d*B*/d*T*) suggests that the structure-breaking (chaotropic) nature of solute-solvent interaction, which is supported by the constant values of Hepler. The increasing trend in the viscosity values is due to the presence of the enhanced frictional resistance between layers. The values of A and B indicate that the solute-solvent interactions are more comparison to the solute-

solute interactions. As a consequence, all the possible combinations of solution chemistry scrutinize the chemistry behind the synergism. R_M^0 values help to determine the interaction taking place between the amino acids and ionic liquid solutions. This makes it easier to pick a mixture of compounds that will generate a synergistic effect in combined state. UV-Visible Spectroscopy helps to determine the molecular association constant between solute and solvent. The ^{1}H NMR spectroscopy showing chemical shifts of BTEAC protons move to higher frequency (downfield) with the addition of both amino acids. The nonpolar alkyl side chains of L-Alanine (methyl group) and L-Isoleucine (*sec*-butyl group) molecules increase the hydrophobic-hydrophobic interactions between the IL and amino acids. The findings indicate that hydrophobic interactions are mainly between the hydrophobic portion (benzyl group and alkyl chains) of BTEAC, and *sec*-butyl group of L-Isoleucine, which is much stronger than that between BTEAC and L-Alanine. Furthermore, employing DFT, evaluation of different parameters like adsorption energy, thermodynamic properties clearly demonstrate that interaction is more prominent with L-Isoleucine+ BTEAC composite than L- Alanine + BTEAC and these observations also corroborate our experimental studies. Overall, in this research, the extent of interaction between amino acids and ILs, will uncover more information about ionic as well as noncovalent interactions, which will help in understanding the potential applications of these ILs and serve as a model system for a better understanding of more complex biological processes and ensures the feasible area in biomedicine chemistry or medicinal industry. Therefore, profitable collaboration achieved in biochemistry, biology, and biotechnology with economic implications.

Declaration of Competing Interest

There are no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper by the authors.

CRediT authorship contribution statement

Baishali Saha: Conceptualization**,** Investigation, Writing – original draft. **Sanjoy Barman:** Data curation, Investigation. **Niloy Roy:** Writing–review & editing. **Biswajit Ghosh:** Visualization. **Rinku Chakrabarty:** Visualization. **Subhankar Choudhury:** Writing – review & editing. **Narendra Nath Ghosh:** Software, Writing – original draft. **Mahendra Nath Roy:** Resources, Supervision.

ACKNOWLEDGMENTS

Prof. M. N. Roy would like to acknowledge UGC, New Delhi, Government of India, for being awarded One Time Grant under Basic Scientific Research via the grant-in-Aid no. F.4-10/2010 (BSR) for providing instrumental facilities according to the research. Baishali Saha is very much grateful to Swami Vivekananda Merit Cum Means Scholarship(SVMCM), reference number WBP211628595537 for financial support in order to continue this research work.

REFERENCE

1. X.Meng, C.Duan, C.Xu, D.Fang, Physicochemical properties and the molar surface Gibbs energy of the aqueous solution of amino acids ionic liquid 1-pentyl-3-methylimidazolium glycine salt [C5mim] [Gly]. Chem.Thermodyn. & Therm. Anal, 2022; 5: 100025.
2. H.Kumar, R.Sharma, Solvation properties of glycine and glycylglycine in aqueous 1-hexyl-3- methylimidazolium bromide solution at different temperatures , J. Chem. Thermodyn, 2021; 152: 106268.
3. R.Tomaš, Z.Kinart, A.Tot, S.Papović, T.T.Borović & M.Vraneš, Volumetric properties, conductivity and computation analysis of selected imidazolium chloride ionic liquids in ethylene glycol, J. Mol. Liq, 2022; *345:* 118178.
4. H.Y.Chen, S.Fang, & L.Wang, Interactions of 1-butyl-2,3-dimethylimidazolium bromide ionic liquid with glycine, L-alanine and L-valine: A volumetric and NMR spectroscopic study,J. Mol. Liq, 2017; *225*: 706-712.
5. J. M.Gomes, S. S.Silva & R. L. Reis, Biocompatible ionic liquids: fundamental behaviours and applications, Chem. Soc. Rev., 2019; *48*: 4317-4335.
6. H.Kumar, R.Sharma & M.Singla, Studies on volumetric and acoustic behfavior of L-alanine and L-leucine in aqueous 1-dodecyl-3-methylimidazolium bromide ionic liquid solutions at different temperatures, J.Mol.Liq, 2021; 342: 117022.
7. R.M.Moshikur, M.R.Chowdhury, M.Moniruzzaman & M.Goto, Biocompatible ionic liquids and their applications in pharmaceutics, Green Chem, 2020; 22: 8116-8139.
8. R.Gaba, A.Pal, D.Sharma, H.Kumar, A.Kumar, Molecular interactions of some non-essential amino acids in aqueous solutions of 1-methylimidazolium chloride at different temperatures, J. Mol. Liq, 2019; 279: 711–718.
9. T.E.Needham Jr, A.N.Paruta, R.J.Gerraughty, Solubility of Amino Acids in Pure Solvent Systems, Journal of Pharmaceutical Sciences, J.Pharm.Sci., 1971; *60*: 565-567.

10. M.Ikeda, S.Takeno, Amino acid production by Corynebacterium glutamicum in Corynebacterium glutamicum, Springer, 2012; 23: 107-147.
11. R.Schweet, H.Lamfrom & E.Allen, The synthesis of hemoglobin in a cell-free system. Proc. Natl. Acad. Sci., 1958; 44: 1029-1035.
12. M.Mondal, S.Basak, B.Rajbanshi, S.Choudhury,N.N.Ghosh,M.N. Roy, Subsistence of diverse interactions of some biologically important molecules in aqueous ionic liquid solutions at various temperatures by experimental and theoretical investigation, J.Mol.Struct, 2022; 1257: 132571.
13. J.Singh, M.Singh, S.Sharma, S.Sharma & M.Sharma, Molecular Interactions of Diphenhydramine-hydrochloride with Some Imidazolium-Based Ionic Liquids in Aqueous Media at T = 293.15−313.15 K: Volumetric, Acoustic, and UV Absorption Studies, ACS Omega, 2021; 6: 22655−22671.
14. S. K.Sharma, G.Singh, H.Kumar & R.Kataria, Study of solvation consequences of glycine, L-alanine and L-valine in aqueous 1-butyl-4-methyl pyridinium chloride ionic liquid solutions probed by physicochemical approach in the temperature interval (288.15-308.15) K, J.Chem. Thermodyn, 2017; 110: 137-153.
15. S.Tunç & O.Duman, Investigation of interactions between some anionic dyes and cationic surfactants by conductometric method, Fluid Ph. Equilibria, 2007; *251*: 1-7.
16. S.E. Friberg, Interactions of Surfactants with Polymers and Proteins, J.Depress.Sci.& Tech, 1994; 15: 1-427.
17. J.E. Lind Jr, J.J. Zwolenik, R.M. Fuoss, Calibration of Conductance Cells at 25° with Aqueous Solutions of Potassium Chloride[1], J.Am.Chem.Soc., 1959; 81: 1557-1559.
18. M. J. Frisch, G. Trucks, H. B. Schlegel, G. Scuseria, M. Robb, J. Cheeseman, G. Scalmani, V. Barone, B. Mennucci and G. Petersson, Gaussian 09, revision A. 1, Gaussian Inc. Wallingford CT, 2009; 27: 34.
19. P.E.Hartnett, C.M.Mauck, M.A.Harris, R.M.Young, Y.L.Wu, T.J.Marks and M.R.Wasielewski, Influence of anion delocalization on electron transfer in a covalent porphyrin donor–perylenediimide dimer acceptor system, J. Am. Chem. Soc., 2017; 139: 749-756.
20. T.Ghosh, S.Mondal, R.Maiti, S.M.Nawaz, N.N.Ghosh, E.Dinda, A.Biswas, S.K.Maity, A.Mallik, D.K Maiti,Complementary amide-based donor–acceptor with unique nano-scale aggregation, fluorescence, and band gap-lowering properties: a WORM memory device. Nanotechnology, 2020; 32: 025208.

21. S. Miertuš, E. Scrocco and J. Tomasi, Electrostatic interaction of a solute with a continuum. A direct utilizaion of AB initio molecular potentials for the prevision of solvent effects, Chem. Phys, 1981; 55: 117-129.
22. M. Cossi, V. Barone, R. Cammi and J. Tomasi, Ab initio study of solvated molecules: a new implementation of the polarizable continuum model, Chem. Phys. Lett., 1996; 255: 327-335.
23. T.Lu, F.Chen, Multiwfn: a multifunctional wavefunction analyzer. J.Comput. Chem, 2012; 33: 580-592.
24. D. Ekka, M.N. Roy, Molecular interactions of α-amino acids insight into aqueous β-cyclodextrin systems, Amino Acids, 2013; 45: 755–777.
25. D.O. Masson, Solute molecular volumes in relation to solvation and ionization, Philos. Mag. J. Sci., 1929; 8: 218–235.

a. Pal, B. Kumar, Volumetric and acoustic properties of binary mixtures of the ionic liquid 1-butyl-3-methylimidazolium tetrafluoroborate [bmim][BF4] with alkoxyalkanols at different temperatures, J. Chem. Eng. Data, 2012; 57: 688–695.

26. M.T. Zafarani-Moattar, B. Asadzadeh, A. Shahrisa, M.G. Nazari, Study of thermodynamic properties of l-serine in aqueous 1-carboxymethyl-3-methylimidazolium chloride solutions at 298.15 K, Fluid Phase Equilib, 2014; 363: 32–40.
27. L.G. Hepler, Thermal expansion and structure in water and aqueous solutions, Can. J. Chem, 1969; 47: 4613–4617.
28. M.N. Roy, V.K. Dakua, B. Sinha, Partial molar volumes, viscosity B-coefficients, and adiabatic compressibilities of sodium molybdate in aqueous 1, 3-dioxolane mixtures from 303.15 to 323.15 K, Int. J. Thermophys, 2007; 28: 1275–1284.
29. G. Jones, M. Dole, THE VISCOSITY OF AQUEOUS SOLUTIONS OF STRONG ELECTROLYTES WITH SPECIAL REFERENCE TO BARIUM CHLORIDE, J. Am. Chem. Soc, 1929; 51: 2950–2964.
30. Q. Liu, L. Ma, K. Li, J. Wang, Y. Zang, Apparent molar volumes of hydrophobic imidazolium type ionic liquids with dimethyl carbonate, J. Mol. Liq, 2020; 309: 113010.
31. F.J. Millero, Molal volumes of electrolytes, Chem. Rev., 1971; 71: 147–176.
32. Y. Marcus, ViscosityB-coefficients, structural entropies and heat capacities, and the effects of ions on the structure of water, J. Solut. Chem, 1994; 23: 831–848.
33. H.D.B. Jenkins, Y. Marcus, Viscosity B-coefficients of ions in solution, Chem. Rev., 1995; 95: 2695–2724.

34. H. Friedman, C. Krishnan, in: Thermodynamics of Ionic Hydration, Aqueous Solutions of Simple Electrolytes, Springer, 1973; 351: 1–118.
35. H.J.V.Tyrrell, M. Kennerley, Viscosity B-coefficients between 5° and 20° for glycolamide, glycine, and N-methylated glycines in aqueous solution, J. Chem. Soc. A, 1968; 2724–2728.
36. R.H. Stokes, R. Mills, Viscosity of Electrolytes and Related Properties, Pergamon Press, 1965.
37. J.M. Tsangaris, R.B. Martin, Viscosities of aqueous solutions of dipolar ions, Arch. Biochem. Biophys, 1965; 112: 267–272.
38. T. Sarma, J. Ahluwalia, Experimental studies on the structures of aqueous solutions of hydrophobic solutes, Chem. Soc. Rev., 1973; 2: 203–232.
39. W. Devine, B. Lowe, Viscosity B-coefficients at 15 and 25° C for glycine, β-alanine, 4-amino-n-butyric acid, and 6-amino-n-hexanoic acid in aqueous solution, J. Chem. Soc. A, 1971; 2113–2116.
40. S. Glasstone, K.J. Laidler, H. Eyring, The Theory of Rate Processes, McGraw-hill, 1941.
41. D. Feakins, F.M. Bates, W.E. Waghorne, K.G. Lawrence, Relative viscosities and quasi-thermodynamics of solutions of tert-butyl alcohol in the methanol–water system: a different view of the alkyl–water interaction, J. Chem. Soc. Faraday Trans, 1993; 89: 3381–3388.
42. D. Feakins, D.J. Freemantle, K.G. Lawrence, Journal of the Chemical Society, Faraday Transactions 1: Physical Chemistry in Condensed Phases, 1974; 70: 795-806.

a. Ali, S. Hyder, S. Sabir, D. Chand, A.K. Nain, Volumetric, viscometric, and refractive index behaviour of α-amino acids and their groups' contribution in aqueous d-glucose solution at different temperatures, J. Chem. Thermodyn, 2006; 38: 136–143.

43. H.L. Friedman, C. Krishnan, Studies of hydrophobic bonding in aqueous alcohols: enthalpy measurements and model calculations, J. Solut. Chem, 1973; 2: 119–140.
44. U. Dash, B. Samantaray, S. Mishra, Volumetric and viscometric studies of sodium nitroprusside in aqueous solutions at different temperatures, J. Teach. Res. Chem, 2005; 11: 87–90.
45. V.I. Minkin, Dipole Moments in Organic Chemistry, Springer Science & Business Media, 2012.
46. M. Born, E. Wolf, Principles of Optics: Electromagnetic Theory of Propagation, Interference and Diffraction of Light, Elsevier, 2013.

47. M. Deetlefs, K.R. Seddon, M. Shara, Predicting physical properties of ionic liquids, Phys. Chem. Chem. Phys, 2006; 8: 642–649.
48. D. Ekka, M.N. Roy, Conductance, a contrivance to explore ion association and solvation behavior of an ionic liquid (tetrabutylphosphonium tetrafluoroborate) in acetonitrile, tetrahydrofuran, 1, 3-dioxolane, and their binaries, J. Phys. Chem. B, 2012; 116: 11687–11694.
49. D. Ekka, T. Ray, K. Roy, M.N. Roy, Exploration of Solvation Consequence of Ionic Liquid [Bu4PCH3SO3] in Various Solvent Systems by Conductance and FTIR Study, J. Chem. Eng. Data, 2016; 61: 2187–2196.
50. X.M. Shen, F. Zhang, G. Dryhurst, Oxidation of dopamine in the presence of cysteine: characterization of new toxic products, Chem. Res. Toxicol, 1997; 10: 147–155.
51. C.M. Romero, E. Jiménez, F. Suárez, Effect of temperature on the behavior of surface properties of alcohols in aqueous solution, J. Chem. Thermodyn, 2009; 41: 513–516.
52. C.M. Romero, M.S. Paéz, Surface tension of aqueous solutions of alcohol and polyols at 298.15 K, Phys. Chem. Liq, 2006; 44: 61–65.
53. M. Mondal, S. Basak, S. Choudhury, N.N. Ghosh, M.N. Roy, Journal of Molecular Liquids, 2021; 116933.
54. R.I. Slavchov, J.K. Novev, Surface tension of concentrated electrolyte solutions, J. Colloid Interface Sci., 2012; 387: 234–243.
55. J.V. Caso, L. Russo, M. Palmieri, G. Malgieri, S. Galdiero, A. Falanga, C. Isernia, R. Iacovino, Investigating the inclusion properties of aromatic amino acids complexing beta-cyclodextrins in model peptides,, Amino Acids, 2015; 47: 2215–2227.
56. H.A. Benesi, J.H.J. Hildebrand, A spectrophotometric investigation of the interaction of iodine with aromatic hydrocarbons,, J. Am. Chem. Soc., 1949; 71: 2703–2707.
57. B. Rajbanshi, S. Saha, K. Das, B.K. Barman, S. Sengupta, A. Bhattacharjee, M.N. Roy, Study to probe subsistence of host-guest inclusion complexes of α and β- cyclodextrins with biologically potent drugs for safety regulatory dischargement, Sci. Rep., 2018; 8: 1–20.
58. M.N. Roy, S. Saha, S. Barman, D. Ekka, Host–guest inclusion complexes of RNA nucleosides inside aqueous cyclodextrins explored by physicochemical and spectroscopic methods,, RSC Adv., 2016; 6: 8881–8891.
59. S. Kumar, S. Sagar, M. Gupta, A thermodynamic and 1H NMR spectroscopy study of binary mixtures of polyethylene glycol butyl ether (PEGBE) 206 with 1-Butanol and 2-(Methylamino) ethanol (MAE), J. Mol. Liq, 2016; 214: 306–312.

Highlights

- Investigated systems are (L-Alanine + aq. solution of BTEAC) and (L-Isoleucine + aq. solution of BTEAC).
- The limiting molar apparent volume, viscosity B-coefficient, limiting molar refraction, molar conductance, surface tension are determined.
- Ion-Ion, Hydrophobic-Hydrophobic, Ion-Hydrophobic interactions have been studied.
- UV-Visible spectroscopy, ^{1}H-NMR and theoretical studies have also been done.
- The interaction is higher incase of (L- Isoleucine + BTEAC + H2O) comparison to (L-Alanine +BTEAC+H2O) solution.

Supporting Information

Table S1. Density (ρ), Viscosity (η), Refractive index (n_D) of aqueous pure BTEAC solutions of concentrations(molality) 0.001m, 0.003m, 0.005m at temperatures 298.15 K, 303.15 K 308.15 K and 313.15 K.

BTEAC + H_2O				
Conc.of aq. IL(m^a)	**Temperature (K^b)**	**Density (ρ^c) × 10^{-3}kg.m^{-3}**	**Viscosity (η^e) mPa.s**	**Refractive index ($n_D{}^d$)**
0.001	298.15	0.99711	0.970	1.3318
	303.15	0.99572	0.874	1.3313
	308.15	0.99411	0.835	1.3308
	313.15	0.99228	0.772	1.3305
0.003	298.15	0.99715	0.978	1.3321
	303.15	0.99576	0.903	1.3314
	308.15	0.99415	0.834	1.3309
	313.15	0.99232	0.779	1.3306
0.005	298.15	0.99720	0.984	1.3323
	303.15	0.99579	0.917	1.3315
	308.15	0.99418	0.849	1.3305
	313.15	0.99236	0.786	1.3307

[a] molality (m) of aqueous BTEAC solution. Uncertainty in molality u(m) = ± 0.0001 mol kg^{-1}; ;[b]Standard uncertainties in temperature u(T) = ± 0.01 K; [c]Standard uncertainties in density u(ρ) = ± 0.00001 kg m^{-3}; [d]Standard uncertainties in refractive indices u(n_D) = ± 0.0002; [e]Standard uncertainties in Viscosity u(η) = ± 0.003mPa.s

Table S2. Density (*ρ*), Viscosity (*η*), Refractive index (n_D) of aqueous pure (L-ALANINE + BTEAC + H_2O) and (L-ISOLEUCINE + BTEAC + H_2O) systems in aqueous BTEAC solutions of concentration(molality), 0.001m at temperatures 298.15 K, 303.15 K 308.15 K and 313.15 K

(L-ALANINE + BTEAC + H_2O) system, 0.001m				(L-ISOLEUCINE+ BTEAC + H_2O) system,0.001m		
Temperature = 298.15 K				Temperature = 298.15 K		
Molality (mol/kg)	Density (*ρ*) $\times 10^{-3}$ kg.m^{-3}	Viscosity (*η*) mPa.s	Refractive Index (n_D)	Density (*ρ*) $\times 10^{-3}$ kg.m^{-3}	Viscosity (*η*) mPa.s	Refractive Index (n_D)
0.0100	0.99739	0.980	1.3320	0.99736	0.984	1.3321
0.0251	0.99782	0.988	1.3322	0.99775	0.998	1.3323
0.0402	0.99826	0.995	1.3324	0.99815	1.011	1.3325
0.0554	0.99870	1.002	1.3326	0.99856	1.024	1.3327
0.0705	0.99915	1.008	1.3328	0.99897	1.037	1.3329
0.0858	0.99960	1.014	1.3330	0.99939	1.049	1.3331
Temperature = 303.15 K				Temperature = 303.15 K		
0.0100	0.99599	0.874	1.3318	0.99596	0.887	1.3320
0.0251	0.99641	0.884	1.3320	0.99634	0.900	1.3322
0.0402	0.99684	0.893	1.3322	0.99674	0.913	1.3324
0.0554	0.99728	0.901	1.3324	0.99715	0.925	1.3326
0.0705	0.99773	0.908	1.3326	0.99757	0.937	1.3328
0.0858	0.99818	0.915	1.3328	0.99800	0.950	1.3330
Temperature = 308.15 K				Temperature = 308.15 K		
0.0100	0.99437	0.845	1.3316	0.99434	0.848	1.3318
0.0251	0.99478	0.854	1.3318	0.99471	0.861	1.3320
0.0402	0.99521	0.863	1.3320	0.99510	0.874	1.3322
0.0554	0.99565	0.871	1.3322	0.99550	0.887	1.3324
0.0705	0.99610	0.879	1.3324	0.99592	0.900	1.3326
0.0858	0.99656	0.887	1.3326	0.99634	0.913	1.3328
Temperature = 313.15 K				Temperature = 313.15 K		
0.0100	0.99253	0.782	1.3314	0.99250	0.785	1.3315
0.0251	0.99293	0.791	1.3316	0.99286	0.799	1.3317
0.0402	0.99335	0.800	1.3318	0.99324	0.812	1.3319
0.0554	0.99379	0.809	1.3320	0.99364	0.826	1.3321
0.0705	0.99423	0.817	1.3322	0.99406	0.839	1.3323
0.0858	0.99469	0.826	1.3324	0.99448	0.851	1.3325

[a] molality (*m*) of aqueous BTEAC solution. Uncertainty in molality u(*m*) = ± 0.0001 mol kg^{-1}; ;[b]Standard uncertainties in temperature u(T) = ± 0.01 K; [c]Standard uncertainties in density u(*ρ*) = ± 0.00001 kg m^{-3}; [d]Standard uncertainties in refractive indices u(n_D) = ± 0.0002; [e]Standard uncertainties in Viscosity u(*η*) = ± 0.003 mPa.s

Table S3. Density (*ρ*), Viscosity (*η*), Refractive index (n_D) of aqueous pure (L-ALANINE + BTEAC + H_2O) and (L-ISOLEUCINE + BTEAC + H_2O) systems in aqueous BTEAC solutions of concentration(molality), 0.003m at temperatures 298.15 K, 303.15 K,308.15 K and 313.15 K

(L-ALANINE + BTEAC + H_2O) system, 0.003m				**(L- ISOLEUCINE + BTEAC + H_2O) system, 0.003m**		
Temperature = 298.15 K				**Temperature = 298.15 K**		
Molality (mol/kg)	**Density (*ρ*) ×10^{-3} kg.m^{-3}**	**Viscosity (*η*) mPa.s**	**Refractive Index (n_D)**	**Density (*ρ*) ×10^{-3} kg.m^{-3}**	**Viscosity (*η*) mPa.s**	**Refractive Index (n_D)**
0.0100	0.99742	0.989	1.3322	0.99739	0.993	1.3322
0.0251	0.99784	0.999	1.3324	0.99777	1.008	1.3324
0.0402	0.99827	1.006	1.3326	0.99817	1.023	1.3326
0.0554	0.99871	1.014	1.3328	0.99857	1.038	1.3328
0.0705	0.99916	1.022	1.3330	0.99899	1.052	1.3330
0.0858	0.99961	1.029	1.3332	0.99941	1.067	1.3332
Temperature = 303.15 K				Temperature = 303.15 K		
0.0100	0.99602	0.914	1.3320	0.99599	0.917	1.3321
0.0251	0.99643	0.924	1.3322	0.99636	0.932	1.3323
0.0402	0.99686	0.933	1.3324	0.99676	0.945	1.3325
0.0554	0.99730	0.941	1.3326	0.99717	0.959	1.3327
0.0705	0.99775	0.949	1.3328	0.99759	0.974	1.3329
0.0858	0.99821	0.957	1.3330	0.99803	0.988	1.3331
Temperature = 308.15 K				Temperature = 308.15 K		
0.0100	0.99440	0.844	1.3318	0.99437	0.847	1.3319
0.0251	0.99480	0.855	1.3320	0.99473	0.863	1.3321
0.0402	0.99523	0.865	1.3322	0.99512	0.877	1.3323
0.0554	0.99567	0.874	1.3324	0.99553	0.892	1.3325
0.0705	0.99612	0.883	1.3326	0.99595	0.906	1.3327
0.0858	0.99659	0.891	1.3328	0.99638	0.919	1.3329
Temperature = 313.15 K				Temperature = 313.15 K		
0.0100	0.99256	0.722	1.3316	0.99253	0.792	1.3317
0.0251	0.99295	0.730	1.3318	0.99288	0.807	1.3319
0.0402	0.99337	0.737	1.3320	0.99326	0.822	1.3321
0.0554	0.99381	0.744	1.3322	0.99367	0.836	1.3323
0.0705	0.99426	0.749	1.3324	0.99408	0.850	1.3325
0.0858	0.99472	0.754	1.3326	0.99451	0.864	1.3327

[a] molality (*m*) of aqueous BTEACl solution. Uncertainty in molality u(*m*) = ± 0.0001 mol kg^{-1}; ;[b]Standard uncertainties in temperature u(T) = ± 0.01 K; [c]Standard uncertainties in density u(*ρ*) = ± 0.00001 kg m^{-3}; [d]Standard uncertainties in refractive indices u(n_D) = ± 0.0002; [e]Standard uncertainties in Viscosity u(*η*) = ± 0.003 mPa.s

Table S4. Density (*ρ*), Viscosity (*η*), Refractive index (n_D) of aqueous pure (L-ALANINE + BTEAC + H_2O) and (L-ISOLEUCINE + BTEAC + H_2O) systems in aqueous BTEAC solutions of concentration(molality), 0.005m at temperatures 298.15 K, 303.15 K 308.15 K and 313.15 K

(L-ALANINE + BTEAC + H_2O) system, 0.005m				**(L- ISOLEUCINE + BTEAC + H_2O) system, 0.005m**		
Temperature = 298.15 K				**Temperature = 298.15 K**		
Molality(mol/kg)	**Density (*ρ*) $\times 10^{-3}$ $kg.m^{-3}$**	**Viscosity (*η*) mPa.s**	**Refractive Index (n_D)**	**Density (*ρ*) $\times 10^{-3}$ $kg.m^{-3}$**	**Viscosity (*η*) mPa.s**	**Refractive Index (n_D)**
0.0100	0.99746	0.995	1.3324	0.99743	0.999	1.3324
0.0251	0.99787	1.005	1.3326	0.99780	1.017	1.3326
0.0402	0.99830	1.014	1.3328	0.99819	1.034	1.3328
0.0554	0.99873	1.022	1.3330	0.99859	1.050	1.3330
0.0705	0.99919	1.030	1.3332	0.99901	1.064	1.3332
0.0858	0.99964	1.038	1.3334	0.99943	1.081	1.3334
Temperature = 303.15 K				Temperature = 303.15 K		
0.0100	0.99604	0.928	1.3322	0.99601	0.932	1.3322
0.0251	0.99644	0.939	1.3324	0.99638	0.948	1.3324
0.0402	0.99687	0.950	1.3326	0.99677	0.964	1.3326
0.0554	0.99731	0.960	1.3328	0.99718	0.980	1.3328
0.0705	0.99777	0.970	1.3330	0.99761	0.995	1.3330
0.0858	0.99823	0.979	1.3332	0.99805	1.011	1.3332
Temperature = 308.15 K				Temperature = 308.15 K		
0.0100	0.99442	0.860	1.3320	0.99439	0.864	1.3321
0.0251	0.99482	0.871	1.3322	0.99474	0.880	1.3323
0.0402	0.99524	0.881	1.3324	0.99513	0.896	1.3325
0.0554	0.99568	0.892	1.3326	0.99554	0.912	1.3327
0.0705	0.99614	0.901	1.3328	0.99597	0.928	1.3329
0.0858	0.99661	0.911	1.3330	0.99640	0.943	1.3331
Temperature = 313.15 K				Temperature = 313.15 K		
0.0100	0.99259	0.797	1.3318	0.99256	0.800	1.3318
0.0251	0.99298	0.809	1.3320	0.99291	0.816	1.3320
0.0402	0.99339	0.820	1.3322	0.99328	0.833	1.3322
0.0554	0.99383	0.831	1.3324	0.99369	0.848	1.3324
0.0705	0.99428	0.842	1.3326	0.99410	0.863	1.3326
0.0858	0.99475	0.852	1.3328	0.99454	0.878	1.3328

[a] molality (*m*) of aqueous BTEAC solution. Uncertainty in molality u(*m*) = ± 0.0001 mol kg^{-1}; ;[b]Standard uncertainties in temperature u(T) = ± 0.01 K; [c]Standard uncertainties in density u(*ρ*) = ± 0.00001 kg m^{-3}; [d]Standard uncertainties in refractive indices u(n_D) = ± 0.0002; [e]Standard uncertainties in Viscosity u(*η*) = ± 0.003 mPa.s

Table S5. Apparent molar volume (ϕ_V), (η_r-1)/√c and Molar Refraction (R_M)of aqueous pure (L-ALANINE + BTEAC + H_2O) and (L-ISOLEUCINE + BTEAC + H_2O) systems in aqueous BTEAC solutions of concentration(molality), 0.001m at temperatures 298.15 K, 303.15 K, 308.15 K and 313.15 K.

(L-ALANINE + BTEAC + H_2O) system, 0.001m				**(L- ISOLEUCINE + BTEAC + H_2O) system, 0.001m**		
Temperature = 298.15 K				**Temperature = 298.15 K**		
Molality (mol/kg)	**ϕ_V× 10^6 (m^3 mol^{-1})**	**(η_r-1)/√c $dm^{3/2}$ $mol^{-1/2}$**	**R_M (m^3 mol^{-1})**	**ϕ_V× 10^6 (m^3 mol^{-1})**	**(η_r-1)/√c $dm^{3/2}$ $mol^{-1/2}$**	**R_M (m^3 mol^{-1})**
0.0100	61.2423	0.103	18.3233	106.4440	0.144	26.9861
0.0251	60.8030	0.120	18.3254	105.7904	0.182	26.9903
0.0402	60.4131	0.130	18.3273	105.3367	0.210	26.9942
0.0554	60.2147	0.141	18.3293	104.9187	0.236	26.9979
0.0705	59.9405	0.149	18.3310	104.6573	0.260	27.0015
0.0858	59.7491	0.154	18.3327	104.3505	0.277	27.0049
Temperature = 303.15 K				Temperature = 303.15 K		
0.0100	62.2945	0.112	18.3390	107.5643	0.149	27.0167
0.0251	61.6538	0.137	18.3413	106.7089	0.187	27.0211
0.0402	61.2140	0.153	18.3434	105.9521	0.221	27.0250
0.0554	60.8104	0.166	18.3453	105.3958	0.249	27.0287
0.0705	60.4197	0.177	18.3471	104.9106	0.272	27.0321
0.0858	60.1535	0.188	18.3488	104.4583	0.296	27.0351
Temperature = 308.15 K				Temperature = 308.15 K		
0.0100	63.3609	0.120	18.3588	108.7094	0.155	27.0459
0.0251	62.5170	0.142	18.3613	107.6505	0.196	27.0506
0.0402	61.7730	0.166	18.3634	106.8419	0.233	27.0548
0.0554	61.2306	0.182	18.3653	106.2619	0.264	27.0587
0.0705	60.7600	0.197	18.3671	105.6179	0.293	27.0621
0.0858	60.3228	0.211	18.3686	105.1820	0.318	27.0654
Temperature = 313.15 K				Temperature = 313.15 K		
0.0100	64.4425	0.128	18.3828	109.8808	0.162	27.0738
0.0251	63.3937	0.155	18.3854	108.6166	0.219	27.0788
0.0402	62.5969	0.181	18.3877	107.7555	0.258	27.0832
0.0554	61.8453	0.204	18.3896	106.9568	0.297	27.0871
0.0705	61.3999	0.220	18.3916	106.2013	0.326	27.0905
0.0858	60.8589	0.239	18.3931	105.6873	0.349	27.0939

Table S6. Apparent molar volume (ϕ_V), $(\eta_r-1)/\sqrt{c}$ and Molar Refraction (R_M)of aqueous pure (L-ALANINE + BTEAC + H_2O) and (L-ISOLEUCINE + BTEAC + H_2O) systems in aqueous BTEAC solutions of concentration(molality), 0.003m at temperatures 298.15 K, 303.15 K, 308.15 K and 313.15 K.

(L-ALANINE + BTEAC + H_2O) system, 0.003m				(L-ISOLEUCINE + BTEAC + H_2O) system, 0.003m		
Temperature = 298.15 K				**Temperature = 298.15 K**		
Molality (mol/kg)	**$\phi_V \times 10^6$ ($m^3\ mol^{-1}$)**	**$(\eta_r-1)/\sqrt{c}$ $dm^{3/2}\ mol^{-1/2}$**	**R_M ($m^3\ mol^{-1}$)**	**$\phi_V \times 10^6$ ($m^3\ mol^{-1}$)**	**$(\eta_r-1)/\sqrt{c}$ $dm^{3/2}\ mol^{-1/2}$**	**R_M ($m^3\ mol^{-1}$)**
0.0100	62.2441	0.110	18.3327	107.4447	0.153	26.9927
0.0251	61.6052	0.132	18.3350	106.5918	0.194	26.9971
0.0402	61.1666	0.143	18.3372	105.8371	0.229	27.0011
0.0554	60.7642	0.156	18.3391	105.4657	0.260	27.0050
0.0705	60.3746	0.168	18.3408	104.9428	0.284	27.0084
0.0858	60.1091	0.176	18.3426	104.5855	0.310	27.0117
Temperature = 303.15 K				Temperature = 303.15 K		
0.0100	63.2994	0.117	18.3485	108.5677	0.155	27.0232
0.0251	62.4582	0.144	18.3510	107.5102	0.203	27.0280
0.0402	61.7167	0.165	18.3531	106.4539	0.232	27.0319
0.0554	61.1760	0.180	18.3550	105.7605	0.263	27.0355
0.0705	60.7069	0.193	18.3567	105.1969	0.295	27.0389
0.0858	60.2712	0.205	18.3583	104.5745	0.321	27.0417
Temperature = 308.15 K				Temperature = 308.15 K		
0.0100	64.3690	0.124	18.3683	109.7834	0.160	27.0525
0.0251	63.3240	0.159	18.3710	108.5290	0.219	27.0575
0.0402	62.2773	0.185	18.3731	107.4215	0.257	27.0616
0.0554	61.5973	0.205	18.3750	106.5218	0.295	27.0653
0.0705	61.0481	0.221	18.3767	105.8396	0.324	27.0686
0.0858	60.4408	0.233	18.3781	105.2591	0.347	27.0717
Temperature = 313.15 K				Temperature = 313.15 K		
0.0100	65.4544	0.132	18.3923	110.8911	0.167	27.0878
0.0251	64.2036	0.170	18.3951	109.4257	0.227	27.0931
0.0402	63.1031	0.196	18.3974	108.2607	0.275	27.0975
0.0554	62.2134	0.222	18.3993	107.1481	0.310	27.1011
0.0705	61.5438	0.240	18.4011	106.4896	0.342	27.1048
0.0858	60.9773	0.262	18.4026	105.8043	0.372	27.1078

Table S7. Apparent molar volume (ϕ_V), (η_r-1)/√c and Molar Refraction (R_M) of aqueous pure (L-ALANINE + BTEAC + H_2O) and (L-ISOLEUCINE + BTEAC + H_2O) systems in aqueous BTEAC solutions of concentration(molality),0.005m at temperatures 298.15 K, 303.15 K, 308.15 K and 313.15 K.

(L-ALANINE + BTEAC + H_2O) system, 0.005m				**(L-ISOLEUCINE + BTEAC + H_2O) system,0.005m**		
Temperature = 298.15 K				**Temperature = 298.15 K**		
Molality (mol/kg)	**ϕ_V× 10^6 (m^3 mol^{-1})**	**(η_r-1)/√c $dm^{3/2}$ $mol^{-1/2}$**	**R_M (m^3 mol^{-1})**	**ϕ_V× 10^6 (m^3 mol^{-1})**	**(η_r-1)/√c $dm^{3/2}$ $mol^{-1/2}$**	**R_M (m^3 mol^{-1})**
0.0100	63.2457	0.113	18.3420	108.4443	0.156	27.0063
0.0251	62.4069	0.134	18.3445	107.3920	0.212	27.0111
0.0402	61.6675	0.153	18.3466	106.5884	0.253	27.0153
0.0554	61.3113	0.166	18.3487	106.0119	0.285	27.0192
0.0705	60.6607	0.176	18.3503	105.3718	0.305	27.0225
0.0858	60.3447	0.188	18.3520	104.9226	0.336	27.0259
Temperature = 303.15 K				Temperature = 303.15 K		
0.0100	64.3045	0.124	18.3581	109.5718	0.163	27.0301
0.0251	63.2629	0.154	18.3608	107.9131	0.213	27.0348
0.0402	62.2197	0.180	18.3629	106.9564	0.255	27.0390
0.0554	61.5419	0.199	18.3648	106.1163	0.292	27.0426
0.0705	60.8502	0.217	18.3664	105.3393	0.320	27.0457
0.0858	60.3891	0.230	18.3679	104.6915	0.348	27.0485
Temperature = 308.15 K				Temperature = 308.15 K		
0.0100	65.3773	0.130	18.3780	110.7234	0.171	27.0667
0.0251	63.7270	0.163	18.3807	109.2633	0.230	27.0720
0.0402	62.7820	0.187	18.3829	107.8494	0.276	27.0762
0.0554	61.9644	0.214	18.3849	106.8004	0.315	27.0798
0.0705	61.1919	0.232	18.3864	105.9026	0.350	27.0829
0.0858	60.5591	0.249	18.3877	105.2963	0.377	27.0859
Temperature = 313.15 K				Temperature = 313.15 K		
0.0100	66.4659	0.140	18.4018	111.9012	0.173	27.0944
0.0251	64.6077	0.185	18.4046	109.8282	0.242	27.0996
0.0402	63.6092	0.215	18.4071	108.7658	0.298	27.1044
0.0554	62.5815	0.243	18.4090	107.5152	0.335	27.1080
0.0705	61.8330	0.267	18.4107	106.7778	0.368	27.1116
0.0858	61.0958	0.288	18.4121	105.9213	0.399	27.1144

Table S8. Molar conductivities of (L-ALANINE + BTEAC + H_2O) and (L-ISOLEUCINE + BTEAC + H_2O) systems in aqueous BTEAC solutions of concentration(molality), 0.001m, 0.003m, 0.005m at temperatures 298.15 K, 303.15 K, 308.15 K and 313.15 K.

Molar conductivities (mS $cm^2 mol^{-1}$)				
(**L-ALANINE + BTEAC + H_2O**) system,0.001m				
Molarity (moles/litre)	298.15 K	303.15 K	308.15 K	313.15 K
0.0100	15880	18250	20230	22550
0.0251	7536	8644	9620	10732
0.0402	5100	6020	6792.5	7825
0.0554	4025.455	4734.545	5394.545	6241.818
0.0705	3284.286	3898.571	4435.714	5387.143
0.0858	2735.294	3311.765	3789.412	4678.824
(**L-ISOLEUCINE + BTEAC + H_2O**) system,0.001m				
0.0100	12550	13740	15570	16340
0.0251	5492	6036	6840	7184
0.0402	3582.5	3942.5	4460	4732.5
0.0554	2774.545	3085.455	3436.364	3645.455
0.0705	2280	2554.286	2871.429	3052.857
0.0858	2036.471	2260	2543.529	2675.294
(**L-ALANINE + BTEAC + H_2O**) system,0.003m				
0.0100	33150	37140	39880	43470
0.0251	14176	15984	17476	18956
0.0402	9172.5	10312.5	11167.5	12377.5
0.0554	7049.091	7758.182	8385.455	9565.455
0.0705	5662.857	6458.571	6851.429	7798.571
0.0858	4749.412	5575.294	5774.118	6897.647
(**L-ISOLEUCINE + BTEAC + H_2O**) system, 0.003m				
0.0100	30320	33470	37430	40740
0.0251	12652	14060	15768	16976
0.0402	8105	8967.5	10052.5	10740
0.0554	6089.091	6769.091	7590.909	8063.636
0.0705	4882.857	5435.714	6111.429	6415.714
0.0858	4191.765	4667.059	5245.882	5495.294
(**L-ALANINE + BTEAC + H_2O**) system, 0.005m				
0.0100	51360	57270	63670	68580
0.0251	22048	24092	27116	28784
0.0402	14490	15697.5	17282.5	18757.5
0.0554	10730.91	11845.45	13121.82	14509.09
0.0705	8620	9720	10624.29	11797.14
0.0858	7318.824	8181.176	9165.882	10135.29
(**L-ISOLEUCINE + BTEAC + H_2O**) system, 0.005m				
0.0100	47960	53770	59320	64670
0.0251	19572	21980	24068	26140
0.0402	12135	13567.5	15050	16347.5
0.0554	9036.364	10109.09	11072.73	11947.27

0.0705	7087.143	7928.571	8752.857	9468.571
0.0858	5951.765	6694.118	7318.824	7840

Table S9. Surface Tension σ of L-Alanine and L-Isoleucine as a function of different concentration (molality) of aqueous BTEAC solutions at temperatures 298.15 K.

(L-ALANINE + BTEAC + H_2O) system, 0.001m		**(L-ISOLEUCINE + BTEAC + H_2O) system, 0.001m**	
Temperature = 298.15 K		**Temperature = 298.15 K**	
Molality(mol/kg)	**Surface Tension(mN/m)**	**Molality (mol/kg)**	**Surface Tension(mN/m)**
0.0100	64.8	0.0100	69.6
0.0251	66.3	0.0251	68.7
0.0402	68.2	0.0402	67.9
0.0554	69.8	0.0554	67.3
0.0705	71.5	0.0705	66.2
0.0858	72.6	0.0858	65.5
(L-ALANINE + BTEAC + H_2O) system, 0.003m		**(L-ISOLEUCINE + BTEAC + H_2O)** system, 0.003m	
Temperature = 298.15 K		Temperature = 298.15 K	
Molality(mol/kg)	Surface Tension(mN/m)	Molality (mol/kg)	Surface Tension(mN/m)
0.0100	63.2	0.0100	70.9
0.0251	64.7	0.0251	69.9
0.0402	66.2	0.0402	68.8
0.0554	67.6	0.0554	68.1
0.0705	69.2	0.0705	67.3
0.0858	70.4	0.0858	66.1
(L-ALANINE + BTEAC + H_2O) system, 0.005m		**(L-ISOLEUCINE + BTEAC + H_2O)** system, 0.005m	
Temperature = 298.15 K		Temperature = 298.15 K	
Molality(mol/kg)	Surface Tension(mN/m)	Molality (mol/kg)	Surface Tension(mN/m)
0.0100	61.6	0.0100	72.3
0.0251	63.2	0.0251	71.9
0.0402	64.3	0.0402	70.9
0.0554	65.6	0.0554	69.8
0.0705	66.9	0.0705	68.9
0.0858	68.1	0.0858	67.1

*u (T) =0.01 K

Table S10. UV-vis spectroscopic data for the Benesi-Hildebrand double reciprocal plot of (L-Alanine+BTEAC) system at 298.15 K.

temp/k	[IL]/µM	[AA]/µM	A_0	A	ΔA	1/[AA]/M^{-1}	1/ΔA	Intercept	Slope	Ka/M^{-1}
	200	20	1.35881	1.27176	0.08705	50000	11.488			
	200	40	1.35881	1.18427	0.17454	25000	5.729			
298.15	200	60	1.35881	1.03157	0.32724	16666.667	3.056			
	200	80	1.35881	0.95082	0.40799	12500	2.451	0.18499	0.000221	837.0588
	200	100	1.35881	0.94882	0.40999	10000	2.439			
	200	120	1.35881	0.88389	0.47492	8333.333	2.106			
	200	140	1.35881	0.84842	0.51039	7142.857	1.959			
	200	160	1.35881	0.82742	0.53139	6250	1.882			
	200	180	1.35881	0.80897	0.54984	5555.556	1.819			

Table S11. UV-vis spectroscopic data for the Benesi-Hildebrand double reciprocal plot of (L-Isoleucine+BTEAC) system at 298.15 K.

temp/k	[IL]/µM	[AA]/µM	A_0	A	ΔA	1/[AA]/M^{-1}	1/ΔA	Intercept	Slope	Ka/M^{-1}
	200	20	1.35881	1.25306	0.10575	50000	9.456			
	200	40	1.35881	1.14359	0.21522	25000	4.646			
298.15	200	60	1.35881	1.04682	0.31199	16666.667	3.205			
	200	80	1.35881	0.98123	0.37758	12500	2.648	0.57895	0.000174	3327.299
	200	100	1.35881	0.9607	0.39811	10000	2.512			
	200	120	1.35881	0.87501	0.4838	8333.333	2.067			
	200	140	1.35881	0.83089	0.52792	7142.857	1.894			
	200	160	1.35881	0.80873	0.55008	6250	1.818			
	200	180	1.35881	0.7204	0.63841	5555.556	1.566			